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U. S. DEPARTMENT OF AGRICULTURE.

BUREAU OF PLANT INDUSTRY—BULLETIN NO. 261.

B. T. GALLOWAY, *Chief of Bureau.*

SEEDS AND PLANTS IMPORTED

DURING THE PERIOD FROM OCTOBER 1 TO DECEMBER 31, 1911:

INVENTORY No. 29; Nos. 31939 to 32368.

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BUREAU OF PLANT INDUSTRY.

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LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry,
Office of the Chief,
Washington, D. C., June 12, 1912.

Sir: I have the honor to transmit herewith and to recommend for publication as Bulletin No. 261 of the series of this Bureau the accompanying manuscript, entitled "Seeds and Plants Imported during the Period from October 1 to December 31, 1911: Inventory No. 29; Nos. 31939 to 32368."

This manuscript has been submitted by the Agricultural Explorer in Charge of Foreign Seed and Plant Introduction with a view to publication.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

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SEEDS AND PLANTS IMPORTED DURING THE PERIOD FROM OCTOBER 1 TO DECEMBER 31, 1911: INVENTORY NO. 29; NOS. 31939 TO 32368.

INTRODUCTORY STATEMENT.

This number of the inventories contains some of Mr. Frank N. Meyer's collections made in central Asia. They should attract the attention of experimenters whose work lies in the creation of hardier or more drought-resistant fruits, forage crops, or grains for the North and West. The difficulties of travel outside the railroad zone in Siberia and the still greater difficulty of getting the living plants he found there through to America alive give to these collections a special value. They represent only a small fraction of the interesting plants which Mr. Meyer found during his two and one-half years' absence. Some of his most interesting material will be described in later issues of the inventories.

Mr. Meyer is now in America and will spend the season studying the hundreds of plants which he secured both in China and central Asia, many of which are far enough advanced to show their adaptability to the American climate.

Mr. Meyer's new durum wheat, the Teiskaia (No. 32157), which was originated at the Bezenshook Agricultural Experiment Station in Russia, can not fail to attract the attention of the cerealists, in as much as it has proved extremely hardy at Samara, not being injured at all, while other varieties tested at the same time were killed out or at least severely injured.

The problem of studying *Medicago falcata*, the Siberian alfalfa, on the steppes of Siberia was given to Mr. Meyer with the result that he has found at least two distinct wild forms which are worthy of special attention. One is a bunchy upright form (No. 32178) from Ust Kamenogorsk, southwestern Siberia, and the other a very vigorous prostrate form (No. 32179), found on sandy, level stretches of land along the River Tom. This is suited, he thinks, for cultivation in meadows where grass is grown for hay production.

For introduction on the western ranges Mr. Meyer has secured a species of Astragalus (No. 32184) which is of bunchy erect habit, is eaten by cattle and horses, and which he believes may have value as a forage plant for cool semiarid climates.

His collection of four hardy Siberian relatives of the sulla (Nos. 32187 to 32189 and 32307), a remarkable forage crop of Spain and Tunis, brings up the question as to whether hardy hybrids which will grow in the South can not now be created. Sulla itself (*Hedysarum coronarium*) has never been made a success in America, owing presumably to its extreme susceptibility to frost. Mr. Swingle recently made the suggestion that the culture of sulla has probably had much to do with keeping up the fertility of the soil in the great sherry-wine region of Spain, which has produced famous wines since Shakespeare's time.

Among the 12 species of vetches (Nos. 32195 to 32206) which were found on the steppes of southwestern Siberia, some are thought by Mr. Meyer to have great promise as forage plants and should be introduced into the northwestern ranges.

Perhaps nothing which he found will create a more general interest than the Siberian cherry from the Ural district and western Siberia (No. 32224). This cherry, identified as a form of *Prunus fruticosa*, is a low bush not over 4 feet high, perfectly hardy and extremely drought resistant, so resistant in fact to extreme cold and drought that it can doubtless be grown throughout the entire Northwest. A plantation of these cherries resembles a tea plantation. The fruits are about the size of currants, are borne in great quantities, and make a most delicious preserve. Not only is this likely to prove a valuable plant as it stands, but two improved varieties have already been produced in Russia and Mr. Meyer has secured these (Nos. 32225 to 32226). The possibility of creating a race of perfectly hardy bush cherries by the use of this species is suggested by Mr. Meyer.

Fruit plants which will live in the interior of Alaska, where the temperature falls to −58° F. and the summers are short and cool, are difficult to find. It seems probable, however, that Nos. 32227 to 32228, two varieties of a large-fruited black currant called the Aldansky Vinograd from the Aldan Mountains of the Yakutsk Province of Siberia, will grow and fruit there and help to better the living conditions of such northern regions as Alaska and Labrador.

Of material secured through correspondence the caroá (No. 32260), a remarkable fiber plant from central Brazil, is worthy of special notice. If the information we have is correct, here is a plant related to the bromelias, which occurs on the plateau back of Bahia, is subject to an extremely dry climate, is capable of cultivation, yields a fiber which is much stronger and more resistant to sea water than manila hemp, and will produce a large quantity of the fiber per acre.

Sugar canes for fodder purposes have attracted considerable attention in the South, and the introduction of the Indian cane (No. 32257) from New South Wales, where it has proved very successful and is coming into great favor among the farmers, is likely to interest forage-crop men.

The asparagus is already such a highly developed vegetable that it may be questioned whether it can be much improved upon. However, the finding by Mr. Meyer of a form growing on dry mountain sides in the southeastern part of the Caucasus, the shoots of which have a special piquancy (No. 32091), and the introduction through Sir Percy Fitzpatrick of a wild form (No. 32271) from Table Mountain, South Africa, which he declares "is a great delicacy and to my taste better than any of the cultivated kinds," can hardly fail to arouse the interest of progressive asparagus growers.

We have scarcely begun to make the acquaintance of the cherimoya, although it is certainly a remarkable subtropical fruit, the cultivation of which in Madeira is a valuable industry. It is claimed that the annona does not bear well at sea level, but one of five varieties just introduced from Costa Rica (Nos. 32298–32301 and 32319) is reported to thrive on the coastal plain of that country.

Dr. Gustav Eisen, of the California Academy of Sciences, during his work in Italy for the Academy, has sent in from near Naples a remarkable plum called the Papagone (No. 32328), which seems not to have been previously introduced. According to Dr. Eisen it is 3 inches long, of a greenish yellow color, has a thin, slender stone, and is the finest plum he has ever eaten in any country.

As heretofore, this inventory has been prepared by Miss Mary A. Austin. For the nomenclature and the notes on the general geographical distribution of the various species Mr. H. C. Skeels is responsible, working, however, under the general direction of Mr. Frederick V. Coville, of the Office of Taxonomic and Range Investigations. The general supervision of this inventory, as of all the publications of this office, has been in the hands of Mr. S. C. Stuntz.

David Fairchild,
Agricultural Explorer in Charge.

Office of Foreign Seed and Plant Introduction,
Washington, D. C., April 29, 1912.

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INVENTORY.

31939. MEDICAGO SATIVA L. **Alfalfa.**

From Gilghit, Kashmir, India. Received through Mr. F. Booth Tucker, Salvation Army, Simla, India, October 2 and 4, 1911.

31940. TRITICUM AESTIVUM L. **Wheat.**

From near Cumpas, Sonora, Mexico. Presented by Mr. Alexander V. Dye, American consul, Nogales, Sonora, Mexico. Received October 4, 1911.

"This is known locally as *Sonora* wheat; it is a hard variety and the only one grown in this consular district." (*Dye.*)

31941. CROTALARIA sp.

From Puerto Bertoni, Paraguay. Presented by Dr. Moises S. Bertoni, Estacion Agronomica. Received October 5, 1911.

31943 to 31945.

From Quetta, India. Presented by Mr. A. Howard, Imperial Economic Botanist, Agricultural Research Institute. Received October 9, 1911.

Seeds of the following:

31943 and 31944. MEDICAGO SATIVA L. **Alfalfa.**

31943. *Momgchiri.*

31944. *Qandhari.*

31945. TRIFOLIUM SUAVEOLENS Willd. **Shaftal clover.**

31946 to 31950. MANGIFERA INDICA L. **Mango.**

From Monghyr, North India. Purchased from Mr. Lalit Mohan Sinha, Lalloo Pokhar Road. Received October 9, 1911.

Cuttings of the following:

31946. *Malda* No. 1.

31947. *Malda* No. 2.

31948. *Malda* No. 3.

31949. *Fazli* No. 1.

31950. *Fazli* No. 3.

31951 and 31952. CYTISUS PROLIFERUS L. f. **Escobon.**

From Canary Islands. Presented by Dr. George V. Perez, Puerto Orotava, Teneriffe. Received October 9 and 11, 1911.

Seeds of the following; quoted notes by Dr. Perez:

31951. Variety *canariae.* "This is the escobon of Grand Canary Island. It is an excellent fodder for goats and a sister plant of the tagasaste. I would suggest it being tried in the mountains of extreme southern California."

31952. "A tall shrub that grows here at from 4,000 to 6,000 feet above the level of the sea. Sometimes it is spoken of as variety *angustifolia.* Goats eat it, but the tagasaste (No. 28827) is better, also the variety from Grand Canary Island (No. 31951). However, the Teneriffe escobon, which is, as it were, the type plant, has many uses. Cartwrights use it for wheels in preference to any other timber. Its height is 20 to 25 feet, and the diameter of the trunk 1 foot."

31953. SPATHODEA CAMPANULATA Beauv.

From Java. Received through Dr. B. T. Galloway, Chief of the Bureau of Plant Industry, United States Department of Agriculture, October 10, 1911.

Distribution.—A tree bearing racemes of scarlet or crimson flowers, found in the countries along the western coast of Africa, from Sierra Leone southward to Angola in Portuguese West Africa. Cultivated as a street tree in Java.

31954 to 31956.

Presented by Dr. F. Mader, Nice, Alpes Maritimes, France. Received October 5, 1911.

Seeds of the following; quoted notes by Dr. Mader:

31954. PRUNUS BRIGANTINA Villars.

"Seed collected from a little group growing between the hazel (*Corylus avellana*) in the Miniera Valley, 1,200 meters [3,930 feet] above sea level, and with a climate like that of your Alleghenies. Of course, the species, found here up to 2,000 meters [6,560 feet], is very hardy, as the Barcelonnette Valley, where it especially abounds, has an almost Siberian climate, frequently −25° C. (−13° F.) and lower in the winter, and up to 35° C. (95° F.) in the summer. It is the true Briançon plum of French foresters, being now extensively planted in the high valleys, also on the Italian side, and has proved to be excellent for sheltering river banks, roadsides, stony ravines, or avalanche beds. The wood, which is very scarce, seems very much like that of *Prunus* (*Cerasus*) *mahaleb*. The fruit is free from sweetness and nearly insipid, but would be suitable for marmalades, etc. The seeds yield an oil used by poor mountaineers for cooking purposes, under the name 'huile de marmotte.' The species is not uncommon in the high valleys of the dry western part of the Maritime and Cottian Alps, from the Var to Briançon; in the more rainy eastern valleys it is much rarer, there being only scattered plants or little patches on shady rocks or other places."

31955. MEDICAGO SATIVA L. **Alfalfa.**

31956. MEDICAGO FALCATA L.

31957 to 31975. PELARGONIUM spp. **Geranium.**

From Kew, England. Presented by Dr. David Prain, director, Royal Botanic Gardens. Received October 12, 1911.

Cuttings of the following, procured for purposes of breeding with the varieties commonly cultivated, with a view to adding to their bedding qualities:

31957. PELARGONIUM CAPITATUM (L.) L'Herit.

Distribution.—A trailing, partly shrubby plant with rosy-purple flowers in dense heads, found in the vicinity of Table Mountain and on the flats around Cape Town, South Africa.

31958. PELARGONIUM QUERCIFOLIUM (L. f.) L'Herit.

Distribution.—A hairy, much-branched shrub, found in South Africa, and well known in cultivation as the "oak-leaf geranium."

31959. PELARGONIUM VIOLAREUM Jacq.

Distribution.—A diffuse shrub with the two upper petals dark red and the three lower ones white, growing on the slopes of the mountains in South Africa.

31960. PELARGONIUM RAPACEUM (L.) Jacq.

Distribution.—On dry stony mountain sides in the vicinity of Cape Town and in the Stellenbosch and Swellendam districts of South Africa.

31957 to 31975—Continued.

31961. × PELARGONIUM TRICUSPIDATUM L'Herit.

Apparently a hybrid of obscure origin.

31962. PELARGONIUM CORDIFOLIUM (Cav.) Curtis.

Distribution.—A shrub found on the slopes of the mountains from the valley of the Olifant's Vlei River southward to the Cape, in South Africa.

31963. × PELARGONIUM BLANDFORDIANUM (Andr.) Sweet.

Apparently a garden hybrid of unknown origin.

31964. PELARGONIUM QUERCIFOLIUM (L. f.) L'Herit.

Variety *major.*

31965. PELARGONIUM RADULA (Cav.) L'Herit.

Variety *major.*

Distribution.—A large, densely branched bush found on the mountain sides in the Tulbagh, Uitenhage, and Albany districts of South Africa.

31966. PELARGONIUM RADULA (Cav.) L'Herit.

31967. PELARGONIUM UNICOLORUM Willd. (?)

These were received under the name *Pelargonium unique aurora*, which may be a varietal name, but the possibility of error between that name and *unicolorum* is very suggestive. *P. unicolorum* is apparently a hybrid of garden origin.

31968. PELARGONIUM MALVAEFOLIUM Jacq. f.

Distribution.—Described from cultivated plants and is probably a garden hybrid.

31969. PELARGONIUM CRISPUM (Bergius) L'Herit.

Distribution.—A slender shrub with strongly scented leaves found on shrubby mountain slopes in South Africa.

31970. PELARGONIUM DENTICULATUM Jacq.

Variety *major.*

Distribution.—A tall weak-stemmed plant found on the mountain slopes in the southern part of South Africa.

31971. PELARGONIUM VISCOSISSIMUM Sweet.

Distribution.—Described from garden plants grown from seed received from the Cape.

31972. PELARGONIUM ZONALE (L.) L'Herit.

Distribution.—A large shrub found on hillsides in the western districts of South Africa.

31973. PELARGONIUM BALBISIANUM Spin.

Distribution.—Probably a garden hybrid.

31974. PELARGONIUM CORDIFOLIUM (Cav.) Curtis.

31975. PELARGONIUM GRANDIFLORUM Willd.

Distribution.—A shrubby plant with leaves palmately lobed and coarsely toothed, bearing large white flowers, found in the vicinity of Giftberg, in South Africa.

31976. COFFEA LIBERICA Bull. **Coffee.**

From Liberia, West Africa. Presented by Mr. Henry O. Stewart, Monrovia, Liberia. Received October 14, 1911.

Distribution.—Liberia and Sierra Leone in Upper Guinea and in the Angola district of Portuguese West Africa in Lower Guinea, on the west coast of tropical Africa.

31977. Phormium tenax Forst. **New Zealand flax.**

From California. Presented by Mr. P. D. Barnhart, Los Angeles. Received October 9, 1911.

Variegated variety.

Seeds.

31978. Annona muricata L. **Soursop.**

From Cuba. Presented by Mr. Robert L. Luáces, agricultural engineer, Camaguey, Cuba. Received October 9, 1911.

"Seed of a wild variety little known even here in Cuba. It is a beautiful tree, growing as much as 30 feet high on the banks of streams and ponds. The fruit is more rounded in shape than the cultivated, and although acid in taste can be eaten. This I believe will make a good stock for *Annona squamosa.*" (*Luáces.*)

31979. Medicago sativa varia (Mart.) Urban. **Sand lucern.**

From Schoeningen, near Colbitzow, Pomerania, Prussia, Germany. Grown on the farm of Mr. Ernest Schlange. Presented by Mr. Joseph E. Wing, Mechanicsburg, Ohio. Received October, 1911.

"This wild yellow alfalfa was found growing on waste sandy ground near a field of cultivated lucern." (*Wing.*)

31980. Petasites laevigatus (Willd.) Reichenb.

From near Sminogorsk, southwestern Siberia. Received through Mr. Frank N. Meyer, agricultural explorer, Bureau of Plant Industry, October 11, 1911.

"(No. 988.) An interesting hardy aquatic perennial, growing on the banks of swift-flowing, shallow streamlets, extending often for several yards in the water, but not occurring in places over 2 feet in depth. Of value as a decorative plant along watercourses in parks in the cooler sections of the United States." (*Meyer.*)

Rhizomes.

Distribution.—In Bohemia and the southwestern part of Siberia.

31981. Citrus sp. **Sour citron.**

From Nagpur, Central Provinces, India. Procured by Mr. R. S. Woglum, Bureau of Entomology, United States Department of Agriculture. Received October 14, 1911.

"*Zamburi*, sour citron. The chief stock used for budding in the Central Provinces, India. The fruit is sour like a lemon, and has a yellow-covered rind. In appearance, color, taste, and character of flesh it is very similar to a California Eureka lemon allowed to overmature on the tree." (*Woglum.*)

Seeds.

31982. Bryophyllum pinnatum (L. f.) Kurz.

From Paraguay. Presented by Mr. C. F. Mead, Villa Encarnacion, Paraguay. Received October 14, 1911.

"This is called locally *La Milagra* (the miracle). A curiosity even for these parts. A low-growing shrub with waxlike leaves of which snails, etc., are very fond. Leaves dropping off from the plant reproduce themselves around the outer edges, or if you pin a leaf on a wall or other place it will start growing." (*Mead.*)

Distribution.—Probably a native of tropical Africa and generally cultivated throughout the Tropics.

31983. Castilla sp. **Central American rubber.**

From Costa Rica. Presented by Mr. Carlos Wercklé, San Jose. Received October 20, 1911.

Seeds.

31984. Persea americana Miller. **Avocado.**

Presented by Mr. C. F. Mead, Villa Encarnacion, Paraguay. Received October 23, 1911.

"*Abogado*, otherwise called *abagate, pagua pauta, abaji, aguacate, ahuaca,* or alligator pear. Seeds from fruit found in the market at Buenos Aires. Fruit small, pear shaped, and dark red in color. Sold in market under the name of 'red Chilian abacate,' or 'red abacate from Chile.'" (*Mead.*)

31985 to 31998. Ipomoea batatas (L.) Poir. **Sweet potato.**

From Peru. Presented by Mr. Antonio Graña, Huando, Chancay, Peru. Received October 10, 1911.

Tubers of the following; quoted notes by Mr. Graña:

"These sweet potatoes bear different names by which they are known by the people of the country without indicating thereby any scientific classification. Neither are they grown as distinct varieties, as they are produced mixed together."

31985. "*Vapor.* Produces in four months."

31986. "*Huamino.* Produces in five months."

31987. "*Cochino.* Produces in six months."

31988. "*Azadura de vaca.* Produces in five months."

31989. "*Romero.* Produces in five months."

31990. "*Supano blanco* (white). Produces in five months."

31991. "*Supano prieto* (black). Produces in five months."

31992. "*Papa.* Produces in four months."

31993. "*Limeño.* Produces in five months."

31994. "*Plaza.* Produces in four months."

31995. "*Niño.* Produces in five months."

31996. "*Cambrax.* Produces in five months."

31997. "*Tabardio.* Produces in five months."

31998. "*Yemade huevo.* Produces in five months."

31999 to 32001. Crotalaria spp.

From Buitenzorg, Java. Sent in by the Java Department of Agriculture, at the request of Mr. C. V. Piper, Bureau of Plant Industry. Received October 5, 1911.

Seeds of the following; under trial at the Buitenzorg garden as green-manure crops and will be tested here for the same purpose.

31999. Crotalaria alata Hamilton.

Distribution.—From the mountains in the Province of Assam in northeastern India southeastward to Java.

32000. Crotalaria leioloba Bartl.

Distribution.—On the lower mountain slopes in northern India from Nepal to Assam, and eastward through the Malay Archipelago as far north as the Philippines.

32001. Crotalaria saltiana Andrews.

32002 and 32003.

From Guatemala. Presented by Mr. S. Billow, Guatemala, Central America. Received October 12, 1911.

Seeds of the following; quoted notes by Mr. Billow:

32002. Cereus sp. **Pitaya.**

"This fruit is produced about 30 miles from Guatemala City, and I understand that it is closely allied to the cactus family. It is red colored and is very delicious. The blossom of the plant, as well as the fruit, is eaten."

32003. Passiflora ligularis Juss. **Passion fruit.**

"This is about the size of a large egg and the seeds are surrounded with a gelatinous substance. When ripe the seeds and this jellylike substance are eaten; when green the whole fruit is stewed in sugared water and eaten, and the seeds and jelly substance are thrown away. It grows on a vine and is largely consumed."

32004. Phytolacca acinosa Roxb.

From Yokohama, Japan. Purchased from the Yokohama Nursery Co. Received October 17, 1911.

Variety *kaempferi.*

Seeds.

See No. 29133 for distribution of this species.

32005. Solanum haematocladum Dunal.

From Brussels, Belgium. Obtained by Prof. William R. Lazenby, of the Ohio State University, Columbus, Ohio. Received October 18, 1911.

"This is a vigorous growing, red-fruited species." (*Lazenby.*)

Distribution.—Not known except from Bolivia.

32006. Dimocarpus longan Loureiro. **Longan.**

The seeds of this Chinese sapindaceous tree were received under the name *Euphoria longana*, which was published by Lamarck (Encyclopédie Méthodique Botanique, vol. 3, p. 574) in 1791. The generic name Euphoria was used by Jussieu (Genera Plantarum, p. 247) in 1789, who characterized the genus and mentioned the plants known by the Chinese names litchi and longan as belonging to it. If the litchi is regarded as the type of the genus Euphoria, the name Euphoria becomes a synonym of Litchi, the generic name of the litchi tree. If the longan be regarded as the type species of Euphoria the name Euphoria can not be maintained because the longan had not at this time received a binomial name, and as Jussieu does not describe it nor give it a binomial name he can not be said, according to present rules of botanical nomenclature, to have published the generic name Euphoria. The first generic name published for the longan is Dimocarpus, published in 1790 by Loureiro (Flora Cochinchinensis, vol. 1, p. 233) and Loureiro's name for this species is here used.

From Kiayingchow, China. Presented by Mr. George Campbell. Received October 19, 1911.

"Seeds from some particularly large and fine fruit." (*Campbell.*)

The tree is handsome and may be used as a shade tree, also as a stock on which to bud the litchi.

Distribution.—Found in India, where it is probably native, and eastward to China and through the Malay Archipelago.

32008. OCIMUM VIRIDIFLORUM Roth. **Mosquito plant.**

From Southern Nigeria, West Africa. Presented by Mr. W. H. Johnson, director, Agricultural Department, Ibadan, Southern Nigeria. Received October 20, 1911.

"This is the West African mosquito plant. This plant is known locally to possess valuable qualities as an insectifuge, but I think its use is really not much practiced by the natives. The foliage is usually hung up in dwelling houses in the green state to keep away mosquitoes. The dried plant is also burned and the resulting smoke therefrom is considered to be useful for the same purpose." (*J. W. Henderson, acting director.*)

Distribution.—A herbaceous perennial found along the western coast of Africa from Sierra Leone southward to Angola.

32009 to 32011. HIBISCUS spp.

From Gold Coast, West Africa. Presented by Mr. A. R. Gould, curator, Botanic Garden, Aburi. Received October 20, 1911.

Seeds of the following; quoted notes by Mr. Gould:

32009. HIBISCUS CANNABINUS L. **Ambari.**

"White, large leafed."

Distribution.—Naturalized or cultivated throughout the Tropics; probably wild in India.

32010 and 32011. HIBISCUS SABDARIFFA L. **Roselle.**

32010. "Red-stalked variety."

32011. "White-stalked variety."

"Interesting indigenous fiber plants cultivated by the natives in the interior."

32012 to 32013. CHRYSANTHEMUM CINERARIAEFOLIUM (Trev.) Vis. **Pyrethrum.**

From Dalmatia, Austria-Hungary. Presented by Mr. K. Portele, Imperial Ministry of Agriculture, Vienna, Austria-Hungary. Received October 20, 1911.

Seeds of the following:

32012. From Cattaro.

32013. From Ragusa.

"This species grows wild here in Dalmatia and is also cultivated."

32014. CRATAEGUS COCCINEA L. **Hawthorn.**

From Seattle, Wash. Collected by Messrs. P. H. Dorsett and Peter Bisset, of the Bureau of Plant Industry. Received October 25, 1911.

"A large-fruited Crataegus collected in Woodland Park, Seattle." (*Bisset.*)

32015 and 32016. PHOENIX DACTYLIFERA L. **Date.**

Grown at the Cooperative Date Garden, Tempe, Ariz., Mr. F. H. Simmons in charge. Received in the spring of 1911. Numbered October 25, 1911.

Seeds of the following:

32015. *Rhars.*

32016. *Deglet Noor.*

32017. PRUNUS sp. **Plum.**

From Pietermaritzburg, Natal, South Africa. Presented by Mr. T. R. Sim. Received October 28 and November 3, 1911.

Methley. See No. 31652 for description.

32018. ANNONA SQUAMOSA L. **Sweetsop.**

From Cuba. Presented by Mr. Roberto L. Luáces, Camagüey, Cuba. Received October 24, 1911.

Seeds.

32019. BOSWELLIA sp. **Frankincense.**

From the island of Socotra. Procured by Mr. Charles K. Moser, American consul, Aden, Arabia. Received November 17, 1911.

Mr. Moser made a special expedition from Aden to the island of Socotra at the mouth of the Red Sea at the request of the Office of Foreign Seed and Plant Introduction, and secured two trees of this so-called incense tree. He was enabled to do this through the assistance of the British Resident at Aden, who invited Mr. Moser to accompany him as a guest on a British Government vessel upon which he himself made a special trip to the island of Socotra for the British Government. Two trees were obtained by Mr. Moser during a 26-hour wait of the vessel, being dug out of the soil by native Socotran boys, at an elevation of 2,500 feet above the sea. These were packed in kerosene cases filled with Socotran soil and were taken by caravan to the boat and transported to Aden. They remained in these tins until October 7, 1911, when they were sent to the Department. The following report is taken from Mr. Moser's letter of April 29, 1911:

"We went into the mountains directly south of Tamarida Bay to a place called Adho Dimellus in the Haghier Range. We first saw the tree about 9 miles in a direct line from the sea at an altitude of about 2,800 feet. The trees were apparently in full flower, with immature fruits intermingled. There were no signs of old seeds, and the Socotrans pretended never to have seen any seeds, but they could be easily gathered, and I have made arrangements to procure some during the summer. The soil was very rocky, a red, rich-looking, easily disintegrated granite, out of which, higher up, arose limestone escarpments. The temperature during the night was about 65° F., but at noon it was nearly 90° F. All the trees we saw were nearly the same size, and I was astonished to find in an area of several miles no small shoots or saplings.

"The natives call the frankincense tree *tee-lah-ah* (spelled phonetically) and its product *lu-ban*. From the samples which I saw and from the reputation it bears in the Aden market, it seems certain that the quality of Socotran *lu-ban* is distinctly inferior to that of Somali and the Hadramaut, and the produce per tree is much smaller. The trees we saw were from 20 to 30 feet in height and from 8 to 10 inches in diameter, with scraggly, swollen branches, which scarcely tapered and ended in tufts of sumac-like leaves of a general yellowish color clotted with autumnal reds. The leaves were multifoliate, 7 to 11 leaflets, 1½ to 2½ inches long, elliptic, regularly crenated, and medium ovate. The flowers are very thickly clustered on thick stalks, 5 to 8 inches long, with nearly a uniform diameter of five-eighths to three-fourths of an inch; in color the flowers were a deep pink or bright magenta, much resembling a thick cluster of double geraniums. The immature fruits among them were one-half to five-eighths of an inch long and pear shaped. The bole and branches were of a livid greenish, almost translucent hue, smooth and covered with blotches, resembling gangrenous human flesh; the outer bark sheds in thin, yellowish white, papery strips or peels. When cut, even with the thumb-nail, the bole, the boughs, and the stalks yield a clear, sticky, viscous fluid with a rich aromatic odor. This exudation usually remains clear until and after it hardens, but I saw some trees with clear 'tears' and on others brownish or amber-colored ones. Every day I have cut my two specimens with the thumb-nail, and with but two exceptions the exudation has been clear and colorless; on those two occasions the wound was followed by a milky drop, but I have been unable to discover why.

"The Socotra olibanum flowers in April and the gum is collected any time after May, which is the beginning of the wet season. It is usually collected, however, during June, July, and August. The Socotran merely goes among the wild trees, giving each about a dozen deep, oblique slashes, 2 or 3 inches long, which he tears open into a kind of pocket at the lower end with a wrench of his knife. The tree is then left to deposit its sap in these pockets for three weeks or a month, at the end of which time the collector comes with his knife and basket and cuts the collected nodules 'or tears' away with pieces of bark. He then either makes new cuts or deepens the old ones and again awaits the harvest in another three or four weeks. The process is repeated until September. The *lu-ban*, which overflows the wound and runs down the tree, is regarded as of less value than that which remains in the pockets. A Socotran average tree is said to yield from 1 to 5 pounds of *lu-ban* per season, while the yield in Somali is much larger. Its value in the island is simply anything which the collector, who has little use for money, can persuade the Arab trader to give him for it in rice, goats, or cotton shirting. In Aden it is worth from 6 to 12 cents per pound, according to quality, while the Somali *lu-ban* is worth from 10 to 24 cents per pound.

"I must add that we found the olibanum growing only on the inside, protected slopes of the mountain, that its range seemed to be from 2,000 to 4,000 feet, and that while we only saw it in a red granite soil, we were told that it grew equally well out of fissures in the limestone heights above us. The climate of Socotra is, of course, very dry and not so hot as Arabia. We found the earth exceedingly dry, and were informed that rain never falls in the Haghier Hills except during the rainy months from May to August or September.

"There is no cleaning of the collected *lu-ban*, but as soon as it hardens a little after being cut from the trees it is ready for market." (*Moser.*)

"The frankincense tree is supposed to have been the tree which furnished the frankincense of the ancients, and the hardened drops of gum are now used very extensively in Roman Catholic churches as incense, being burned in the censers. The tree will probably thrive only in the dry, almost frostless, areas of the Southwest." (*Fairchild.*)

32020. CITRUS LIMETTA Risso. **Lime.**

From Burringbar, New South Wales, Australia. Presented by Mr. B. Harrison, Burringbar. Received July 31, 1911. Numbered October 15, 1911.

Seeds.

32021. STIZOLOBIUM CINEREUM Piper and Tracy.

From Amani, German East Africa. Presented by Dr. A. Zimmermann, director, Biologisch Landwirtschaftliches Institut, Amani. Received October 25, 1911.

32022 to 32025. STATICE spp.

From Canary Islands. Presented by Dr. George V. Perez, Puerto Orotava, Teneriffe. Received October 5, 1911.

Seeds of the following; quoted notes by Dr. Perez:

32022. STATICE ARBOREA Brouss.

Variety *frutescens*. "Native of Teneriffe. Botanically this is a dwarf form of *arborea*."

32023. STATICE MACROPTERA Webb and Berth.

"Native of the island of Hierro."

32024. STATICE PEREZII Stapf.

"Native of Teneriffe. Newly discovered species."

32025. STATICE PUBERULA Webb.

"Gathered wild at Famara, Lanzarote. This seed keeps 2 or 3 years."

32026 and 32027.

From near Ust Kamenogorsk, southwestern Siberia. Received through Mr. Frank N. Meyer, agricultural explorer, Bureau of Plant Industry, October 30, 1911.

Roots of the following:

32026. HEDYSARUM SPLENDENS Fisch.

"(No. 989, October 2, 1911.) A rare and interesting legume occurring on stony mountain slopes here and there along the Irtish River. May possess value as a forage plant in semiarid regions where irrigation is not practicable and where the soil is stony and sterile." (*Meyer.*)

32027. ASTRAGALUS sp.

"(No. 990, October 2, 1911.) A small Astragalus, quite common on rocky and sterile places. Is eagerly browsed by horses and cattle. Of value like the preceding number." (*Meyer.*)

32028 to 32032.

Presented by Dr. A. Robertson Proschowsky, Nice, France. Received October 30, 1911.

Seeds of the following; quoted notes by Dr. Proschowsky:

32028. OXYTENANTHERA ABYSSINICA (Rich.) Munro.

From Abyssinia.

"The following plants from Nice, France, are exceedingly drought resistant and are liked by herbivorous animals."

32029. CORONILLA VALENTINA L.

Distribution.—The countries bordering on the western part of the Mediterranean from Spain to Italy, Corsica and Sardinia, and in northern Africa.

32030. LOTUS ORNITHOPODIOIDES L.

See No. 7730 for description.

Distribution.—The countries bordering on the Mediterranean from Spain to Syria and in northern Africa.

32031. MEDICAGO SATIVA L. **Alfalfa.**

"Seeds of the wild-growing form."

32032. MEDICAGO ORBICULARIS MARGINATA (Willd.) Benth.

32033 and 32034. ANNONA CHERIMOLA Miller. **Cherimoya.**

From Costa Rica. Presented by Mr. Carlos Wercklé, San Jose. Received October 27, 1911.

Seeds of the following; quoted notes by Mr. Wercklé:

32033. "From Don Buenaventura Corrales."

32034. "First-class Annona from Vindas, in San Pedro del Mojon."

32035. PHASEOLUS sp.

From South Sea Islands (Oceania). Presented by Rev. C. N. Field, 33 Bowdoin Street, Boston, Mass. Received November 3, 1911.

"I have never tasted a variety as delicious as this one. The seeds were given to me by a man who had traveled around the world. They thrived much better than ordinary scarlet-runner beans; on very poor soil near Boston they grew 10 feet high and were remarkably productive. They are eaten baked after removing the pods and found especially sweet. They have a very pretty purple flower." (*Field.*)

32036 and 32037. LANGUAS GALANGA (L.) Stuntz. **Galangale.**

Roots of this plant were received under the name *Alpinia galanga* (L.) Willd. The genus Alpinia, however, was based on a single species, *Alpinia racemosa* L. (Species Plantarum, vol. 1, 1753, p. 2). This is not now considered congeneric with the present plant, which was first published as *Maranta galanga* L. (Species Plantarum, ed. 2, vol. 1, 1762, p. 3). As the type of the genus Maranta is *Maranta arundinacea*, also not congeneric with the galangale under discussion, it is necessary to adopt for this plant the next later generic name, Languas, published in 1783 by Koenig in Retzius, Observationes, vol. 3, p. 64.

From Buitenzorg, Java. Presented by the Director of Agriculture at the request of Mr. C. V. Piper. Received November 2, 1911.

Distribution.—Throughout India from the foot of the Himalayas to Ceylon and Malakka; generally cultivated in the Tropics.

Roots.

32038 to 32042.

From Chinese Turkestan. Received through Mr. Frank N. Meyer, agricultural explorer, Bureau of Plant Industry, November 3, 1911.

Seeds of the following:

32038. TRITICUM AESTIVUM L. **Wheat.**

From Kizil Bulak, Tien Shan Mountains, Chinese Turkestan. Altitude, 6,650 feet.

"(No. 1583a, March 4, 1911.) A summer wheat of a very dark color, called *Kara boogdai*, meaning black wheat. Sown in early April and grown under irrigation. Of value probably for sections of the United States where the summers are not only short but hot and dry." (*Meyer.*)

32039. TRITICUM AESTIVUM L. **Wheat.**

From Kara Tugai, Tekes Valley, Tien Shan Mountains, Chinese Turkestan. Altitude, 3,900 feet.

"(No. 1584a, March 6, 1911.) A rare local variety of summer wheat of great excellence. Grains large, of a pale-yellow color, ears very long. The flour made from this wheat makes a fine quality of substantial bread. Sown in April and raised under slight irrigation. To be tested in the western sections of the United States." (*Meyer.*)

32040. ORYZA SATIVA L. **Rice.**

From Aksu, Chinese Turkestan.

"(No. 1585a, February 27, 1911.) A local variety of wet-land rice, called *Kara kiltrick*. The variety absorbs a large quantity of water in cooking; the grains always remain separate, are of a snow-white color, and a very large size. Although expensive in comparison to the lower grades, yet it is considered economical, as only one-third to one-half the quantity is needed to fill the same cooking vessel. If a specially fine quality of rice is wanted, the plants are set out by hand, with the result that the rice treated in this manner is larger, of finer quality, and greater in yield. However, as the labor connected with such practice is too expensive to justify the returns, this variety is usually sown broadcast, like all rices in this part of the world.

"As the soil around Aksu is decidedly alkaline, this rice will be able to stand a fair amount of alkali. Otherwise it can be tested in the same way as Nos. 1571a to 1580a, inclusive (S. P. I. Nos. 31823 to 31832)." (*Meyer.*)

32038 to 32042—Continued.

32041. Oryza sativa L. **Rice.**

From Aksu, Chinese Turkestan.

"(No. 1586a, February 27, 1911.) A local variety of wet-land rice, called *Ak kiltrick*. In looks and yield very much the same as the preceding number, but not near so good in quality; swells but little in cooking and is not so white and large. To be tested like the preceding number." (*Meyer.*)

32042. Hordeum sp. **Barley.**

From Yengi Malah, Tien Shan Mountains, Chinese Turkestan. Altitude, 7,950 feet.

"(No. 1587a, March 4, 1911.) A black hull-less summer barley, grown under irrigation on rocky fields at high altitudes. A very rare local variety, apparently a mutation which has not yet been fixed. It is intermixed with other varieties and with wild black and white oats, which may also prove to be interesting. Locally used as a feed for horses and may possibly be of value for growing for this purpose in elevated arid and semiarid regions. To be tested especially in the intermountain sections of the United States." (*Meyer.*)

Note.—See Nos. 32280 and 32281 for oats and barley picked out of this lot.

32043 to 32060.

From Brazil. Presented by Mr. Welman Bradford, Crowley, La. Received October 2, 1911.

Seeds of the following; quoted notes by Mr. Bradford:

32043. Michelia champaca L. **Champac.**

"Magnolialike tree having yellow blooms. Not as sweet as our magnolia. Grows 30 feet high. It is being planted in Sao Paulo as an ornamental street tree."

Distribution.—A tall tree found wild in the forests on the temperate slopes of the Himalayas in northern India; often cultivated.

32044 to 32046. Annona squamosa L. **Sweetsop.**

"*Fruta de conde.* This is known as the *Princess fruit;* there is another variety called the *Prince*. In my estimation it is the best fruit that ever grew. The largest plants I have noticed are about 12 feet high, and the largest fruit about 5 inches in diameter."

32047. Passiflora sp. **Passion fruit.**

"*Maracuja roxo* (purple maypop). This is round, very hard, and stiff. Should not be eaten until quite ripe, as it is too sour."

32048 and 32049. Passiflora sp. **Passion fruit.**

"Said to be the best to eat."

32048. "*Maracuja amarello* (yellow maypop). Large."

32049. "*Maracuja amarello* (yellow maypop). Small."

32050. Passiflora ligularis Juss. **Passion fruit.**

"*Maracuja guasu* (large maypop)."

32051. Erythrina crista-galli L.

See No. 29655 for description.

32052. Rollinia sp.

32053. Toluifera sp.

"A tall forest tree. Wood deep purple, oily, very sweet scented, proof against the attack of ants, absolutely everlasting. Posts made from it never rot, and trunks and furniture are insect proof."

32043 to 32060—Continued.

32054. Dolicholus phaseoloides (Swartz) Kuntze.

"A little vine that grows wild and bears a very pretty bean. All the people of the East Indies use these beans for ornament, stringing, etc."

Distribution.—In the West Indies and from Nicaragua southward through tropical South America.

32055. Ormosia monosperma (Swartz) Urban.

"A hardwood timber forest tree growing on the banks of rivers."

32056. Gossypium sp. **Cotton.**

32057. Oryza sativa L. **Rice.**

"Black rice. Planted here in fields to fool the birds."

32058. Canavali sp. (?)

"This is known throughout the Parahyba Valley, also between Sao Paulo and Rio Janeiro. It is an easy and luxuriant grower found in the woodlands and in the timber, thrives in the dense shade, the vines climbing up to the tops of the trees at least 20 feet until they find the sun. Not cultivated at all. Some people seem to think it is poisonous and say that the cattle after eating the beans and pods and drinking water will die. It is a prolific bearer; the pods are about 8 inches long, and as well as I can remember are smooth. It has three leaflets somewhat separated from each other, not close, as with the cowpea."

32059. Astragalus sinicus L.

"From Parahyba Valley. A giant clover growing 3 or 4 feet high. Flowers pink to yellow. Bears a slightly curved, fuzzy pod 1½ inches long. The roots are well covered with nodules. It is a weed at present, but by planting closely it will get finer and softer. May be of value for plowing under."

32060. Vigna sinensis (Torner) Savi. **Cowpea.**

"From a bean exposition in Sao Paulo."

32061 and 32062. Andropogon sorghum (L.) Brot. **Sorghum.**

From Buitenzorg, Java. Presented by the Department of Agriculture, at the request of Mr. C. V. Piper, Bureau of Plant Industry. Received October 5, 1911.

Seeds of the following; quoted notes by Mr. Carleton R. Ball:

32061. "Seeds brown (clay on the included portion and burnt sienna to claret brown on the exposed tips), narrowly obovate, cuneate at the base; 3 by 5 millimeters in diameter. Glumes transversely shouldered, black, and indurated below the shoulder, somewhat scarious above; more or less pubescent."

32062. "A form of white durra. Seeds white, sublenticular; 3½ by 4 millimeters to 4 by 4½ millimeters in diameter. No glumes present."

32063. (Undetermined.)

From Brazil. Presented by Mr. Fred. Birch, Theophilo Ottoni, Minas Geraes, Brazil. Received November 4, 1911.

"Seeds of a forest shrub which I have discovered here. It is a rare pleasure to find a new fruit thus. It is a shrub about 4 feet high, of very compact growth; stem and branches tough and elastic, leaves dark green, glossy, and fairly tough, about 1½ to 2 inches long by 1 inch wide, built like so many of the leaves here for living through a long dry season. I have not yet seen the flower. The fruit is a sort of elongated cherry, about seven-eighths of an inch long by three-eighths to seven-sixteenths of an inch thick in the middle, black or purple black, and with a bright glossy skin. They usually occur singly, strung along the branches, but sometimes there are two together. From one small shrub we have eaten. I think, about 300 fruits. The flesh is about one-eighth of an inch thick over the seed and it is soft and juicy, tasting more like a black cherry than any other northern fruit I can think of. When I recall the size of the wild fruit from which the common cherry is supposed to have originated, I think this little fruit promises well to repay cultivation. I would suggest trying it in Florida or California or Texas; the last-named State may be the best for it. From mid-August until the end of September is the time of fruiting—i. e., in early spring here." (*Birch.*)

32064 to 32069.

From Mexico. Presented by Dr. C. A. Purpus, Zacuapam, Huatusco, Vera Cruz, Mexico. Received November 6, 1911.

Seeds of the following; quoted notes by Dr. Purpus:

32064 to 32067. SOLANUM NIGRUM L. **Nightshade.**

32064. "From Minas San Rafael, San Luis Potosi, 1,600 meters [5,250 feet]."

32065. "From Esperanza, Puebla, 2,700 meters [9,850 feet]."

32066. "From Tehuacan, Puebla, 1,700 meters [5,575 feet]."

32067. "From Rascon, San Luis Potosi, 400 to 500 meters [1,300 to 1,650 feet]."

32068 and 32069. NICOTIANA spp. **Wild tobacco.**

32068. "From Guascama, San Luis Potosi."

32069. "From Minas San Rafael, San Luis Potosi."

32070. CACARA EROSA (L.) Kuntze.

From Tampico, Mexico. Presented by Mr. Clarence A. Miller, American consul, through Mr. E. C. Green, in charge, South Texas Garden, Brownsville, Tex. Received December 18, 1911.

"*Hicama de Agua.*" See No. 27959 for previous introduction.

32071. CALLITRIS CUPRESSIFORMIS Ventenat.

Tasmanian cypress pine or Oyster Bay pine.

From Tasmania. Presented to the United States Forest Service by Mr. L. A. Evans, editor of the Agricultural Gazette, Hobart, Tasmania, and to the Office of Foreign Seed and Plant Introduction by Mr. Raphael Zon, Chief of Silvics, United States Forest Service. Received November 8, 1911.

"This pine is described by Col. W. V. Legge in a report on the 'Tasmanian cypress pine,' published this year [1911]. According to this paper, the tree is confined mainly to the coast, where it does well on poor soils. It seems to have a slow growth, but in time reaches a height of 100 feet and a diameter of about 2½ feet. In spite of the fact that it is chiefly a warm climate tree, it also thrives in some of the colder parts of Tasmania, where there is considerable frost. It has a plain whitish wood, without figure, and with little difference in color between the sapwood and the heartwood. Its grain is hard and close, and the wood is exceedingly durable. It is largely used for piles, telegraph poles, and in general construction work. It not infrequently grows in mixture with eucalypts and when grown in the forest under moderate light conditions its form is that of a sharp cone, which is tall both in proportion to the diameter and the spread of the lateral branches. There are all gradations from this form to the spreading bushy tree found in the open.

"Since Florida is apparently the region in the United States best adapted to this species, I would advise growing some at Miami for experimental planting by the Forest Service in the Florida National Forest. Although the tree is widely used for a great variety of purposes in Tasmania, I doubt if it would prove superior to our own conifers and believe that the chief advantage in introducing it into Florida would probably be to furnish a comparatively soft, light wood for local use." (*Zon.*)

32072. SYZYGIUM CUMINI (L.) Skeels. **Jambu.**

From Algiers, Algeria. Presented by Dr. L. Trabut. Received November 10, 1911.

"Originally from Madagascar. A very vigorous tree, bearing large leaves and large, sweet fruits." (*Trabut.*)

See No. 31571 for previous introduction.

32073. SECALE CEREALE L. **Rye.**

Purchased from Landwirtschaftsvereins, Insterburg, Germany. Brought in by Mr. E. Brown, Botanist in Charge, Seed Laboratory, Bureau of Plant Industry. Numbered November 11, 1911.

Variety *multicaule.* "*Johannisroggen, Johannistagroggen, Seigle de la Saint-Jean.* A variety commonly cultivated in northern Germany in mixture with *Vicia villosa* for green forage and hay. It is also a favorite grain variety in East and West Prussia. It can be seeded any time between June 15 and October 15, and when seeded early furnishes abundant green fodder or pasture in the fall and green fodder, hay, or grain in the spring. It is an especially hardy variety adapted to a wide variety of soils, and on account of its stooling habit less seed is required than for other varieties of rye. This should prove an especially valuable forage crop throughout the sections of the South where it is difficult to maintain a good turf for pasture or meadow on account of the lack of vegetable matter or because of an acid condition of the soil." (*Brown.*)

32074 to 32077.

From Alhajuela, Panama. Collected by Mr. August Busck. Presented by Mr. William R. Maxon, United States National Museum, Washington, D. C. Received November 10, 1911.

Plants of the following:

32074 to 32076. (Undetermined.) **Orchid.**

32077. HIERACIUM sp.?

32078. MEDICAGO FALCATA L.

From Tomsk, Siberia. Received through Mr. Frank N. Meyer, agricultural explorer, Bureau of Plant Industry, November 13, 1911.

"(No. 1636a, August 22, 1911.) A tall semierect form of *Sholteek* growing 4 to 5 feet in height, having much foliage and bearing large pods containing heavy seeds. Apparently shatters very little. Collected in the botanical garden of the University of Tomsk. To be tested for forage purposes and for hybridization exclusively." (*Meyer.*)

32079. CHRYSOPHYLLUM sp.

From Paraguay. Presented by Mr. C. F. Mead, Villa Encarnacion. Received December 19, 1911.

"In Guarany this is called *aguay.* A quick-growing tree reaching a height of 20 meters [65 feet]; it fruits in the fourth year. The bark is smooth, the wood white and very light. The fruit is something the size and shape of an olive, very astringent and not relished when fresh, but is very widely used hereabouts in preserves, for which purpose it is excellent." (*Mead.*)

32080 and 32081. Asparagus sublatus Steud. **Asparagus.**

From Jeolikote, United Provinces, India. Presented by Mr. Norman Gill, superintendent, Kumaon Government Gardens. Received November 13, 1911.

Seeds of the following; quoted notes by Mr. Gill:

32080. "Barb asparagus."

Distribution.—On the subtropical slopes of the Himalayas at an altitude of 2,000 to 4,000 feet, from Kumaon to Nepal in northern India.

32081. "Without barbs."

32082. Garcinia sp.

From China. Presented by Mr. George Campbell, Kiayingchow, on native boat en route to Swatow, China. Received November 14, 1911.

"This morning (October 6, 1911) my boat stopped at a market town and I strolled through it. I found a few specimens of a fruit called *Sann pee pah,* or wild loquat. I brought one back to the boat with me. It was the size of an unhulled walnut and looked like a yellow apple, save that it was spherical and marked into seven segments. It peeled like an apple.

"The flesh was about as thick as the hull of a walnut and very sour, but inclosed a core of seven lobes, each, with the exception of one, containing a seed. Each seed was inclosed in a sweet pulp very pleasant to the taste and suggesting the mangosteen to me." (*Campbell.*)

32083. Annona reticulata L. **Bullock's-heart.**

From Cairns, North Queensland, Australia. Presented by Prof. Charles E. Wood, manager, Kamerunga State Nursery. Received November 15, 1911.

Cuttings of this species introduced for the work of the Office of Foreign Seed and Plant Introduction in bringing together all the improved varieties of this genus for trial.

32084 to 32086. Ipomoea batatas (L.) Poir. **Sweet potato.**

From Port Moresby, Papua. Presented by Mr. A. C. English, Barodobo Plantation, Kapa Kapa, Port Moresby. Received November 15, 1911.

"Seeds of three varieties that we have here in this locality, which are great tuber producers. One has a white skin and white flesh, one white skin and deep yellow flesh, and one a pink skin and white flesh. Seed from sweet potatoes are rarely known here, even amongst the natives who plant them extensively." (*English.*)

32087. Castanospermum australe Cunn. and Fraser.
Moreton Bay chestnut.

From Brisbane, Queensland, Australia. Presented by Mr. Frederick Manson Bailey, Colonial Botanist, Department of Agriculture and Stock, Brisbane. Received November 16, 1911.

"In the scrub near Kuranda we noticed trees bearing pods about the size and shape of a banana, but at least twice the diameter. Upon opening the pods they were found to contain huge beans that look very much like chestnuts. They have a leathery skin, and the interior is white and not very hard, about the consistency of a nut. I tasted one of the beans, although I was told that it was poisonous. It tasted very much like a nut, but had no distinctive flavor. In spite of the poisonous nature of the bean, the 'black fellows' have learned to use it as a food. They first roast the beans in hot ashes, then skin them and pound the white flesh into coarse flour. They fill a basket with this flour and place it in running water all night. In this way the poisonous principle is washed out." (*Dr. Alexander Graham Bell.*)

Distribution.—A tall tree found in the valleys of the Endeavour and Brisbane rivers in the State of Queensland, and in the valley of the Clarence River in the province of New South Wales in Australia.

32088. CICER ARIETINUM L. **Chick-pea.**

From Nogales, Sonora, Mexico. Presented by Mr. Alexander V. Dye, American consul. Received November 11, 1911.

"From the Mayo River Valley. Known locally as *Garbanzo gordo.* While there is only one variety grown for commercial purposes, those raised in the Mayo River Valley are usually considered larger and better." (*Dye.*)

32089. MEDICAGO SATIVA VARIA (Mart.) Urb. **Sand lucern.**

From Isere, France. Presented by Dr. L. Trabut, Algiers, Algeria. Received November 18, 1911.

32090. LEUCADENDRON MELLIFERUM (Thunb.) W. F. Wight. See Botanical Notes, etc., p. 60.

From the slopes of Table Mountain, Cape Colony, South Africa. Presented by the Assistant Conservator of Forests, Western Conservancy, Cape Town, at the request of Prof. J. Burtt Davy, Pretoria, Transvaal. Received November 20, 1911.

See No. 28016 for previous introduction.

32091. ASPARAGUS sp. **Asparagus.**

From Geok Tepa, Aresch District, Elisabethpol Government, Trans-Caucasia, Russia. Presented by Mr. A. Schelkownikow, Chaldan station, Trans-Caucasia, Russia, at the request of Mr. Frank N. Meyer, agricultural explorer, Bureau of Plant Industry. Received November 21, 1911.

"Seeds of the large wild asparagus, which grows so well in this vicinity." (*Schelkownikow.*)

A large asparagus which so impressed Mr. Meyer that he made a special effort to have seeds secured for the work now being undertaken in breeding rust-resistant forms and also improved forms for the use of florists.

32093 to 32110.

From India. Collected by Mr. C. V. Piper, Bureau of Plant Industry. Received November 17, 1911.

The following material; quoted notes by Mr. Piper:

32093. ALOCASIA MACRORRHIZA (L.) Schott.

"(No. 104.)"

32094. PHASEOLUS VULGARIS L. **Bean.**

"(No. 97.) White beans purchased in the market of Calcutta, September 12, 1911."

32095. PHASEOLUS VULGARIS L. **Bean.**

"(No. 98.) White beans purchased in the market of Calcutta, September 12, 1911."

32093 to 32110—Continued.

32096. Cicer arietinum L. **Chick-pea.**

"(No. 69.) Bought in the market at Trichinopoli, August 29, 1911."

32097. Cucurbita pepo L. **Squash.**

"(No. 95.) Bought on the market at Trichinopoli, August 31, 1911. Large, dull, pale orange; subglobose; deeply ribbed with about ten ribs."

32098. Elaeocarpus serratus L.

"(No. 81.) An olivelike fruit bought in the market at Colombo, August 28, 1911."

Distribution.—A tree found from the tropical slopes of the Himalayas, where it grows as far up as 3,000 feet in the province of Sikkim, southeastward through India and the Malay Islands to Java.

32099. (Undetermined.)

"(No. 82, August 29, 1911.) A shrub, bearing large pink flowers. This grows 3 to 10 feet high and is very ornamental. It is abundant at Newara Eliya, Ceylon, altitude of 6,000 feet."

32100. (Undetermined.)

"(No. 83, August 27, 1911.) An irislike plant with loose panicles of blue flowers. Decidedly ornamental. Native at Newara Eliya, Ceylon."

32101. Rubus sp.

"(No. 84, August 27, 1911.) A shrub much like the common blackcap. Fruits in large clusters, red but tomentose. Flavor good. Flowers pink. Abundant at Newara Eliya, Ceylon. Altitude 6,000 feet."

32102. Berberis sp. **Barberry.**

"(No. 86, August 27, 1911.) A common species at Newara Eliya, Ceylon, altitude 6,000 feet. It closely resembles *Berberis vulgaris*, but the fruits are black."

32103. Rhodomyrtus tomentosa (Ait.) Wight.

"(No. 87, August 27, 1911.) A melastomaceous shrub with pink flowers and canescent leaves. Grows 3 to 6 feet high. Quite ornamental. Common at Newara Eliya, Ceylon, 6,000 feet altitude."

32104. Benincasa hispida (Thunb.) Cogniaux. **Wax gourd.**

The wax gourd has heretofore been listed in the inventories as *Benincasa cerifera*, a name published in 1818 by Savi (Bibliotheca Italiana, vol. 9, p. 158), who cited *Cucurbita cerifera* Fischer (Catalogue du Jardin des Plantes à Gorenki, 1812, p. 32), a name not accompanied by a description. The attention of the Office of Foreign Seed and Plant Introduction has recently been called to the publication in 1881 of the name *Benincasa hispida* Cogniaux (De Candolle, Monographiae Phanerogamarum, vol. 3, p. 513) based on *Cucurbita hispida* Thunberg (Flora Japonica, 1784, p. 322) which appears to be the correct name for this species.

"(No. 94.) Purchased in the Trichinopoli market, August 31, 1911. Large, cylindrical, 12 to 18 inches long by 6 to 8 inches in diameter. Very white and waxy fruit."

This vegetable, commonly used by the Chinese for vegetable soups and also candied as a sweetmeat wherever it grows, is recommended to amateurs who are experimenting with new vegetables. It has been grown successfully near Washington, D. C.

32093 to 32110—Continued.

32105. PHYLLANTHUS ACIDA (L.) Skeels.

"(No. 96.) A round, green, gooseberrylike fruit bought in the market at Colombo, Ceylon, August 28, 1911."

See No. 23472 for previous introduction.

32106. CITRUS sp. **Orange.**

"(No. 88.) Best orange at Colombo. Round, green, light skin, very juicy, subacid, much rag, many seeds."

32107. CITRUS DECUMANA (L.) Murray. **Pomelo.**

"(No. 99, September 4, 1911.) From Calcutta. Large globose pomelo of the dry type. Flesh yellow; good quality, but not as good as the Siam."

32108. BELOU MARMELOS (L.) Lyons. **Bael.**

"(No. 90, August 29, 1911.) From Colombo."

See No. 24450 for description.

32109. CALAMUS VIMINALIS Willd. **Rattan.**

"(No. 93, August 31, 1911.) From Trichinopoli. A peculiar fruit said to be used in curries. Flesh very acid. A rattan palm."

"It is a stout scrambling and climbing species, with cane thin but strong. It makes excellent walking sticks and is the chief rattan of the Malay Peninsula." (*Watt, Commercial Products of India.*)

Distribution.—A climbing palm found in the Malay Islands.

32110. SPONDIAS CYTHEREA Sonnerat. **We fruit.**

"(No. 91.) Bought in the market at Colombo, August 28, 1911. Fruit smooth, oval, size of a large hen's egg, each containing one stone. Flesh firm, yellowish, subacid, pleasant to taste, odor of pineapple."

32111 to 32135.

Collected by Mr. C. V. Piper, of this Department. Received November 20, 1911.

The following material; quoted notes by Mr. Piper:

32111. POLYTRIAS DIVERSIFLORA (Steud.) Nash.

"(No. 41.) Collected at Batavia, July 23, 1911. The common lawn and pasture grass of Java at low altitudes. Makes a good lawn. Horses as well as cattle eat it readily and seem to thrive on it. Introduced in the Philippines, where it is spreading."

Distribution.—Java, and introduced in other tropical regions.

32112. STIZOLOBIUM sp.

"(No. 52, July 27, 1911.) The native Java form cultivated in the botanic garden at Buitenzorg."

32113. PANICUM NUMIDIANUM Lam.

"(No. 55, August 3, 1911.) Collected at Karanganjar, Java. The same as Para grass. To be tested in comparison with the American form."

Distribution.—Northern Africa, India, and generally distributed in the Tropics.

32114. MEIBOMIA HETEROPHYLLA (Willd.) Kuntze.

"(No. 65, August 24, 1911.) From Peradeniya, Ceylon. Similar to but much larger than *Meibomia triflora*, now abundantly established in Florida. If this proves equally aggressive it will be a valuable pasture plant."

Distribution.—From the plains of India eastward through the Malay Archipelago, including the Philippines, and in China.

32111 to 32135—Continued.

32115. Arundinella sp.

"(No. 70, August 11, 1911.) From mountains near Garoet, Java. Makes a good sward and is probably valuable as pasturage. Seed habits good."

32116. Axonopus compressus (Swartz) Beauv.

"(No. 71, July 26, 1911.) Best lawn grass in Buitenzorg; will grow in dense shade."

Distribution.—The West Indies and southward to Brazil; introduced into the Tropics of the Old World.

32117. Syntherisma sanguinalis (L.) Dulac.

"(No. 72, July 25, 1911.) Collected in shady ground at Hotel des Indes, Batavia, Java. A lawn grass much like St. Augustine, but hardly as good."

32118. Syntherisma debilis (Desf.) Skeels.

(*Panicum debile* Desf., 1800, Flora Atlantica, vol. 1, p. 59.)

The seeds of this grass, received from Java as an unidentified species of Syntherisma, belong to *Panicum debile* Desfontaines. This species seems not to have been placed in the genus Syntherisma heretofore.

"(No. 73, July 26, 1911.) Growing in an orchard near Buitenzorg. Much smaller and more slender than our crab-grass, but perhaps of similar value. Makes a good turf."

Distribution.—Found on the plains and low hills of India and extends eastward through the Malay Archipelago as far as the Philippines.

32119. Panicum maximum hirsutissimum Nees.

"(No. 74, July 2, 1911.) Growing at Singalong Experiment Station, Manila, P. I. Smaller than Guinea grass. Decumbent at base, and roots at nodes."

32120. Syntherisma longiflora (Retz.) Skeels.

(*Paspalum longiflorum* Retz., 1786, Observationum Botanicarum, vol. 4, p. 15.)

The seeds of this grass, received from Ceylon, were identified as *Paspalum longiflorum* Retz., which seems not to have been heretofore placed in the genus Syntherisma.

"(No. 75, August 25, 1911.) From Peradeniya, Ceylon. A creeping species rooting at the joints, which makes a good but thin turf."

Distribution.—Found in India from Kashmir to Ceylon, and in Malakka; is generally distributed in the tropical and subtropical regions of the Eastern Hemisphere.

32121. Pterocarpus indicus Willd.

"(No. 42, August 15, 1911.) From Singapore. A beautiful shade tree, elm-like in form, but with drooping branches. Abundantly planted in the Malay Peninsula, but, according to Mr. Ridley, is not native. Said to differ from the true *Pterocarpus indicus* in having larger pods."

Distribution.—A tall tree found throughout India and eastward through China and the Malay Archipelago as far north as the Philippines.

32122. Salakka edulis Reinw.

"(No. 44, July 24, 1911.) From Batavia, Java. A palm cultivated in Java. The fruit consists of three carpels enclosed in a scaly envelope. Each carpel is fleshy, with a large central seed. The flesh is firm and much like quince in flavor. Abundant in the Java markets in July. Malay name *Salak*."

Distribution.—Known only from the islands of the Malay Archipelago.

32111 to 32135—Continued.

32123. Sesban sp.

"(No. 45, August 9, 1911.) A large shrub or small tree grown on the dikes surrounding the rice paddies near Surabaya, Java. *Agati grandiflora* and *Cajan indicum* are also grown in the same way."

32124. Myristica sp.

"(No. 46, July 26, 1911.) Purchased in the market at Batavia, Java. A seed used by the Javanese as a cheap substitute for the nutmeg."

32125. Ptychoraphis augusta (Kurz) Beccari.

"(No. 51, July 20, 1911.) An ornamental palm from Singapore."

Distribution.—Found in the Nicobar Islands in the Indian Ocean.

32126. Dialium indum L.

"(No. 53, July 20, 1911.) Fruit like a tamarind in structure and flavor. Purchased in the market at Singapore."

Distribution.—A tree with alternate pinnate leaves found in the island of Java.

32127. Passiflora edulis Sims. **Passion fruit.**

"(No. 57, July 19, 1911.) Bought in the market at Singapore. Fruit yellow, ovoid, 2 inches long. Pulp subacid, seedy."

32128. Impatiens sp.

"(No. 58, August 24, 1911.) From Peradeniya, Ceylon. A species with rose-colored flowers."

32129. Impatiens sp.

"(No. 59, August 1, 1911.) Collected on high mountains near Garoet, Java. Flowers rose colored, perhaps two species mixed."

32130. Rubus sp. **Raspberry.**

"(No. 63, August 1, 1911.) Collected in the mountains near Garoet, Java. Vines very large, ascending. Leaves grapelike. Flowers and fruits in large panicles. Fruit a raspberry, salmon yellow, subacid, of fair quality."

32131. Rubus sp. **Raspberry.**

"(No. 64, August 1, 1911.) Collected in the mountains near Garoet, Java. Leaves digitate, 5 to 7 leaflets. Fruit a raspberry red, with large pyrene, subacid, juicy."

32132. Citrus sp. **Orange.**

"(No. 66, July 24, 1911.) From Batavia, Java. Fruit dark green, 2½ inches in diameter. Stem rather loose. Flesh yellow, subacid, juicy, but much rag and many seeds."

32133. Citrus sp. **Orange.**

"(No. 67, July 20, 1911.) From Singapore. A loose-skinned, yellow, very seedy, sour kid-glove orange."

32134. Triphasia trifoliata (L.) DC.

"(No. 68, July 23, 1911.) A common hedge plant at Batavia, Java. Fruit red, pyriform, about 1 inch long."

32135. Elettaria speciosa Blume.

"(No. 69, August 2, 1911.) From Garoet, Java. A fruit commonly seen in Javanese markets."

Distribution.—A herbaceous perennial found in damp woods on the island of Java.

32136. MEDICAGO SATIVA L. **Alfalfa.**

From Tulare, Cal. Presented by Mr. J. T. Bearss, Agricultural Experiment Station, Kearney Park, Fresno Co., Cal. Received November 23, 1911.

"One of the surviving plants from the plat of Arabian alfalfa (No. 8823?), but presumably a hybrid between this and the Turkestan (No. 1151) on an adjoining plat." (*Bearss.*)

32137 and 32138. PASSIFLORA INCARNATA L. **Maypop.**

From Salem, Mass. Purchased from Mr. Harlan P. Kelsey, Higginson Square. Received November 22 and 24, 1911.

Seeds and plants procured for breeding purposes and for the collection being made by the Office of Foreign Seed and Plant Introduction of all the species of this genus having edible fruits for use in hybridization work.

32141 and 32142. PHOENIX DACTYLIFERA L. **Date.**

From Cairo, Egypt. Received through Mr. George J. Salem, November 22, 1911.

Seeds of the following:

32141. *Zaglool.* **32142.** *Samany.*

"These are supposed to be the best in the market and are grown extensively in the delta, particularly in the gardens of Alexandria, Rosetta, Edku, and Cairo. They are used for the table, confectionery, and exportation." (*Salem.*)

32143. CUCUMIS MELO L. **Muskmelon.**

From Beni Suef, Egypt. Received through Mr. George J. Salem, Cairo, November 22, 1911.

"*Domiri.* This melon is somewhat larger and sweeter than our cantaloupes, and could keep for several days. It is grown in the province of Beni Suef and sold in the markets of the large cities for only a few days." (*Salem.*)

32144 to 32150. SOLANUM NIGRUM L. **Nightshade.**

Presented by Mr. K. Portele, agricultural ministry, Vienna, Austria. Received November 23, 1911.

Seeds of the following:

32144 to 32149. From Austria.

32144. From Krakow.

32145. From Linz, Upper Austria.

32146. From Halterthal, near Vienna.

32147. From Trieste.

32148. Cultivated in the botanic garden at Vienna. Origin unknown.

32149. From Pratu, near Vienna.

32150. From Breslau, Germany.

32151 and 32152. FEIJOA SELLOWIANA Berg.

From Los Angeles, Cal. Presented by Mr. H. Hehre. Received November 15, 1911.

Seeds of the following:

32151. "The fruits of this variety weighed nearly 1½ ounces each. The quality is good when the fruit is properly ripened. The flesh is somewhat granular." (*R. A. Young.*)

32152. "The fruits of this variety weighed nearly 2 ounces each. The quality is very good when the fruit is properly ripened, the flavor being mild and the flesh less granular than in some forms." (*R. A. Young.*)

32153 and 32154. Convolvulus spp. **Rose-root.**

From Puerto Orotava, Teneriffe, Canary Islands. Presented by Dr. George V. Perez. Received November 23, 1911.

Seeds of the following:

32153. Convolvulus floridus L. f. **Rose-root.**

"One of the most interesting and striking plants in the Canary Islands is this shrubby Convolvulus, known locally as *guadil*. This, together with a closely allied species, *C. scoparius,* is sometimes known as rose-root and is said to yield oil of roses. The *guadil* is, however, more interesting as an ornamental flowering shrub than for any economic purposes to which at one time it may have been applied. It belongs to the bindweed order (Convolvulaceæ), although nothing can be less like a Convolvulus in habit, and is one of the comparatively few examples of this order (containing the jalap plant, the sweet potato, the scammony, and the showy ipomoeas), possessing an upright stem and a treelike habit. It may be described as a compact shrub from 4 to 6 feet in height, with a stout woody stem and numerous branches. The branches, especially in the young state, are white powdered, as also are the abundant linear undulate leaves. The latter are about 3 or 4 inches long, attenuated toward the petiole, with rounded and somewhat emarginate tips. The flowers are abundantly produced in large loose terminal panicles. The sepals are ovate acuminate, about 2 lines long. The corolla is about one-half to three-quarters of an inch in diameter and pure white. The capsule is conical and slightly villose at the apex. When in flower the plant appears as if covered with newly fallen snow. It is one of the few native plants which awakens the enthusiasm of local residents of Teneriffe. According to Dr. Perez the *guadil* was an object of high regard by the Guanches, the aboriginal race of the island. This can well be understood, for when in flower it is one of the brightest and most attractive objects in the Teneriffe landscape. The pale powdered green of the leaves forms an excellent background for the masses of pure white flowers. The only pity is it is so rarely seen. In the search for rose-roots for export purposes this and the allied species have been nearly exterminated in the wild state. In Teneriffe it grows from sea level up to elevations of about 1,000 feet. It evidently prefers a free porous soil and thrives in exposed situations on rocks and slopes. In rich soil the plant appears to put on a looser habit and when laden with flowers the panicles become almost pendulous. It is readily increased by seed, and as it is a fast grower it should flower in the second or third year." (*Extract from D. Morris, Kew Bulletin of Miscellaneous Information, 1893, pp. 133–134.*)

32154. Convolvulus scoparius L. f. **Rose-root.**

"This, as its name denotes, is a Convolvulus with the habit of the common broom. It is an erect shrub 7 to 8 feet high with a few long narrow branches and somewhat short linear leaves. The flowers are produced in many flowered axillary cymes. The corolla is slightly plaited with a 5-partite limb; it is tinged with red outside and white within. This is known locally as *Leña Noel* or *Leña Loel.* It is only rarely met with; Teneriffe specimens in the Kew Herbarium are from Guia on the southwestern slopes of the island and from Barranco Santo. It is said to be more common at Palma, but owing to the excessive digging of the roots many years ago for export purposes it is restricted to very few localities.

"There is a single specimen of the root of *Convolvulus scoparius* in the Kew Museum. It is about 8 inches long, 2½ inches wide, and 1 inch thick. The heartwood occupies about three-fourths of the entire diameter of the specimen and is of a distinct orange-yellow color, very dense, marked by closely lying

32153 and 32154—Continued.

32154—Continued.

annual rings and numerous very fine medullary rays, the spaces between them being filled with small, partially open vessels. The sapwood is very much lighter in color and is comparatively soft and easily cut. It was sent to the museum about 1850 and still retains its roselike smell. A sample of oil in the museum is of clear amber color and possesses a distinct though not a penetrating odor of roses.

"In Lindley's Flora Medica (1838), p. 400, there is the following note on this plant:

"Wood perfumed, smelling strongly of roses, yellowish fawn color veined with red, burning readily when lighted. Taste bitter, balsamic. Yields by distillation an essential oil of bitter balsamic flavor; little used, except, according to Feé, for adulterating oil of roses."

"The latest information on the subject is probably contained in Piesse's The Art of Perfumery (1879), p. 188, as follows:

"When rosewood, the lignum of the *Convolvulus scoparius*, is distilled, a sweet-smelling oil is procured, resembling in some slight degree the fragrance of the rose, and hence its name. At one time, that is, prior to the cultivation of the rose-leaf geranium, the distillates from rosewood and from the root of the *Genista canariensis* (Canary rosewood) were principally drawn upon for the adulteration of real otto of roses; but as the geranium oil answers so much better the oil of rhodium has fallen into disuse, hence its comparative scarcity in the market at the present day, though our grandfathers knew it well. One hundredweight of wood yields about 3 ounces of oil.

"Ground rosewood is valuable as a basis in the manufacture of sachet powders for perfuming the wardrobe." (*Extract from D. Morris, Kew Bulletin of Miscellaneous Information, 1893, pp. 134–136.*)

32155 to 32157.

From southeastern Russia. Received through Mr. Frank N. Meyer, agricultural explorer, Bureau of Plant Industry, November 25, 1911.

Seeds of the following:

32155. HELIANTHUS ANNUUS L. **Sunflower.**

From Bezenshook (Besentschuk), Government of Samara, southeastern Russia.

"(No. 1704a, November 8, 1911.) A variety of sunflower called *Pantsernara*, meaning armor-plated; originated by Mr. Karsin in Russia. The seeds of this remarkable variety are provided with an extremely hard shell, being thickened with silica, and the insects that have been playing such havoc heretofore with the sunflower-seed crop in Russia find it beyond their power to penetrate the hulls of this variety. Obtained from Prof. N. M. Tulaikoff, director of the agricultural experiment station at Bezenshook." (*Meyer.*)

32156. TRITICUM DURUM Desf. **Wheat.**

From Bezenshook (Besentschuk), Government of Samara, southeastern Russia.

"(No. 1705a, November 8, 1911.) A large-grained, uncommon, summer variety of durum wheat, called *Amerikanka*. Grown here and there in southeastern Russia, having apparently originated in these regions. Obtained like the preceding number." (*Meyer.*)

32157. TRITICUM DURUM Desf. **Wheat.**

From Bezenshook (Besentschuk), Government of Samara, southeastern Russia.

"(No. 1706a, November 8, 1911.) A new and valuable winter variety of black-bearded durum wheat, having very long open ears. It is called *Teiskaia*

32155 to 32157—Continued.

32157—Continued.

and originated at the Bezenshook Agricultural Experiment Station. This variety is proving extremely hardy, having survived snowless winters, when other winter wheats were either killed out entirely or severely injured. It is expected that within a few years this wheat will play a big rôle in the agricultural development of the Government of Samara. Obtained like the preceding numbers." (*Meyer.*)

32158 and 32159. CARICA PAPAYA L. **Papaya.**

From Miami, Fla. Grown at the Subtropical Plant Introduction Garden. Received November 25, 1911.

Seeds of the following:

32158. "Grown from No. 28536. Fruit medium size, globular, from 5 to 6 inches in diameter, with very tender meat of excellent flavor and pale yellow or orange colored. The parent tree of this variety produced pear-shaped fruits of large size, demonstrating the great variation of papayas in the shape and size of fruit when raised from seed." (*H. F. Schultz.*)

32159. "Grown from No. 28534. Fruit pyriform, of large size, 10¼ inches long and 5½ inches in diameter, weighing 10 pounds. The meat is rich yellow and of excellent flavor, nearly 2 inches thick. Seeds quite numerous, but easily separated from the meat. The tree matured its fruit within 16 months from seed." (*H. F. Schultz.*)

32160 to 32162. COFFEA spp. **Coffee.**

From Mayaguez, Porto Rico. Presented by Mr. D. W. May, director, Agricultural Experiment Station. Received November 29, 1911.

Seeds of the following:

32160. COFFEA sp.

"Ceylon hybrid."

32161. COFFEA sp.

"*Maragogipe.* Java variety."

32162. COFFEA ARABICA L.

"Native variety."

32163. ANNONA DIVERSIFOLIA Safford. **Ilama, or anona blanca.**

From Acapulco, Mexico. Presented by Mr. Marion Letcher, American consul, through Mr. W. E. Safford, Bureau of Plant Industry. Received December 1, 1911.

Seeds.

32164. COLOCASIA sp. **Dasheen.**

From Kiayingchow, China. Presented by Mr. George Campbell. Received December 1, 1911.

"*Penang.* The tubers of this variety are quite uniform in size and shape, and are about the size of a goose egg. The people here think them far superior to the kinds I sent you previously (Nos. 27297 and 27298)." (*Campbell.*)

"The tubers resemble the Japanese dasheens in appearance and are very slightly acrid when raw. The cooked tubers are mealy, grayish white in color, and the flavor is good, though a little strong and suggestive of the taro." (*R. A. Young.*)

32167. MENTHA PIPERITA L. **Peppermint.**

From Kobe, Japan. Presented by Mr. George N. West, American consul. Received December 2, 1911.

Roots.

32168. Prunus subcordata Benth.

From Lassen County, Cal., west of Honey Lake, at an altitude of about 4,700 feet. Collected by Mr. Karl Kair; presented by Mr. Marsden Manson, San Francisco. Received September 7, 1909. Numbered for convenience in recording distribution December 4, 1911.

Variety *kelloggii*. "A small tree, native of dry rocky hills of northern California, with thick leaves and white flowers changing to rose. The dark-red, clingstone fruits contain a subacid flesh and are used for drying and preserving. For testing and breeding purposes in dry regions of the United States."

Plants of this variety were formerly distributed under No. 25933.

32169. Aralia californica S. Watson. **California spikenard.**

From Mill Valley, Marin Co., Cal., Presented by Mr. Charles G. Adams, at the request of Mr. G. P. Rixford, Bureau of Plant Industry. Collected November 15, 1911. Numbered December 6, 1911.

"In moist, cool ravines where the sun only slants athwart the branches and a certain dankness always lingers the California spikenard scents the air with its peculiar odor, It closely resembles *Aralia racemosa* of the Eastern States, but it is a larger, coarser plant in every way. It throws up its tall stems with a fine confidence that there will be ample space for its large leaves to spread themselves uncrowded. Its feathery panicles of white flowers are followed by clusters of small purple berries and are rather more delicate than we should expect from so large a plant." (*M. E. Parsons, The Wild Flowers of California, pp. 77–78.*)

"Introduced for the breeding experiments with the Japanese udo (*Aralia cordata*) and the American spikenard (*Aralia racemosa*) in an effort to improve the character of the Japanese vegetable." (*Fairchild.*)

32170 to 32172.

From San Jose, Costa Rica. Presented by Mr. Carlos Wercklé. Received December 4, 1911.

The following material; quoted notes by Mr. Wercklé:

32170. Dioscorea sp.

"*Papa caribe.*"

32171. (Undetermined.)

"A very large and good green sapote."

32172. Persea pittieri Mez. **Avocado.**

"A very early variety, but rather poor. Good quality, but little flesh."

32173. Aralia cordata Thunb. **Udo.**

From Chevy Chase, Md. Grown by Mr. David Fairchild, Bureau of Plant Industry, on his place, "In the Woods."

Collected November 5, 1911.

"Seeds collected from plants 2, 3, and 4 years old." (*Fairchild.*)

32174. Thea sp. **Tea.**

Presented by Mr. J. R. C. Boyer, Cranford, N. J. Received through Mr. E. C. Green, Pomologist in Charge, South Texas Plant Introduction Garden, Brownsville, Tex., December 2, 1911.

"Chinese tea seed, not of the regular tea plant, but, as I understand it, of the Camellia family. The seeds are crushed for the oil, which is used as an edible oil and for many other purposes." (*Boyer.*)

32175 to 32245.

From Siberia. Collected by Mr. Frank N. Meyer, agricultural explorer, Bureau of Plant Industry. Received November 29, 1911.

Seeds of the following:

32175. Triticum polonicum L. **Wheat.**

From Omsk, Siberia.

"(No. 1631a, August 9, 1911.) A very large hard wheat, obtained at the agricultural exhibition held in Omsk during July and August, 1911. Said to have been grown at Atbasar, Akmolinsk Government. Called *Afrikanski*, or *Africanum congum*. To be tested in the semiarid Northwest." (*Meyer.*)

32176. Triticum polonicum L. **Wheat.**

From Omsk, Siberia.

"(No. 1632a, August 9, 1911.) A very large hard wheat, obtained like the preceding number. Locality from whence it came not given." (*Meyer.*)

32177. Triticum durum Desf. **Wheat.**

From Omsk, Siberia.

"(No. 1633a, August 9, 1911.) A hard summer wheat having blue ears; called *Sineshka*. Said to be very drought resistant and to stand the Siberian climate well, although constant selection has to be practiced to prevent it from deteriorating. Obtained like the preceding numbers." (*Meyer.*)

32178. Medicago falcata L.

From near Ust Kamenogorsk, southwestern Siberia.

"(No. 1635a, October 4, 1911.) A bunchy, upright form of sholteek growing about 2 to 3 feet in height. Pods rather broad and flat, apparently nonshattering. Only two plants found of this variety. These were growing in decomposed rock on a mountain slope facing the Irtish River. To be tested exclusively for forage purposes and hybridization." (*Meyer.*)

32179. Medicago falcata L.

From near Tomsk, Siberia.

"(No. 1637a, August 24 and 25, 1911.) A sholteek of prostrate habit and very vigorous growth, shoots being over 6 feet long. Pods very large, apparently nonshattering. Found on sandy level stretches of land along the River Tom, 20 to 30 feet above the water. To be tested in meadows where grass is grown for hay production." (*Meyer.*)

32180. Medicago falcata L.

From Barnaul, Siberia.

"(No. 1638a, September 1, 1911.) A sholteek collected in the Kuznetsk district to the east of Barnaul, said to be of vigorous growth. Presented by Mr. N. B. Sokoloff, agricultural instructor at Barnaul. To be tested like No. 1634a (S. P. I. No. 32389)." (*Meyer.*)

32181. Medicago platycarpa (L.) Trautv.

From near Sminogorsk, southwestern Siberia.

"(No. 1639a, September 21, 1911.) A strange wild alfalfa having yellow flowers and large flat pods of black color when ripe. Seeds very large. Prefers to grow between shrubbery and tall grass on the north slopes of hills. Of erect growth, but needs some support. Apparently not of as great value as a fodder plant as other members of the same genus. To be tested in cool, moist-air sections of the United States." (*Meyer.*)

32175 to 32245—Continued.

32182. Onobrychis vulgaris Hill 1756.

(*Onobrychis viciaefolia* Scop. 1772.)

From between Chistunka and Sminogorsk, southwestern Siberia.

"(No. 1640a, September 8 to 24, 1911.) The wild Siberian esparcet found along the edges of wheat fields and on stretches of level land that was once in cultivation. Apparently able to stand an unusual amount of cold and drought. Recommended as a forage plant in those sections of the semiarid belt, where the ordinary alfalfa gets winterkilled." (*Meyer.*)

32183. Onobrychis vulgaris Hill.

From Omsk, Siberia.

"(No. 1641a, August 9, 1911.) A large-seeded esparcet being tested at the agricultural experiment station near Omsk and promising to become an important forage plant for semiarid western Siberia." (*Meyer.*)

32184. Astragalus sp.

From near Chistunka, southwestern Siberia.

"(No. 1642a, September 9, 1911.) A species of Astragalus bearing a close resemblance to esparcet, found on abandoned wheat fields. Of bunchy erect habits, making many stems, well supplied with almost glabrous foliage. Flowers purplish blue, seed racemes persistent. Eaten by horses and cattle. Of value possibly along with sholteek and esparcet as a forage plant for cool semiarid climates." (*Meyer.*)

32185. Astragalus sp.

From near Chistunka, southwestern Siberia.

"(No. 1643a, September 8, 1911.) A species of Astragalus of somewhat open growth. Main stems being erect, side stems lying more or less on the ground. Foliage not very dense and quite hairy, flowers yellow, seed racemes persistent. Occurring on abandoned wheat fields and along ditches. To be tested like the preceding number, but it is not so promising." (*Meyer.*)

32186. Astragalus sp.

From near Sminogorsk, southwestern Siberia.

"(No. 1644a, September 24, 1911.) A small serradellalike species of Astragalus, occurring on sandy pasture grounds; is browsed by cattle. Of value possibly as a pasture plant on sandy lands in semiarid regions." (*Meyer.*)

32187. Hedysarum sibiricum Poir.

From Tomsk, Siberia.

"(No. 1645a, August 22, 1911.) An upright-growing variety that throws up many stems and is well supplied with rather large glabrous foliage. May possess value as a forage plant for the northern sections of the United States and may also serve as a factor in hybridization experiments to be made with the famous sulla (*Hedysarum coronarium*) in making it hardier and available to regions outside the Mediterranean." (*Meyer.*)

Distribution.—Central Siberia, extending eastward to the region of Lake Baikal.

32188. Hedysarum sp.

From near Sminogorsk, southwestern Siberia.

"(No. 1646a, September 21 to 24, 1911.) A rare legume found here and there in southern Siberia. Makes a luxuriant growth of leaves when planted in rich soil. Flowers of a purple-violet color and attractive to the eye. Of value like the preceding number, and perhaps of even more importance. Should be tested with great care." (*Meyer.*)

32175 to 32245—Continued.

32189. HEDYSARUM SPLENDENS Fisch.

From near Ust Kamenogorsk, southwestern Siberia.

"(No. 1647a, October 2 and 4, 1911.) A very rare Hedysarum occurring on rocky mountain slopes facing south and southeast. Of value possibly as a forage and pasture plant on dry and rocky places. Roots sent under No. 989 (S. P. I. No. 32026); see the latter number for further remarks." (*Meyer.*)

Distribution.—The Altai region of Siberia.

32190. LATHYRUS GMELINI (Fisch.) Fritsch. (?)

From Tomsk, Siberia.

"(No. 1648a, August 20, 1911.) Variety *orientalis.* A very vigorous-growing legume found on moist hill slopes here and there in central and southern Siberia and in the Ural Mountains. It is a fodder of great value and eagerly sought by horses and cattle, who devour every bit of it. In protected localities, i. e., between shrubbery, the plants attain a height of 8 feet, but generally they are 3 to 4 feet high. Of value as a very promising forage plant for the cooler sections of the United States." (*Meyer.*)

32191. VICIA UNIJUGA A. Braun.

From near Tomsk, Siberia.

"(No. 1649a, August 25, 1911.) An erect-growing plant which attains its greatest luxuriance on fields from which light forest has recently been cut. Seems to possess value as a forage plant on wooded pasture grounds for the cooler sections of the United States." (*Meyer.*)

32192. LATHYRUS PISIFORMIS L.

From near Tomsk, Siberia.

"(No. 1650a, August 24, 1911.) A species of wild pea found mostly at the edges of clumps of shrubbery. Grows from 2 to 5 feet tall. Of erect growth, but when tall it needs some support. Eagerly eaten by horses and cattle. Apparently valuable for forage purposes in the cooler sections of the United States." (*Meyer.*)

32193. LATHYRUS PRATENSIS L.

From Tomsk, Siberia.

"(No. 1651a, August 18 to 20, 1911.) The well-known meadow pea, which is grown here and there in northwestern Europe for forage purposes. The Siberian form may prove to be extremely hardy and deserves therefore to be tested for forage purposes in the cooler sections of the United States. This plant thrives apparently best when allowed to overrun small scrub in slightly shaded places." (*Meyer.*)

32194. LENTILLA LENS (L.) W. F. Wight. **Lentil.**

The lentil has heretofore been listed in these inventories as *Lens esculenta*, which was published in 1794 by Moench (Methodus, p. 131). The first binomial name given to the lentil was *Ervum lens*, which was published in 1753 by Linnæus (Species Plantarum, vol. 2, p. 738). As the type species of the genus Ervum, *E. ervilia*, is not congeneric with the lentil but belongs to the genus Vicia, which was published on page 734 of Species Plantarum, the generic name Ervum can not be used for the lentil. The generic name Lens was first published in 1754 by Stickman (Herbarium Amboinense, reprinted in Linnæus's Amoenitates Academicae, 1759, vol. 4, p. 128) and was based on the plant described and figured in 1747 by Burmann (Herbarium Amboinense, vol. 5, p. 5, pl. 4) which

32175 to 32245—Continued.

32194—Continued.

is generally called *Entada scandens*. This use of the generic name Lens by Stickman prevents the use of the same name for the lentil. As no other generic name had been published for the lentil up to 1909, Mr. W. F. Wight (Century Dictionary Supplement, p. 719) published the name Lentilla by referring to "lens" and "lentil," under which names on pages 3409 and 3410 of volume 4 of the 1895 edition the lentil is described as *Lens esculenta*. When the new edition of 1911 appeared, in which the information first published in the two supplementary volumes 11 and 12 of 1909 was incorporated with the main body of the work, the name *Lentilla lens* was published (vol. 5, p. 3410) under the heading "lentil."

From Barnaul, Siberia.

"(No. 1652a, August 31, 1911.) A small-seeded variety of lentil that is grown by some progressive farmers in southern Siberia as a fodder for horses and milch cows. Prefers a light, sandy soil. Of value possibly for forage purposes in the cooler sections of the semiarid belt." (*Meyer.*)

32195. VICIA SATIVA L. **Spring vetch.**

From Barnaul, Siberia.

"(No. 1653a, August 31, 1911.) A vetch that is grown here and there in Siberia for forage purposes. It is said to stand drought very well and promises to be a good fodder plant in semiarid southwestern Siberia." (*Meyer.*)

32196. VICIA CRACCA L. **Vetch.**

From Tomsk, Siberia.

"(No. 1654a, August 18 to 20, 1911.) A wild vetch found on slightly shaded places. Likes to overrun low scrub. Is eagerly eaten by horses and cattle. Of value in pastures of the northern United States where some scrub is scattered over the land." (*Meyer.*)

32197. VICIA AMOENA Fisch. **Vetch.**

From near Chistunka, southwestern Siberia.

"(No. 1655a, September 8 and 9, 1911.) A sturdy vetch with rather dark green foliage found on dry earth banks. Does not grow over 1 foot in height, but on favorable locations spreads out considerably. Of value possibly as a forage or pasture plant in the northern sections of the United States." (*Meyer.*)

32198. VICIA AMOENA Fisch. **Vetch.**

From near Barnaul, Siberia.

"(No. 1656a, September 1 to 3, 1911.) A wild vetch, occurring along the edges of abandoned fields. Apparently a variety of the preceding number and as such the same remarks apply to it." (*Meyer.*)

32199. VICIA MEGALOTROPIS Ledeb. **Vetch.**

From near Sminogorsk, southwestern Siberia.

"(No. 1657a, September 21 to 24, 1911.) An exceedingly vigorous species of vetch, often 8 to 10 feet in height when running up into bushes; in open places, however, it reaches only a couple of feet in height. Much relished by horses and cattle and apparently of great promise as a forage plant. To be tested in the cooler sections of the United States." (*Meyer.*)

Distribution.—Slopes of the mountains in the Altai region of southwestern Siberia.

32175 to 32245—Continued.

32200. VICIA CRACCA L. **Vetch.**

From near Chistunka, southwestern Siberia.

"(No. 1658a, September 8, 1911.) A dense-growing variety of climbing vetch found between Artemisia growth on a dry plain. Of value for forage purposes in the semiarid belt of the United States." (*Meyer.*)

32201. VICIA CRACCA L. **Vetch.**

From near Sminogorsk, southwestern Siberia.

"(No. 1659a, September 21, 1911.) Variety *hircina.* A variety of wild climbing vetch, occurring mostly between low scrub, foliage slightly hairy. Of value like No. 1654a (S. P. I. No. 32196)." (*Meyer.*)

32202. VICIA CRACCA L. **Vetch.**

From near Sminogorsk, southwestern Siberia.

"(No. 1660a, September 21, 1911.) A variety of vetch of more upright and vigorous growth than the ordinary *Vicia cracca;* apparently a hybrid. Found between scrub on the north slope of a hill. Valuable possibly for forage purposes in the northern United States." (*Meyer.*)

32203. VICIA sp. **Vetch.**

From near Sminogorsk, southwestern Siberia.

"(No. 1661a, September 21, 1911.) A vetch of vigorous yet graceful growth, having deep-green, finely pinnated foliage and bearing many seed pods on long stems. Found between scrub on the north slope of a hill. Valuable apparently for forage purposes." (*Meyer.*)

32204. VICIA SEPIUM L. **Vetch.**

From near Tomsk, Siberia.

"(No. 1662a, August 24, 1911.) A rather small-growing vetch occurring on open sandy plains. Of value possibly as a forage plant on sandy lands in cool semiarid regions." (*Meyer.*)

Distribution.—Throughout Europe and eastward to the region of Lake Baikal in Siberia.

32205. VICIA AMOENA Fisch. **Vetch.**

From near Chistunka, southwestern Siberia.

"(No. 1663a, September 9, 1911.) A rather large vetch found along a ditch on a dry plain. Apparently of value for forage purposes in semiarid regions." (*Meyer.*)

32206. VICIA sp. **Vetch.**

From near Sminogorsk, southwestern Siberia.

"(No. 1664a, September 21, 1911.) A vetch growing in dense shade between shrubs on the north slope of a hill. Has an abundance of juicy light-green foliage that seems to make this plant desirable for forage purposes in cool shady locations." (*Meyer.*)

32207. TRIFOLIUM LUPINASTER L. **Clover**

From western Siberia.

"(No. 1665a, August 24 to September 21, 1911.) A wild clover not growing to large size and but scantily furnished with foliage. Withstands extreme droughts and is able to grow on very sterile soils. Of value as an element in pasture grounds in cold semiarid regions." (*Meyer.*)

32175 to 32245—Continued.

32208. Trifolium medium Huds. **Clover.**

From Tomsk, Siberia.

"(No. 1666a, August 22, 1911.) A clover resembling the ordinary red clover very much, but the leaves are almost glabrous and the whole plant of a denser, more even growth. Apparently able to stand considerable drought. Of value possibly as a forage plant in semiarid regions." (*Meyer.*)

32209. Trifolium hybridum L. **Alsike clover.**

From near Sminogorsk, southwestern Siberia.

"(No. 1667a, September 18 to 20, 1911.) A clover found in moist places and along low ditches in peaty and clayey soil. Apparently of value on pasture lands that are subject to inundation at times. Seems to make its heaviest growth toward the end of summer, which indicates that it will probably thrive better in cool moist-air localities than in warm and dry regions." (*Meyer.*)

32210. Trifolium pratense L. **Red clover.**

From near Sminogorsk, southwestern Siberia.

"(No. 1668a, September 18, 1911.) A rather vigorous form of red clover found in a ditch running through stony land. Possibly somewhat hardier than the ordinary clover." (*Meyer.*)

32211. Trifolium incarnatum L. **Crimson clover.**

From Omsk, Siberia.

"(No. 1669a, July 22, 1911.) A clover obtained by my interpreter from the agricultural exhibition held in Omsk during the summer of 1911." (*Meyer.*)

32212. Trifolium hybridum L. **Alsike clover.**

From Omsk, Siberia.

"(No. 1670a, July 22, 1911.) A clover that passes under the name *black clover*. Obtained like the preceding number." (*Meyer.*)

32213. Trifolium repens L. **White clover.**

From Omsk, Siberia.

"(No. 1671a, July 22, 1911.) A clover that passes under the name *yellow clover*. Obtained like the preceding numbers. (*Meyer.*)

32214. Trifolium pratense L. **Red clover.**

From Omsk, Siberia.

"(No. 1672a, July 22, 1911.) A sample of clover obtained like preceding numbers." (*Meyer.*)

32215. Trifolium repens L. **White clover.**

From Tomsk, Siberia.

"(No. 1673a, August 19, 1911.) A white clover collected in the vicinity of Tomsk, Siberia." (*Meyer.*)

32216. Festuca elatior L. **Meadow fescue.**

From near Widrichta, southwestern Siberia.

"(No. 1674a, September 30, 1911.) A grass of tall growth, with many spikes; found on earth banks. Of value possibly as a fodder plant in the colder sections of the United States." (*Meyer.*)

32175 to 32245—Continued.

32217. Agrostis alba L. **Creeping bent-grass.**

From near Widrichta, southwestern Siberia.

"(No. 1675a, September 30, 1911.) A grass of tall, airy growth, found at the foot of an earth bank. Possibly of value like the preceding number." (*Meyer.*)

32218. Dactylis glomerata L. **Orchard grass.**

From near Widrichta, southwestern Siberia.

"(No. 1676a, September 30, 1911.) A coarse grass found in an earth bank. Of value like the preceding number." (*Meyer.*)

32219. Festuca sp.

From Omsk, Siberia.

"(No. 1677a, July 18, 1911.) Obtained by my interpreter at Omsk, said to come from western Akmolinsk, and not to send up new shoots after having once been plowed under. Possibly of value for forage purposes on dry plains." (*Meyer.*)

32220. Phleum pratense L. **Timothy.**

From Omsk, Siberia.

"(No. 1678a, July 18, 1911.) A strain of timothy coming from Kungur, Perm Government, said to be superior to the ordinary kind. Obtained at Omsk by my interpreter. Of value like the preceding number." (*Meyer.*)

32221. Bromus inermis Leyss. **Brome-grass.**

From Omsk, Siberia.

"(No. 1679a, July 18, 1911.) A coarse grass said to be a good fodder and able to stand much drought. Obtained by my interpreter. Of value like the preceding numbers." (*Meyer.*)

32222. Trifolium pratense L. **Red clover.**

From Omsk, Siberia.

"(No. 1680a, July 18, 1911.) A species of clover said to stand much drought. Obtained by my interpreter. Of value like the preceding numbers." (*Meyer.*)

32223. Malus sp. **Crab apple.**

From Barnaul, Siberia.

"(No. 1681a, September 5, 1911.) A low-growing variety of Siberian crab apple. The fruits are the size of a plum, of a yellowish color with a red blush. Thrives best on hill slopes facing north; suitable for cold regions." (*Meyer.*)

32224. Prunus fruticosa Pallas. **Cherry.**

From Omsk, Siberia.

"(No. 1682a, August 5, 1911.) The so-called Kurgan cherry, grown quite extensively throughout the Ural district and in western Siberia as a home fruit. The dark-red individual cherries are only as large as good-sized red currants and are sour. However, they make delicious compote and preserves, having a spicy, nutty flavor, and are in great demand for this purpose. In Omsk they are sold during July and August at 4 to 5 rubles ($2.06 to $2.57) per pood (36 pounds). These cherry bushes grow from 2 to 4 feet in height, bear glossy dark-green leaves, and stand a remarkable amount of drought, cold, and neglect. A well-kept plantation is very pleasing to the eye and resembles a tea plantation more than anything else. This plant deserves to be given an extensive and thorough trial as a home fruit in the cold and semiarid sections of the United States. Is recommended also as a factor in hybridization experiments to

32175 to 32245—Continued.

32224—Continued.

create a perfectly hardy large-fruited cherry fit to thrive in the upper Mississippi Valley and the regions west of it." (*Meyer.*)

Distribution.—Eastern Russia and western Siberia from the valley of the Volga to the Altai region.

32225. PRUNUS FRUTICOSA Pallas. **Cherry.**

From Tomsk, Siberia.

"(No. 1683a, August 25, 1911.) An improved form of the Kurgan cherry. Obtained from Prof. N. F. Kastchenko, of the University of Tomsk. See the preceding number for further remarks." (*Meyer.*)

32226. PRUNUS FRUTICOSA Pallas. **Cherry.**

From Tomsk, Siberia.

"(No. 1684a, August 25, 1911.) A large-fruited variety of the Kurgan cherry almost as large as ordinary cherries, flesh blackish red. This introduction deserves extra care. Obtained like the preceding numbers." (*Meyer.*)

32227. RIBES DIKUSCHA Fisch. **Currant.**

From Tomsk, Siberia.

"(No. 1685a, August 18, 1911.) The native name of this remarkable currant is *Aldansky vinograd,* meaning Aldan grape, on account of the resemblance of the berries to grapes. It is a black-fruited variety native to the Aldan Mountains in Yakutsk Government. Berries large, bluish black in color, and of sour flavor. They are fine for preserves and are also said to produce a sparkling wine resembling champagne. As the summer is remarkably short in Yakutsk Government and the winter's cold most intense, this currant may be expected to thrive in even the coldest sections of the United States. To be tested in the mountains of New England and in Alaska. Collected in the botanic garden of the University of Tomsk." (*Meyer.*)

32228. RIBES DIKUSCHA Fisch. **Currant.**

From Tomsk, Siberia.

"(No. 1686a, August 25, 1911.) Variety *appendiculata.* An improved form of the Yakutsk black currant, being more prolific and bearing larger berries than the type. Obtained from Prof. N. F. Kastchenko of the University of Tomsk, who states that the climate in Tomsk is actually too hot for this remarkable currant. This number should be tested with extreme care." (*Meyer.*)

32229. RIBES NIGRUM L. **Black currant.**

From Tomsk, Siberia.

"(No. 1687a, August 18, 1911.) A native Siberian variety of black currant of vigorous growth; fruits large, but not over juicy. Able to stand intense cold and recommended as a home fruit for the cool and somewhat moist sections of the United States. Obtained from the botanical garden of the University of Tomsk, Siberia." (*Meyer.*)

32230. RIBES sp. **Currant.**

From near Sminogorsk, southwestern Siberia.

"(No. 1688a, September 24, 1911.) A wild red currant of thrifty growth and able to exist under adverse conditions. Fruits small and sour. This fruit is recommended for testing only in the sections of the United States where red currants do not grow at present." (*Meyer.*)

32175 to 32245—Continued.

32231. Rubus arcticus L. **Nimbleberry.**

From Barnaul, Siberia.

"(No. 1689a, September 5, 1911.) A wild nimbleberry from Finland, called in Swedish *Okerbar*. Said to possess a remarkably delicious fragrance, and is used in small quantities for flavoring compotes and preserves. Obtained from a Finnish family in Barnaul. To be tested in some cool and moist section of the United States, preferably southern Alaska." (*Meyer.*)

32232. Solanum dulcamara L. **Bittersweet.**

From Tomsk, Siberia.

"(No. 1690a, August 24, 1911.) Variety *persicum.* A vigorously growing variety of bittersweet, growing in shrubbery up to a height of over 10 feet. Stands extreme cold and may be used as an ornamental porch and pillar vine. To be tested in the cool and moist sections of the United States." (*Meyer.*)

32233. Crataegus sanguinea Pallas. **Hawthorn.**

From near Tomsk, Siberia.

"(No. 1691a, August 24, 1911.) An ornamental native Siberian haw, much used in Tomsk as a hedge plant. When left alone this haw develops into a tall shrub and becomes loaded in the fall with masses of orange-red berries, which make these shrubs very ornamental. The berries are often collected by the Russian peasants and after having been boiled with sugar a passable haw butter is made from them. To be tested in the cool and moist sections of the United States." (*Meyer*)

Distribution.—Throughout Siberia from the Ural Mountains to the region of Lake Baikal.

32234. Cotoneaster sp.

From near Sminogorsk, southwestern Siberia.

"(No. 1692a, September 18, 1911.) A cotoneaster growing 2 to 3 feet in height. Occurs on dry stony hill slopes. Possibly of value as a small ornamental shrub in gardens and parks in the cool semiarid sections of the United States." (*Meyer.*)

32235. Berberis sibirica Pallas. **Barberry.**

From near Sminogorsk, southwestern Siberia.

"(No. 1693a.) A rare low-growing species of barberry, occurring on rocky mountain slopes facing north or northeast. Rarely seen over 1 foot in height. Berries hang solitary and are of coral-red color. Suitable for planting in rockeries and as an ornamental ground cover on rocky places in the colder sections of the United States." (*Meyer.*)

Distribution.—The Altai Mountain region of southwestern Siberia and northern Mongolia.

32236. Juniperus sabina L. **Juniper.**

From near Sminogorsk, southwestern Siberia.

"(No. 1694a, September 24, 1911.) A pretty, dense-growing variety of juniper, found on wind-swept, rocky mountain slopes, facing south or southeast. Often very spreading in habit. Of value like the preceding number." (*Meyer.*)

32237. Artemisia sp.

From near Kalmukski Meesi, southwestern Siberia.

"(No. 1695a, September 15, 1911.) A perennial semiwoody wormwood having gracefully divided light-green foliage and possessing a very attractive, aromatic odor. Grows to a height of 3 to 4 feet and occurs on dry waste places.

32175 to 32245—Continued.

32237—Continued.

Of value as a lining plant along paths and as low hedge material for the cold semiarid sections of the United States." (*Meyer.*)

32238. Hyssopus officinalis L. **Hyssop.**

From near Ust Kamenogorsk, southwestern Siberia.

"(No. 1696a, October 2, 1911.) Variety *ambigua*. A very aromatic herbaceous perennial growing in rocky cliffs and between stony débris along the Irtish River. Flowers blue and apparently very rich in honey. Of value as a useful bee plant and as an ornamental perennial for the hardy border. Will probably do especially well in the drier sections of the United States." (*Meyer.*)

32239. Clematis integrifolia L. **Clematis.**

From near Sminogorsk, southwestern Siberia.

"(No. 1697a, September 18, 1911.) An erect-growing herbaceous clematis, making stems 2 to 4 feet in height and bearing large deep-blue flowers of somewhat roselike form; blooms in June. Occurs between shrubbery or in slightly shaded places where the soil remains damp. Of value as an ornamental hardy perennial for the northern United States and may also be of use as a factor in hybridization experiments in trying to create races of erect-growing large-flowered forms of clematis." (*Meyer.*)

32240. Paeonia anomala L. **Peony.**

From Tomsk, Siberia.

"(No. 1698a, August 19, 1911.) A strong-growing wild peony found on well-drained hill slopes, mostly between scrub. Flowers large, of pale-rose color; foliage somewhat coarse. Of interest as a perfectly hardy herbaceous perennial; will thrive in regions with short cool summers and long cold winters." (*Meyer.*)

Distribution.—Throughout central and southern Siberia from the Ural Mountains eastward to the region of Lake Baikal.

32241. × Rhododendron praecox Davis.

From Tomsk, Siberia.

"(No. 1699a, August 20, 1911.) A rare deciduous shrubby rhododendron, able to stand extreme cold. Obtained from the botanical garden at Tomsk, Siberia." (*Meyer.*)

Considered to be a hybrid between *Rhododendron dauricum* L. and *R. ciliatum* Hook. f.

32242. Gueldenstaedtia monophylla Fisch.

From Tomsk, Siberia.

"(No. 1700a, August 20, 1911.) A very rare legume, up to the present having been found only in one place in Mongolia and in another spot in southern Siberia, occurring on dry, rocky places. Leaves and flowers small. This plant is of botanical interest only, although it possibly might enter as an item in the flora of very dry pasturing grounds in the western sections of the United States. Obtained like the preceding number." (*Meyer.*)

Distribution.—Dry rocky places in the Altai Mountain region of southwestern Siberia.

32243. Asparagus officinalis L. **Asparagus.**

From Chistunka Steppe, southwestern Siberia.

"(No. 1701a, September 8, 1911.) A vigorously growing wild asparagus, found here and there on a dry plain. Seems to be able to withstand more drought and adverse conditions than the ordinary forms of asparagus. To be tested in rustproof breeding experiments." (*Meyer.*)

32175 to 32245—Continued.

32244. Citrullus vulgaris Schrad. **Watermelon.**

From Ust Kamenogorsk, southwestern Siberia.

"(No. 1702a, September 30, 1911.) A watermelon of medium size; rind dark green, quite thin; flesh pale red, of fresh sweet taste; seeds brown. Keeps well and is a good variety in general. Of value possibly in extending the watermelon belt farther north." (*Meyer.*)

32245. Citrullus vulgaris Schrad. **Watermelon.**

From Ust Kamenogorsk, Siberia.

"(No. 1703a, October 1, 1911.) A watermelon of medium size; rind light green, very thin; flesh dark red and very sweet; seeds very small, of brownish black color. Is a good keeper. To be tested like the preceding number." (*Meyer.*)

32247. Gossypium sp. **Cotton.**

From Addis Abeba, Abyssinia. Presented by Mr. Guy R. Love, American vice consul general, Addis Abeba. Received November 23, 1911.

"Native Abyssinian cotton." (*Love.*)

32248. Annona muricata L. **Soursop.**

From Camaguey, Cuba. Presented by Mr. Roberto L. Luáces. Received December 5, 1911.

"I have found here in this part of Cuba two trees of *Annona muricata*, growing in primitive woods on the banks of the Caunao River. From fruit of these trees I collected some seed. These seeds are not from cultivated plants escaped from cultivation, for, in the first place, the general mode of carriage of seeds from cultivation to wildness, such as birds, is wanting in this case, for neither birds nor beasts (except man) will eat the *A. muricata*, and, besides, the tree itself and its fruits are very different from what we call the cultivated." (*Luáces.*)

32249 to 32255. Citrus trifoliata × aurantium. **Citrange.**

The following plants, propagated by Mr. G. L. Taber, Glen St. Mary Nursery Co., Glen St. Mary, Fla., for distribution by the Office of Crop Physiology and Breeding Investigations, were numbered December 7, 1911.

Seedling plants as follows:

32249. *Willits.* See No. 13003 for description.

32250. *Rustic.* See No. 19608 for description.

32251. *Savage.* See No. 21594 for description.

32252. *Norton.* "Fruits small and somewhat ribbed, resembling the *Savage* (No. 21594)." (*S. C. Mason.*)

32253. *Saunders.* "Fruits small, orange yellow, with unusually thick skin. Pulp vesicles rather large, adhering compactly, making the interior of the fruit seem hard and dry as compared to the other citranges. What juice there is is sharply acid and entirely free from the bitter taste so prominent in some of the citranges." (*S. C. Mason.*)

Grafted plants as follows:

32254. *Cunningham.* "Resembles a miniature *Colman* (No. 19609), having the same fuzzy skin possessed by that citrange." (*S. C. Mason.*)

32255. *Sanford.*

32256. Mangifera indica L. **Mango.**

From Chiloane Island, Africa. Presented by Mr. R. H. B. Dickinson, Assistant Director of Agriculture, Beira, Portuguese East Africa. Received December 8, 1911.

"Cuttings taken from a tree said to be 50 or 60 years old, growing near a small Mohammedan temple. It bears large fruits, which may be expected to ripen in January." (*Dickinson.*)

This may possibly be the *Lathrop* mango described under Nos. 9486 and 9669.

32257. (Undetermined.) **Indian cane.**

From Burringbar, New South Wales, Australia. Presented by Mr. B. Harrison. Received July 3, 1911. Numbered December 15, 1911.

"I wish to draw your attention to the value of Indian cane as a heavy yielding fodder plant for dairy or other stock. It is rapidly coming into great favor here and yields from 40 to 100 tons of fodder per acre, while it is said to be superior to any of the sorghum species for resisting drought and frost and is not injurious to stock during any period of its growth. One writer says: 'After some forty years' practical experience in fodder growing I consider this cane miles ahead of anything else I have tried before, the great advantage being that you can cut it as you require it; secondly, the enormous yield. I estimate the yield of my crop at 56½ tons per acre, but the land was manured heavily.' There is a large area of land in many of the States where this cane would thrive well and would without doubt prove of incalculable benefit to many stock owners." (*Harrison.*)

32258. Scopolina japonica (Maxim.) Kuntze.

From Yokohama, Japan. Purchased from the Yokohama Nursery Co. Received December 6, 1911.

Distribution.—A herbaceous perennial found along the banks of streams in the vicinity of Nikko in the province of Tozando, Japan.

32259. Garcinia tinctoria (DC.) W. F. Wight.

From Port Louis, Mauritius. Presented by Mr. G. Regnard. Received December 8, 1911.

Introduced as a possible stock for the mangosteen.

32260 and 32261. Neoglaziovia spp.

From Bahia, Brazil. Presented by Mr. Omar E. Mueller, American vice consul. Received November 29, 1911.

Plants of the following; quoted notes from Bulletin of the Pan American Union, 1910:

"These fiber plants are of great commercial worth. They grow abundantly in this section, but other than making a few cords for local use nothing is being done with them."

32260. Neoglaziovia variegata (Arrudo) Mez. **Caroá.**

"This is half round, light green, white banded, snakelike, produces an excellent fiber, and flourishes regardless of droughts.

"Mr. Louis Raposo, a Brazilian gentleman resident in Philadelphia, gives the following information concerning this plant:

" 'Among the new things found growing in the vast wilds of Brazil is a fibrous plant called *caroá*, of which the supply is apparently inexhaustible. The plant is produced from a bulb and is of rapid growth. When stripped at maturity of its fiber it takes but six months under the coaxing influence of the Brazilian sun and soil to reproduce a full-length crop from 6 to 8 feet in length.

32260 and 32261—Continued.

32260—Continued.

"Mr. Raposo states that shipments of the fiber sent to London and manufactured into rope, as tested by English engineers, show a tensile strength 10 times greater than manila rope of like dimension. As compared with other products from which rope and twine are made, the caroá gives a 10 per cent greater outturn of finished material from the same weight of raw material.

"'Tests of caroá rope,' he says, 'for use on shipboard show great wearing quality, as well as other superiorities. The rope does not swell from wetting. In three round trips from London to Bombay the rope gave no appearance of serious damage.

"'It takes 20 tons of the green caroá to make 1 ton of the fiber ready for shipment, but a large part of the wastage is said to be suitable for making paper. This if true would give the plant a far more considerable value. The cost of the fiber as rudely produced in Brazil and laid down in London was $80 per ton. This cost would be largely reduced by economical handling. The discovery of this new fiber, if what is claimed for it be true, is a most important addition to the raw material of manufacture.' " (*Bulletin, Pan American Union, 1910.*)

"This species and the closely related following one occur throughout the arid districts along the Rio Sao Francisco in a climate said to resemble that of our arid Southwest. Mr. Tennant Lee, who saw 2,000 acres of these plants in a wild state and who tested their fiber, says this is one of the finest fibers ever brought into the United States. Ropes made of it will stand salt water longer than manila hemp; it is 28 per cent stronger than manila, yields a larger percentage of fiber than the abacá, and the waste is suitable for paper making." (*Fairchild.*)

32261. Neoglaziovia concolor C. H. Wright. **Makimbira.**

"This plant has leaves protected by stout incurved spines upon their edges, thereby rendering the handling both difficult and dangerous."

32262. Casimiroa sapota Oersted. **Sapota blanco.**

From San Jose, Costa Rica. Presented by Mr. Carlos Wercklé, Museo Nacional. Received August 4, 1911. Numbered December 15, 1911.

32263. Echium pininana Webb and Berth.

From Palma, Canary Islands. Presented by Dr. George V. Perez, Puerto Orotava, Teneriffe, Canary Islands. Received December 8, 1911.

"This is a most striking ornamental plant with a very tall single spike of light-blue flowers. I believe the leaves will turn out to be an excellent forage, better than the prickly comfrey (No. 2152). These seeds are from a wild plant in Palma, where it is native. Try it in southern California or Florida." (*Perez.*)

32264. Garcinia venulosa (Blanco) Choisy.

From the Limay Forest Station, Philippine Islands. Presented by Maj. George P. Ahern, Director of Forestry, Department of the Interior, Manila. Received December 9, 1911.

"The seeds from which these plants were grown were collected in Bataan Province." (*Ahern.*)

Distribution.—A tree found in the Philippines.

32265 and 32266. CICER ARIETINUM L. **Chick-pea.**

From Chihuahua, Mexico. Presented by Mr. Marion Letcher, American consul. Received October 9, 1911.

Seeds of the following:

32265. "*Garbanzo grande.*" 32266. "*Garbanzo chico.*"

32267. DIOSPYROS KAKI L. f. **Persimmon.**

From Fayetteville, N. C. Presented by Mr. J. S. Breece. Received October, 1911.

"Seeds taken from a Tamopan persimmon, grown by Mr. Breece under No. 17172. The other fruits received at the same time were seedless and it is thought that the seeds in the single specimen were the result of accidental cross-pollination." (*Fairchild.*)

The fruit from which these seeds were taken is identical with Mr. Frank N. Meyer's description of No. 16921, and it is thought that there may have been some mistake made in sending it out under No. 17172.

32268 to 32271. ASPARAGUS spp. **Asparagus.**

From South Africa. Presented by Sir Percy Fitzpatrick, Johannesburg, Transvaal, South Africa. Received December 6, 1911.

Seeds of the following; quoted notes by Sir Percy Fitzpatrick:

32268 to 32270. ASPARAGUS OFFICINALIS L.

32268. "From Harrismith, Orange Free State."

32269. "From cultivated plants, Johannesburg."

32270. "From cultivated plants near Johannesburg."

32271. ASPARAGUS sp.

"Seed gathered on slopes of Table Mountain from wild plants in native bush forest. This asparagus is a great delicacy and to my taste is better than any of the cultivated kinds. Table Mountain, near Cape Town, is 1,000 miles away. The asparagus is gathered by colored people, who are jealous of the monopoly and most ignorant of methods of seed collecting. I got some seed in March last, but the berries were quite green in color. I know that our wild asparagus (which is here known as *Wach-een-beetze*, or Wait-a-bit, on account of its hooked thorns) ripens red, as do the others. I tried again during all April, but the 'children of the sun' smilingly replied that I was mistaken and that the green berries were quite ripe."

32272 to 32277.

From Darmstadt, Germany. Presented by Prof. William R. Lazenby, of the Ohio State University, Columbus, Ohio. Received November 23, 1911.

Seeds of the following; quoted notes by Mr. Lazenby:

32272. LYCIUM BARBARUM L.

"This is used very largely here as a hedge plant around yards and small orchards. It grows so dense that rabbits can not get through it. I am told it is easily pruned and kept within bounds. It is also planted on sandy railroad embankments to bind the soil and prevent washing or sliding."

Distribution.—A shrub found in southwestern Asia from Mesopotamia eastward through Persia to the Punjab region of northern India.

32272 to 32277—Continued.

32273. Rosa sp. **Rose.**

"A rose used for the same purpose as the preceding number. The fruits or hips of this rose are very ornamental. It grows along the edges of the forest. I have no idea what the flower is, but the plant seems to have merit. May be useful for breeding purposes."

32274. Euonymus sp.

"A fruitful and beautiful Euonymus."

32275. Viburnum sp.

"A strong-growing variety."

32276. Prunus sp.

"This seems to have merit as a hedge plant."

32277. Ptelea sp.

"A Ptelea whose large-winged seeds are quite attractive. It seems to be of much more vigorous growth than our common *Ptelea trifoliata.*"

32278. Belou marmelos (L.) Lyons. **Bael.**

From Zafarwal, India. Presented by Mr. H. S. Nesbitt, American Mission. Received July 20, 1911. Numbered December 14, 1911.

"This bael is used in India as an astringent curative of dysentery, etc." (*Nesbitt.*) See No. 24450 for further description.

32280 and 32281.

From Yengi Malah, Tieñ Shan Mountains, Chinese Turkestan. Collected by Mr. F. N. Meyer, agricultural explorer, Bureau of Plant Industry. Received November 3, 1911.

Seeds of the following; picked out of No. 32042. See that number for remarks.

32280. Avena sativa L. **Oat.**

32281. Hordeum sp. **Barley.**

32282. Annona diversifolia Safford. **Ilama, or anona blanca.**

From Salvador, Central America. Presented by Don Rafael B. Castillo, Director General of Agriculture, through Mr. W. E. Safford. Received December 12, 1911.

Seeds.

32283 to 32294.

From Manila, Philippine Islands. Presented by Mr. E. D. Merrill, Bureau of Science. Received November 11, 1911. Numbered December 15, 1911.

Seeds of the following:

32283. Pahudia rhomboidea (Blanco) Prain.

See No. 31586 for description.

32284. Parkia timoriana Merrill.

32285. Pithecolobium acle (Blanco) Vidal.

32286. Phytocrene blancoi Merrill.

32287. Pandanus luzonensis Merrill.

Distribution.—A treelike species reaching a height of 25 feet, found in the valley of the Lamao River in the island of Luzon in the Philippines.

32283 to 32294—Continued.

32288. Mucuna sp.

32289. Barringtonia sp. (?)

32290. Terminalia ovocarpa Merrill.

Distribution.—A large tree growing at an elevation of 300 to 400 feet in the dry hill forests in the island of Luzon, in the Philippines.

32291. Knema heterophylla (Vill.) Warburg. (?)

Distribution.—A tall tree found on the islands of Luzon, Panay, Mindanao, and Sulu in the Philippines.

32292. Caryota cumingii Lodd.

Distribution.—Known only from the Philippines.

32293. Dracontomelon edule (Blanco) Skeels. **Lamio.**

(*Paliurus edulis* Blanco, 1837, Flora de Filipinas, p. 174.)

The seeds of this Philippine tree, which belongs to the family Anacardiaceæ, were received under the name *Dracontomelon cumingianum*. This name is attributed in the Index Kewensis to Baillon (Bulletin de la Société Linnéenne de Paris, vol. 1, 1877, p. 122), who, although he did not use the binomial credited to him, cited Adansonia, vol. 10, 1872, p. 329, where the genus Comeurya is characterized and one species, *C. cumingiana*, is listed. Merrill (A Review of the Identifications of the Species Described in Blanco's Flora de Filipinas, 1905, p. 36) has identified *Paliurus edulis* Blanco as being, for the most part, the same as *Dracontomelon cumingianum* Baillon. Blanco's specific name, being earlier, is here restored.

32294. Chalcas crenulata (Turcz.) Mueller.

Distribution.—A shrub or small tree found in the Philippines and in Queensland, Australia.

32295. Coffea arabica L. **Coffee.**

Grown not far from the town of Villeta, at an altitude of 3,000 feet, in the province of Cundinamarca, Colombia. Presented by Mr. Arthur Hugh Frazier, Chargé d'Affaires ad Interim, American Legation, Bogota. Received November 29, 1911.

"Actually no coffee is grown at or near Bogota, the altitude being much too high for the plant. The so-called Bogota coffee is grown at altitudes varying from 3,000 to 6,000 feet and, roughly speaking, is confined to the Department of Cundinamarca, although in the trade the coffees of Antioquia and Santander are sometimes confused with the product of Cundinamarca." (*Frazier.*)

32296 and 32297.

From Costa Rica. Presented by Mr. Carlos Wercklé, Museo Nacional. Received December 18, 1911.

Seeds of the following:

32296. Lucuma sp.

Seed small, roundish.

32297. Achras zapota L. **Mamee sapota.**

32298 to 32301. ANNONA CHERIMOLA Miller. **Cherimoya.**

From Costa Rica. Presented by Mr. Carlos Wercklé, Museo Nacional, San Jose. Received December 9, 1911.

Seeds of the following; quoted notes by Mr. Wercklé:

32298. "Fruit medium to large, round, with well-marked mottling only or a slight depression in upper part of carpel; green with a brownish gray tinge. Highest quality, less seeds than any other I have seen (eight in a good-sized fruit). The fruit from which these seeds were taken was quite distinct in form, very plump and thick, short, hardly tapering, not compressed, medium sized, brownish black."

32299. "Fruit medium to large, similar to the preceding number, but without the brownish gray tinge, not inferior to it, but more seedy (still few seeds). Seeds of common shape and color."

32300.

"Fruit large, cordiform ovate, completely even, with no depressions at all in the carpels and netting; i. e., the edge of carpels or rather the division lines of the carpels not noticeable, except in a very few places. Color yellowish green to greenish yellow, suffused with a very light brownish tinge; on the sun side the color passes to greenish orange, which is very rare in this species. In many places the skin is a little russety. Seeds few; black, long, and narrow. Quality the highest."

32301.

"Quite a good annona from San Pedro."

32302. ANNONA MURICATA L. **Soursop.**

From Camaguey, Cuba. Presented by Mr. Roberto L. Luáces. Received December 18, 1911.

"Seeds from a cultivated tree with sweet fruits." (*Luáces.*)

32303 to 32308.

From near Saratoff, southeastern Russia. Received through Mr. Frank N. Meyer, agricultural explorer, Bureau of Plant Industry, December 19, 1911.

The following material:

32303. MALUS sp. **Apple.**

"(No. 991, November 23, 1911.) A wild apple of shrubby growth occurring on dry, well-drained hills, in company with such shrubs as *Acer tataricum*, *Rhamnus cathartica*, *Prunus spinosa*, *Spiraea crenifolia*, etc. Of value possibly in breeding work." (*Meyer.*)

32304. SALIX sp. **Willow.**

"(No. 992, November 23, 1911.) A medium-sized willow of slightly drooping habits. The young branches are of a shiny brown color, while the trunk and heavy branches assume a grayish green color. Occurring on rather dry places. Of value as an ornamental park and garden tree in those semiarid sections of the United States where the summers are hot and the winters cold." (*Meyer.*)

32305. CORONILLA VARIA L. (?).

"(No. 993, November 23, 1911.) A legume found on dry hill slopes and in loess ravines. Apparently very drought resistant. Of possible value as a forage and pasture plant for the drier sections of the United States." (*Meyer.*)

32303 to 32308—Continued.

32306. MEDICAGO sp.

"(No. 994, November 23, 1911.) A wild alfalfa of erect growth occurring on stony places. May possibly be *Medicago caerulea*."

32307. HEDYSARUM GRANDIFLORUM Pallas (?).

"(No. 995, November 24, 1911.) This plant occurs on hill slopes which are mainly composed of limestone. It is of low growth and may possibly possess value as a pasturing plant in semiarid regions; also as an ornamental garden perennial. See Nos. 1645a to 1647a (Nos. 32187 to 32189) for further remarks." (*Meyer*.)

32308. PYRUS COMMUNIS L. **Pear.**

"(No. 996, November 24, 1911.) A wild pear of shrubby growth, growing on dry, pebbly hill slopes in company with *Prunus spinosa*, *Crataegus sanguinea*, etc. Possibly of value in breeding experiments." (*Meyer*.)

32309. LAGERSTROEMIA SPECIOSA (L.) Pers.

From Lamao, Bataan, Philippine Islands. Presented by Mr. O. B. Burrell, superintendent, Lamao Experiment Station, Bureau of Agriculture. Received December 20, 1911.

Distribution.—A tall tree found from northwestern India, where it is cultivated, southward and eastward to China and the Malay Archipelago.

32310. ASPARAGUS FILICINUS Hamilton. **Asparagus.**

From Sibpur, near Calcutta, India. Presented by Maj. A. T. Gage, superintendent, Royal Botanic Garden. Received December 18, 1911.

32311 to 32315.

From Dehra Dun, India. Presented by Mr. A. C. Hartless, superintendent, Government Botanic Gardens, United Provinces, Seharunpur. Received December 18, 1911.

Seeds of the following:

32311. ALYSICARPUS VAGINALIS (L.) DC.

"Variety No. 2."

32312. ALYSICARPUS RUGOSUS (Willd.) DC

"Variety No. 1. Narrow leaved."

32313. CASSIA MIMOSOIDES L.

32314. ZORNIA DIPHYLLA (L.) Persoon.

Distribution.—On the plains of India and cosmopolitan throughout the Tropics.

32315. ANDROPOGON MONTICOLA Schultes.

Distribution.—On the slopes of the Himalayas in northern India, where it ascends to an elevation of 6,000 feet, and southward to Ceylon and Burma.

32316 and 32317.

From India. Procured by Mr. R. S. Woglum, Bureau of Entomology, United States Department of Agriculture. Received December 12, 1911.

Seeds of the following:

32316. Cassia fistula L.

"A large tree with very pretty, yellow flowers. Seed pods cylindrical, 1 to 1½ feet long, ¾ inches in diameter. My boy says that the natives use the partitions between the seeds to steep into tea which is used for fever, etc. Seeds are not used." (*Woglum.*)

See No. 29182 for previous introduction.

32317. Chalcas paniculata L.

See No. 25350 for previous introduction.

32318 and 32319.

From Costa Rica. Presented by Mr. Carlos Wercklé, Museo Nacional, San Jose. Received December 26, 1911.

The following material; quoted notes by Mr. Wercklé:

32318. Musa sp.

"*Banana-guinea*, intermediate between the banana and the guinea; better than the banana. From the Coyalar."

32319. Annona sp.

"*Anona de San Francisco.* Bears the second or third year; highest quality; stands the climate of the coast perfectly. Takes as splice graft on anona, guanávana, and soncoya."

See Nos. 31574 to 31576 for previous introduction of this variety.

32322. Annona sp.

From Costa Rica. Presented by Mr. Carlos Wercklé, Museo Nacional, San Jose. Received December 26, 1911.

"Mixed lot of seed of some very good varieties of anonas." (*Wercklé.*)

32323. Castanea sativa Miller. **Chestnut.**

From Kutien, Fukien, China. Presented by Dr. T. H. Coole. Received December 22, 1911.

32324. Aralia cordata Thunb. **Udo.**

From Sapporo, Japan. Presented by Mr. Y. Takahashi, botanist and vegetable pathologist, Hokkaido Agricultural Experiment Station. Received December 28, 1911.

"Roots of a good cultivated strain of native udo." (*Takahashi.*)

32325. Ficus rigo F. M. Bailey.

From Barodobo, Kapa Kapa, Papua. Presented by Mr. A. C. English. Received September 27, 1911. Numbered December 15, 1911.

"A good rubber-producing tree, hardy and will grow well in our dry belt here. The rubber from this tree is on a par with Para rubber. I have not yet been able to get the seed to germinate. I started my plantation with plants of this species obtained from the scrubs growing as a parasite on other trees, the seed being carried by birds and animals, and after passing through the bowels, it then germinates in the forks of trees and in decomposed timber. It readily strikes from cuttings and it appears to be free from all diseases." (*English.*)

Distribution.—The Rigo district of southeastern British New Guinea.

32326. ANDROPOGON sp. **Lemon grass.**

From Tampico, Mexico. Presented by Mr. Clarence A. Miller, American consul. Received November 22, 1911. Numbered December 30, 1911.

"*Zacate Limon.* This is said to be a domestic plant. Each stalk should be separated, as it grows up like a multiplying onion and bears no seed. Each stalk with ordinary care usually produces a bunch about the size of a dish pan in a year. By many the leaves or blades are picked fresh. The leaves are then steeped and the resulting tea is used as a table beverage. This grass is very plentiful in this section." (*Miller.*)

32327 and 32328.

From Rome, Italy. Presented by Dr. Gustav Eisen, California Academy of Sciences, San Francisco, Cal. Received December 26, 1911.

Seeds of the following; quoted notes by Dr. Eisen:

32327. PHOENIX DACTYLIFERA L. **Date.**

"*Zagloul.* The very best date that I have eaten anywhere. It is very large, in fact the largest date I have seen. Medium dark-brown, sweet, and remarkably tender flesh. Said to come from Fayum, but the dates were bought in Cairo. Ripe in January and lasts fresh until April. Best in March and February. Said to be the best date in Egypt."

32328. PRUNUS DOMESTICA L. **Plum.**

"*Papagone.* The finest plum I have eaten in any country. Native of the campagna around Naples, especially Boscotrecase and other towns around Vesuvius. About 3 inches long, greenish yellow, oblong, with a remarkably long, thin, and slender stone compared to the size of the fruit. The quality of this plum can not be too highly praised. Have not seen it in any other part of Europe, nor in California."

32329 to 32347.

From Buitenzorg, Java. Presented by the Director of Agriculture, at the request of Mr. C. V. Piper. Received November 2, 1911.

Seeds of the following; quoted notes by Mr. Piper:

32329. ARUNDINELLA sp.

"A rather coarse but leafy species and probably valuable for green feeding."

32330. AXONOPUS COMPRESSUS (Swartz) Beauv.

"A carpet grass."

Distribution.—Jamaica and other islands of the West Indies; naturalized in the Tropics.

32331. BRADBURYA PUBESCENS (Benth.) Kuntze.

"A twining legume forming a dense mass 6 feet tall when supported."

Distribution.—In southern Mexico, from the vicinity of Vera Cruz and Orizaba southward, through tropical South America, and in the West Indies.

32332. BOTOR PALUSTRIS (Desv.) Kuntze.

"Grown in the Buitenzorg Botanic Garden under the name of *Psophocarpus robustus.* A climbing vine much larger and more vigorous than the ordinary Goa bean."

Distribution.—Considered to be a native of Africa; generally cultivated in the Tropics.

32329 to 32347—Continued.

32333. Cassia rotundifolia Pers.

"A low-growing legume having much the same habit as *Lotus corniculatus*. It produces seed in abundance."

Distribution.—From the region of Orizaba in southern Mexico southward to Uruguay.

32334. Cracca villosa argentea (Lam.) Kuntze.

"An open bushy species growing 3 feet high. This seed shatters easily."

Distribution.—On the plains of India from the Himalayas to Ceylon.

32335. Dolicholus minimus (L.) Medic.

"A slender vine climbing to a height of 12 or 15 feet in the trees. In cultivated ground it often makes a dense covering a foot or more deep and is inclined to be somewhat weedy."

Distribution.—Throughout the plains of India and generally distributed in the Tropics.

32336. Dracontomelon dao (Blanco) Merrill and Rolfe. **Dao.**

Distribution.—A tree found on the islands of Luzon and Mindoro in the Philippines.

32337. Eragrostis sp.

32338. Meibomia sp.

"A species having much the same habit as the Florida beggarweed and perhaps of equal value."

32339. Panicum argenteum R. Brown.

Distribution.—On the islands in the Gulf of Carpentaria on the northern coast of Australia.

32340. Panicum radicans Retz.

Distribution. Throughout India and eastward to tropical China and the Malayan and Polynesian Islands.

32341. Paspalum conjugatum Berg.

"This species is called *Carabao* grass in the Philippines, and *Bitter* grass in Java. It forms a vigorous, often pure, growth in low-lying lands, and also grows abundantly in dense shade. Stock are not very fond of it."

Distribution.—From San Luis Potosi in northern Mexico southward to Brazil; also in tropical Africa and Asia.

32342. Phaseolus sublobatus Wall.

"A twining species with slender stems."

Distribution.—Found on the estuary of the Irrawaddy River in southern Burma, India.

32343. Syntherisma ciliaris (Retz.) Schrad.

32344. Tricholaena rosea Nees.

"A grass very closely resembling tall grass and possibly identical."

32345. Tricholaena rosea Nees.

See previous number for description.

32346. Vigna sp. (?)

"An annual legume with the habit of the cowpea, but less vigorous in growth and the stems more slender and twining."

32329 to 32347—Continued.

32347. (Undetermined.)

"A coarse species with the general appearance of Para grass, but the stems are erect."

32348 to 32350. Prunus armeniaca L. Apricot.

From Imperial Estate "Murgab," Bairam Ali, Oasis of Merv, Russian Turkestan. Collected by Mr. Frank N. Meyer, agricultural explorer, Bureau of Plant Industry, November, 1911. Received December 29, 1911.

Cuttings of the following; quoted notes by Mr. Meyer:

32348. "(No. 1001.) A native central Asian variety of apricot, called *Murgab*. Said to be of fine qualities."

32349. "(No. 1003.) A native central Asian variety of apricot, called *Abutalibe*. Said to be of fine qualities."

32350. "(No. 1004.) A native central Asian variety of apricot, called *Acha-djanowood*. Said to be of fine qualities."

32351. Blighia sapida Koenig. Akee.

From Jamaica, British West Indies. Presented by Mr. W. Harris, superintendent, Hope Botanic Gardens, Kingston. Received December 29, 1911.

See No. 24592 for description.

Seeds.

32352. Quercus suber L. Cork oak

From Algeciras, Spain. Procured through Mr. R. L. Sprague, American consul, Gibraltar, Spain. Received December 29, 1911.

32353 to 32358.

From Mexico. Presented by Dr. C. A. Purpus, Zacuapam, Huatusco, Vera Cruz. Received December 26, 1911.

Seeds of the following; quoted notes by Dr. Purpus:

32353. Nicotiana sp. **Tobacco.**

"From Tehuacan, Puebla. Altitude 1,000 meters [3,280 feet]."

32354. Pinus montezumae rudis (Endl.) Shaw. **Pine.**

"From Esperanza. Altitude 2,700 to 2,800 meters [8,800 to 9,100 feet]."

Distribution.—From the southern part of the province of Chihuahua, in central Mexico, southeastward to Central America.

32355. Pinus leiophylla Schlecht. and Cham. **Pine.**

"From Amecameca. Altitude 2,500 to 2,600 meters [8,200 to 8,500 feet]."

Distribution.—Subtropical and warm temperate altitudes from the province of Zacatecas southeastward to the province of Oaxaca in Mexico.

32356. Pinus patula Schlecht. and Cham. **Pine.**

"From Boca del Monte, Puebla. Altitude 2,100 meters [6,900 feet]."

Distribution.—Warm temperate altitudes in the provinces of Hidalgo, Puebla, and Vera Cruz, in Mexico.

32353 to 32358—Continued.

32357. Pinus pseudostrobus Lindl. **Pine.**

"From Zacuapam, Huatusco."

Distribution.—From the province of Vera Cruz, in southern Mexico, southeastward to Nicaragua.

32358. Solanum nigrum L. **Nightshade.**

"From Zacuapam, Vera Cruz. Altitude, 3,000 feet."

32359. Coffea laurentii Wildem. Coffee.

From Kingston, Jamaica. Presented by Mr. William Harris, superintendent, Hope Botanic Gardens. Received December 30, 1911.

Procured for experimental growing in Arizona, southern California, and Florida.

32360. Malus sp. Crab apple.

From Jamaica Plain, Mass. Presented by Mr. Charles W. Livermore, Brookline, Mass. Received December, 1911.

"Seeds from an excellent crab apple. The tree from which these came is very hardy and bears abundantly. The apples are so generally superior that I hope you will be able to test the seeds at some station for the benefit of the Pacific coast." (*Livermore.*)

32361 to 32366.

From China. Presented by Mr. Nathaniel Gist Gee, Soochow University, Soochow. Received December 30, 1911.

Seeds of the following; quoted notes by Mr. Gist Gee:

32361. Glycine hispida (Moench) Maxim. **Soy bean.**

Olive green. "From Soochow. Hairy green beans planted in second or third month. Eaten green or dried."

32362. Vigna sinensis (Torner) Savi. **Cowpea.**

Black. "*Kaung doen*. About the same as red beans."

32363. Phaseolus radiatus L.

"Small green beans planted in the fifth moon, ready to harvest in two months. Eaten only after being dried."

32364. Phaseolus angularis (Willd.) W. F. Wight.

"Red beans planted the third or fourth month. Ready to eat in eight months. Dried before eating."

32365. Castanea sativa Miller. **Chestnut.**

"Large chestnut from Dong Ding Mountain, near Soochow."

32366. Castanopsis sp.

"From Tientsin."

32367 and 32368. Hordeum spp. Barley.

From Transvaal, South Africa. Presented by Prof. J. Burtt Davy, agrostologist and botanist, Department of Agriculture, Pretoria. Received December 26, 1911.

Seeds of the following:

32367. Hordeum vulgare himalayense Rittig.

32368. Hordeum vulgare cornutum Schrader. **Awnless barley.**

BOTANICAL NOTES AND PUBLICATION OF NEW NAMES.

32006. Dimocarpus longan Loureiro.

32036 and 32037. Languas galanga (L.) Stuntz.

32090. Leucadendron melliferum (Thunb.) W. F. Wight.

This South African proteaceous shrub has been listed in previous issues of the Inventories as *Protea mellifera* Thunberg (Dissertatio botanica de Protea, 1781, p. 34). In establishing the genus Protea (Species Plantarum, 1753, p. 94) Linnaeus published but two species, *P. argentea* and *P. fusca*. Both of these have since been referred to the genus Leucadendron of Berg (Handl. Vet. Akad. Stockholm, 1766, p. 325), which, however, is not the Leucadendron of Linnaeus (Species Plantarum, 1753, p. 91), the type of which is *L. lepidocarpodendron*. In his Mantissa (vol. 2, 1771, p. 189) Linnaeus, however, changed his conception of the genus Protea to include his *Leucadendron lepidocarpodendron*. This has resulted in the confusion and ultimate exact exchange of name between the two genera Protea and Leucadendron. Since the plant under discussion is congeneric with *Leucadendron lepidocarpodendron* and not with *Protea argentea*, the type of the genus Protea, it is necessary to retain it in the Linnaean genus Leucadendron. This was recognized in 1911 by Mr. W. F. Wight, who in the revised edition of the Century Dictionary, vol. 5, p. 3422, under "Leucadendron," published the name *Leucadendron melliferum*, based on *Protea mellifera* Thunberg.

32104. Benincasa hispida (Thunb.) Cogniaux.

32118. Syntherisma debilis (Desf.) Skeels.

32120. Syntherisma longiflora (Retz.) Skeels.

32194. Lentilla lens (L.) W. F. Wight.

32293. Dracontomelon edule (Blanco) Skeels.

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Issued December 17, 1912.

U. S. DEPARTMENT OF AGRICULTURE.

BUREAU OF PLANT INDUSTRY—BULLETIN NO. 262.

B. T. GALLOWAY, *Chief of Bureau.*

ORNAMENTAL CACTI:

THEIR CULTURE AND DECORATIVE VALUE.

BY

CHARLES HENRY THOMPSON,
Assistant Botanist, Missouri Botanical Garden.

WASHINGTON:
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1912.

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LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry,
Office of the Chief,
Washington, D. C., July 9, 1912.

Sir: I transmit herewith and recommend for publication as Bulletin No. 262 of this Bureau an illustrated manuscript entitled "Ornamental Cacti: Their Culture and Decorative Value." This manuscript was prepared by Mr. Charles Henry Thompson, who carried on the investigations under the direction of the Agriculturist in Charge of Cactus Investigations in the Office of Farm Management. Owing to its strictly horticultural nature, it was deemed wise that it be issued from the Office of Horticultural Investigations and Arlington Farm.

Mr. Thompson has long been in charge of the Succulent Collections of the Missouri Botanical Garden, St. Louis. His position and experience eminently qualify him to discuss this subject.

It is believed that while this bulletin may be of interest to cactus fanciers throughout the country, its particular field of usefulness will prove to be the warmer and drier southwestern regions where these plants are extensively used for decorative gardening.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

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ORNAMENTAL CACTI: THEIR CULTURE AND DECORATIVE VALUE.

INTRODUCTION.

With the exception of a few species of Rhipsalis the cacti are strictly indigenous to the Western Hemisphere. The introduction of these plants into Europe evidently began soon after the discovery of America. The English, Dutch, and Spanish traders, who early carried on a commercial business in the West Indies, South America, Central America, and Mexico, took back to their respective countries many interesting and curious plants then new to the gardens and plant lovers of Europe.

In the earliest published reports of the introduced and cultivated plants of European gardens we frequently find accounts and in many instances illustrations of cacti. Gradually additional plants were introduced, until at the time Linnæus published his Species Plantarum (1753) he recognized 22 species, all of which he included under the generic name of Cactus. They were commonly known as thistles, probably from the spiny character of their protective armor. The smaller, more or less globose forms, were called "melon" thistles, while the taller ones were called "torch" thistles or "candle" thistles. The *Ficus indica*, or Indian fig, and several other species of Opuntia were introduced into the Mediterranean region at a very early date.

From the time of the publication by Linnæus the steady introduction of new plants was continued from the Western Hemisphere into Europe. These importations included many forms of cacti. Miller, in his dictionary, enumerates a number of species distinct from those recognized by Linnæus. Others were described and published from time to time by Haworth, Link and Otto, Salm-Dyck, P. De Candolle, Lemaire, Pfeiffer, and others. The most extensive modern systematic work is Gesammtbeschreibung der Kakteen, by Dr. Karl Schumann.

It was not until within the past half century that any special interest in cacti was manifested in America. A few species, such as the night-blooming cereus (*Cereus grandiflorus* and *Cereus nycticalus*), queen of the night (*Phyllocactus acuminatus*), crab cactus (*Epiphyllum truncatum*), and the rat-tail cactus (*Cereus flagelliformis*), had become favorites as house plants. General collections of this group of the plant world by Dr. George Engelmann laid the foundation for the large collection at the Missouri Botanical Garden at St. Louis, Mo. Similar interest manifested by Dr. Asa Gray added materially to the collection at the Botanical Garden at Cambridge, Mass. As the public became more acquainted with these bizarre forms of vegetation, a livelier interest in them sprang up, and many persons throughout the country began to gather private collections. Fanciers became so numerous that in certain localities clubs or societies were organized among them, where ideas and experiences as to the culture of these plants could be discussed and specimens exchanged. Experience was the high-priced teacher from whom these collectors had to gain their knowledge. Similar organizations were formed in Germany, where amateur collectors were numerous, and also in France and in England. Many articles have been published in the horticultural journals of these countries describing proper methods of propagation and culture, and Mr. Watson, of the Kew Gardens, England, issued a Handbook of Cactus Culture. These helps have disseminated a better general knowledge of methods to be employed, but the soil and climatic conditions of Europe differ so materially from those of various parts of America that their rules are not well adapted to our own special needs.

The growing interest in this group of plants in America and the inadaptability of rules for general gardening in growing them, as well as of rules laid down for their special care by European growers, have created a demand in this country for a work that will include both general and special rules that may be applied to any part of our country. To meet this demand is the writer's purpose in putting forth this bulletin. Naturally, much of the material herein contained is compiled from the experiences of others, but use of it is made only in so far as it agrees with the writer's experience and observations, gained during the years spent in caring for the collection at the Missouri Botanical Garden and in traveling through the native haunts of these plants throughout the Southwestern States and in Mexico, as well as in the examination of many private collections.

PROPAGATION OF CACTI FROM SEEDS.

Most cacti yield seeds abundantly. Ordinarily, few of these seeds germinate and develop into mature plants, because of unfavorable environment. The seeds are usually fertile, however, and when planted under proper conditions a large percentage of them will germinate and with a little care will produce plants in abundance.

The best soil for growing cacti from seed has proved to be a thoroughly decomposed sod mixed with at least its own volume of sand. After these ingredients have been carefully mixed, they are run through a sieve of about $\frac{1}{4}$-inch mesh, which removes any large particles and all superfluous root fibers, making a loose soil which drains very readily. It is not necessary that the soil be very rich in humus, and manured soils should always be avoided because of their undue tendency to hold moisture. They are also a medium for producing germs of decay. An open, drainable soil is the chief requisite for cultivating cacti.

For germinating the seeds an ordinary 4-inch pot is very convenient. New pots are preferable, but old pots may be used with safety if thoroughly sterilized. Porous pots are soon covered with green algæ when left in a moist place for any considerable time. This growth will spread over the surface of the soil in a close blanket which precludes the free access of air and seriously retards the drainage of superfluous water. These algæ will in time grow over the little seedlings that have survived other adverse conditions and will smother them to death. To combat the algæ the pots should be thoroughly sterilized just previous to being used, and to accomplish this object two efficient methods have been found. One method is to bake or burn the pots, so that all life on them or in their pores may be destroyed. The other method is to soak the pots for a time in a weak solution of copper sulphate and then thoroughly wash them in the same solution. If a very strong solution of the copper sulphate is used, some of it will be left in the pores of the clay, and later, when the seedlings are being watered, enough may pass through the soil to injure the tender young plants.

Reasonable care should be exercised in preparing the pot for planting. As a rule the drain hole in the bottom of the pot is too small and is easily clogged. This hole should be enlarged, as thorough drainage must be maintained in growing cacti. The pot should be filled to one-fourth its depth with small bits of broken pots, and on these the prepared soil should be placed and pressed or shaken together firmly but not packed hard. The soil surface is then leveled by the use of a round, flat-faced tamper of a diameter to just fill the pot. The soil should not be packed but should be lightly tamped, with only sufficient effort to produce a smooth, level surface. This surface should be about half an inch from the top of the pot. Over it the seeds are evenly distributed and then covered with a very thin layer of soil, upon which is spread a layer of fine gravel to a depth of about one-fourth of an inch. This layer of gravel is important in many ways. As the pots are later watered with a fine spray, it prevents the surface of the soil from washing and consequently keeps the seeds from being disarranged. It also promotes the free passage of moist air through the spaces between the bits of gravel, which, together with the shading by the gravel, prevents the surface of the soil from becoming dry and baked. It also checks the growth of algæ over the soil surface.

As the seedlings grow they easily force their way through the gravel to the sunlight. For the first few months of their existence, cactus seedlings are but small, globular, balloon-shaped or cylindrical bodies, composed of very thin-walled cells filled to turgidity with water. They are so tender and delicate that they readily "damp off" if subjected to a sudden change from a high to a low temperature. The death rate of seedlings from this cause has been greatly minimized or almost wholly checked by the use of the gravel over the surface of the soil. This layer, with its intervening spaces, acts as a protection from sudden changes in temperature during that period of their growth when the seedlings are most susceptible to injury. By the time they have grown sufficiently large to project beyond the gravel they have become hardier and more robust in structure. Again, the gravel layer is of great value in that it keeps the surface of the soil moist. The little seedlings have exceedingly fine and delicate roots which spread out near the surface of the soil. If this surface is allowed to dry out to the depth of one-eighth of an inch or more, these delicate rootlets will be destroyed and the seedlings will be damaged or killed. In most instances the diminutive plant has not enough food stored up in its body to keep it alive until another set of feeding roots can be produced, and it starves to death. For watering, a vessel should be used that gives a fine, gentle spray, in order to avoid the danger of washing the seeds from their position or of injuring the delicate young seedlings. Watering should be done at least once a day. The temperature of the propagating house or frame should be kept as nearly uniform as possible and should not vary much from 70° F.

The proper time for transplanting the seedlings differs for different genera and species, but they should usually be left in the germination pot until the plant shows at least three or four clusters of spines. By that time the tissues will have become considerably hardened and a very good root system will have been formed. The taller growing species, such as Cereus, will be the first ones ready for transplanting. Mamillaria and kindred genera and plants of similar growth will be the last. The seedlings should be transplanted into a flat sufficiently small for convenient handling, which should be provided with drainage openings in the bottom. It should be filled with the same kind of material and soil as used in the germination pots, the surface to be carefully leveled in the same way. The rows should be about an inch apart, with the same interval between seedlings in the row. After the flat has been filled with the seedlings a thin layer of clean gravel should be placed all over the soil surface and close up around the plants. The flats should then be placed in a perfectly level position, so that the soil will not shift from one side toward the other when watered. Cactus plants are of slow growth and may remain in the flat for several months before being potted. The proper time for potting is when the plants have grown so large that they begin to crowd each other or when the roots of adjacent plants begin to intermingle. In preparing pots for individual plants the same method should be followed as for the preparation of the germination pots, except that a coarse soil may be used to advantage. It is not advisable to begin with pots smaller than 2½ inches, as they dry out too rapidly.

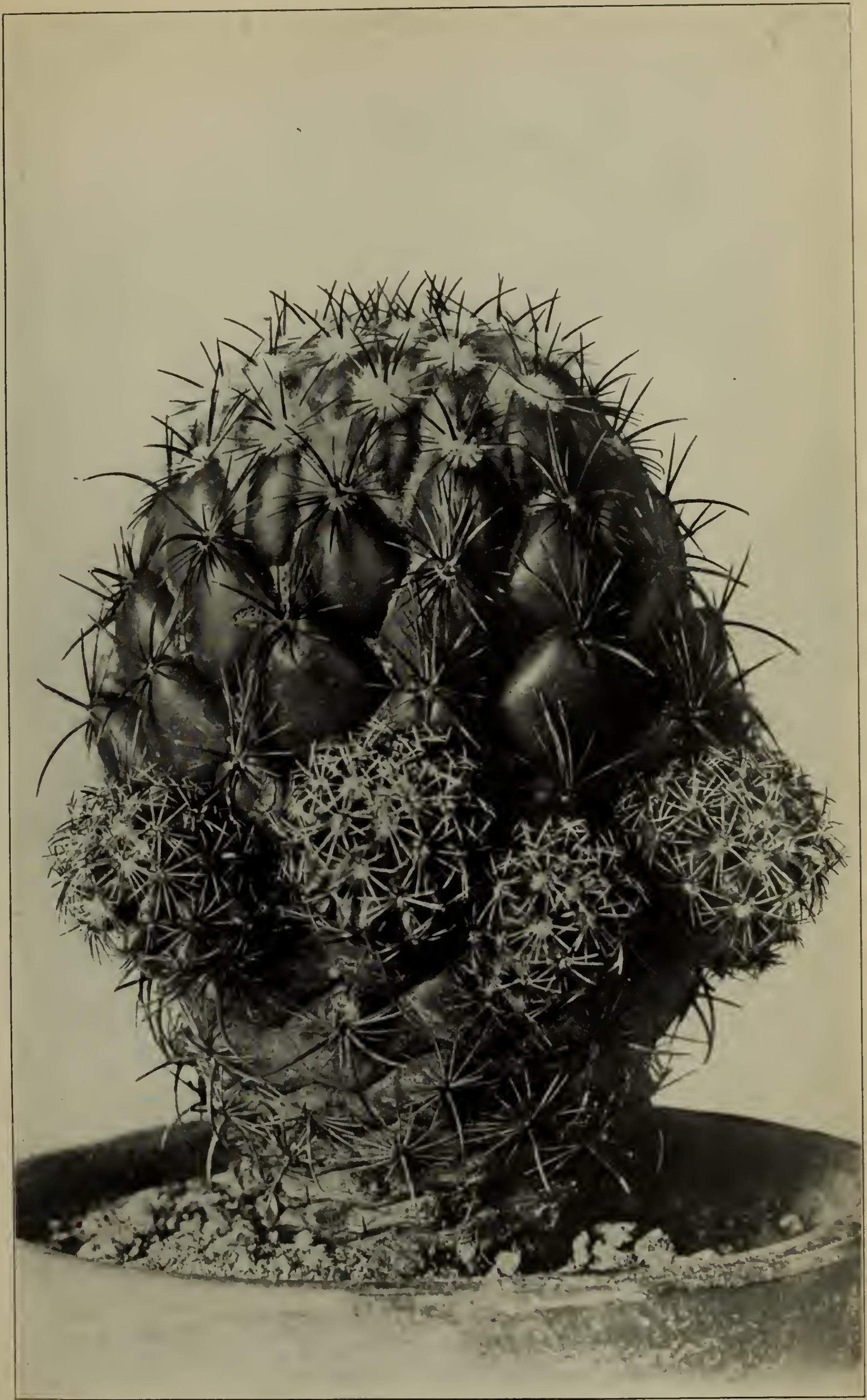
VEGETATIVE PROPAGATION.

Almost all cacti may be readily propagated from cuttings. The plants are so soft in tissue and so filled with water that any bruise or mutilation is likely to be the point of attack of a rot fungus, which quickly destroys them; so, in making the cutting, a clean, sharp knife must be used and a smooth surface left on the cut end. The cutting should then be placed in a dry atmosphere for a day or more, until, by drying, a kind of cuticle has formed over the cut surface. The cutting may then be rooted in sand on a bench or planted directly in pots. In the warmer, drier regions it may be placed directly in the open ground, provided the soil has perfect drainage. In greenhouse culture it is best not to place much of the cutting below the surface of the soil or sand; 1 inch is sufficient for large plants, and less than that for smaller ones, in proportion to the size of the cutting. When the cutting is long and likely to fall over, a stick should be inserted in the soil by its side and the two securely tied together until roots have been formed. When mature plants are shipped in from the field the roots are always more or less injured. It is always best to cut away the roots, let the wounds dry and heal for a time, and then plant them as cuttings. Many of the opuntias are naturally adapted to propagate themselves vegetatively. The stems are readily detached at the joints. These stems fall to the ground and in a short time develop roots and begin to grow as independent plants. Some are adapted for even wider dissemination. The spines which they bear are very sharp and stiff, and are barbed. These spines penetrate the skins of passing animals and cling so tenaciously that the joints bearing them are readily detached from the parent plant and may be carried a considerable distance before being released from their carrier. Once lodged in proper soil under favorable climatic conditions, they soon become new individual plants. In many of the opuntias the fruits are sterile but proliferous. They may be removed and treated as cuttings and will readily produce new plants. Many of the smaller forms, such as Echinocactus, Echinocereus, and Mamillaria, produce branches (Pl. I) which are readily detachable and are easily rooted as cuttings. Some species of Mamillaria have side shoots which are so lightly attached that they break off by a slight touch. Such plants depend almost entirely on vegetative propagation and rarely produce flowers and fruit.

GRAFTING.

Grafting is easily accomplished throughout this whole group of plants. The possibilities of uniting both species and genera seem to be unlimited. For a long time it has been a practice to graft Epiphyllum on Peireskia or some upright, stiff-stemmed Cereus in order to produce a more decorative bush plant. The rat-tail cactus (*Cereus flagelliformis*) is frequently treated in the same manner. Aside from its ornamental possibilities, grafting may be resorted to profitably as a means of propagation. It not infrequently happens that a plant becomes decayed at its base, and when all evidence of decay or disease has been removed there will be so little healthy tissue left that it is next to impossible to get it to grow as a cutting. Such a piece may be grafted on a healthy stock and the plant be preserved, if the growing tip is intact. A cleft graft or a saddle graft is more desirable where either of these can be employed, since they require less work in preparation and give a good large surface for the union of the tissues. The mucilaginous sap that exudes from the cut surface of a cactus plant allows the stock and scion to slip apart very easily, and the parts become disarranged unless proper precaution is used to prevent it. For this purpose the needle-like spines of Peireskia or Opuntia may be used. The two parts are pressed firmly together into the desired position, and then a spine is thrust through the united portions, securely pinning them in that position. No wax is required, but it is best to closely wrap the graft with raffia to exclude the air. The grafted plants are then placed in a warm, moderately moist place until the tissues have become thoroughly knitted together. They should not be placed where they might be subject to drying, for under such conditions the cut surface will be the first to dry, and consequently a perfect union will be prevented.

With small globose or thick plants, such as Mamillaria (Pl. II), Echinocactus, Echinocereus, etc., a different method is preferable. The head of the plant is cut away with a perfectly smooth transverse cut. (Pl. III.) A stock is selected which has about the same diameter as the scion, and it is also given a smooth transverse cut. The two flat surfaces are then pressed firmly together and held in place by tying them together with a cotton or other soft cord. It is quite essential that clean instruments be used to prevent



MAMILLARIA CORNIFERA, MISSOURI BOTANICAL GARDEN, ST. LOUIS, 1899.

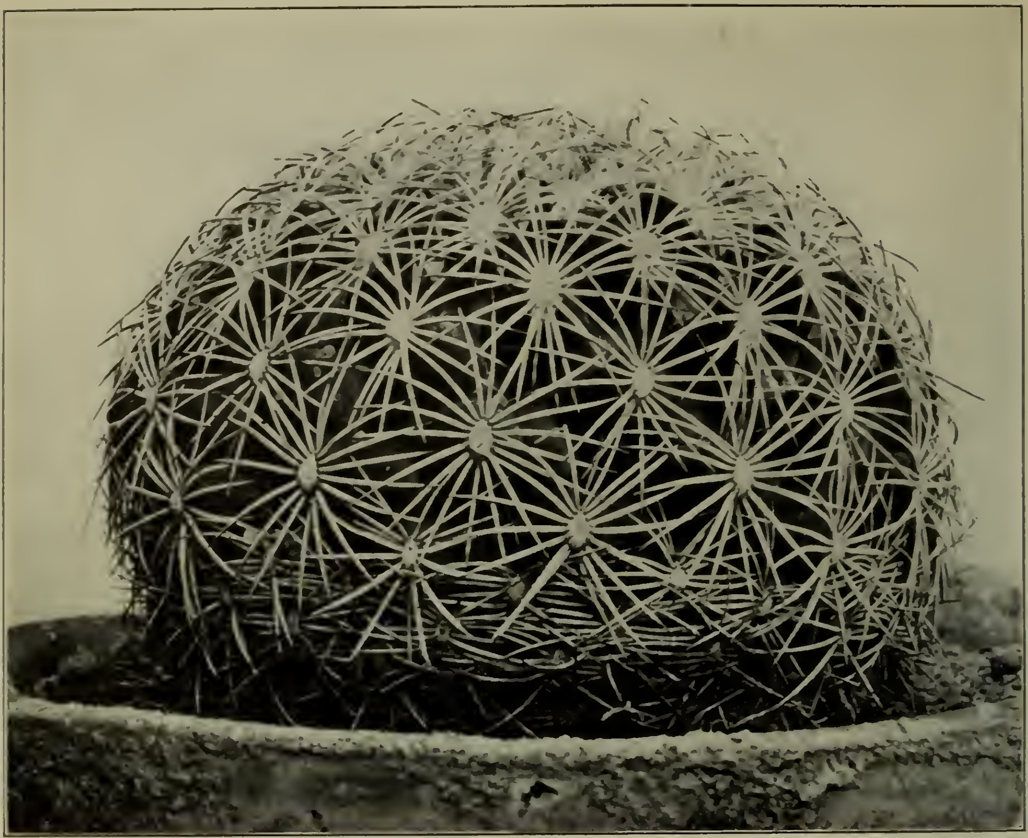


FIG. 1.—MAMILLARIA COMPACTA, MISSOURI BOTANICAL GARDEN, ST. LOUIS, 1911.

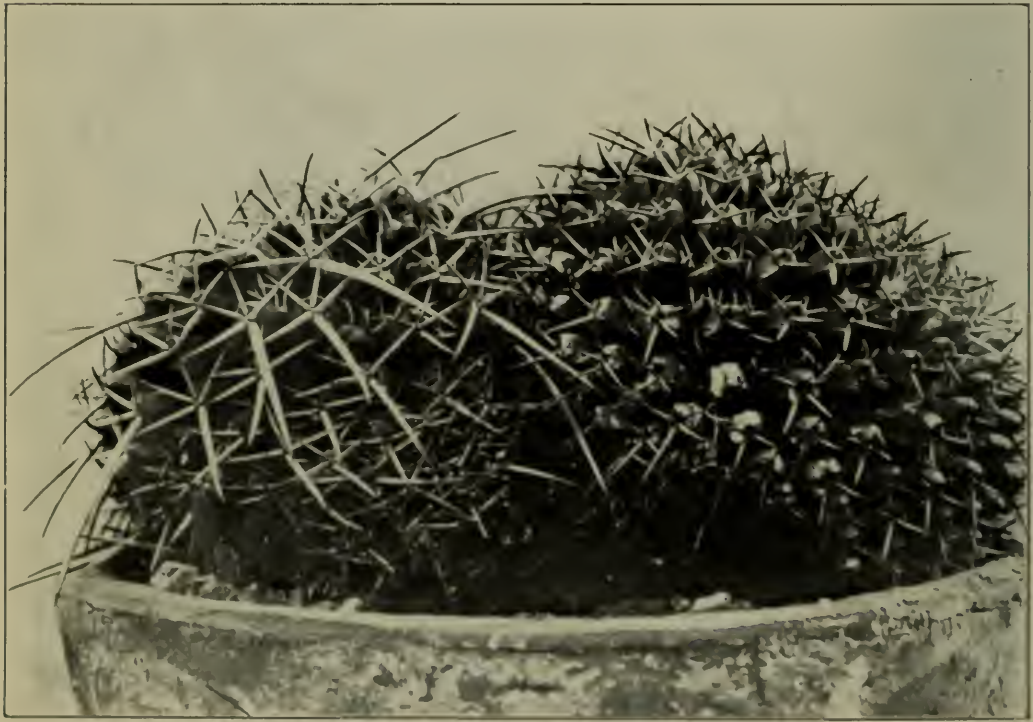


FIG. 2.—MAMILLARIA PYRRHOCEPHALA, MISSOURI BOTANICAL GARDEN, ST. LOUIS, 1910.

the inoculation of the plant with disease germs. A number of the upright-growing species of Cereus have been used successfully for stocks, and there seems to be no limit to the number of species that may be used. It has been found, however, that some are better than others for the purpose. When it is desired to have the scion a foot or more high, good stocks may be obtained from *Cereus stellatus*, *C. serpentinus*, and other species of similar habits of growth. These stocks are preferable for use in grafting *Cereus flagelliformis* and species of Epiphyllum and Rhipsalis, which normally grow in a pendent position. Where only short stocks are desired the above may be used, and also *Cereus nycticalus*, *C. tortuosus*, *C. bomplandii*, *C. macdonaldiae*, and *C. grandiflorus*. These latter plants are weak stemmed when allowed to grow tall; hence, they can not be used for high grafts unless supported by a stake of some kind. All these species are readily grown from cuttings, which should be somewhat longer than the stock is to be. When the cutting is thoroughly rooted it should be potted and kept in good growing condition until a new root system has formed. It will then be ready to receive the scion after having been cut back to the desired height.

CULTURE.

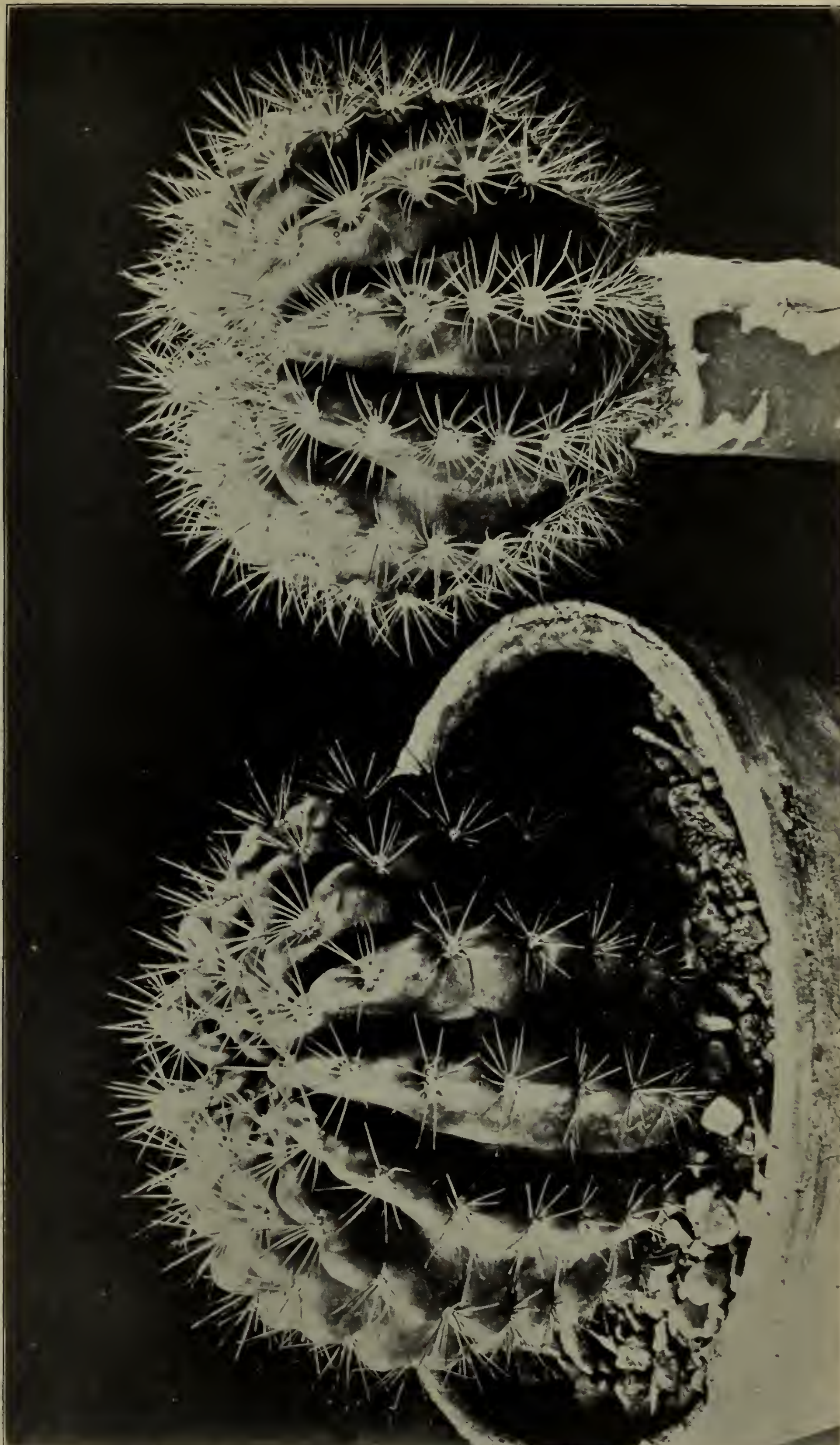
Cacti thrive from southern Canada to far down in South America. Between these extreme points there is scarcely any combination of atmospheric and soil conditions that does not support one or more species of the family. They are found near the seashore in the Tropics, as well as high up on the mountains, where in winter they are subjected to severe frosts. They are most abundant, however, in the higher semiarid tablelands. With these facts in mind, it is clear that when collected they can not all be treated alike, but must be grouped according to the conditions under which the individuals grow in their native haunts, and each group must receive a different treatment to accord therewith.

Epiphyllum, Rhipsalis, a few species of Phyllocactus, and some species of Cereus are epiphytic in their tropical homes and should be given like treatment in conservatories. They should be grown under practically the same warm, moist atmospheric conditions as are tropical orchids, which may be grown in baskets of peat and moss, or be trained on blocks or stumps, or on walls, wherever the roots have opportunity to penetrate a moisture-laden medium. Most species of Phyllocactus and of the climbing species of Cereus should be grown in orchidlike conditions of temperature and humidity but in very loose, moderately rich soil. For this purpose a mixture of loam, sand, and an abundance of thoroughly decomposed leaf mold makes an excellent soil. By far the greater number of species of cacti are terrestrial in their habits and are indigenous to warm, semi-arid regions. The annual rainfall in these regions is very slight and continues for only a brief period. It is difficult to reproduce such conditions in our northern climate, and it is found that cacti can best be grown here by minimizing the action of our abundant rains by having the plants placed in a thoroughly well-drained situation. It is equally difficult to reproduce the conditions in our conservatories, where they are heated artificially, because of the drying effect of the heat. This condition may be largely counteracted by a judicious watering of the soil about the plants. For this group of plants it is not necessary that the soil be very rich, but it is essential that it be very open and thoroughly drained.

In repotting older plants it is best to disturb the roots as little as possible. Enlarge the drain opening in the bottom of the pot and place over it broken pots or other coarse material to not less than 1 inch in depth, to insure perfect, uniform drainage. Over the coarse material put a layer of soil. Remove the plant to be repotted by inverting the pot and gently tapping its rim on the edge of a bench or some such solid structure. The whole body of dirt will come out in a lump. Remove any bits of broken pots that may be attached to the bottom, but leave the soil in place about the roots. The surface soil should be removed if it shows any evidence of containing algæ or fungous growths. Place this ball of dirt and roots in the next pot and pack fresh soil about it, leaving sufficient space at the top to receive water. In conservatories pots are apt to become coated with green algæ, and old pots especially so, because the spores of the algæ are likely to remain in the pores of a pot from its previous use. Old pots should be thoroughly sterilized, as heretofore explained for the germination pots. After the plant is potted the surface of the soil should be covered with fine gravel to a depth of at least half an inch.

The soil about the plants should never be allowed to become absolutely dry for any great length of time or the roots will be seriously injured; on the other hand, it must not be kept saturated, but should be kept moist at all times. Any superfluous water standing about the base of the plant or in the soil about its roots is a serious menace, since it acts as a medium through which germs of rot enter the plant and soon destroy it. Cactus plants contain so much liquid that decay works very rapidly through them. When decay is once started it is difficult to save the plant; hence, the urgent necessity for having thorough drainage below the plant and a thoroughly drainable soil. Failures in the growing of cacti are undoubtedly due more to the neglect of this precaution than to all other causes combined.

Cacti do not require to be pruned beyond the removal of dead portions and to keep the plants in shape within the space allotted to



ECHINOPSIS CALOCHLORA, SHOWING THE DEVELOPMENT OF MORE SPINES IN THE GRAFTED PLANT THAN IN ITS PARENT. THE GRAFTED PLANT IS AN OFFSET FROM THE OTHER. MISSOURI BOTANICAL GARDEN, ST. LOUIS, 1907.

OPUNTIA LEUCOTRICHA, NATURAL LANDSCAPE AT DURANGO, MEXICO, 1909.

(Photographed by Griffiths.)

them. Pruning may be done at any time, but preferably when the atmosphere is dry, so that the cut surface may dry and heal quickly.

In conservatories during the colder season, in order that the air may be sufficiently dry, a temperature ranging from 60° to 70° F. should be maintained. A temperature lower than 60° for any considerable length of time would hold the moisture about the plants too long and invite decay. During the warmer season, if the plants are kept in the house it is necessary that it be kept fully ventilated. The aim is to have at all times a dry atmosphere and a moderately moist well-drained soil. If the plants are placed in open ground during the summer months and their pots plunged in the beds, these beds likewise must be thoroughly drained. In placing a collection out of doors as a permanent planting, a situation should be selected, if possible, where the ground slopes sufficiently to insure perfect drainage. If natural drainage is impossible, a system of drain tiles should be placed throughout the area to be planted and the soil above the tiles should be made loose and porous by the abundant addition of gravel and sand. Out-of-door planting is preferably done during the dry season, so that the cut surfaces or any injured portions of the plants will dry over quickly and be less easily infected with rot.

DISEASES.

The one disease from which cacti suffer more than any other is rot. The plant body is so saturated with water that it forms an excellent medium for the growth of this disease. It is liable to attack the plant at any point where the germs have opportunity to reach the interior. Any cut or bruised place presents the most favorable point for infection, from which the disease rapidly spreads and destroys the plant. Water dripping on a plant for even a short time may induce infection. By far the greater number of plants receive the infection through their bases or roots, whence it works upward through the center of the plant. By the time it has reached the surface the plants are usually too far gone to be saved. If the disease is detected before it has reached the crowns of the plants, they may be saved by cutting away all the diseased portions and then grafting the crowns on some healthy stock. Otherwise, it is best to remove the plants at once and burn them. The soil in which they were potted and also the pots, if to be used again, should be sterilized, so that other plants may not be infected from them.

Another disease more common to species of Mamillaria and to a less extent found on Echinocactus and Cereus makes its first appearance as a small, light orange-colored spot on any portion of the plant surface, usually starting at a pulvinus, which seems to be the point at which the infecting germ enters. This spot steadily grows until the plant is totally destroyed. The disease travels inward, toward the center of the plant, following fibrovascular bundles. The colored tissue readily separates from the healthier portion of the plant and is easily removed, but this merely checks its ravages for a time. The disease penetrates every portion of the plant and in time will make itself manifest again in other orange-colored spots on the surface. It is a contagious disease, and the only hope for saving a collection of plants is to destroy all the infected individuals, preferably by burning them. Many remedies for this disease have been applied, but without success.

INSECT PESTS.

The Bureau of Entomology of this Department has investigated cactus insects extensively. The results of this work appear in a bulletin of that Bureau (No. 113), which may be had upon application.

ECONOMIC VALUE OF CACTI.

MEDICINES.

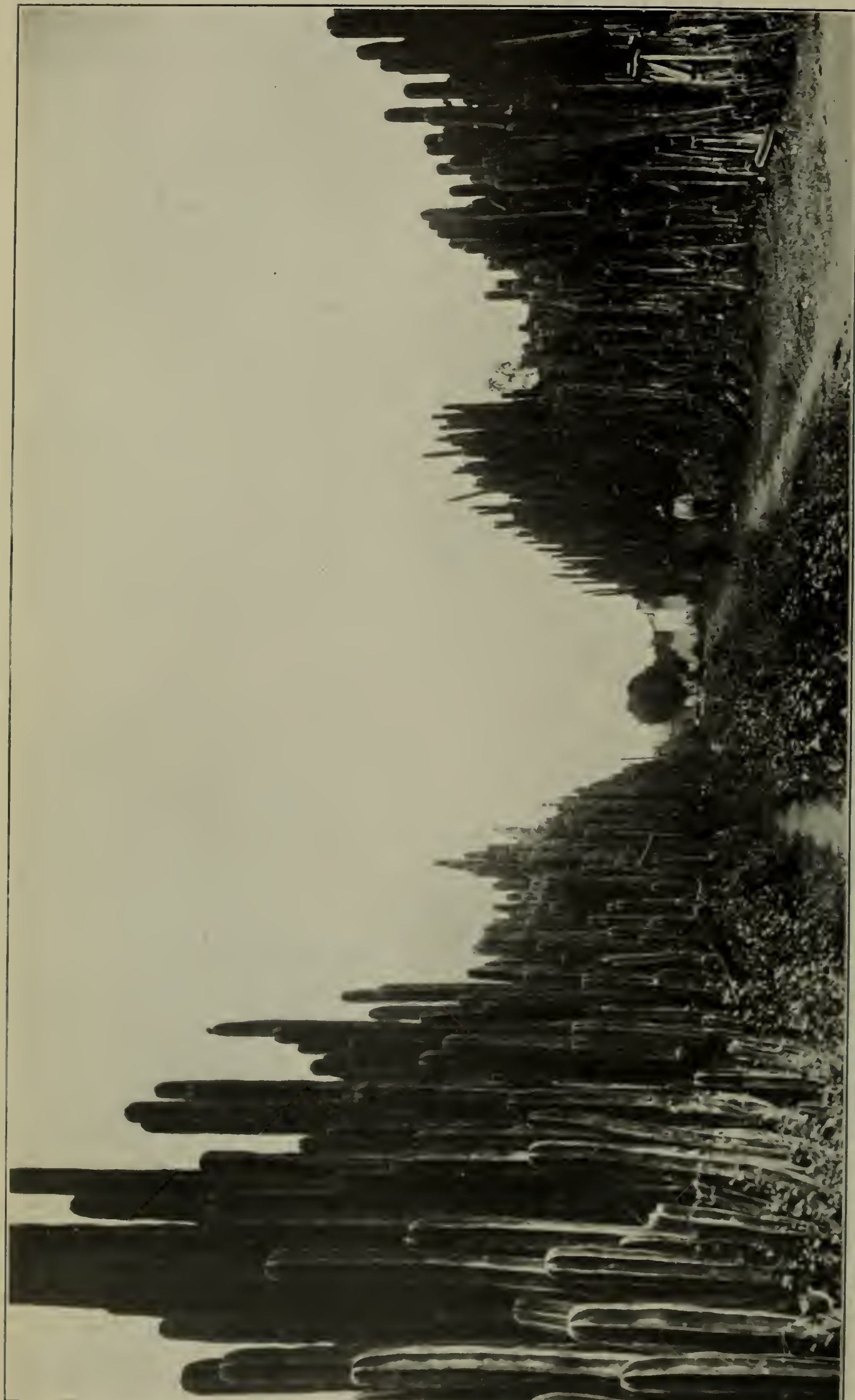
To a limited extent *Cereus grandiflorus* and *C. nycticalus* have been used in the preparation of certain compounds. Other cacti are known to contain characteristic alkaloids which from their peculiar action on the human system may yet prove of value in treating special disorders. Most notable of these forms is the so-called piote bean or mescal button, also known as the dumpling cactus (*Lophophora williamsii* and *L. lewinii*). Since remote times the aborigines of America have used this plant in certain of their religious rites. When the plants are eaten raw, dried or fresh, with water, the optic nerve is so affected that by closing the eyes the user is made to see visions illuminated in the brightest of colors. An alkaloid of this plant has been separated from it and found to contain the same properties. It is not impossible that in time it may be found of value in the treatment of certain ocular disorders. However, no member of the family seems as yet to have yielded a drug that has been used to any considerable extent as a medicine.

GARDEN VEGETABLE.

Among the poorer classes of Mexico the very tender growths of Opuntia are eaten raw, made into a sort of salad, or are cooked, as may be desired. There is little to commend this cactus to those having access to the common and more palatable vegetables of the garden.

FIG. 1.—OPUNTIA LEUCOTRICHA, A. S. WHITE PARK, RIVERSIDE, CAL., 1909.

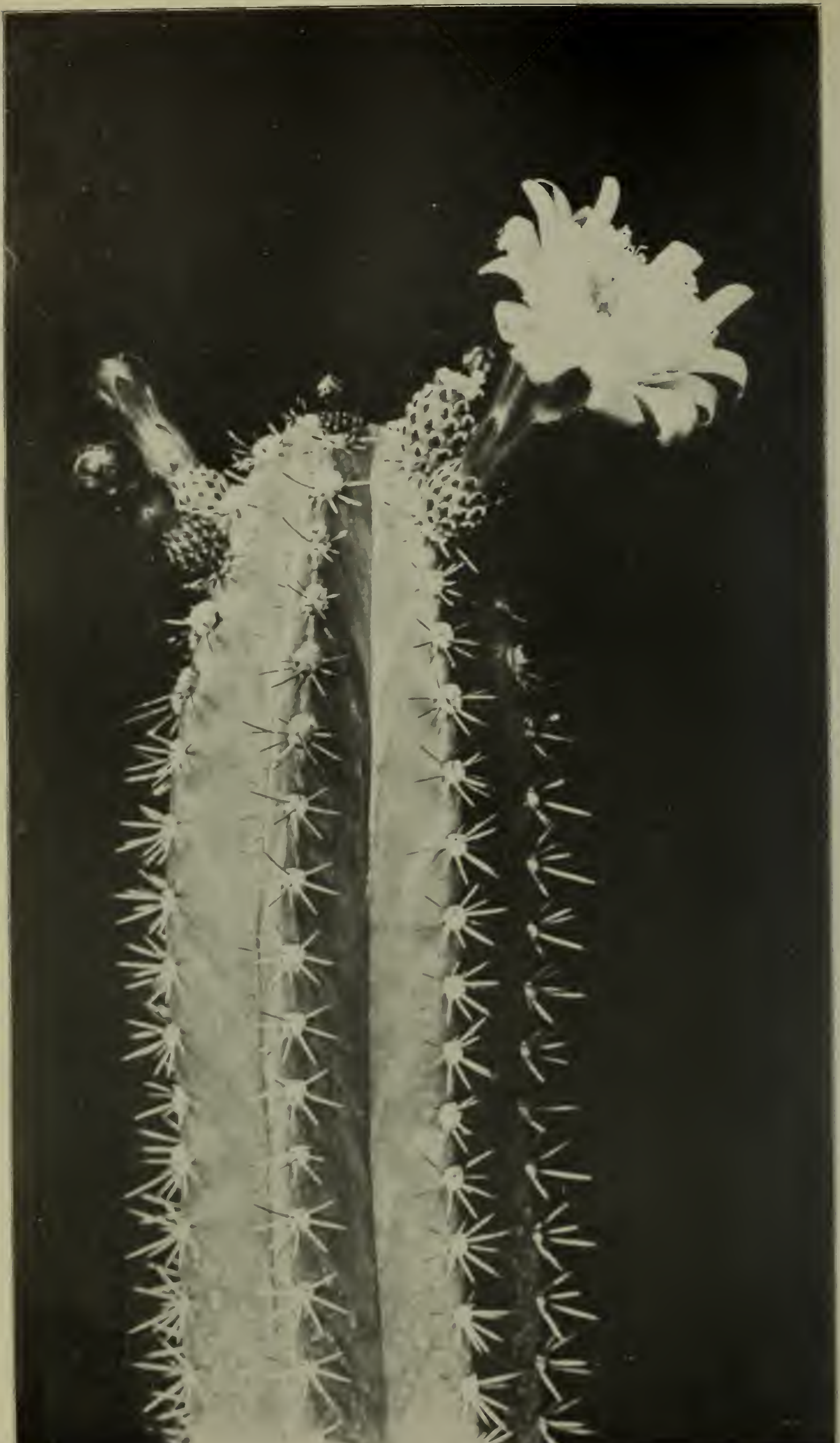
FIG. 2.—OPUNTIA MICRODASYS (IN FOREGROUND), A. S. WHITE PARK, RIVERSIDE, CAL., 1909.



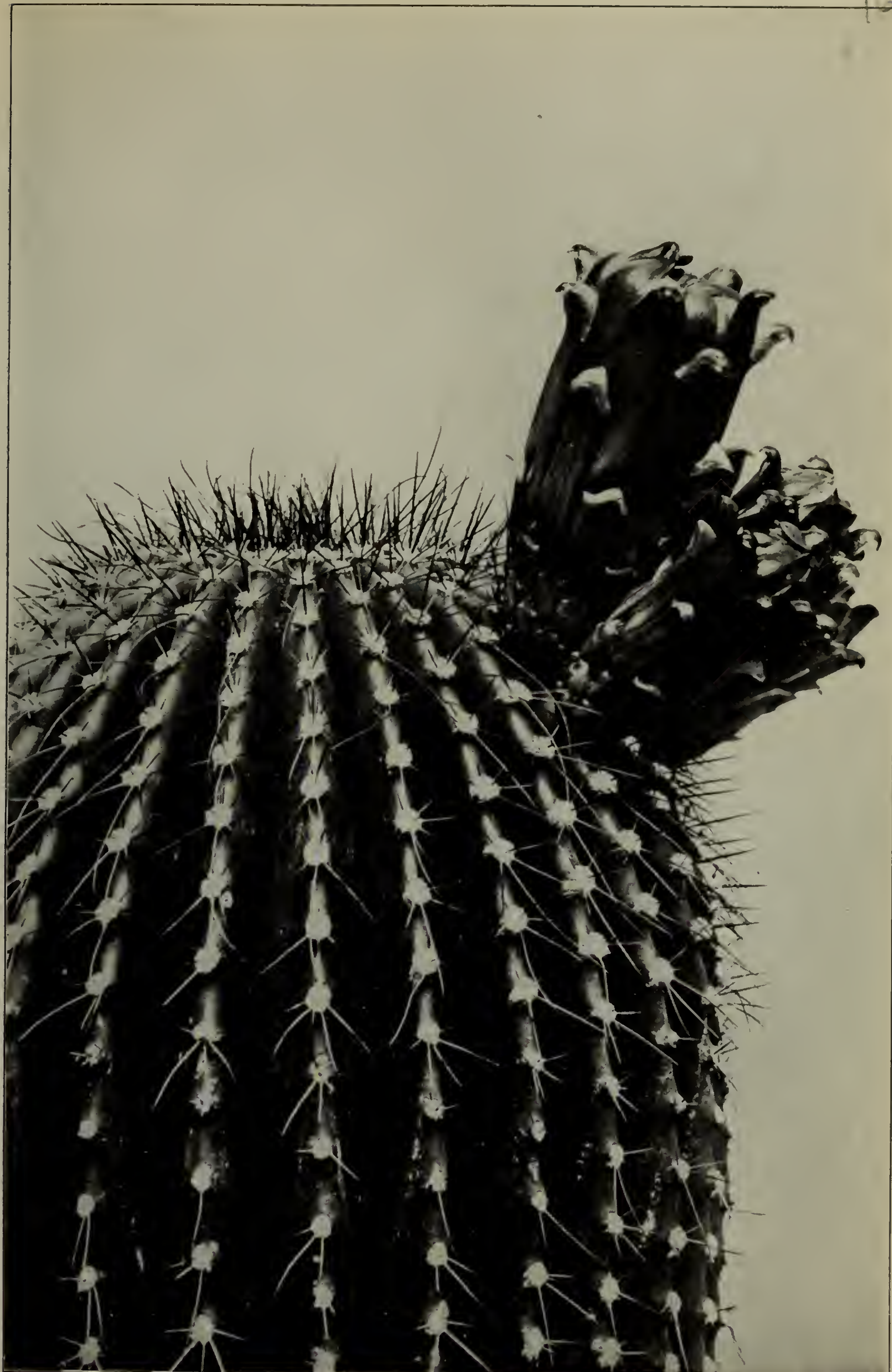
CEREUS MARGINATUS USED AS A HEDGE-PLANT, ENCARNACION, JALISCO, MEXICO.

(Photographed by Griffiths.)

CEREUS TORTUOSUS, MISSOURI BOTANICAL GARDEN, ST. LOUIS, 1898.



CEREUS EBURNEUS. MISSOURI BOTANICAL GARDEN, ST. LOUIS, 1905.



PILOCEREUS POLYLOPHUS (CEREUS NICKELSII), MISSOURI BOTANICAL GARDEN, ST. LOUIS, 1905.

(Natural size.)

ECHINOPSIS GEMMATA, MISSOURI BOTANICAL GARDEN, ST. LOUIS, 1898.

FRUITS.

The fruits of a great many species of cacti are very agreeable to the taste, as well as refreshing and nourishing. This is especially true of many species of Opuntia known as tunas. (Pls. IV and V.) In certain parts of Mexico the tuna forms a considerable part of the diet of the poorer natives. In many places it is grown for the market and finds ready sale among all classes. The outer part is peeled away in the same manner as in paring an apple or peach for consumption raw. The inner pulp, containing the seeds, not only possesses a pleasant flavor, but it also creates the impression of being cool even in the hottest weather. Forms of tuna have as wide a range in color, flavor, and size as many of our northern fruits.

Numerous smaller fruits are gathered from wild plants, either for home consumption or for sale in the market. Among these may be mentioned the small, globose, red fruit of *Cereus geometrizans* and kindred species, which is very sweet. It is called "garambullo" by the Mexicans. A number of species of Mamillaria produce an abundance of smaller club-shaped red fruits which have a very pleasant, sweet taste, combined with a slightly acid tang, and are eaten by the natives, who call them "chilitos." The above-mentioned fruits, and also that of *Cereus giganteus*, are quite commonly used in the preparation of preserves, jams, and cakes of somewhat jellylike consistency. These preparations represent some of the choice delicacies of the natives and are to be found on sale in the markets and on the streets of Mexican cities. The fleshy interior of certain species of Echinocactus is used in the production of the so-called cactus candy. The flesh is cut into layers and cured in sugar sirup and allowed to dry, similar to the manner in which citron is prepared for market. The cactus flesh merely forms a foundation, adding perhaps a little flavor.

WOOD.

For the most part all cactus plants are composed of soft, water-laden tissue, but the axis of the plant is composed of a woody core, which in some species makes a considerable development, especially in Opuntia and the large species of Cereus. This woody portion is always more or less porous and usually of an open, lacelike structure, so that it is of little value as compared with other woods. Nevertheless, it is used to some extent in the manufacture of ornaments and rustic work, but more for its curious structure than for any real value the wood may possess. Some of the cylindrical forms of Opuntia yield rather grotesque and ornamental walking canes, as do also a few of the slender-growing columnar species of Cereus. These forms also furnish wood for rustic picture frames, ornamental pincushions, trays, inkwell stands, and the like. To a limited extent the wood of the taller growing species forms material in the shape of poles for the construction of fences and temporary huts.

HEDGES.

Because of their animal-resisting armor of spines, combined with their habit of growth, certain species of cacti are naturally adapted for use as efficient hedge plants wherever they grow in the open throughout the year. The one species most commonly used in Mexico for this purpose is the organo (*Cereus marginatus*), Pl. VI, so called because of its fancied resemblance to the pipe of an organ. It branches freely from the base near the surface of the ground, and these branches immediately assume an upright habit of growth. Growing closely together they soon produce an impenetrable barrier. Its habits of growth recommend it, since there are scarcely any branches above the base and these never spread and cover any great area, thus making a compact, dense, and comparatively narrow hedge. *Cereus stellatus* and *C. weberi* are also used in the regions where they are abundant as native plants, but they have the disadvantage of making a thicker and more open hedge and consequently cover more ground. Where narrowness of the hedge is of minor importance, many of the taller growing species of Opuntia make an equally serviceable barrier and are at all times decorative, especially when bearing an abundance of flowers and fruits.

DECORATIVE VALUE OF CACTI.

It is not intended to convey the idea that cacti, as a whole, can hope to rival many other groups of plants in gorgeous display. For the most part they lack the foliage that lends so much to the value of other plants, and in most instances the flowers, when present, are either too small, too few (Pls. VII, VIII, IX, and X), or too short lived to be considered of any great worth. In some of the climbing species of Cereus and in Phyllocactus the flowers attain a very considerable size, and their waxlike texture and pure whiteness or delicately tinted red, pink, or cream colors present a combination that always calls forth exclamations of wonder and pleasant surprise. Many forms bloom at night, and their flowers are always white and to a slight degree pleasantly fragrant. The flowers are usually produced in periods, each period lasting from one to three or four days. At such a period the plants, if mature and vigorous, will bear a large number of flower buds, which open in the evening after sunset and close with the approach of strong morning light, never again to open. The following night other buds will bloom, and so on until in a few days all will have passed the blooming period, which, after an interval of time, will recur. In our northern conservatories there are usually three or four such periods during the summer season, averaging about four or five weeks apart. On these occasions the display of large white flowers in abundance in the moonlight is a wonderful sight. Most of the species of Echinocereus produce comparatively large showy flowers in a crown about the ends of the branches. They are very attractive in their highly colored (yellow, orange, red, and purple) waxy flowers, but they do not respond so readily to cultivation as many others, especially in greenhouses. Some species of Echinopsis also produce flowers in abundance for a period of a few days. These are trumpet shaped, upright, about 8 inches long, forming a crown about the top of the plant. They range in color from pure white to pale yellow or rosy pink.

The chief attractiveness and beauty in cacti as a group[1] is the remarkable symmetry of growth in the individual plants. The columnar, and most of the genera of smaller cylindrical or globular forms, have clean-cut, longitudinal, parallel angles, ribs, or wings, and located on them at regular intervals are the buds, or pulvini, which bear the spines and flowers, and from which side branches may be developed. The coloring of the epidermis of the plant is frequently very attractive. While in most species this color is some shade of green, many specimens are coated over to a greater or lesser extent with white or bluish glaucousness. In some species the surface is dotted over with very small bunches of velvety white hair, as in *Echinocactus myriostigma, E. ornatus*, and *E. capricornus*. Other species are mottled with purple, which in the young growths of *E. ingens* is arranged in transverse bands, alternating with bright green. The coloring of the spines, too, is often exceedingly attractive, especially in the younger growths. It ranges from pure white to amber, yellow, red, and black. Frequently some of these colors are combined on one spine in either longitudinal stripes or transverse bands, and the perfectly uniform variegation is very striking. The form, structure, and arrangement of the spines are in most instances remarkable and show a wonderful adherence to a definite plan of symmetrical arrangement. In certain species some of the spines have a structure of soft and hard transverse layers from base to tip, giving an uneven though uniformly wavy surface much like that of a goat's horn. The larger number of spines are straight or only slightly curved; others have the end curved in the form of a fishhook. Nearly all of them are rather stiff, but some are soft and featherlike

[1] See Safford, W. E., "Cactaceæ of Northeastern and Central Mexico, Together with a Synopsis of the Principal Mexican Genera," in Annual Report, Smithsonian Institution, 1908, pp. 525-563, in which are illustrations of many species of Cactaceæ.

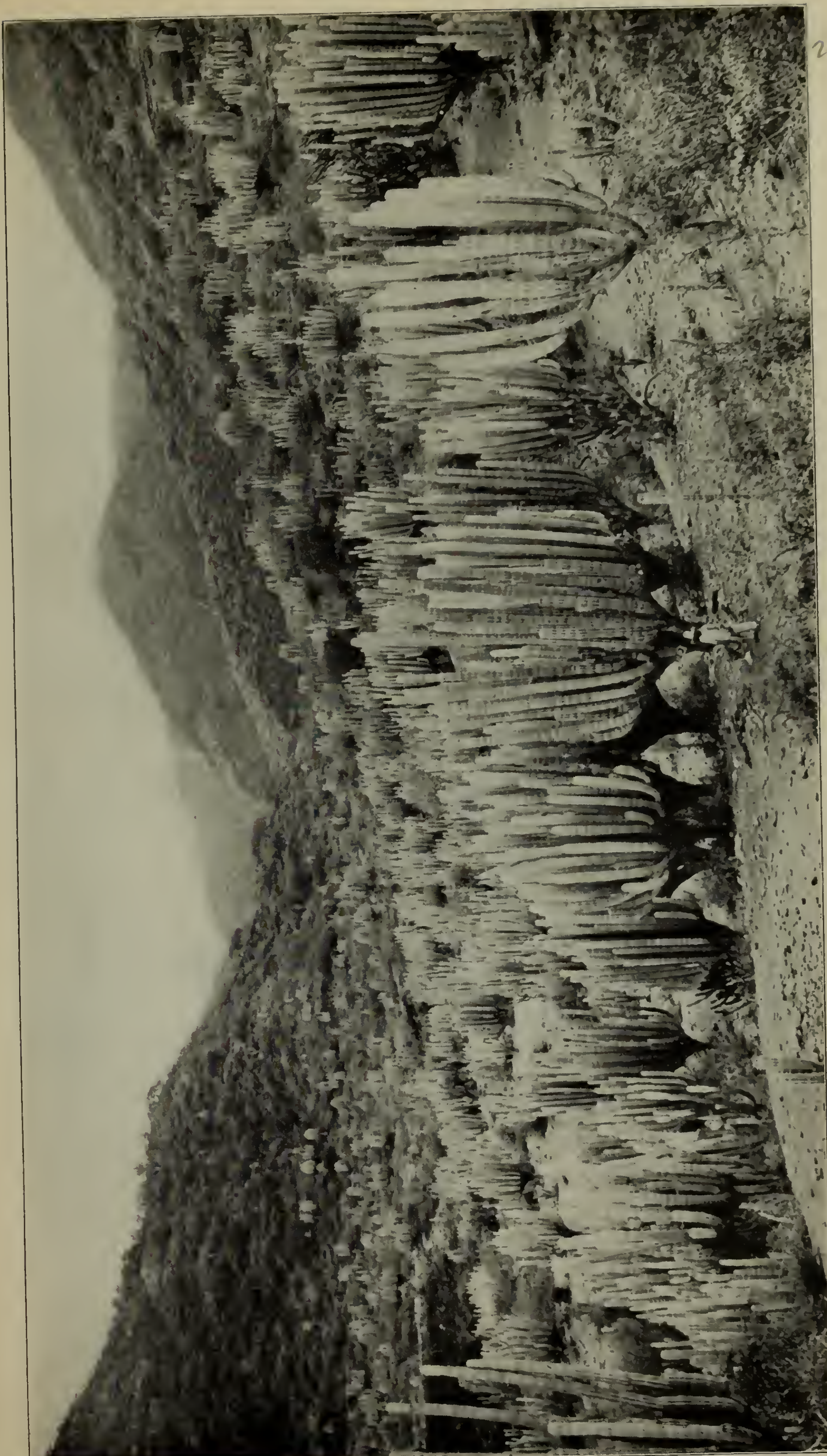
in structure and others are thin, flat, paperlike, and flexible. Again, in some species the spines are entirely absent. Mamillaria (Pls. I and II) and some groups of Echinocactus have all the variations of characters already described, but differ materially in body structure. In them the ribs or angles have entirely disappeared and are represented by rows of tubercles or mammæ, each bearing at its summit a cluster of spines. In this group the tubercles are not arranged in longitudinal rows, but are geometrically tesselated over the plant surface, so arranged as to form spirals running in both directions about the plant.

A remarkable and interesting feature is the regularity in number with which these spiral rows appear. As a rule they fall into the numbers 5, 8, 13, 21, 34, 55, and possibly higher numbers. For instance, if it is found that there are 13 parallel rows of tubercles running obliquely around the plant in one direction, there will be either 8 or 21 such parallel rows running obliquely around it in the other direction. Whatever the number of rows counted in one direction, the number counted in the opposite direction will be the one either preceding or following it in the series. Exceptions to this rule are rare, and when one is noted the numbers are usually found to be the doubles of two adjacent numbers in the above series, as 10, 16, 26, 42, and so on. Another interesting fact is that each number in the series is the sum of the two immediately preceding it.

Symmetry is the greatest attraction in this group of plants. Monstrosities are, however, not infrequent in the family, usually assuming a cristate or cockscomb form of growth. These forms are so odd in appearance that they are frequently sought after, and it is not uncommon to find them represented in the collections of amateurs. Their very grotesqueness commends them to the consideration of collectors.

SINGLE PLANT DISPLAYS.

Each individual plant has an attraction of its own. (Pls. XI, XII, and XIII.) Whether it be the symmetrical order of its trunk, its color, its versicolored or versiform spines, or a combination of all these features, supplemented in its proper seasons by the production of flowers and fruits, each normal, healthy plant is well worthy of consideration as an individual specimen. Their adaptability is such as to commend them for situations where many other plants could not exist. They do not require frequent repotting and replenishing of soil, and subsist best on a minimum of water, so that if necessarily neglected for a time they do not materially suffer. A single plant is well worth the little trouble required for its keeping. It occupies a very small amount of space in comparison to its weight, which is an advantage in many instances.



CEREUS WEBERI PREDOMINATING IN NATURAL LANDSCAPE AT TOMELLIN, MEXICO, 1909.

(Photographed by Griffiths.)

CEREUS STELLATUS (IN FOREGROUND), NATURAL LANDSCAPE AT TOMELLIN, MEXICO, 1909.

(Photographed by Griffiths.)

FIG. 1.—PILOCEREUS FULVICEPS, TEHUACAN, MEXICO, 1910.

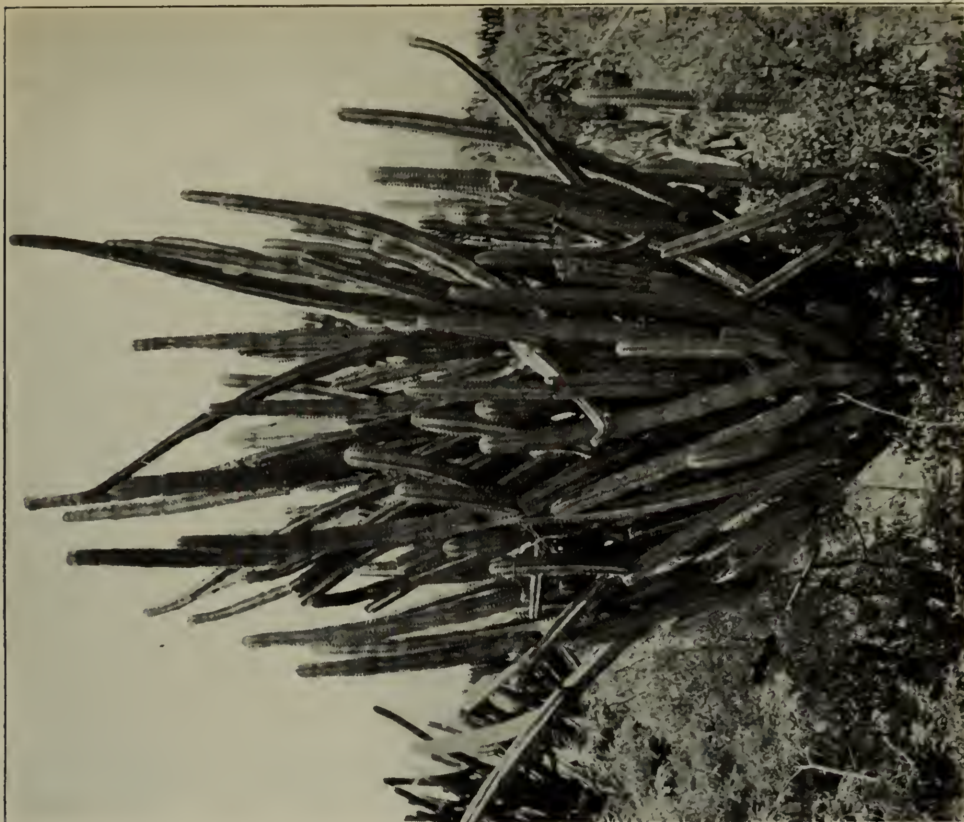


FIG. 2.—CEREUS EBURNEUS, TOMELLIN, MEXICO, 1910.

ORNAMENTAL PLANTING OF CACTI, MISSOURI BOTANICAL GARDEN, ST. LOUIS, 1905.

(Photographed by Griffiths.)

A pretty and interesting display may be had by arranging the plants in groups on benches, on window sills, or on bracket shelves on either side of a window. Pots of individual plants of various sizes lend themselves very readily to artistic arrangement. In any banking effect the larger and taller ones should be placed in the background, and the rest graded down to the front according to the size of the plants. Should the plants be too uniform in size for such arrangement, those in the background may be elevated on inverted pots or blocks of wood of suitable height.

GROUPINGS.

Pleasing effects may be obtained by placing a number of plants in one pot or small box. For this purpose it is necessary to choose small plants, preferably the low-growing globular or short, cylindrical forms of Mamillaria and Echinocactus. With a little care in the selection of perfectly symmetrical plants with well-developed spines, and some taste in arrangement, a compact group may be built up which will make an excellent ornament for the table or window and can easily be moved to any place desired. In the diversity of designs which may be followed there is a wide range of possibilities, ornamental pots or boxes lending an artistic touch to the composition.

PLANTINGS IN OPEN GROUND.

Cactus roots naturally penetrate deep into the soil, and at the same time some of them spread widely from the plant stem. This tendency is necessarily limited in potted plants, and the plant does not receive the nourishment or water that it should have; hence it is always better to place them in the open ground if possible. In the Northern States it is necessary that the plants be protected from frost in winter. In such localities a room in a greenhouse may be set aside for this group of plants, and beds made in the native earth to receive them. Here they may be placed close together, as they shade one another very little and do not have the abundant foliage of other plants. The roots may intermingle, but to no greatly detrimental extent, since the main feeding roots penetrate deep down into the soil. Furthermore, cactus plants need comparatively little nourishment, and it would require a long period of time to exhaust the soil. An effective arrangement is to build up rocks and soil, leaving the surface more or less covered with rocks, making a genuine rockery. (Pl. XVII.) This treatment lends a natural aspect to the surroundings and furthermore adds a greater degree of drainage, so necessary to cacti at all times.

Cacti may be used as good decorative plants in outdoor beds, planted either temporarily or permanently. (Pl. XVI.) Where one has a number of individual potted plants that have to be housed for protection in the winter season, it is always desirable that they be placed in the open during the summer months. They should be taken out as soon as all danger from frost is past and left till danger from frost threatens in the fall. The beds should be either high or on a sloping surface, to insure thorough drainage about the plants. With such plants it is better to leave them potted and plunge the pots into the soil. (Pls. XIV and XV.) Plants thus exposed to the sunshine and rain during the summer months will do far better than those kept indoors and given house treatment. A judicious arranging of the plants in such beds will have an attractive and pleasing effect. Where a large number of individuals of a few species are available, some artistic designs may be worked out in these summer beds.

In the warmer southern or southwestern portions of our country a very large number of cacti will thrive out of doors the year round. In such localities the possibilities for bed planting have a much wider range. More area may be given to them there than would be necessary in the conservatories of the North. They will require greater space, because plants that grow in the open thrive much better than potted ones and consequently branch and spread over a greater area. In such localities, with plenty of room, it is possible to produce decidedly realistic landscape effects. Especially is this true in parks (Pls. XVI and XVII), where the semiarid character of the native home of the cacti may be reproduced with wonderful accuracy. Winding paths may be laid out through the tract and the borders planted in irregular groups, so that the effect will change as one passes along any of the walks.

CULTIVATED FORMS OF CACTI.

The following list contains the names of most of the cacti now in cultivation in the United States. Many other forms are to be found in collections but are not at all common. They are grouped with reference to their habits of growth. Measurements, where given, refer to mature plants and are only approximate. The list, arranged as it is with reference to size, will serve as a guide to prospective purchasers in dealing with collectors and traders.

COLUMNAR FORMS OF CACTI.

Tall—over 6 feet in height.

Cephalocereus:
- chrysomalus.
- senilis.

Cereus:
- azureus.
- baumannii.
- chiotilla.
- coerulescens.
- columnaris.
- dumortieri.
- eburneus.

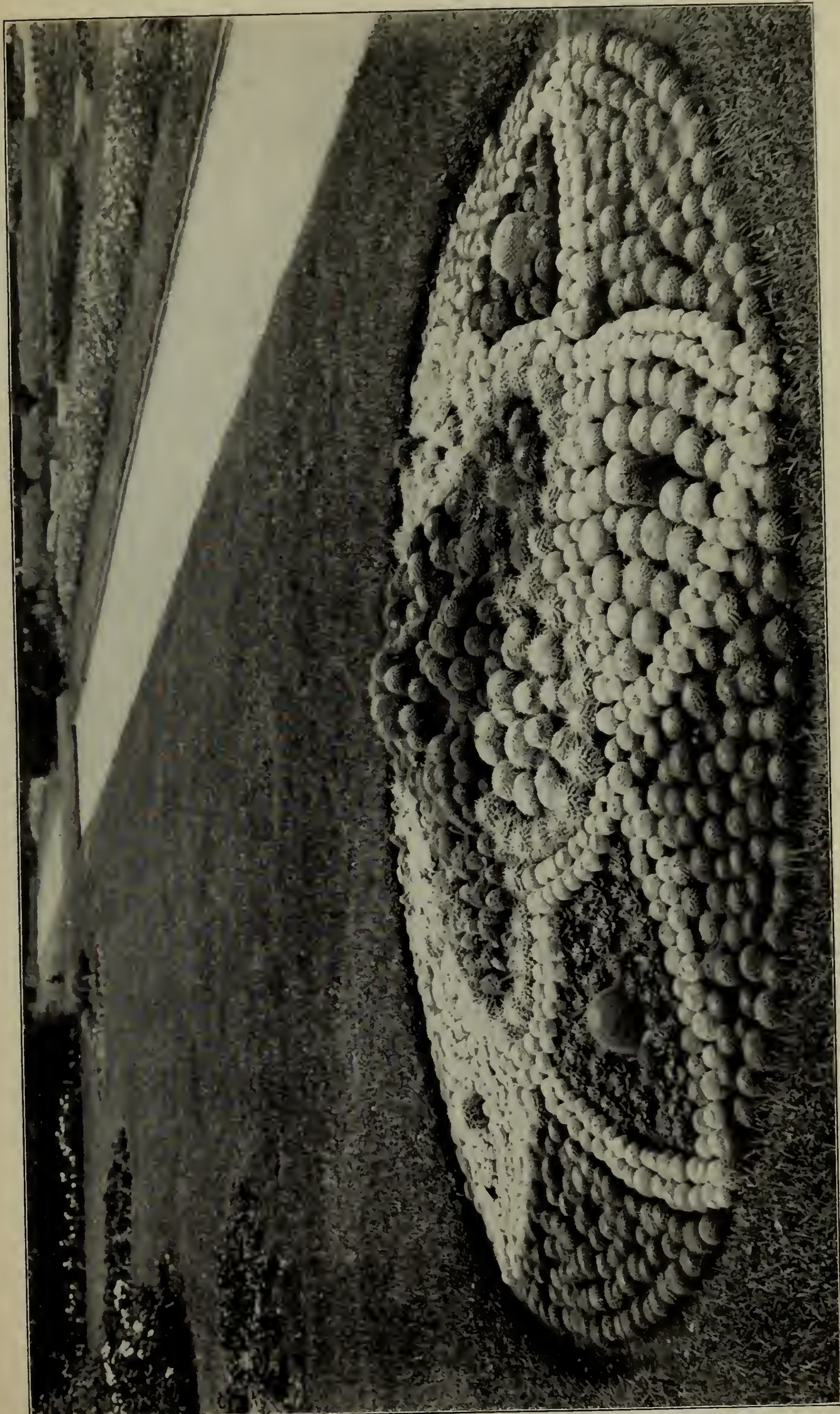
Cereus—Contd.
- euphorbioides.
- forbesii.
- geometrizans.
- giganteus.
- hankeanus.
- hildmannianus.
- jamacaru.
- macrogonus
- marginatus.
- pecten-aboriginum.

Cereus—Contd.
- peruvianus.
- pringlei.
- serpentinus.
- stellatus.
- thurberi.
- weberi.

Opuntia:
- cereiformis.

Pilocereus:
- chrysacanthus.

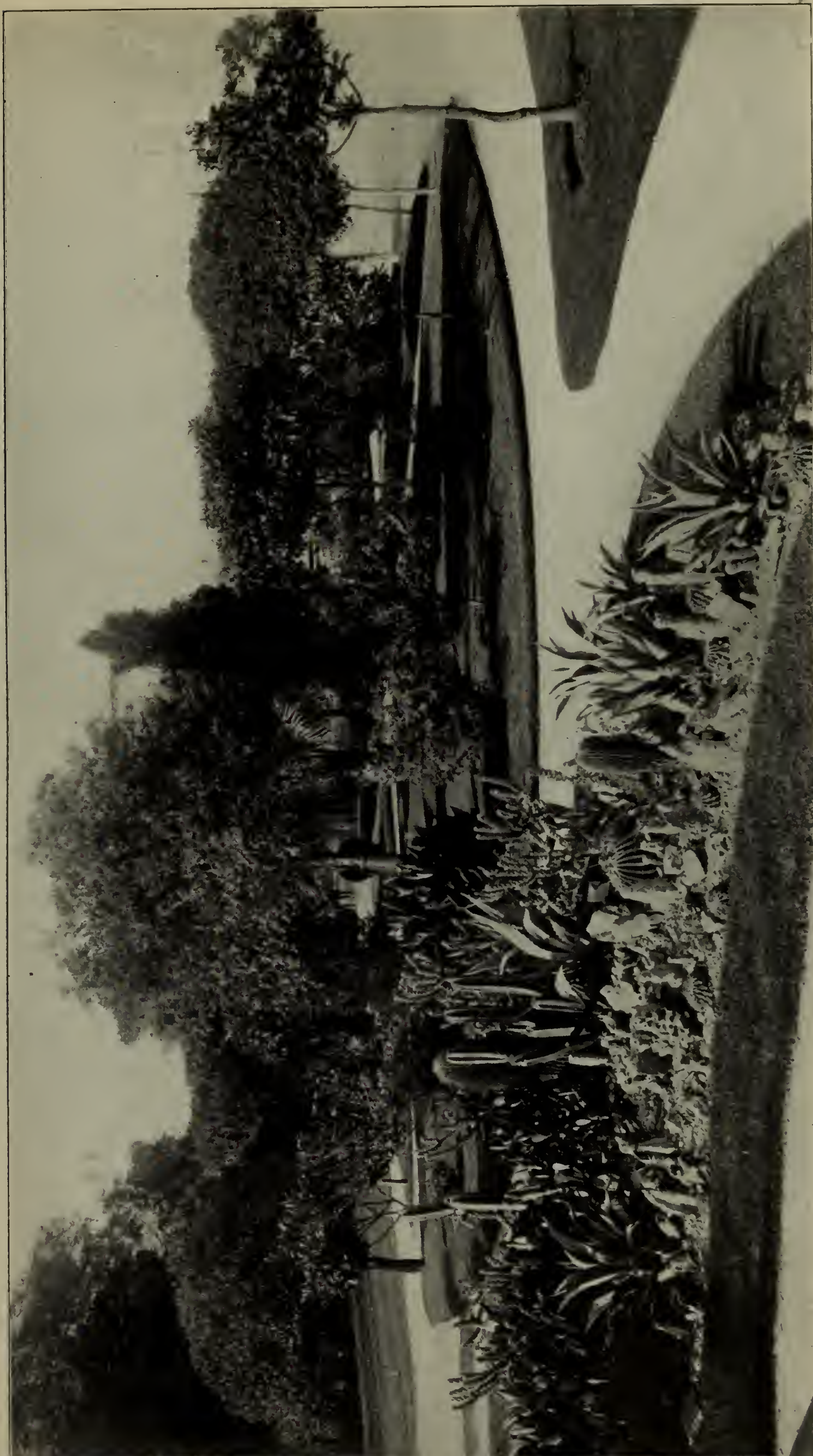
Pilocereus—Contd.
- cometes.
- exerens.
- fulviceps.
- hoppenstedtii.
- houlletii.
- lanuginosus.
- polylophus.
- russelianus.
- schottii.
- strictus.



MAMILLARIAS PLANTED IN FORMAL DESIGN, MISSOURI BOTANICAL GARDEN, ST. LOUIS, 1905.

CACTUS GARDEN, A. S. WHITE PARK, RIVERSIDE, CAL., 1905.

(Photographed by Griffiths.)



SUCCULENT ROCKERY IN ALAMO PARK, SAN ANTONIO, TEX., 1904.

(Photographed by Griffiths.)

ECHINOCACTUS INGENS, SHOWING THE ZEBRALIKE MARKINGS OF YOUNG OFFSHOOTS, TEHUACAN, MEXICO, 1905.

(Photographed by Trelease.)

Columnar Forms of Cacti—Continued.

Lower—from 1 to 6 feet in height.

Cereus:
- candicans.
- emoryi.
- eruca.
- gummosus.
- mamillatus.

Cereus—Contd.
- spachianus.
- speciosus.
- thelegonus.

Echinocactus:
- cylindraceus.
- ingens. (Pl. XVIII.)
- johnsonii.
- ornatus.
- peninsulae.

Echinocactus—Contd.
- pilosus.
- recurvus.

Echinopsis:
- eyriesii.

Short—less than 1 foot in height.

Echinocactus:
- anfractuosus.
- beguinii.
- bicolor.
- capricornus.
- gibbosus.
- intertextus.
- krausei.
- lenninghauseii.
- leucacanthus.
- longihamatus.
- scheeri.
- uncinatus.
- whipplei.

Echinocereus:
- acifer.
- berlandieri.
- chloranthus.
- cinerascens.
- conglomeratus.
- ctenoides.
- dasyacanthus.
- dubius.
- engelmannii.
- fendleri.
- knippelianus.
- mojavensis.
- paucispinus.
- pectinatus.

Echinocereus—Contd.
- phoeniceus.
- procumbens.
- rigidissimus.
- roemeri.
- stramineus.
- viridiflorus.

Mamillaria:
- carnea.
- clava.
- conoidea.
- cornifera.
- dolichocentra.
- elegans.
- elongata.

Mamillaria—Contd.
- erecta.
- eriacantha.
- grahamii.
- gracilis.
- halei.
- leona.
- macromeris.
- microthele.
- radiosa.
- raphidacantha.
- roseana.
- setispina.
- spinosissima.
- strobiliformis.

Globose Forms of Cacti.

Large—more than 1 foot in diameter.

Echinocactus:
- electracanthus.
- emoryi.

Echinocactus—Contd.
- grusonii.
- ingens.

Echinocactus—Contd.
- longihamatus.

Echinocactus—Contd.
- wislizeni.

Medium—from 3 inches to 1 foot in diameter.

Ariocarpus:
- fissuratus.
- retusus.

Echinocactus:
- albatus.
- capricornus.
- coptonogonus.
- corniger.
- crispatus.
- heterochromus.
- hexaedrophorus.

Echinocactus—Contd.
- horizonthalonius.
- lophothele.
- multicostatus.
- myriostigma.
- polycephalus.
- robustus.
- setispinus.
- texensis.
- unguispinus.

Echinopsis:
- gemmata.
- multiplex.
- nigricans.
- oxygona.

Leuchtenbergia:
- principis.

Mamillaria:
- celsiana.
- gigantea.
- heeseana.

Mamillaria—Contd.
- melanocentra.
- mutabilis.
- radiosa.
- robustispina.
- scheeri.
- seitziana.

Melocactus:
- communis.
- ferox.

Small—less than 3 inches in diameter.

Ariocarpus:
- kotschubeyanus.

Echinocactus:
- denudatus.
- humilis.
- intertextus.
- macdowellii.
- mammulosus.
- minusculus.
- ottonis.
- schickendantzii.
- schilinzkyanus.
- simpsoni.
- submammulosus.
- tabularis.
- turbiniformis.

Lophophora:
- lewinii.
- williamsii.

Mamillaria:
- angularis.
- bicolor.
- bocasana.
- candida.
- caput-medusae.
- carretii.
- centricirrha.
- decipiens.
- dioica.
- elegans.
- elephantidens.

Mamillaria—Contd.
- formosa.
- gummifera.
- heyderi.
- lasiacantha.
- lesaunieri.
- longimamma.
- meiacantha.
- micromeris.
- missouriensis.
- parkinsonii.
- perbella.
- phellosperma.
- plumosa.
- polyedra.

Mamillaria—Contd.
- pusilla.
- radians.
- recurvata.
- rhodantha.
- schelhasei.
- sempervivi.
- senilis.
- sphaerica.
- uncinata.
- wildii.
- zephyranthoides.

Pelecyphora:
- aselliformis.
- pectinata.

Platyopuntias and Nopaleas.

Tall forms—over 6 feet in height.

Opuntia:
brasiliensis.
chlorotricha.
engelmannii.

Opuntia—Contd.
ficus indica.
leucotricha.
puberula.

Opuntia—Contd.
robusta.
tomentosa.
tuna.

Nopalea:
auberi.
dejecta.

Medium forms—2 to 6 feet in height.

Opuntia:
camanchica.
curassavica.

Opuntia—Contd.
microdasys.
monacantha.

Opuntia—Contd.
monacantha variegata.
rafinesquei.

Nopalea:
coccinellifera.

Low or decumbent forms—less than 2 feet in height.

Opuntia:
arenaria.
basilaris.
decumbens.

Opuntia—Contd.
fragilis.
missouriensis.
pes-corvi.

Opuntia—Contd.
procumbens.
rutila.
strigilis.

Opuntia—Contd.
ursina.
vulgaris.

Cylindropuntias.

Tall forms—over 6 feet in height.

Opuntia:
acanthocarpa.
arbuscula.

Opuntia—Contd.
bigelowii.
fulgida.

Opuntia—Contd.
imbricata.

Opuntia—Contd.
prolifera.

Medium forms—1 to 6 feet in height.

Opuntia:
alcahes.
bernardina.
cholla.

Opuntia—Contd.
echinocarpa.
leptocaulis.

Opuntia—Contd.
salmiana.
subulata.

Opuntia—Contd.
versicolor.
whipplei.

Low or prostrate forms—less than 1 foot in height.

Opuntia:
clava.
davisii.

Opuntia—Contd.
diademata.
emoryi.

Opuntia—Contd.
grahamii.
parryi.

Opuntia—Contd.
schottii.

Foliage-Bearing Cacti.

Climbing or clambering forms.

Peireskia:
aculeata.
godseffiana.

Peireskiopsis:
brandegeei.
spathulata.

Shrubs or small trees.

Peireskia:
amapola.
bleo.

Peireskia—Contd.
nicoyana.

Climbing, Night-Blooming Forms of Cereus.

Cereus:
baxaniensis.
bonplandii.
grandiflorus.
hamatus.

Cereus—Contd.
irradians.
macdonaldiae.
martini.

Cereus—Contd.
nycticalus.
ocamponis.
setaceus.

Cereus—Contd.
spinulosus.
tortuosus.
triangularis.

Plants Native to Moist Tropical Regions.

Terrestrial.

Phyllocactus:
ackermannii.
acuminatus.
anguliger.

Phyllocactus—Contd.
crenatus.
grandis.

Phyllocactus—Contd.
hookeri.
phyllanthoides.

Phyllocactus—Contd.
stenopetalus.
strictus.

Epiphytic.

Cereus:
flagelliformis.

Epiphyllum:
gaertneri.
russellianum.
truncatum.

Hariota:
salicornioides.

Rhipsalis:
anceps.
cassytha.

Rhipsalis—Contd.
conferta.
grandiflora.
houllitiana.
mesembryanthemoides.

Rhipsalis—Contd.
paradoxa.
pentaptera.
rhombea.
saglionis.

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U. S. DEPARTMENT OF AGRICULTURE.

BUREAU OF PLANT INDUSTRY—BULLETIN NO. 263.

B. T. GALLOWAY, *Chief of Bureau.*

METHODS USED IN BREEDING ASPARAGUS FOR RUST RESISTANCE.

BY

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LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry,
Office of the Chief,
Washington, D. C., July 29, 1912.

Sir: I have the honor to transmit herewith a manuscript entitled "Methods Used in Breeding Asparagus for Rust Resistance," by Mr. J. B. Norton, Physiologist in the Office of Cotton and Truck Disease and Sugar-Plant Investigations, and to recommend its publication as Bulletin No. 263 of the Bureau series.

This paper deals with the methods developed in the selection, pollination, and breeding of asparagus at Concord, Mass. The work was done in connection with the rust-resistant asparagus breeding investigations being conducted at Concord by this Bureau in cooperation with the Massachusetts Agricultural Experiment Station.

The author desires to acknowledge the assistance of Mr. C. W. Prescott, of Concord, Mass., who, since the beginning of the work, has done everything possible to aid the breeding work. Mr. Frank Wheeler, of Concord, Mass., together with many other asparagus growers from various sections, has given active assistance in the work.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

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METHODS USED IN BREEDING ASPARAGUS FOR RUST RESISTANCE.

INTRODUCTION.

History of asparagus rust.—Asparagus rust (*Puccinia asparagi* DC.), which has caused severe losses to asparagus growers in the United States during the past 16 years, is a native of Europe. Common asparagus (*Asparagus officinalis*) grows wild over the greater portion of Europe and parts of western Asia and northern Africa. This species, with several closely allied forms, is the normal host plant of the asparagus rust. There is nothing in the literature of the subject to indicate that the rust is in any way nearly as serious a pest in Europe as it has been in America. The comparative immunity of the crop in Europe is partly explained on the ground that the rust is held in check by its natural enemies and by climatic conditions. The gradual development of resistant varieties in Europe has had something to do with this apparent difference in severity of attack.

Occurrence of rust in America.—The definite occurrence of asparagus rust in America was unknown until 1896, when its discovery was recorded by Prof. B. D. Halsted,[1] of the New Jersey Agricultural Experiment Station. The same year brought reports of its occurrence on Long Island and in Massachusetts and Connecticut. It is very probable that the rust had been introduced in some way from Europe a year or so previously and had spread without being discovered. Since 1896 it has spread over practically the entire area of the United States where asparagus is grown. It became a factor in asparagus growing in the large fields of California in 1902.

Failure of spraying methods in the East.—In spite of the general interest and alarm felt by growers and station workers at the sudden appearance and rapid spread of this disease, satisfactory methods of rust control have not been developed for the eastern regions. Smith says:[2]

> In regard to methods of treatment for the control of the rust it may fairly be said that up to the time of the appearance of the disease in California nothing effective and satisfactory had been developed in other portions of the country previously affected.

[1] Halsted, B. D. Garden and Forest, vol. 9, 1896, p. 394.

[2] Smith, R. E. Bulletin 172, California Experiment Station, 1906, p. 1.

Many elaborate experiments with sprays have been carried out by different experiment stations, but for various reasons the growers as a class have failed to take up the practice of spraying. Here and there an isolated case exists where a farmer put in a spraying outfit at considerable cost and managed to check the rust so as to make his beds continue to yield paying crops. Most growers did not take up this work on account of its extra cost and trouble, either letting their old beds die out (Pl. I, fig. 1) or planting new fields (Pl. II) of such semi-resistant varieties as Argenteuil or Palmetto.

Previous attempts at breeding.—Some preliminary attempts have been made to start breeding work to develop resistant strains, but so far as the writer knows none of these attempts have been successful. Smith [1] makes the following statement:

> A beginning has been made by the writer toward breeding desirable rustproof varieties by saving seed of such plants from various States, which is being carefully planted for such a purpose. Quite a collection is already on hand from promising sources. Seed has also been imported from Europe of a number of varieties grown there and plants have been obtained from all of these.

The preliminary work on spraying and variety testing brought out the fact that certain European varieties were more resistant to rust than the ordinary strains grown in the United States at the time the rust was introduced.

Hexamer [2] in his book on asparagus says:

> All the cultivated varieties of asparagus are readily affected by the rust, although it has been found that some varieties, notably Palmetto, are less susceptible to its attacks than others.

Smith,[3] under a discussion of varieties, makes the following statement:

> There is no question that some varieties of asparagus are more resistant to rust than others. This difference appears much more in new beds, planted after the rust outbreak started, than in those which existed at the time. So much is this true that in the East the rust problem seems well-nigh solved by the growing of Palmetto asparagus, yet in the first years of rust the difference in favor of this variety was slight and often not at all apparent. In 1900 Sirrine wrote that "The fields of Long Island have been watched every year since 1896, with the result that only slight, if any, differences in favor of the Palmetto were to be noticed, except that in some cases it did not succumb as early;" yet at present in the same fields the Palmetto alone remains and is being extensively planted. Mr. William Conover planted a field on his place in New Jersey with three rows of Palmetto, then three rows of Conover's Colossal, alternately, and after a few years the Palmetto was still green when the other variety was practically exterminated so that those rows had to be replanted with Palmetto. There is no doubt whatever that this variety is much less affected and less injured by rust in the long run, even though it does not always appear at first. The Argen-

[1] Smith, R. E. Asparagus and Asparagus Rust in California. Bulletin 165, California Agricultural Experiment Station, 1905, p. 95.

[2] Hexamer, F. M. Asparagus, Its Culture for Home Use and for Market, 1901, p. 140.

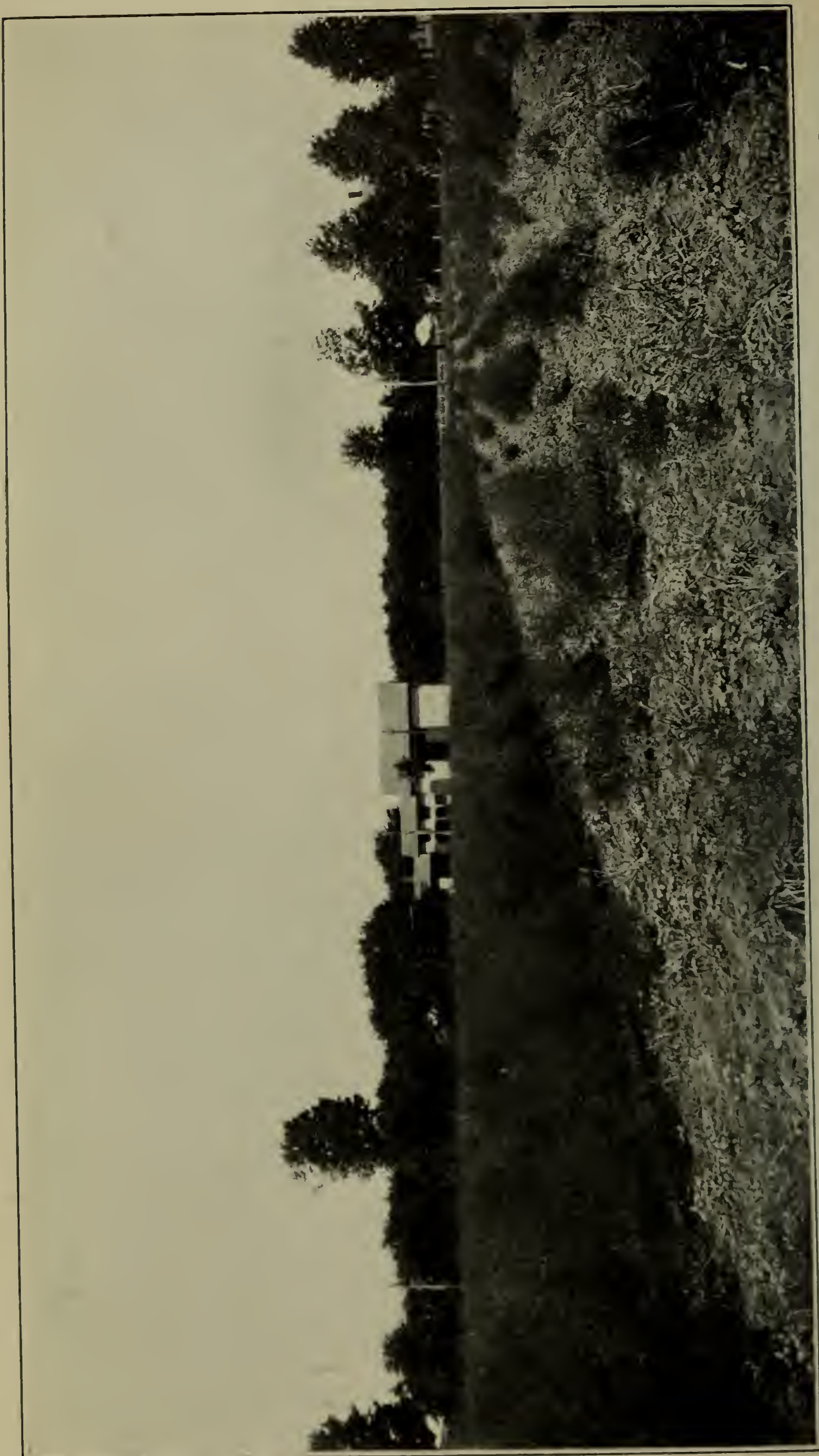
[3] Smith, R. E. Op. cit., p. 94.

FIG. 1.—AN ASPARAGUS FIELD ON CAPE COD, MASS., KILLED OUT BY SEVERE ATTACKS OF RUST AND NEVER REPLANTED.

FIG. 2.—FIELD SHOWING THE EFFECT OF RUST ON A NONRESISTANT VARIETY OF ASPARAGUS. ROW B24 (IN THE CENTER) WOULD HAVE BEEN AS VIGOROUS AS THE ROWS ON EITHER SIDE, BUT FOR RUST.

(From a photograph taken September, 1908, after the second season's growth.)

EFFECT OF RUST ON ASPARAGUS.



AN OLD ASPARAGUS FIELD AT CONCORD, MASS., KILLED OUT BY RUST. THE NEW FIELD OF READING GIANT ON THE LEFT WAS GROWN AS A BREEDING FIELD IN RUST-RESISTANT BREEDING WORK.

teuil (Bonvallet's Giant, French, and other trade names) is, if not identical with Palmetto, indistinguishable from it, and equally rust proof. Among all the American varieties no great difference exists, and they are in fact probably all selections from the old Conover's Colossal. * * * In a rusty field of any variety plants can be seen here and there which are greener, less affected, and more nearly rust proof than the average of the field.

Massachusetts Asparagus Growers' Association.—The publication in 1905 of Bulletin 165 of the California Agricultural Experiment Station by Prof. Ralph E. Smith, demonstrating the value of spraying and preventive measures in fighting rust, reawakened the interest of eastern growers. The general interest in plant breeding at this time, particularly in its application to the breeding of disease-resistant varieties of crops, suggested the possibility of its application to asparagus in fighting the rust. The fact that individual plants of a variety as well as different varieties as a whole varied in their rust resistance showed that the breeding proposition was not impossible. These facts led in 1906 to the organization of the Massachusetts Asparagus Growers' Association, with the object of securing by plant breeding a rust-resistant variety of asparagus. This association enlisted the cooperation of the Massachusetts Experiment Station and the United States Department of Agriculture. A cooperative plan of work was drawn up, under which, with some modification, the work has been carried out. The Department made collections of seed and plants from various sources and furnished the services of its experts in carrying out the breeding work. The Massachusetts station furnished the funds to run the work at Concord, Mass., where a branch experiment station was established on the farm of Mr. Charles W. Prescott. Beginning with the fall of 1908, the department has borne all of the expenses in connection with the breeding work, the funds formerly supplied by the Massachusetts station being needed for the proper development of the fertilizer and nutrition work on asparagus at the Concord station.

It must be understood at the outset that this work was intended to develop a rust-resistant strain of asparagus and not to discover remedial measures to help the nonimmune varieties already growing. Spraying treatments, etc., have been recommended by plant pathologists for years, but none have been generally adopted by the growers. The Massachusetts Asparagus Growers' Association started out with one idea, namely, the production of a strain that would be so immune to rust that the farmer would need to pay no attention to fighting the disease. This object has been kept constantly in view, and at present the prospect of success is so certain that no experiments with sprays will be undertaken. Nine out of ten growers in the East will not spray, anyway. Breeding work will produce better yielding types of commercially rust-immune asparagus, which will drive out the older fields as fast as it is possible to produce the stock.

PRELIMINARY WORK.

NATURE OF THE DISEASE.

In taking up breeding work for disease resistance, a knowledge of the life history of the organism causing the trouble is usually considered necessary. Previous work done in America on the life history of the asparagus-rust fungus by Halsted, Stone, Smith, and others has given us a basis of sufficient breadth to go ahead without further work. As a matter of fact, the methods of breeding used in this work have not depended on a knowledge of the life history of the rust fungus, except in a minor way. As yet we have found no constant differences in structure or physiology between the resistant and nonresistant plants.

Asparagus rust was described in 1805 by De Candolle as *Puccinia asparagi.* It belongs to the order Uredineæ, which comprises the group of fungi known as "rusts." These fungi are all parasitic on the higher plants, the most familiar examples being the common grain rusts. Asparagus rust differs from the grain rusts in being autœcious—that is, all stages of the rust occur on the asparagus plant, while the grain rusts are heterœcious, the spring stages occurring on a widely different host, wheat rust, for example, having its spring stage on barberry. The disease known to growers as asparagus rust is always the direct result of an infection from spores of *Puccinia asparagi* and, contrary to opinions held by many growers, is not caused by fertilizers, soil or weather conditions, insects, or anything else of this nature. However, once the disease is introduced, these other factors may influence its development and intensity.

The first activity shown in the spring by the rust occurs about the same time that the shoots of asparagus begin to appear above ground. At this time the resting spores of the fungus begin to germinate. From each cell of these spores there arises a short segmented filament, bearing four small sporidia. These sporidia are carried by air or water into contact with the young shoots just coming through the ground and on germinating send their mycelium through the epidermis of the shoot and establish themselves in the asparagus shoot. This mycelium after a growing period of less than a month, varying with weather conditions, starts to produce spores. These spores are located under the epidermis in groups commonly known as cluster cups or æcidia (Pl. III, fig. 2). These æcidia finally rupture the epidermis of the asparagus shoot and the light-orange spores are liberated. Accompanying the cluster-cup stage on asparagus are found honey-yellow spots of smaller size, known as spermogonia. These spermogonia produce small sporelike bodies that resemble the male spores of related orders, but which are now apparently functionless.

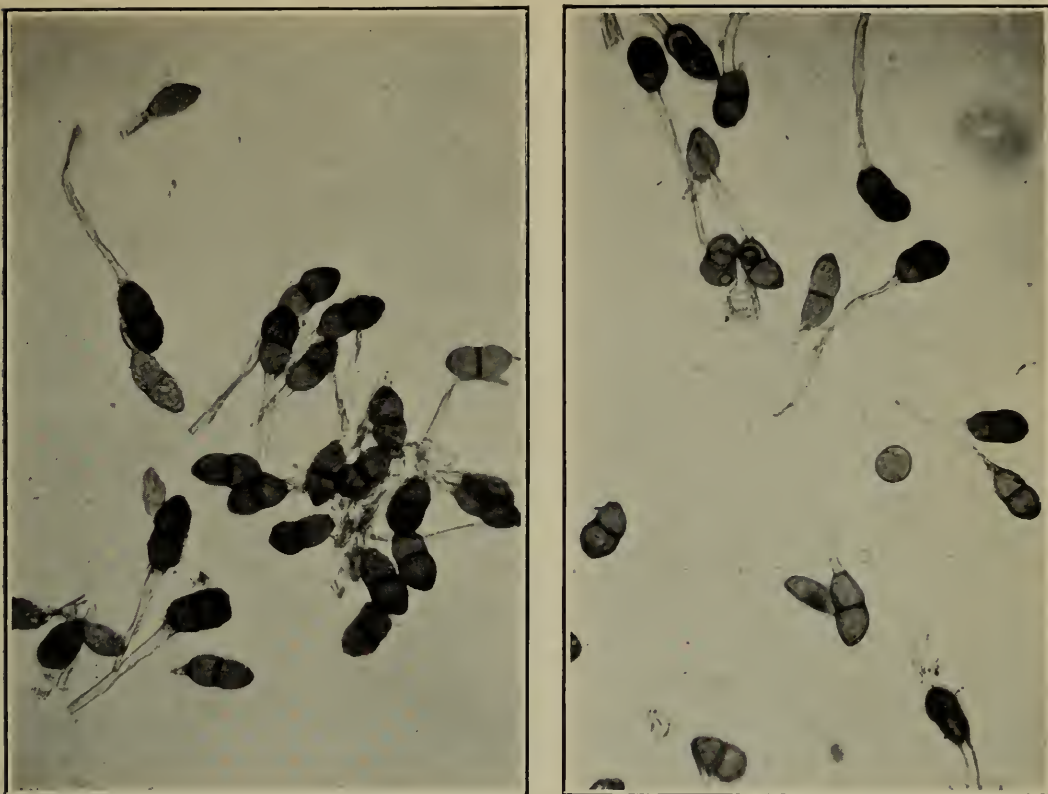


FIG. 1.—TELEUTOSPORES OF ASPARAGUS RUST. NEAR THE CENTER OF THE CUT ON THE RIGHT IS A UREDOSPORE. ×180.

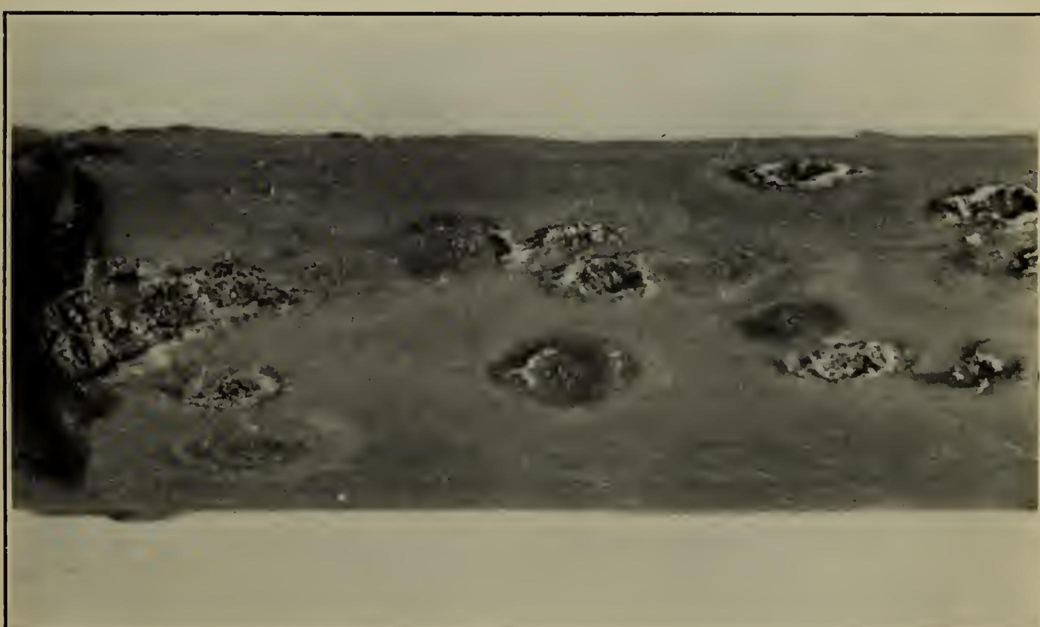


FIG. 2.—CLUSTER-CUP STAGE OF RUST ON AN ASPARAGUS STEM. ×3.

DIFFERENT STAGES OF PUCCINIA ASPARAGI.

ASPARAGUS BRANCH WITH SORI OF PUCCINIA ASPARAGI IN UREDO AND TELEUTO STAGES. ×5.

The spores from the cluster cups are usually rounded, orange yellow, and 1-celled. They are carried by air currents and lodge on the stems or cladodes of the asparagus plants. These spores germinate in the presence of moisture and produce a mycelium which grows into the asparagus plant through the stomata or breathing pores. These spring spores provide the infection which causes the summer rust or uredo stage. It is in this stage that the serious damage is done. The first signs of summer rust appear in about two or three weeks after the development of the cluster cups. The uredo spores appear in clusters of single-celled, red-brown spores which rupture the epidermis and are carried away by the air currents to reinfect other plants. At this stage the rust spreads rapidly. In warm weather accompanied by dew at night, the life cycle from uredo spore to uredo spore is often less than 12 days. This is shown by observations on seedling asparagus plants in the summer of 1909, when rust was repeatedly found on shoots that had been out of the ground less than 12 days. Beginning its attacks in the region of Concord about the latter part of June or first part of July, the uredo stage continues into October. Accompanying it and sometimes occurring alone is the teleuto stage (Pl. III, fig. 1), the fall stage, which goes through the winter to provide the spring infection. From the common name, fall rust, it might be assumed that this stage would be found only in the fall, but it has appeared regularly with the uredo stage at Concord during the summers since 1908 (Pl. IV). This is due to the fact that it appears under unfavorable circumstances, such as dry weather or prolonged periods of cool weather.

Asparagus rust has an active parasite in the fungus *Darluca filum*, a parasite of rusts in general, which is usually present in the rusted fields and is found attacking the rust in all its stages. During foggy or rainy periods in summer when the rust suffers most from Darluca, its attacks are reenforced by several saprophytic fungi, which often give a mildewed or molded appearance to shoots that have been injured by rust.

EFFECT OF THE DISEASE.

While the effect of asparagus rust is not seen directly in the marketed product, nevertheless it is quite injurious. The damage is due to the weakening of the plants by the attacks of the rust on the shoots during the summer after the cutting season is over. It is during the growing season that the plants store up food for the next spring, and if the shoots are injured or broken off the next season's food supply is accordingly diminished. When the rust attacks a plant no injury is apparent until the formation of the spore clusters ruptures the epidermis and allows excessive evaporation from the stems. The shoots then wither and die. Attacks on the smaller branches and cladodes show a deadening effect shortly after the rust sori appear. On the whole the mechanical injury seems to be greater than any other. The attacks on young shoots in the late summer when the rust has become abundant are apt to be quite severe. This is due to the almost complete infection that takes place owing to the large number of uredospores blowing about while the growth is quite tender just as it comes through the soil. Spores are often developed from these infections before the shoot has had time to branch out and produce cladodes. These spore clusters or sori often are so numerous that they crack off the epidermis from large areas and the plants rapidly wither or stop growing.

COLLECTION OF VARIETIES.

The first work in starting breeding experiments was the accumulation of a collection of varieties from different sources. It was the aim to include in this variety test all possible sources of rust-resistant plants. In order to get the work started as soon as possible, eight rows of yearling roots were planted on the trial grounds at Concord, Mass., during May, 1906. These roots were contributed in lots of 100 by local members of the Asparagus Growers' Association, as shown in Table I. Row 9 was added the next season from roots obtained by Mr. Prescott.

Table I.—*Roots planted for breeding experiments in field A.*

Row.	Variety.	Source.
	Set May 5, 1906:	
1	New American (Geneva grown)	Anson Wheeler, Concord, Mass.
2	Do	Do.
3	Argenteuil (Concord grown)	F. E. Foss, Concord, Mass.
4	Do	Do.
6	Palmetto (Long Island grown)	Anson Wheeler, Concord, Mass.
7	New American (Concord grown)	Do.
8	(?) (Concord grown)	Wilfrid Wheeler, Concord, Mass.
	Set April 17, 1907:	
9	Palmetto (Long Island grown)	W. H. Reeve, Mattituck, Long Island.

During the fall and winter of 1906 a larger collection of seed and roots was obtained from most of the seedsmen in America and Europe and from interested growers in the asparagus region of the East. Mr. C. W. Prescott made a trip in the fall of 1906 through the asparagus regions of Long Island and New Jersey, securing seed from resistant stocks and fields. These lots of seed from all sources were germinated in flats in the greenhouses at Washington, D. C., in March, 1907; shipped to the field at Concord, Mass., in May, 1907; and planted in their permanent place in the trial rows. This treatment was very severe and many plants failed to grow. Judging by the resistance and vigor of the plants in 1907, larger orders were placed that fall for seed of Late Argenteuil from Vilmorin-Andrieux & Co., of Paris, and for Reading Giant from Sutton & Sons, Reading, England. These lots of seed were grown in 1908 at Concord and were planted in 1909, 2½ acres of Argenteuil and practically the same of Reading Giant. No new strains have since been planted.

Table II shows the varieties planted in 1907 from the seed and roots obtained the previous fall and winter. It was the intention to have about 100 seedlings or about 10 roots from each lot.

TABLE II.—*Asparagus varieties planted in field B at the Concord Asparagus Experiment Station.*

Row.	Name of variety.	Source.
	FROM SEEDLINGS GROWN AT WASHINGTON.	
1	Dreer's Eclipse	Henry A. Dreer, Philadelphia, Pa.
2	Palmetto (Prescott 10)	Walter Van Fleet, Little Silver, N. J.
4	Hub	Joseph Breck & Sons, Boston, Mass.
6	Moore's Giant	Schlegel & Fottler, Boston, Mass.
8	.....do	W. W. Rawson & Co., Boston, Mass.
10	Palmetto	J. M. Mitchell, Mount Pleasant, S. C.
12	Mammoth Prolific	Moore & Simon, Philadelphia, Pa.
14	Donald's Elmira	Johnson & Stokes, Philadelphia, Pa.
16	Colossal	J. M. Thorburn & Co., New York, N. Y.
18	Rust Resistant	B. R. Tillman, Trenton, S. C.
20	Colossal	Jas. Barr & Sons, London, England.
22	Perfection	H. W. Buckbee, Rockport, Ill.
24	Seedling	R. P. Wakeman, Southport, Conn.
26	Late Argenteuil	Vilmorin-Andrieux & Co., Paris, France.
28	Vick's Mammoth	James Vick & Sons, Rochester, N. Y.
30	Sutton's Perfection	Sutton & Sons, Reading, England.
32	Reading Giant	Do.
34	Early Argenteuil	Vilmorin-Andrieux & Co., Paris, France.
36	Barr's Canadian	Barr & Sons, London, England.
38	Mammoth Emperor	James Carter & Co., London, England.
40	Barr's Mammoth	Jas. Barr & Sons, London, England.
42	Columbian Mammoth White	D. M. Ferry & Co., Detroit, Mich.
44	.....do	J. M. Thorburn & Co., New York, N. Y.
46	White German	Vilmorin-Andrieux & Co., Paris, France.
48	Erfurt Giant White	Ernest Benary, Erfurt, Germany.
50	Snow Cap Giant	Do.
52	Giant White Head	Heinemann, Erfurt, Germany.
54	Barr's New White	Jas. Barr & Sons, London, England.
56	Sutton's Giant French	Sutton & Sons, Reading, England.
58	Snow Head	Platz & Son, Erfurt, Germany.
60	Glory of Brunswick	Ernest Benary, Erfurt, Germany.
62	Palmetto	N. L. Willet Drug Co., Augusta, Ga.
64	Batavian	James Carter & Co., London, England.
66	Purple Dutch	Vilmorin-Andrieux & Co., Paris, France.
68	French Giant	Johnson & Stokes, Philadelphia, Pa.
70	Erfurt Giant	Heinemann, Erfurt, Germany.
72	Bonvallet's Giant	Vaughan's Seed Store, Chicago, Ill.
74	Early Argenteuil (Prescott 14)	Peter Henderson & Co., New York, N. Y.
76	Giant Argenteuil	Vilmorin-Andrieux & Co., Paris, France.
78	Palmetto	George Tait & Sons, Norfolk, Va.
80	Palmetto (Prescott 12)	Jas. J. H. Gregory & Sons, Boston, Mass.
82	Palmetto (Prescott 2)	W. H. Reeve, Mattituck, Long Island, N. Y.
84	Palmetto (Prescott 3)	Joseph Cooper, Mattituck, Long Island, N. Y.
86	Palmetto (Prescott 4)	A. L. Downs, Mattituck, Long Island, N. Y.
88	Palmetto (Prescott 5)	J. G. Downs, Mattituck, Long Island, N. Y.
90	Bonvallet (Prescott 13)	Vaughan's Seed Store, Chicago, Ill.
92	Bay State	A. D. Shamel, U. S. Dept. of Agriculture.
94	Palmetto (Prescott 1)	Joseph Cooper, Mattituck, Long Island, N. Y
96	Palmetto (Prescott 6)	Long Island Seed Co., Mattituck, N. Y.
98	Palmetto (Prescott 7)	Edwin Beeckman, Middletown, N. J.
100	Palmetto (Prescott 8)	W. B. Conover, Red Bank, N. J.
102	Palmetto (Prescott 9)	Dr. S. L. De Fabry, Little Silver, N. J.
104	Palmetto (Prescott 11)	Hiram Worthley, Concord, Mass.
114	Nutmeg State	A. D. Shamel, U. S. Dept. of Agriculture.

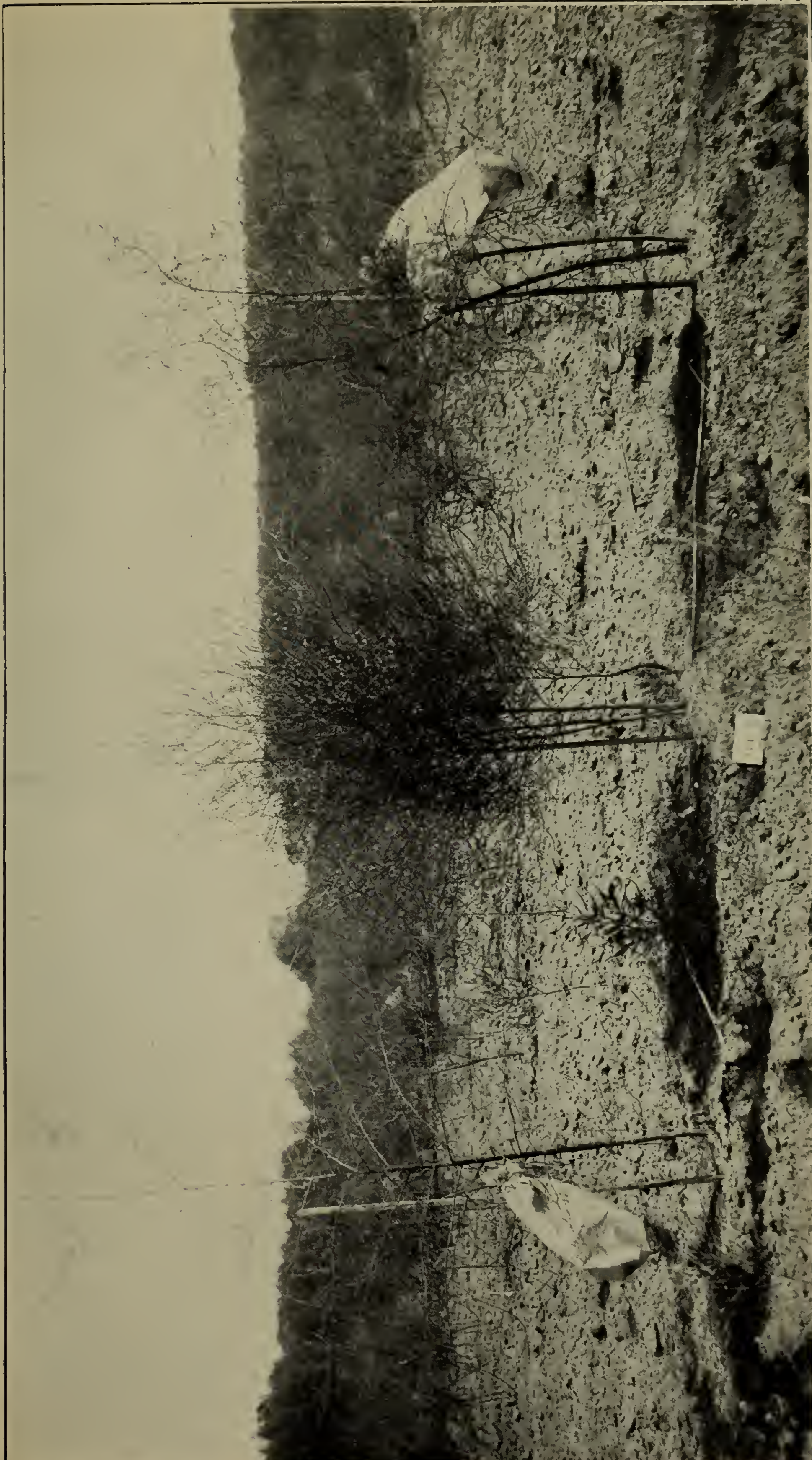
TABLE II.—*Asparagus varieties planted in field B at the Concord Asparagus Experiment Station*—Continued.

Row.	Name of variety.	Source.
	ROOTS OBTAINED FROM GROWERS AND SEEDSMEN.	
1	Dreer's Eclipse	Henry A. Dreer, Philadelphia, Pa.
4	Hub	Joseph Breck & Sons, Boston, Mass.
12	Mammoth Prolific	Moore & Simon, Philadelphia, Pa.
14	Donald's Elmira	Johnson & Stokes, Philadelphia, Pa.
15	.....do	Dr. B. T. Galloway, U. S. Dept. of Agriculture.
16	Colossal	J. M. Thorburn & Co., New York, N. Y.
20	.....do	Jas. Barr & Sons, London, England.
22	Perfection	H. W. Buckbee, Rockport, Ill.
26	Late Argenteuil	Vilmorin-Andrieux & Co., Paris, France.
28	Vick's Mammoth	James Vick & Sons, Rochester, N. Y.
30	Sutton's Perfection	Sutton & Sons, Reading, England.
32	Reading Giant	Do.
34	Early Argenteuil	Vilmorin-Andrieux & Co., Paris, France.
36	Barr's Canadian	Jas. Barr & Sons, London, England.
38	Mammoth Emperor	Jas. Carter & Co., London, England.
40	Barr's Mammoth	Jas. Barr & Sons, London, England.
41	.....do	Johnson & Stokes, Philadelphia, Pa.
42	Columbian Mammoth White	D. M. Ferry & Co., Detroit, Mich.
44	.....do	J. M. Thorburn & Co., New York, N. Y.
48	Erfurt Giant White	Ernest Benary, Erfurt, Germany.
49	Erfurt Giant	Heinemann, Erfurt, Germany.
50	Snow Cap Giant	Ernest Benary, Erfurt, Germany.
52	Giant White Head	Heinemann, Erfurt, Germany.
54	Barr's New White	Jas. Barr & Sons, London, England.
56	Sutton Giant French	Sutton & Sons, Reading, England.
58	Snow Head	Platz & Son, Erfurt, Germany.
60	Glory of Brunswick	Ernest Benary, Erfurt, Germany.
62	Palmetto	N. L. Willet Drug Co., Augusta, Ga.
64	Batavian	James Carter & Co., London, England.
66	Purple Dutch	Vilmorin-Andrieux & Co., Paris, France.
68	French Giant	Johnson & Stokes, Philadelphia, Pa.
72	Bonvallet's Giant	Vaughan's Seed Store, Chicago, Ill.
78	Palmetto	George Tait & Sons, Norfolk, Va.
107	.....do	T. S. Williams, Hattieville, S. C.
108	Giant Emperor	John Lewis Childs, Floral Park, N. Y.
109	(Unnamed)	South Carolina.

In addition to the above-mentioned lots, selections were begun in 1908 from about 5 acres of Imported Argenteuil and 2 acres of selected stock from the imported lot, both on the farm of Mr. Frank Wheeler, and from 3 acres of selected stock from Mr. Wheeler planted on the station grounds. This last field is being used for fertilizer and nutrition trials by the Massachusetts station.

VARIETAL UNIFORMITY.

Although many names are included in the collection of varieties, few distinguishing characters are to be found to separate the so-called varieties. A lot of seedlings would show nearly all the variations found in the whole trial field (Pl. V). One lot of Columbian Mammoth White that could have been the purest stock in the field in the character of whiteness showed no pure white plants in the whole trial row. To judge from the observations made on the varietal lots at Concord there are at present no pure strains of asparagus, the difference between the various lots being on a percentage basis. Thus, one lot may have more large stalks than another, hence it may



THREE PLANTS OF THE SAME VARIETY, B114, ARGENTEUIL, SHOWING WIDE DIFFERENCES IN TYPE OF MATURE GROWTH. IN SOME LOTS THE DIFFERENCES ARE EVEN MORE MARKED.

TYPES OF ASPARAGUS SHOOTS. NOS. 1 AND 3, GOOD; NO. 2, TOO LARGE AT BASE; NO. 4, ROUGH WITH PROJECTING SCALES; NO. 5, FROM SHORT SLOW-GROWING PLANT.

be called Giant; another lot may have more white plants and therefore may be given the name "White" or some similar term.

The observations at Concord showing lack of uniformity in varieties correspond to those of many experimenters and growers elsewhere who have written on the subject.

Ilott,[1] writing from the standpoint of an English grower, says:

There are many so-called varieties, yet they differ but little. Messrs. Sutton & Sons of Reading have two—Perfection and Giant French—which are somewhat distinct. They are both excellent kinds, but whether they differ from others going by different names I do not know, for culture has a great deal to do with the appearance of asparagus, as of human beings. A variety which is sometimes well grown, and sometimes the reverse, varies much in appearance, thus favoring the idea of a difference of variety. Two other possibly distinct varieties are Argenteuil Early Giant and Argenteuil Late Giant, which latter probably keeps longer in the cutting season by furnishing shoots later than the first named. Conover's Colossal is another good kind, but not superior to those named above. Palmetto reached me a few years ago with a startling character. It is said to be both earlier and larger than any other, but planted side by side with all the kinds above mentioned I have not yet found it [to] display its alleged virtues. It came from America, and it is possible that it went over there first from Europe, probably from England, for I find it about as good as many others. As to size, it is smaller than Sutton's Giant French. The only other variety which I am going to mention is one which was sent out by Messrs. Bunyard. They named it Harwood's Early, and it is noteworthy as being alleged to be the earliest to become fit for the markets. It certainly has in my experience for three years in succession started before the other kinds. There is, however, as I consider, far more importance in soils, sites, and general cultivation than in difference of variety, and whereas the cultivation differs materially, the varieties do not, in any great measure, differ from one another.

H. W. Ridgway,[2] of Swedesboro, N. J., one of the best growers in the East, in a recent discussion of asparagus growing, says:

Variety is the principal thing, but in making our selection of variety let us not put too much dependence on the name. It may be misleading, owing to the fact that many growers are not acquainted with the varieties and accept the name given them without questioning its authenticity. There is only one species and several varieties; one-half of the names that we hear are not varieties. The grass so named has been caused by methods of cultivation, highly-manured land, and climatic conditions, and differ from each other only by a single characteristic which will rapidly disappear when grown under climatic and soil conditions different from that in which they originated.

Smith in his work in California found no uniform varieties and many names applied to strains that differed from each other in no appreciable way. The same opinion as to varietal differences is held by most growers who have been interviewed.

One thing is apparent in looking over tests of varieties, namely, that no real pedigree breeding has been done. A search through

[1] Ilott, Charles. The Book of Asparagus, 1901, p. 2.

[2] Ridgway, H. W. Extract from Thirty-sixth Annual Report, New Jersey State Board of Agriculture, 1909, pp. 111–115.

available literature reveals no pedigree work nor even plans for any. The variability of the best imported strains seems to settle the matter (Pl. VI), at least as far as Europe is concerned. The results from one generation of pedigree breeding at Concord show that uniformity can be obtained in many characters by proper selection of parent types, yet it is highly probable that little real advance in asparagus varieties has been made since the time of the Roman gardener who grew stalks of such size that three weighed over a pound.

A quotation from Hexamer[1] shows the best method recommended by authors in giving advice as to seed growing. This method is practically the same as that recommended by most European writers and is that followed by some of the best growers in this country.

> In order to insure the production of the very best asparagus seed a sufficient number of pistillate or seed-bearing plants, which produce the strongest and best spears, should be selected and marked so that they may be distinguished the following spring when the shoots appear. These clumps should be close together and near some staminate or male plants which have to be marked likewise, as without their presence fertile seed can not be produced. The number of the male to the female plants should be about one to four or five. The following spring all the sprouts of the selected male plants are allowed to grow without cutting. On each hill of the female plants the two strongest and earliest stalks are allowed to grow, cutting the later appearing spears with the others for market or home use. Thus these early stalks of both male and female plants bloom together before any other stalks, and the blooms on the female plants will be fertilized with the pollen of the selected male plants. This last is of prime importance, for on proper fertilization depends the purity of the seed as well as the vigor of the resultant plant. Not all seed of even a good plant properly fertilized should be used for reproduction, as of the seeds gathered from any plant some will be better than others. Only the largest, plumpest, and best-matured seeds should be used, for by saving these the most nearly typical plants of the sort will be most certainly produced. The selection of the best seed from typical plants is as essential to success as are good soil, thorough cultivation, and heavy manuring.

VALUE OF UNIFORMITY.

The uniform distribution of good asparagus over the field is a matter that has received some attention from growers. The yield and type of the individual plants in most varieties differ widely, and it is probable that less than half of the plants pay a profit. This difference in yield is illustrated graphically in figure 1. A separate record was kept of the cut of each hill in row A1, New American, Geneva-grown stock, in its fifth season in permanent place in the bed. The diameter of each stalk was measured, this method being considered more reliable than to take the weight. This diagram shows that 37 of the hills cut above the average and that unquestionably many plants in the row were not paying for ground rent, fertilizer, and labor.

[1] Hexamer, F. M. Asparagus: Its Culture for Home Use and for Market, 1901, p. 27.

This row represents an "average" lot of plants. Some beds in Concord and vicinity show a higher average efficiency, but many are lower.

INTRODUCTION OF UNCULTIVATED SPECIES.

In connection with the introduction of asparagus varieties for the rust-resistant work, several wild species have been brought in by the Office of Foreign Seed and Plant Introduction from different regions for distribution. *Asparagus* (*Asparagopsis*) *virgatus* from South Africa was tested at Concord in 1908 and proved to be entirely free from rust. The following winter all the plants were killed by cold. Later trials at Washington have shown that this species is quite tender. Mr. George W. Oliver, of this Bureau, has tried to cross this species with *A. officinalis*, but without success. *A. officinalis pseudoscaber* was tested in 1911 at Concord but proved quite susceptible to rust. So far the attempted crosses between this variety and the parent form have failed to give hybrid plants. *A. davuricus*, a related form from China, was pollinated in 1911 with *A. officinalis* pollen and has given seedlings that show hybrid characters. *A. davu-*

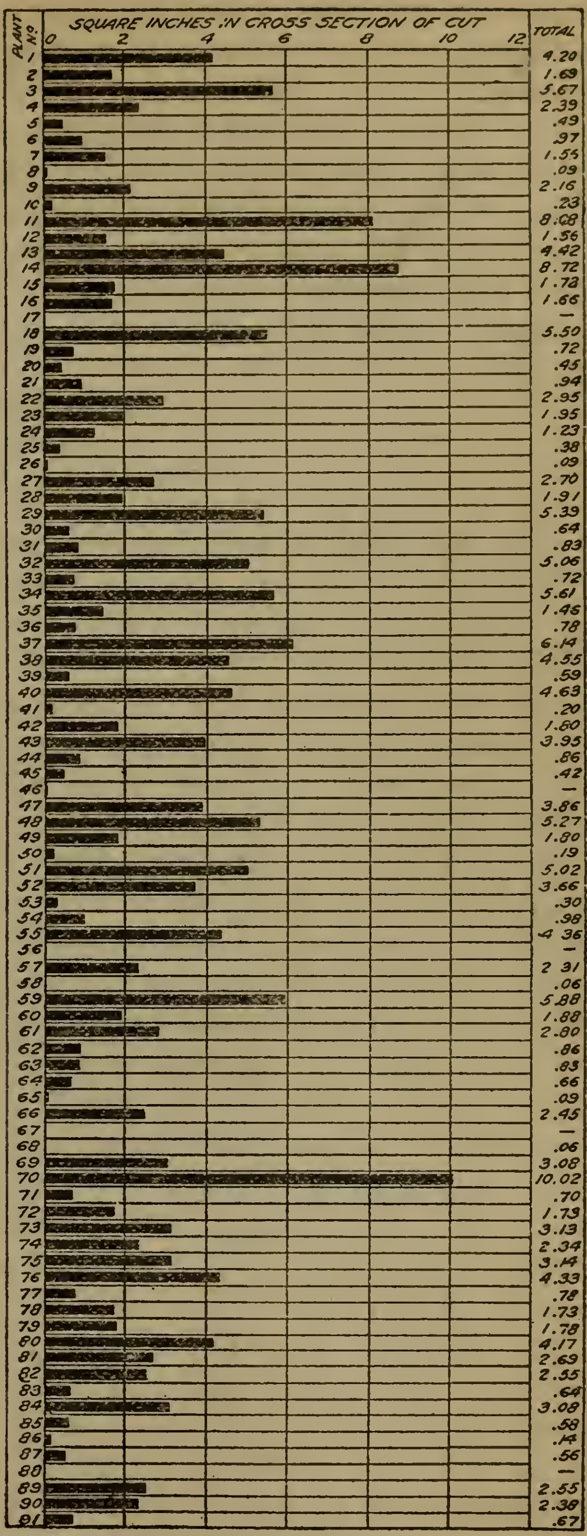


FIG. 1.—Diagram showing the comparative yields of individual plants of row A1, season of 1910. The yield is shown in the sum of the squares of the diameters of the stalks cut from each hill.

ricus has not been tested for rust resistance. Its size and hardiness indicate that valuable forms might possibly come from hybrids between it and *A. officinalis*.

A collection of forms of asparagus from all over the Old World is being made, and as these plants come into bloom hybrids will be made with *Asparagus officinalis* wherever possible. Of course, many species are not closely related to the cultivated forms and will not give fertile hybrids. These forms will be grown to determine their possible ornamental value and to aid in a systematic study of the group.

SELECTION.

PRELIMINARY METHODS.

The first work of selecting rust-resistant plants was begun in the fall of 1908. At that time no definite information was obtainable in regard to rust-resistant plants. It was not definitely known that the resistance was due to a character inherent in the plant, or to some local condition that rendered the plant immune for that environment or season only. The relative value of a resistant plant in a resistant lot as compared to an equally resistant plant in a lot or strain that averaged more rusty was unknown.

Several hundred marking stakes were made from ordinary lath sharpened at one end. These lath can be readily seen at some distance in an ordinary asparagus field. The experimental fields were gone over and every plant showing exceptional resistance was marked. In the fall of 1908 the variety test plats in fields A and B, comprising about 2 acres of different strains, were gone over in this way. Three acres of Argenteuil, Mr. Frank Wheeler's select stock, on the station grounds and several acres on the farm of Mr. Wheeler were also included. This work was repeated in 1909 and again in 1910. In 1909, selections were first made from the new plantings of Reading Giant and Late Argenteuil set out in permanent beds that spring. These beds have been included in the selection areas since that time. Besides some new untested plants, the selections of 1910 included only those plants of past years that had been progeny tested and proved of value as breeders. In 1909 many cross-pollinations were made between the select plants of 1908. Eleven hundred of these seedlings grown in the greenhouse were planted on the station grounds in 1910 in four rows with hills 1 foot apart in the row. These pedigree lots were included in the selections of 1910 and 1911.

GREENHOUSE INFECTION.

When the plans for rust-resistant breeding work were laid out in the fall of 1908 it was intended that the experimental infection of seedling plants to determine their relative resistance would be carried on during the winter months in the greenhouses at Washington. In this way it would be possible to gain a season. For various reasons this plan proved impracticable. The rust is not readily transferable in the greenhouses, owing to the lack of dew. The fact that uredospores do not germinate unless properly ripened on the host is another factor. During the fall of 1908 and again in 1909 the rusty plants brought into the greenhouses died back, so that the rust infection was lost and the new shoots coming up had no rust on them. The feeling that the different conditions of moisture, heat, etc., existing in the greenhouse might cause an entirely different rust relation as compared to that of the field has led to the dropping of this part of the plan. The work is now so far along that the greenhouse infection work is unnecessary.

JUDGING RUST RESISTANCE.

In the work at Concord the preliminary selection of breeding stock was begun in the fall of 1908 on fields A and B, planted in 1906 and 1907. The plants were marked for rust resistance on a scale of 0 to 10, the zero mark being used for a plant practically nonresistant and at the time of selection showing no green whatever as a result of a strong rust infection, 10 being the mark for a plant having no rust. The intermediate grades were assigned to plants showing intermediate degrees of infection according to the personal judgment of the observer. Experience in judging amounts of rust is required. The first season's marks are not as accurate as those of later years, because the plant as a whole rather than the rustiest stalk was then considered. The experience of later years has shown that the rustiest stalk in the hill is the best index, as many of the earlier stalks do not rust badly. They become hardened and seem more immune than the shoots that come up when the rust has become prevalent and is giving a strong infection. In making breeding-stock selections in the future no plants will be saved that do not show practical immunity to the rust.

In connection with the selection work the question was raised by visiting experimentalists as to the reliability of the methods used in marking resistant plants. In order to test the value of the marks assigned, row A1 was scored on two successive days, the second marking beginning at the other end of the row from the first so as to eliminate the factor of memory as far as possible. The result of the score is shown in a correlation table (Table III).

TABLE III.—*Correlation between two independent gradings of row A1 for rust resistance, September, 1911, to test reliability of values assigned by observer.*

[Coefficient of correlation 0.925±0.013.]

Grades.	First marking (grades).										Frequency.	Departure from mean.
	1	2	3	4	5	6	7	8	9	10		
Second marking:												
1	4										4	−5
2		3									3	−4
3			4	3							7	−3
4				3	2	1					6	−2
5				1	5	1	7				14	−1
6					2	3	2	2			9	0
7							3	7			10	+1
8							2	15	8		25	+2
9									2		2	+3
10										1	1	+4
Frequency	4	3	4	7	9	5	14	24	10	1	81	
Departure from mean	−5	−4	−3	−2	−1	0	+1	+2	+3	+4		

The greatest deviations are in the middle grades, but as these plants are not valuable for breeding parents the high correlation between the grading in the two sets of observations from a practical standpoint is higher yet. This method of checking up the value of grades assigned in selection work where the personal equation largely enters should be followed more extensively than it is at present. The ability to accurately judge differences of minor degree adds greatly to the value of the work.

RECORD OF SELECT PLANTS.

In making selections some permanent record is necessary. Permanent stakes in the field are not desirable, on account of the spring cultivation with a disk harrow. In our experimental plats the plants are placed at definite distances apart and a record made of a select plant, giving its row number and plant number to enable it to be relocated in the spring. Four-foot lath make cheap and convenient stakes which can be seen for some distance in the field. When plants in a cutting bed are to be left for seed, it is necessary to mark them so the cutters will let them alone.

PROTECTION OF SELECT PLANTS FROM FROST.

On account of late frosts occurring after the selected plants are up a foot or so, it is sometimes necessary to cover them over night. Manila paper bags are very good for this purpose. When the shoots were covered with moist earth at times when the temperature went low enough to make ice one-fourth of an inch thick, the stalks froze and when the sun came out they thawed out rapidly enough to be killed. At the same time the shoots under the manila bags escaped injury. A peculiar frost phenomenon was observed in June, 1910, when a light frost occurred in the lowest portions of one of the breeding fields. Glassine paper bags, which are used to cover flowering branches on female plants in breeding work, instead of protecting the flowers from frost actually increased the injury, so that the flower buds dropped off under the bags, while the unprotected buds outside remained uninjured.

RUST INFECTION TO SECURE SELECT RESISTANT PLANTS.

During the past three seasons there has been an abundance of rust during July, August, and September on the experimental grounds at Concord. This abundant attack of rust is necessary to obtain selections of any practical value. The attack of rust should deaden the tops of practically 95 per cent of the young seedlings from standard Argenteuil stock in the seed bed, or in its ravages in a commercial field of good Argenteuil make it possible for a beginner to pick out less than 10 resistant plants per acre as being plainly superior in rust resistance to the other five or six thousand.

So far in this work no asparagus plants have been found that will not rust to some extent. There is a wide difference in susceptibility in different varieties. The old American sorts represented in Concord by Moore's crossbred are practically nonimmune, while Argenteuil and other related European varieties are highly immune, so much so that they are not troubled by rust unless a new bed planted near by is not being cut in the spring.

There is no question that the spread of the rust from one field to another depends on the direction and intensity of air currents. On the experimental field at Concord the prevailing winds are from the northwest. This fact, combined with the circumstance that the dew dries up last on the northwest side of the plant, makes the heaviest attacks of rust on the shady side. On account of the direction of the prevailing wind at Concord, it is highly advisable to have any infection area on the north or west of the seedling bed to be infected. Once a field has had a good infection of rust and the resistant plants marked, it is not necessary to provide rust in future seasons, as the select plants can be tested by growing pedigree seedlings.

In fact, it is not necessary to test the individual resistance of a plant in order to determine its value as a breeding parent. All that is necessary is to test a small lot of its seedlings.

CAUSES OF RESISTANCE.

Æcidial stage on resistant plants.—An interesting feature of the rust-resistant breeding work developed in the spring of 1909, when the cluster-cup stage of the rust appeared. The plants that had been selected as rust resistant in 1908 were allowed to grow without being cut. In addition to these selected plants several nonresistant plants were allowed to grow up to be used in crossing tests to determine the dominance of the resistant character. No uniform differences in the æcidial infection could be noticed, many of the most resistant plants having a better infection than the rusty plants. This development caused some doubt as to the nature of resistance and made it seem possible that the immunity of the year before might be due to some temporary factor. Later in the season when the summer stages of the rust appeared this doubt was dissipated, as the resistance again appeared in the select plants of the year before in about the same degree as in the previous season.

Relation of structure to resistance.—The fact that sporidia from germinating teleutospores can infect through the epidermis without necessarily entering through the stomatal openings gives an explanation for the phenomenon just noted and sheds a possible light on the cause of rust resistance in asparagus.

It is a well-known fact that in the heterœcious rusts the æcidial stages occur on plants widely different in general character from their hosts when in the uredo or teleuto stages. Thus there is reasonable ground on which to oppose the theory that rust resistance is due to structural differences simply because the æcidial stage appears on resistant plants as freely as on nonresistant ones. However, the theory that in asparagus resistance has a morphological cause is reenforced by several other points. While little work has been done on this problem in asparagus, the evidence tends to show that resistant plants have smaller stomata than the nonresistant ones. It may be, of course, that the size of guard cells is not closely correlated with the actual size of the opening through which the mycelium must pass, but it gives a suggestive point of attack in solving the problem. When the rust develops in a field in summer, the shoots that came up first and have fully matured and hardened develop a lighter attack of rust than the shoots which appear during the height of the rust epidemic. Once the rust gets started in the plant it goes ahead in its development equally well in resistant and rusted plants, no difference being discernible in the type or vigor of the individual sori on plants of different degrees of resistance.

Ward,[1] in his studies of rust resistance in the genus Bromus, comes to the conclusion that resistance is not due to structural causes. He says: [2]

> We are hence driven to conclude that the factors which govern predisposition on the one hand and immunity on the other are similar to those which govern fertility and sterility of stigmas to pollen * * *.

[1] Ward, H. M. On the Relations between Host and Parasite in the Bromes and Their Brown Rust, Puccinia Dispersa (Erikss.). Annals of Botany, vol. 16, 1902, pp. 233–315.

[2] Ward, H. M. Op cit., p. 314.

In a later study of rust resistance Ward[1] says tha this researches "clearly led to the conclusion that the matter has nothing to do with anatomy, but depends entirely upon physiological reactions of the protoplasm of the fungus and of the cells of the host."

Until sufficient evidence has been accumulated on the correlations between structure and rust resistance in asparagus the writer does not care to claim definitely that the size of the stomata is related to the phenomenon of resistance to the attacks of the uredo stage of the asparagus rust. The presence of the æcidial stage on the asparagus plant gives a point of attack in the search for the cause of immunity that is not found in the heterœcious rusts of grasses, and when the studies on this point have been completed it is hoped that new light, at least, will be thrown on the question of disease resistance.

Relation of vigor to resistance.—The theory that vigor of growth is correlated with resistance, as suggested by some American writers on the subject, can not be accepted, for many resistant plants are quite small and never produce strong shoots. The trials of the last two seasons of two equally resistant strains of Argenteuil stock from local growers at Concord show no relation between resistance and vigor. About 450 one-year-old seedlings of each strain were planted in 1908 side by side on uniform land and under uniform treatment. When they were cut for a short time in 1910 each day's yield was separated into giant and common grades, using the local grading system. One lot gave a total yield over a period of 35 days, from April 23 to May 28, of $142\frac{9}{16}$ pounds divided into $106\frac{13}{16}$ pounds of giant and $35\frac{3}{4}$ pounds of common. The second lot gave in the same period only $65\frac{13}{16}$ pounds total cut divided into $14\frac{1}{2}$ pounds giant and $51\frac{5}{16}$ pounds common. The details of the record are presented in Table IV.

[1] Ward, H. M. Recent Researches on the Parasitism of Fungi. Annals of Botany, vol. 19, 1905, p. 21.

TABLE IV.—*Yield from five 300-foot rows of Argenteuil asparagus, showing comparison of large and small strains, seasons of 1910 and 1911.*

Date.	Large strain.						Small strain.					
	Giant.			Common.			Giant.			Common.		
	Weight.			Weight.			Weight.			Weight.		
	Pounds.	Ounces.	Stalks.	Pounds.	Ounces.	Stalks.	Pounds.	Ounces.	Stalks.	Pounds.	Ounces.	Stalks.
1910.[1]												
April:												
23	1	14	20	0	8	15	0	0	0	0	11	20
25	1	13	20	0	9	19	0	8	7	1	3	29
27	5	10	55	2	8	57	0	13	11	2	1	61
28	6	2	63	1	4	34	0	10	9	2	1	76
May:												
2	14	7	126	2	8	65	1	14	23	6	5	182
4	7	0	73	2	2	45	1	9	20	2	8	60
10	5	7	45	1	4	29	0	12	9	1	11	41
13	11	6	96	3	2	73	0	10	9	4	4	135
16	7	10	70	2	5	74	1	5	16	3	12	120
18	8	3	72	2	6	66	1	14	25	4	7	142
20	4	2	38	1	5	35	0	15	12	2	15	91
21	4	10	45	2	10	66	0	10	8	2	13	108
23	6	1	55	2	4	60	0	13	10	3	13	123
24	4	10	42	6	2	54	0	7	6	2	14	101
25	6	15	71	3	1	90	0	8	7	4	2	146
26	3	12	40	1	6	48	0	7	6	2	3	85
27	2	15	31	2	5	51	0	8	6	1	9	50
28	3	4	31	1	15	55	0	5	4	2	1	69
Total	106	13	1,073	35	12	936	14	8	188	51	5	1,739
1911.[2]												
May:												
6	0	11	10	0	5	10	0	2	2	1	0	32
8	6	10	71	3	7	82	1	8	22	4	7	153
9	8	9	82	3	8	85	1	4	15	5	0	133
10	15	5	127	3	12	86	2	8	28	8	10	208
11	21	8	189	4	8	105	4	11	51	8	10	233
12	25	7	211	3	6	84	4	4	50	7	10	214
13	16	10	142	2	13	61	2	12	30	5	11	147
14	12	3	99	1	8	41	2	8	34	2	2	65
15	8	1	72	2	0	45	2	11	33	2	11	80
16	10	13	89	2	7	61	2	1	25	4	8	129
17	8	4	67	2	7	59	2	7	30	5	1	132
18	9	3	83	2	10	62	1	9	18	4	12	132
19	8	3	74	2	13	80	1	10	18	5	11	146
20	6	0	58	1	7	33	1	6	17	2	14	78
21	11	10	103	3	8	88	2	9	27	7	10	192
22	8	9	77	5	1	117	2	11	28	9	5	246
23	8	5	74	2	8	69	2	0	23	5	4	161
24	1	3	13	0	10	20	0	1	1	0	9	23
26	8	14	79	1	10	42	3	6	39	4	0	112
27	7	14	72	1	13	44	2	13	34	4	0	108
28	13	9	121	5	1	113	3	4	38	8	3	224
29	9	7	89	5	10	140	2	10	33	5	13	184
30	7	4	60	4	13	125	1	5	16	4	8	128
31	5	6	48	3	3	70	1	11	19	3	14	101
June:												
1	7	0	60	3	5	83	1	11	21	6	3	176
2	6	6	53	2	8	67	0	11	9	3	12	114
3	8	2	80	3	6	98	1	10	20	3	3	108
4	4	12	48	2	11	67	0	11	9	1	15	49
5	6	4	62	1	13	56	1	6	18	3	6	104
7	5	9	51	1	14	55	2	0	23	3	14	113
8	3	0	30	1	3	37	1	9	20	2	12	103
9	5	14	59	2	9	67	1	7	20	3	10	105
10	7	10	67	4	4	111	1	15	23	5	9	164
11	4	0	40	3	5	88	1	13	20	2	1	50
12	4	12	47	1	1	20	1	2	16	4	2	137
14	2	13	30	1	11	48	1	3	15	2	6	70
Total	305	10	2,737	100	6	2,519	70	12	845	164	10	4,654

[1] Total yield in 1910: Large strain—2,009 stalks, weighing 142 pounds 9 ounces (average weight per stalk, 1.14 ounces); small strain—1,927 stalks, weighing 65 pounds 13 ounces (average weight per stalk, 0.54 ounce).

[2] Total yield in 1911: Large strain—5,256 stalks, weighing 406 pounds (average weight per stalk, 1.23 ounces); small strain—5,449 stalks, weighing 235⅜ pounds (average weight per stalk, 0.69 ounce).

The results in 1911, showing the yield for a full season, were more conclusive. Again lot 1 was far superior in size and total yield, giving 406 pounds total, of which 305$\frac{5}{8}$ pounds were of giant grade and 100$\frac{3}{8}$ pounds common. Lot 2, while actually producing more stalks in the season, had only 235$\frac{3}{8}$ pounds total, 70$\frac{3}{4}$ pounds giant and 164$\frac{5}{8}$ pounds common. The record for 1911 is also shown in Table IV. If there was any difference, the advantage in rust resistance is in favor of lot 2; moreover, about 10 of the best plants were reserved out of lot 1 as breeding parents and the cut is thus perceptibly reduced. In the region around Concord it has been noticed frequently that the poorer parts of the field had less rust when other conditions were equal, so that the application of chemical fertilizers has been held by some farmers to be the cause of the rust.

BREEDING.

The real work of breeding started in the spring of 1909. Many questions of importance in regard to methods had to be settled, for to a certain extent we were on unknown ground. Asparagus was generally recognized as a diœcious plant, but several writers and observers had suggested that parthenogenetic seeds were sometimes produced. The relative dominance of rust resistance in heredity was uncertain in asparagus. Biffen[1] in his work with disease resistance in the small grains had shown an apparent dominance of susceptibility, but in asparagus there is no question at present that the heterozygous forms are intermediate in resistance. The possibility of obtaining a combination between strains that would give first-generation hybrid vigor was important, and above all was the hope of finding two parent plants that would give a highly uniform progeny in rust resistance and vigor.

In starting the work a study had to be made of the natural and artificial methods of pollination. Means had to be devised to control the pollination work so that reliable pedigrees could be established. The paragraphs that follow comprise an account of the observations made and the resulting methods developed and now in practice on the different phases of these problems.

SEX.

Asparagus officinalis is functionally diœcious, but the flowers on both types of plants contain rudiments of the organs of the opposite sex. Under field conditions asparagus apparently requires the aid of insects to secure proper pollination. As a rule, no seed is set without the aid of bees and other insects carrying pollen from the flowers

[1] Biffen, R. H. Journal of Agricultural Science, vol. 1, 1905, p. 40; vol. 2, 1907, pp. 109–128.

of male plants to the stigmas of the flowers on the female plants. Hermaphrodite plants occur now and then, but so far in our experiments can not be considered a factor in seed production. In the flowers of the typical female plant the rudiments of stamens (Pl. VII, fig. 2) exists, but the writer has never seen one developed sufficiently to even suggest the possibility of self-pollination. On the other hand, the male plants often show a well-developed ovary with style and stigma and sometimes even a typical stigmatic surface. The great majority of male flowers, however, lack a well-developed ovary, the rudiment being about half the size of the normal ovary of the female flower and lacking any stigmatic development (Pl. VII, fig. 1), the style often being completely abortive. The hermaphrodite plants mentioned above are always of the male type, the flowers being for the greater part pure male in that they lack the complete and functional ovary. In one wild male plant examined the flowers at the extremities of the branches were typically female with well-developed stigma and abortive anthers. This male had been used for pollination work in testing rust resistance of select plants. Another hermaphrodite plant which produced seed that would germinate and make healthy, vigorous plants had many flowers whose ovaries lacked the stigmas. The berries on these hermaphrodite plants are very small and rarely have more than one seed in them. The seeds are usually peculiar in that the seed coats are not entirely developed. The seeds appear mottled black and white, varying from the white of the uncovered endosperm in the smaller seeds to well-covered, entirely black seeds in which the coats have had their normal development and have completely covered the endosperm (Pl. IX, fig. 1). These small seeds make weak plants and in many cases abnormal ones, but the larger, better developed seeds make healthy seedlings of normal type. As yet these seedlings have not been observed in bloom, so the sex inheritance is unknown.

POLLINATION.

During the blooming period of 1909 and again in 1910 branches of pistillate plants were covered with paraffin paper bags to exclude pollen-carrying insects (Pl. VIII). Although more than a hundred of these check trials were made, in no case did any seed set from flowers that opened under the bags. The ovaries would swell and apparently start to develop good berries, but after reaching about one-third of the ordinary diameter they would turn yellow and drop off. The uncovered flowers on the same stem set seed abundantly (Pl. IX, fig. 2). That this failure to set seed is due to a lack of pollination is shown by the large number of seeds secured under bags when

FIG. 1.—MALE FLOWERS, THE LOWER WITH SOME OF ITS PERIANTH LOBES REMOVED TO SHOW THE STAMENS AND RUDIMENTARY OVARY. THE PERIANTH LOBES ARE LONGER THAN IN THE FEMALE FLOWER. X 5.

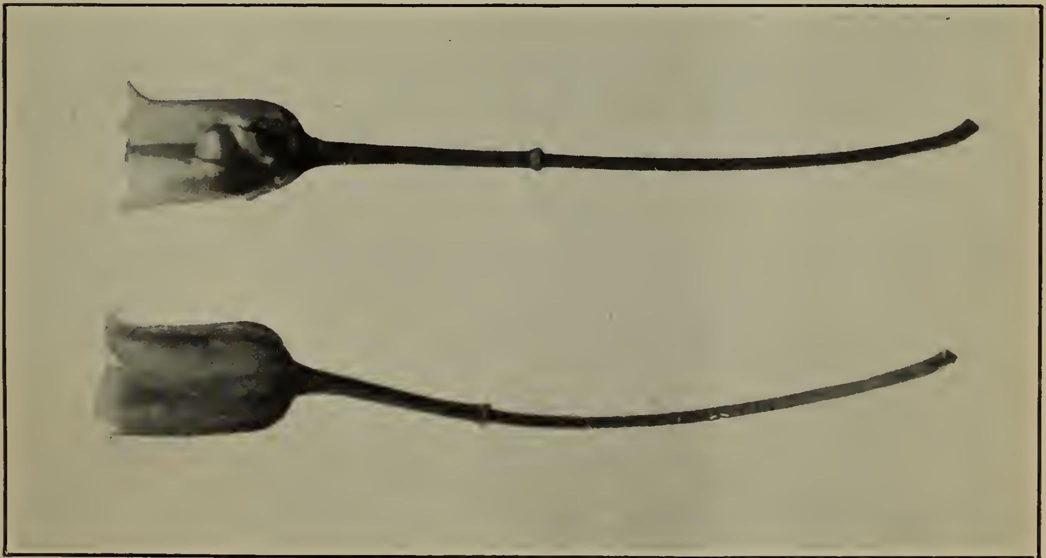


FIG. 2.—FEMALE FLOWERS, THE UPPER WITH SOME OF ITS PERIANTH LOBES REMOVED TO SHOW THE OVARY AND RUDIMENTARY STAMENS. THE PERIANTH LOBES ARE SHORTER THAN IN THE MALE FLOWERS. X 5.

FLOWERS OF ASPARAGUS OFFICINALIS.

FEMALE ASPARAGUS PLANT WITH BRANCHES COVERED BY "GLASSINE" BAGS TO KEEP INSECTS FROM POLLINATING THE FLOWERS WITH POLLEN FROM UNKNOWN MALES, THIS PLANT BEING USED TO TEST THE COMPARATIVE RESISTANCE TRANSMISSION OF SEVERAL MALES.

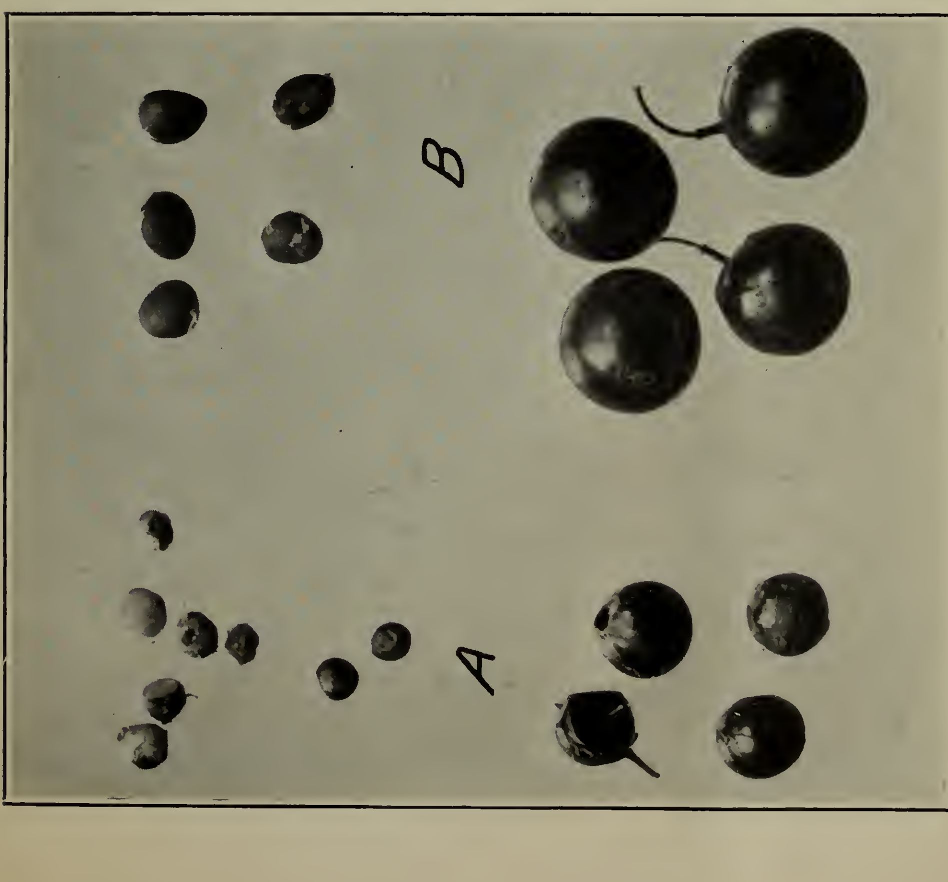


FIG. 1.—ASPARAGUS FRUIT AND SEED. *A*, FROM A HERMAPHRODITE PLANT. EACH BERRY CONTAINS ONE SEED WHICH OFTEN LACKS ALL OR PART OF THE BLACK SEED COAT. *B*, FROM A NORMAL PLANT.

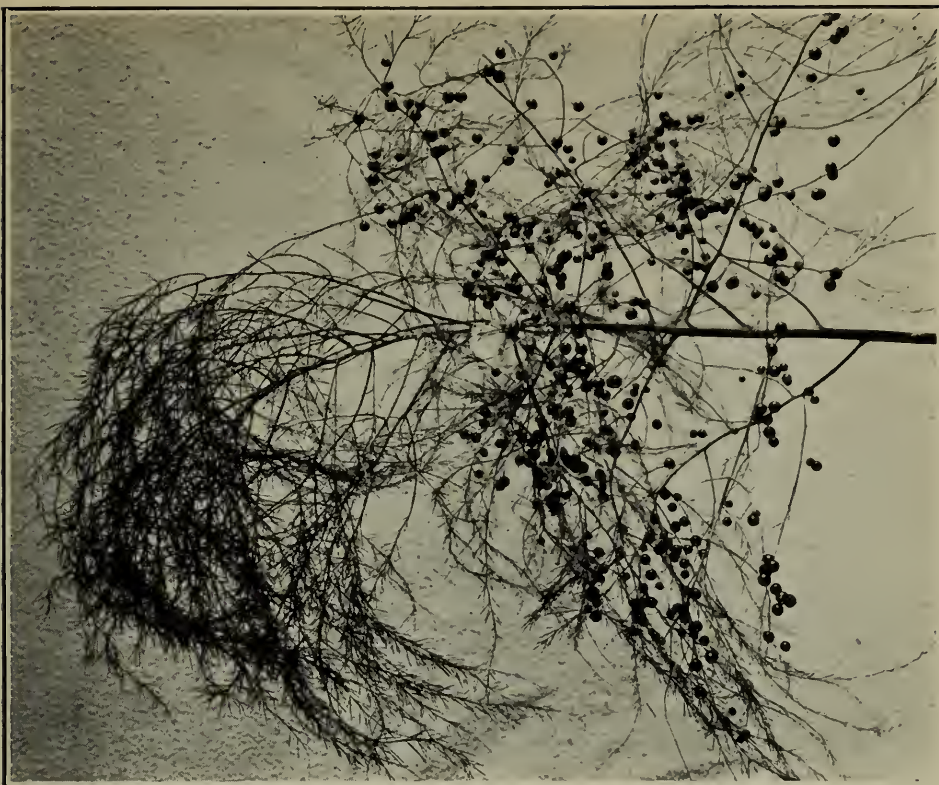


FIG. 2.—ASPARAGUS STEM, SHOWING THE EFFECT OF BAGGING FLOWERS WITHOUT POLLINATION. THE TOP OF THE STEM WAS COVERED DURING THE BLOOMING SEASON, WHILE THE LOWER BRANCHES BLOOMED IN THE OPEN AND WERE POLLINATED BY BEES.

FIG. 1.—SEEDLINGS OF 1909, IN SEPTEMBER, 1909, AFTER A SEVERE ATTACK OF RUST. ROWS 3, 5, 6, 7, AND 12 SHOW RESISTANCE.

FIG. 2.—PEDIGREE SEEDLINGS OF 1910, IN SEPTEMBER, 1910, B136-4 × A7-83, SHOWING RUST RESISTANCE.

FIG. 3.—PEDIGREE SEEDLINGS OF 1910, IN SEPTEMBER, 1910, B136-4, OPEN FERTILIZED, SHOWING LACK OF RESISTANCE AND VIGOR.

ASPARAGUS SEEDLINGS, SHOWING COMPARATIVE RUST RESISTANCE.

the flowers were hand-pollinated. The insect visitors to the asparagus flowers are largely bees of different kinds. The honey bee is most plentiful during the blooming season, and at this time of the year practically all of the pollen that comes into hives that are near asparagus fields is the rich, orange pollen from the staminate asparagus flowers. Apparently a large amount of nectar is also produced. This is shown particularly in flowers under bags where the bees have been kept away until the flower is old, when it is so abundant as to interfere with pollination. In addition to the honey bee many small wild bees frequent the asparagus flowers. Some of these small bees are a nuisance in the pollination work, apparently being especially attracted by the extra quantity of honey in the protected flowers.

The wind seems to play little part in pollination, as the male flowers retain their load of pollen until they begin to wither unless it is removed by a bee. The pollen hangs together in masses and does not become powdery until the flower dries up.

The anthers of the staminate flowers dehisce throughout the day, but by far the greater portion open in the morning hours. On a bright, sunny day they are nearly all open by the time the dew is gone. Cold, damp weather seems to prevent the anthers from opening, so that after a long spell of rainy weather a large number of flowers will be spoiled, as the anthers do not open well except when fresh. There is quite a range of variability in this respect and also as to the time the anthers open in the morning. Some days when the atmosphere was moist some staminate plants could not be used in pollination work until an hour or so after most of the others, and on misty or damp, cloudy days these plants would refuse to shed their pollen at all, while plants near them would yield abundant supplies.

METHODS OF HAND POLLINATION.

In the work of hand pollination in asparagus very little apparatus is required. The principal requirement is to prevent the random pollination of the pistillate flowers by insect visitors. So far in this work we have largely depended on paraffin paper bags. A good quality of paper known as "glassine" is used. This paper is nearly transparent and is tougher and wears better than any other paraffin paper obtainable at a reasonable price. During the spring of 1910 many of these bags went through a three-days' storm of wind and rain without injury. The bags are attached and held on by short pieces of No. 18 office wire to which is attached a small eyelet string tag for records. The office wire is purchased in small rolls and cut up with a pair of shears into pieces about 4 inches long. The bags are placed over the branches of the female plant, gathered at the mouth, and firmly held by bending a piece of wire around the bag about an inch or two above the mouth. Ordinarily two branches are included in the bag in order to brace each other and prevent the wind from whipping the bags around or breaking the stems. This wire is a great time saver over twine, as it does away with tying and untying at a time when minutes are valuable. This wire is also used in tying the main stalks to stakes to keep them standing against storms. Plain copper wire of equal weight is apt to cut and wear into the stems or through the bags.

In the season of 1911, cages constructed of ordinary fly-screen wire cloth were used to cover the select plants both male and female. It was discovered later in the season that a very small wild bee was squeezing in through the meshes and pollinating flowers. In the future a finer mesh copper-wire screen will be used. The season of 1911 was exceptional for days of extreme heat. Many of the branches inclosed under the glassine bags died after they had set fruit. Such berries never developed viable seed. Asparagus throughout its development is very partial to good ventilation and any protective measures for pedigree work must take this into account.

The female plants to be tested can be bagged about the time the first flowers are ready to open. At this time the branches are tough enough to bend readily without breaking. Flowers that have opened are picked off before the branch is bagged.

The work of pollination is best done in the morning hours just after the dew is gone. The desired male plants are visited and a collection made of flowers undisturbed by bees or other insect visitors. These flowers are placed in small shell vials lined with absorbent paper and with a stopper of absorbent cotton. The vial keeps them from drying out and losing their pollen and from becoming mixed with flowers from other plants. All vials used should be properly labeled to prevent mistakes.

In selecting male flowers only those are taken whose anthers have dehisced their pollen. The pollen in a freshly opened flower clings in an orange mass around the stamens at the throat of the bell-shaped flower. Flowers that have been visited by bees have lost most of this mass and the lighter color of the anther walls gives a much lighter color to the anther cluster. These flowers should be rejected, as they are liable to be mixed with foreign pollen.

In pollinating, the bag is removed from the female branch. A flower from the bottle of male flowers is taken out and grasped lightly in the right hand, using the thumb and finger or a pair of forceps. With the left hand a flower on the female branch is carefully bent into such a position that it can be touched with the pollen mass in the male flower. If the flowers are brought lightly together so as not to injure the stigma, the stigmatic surface will be well covered with pollen and in the course of a few days the ovary will swell into a full-sized berry. With a little practice pollination becomes a matter of routine work. The whole male flower is used in pollination, and this method has proved very satisfactory, doing away with the use of brushes or other pollinating devices which are apt to cause mixing of pollen from different males. After all the female flowers that are open are pollinated the bag is replaced and the tag marked with identifying data. One male flower will usually pollinate ten or more female flowers, leaving enough pollen on each stigma to be plainly visible to the naked eye.

CARE OF SEED.

After the various pollinations are made on select plants in spring and early summer considerable time must elapse before the seed is ripe and ready for harvest. Cultivation is apt to break off branches unless great care is used. Asparagus beetles must be guarded against. The various accidents considerably reduce the percentage of seed set. The berries should not be harvested until they are ripe and soft, otherwise the seed is apt to be shriveled and of poor quality. The seed when in lots of less than a hundred berries is harvested by picking the berries from the plants in the field and placing them in small manila bags, which are labeled on the outside with sufficient data to distinguish the different lots and stored temporarily in racks in a well-ventilated room. After the berries have begun to wither they can be stored in ventilated storage boxes without injury until such time as it is convenient to clean them. The lots of berries are crushed and washed in water, the pulp and skins washed out, and the clean seed allowed to dry thoroughly. The seed is then placed in shell vials, labeled and corked, and stored for next year's planting. The seed of *Asparagus officinalis* retains its viability for several years if properly handled.

METHOD OF TESTING PROGENY.

The first work in testing transmission of relative rust resistance was in the summer of 1909. The previous fall samples of seed had been saved from various plants of different degrees of resistance. Twelve lots of these seeds were planted in duplicate in short rows in seed flats on July 13. When the shoots appeared, July 26, and for several succeeding days, fresh uredospore material was dusted over the flats. The following table shows the relative rust resistance of the rows of these seedlings from observations made September 3, 1909.

TABLE V.—*Duplicate test of seedlings of 1909 from plants of varying rust resistance to show relative transmission of susceptibility.*

Row.	Source of seed.	Type of plant.	Rank of seedlings in resistance.		
			First lot.	Second lot.	Average.
1	A1–6	Badly rusted, near rusty bed	7	9	8
2	A3–61	Very resistant female	6	5	5.5
3	A4–7	Resistance good	3	7	5
4	A4–17	Resistance fair	10	8	9
5	A7–5	Resistance good	4	3	3.5
6	A7–15	do	2	4	3
7	A7–25	do	5	2	3.5
8	B24–27	Very rusty	11	10	10.5
9	B24–28	do	9	11	10
10	Old field	Rusty	12	12	12
11	do	Resistant	8	6	7
12	Frank Wheeler; old bed	Best resistant female	1	1	1

An inspection of the table shows that the asparagus plants transmit resistance to their offspring in about the same relative degree that they resist the rust themselves. Plate X, figure 1, is from a photograph taken in September, 1909, of lot 1 of this set of duplicates, showing plainly the effect of rust on the lots.

This experiment settled the question of the value of rust resistance in the plant as a mark of transmitting power. After this preliminary trial, plans were laid to test the lots of seed that were obtained from our hand pollinations in 1909. In addition to the hand-pollinated lots a sample of open-fertilized seed has usually been saved for comparison in the progeny tests.

In January, 1910, as many lots of seed as could be conveniently handled were planted in seed flats in the greenhouse at Washington. Studies of the germination and growth of these lots were made and correlations measured between various characters. In the discussion on the following pages these seedlings are referred to as the greenhouse seedlings of 1910 to distinguish them from other lots of asparagus plants under observation.

USE OF BIOMETRY.

CORRELATION STUDIES.

The use of statistical methods in breeding is becoming more and more popular and in many lines is really necessary. The presentation of biometrical studies is now rather common in experiment-station bulletins. If breeding is to be put on a progressive scientific basis this type of work will of necessity become more prevalent. The excessive presentation of correlation studies as such should be discouraged among practical workers. The value of any work in correlation depends on the possible use that can be made of the facts in a practical way. The great number of interrelated biologic phenomena that occur in any plant need to be understood before one character can be used as a basis for selection for a correlated character.

In the study of a plant like asparagus, where selections must be made for rust resistance and yield, two desired characters which do not show simultaneously, in order to do efficient work some other correlated character must be substituted for yield. Again, the young seedling must be compared with the bearing plant several years later so that undesirable stock can be discarded without the necessity of planting large fields with plants of unknown qualities. Before any correlation studies were made all stocks were planted in permanent beds and grown for years without selection. Since 1910 no untested plants have been carried beyond the first season in the seed bed, all the poor stock being discarded the first year. In this way a great saving in field space is accomplished. Of course, care must be used in basing selections on one character to get plants good in another character. Where the correlation runs above 0.75 very good results are usually obtained.

Correlation tables serve a good purpose in checking up the reliability of tests. The use of duplicate plantings or trials year by year to secure data on average performance are valuable only as they show a high correlation.

Correlation tables are used in our breeding wherever possible to show the reliability of the observations made. This feature is illustrated in Table VI, in which the heights of several lots of the greenhouse seedlings on February 7, 1910, are correlated with the heights of the same lots four days later, showing that the observations taken on either of the two dates were satisfactory.

TABLE VI.—*Correlation between the heights of the tallest plants in 66 lots of progeny rows of greenhouse seedlings of 1910, taken four days apart, February 7 and February 11, 1910.*

[Heights on February 11, subject; heights on February 7, relative. Coefficient of correlation, 0.941±0.013.]

Heights on February 11.	Heights on February 7 (¼-inch units).																			Frequency.	Departure from mean.
	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31		
¼-inch units:																					
18	1																			1	−12
19																				0	−11
20			1																	1	−10
21																				0	− 9
22																				0	− 8
23																				0	− 7
24								1												1	− 6
25							1			1										2	− 5
26									2											3	− 4
27										2	2									4	− 3
28										1		3		1	1					6	− 2
29												5	1	1	1					8	− 1
30												1	1	3	1					6	0
31													3	4	4	2				13	+ 1
32														1	4	3		1		9	+ 2
33															2	1	1			4	+ 3
34															1	1	1	1		4	+ 4
35																		2		2	+ 5
36																	1			1	+ 6
37																	1		1	2	+ 7
Frequency	1	0	1	0	0	0	1	1	2	4	2	9	5	10	14	7	4	4	1	66	
Departure from mean	−12	−11	−10	−9	−8	−7	−6	−5	−4	−3	−2	−1	0	+1	+2	+3	+4	+5	+6	....	

DUPLICATE TESTS.

The use of duplicate plats in testing varieties or in breeding or selection is important in checking up results. Unless the correlation is high, say around 60 to 70 per cent, the test should be looked on with doubt. When these data are presented in the form of two overlaid curves, the observer is liable to be misled, as the correlation is hard to judge; but this form of presentation is used by many writers, is easily constructed, and can be used to show the relation of more than two characters at the same time. Still, the fact remains that the measure of the correlation is indefinite.

Duplicate plantings were made of 33 lots of the greenhouse seedlings, and from them many interesting facts were developed. Table VII presents the correlation between the tallest plants in the duplicate rows from measurements taken February 11, 1910. The correlation between the average height of all the plants in the rows on the same date is shown in Table VIII, and Table IX shows the correlation between the tallest plant in the row and the average of all the plants in the row. In dealing under ordinary circumstances with progeny lots of plants varying in a normal way, these tables show whether rapid selections can be based on the best individual in each lot. It appears that there is a high probability that an experimenter is perfectly safe in basing selections of future progeny lots on a comparison of the means or of either the high or the low extreme.

TABLE VII.—*Correlation between the tallest plants in two lots of duplicate plants of progeny lots of greenhouse seedlings of 1910, February 11, 1910.*

[Lot 1, subject; lot 2, relative. Coefficient of correlation 0.876±0.028.]

Height of first lot.	Height of second lot (¼-inch units).																			Frequency.	Departure from mean.
	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40		
¼-inch units:																					
19	1																			1	−14
20																				0	−13
21																				0	−12
22																				0	−11
23																				0	−10
24																				0	− 9
25																				0	− 8
26						1														1	− 7
27																				0	− 6
28																				0	− 5
29							1		1											2	− 4
30								1												1	− 3
31									1		1		1							3	− 2
32									1	1	2									4	− 1
33										3			2							5	0
34										1	1			1						3	+ 1
35													1	2	1		1			5	+ 2
36															1		2			3	+ 3
37													1	1					1	3	+ 4
38															1					1	+ 5
39																				0	+ 6
40																				0	+ 7
41																1				1	+ 8
Frequency	1	0	0	0	0	1	1	1	3	5	4	0	5	4	3	1	3	0	1	33	
Departure from mean	−11	−10	−9	−8	−7	−6	−5	−4	−3	−2	−1	0	+1	+2	+3	+4	+5	+6	+7	......	

TABLE VIII.—*Correlation between the average heights of greenhouse seedlings of 1910 in two duplicate lots of progeny, February 11, 1910.*

[Second lot, subject; first lot, relative. Coefficient of correlation 0.826±0.035.]

Average height of second lot.	Average height of first lot (¼-inch units).																	Frequency.	Departure from mean.
	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33		
¼-inch units:																			
20	1																	1	−8
21																		0	−7
22																		0	−6
23																		0	−5
24								1		2								3	−4
25																		0	−3
26										3			1					4	−2
27										2		1	1	2				6	−1
28										1	2			2	1			6	0
29													2		1			3	+1
30											1			1		2		4	+2
31													1	1	1			3	+3
32																		0	+4
33															1		1	2	+5
34																	1	1	+6
Frequency	1	0	0	0	0	0	0	1	0	8	3	1	5	6	4	2	2	33	
Departure from mean	−11	−10	−9	−8	−7	−6	−5	−4	−3	−2	−1	0	+1	+2	+3	+4	+5	....	

TABLE IX.—*Correlation between height of tallest plant and mean height of progeny rows of greenhouse seedlings of 1910, February 11, 1910.*

[Mean height, subject; height of tallest plant, relative. Coefficient of correlation 0.901±0.016.]

Mean height of row.	Height of tallest plant in row (inches).																							Frequency.	Departure from mean.
	4.75	5.00	5.25	5.50	5.75	6.00	6.25	6.50	6.75	7.00	7.25	7.50	7.75	8.00	8.25	8.50	8.75	9.00	9.25	9.50	9.75	10.00	10.25		
Inches:																									
4.25	1																							1	−2.75
4.50																								0	−2.50
4.75																								0	−2.25
5.00				1																				1	−2.00
5.25																								0	−1.75
5.50																								0	−1.50
5.75								1																1	−1.25
6.00									1	1		1												3	−1.00
6.25																								0	− .75
6.50											3	3	3	2	1									12	− .50
6.75													3	3	1	2				1				10	− .25
7.00													2	3		1								6	0
7.25															2		5			1				8	+ .25
7.50															1	4	3	1	1			1		11	+ .50
7.75																	1	4		1				6	+ .75
8.00																			2					2	+1.00
8.25																			1	2			1	4	+1.25
8.50																		1						1	+1.50
Frequency	1	0	0	1	0	0	0	1	1	1	3	4	8	8	5	7	9	6	4	5	0	1	1	66	
Departure from mean	−3.50	−3.25	−3.00	−2.75	−2.50	−2.25	−2.00	−1.75	−1.50	−1.25	−1.00	− .75	− .50	− .25	0	+ .25	+ .50	+ .75	+1.00	+1.25	+1.50	+1.75	+2.00		

USE OF CORRELATION STUDIES IN BREEDING WORK.

The value of studies in correlation to the practical work of plant improvement, while not questioned by those familiar with statistical methods, has been doubted by those not in the habit of doing accurate work. One or two examples will show the possibilities of the use of correlation.

The relation of the size of the asparagus stalk in the fall to the next year's cut is interesting, as it is necessary in selecting rust-resistant plants in the fall to pick those that will give large-sized shoots. Studies of several plants in row A1 presented in Table X give a fair idea of the value of large-sized fall growth in determining large-sized spring growth. In the same way the total yield should be taken into consideration. This quality is hidden at the time of selection and must be correlated with total production of stalks in the field in the fall. This relationship was worked out with the plants in row A1 and the result is shown in Table XI. Certainly when the correlation is as high as 0.8 the observer should make an almost perfect selection by using the correlated character to pick the very best plants.

TABLE X.—*Correlation between the diameters of the largest stalks in 86 hills of row A1 in the fall of 1909 and in the largest stalks cut in the spring of 1910.*

[Diameters of 1909, subject; diameters of 1910, relative. Coefficient of correlation 0.575±0.050.]

Fall of 1909.	Spring of 1910 (⅛-inch units).											Frequency.	Departure from mean.
	2	3	4	5	6	7	8	9	10	11	12		
⅛-inch units:													
2	....	....	1	....	....	....	....	....	....	....	....	1	−2
3	1	1	8	7	3	....	....	....	....	....	....	20	−1
4	1	4	3	6	12	5	1	....	....	....	....	32	0
5	....	1	2	3	5	7	4	1	....	....	....	23	+1
6	....	....	....	....	2	3	1	....	....	....	1	7	+2
7	....	....	....	....	....	1	2	....	....	....	....	3	+3
Frequency	2	6	14	16	22	16	8	1	0	0	1	86	
Departure from mean	−4	−3	−2	−1	0	+1	+2	+3	+4	+5	+6	......	

TABLE XI.—*Correlation between cross section of stems in fall of 1910 and cut in spring of 1911 on 82 producing plants of row A1.*

[Fall stem area, subject; spring cut area, relative. Coefficient of correlation 0.811±0.025.]

Fall of 1910.	Spring of 1911 (square inches).																	Frequency.	Departure from mean.
	1	3	5	7	9	11	13	15	17	19	21	23	25	27	29	31	33		
⅛-inch square units:																			
10	13	4	....	1	...	...	...	...	...	...	...	...	...	....	....	....	....	18	− 2
30	2	6	6	2	...	2	...	...	...	...	...	...	...	....	....	....	....	18	− 1
50	1	3	5	3	1	2	1	...	...	...	...	...	...	....	....	....	....	16	0
70	....	....	1	6	3	1	1	2	...	...	...	...	...	....	....	....	....	14	+ 1
90	....	....	....	1	1	1	...	...	...	1	...	...	...	....	....	....	....	4	+ 2
110	....	....	1	....	1	2	...	...	...	...	...	...	...	....	....	....	....	4	+ 3
130	....	1	....	....	...	1	1	...	...	...	...	...	...	....	....	....	....	3	+ 4
150	....	....	....	....	...	...	...	...	...	2	...	...	...	....	....	....	....	2	+ 5
170	....	....	....	....	...	...	...	...	...	...	...	1	...	....	....	....	....	1	+ 6
190	....	....	....	....	...	...	...	...	...	...	...	...	...	....	....	....	....	0	+ 7
210	....	....	....	....	...	...	...	...	...	...	...	...	...	....	....	....	....	0	+ 8
230	....	....	....	....	...	...	...	...	...	...	...	...	...	....	....	....	....	0	+ 9
250	....	....	....	....	...	...	...	...	...	...	1	...	...	....	....	....	....	1	+10
270	....	....	....	....	...	...	...	...	...	...	...	...	...	....	....	....	1	1	+11
Frequency	16	14	13	13	6	9	3	2	0	3	1	1	0	0	0	0	1	82	
Departure from mean	−3	−2	−1	0	+1	+2	+3	+4	+5	+6	+7	+8	+9	+10	+11	+12	+13	....	

A most interesting interrelation comes in the size of the seedling to the weight of the seed. It was early noticed that the greenhouse lots of seedlings showed striking differences between progeny lots from different female plants. Wherever enough seed remained unplanted to give a fair average, the seed weight was determined and a comparison made with the average height of the seedlings in the progeny rows. Table XII was obtained with a coefficient of 0.780. Where future size depends on the start the young seedling gets in the bed, the tremendous importance of the use of large seed is at once apparent. To further test this correlation, 100 seeds from plant C13–5–1, open fertilized, were sown in 1910 under uniform conditions of moisture, heat, and light in a soil of uniform texture. Each seed was weighed to 0.001 gram, the germination record was made, and the height of each seedling was measured daily. No effect of size of seed on germination could be determined, but the size of the individual seed showed a very strong influence on the height and rate of growth. (Table XIII.) Where the individual seed is taken into account the correlation is lower than where the average of several is taken, on account of the varying hereditary tendencies in different seed of the same weight.

TABLE XII.—*Correlation between average weight of seed and average height of greenhouse seedlings of 1910 for 42 progeny lots on February 11, 1910.*

[Height, subject; weight, relative. Coefficient of correlation 0.780±0.042.]

Height of seedlings.	Average weight of seed (milligrams).											Frequency.	Departure from mean.
	19	20	21	22	23	24	25	26	27	28	29		
Inches:													
5.75	1	....	....	....	....	....	....	....	....	....	....	1	−1.00
6.00	1	2	....	1	1	....	....	....	....	....	....	5	− .75
6.25	....	1	1	....	....	....	....	....	....	....	....	2	− .50
6.50	....	2	1	1	....	....	....	....	....	....	....	4	− .25
6.75	....	3	1	1	1	1	2	....	....	....	....	9	0
7.00	....	....	....	3	4	2	1	....	....	....	....	10	+ .25
7.25	....	....	....	....	1	....	....	....	1	....	....	2	+ .50
7.50	....	....	....	....	2	1	2	....	....	....	....	5	+ .75
7.75	....	....	....	....	....	....	....	1	1	....	1	3	+1.00
8.00	....	....	....	....	....	....	....	....	1	....	....	1	+1.25
Frequency	2	8	3	6	9	4	5	1	3	0	1	42	
Departure from mean	−4	−3	−2	−1	0	+1	+2	+3	+4	+5	+6	......	

TABLE XIII.—*Correlation between height of shoot and weight of seed in 100 seedlings from weighed seed of C13–5–1, open fertilized, grown under control conditions in greenhouse at Washington in 1910.*

[Height of first shoot April 9, 1910, subject; weight of seed in milligrams, relative. Coefficient of correlation 0.41±0.056.]

Height of shoot.	Weight of seed in milligrams.															Frequency.	Departure from mean.
	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26		
Inches:																	
3			1													1	−2.50
3.25																0	−2.25
3.50					1	1		1								3	−2.00
3.75		1			1											2	−1.75
4		1			1	1		1								4	−1.50
4.25								2								2	−1.25
4.50								1	3	1						5	−1.00
4.75						3	2			1						6	− .75
5				2	1		2	1				1				7	− .50
5.25				2	2	1	1		3	1						10	− .25
5.50	2	1		1		1	1	1	1	1	2			1		12	0
5.75			1	1	4	3	1		1	4				1	1	17	+ .25
6					2	2	1	1	1		1					8	+ .50
6.25				1	1			1	1		1					5	+ .75
6.50							4		2		1	1				8	+1.00
6.75								1			3	1	1			5	+1.25
7												1				1	+1.50
7.25																0	+1.75
7.50											1	1			1	3	+2.00
7.75										1						1	+2.25
Frequency	2	3	2	7	13	12	12	10	12	9	8	5	1	2	2	100	
Departure from mean	−7	−6	−5	−4	−3	−2	−1	0	+1	+2	+3	+4	+5	+6	+7		

Table XIV is introduced in order to show that the size of the young asparagus seedling is important in determining the future size of the plant. This table shows the correlation existing between the average heights of 85 progeny lots while in the seed flats in the greenhouse at an age of three weeks and the average of the same progeny lots in their permanent place in the field. The factor of crowding, which aids the stronger plants and retards the weaker ones, was eliminated by placing the plants in individual pots February 13 and so maintaining them until they were transplanted to the field where they are just beginning to show the effects of crowding in the fall of 1911.

TABLE XIV.—*Correlation between average height of greenhouse seedlings in progeny lots February 11, 1910 (in ¼-inch units), and their average height September, 1910 (in inches).*

[Height in September, subject; height in February, relative. Coefficient of correlation 0.552±0.048.]

Height in September.	Average height of seedlings February 11, 1910 (¼-inch units).															Frequency.	Departure from mean.
	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34		
Inches:																	
9	...	...	1	1	...	...	...	...	...	...	...	...	...	...	...	2	−6
10	1	...	...	...	...	...	2	...	...	...	...	...	...	...	...	3	−5
11	...	...	...	1	1	1	...	3	...	...	...	...	...	...	...	6	−4
12	...	...	...	...	2	...	...	...	1	1	...	...	...	...	...	4	−3
13	...	...	...	...	1	...	3	3	4	1	2	...	...	...	...	14	−2
14	...	...	...	...	3	...	...	1	3	...	...	1	...	...	...	8	−1
15	...	...	...	...	1	...	2	1	3	1	2	1	...	...	1	12	0
16	...	...	...	...	...	1	1	2	3	1	1	1	1	2	...	13	+1
17	...	...	...	...	...	...	1	3	...	...	1	1	1	...	...	7	+2
18	...	...	...	...	...	...	...	1	1	...	...	...	1	...	...	3	+3
19	...	...	...	...	...	...	1	...	1	1	1	2	...	...	...	6	+4
20	...	...	...	...	...	...	...	...	...	1	1	...	...	...	...	2	+5
21	...	...	...	...	...	...	...	1	...	...	...	...	2	...	...	3	+6
22	...	...	...	...	...	...	...	...	1	...	...	...	...	1	...	2	+7
Frequency	1	0	1	2	8	2	10	15	17	6	8	6	5	3	1	85	
Departure from mean	−8	−7	−6	−5	−4	−3	−2	−1	0	+1	+2	+3	+4	+5	+6	....	

In continuing the work with the greenhouse seedlings after they were removed to the field in May, 1910, further correlations have been studied. While these progeny lots are not exactly a random sample, in many respects they answer the purpose of one, the population being distributed in monomodal and often nearly normal variation curves.

In making selections among the young seedlings a general tendency among growers is to assume that height is a good index of large-sized shoots in future years in the cutting bed. According to the tables constructed on the data of height in 1910 and stalk diameter of the greenhouse seedlings in 1911 (Table XV), there is a correlation of 0.634 ±0.013. This result is supported by notes from row A1, where the total area of the cross section of the fall stems in 1910 was compared with the total area of the cross section of the shoots cut in the spring of 1911, as shown in Table XI (p. 36).

TABLE XV.—*Correlation between height in 1910 (inches) and diameter of largest stalk in 1911 ($\frac{1}{16}$-inch units) of the greenhouse seedlings of 1910.*

[Height in 1910, subject; diameter in 1911, relative. Coefficient of correlation 0.634±0.013.]

Height in 1910.	Diameter of stalks in 1911 ($\frac{1}{16}$-inch units).														Frequency.	Departure from mean.
	1	2	3	4	5	6	7	8	9	10	11	12	13	14		
Inches:																
4	....	1	....	1	....	....	....	....	....	....	....	....	....	....	2	−11
5	3	....	1	1	....	....	....	....	....	....	....	....	....	....	5	−10
6	1	2	2	4	3	....	....	....	....	....	....	....	....	....	12	− 9
7	....	4	7	5	8	2	2	1	....	....	....	....	....	....	29	− 8
8	....	3	4	9	4	9	1	1	....	....	....	....	....	....	31	− 7
9	....	4	10	18	12	11	1	3	....	....	....	....	....	....	59	− 6
10	....	....	9	15	18	5	3	....	....	....	....	....	....	....	50	− 5
11	....	3	9	16	20	14	11	5	....	1	....	....	....	....	79	− 4
12	....	....	7	10	24	18	6	2	....	....	....	....	....	....	67	− 3
13	....	1	6	10	20	28	6	3	....	1	....	....	....	....	75	− 2
14	....	1	3	12	13	30	14	9	2	1	....	....	....	....	85	− 1
15	....	....	3	6	15	28	17	10	....	....	....	....	....	....	79	0
16	....	....	2	3	11	29	11	10	4	2	1	....	....	....	73	+ 1
17	....	....	1	....	3	25	9	16	1	1	....	....	....	....	56	+ 2
18	....	....	....	4	11	19	12	11	3	....	....	....	....	....	60	+ 3
19	....	....	....	....	5	14	12	10	1	4	....	....	....	....	46	+ 4
20	....	....	....	2	1	4	11	8	5	3	....	....	....	....	34	+ 5
21	....	....	....	1	1	8	7	14	3	1	1	....	....	....	36	+ 6
22	....	....	....	....	1	5	5	6	1	1	....	....	....	....	19	+ 7
23	....	....	....	....	1	5	1	7	3	1	2	3	....	....	23	+ 8
24	....	....	....	....	....	....	2	2	2	1	3	....	....	1	11	+ 9
25	....	....	....	....	....	2	1	3	....	1	....	....	....	....	7	+10
26	....	....	....	....	....	....	....	....	2	....	1	....	....	....	3	+11
27	....	....	....	....	....	2	....	....	....	....	....	....	....	....	2	+12
28	....	....	....	....	....	....	....	....	....	1	....	1	....	....	2	+13
29	....	....	....	....	....	....	....	1	....	....	....	1	....	....	2	+14
30	....	....	....	....	....	....	....	....	....	....	....	....	....	....	0	+15
31	....	....	....	....	....	....	....	....	....	....	1	....	....	....	1	+16
32	....	....	....	....	....	....	....	....	....	....	....	1	....	....	1	+17
Frequency	4	19	64	117	171	258	132	122	27	19	9	6	0	1	949	
Departure from mean	−5	−4	−3	−2	−1	0	+1	+2	+3	+4	+5	+6	+7	+8	......	

Table XVI shows a comparison of height of the greenhouse seedlings in September, 1910, and in 1911. This table serves as another illustration of the opportunity of making selections on seedlings. Seventy-five per cent of the 1910 lot could have been discarded without losing any of the 10 best plants of 1911. A comparison of the correlation here shown, with the table presented by Clark [1] in his studies of timothy in 1910, shows that the conditions at Concord were more uniform than those at Ithaca.

One striking feature in connection with these studies is that the reliability of the height correlation is almost equaled by the reliability of height with next year's size (Table XV).

Data from mature plants in row A1 show a high correlation (Table XVII) between the amount of tops on the plants in the fall year by year and also a high correlation (Table XVIII) between the cut of individual plants compared in successive years.

[1] Clark, C. F. Bulletin 279, Cornell Agricultural Experiment Station. 1910.

TABLE XVI.—*Correlation between heights of the greenhouse seedlings of 1910 in 1910 and in 1911 (inches).*

[Height in 1910, subject; height in 1911, relative. Coefficient of correlation 0.642±0.0126.]

Height in 1910.	Height of plants in 1911 (inches).																												Frequency.	Departure from mean.
	3	6	9	12	15	18	21	24	27	30	33	36	39	42	45	48	51	54	57	60	63	66	69	72	75	78	81	84		
Inches:																														
4								1	1																				2	−11
5	1			2							2																		5	−10
6				1		2			1	2	5			1															12	− 9
7				2	1	1		1	1	6	6	4	3	1	2		1												29	− 8
8						1	2	1	3	6	2	6	1	6	1	2													31	− 7
9							1	5	8	6	8	11	4	7	5	1	1	2											59	− 6
10						2	3	1	4	6	7	3	11	5	5	1	2												50	− 5
11						1	2	2	6	10	5	12	9	17	3	3	3	3	2	1									79	− 4
12							1		4	10	8	7	11	9	5	6	2	4											67	− 3
13								2	2		7	14	5	17	10	8	3	7											75	− 2
14								1	3	4	6	5	12	15	9	6	9	11	2	2									85	− 1
15									1	1	2	7	14	9	14	14	4	8	5										79	0
16								1	1	2	3	3	8	9	10	8	13	10	2	1	2								73	+ 1
17										1	2	2	3	9	7	10	8	9	5										56	+ 2
18										1	3	3	2	10	9	12	8	6	3	3									60	+ 3
19											1	4	5	1	2	5	15	5	6	1	1								46	+ 4
20											1	2		3	2	4	7	5	4	3	3		1						34	+ 5
21											1	2		3	5	3	5	5	5	6		1							36	+ 6
22													1	2		2	5	4	1	1	2	1							19	+ 7
23													2	1	2	4	4	2	2	3	1	2							23	+ 8
24															1	2	1		2	3	1	1							11	+ 9
25																2	1	1	1	2									7	+10
26																			2	1									3	+11
27														1				1											2	+12
28																						1						1	2	+13
29																				1	1								2	+14
30																													0	+15
31																					1								1	+16
32																										1			1	+17
Frequency	1	0	0	5	1	7	9	15	35	55	69	84	91	126	92	93	92	83	42	28	12	6	1	0	0	1	0	1	949	
Departure from mean	−39	−36	−33	−30	−27	−24	−21	−18	−15	−12	−9	−6	−3	0	+3	+6	+9	+12	+15	+18	+21	+24	+27	+30	+33	+36	+39	+42		

TABLE XVII.—*Correlation between area of cross section of fall stems of row A1 in 1910 and in 1911.*

[Stems in 1910, subject; stems in 1911, relative. Coefficient of correlation, 0.859±0.018.]

Stems in fall of 1910.	Area of cross section of stems in fall of 1911 (⅛-inch square units).																	Frequency.	Departure from mean.
	10	30	50	70	90	110	130	150	170	190	210	230	250	270	290	310	330		
⅛-inch square units:																			
10	8	9	1															18	− 2
30		5	6	4	2		1											18	− 1
50	2	1	3	4	2	2	1	1										16	0
70				1	3	1	4	4		1								14	+ 1
90							2	1			1							4	+ 2
110				1		1	1	1										4	+ 3
130						1			1	1								3	+ 4
150											1	1						2	+ 5
170																	1	1	+ 6
190																		0	+ 7
210																		0	+ 8
230																		0	+ 9
250																	1	1	+10
270																1		1	+11
Frequency	10	15	10	10	7	5	9	7	1	2	2	1	0	0	0	1	2	82	
Departure from mean	−4	−3	−2	−1	0	+1	+2	+3	+4	+5	+6	+7	+8	+9	+10	+11	+12	......	

TABLE XVIII.—*Correlation between the yield in 1910 and in 1911 from the plants in row A1, given in the sum of the squared diameters of the shoots from each hill.*

[Cut of 1911, subject; cut of 1910, relative. Coefficient of correlation, 0.797±0.027.]

Cut of 1911.	Cut of 1910 (square inches).											Frequency.	Departure from mean.
	0 to 1	1 to 2	2 to 3	3 to 4	4 to 5	5 to 6	6 to 7	7 to 8	8 to 9	9 to 10	10 to 11		
Square inches:													
0 to 2	16	....	....	....	....	....	....	....	....	....	....	16	−6
2 to 4	7	6	1	....	....	....	....	....	....	....	....	14	−4
4 to 6	2	6	3	1	1	....	....	....	....	....	....	13	−2
6 to 8	3	2	5	2	....	....	1	....	....	....	....	13	0
8 to 10	....	1	....	2	....	3	....	....	....	....	....	6	+2
10 to 12	....	....	3	1	4	1	....	....	....	....	....	9	+4
12 to 14	....	....	....	....	1	2	....	....	....	....	....	3	+6
14 to 16	....	....	....	1	1	....	....	....	....	....	....	2	+8
16 to 18	....	....	....	....	....	....	....	....	....	....	....	0	+10
18 to 20	....	....	....	....	....	2	....	....	1	....	....	3	+12
20 to 22	....	....	....	....	....	....	....	....	....	....	1	1	+14
22 to 24	....	....	....	....	....	....	....	....	1	....	....	1	+16
Frequency	28	15	12	7	7	8	1	0	2	0	1	81	
Departure from mean	−2	−1	0	+1	+2	+3	+4	+5	+6	+7	+8	......	

Table XIX shows how close a selection can be made for stalk diameter in 2-year-old plants by saving only the tall plants. This method, of course, is much quicker than to measure the diameter of the shoots of every plant in the bed.

TABLE XIX.—*Correlation between height (inches) and diameter of largest stalks ($\frac{1}{16}$-inch units) in 1911 of greenhouse seedlings of 1910.*

[Heights, subject; diameter, relative. Coefficient of correlation, 0.792±0.008.]

Height in 1911.	Diameter in 1911 ($\frac{1}{16}$-inch units).														Frequency.	Departure from mean.
	1	2	3	4	5	6	7	8	9	10	11	12	13	14		
Inches:																
3	1	....	....	....	....	....	....	....	....	....	....	....	....	....	1	−39
6	....	....	....	....	....	....	....	....	....	....	....	....	....	....	0	−36
9	....	....	....	....	....	....	....	....	....	....	....	....	....	....	0	−33
12	3	2	....	....	....	....	....	....	....	....	....	....	....	....	5	−30
15	....	1	....	....	....	....	....	....	....	....	....	....	....	....	1	−27
18	....	4	3	....	....	....	....	....	....	....	....	....	....	....	7	−24
21	....	3	4	2	....	....	....	....	....	....	....	....	....	....	9	−21
24	....	5	6	4	....	....	....	....	....	....	....	....	....	....	15	−18
27	....	2	19	11	3	....	....	....	....	....	....	....	....	....	35	−15
30	....	2	13	27	8	5	....	....	....	....	....	....	....	....	55	−12
33	....	....	10	21	29	8	1	....	....	....	....	....	....	....	69	−9
36	....	....	5	25	29	19	5	1	....	....	....	....	....	....	84	−6
39	....	....	4	15	29	35	6	2	....	....	....	....	....	....	91	−3
42	....	....	....	8	38	47	20	12	1	....	....	....	....	....	126	0
45	....	....	....	2	19	42	14	12	1	1	1	....	....	....	92	+3
48	....	....	....	1	11	41	19	17	2	....	1	1	....	....	93	+6
51	....	....	....	1	4	25	28	22	8	2	2	....	....	....	92	+9
54	....	....	....	....	1	29	23	24	2	4	....	....	....	....	83	+12
57	....	....	....	....	....	7	9	14	6	5	1	....	....	....	42	+15
60	....	....	....	....	....	....	4	14	4	3	2	1	....	....	28	+18
63	....	....	....	....	....	....	3	2	1	2	2	1	....	1	12	+21
66	....	....	....	....	....	....	....	2	1	2	....	1	....	....	6	+24
69	....	....	....	....	....	....	....	....	1	....	....	....	....	....	1	+27
72	....	....	....	....	....	....	....	....	....	....	....	....	....	....	0	+30
75	....	....	....	....	....	....	....	....	....	....	....	....	....	....	0	+33
78	....	....	....	....	....	....	....	....	....	....	....	1	....	....	1	+36
81	....	....	....	....	....	....	....	....	....	....	....	....	....	....	0	+39
84	....	....	....	....	....	....	....	....	....	....	....	1	....	....	1	+42
Frequency	4	19	64	117	171	258	132	122	27	19	9	6	0	1	949	
Departure from mean	−5	−4	−3	−2	−1	0	+1	+2	+3	+4	+5	+6	+7	+8	......	

All these tables go to show that in asparagus we are dealing with a stable plant with a permanent individuality; that an individual characteristic observed one year will persist in nearly unmodified form in future years. Without a study of this kind any breeding work would remain an uncertain proposition for several generations.

VIGOR OF SEEDLINGS OF MALE A7–83.

The several lots of hand-pollinated seed with the check lots of open-fertilized seed which were sown in the greenhouses at Washington in

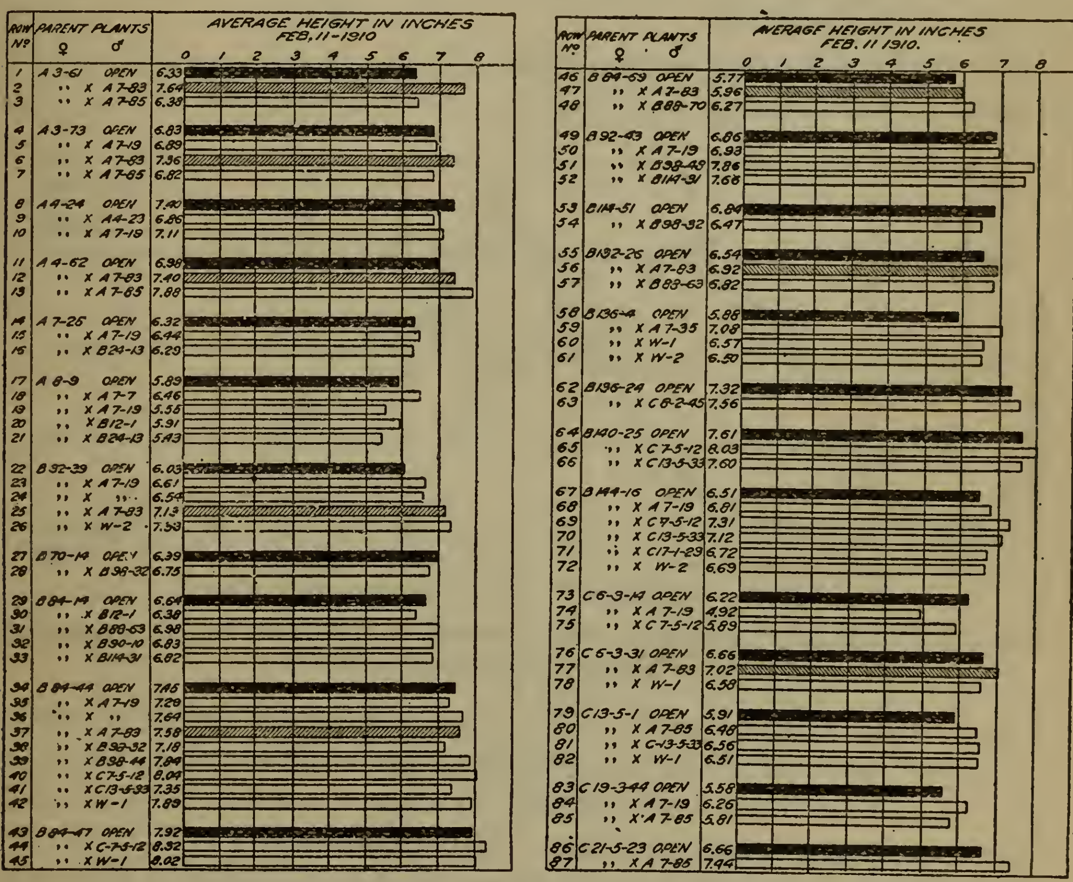


FIG. 2.—Diagram showing the average height of 87 progeny rows of seedlings of 1910 in greenhouse. The measurements were made February 11, 1910, for comparison of open-fertilized and hand-pollinated lots of seed.

January, 1910, were planted to study the effect of the different parents on vigor of the young seedlings. These lots of seedlings varied markedly in average size, and it was easily seen that the open-fertilized lots of seed as a rule were shorter than the hand-pollinated lot from the same female plant. The accompanying diagram (fig. 2) shows the average heights on February 11 of the entire series of seedlings arranged in classes showing the different lots of hand-pollinated seed with the sample check from the same female. The check is shown in black with the different pedigreed lots following in outline. A study of the diagram shows that wherever male A7–83 was used an added vigor is shown in height over the check, which represents an average of the selected males. Of course, this check lot is influenced by the proximity of good or bad male plants, but usually several males would be about equidistant from any select female. Several other males show an added vigor, but the lack of rust resistance shown in these lots when exposed to rust in the fall of 1910 removes them from consideration. This difference of vigor is still maintained in the seedlings of 1910 after growing two years in the field. Of course, rust has entered into the effect now, but it certainly has not been the whole cause of the marked increase in the progeny of male A7–83.

The size of seed being so important a factor in seedling size, it was thought best to continue this study in 1911 on lots of seed of the same

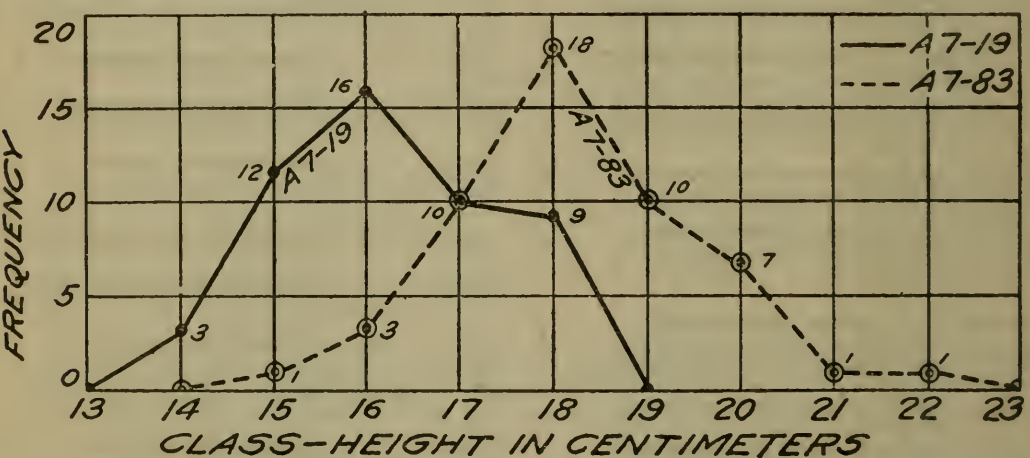


FIG. 3.—Diagram showing the height of 50 seedlings each from A7-25 pollinated with A7-19 (male) and A7-83 (male), seed weighing between 0.021 and 0.024 gram.

average size. Female plant A7–25 was pollinated with both male A7–83 and male A7–19 in the summer of 1910 and carefully weighed seeds of the two lots were planted to show the effect both of seed size and of vigor from the male. The effect of these two factors on the seedlings when 10 days old is shown in Table XX. To show the effect of A7–83 on seedling vigor, 50 seedlings from each lot representing the different males were selected from seed weighing as near as possible the same; in each case the weights ranged between 21 and 24 milligrams. In the diagram shown as figure 3 a comparison is made between the heights 10 days after germination of lots of seedlings from A7–25 pollinated with these two males. The result shows that the added vigor of the lot from A7–83 is due to something besides large seed.

TABLE XX.—*Comparison of weight of seed and height of seedlings at 10 days of age from two lots of 1910 seed from A7–25 female, the two lots having different male parents, A7–19 and A7–83.*

Weight of seed.		Height of seedlings (centimeters).												Weight frequency.	
		12	13	14	15	16	17	18	19	20	21	22	23	A7–19	A7–83
Milligrams:															
13	A7–19	1												1	
	A7–83														
14	A7–19			1										1	
	A7–83														
15	A7–19													0	
	A7–83														
16	A7–19		1											1	
	A7–83														
17	A7–19		1		2									3	
	A7–83														
18	A7–19				1									1	
	A7–83				1										1
19	A7–19			1	1	4	1							7	
	A7–83		1												1
20	A7–19			1	3	6	1							11	
	A7–83														0
21	A7–19				5	5	3	1						14	
	A7–83							5	1						6
22	A7–19			1	6	4	3	3						17	
	A7–83					1	4		1	2					8
23	A7–19					5	4	4						13	
	A7–83				1	1	3	7	4	2					18
24	A7–19			2	1	2		1						6	
	A7–83					1	4	10	5	3	1	1			24
25	A7–19						1	5	4					10	
	A7–83		1			2	2	5	4	4	1				19
26	A7–19						1		2	2				5	
	A7–83							2	2	2	2	2	1		11
27	A7–19				1			3						4	
	A7–83									1	1				2
28	A7–19								1		1			2	
	A7–83									1	1		2		4
29	A7–19														
	A7–83											1	1		2
30	A7–19														
	A7–83											1			1
Height frequency:															
A7–19		1	2	6	20	26	14	17	7	2	1			96	
A7–83			2	0	2	5	13	29	17	15	6	5	4		97

RUST RESISTANCE.

To any one familiar with rusty asparagus fields the injury caused by rust is apparent, yet the actual damage is hard to estimate on account of the different seasons affecting the cut of the crop.

Table XXI shows the relation between the percentage of stem cross section of the crop of 1911 to that of 1909 and the rust resistance of 1910. Several plants growing in 1909 rusted so badly that they died before 1911, and are therefore excluded from the table. It is realized that the increase in size of asparagus hills is influenced by many things other than rust, so that the actual effect of rust is much higher than the coefficient given shows.

TABLE XXI.—*Correlation between the degree of rust resistance in 1910 and the percentage of the stem cross section of 1911 to that of 1909 in row A1.*

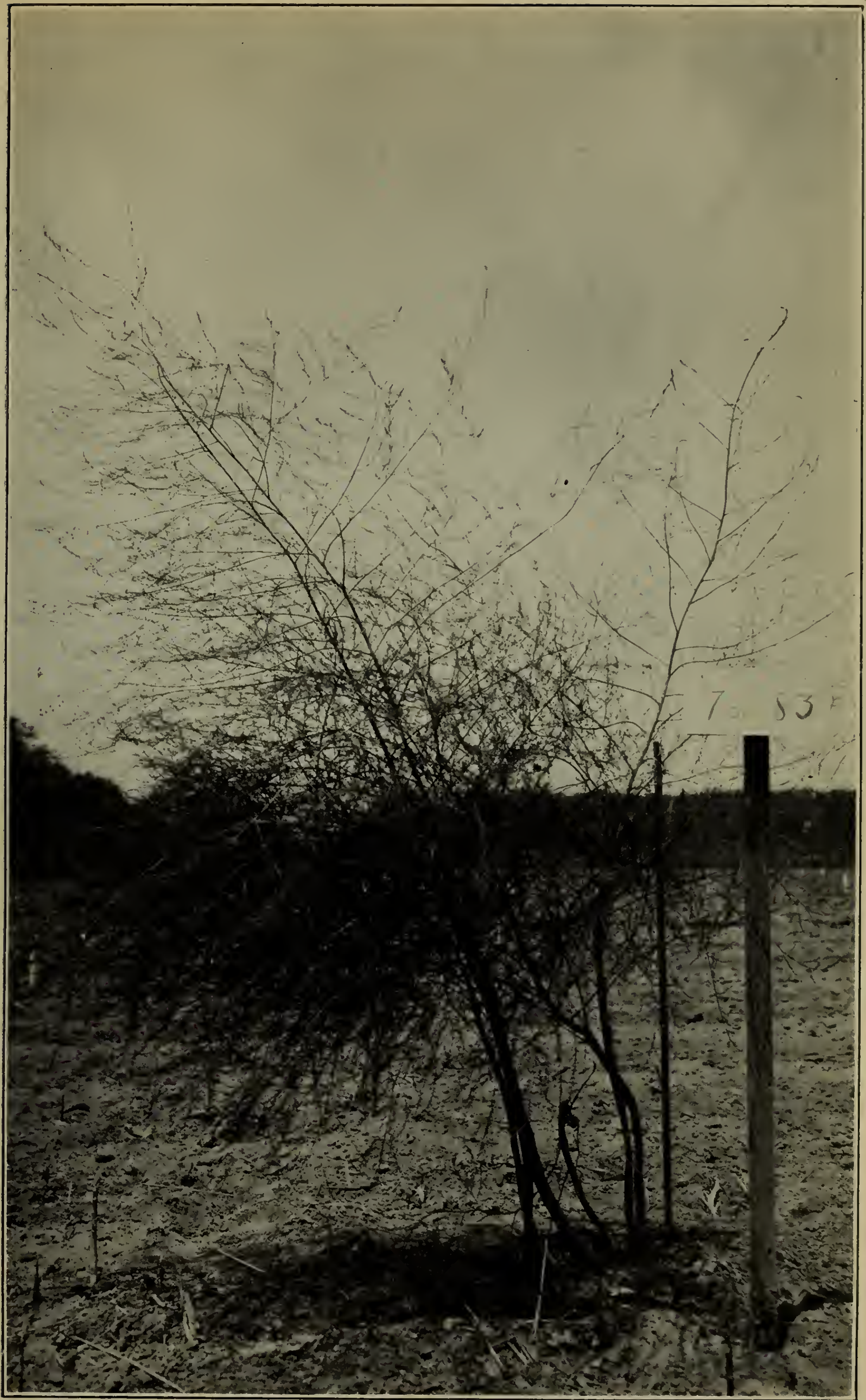
[Rust resistance, subject; percentage of stem cross section, relative. Coefficient of correlation 0.393±0.065.]

Rust resistance in 1910.	Percentage of stem cross section of 1911 to that of 1909.														Frequency.	Departure from mean.
	20	60	100	140	180	220	260	300	340	380	420	460	500	540		
Grade:																
0		6	2	2	1										11	−4
1	1	2	2	3			1								9	−3
2		2		3	2	1									8	−2
3	1	1		2	3	1	1			1					10	−1
4			1	3	1		1								6	0
5		3	1		1	3				1				1	10	+1
6	1	1	3		2	1	2								10	+2
7			1	2	1						1			1	6	+3
8				1	3		3	1							8	+4
Frequency	3	15	10	16	14	6	8	1	0	2	1	0	0	2	78	
Departure from mean	−160	−120	−80	−40	0	+40	+80	+120	+160	+200	+240	+280	+320	+360		

In 1910 a study was made of the rust resistance of the seedlings started in the greenhouse at Washington and later transferred to a permanent place in the field at Concord. The rust attacked the field in August, so that in the latter part of September some of the nonresistant plants were dead. The seedlings of the several lots were then judged as individuals, and Table XXII shows how they ranked in rust resistance. Lots 1, 4, 8, 11, etc., are from open-fertilized seed from the female plants used, while lots 2, 3, 5, 6, etc., are from seed hand pollinated from male plants mentioned in the second column of the table. A study of the table shows A7–83 to be possessed of great power to transmit rust resistance. The results shown in this table are those on which our breeding work is now based. A7–35 is the only other male showing desirable resistance. This male is being tested further and may be selected as a breeding plant. But there is now no question as to the desirability of A7–83 (Pl. X, figs. 1, 2, and 3, and Pl. XI).

TABLE XXII.—*Rust resistance of individual greenhouse seedlings of 1910 in progeny lots in the field, September, 1910.*

Parent plants. ♀	Parent plants. ×♂	No.	Individual rust resistance of seedlings (grades). 0	1		2		3		4		5		6		7		8		9		10	Number of plants.	Mean.
A3-61		1						1		1				1	1	2		3					9	6.25
	A7-83	2														2		5	1	1	5		14	8.50
	B72-85	3				2		1				4		2		4	2						15	5.47
A3-73		4						2				2	1	2		3							10	5.45
	A7-19	5								2		4		1		3							10	5.50
	A7-83	6								1				1		1	1	2	2	1			9	7.39
	A7-85	7								2	1	4		1	1			1					10	5.30
A4-24		8								1	2	4		2		1							10	5.00
	A4-23	9										1	3	3		1		1		1			10	6.35
	A7-19	10										2	2	1	2	4		4					15	6.67
A4-62		11						1		3	1	2	1		1	1		1					11	5.74
	A7-83	12											1			1		1	3	2	1		9	8.19
	A7-85	13									1	1	1	3		2	1	1					10	6.25



PLANT "WASHINGTON," A7-83, SHOWING THE GENERAL TYPE OF THE BEST BREEDING MALE USED IN THE RUST-RESISTANT BREEDING WORK.

(Photograph taken June, 1910.)

PLANT "MARTHA," B32-39, SHOWING THE GENERAL TYPE OF THE BEST BREEDING FEMALE USED IN THE RUST-RESISTANT BREEDING WORK.

(Photograph taken September, 1909.)

TABLE XXII.—*Rust resistance of individual greenhouse seedlings of 1910 in progeny lots in the field, September, 1910*—Continued.

Parent plants. ♀	Parent plants. ×♂	No.	Individual rust resistance of seedlings (grades). 0	1		2		3		4		5		6		7		8		9		10	Number of plants.	Mean.
A7-25		14								1	1	1	2	2	1	2							10	5.70
	A7-19	15										2	3	3		4	1	2					15	6.07
	B24-13	16				5		4				1											10	2.70
A8-9		17						1		1	1	1		3		1							8	5.19
	A7- 7	18						1			2			3		1							7	5.29
	A7-19	19								1	2	6	1	4		4							18	5.58
	B12- 1	20						1		1		3		1	1	3							10	5.55
	B24-13	21		3		4		3															10	2.00
B32-39		22								1	1	2	1	1		2			2				10	6.10
	A7-19	23									1		1	3	2	6	4	1	1				19	6.82
	A7-19	24								1	1	1	1	4	1	3	2	1					15	6.23
	A7-83	25																		1	6	1	8	9.50
	W- 2	26									1	1			4	1	2						9	6.39
B70-14		27									1	5	3					1					10	5.40
	B98-32	28								1	1	2	2	1	2								9	5.39
B84-14		29						2	1	1		2	2			1	1						10	4.90
	B12- 1	30									2	2	1	1	1	1	1	1					10	5.95
	B88-63	31						1				3	1	3		3							11	5.68
	B90-10	32				1		2		3		2		2									10	4.20
	B114-31	33					1	5	2		1					1							10	3.60
B84-44		34				1				3		1	3	1		1							10	4.85
	A7-19	35								1	1	1	1	3			2						9	5.78
	A7-19	36													1	3	1	4	1				10	7.55
	A7-83	37															1	6	2				9	8.05
	B98-32	38				1		1	1	2	2	2		1									10	4.15
	B98-44	39						1	1			3	2	1		2							10	5.25
	C7-5-12	40							5			2		2		1							10	4.65
	C13-5-33	41								2		2	1	2	1		1						9	5.50
	W- 1	42								1				5	1	2							9	6.06
B84-47		43				1		2	1	3			1	2									10	4.10
	C7-5-12	44					1	2	4					2	1								10	4.10
	W- 1	45						1			3	3		5	1		1						14	5.39
B84-69		46				1		3	1	1			1		1								8	3.81
	A7-83	47						1		2		3		3		2	1	2	1				15	6.00
	B88-70	48						2	2	2	1	3											10	4.05
B92-43		49				2		3	2			1		2									10	3.70
	A7-19	50						1	2	3	1	2		2		1	1	1					14	5.07
	B98-48	51		1		1	1	2		2	1			1		1							10	3.70
	B114-31	52								1	1	3	2	1		2							10	5.45
B114-51		53								1		1		1	2	3	1	2					11	6.77
	B98-32	54									4		1	1		2							8	5.44
B132-26		55								1		2		3		4							10	6.00
	A7-83	56														2	1	2	4	1			10	8.05
	B88-63	57						1		1		1		2	1	4							10	5.85
B136-4		58				1		2						3		2	1						9	5.28
	A7-35	59														1	1	3	2	3			10	8.25
	W- 1	60								1		2	1	1		4	1						11	5.96
	W- 2	61		1				5		2	1	1											10	3.35
B136-24		62						1		2		2		2		2	1						10	5.45
	C8-2-45	63				1		2		2		4		1									10	4.20
B140-25		64						2				5		2	1								10	4.95
	C7-5-12	65						1	1	1		1		6									10	5.15
	C13-5-33	66					1		2	1		4	1										9	4.33
B144-16		67						2	1	2		2		2		1							10	4.35
	A7-19	68				1		1	2	4		1		1									10	3.90
	C7-5-12	69		2		3		1	1	2		1											10	2.75
	C13-5-33	70				1	2	2		2	1	2		1									11	3.77
	C17-1-23	71				1	1	4		2		2											10	3.45
	W- 2	72	1	3	1	1	1	1		2													10	2.00
C6-3-14		73				2	1	7															10	2.75
	A7-19	74					1	1	1	1		4		1									9	4.33
	C7-5-12	75		1		1		3	1	1		2		1									10	3.55
C6-3-31		76						1			2	4	1			1							9	4.94
	A7-83	77										1				2	4	2	1				10	7.35
	W- 1	78						1		1		1		3		1	1	2					10	6.05
C13-5-1		79				1					1	3		2		1	1						9	5.33
	A7-85	80						2	2	1	1	3	2	4		3		2					20	5.43
	C13-5-33	81					1	2	4	3	1	3											14	3.86
	W- 1	82					1	2	3	1	3	6		1		1	1						19	4.58
C19-3-44		83		1			1	3	1	3		1											10	3.30
	A7-19	84								3		4	2	6									15	5.27
	A7-85	85		1			1	3	3	2		6		2		1							19	4.21
C21-5-33		86	1			1	1	1		1	1	1		3									10	3.90
	A7-85	87								1		6				3							10	5.50
	C21-5-47	89					1	6		3													10	3.25
	Total	....	2	13	1	33	16	101	44	93	47	160	46	130	27	111	36	51	20	10	12	1	951	.....

Table XXIII shows the height of these greenhouse seedlings at the end of the season's growth in 1910. Again A7–83 is found standing out above the other males in the transmission of vigor. As mentioned on page 44, in respect to the vigor of seedlings of some of the males while in the greenhouse, some good lots are found, but they are poor in rust resistance, and the male parents have been discarded.

Table XXIII.—*Height of individual greenhouse seedlings of 1910, in progeny lots in the field, September, 1910.*

Parent plants.		Row No.	Height in inches.																														Number of plants.	Mean height.
♀	×♂		3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32		
A3-61		1								2	3	1		3	1																		10	12.20
	A7-83	2									2			2	1	2	2	1	2				1		1								14	16.79
	A7-85	3					1				1	1	1		1	3		3	2	1		1											15	16.00
A3-73		4					1				1		5	1	1					1													10	13.20
	A7-19	5						2	2	1	2						1				2												10	12.50
	A7-83	6					1		1			1		1	1	2		1									1						9	14.89
	A7-85	7			1	1		1			1	2		2				1			1												10	12.10
A4-73		8					1		1		1	2	1	3	1																		10	12.10
	A4-23	9									1		1	3	1	1	1	1	1														10	15.10
	A7-19	10			1							1		1	3	1	2	3	2			1											15	16.00
A4-62		11							1	1		1	1		1		2	1	1		2												11	15.64
	A7-83	12									1			1	1	1			1	1		1		1	1								9	18.44
	A7-85	13												1	1	1		4			2	1											10	18.10
A7-25		14									2	1	1	1	2	1		1	1														10	14.40
	A7-19	15					1		1	3	1	2	3	3					1														15	12.07
	B24-13	16				1	1	2	3	1		1		1																			10	9.20
A8- 9		17							2		2		1	1	1	1																	8	12.25
	A7- 7	18						2	1	1		2			1																		7	10.57
	A7-19	19			1		3	2	7	2	2		1																				18	8.89
	B12- 1	20			1		1		2	1	1	3	1																				10	10.00
	B24-13	21		1	1	2		2	1		2		1																				10	8.10
B32-39		22									1		1	1	2	2	3																10	15.10
	A7-19	23							1	1		3	1	3	2		3	2		2		1											19	15.21
	A7-19	24							1	2	2	2	3	2	1		1	1															15	12.80
	A7-83	25				1		2				1	1	1	1		1																8	11.63
	W- 2	26					1				1		1	2	1	1	1				1												9	14.22
B70-14		27	1					1			2	1	2	1	1					1													10	12.00
	B98-32	28				1	1		1		1	2			2			1															9	9.17
B84-14		29					1	1								2		1	2	1		2											10	16.70
	B12- 1	30										1	1	2	2		1	2			1												10	14.90
	B88-63	31							1				1	2		1				2	2	1							1				11	18.09
	B90-10	32									2	2	1		1	1		1	1				1										10	15.00
	B114-31	33							1	2	1	1		1		2							1				1						10	14.80
B84-44		34								1			1				2	2		1	1		1		1								10	18.20
	A7-19	35										1			1				2	1	1		2		1								9	19.67
	A7-19	36												1				1	1	1	3	1	1	1									10	20.30
	A7-83	37													1		1		1	1	1	1	2					1					9	20.44
	B98-32	38					1		1				2		2	2			1					1									10	14.70
	B98-44	39								1				1		3			2	1	1		1										10	17.40
	C7-5-12	40									2			2	1	2		1		1	1												10	15.60
	C13-5-33	41								1	2		1			1		2		1	1												9	15.33
	W- 1	42										1	2	1			1	1	1			1	1										9	16.78
B84-47		43					1			1				3	1		1	1	1										1				10	15.70
	C7-5-12	44						1		1	1				1	3	2	1															10	14.40
	W- 1	45											1	3	2	1	2	3	1	1													14	16.29
B84-69		46				1				3	2						1	1															8	11.63
	A7-83	47						2	3		3	1		3	2						1												15	12.07
	B88-70	48					1	3	2	2	1			1																			10	9.40
B92-43		49							2		2	1	2	1		1		1															10	12.60
	A7-19	50					1	1	2	3	2	2			2	1																	14	11.07
	B98-48	51									1		1	2	2	2		2															10	15.00
	B114-31	52				1			1			2		2	1		1	2															10	13.50
B114-51		53							1	2	1	3	1	1		1	1																11	12.36
	B98-32	54								1	1	3		1	1	1																	8	11.25
B132-26		55											1		4	1	1			1	1	1											10	16.90
	A7-83	56													1	1			2				3	1	1	1							10	21.30
	B88-63	57										1	1		2			2			2	1	1										10	17.80

TABLE XXIII.—*Height of individual greenhouse seedlings of 1910, in progeny lots in the field, September, 1910*—Continued.

Parent plauts.		Row No.	Height in inches.																														Number of plants.	Mean height.
♀	×♂		3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32		
B136- 4	....	58			1			1		1					1	2	1	1	1														9	13.78
	A7-35	59														2	1		1	3	3												10	19.10
	W- 1	60		1			1		1	1				3	1	2	1																11	12.36
	W- 2	61				1					3	1	2	2		1																	10	12.10
B136-24	....	62							1	1			1		1	5	1																10	14.40
	C8-2-45	63									1	1	1		4			1	1	1													10	15.30
B140-25	....	64											1			1	2	2	1				1	2									10	18.90
	C7-5-12	65										1				1	1			2			1	1		1		1				1	10	21.80
	C13-5-33	66											2	2	3	2																	9	14.56
B144-16	....	67									2	1	1		1	1			1		2			1									10	16.30
	A7-19	68					1					2	2	1	1		2				1												10	14.10
	C7-5-12	69						2	1		1	1	1	2			1		1														10	12.50
	C13-5-33	70					2			2	2	1	2	1	1																		11	11.18
	C17-1-23	71				1	1	1	3				1	2	1																		10	10.20
	W- 2	72					3	1	1		3	1					1																10	10.00
C6-3-14	....	73					1	1	2	2	2		1	1																			10	10.20
	A7-19	74					1	1	4	1	1	1																					9	9.33
	C7-5-12	75						1				2		1	1		4	1															10	14.70
C6-3-31	....	76								1		1		2	1	1			1	2													9	15.56
	A7-83	77												1	1	2		1	1			2	2										10	18.80
	W- 1	78									1		2			1	1	1		1	1		2										10	17.50
C13-5- 1	....	79								3		1	1		1	2	1																9	13.22
	A7-85	80									1			2		1	1	2	5	4		1	1	2									20	18.80
	C13-5-33	81						1	1		2		1	2		2	1	3	1														14	14.43
	W- 1	82									1		1		2		2	1	2		3	1	1	1	2	1					1		19	20.16
C19-3-44	....	83		1	1		1	1		1	1	2		1				1															10	10.10
	A7-19	84		2	1			1			2		3		3		1			1			1										15	12.47
	A7-85	85			1				3	1	1	2	5			1	2	1	1	1													19	13.11
C21-5-33	....	86					1		1			1	3			1		2		1													10	13.90
	A7-85	87							1				1		1	1	2		2	1		1											10	16.70
	C21-5-47	89				2				3		1	1		1	1						1											10	12.00
Total.......		..	1	5	9	12	29	33	58	50	75	66	75	84	79	72	57	61	45	35	34	19	24	11	7	3	2	2	2	..	1	1	952	14.42

Among the females, B32–39 (Pl. XII) stands out in the 1910 test as a good parent for rust resistance. Unfortunately the progeny lot of seedlings B32–39×A7–83 was in poor ground and did not show well in vigor compared to the open-fertilized lot which was in normal soil at the other end of the field. As was found in 1911, this apparent lack of vigor was due to poor ground only.

The accompanying diagram (fig. 4) shows a comparison between A7–83 and A7–19, the two males used most in our pollination work in 1909, both in rust resistance and vigor transmission on the various lots of progeny from different females. Accompanying the records for each progeny is that of the check lot open-fertilized from the same females. This table shows strikingly the great advantage in using pedigreed seed of good parents. Attention is again called to the fact that the male plants available for pollination of the open-pollinated seed were, with few exceptions, select plants.

PERMANENCY OF RUST RESISTANCE.

A study of rust resistance year by year shows the same permanence of this character in the plants that was shown in the studies of size,

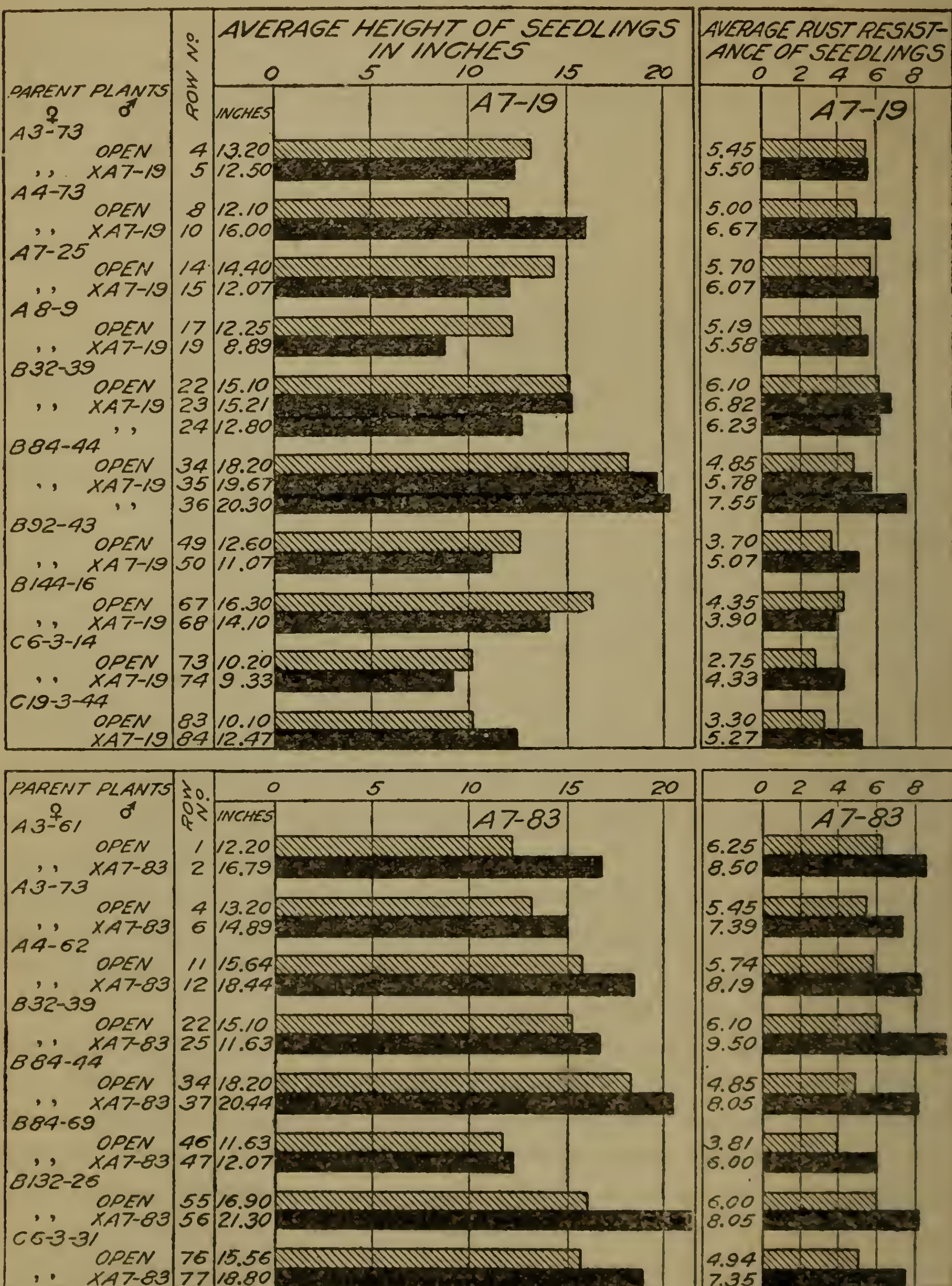


FIG. 4.—Diagram showing the effect on greenhouse seedlings of 1910 of A7-19 and A7-83 with respect to the average heights and average rust resistance of progeny lots in comparison with the progeny from open-fertilized seed from the same female plants.

Parent plants ♀	♂	Row No.	Average height of seedlings (inches)	Average rust resistance of seedlings
A3-73	OPEN	4	13.20	5.45
,,	XA7-19	5	12.50	5.50
A4-73	OPEN	8	12.10	5.00
,,	XA7-19	10	16.00	6.67
A7-25	OPEN	14	14.40	5.70
,,	XA7-19	15	12.07	6.07
A8-9	OPEN	17	12.25	5.19
,,	XA7-19	19	8.89	5.58
B32-39	OPEN	22	15.10	6.10
,,	XA7-19	23	15.21	6.82
	,,	24	12.80	6.23
B84-44	OPEN	34	18.20	4.85
,,	XA7-19	35	19.67	5.78
	,,	36	20.30	7.55
B92-43	OPEN	49	12.60	3.70
,,	XA7-19	50	11.07	5.07
B144-16	OPEN	67	16.30	4.35
,,	XA7-19	68	14.10	3.90
C6-3-14	OPEN	73	10.20	2.75
,,	XA7-19	74	9.33	4.33
C19-3-44	OPEN	83	10.10	3.30
	XA7-19	84	12.47	5.27

Parent plants ♀	♂	Row No.	Average height of seedlings (inches)	Average rust resistance of seedlings
A3-61	OPEN	1	12.20	6.25
,,	XA7-83	2	16.79	8.50
A3-73	OPEN	4	13.20	5.45
,,	XA7-83	6	14.89	7.39
A4-62	OPEN	11	15.64	5.74
,,	XA7-83	12	18.44	8.19
B32-39	OPEN	22	15.10	6.10
,,	XA7-83	25	11.63	9.50
B84-44	OPEN	34	18.20	4.85
,,	XA7-83	37	20.44	8.05
B84-69	OPEN	46	11.63	3.81
,,	XA7-83	47	12.07	6.00
B132-26	OPEN	55	16.90	6.00
,,	XA7-83	56	21.30	8.05
C6-3-31	OPEN	76	15.56	4.94
,,	XA7-83	77	18.80	7.35

yield, etc. Once the individual plants are learned, their individuality is recognized in different seasons. The attack of rust on the greenhouse seedlings in 1910 was very uniform and satisfactory from the standpoint of selection of rust-resistant plants. In 1911 the rust came on very much later and did not get started uniformly over the plat. Some lots were attacked as badly as in 1910, but the ends and outside rows where exposed to the wind and plants that were shaded by trees failed to show as much rust as in 1910. In spite of this fact, the correlation between the rust resistance for the two seasons of the individuals included in the tests of both years is quite high (Table XXIV).

TABLE XXIV.—*Correlation between rust resistance in 1910 and in 1911 of greenhouse seedlings of 1910.*

[Resistance in 1910, subject; resistance in 1911, relative. Coefficient of correlation 0.512±0.015.]

Resistance in 1910.	Rust resistance in 1911 (grades).											Frequency.	Departure from mean.
	0	1	2	3	4	5	6	7	8	9	10		
Grades:													
0	......	......	......	......	......	......	......	1	1	......	......	2	−5
1	......	3	1	1	......	1	2	4	......	......	......	12	−4
2	1	3	1	5	5	6	1	12	2	......	......	36	−3
3	......	1	2	7	5	13	13	34	34	10	......	119	−2
4	......	......	......	3	5	7	12	47	38	23	3	138	−1
5	......	......	1	2	1	15	22	40	74	40	13	208	0
6	......	......	1	......	3	5	13	32	60	39	22	175	+1
7	......	......	......	......	......	3	5	20	52	39	19	138	+2
8	......	......	......	......	......	......	1	9	19	29	26	84	+3
9	......	......	......	......	......	......	......	......	4	9	14	27	+4
10	......	......	......	......	......	......	......	......	1	3	2	6	+5
Frequency	1	7	6	18	19	50	69	199	285	192	99	945	
Departure from mean	−8	−7	−6	−5	−4	−3	−2	−1	0	+1	+2	......	

It should be noted that very few plants show greater rust in 1911 than in 1910. If the rust attacks had been of equal severity both seasons, a much higher value for the coefficient of correlation would have been obtained.

As was shown in Table XXI, there is a definite relation between increase in size and rust resistance. The young seedlings in the greenhouse lot show a relation between rust resistance and size, but to a certain extent the size is dependent even yet on the size of the seed. In Table XXV the heights in September, 1911, are correlated with the rust resistance of plants as observed in September, 1910. The first 25 lots of the greenhouse seedlings were used in this table, as the rust attack in 1911 on this row was more uniform than on the other rows.

TABLE XXV.—*Correlation between height in 1911 (inches) and rust resistance in 1910, row 1 of greenhouse seedlings of 1910.*

[Height in 1911, subject; rust resistance in 1910, relative. Coefficient of correlation 0.484±0.032.]

Height in 1911.	Rust resistance in 1910 (grades).										Frequency.	Departure from mean.
	1	2	3	4	5	6	7	8	9	10		
Inches:												
12	......	......	......	......	1	......	......	......	......	......	1	—9
15	......	......	......	......	......	1	......	......	......	......	1	—8
18	......	1	......	1	......	1	......	......	......	......	3	—7
21	1	......	......	1	3	......	......	1	......	......	6	—6
24	......	1	1	......	......	2	......	......	......	......	4	—5
27	......	3	2	2	6	5	......	......	......	......	18	—4
30	......	1	5	5	7	7	2	2	......	......	29	—3
33	......	2	2	2	9	5	8	3	2	2	34	—2
36	2	1	1	3	8	11	8	4	......	......	38	—1
39	......	3	1	......	7	11	12	8	1	......	43	0
42	......	......	1	1	7	6	13	3	......	1	32	+1
45	......	......	......	......	1	5	9	3	2	......	20	+2
48	......	......	......	1	3	2	3	4	3	1	17	+3
51	......	......	1	......	2	4	1	2	3	3	16	+4
54	......	......	......	......	......	......	4	1	1	......	6	+5
57	......	......	......	......	......	......	......	......	......	1	1	+6
60	......	......	......	......	......	......	1	1	1	......	3	+7
63	......	......	......	......	......	......	......	......	1	......	1	+8
Frequency	3	12	14	16	54	60	61	32	14	7	273	
Departure from mean	— 5	— 4	— 3	— 2	— 1	0	+ 1	+ 2	+ 3	+ 4	......	

SEEDLINGS OF 1911.

In 1910 the work of making pedigree combinations was continued in the spring months. This work was done before the rust developed and was naturally of a more or less hit-or-miss character. Male plants were selected for their individual qualities with the hope that they would transmit these qualities to their offspring. A7–19 and A7–83 were used as check males to test new female plants. The female plants that had given the best resistance both in 1908 and in 1909 were also used in making combinations with these males with the plan that if any of the 1909 combinations showed desirable resistance the 1910 lots of seed would furnish an additional supply of the desirable progeny.

The seed resulting from these pollinations was planted in 1911 at Concord, and when the rust attack developed in August the behavior of the different progenies was much the same as in 1910. A7–19 proved to be a failure in point of transmission of rust resistance and has been discarded. A7–83, however, again performed in a very satisfactory manner. Its progeny proved highly resistant to rust and very vigorous in comparison with seedlings from American stock lacking in rust resistance. Plate XIII shows a row of seedlings from plants in row B24 (fig. 1) compared with the best resistant progeny "Martha Washington" (fig. 2). The striking difference in the two photographs is not so great as the contrast in the field, where the rich green of "Martha Washington" contrasts with the gray brown of the dead seedlings from row B24.

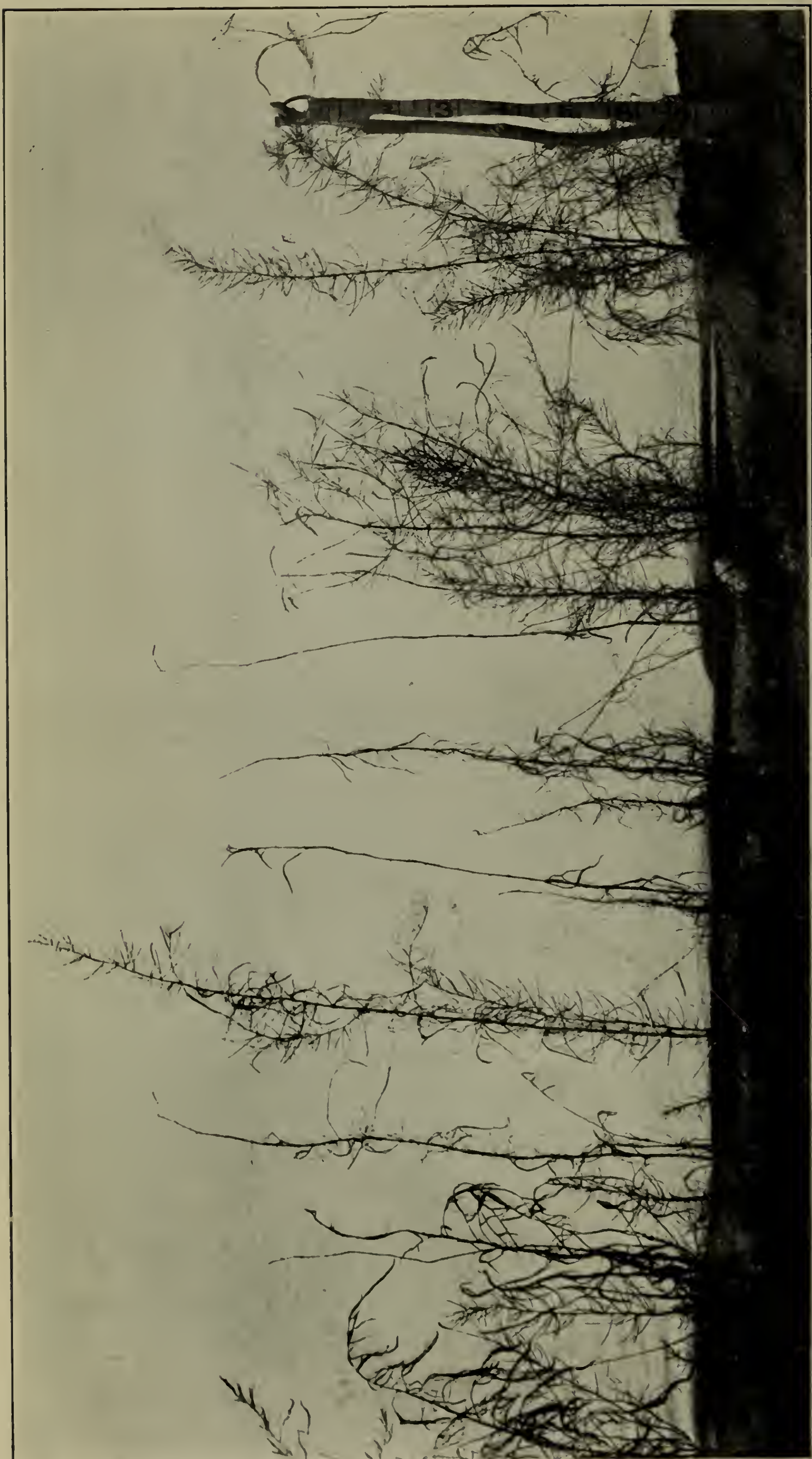
FIG. 1.—WAKEMAN SEEDLING STOCK, SHOWING TOPS ENTIRELY KILLED BY RUST.

(Photograph taken September 25, 1911.)

FIG. 2.—"MARTHA WASHINGTON" STOCK (PROGENY B32-39 × A7-83), COMMERCIALLY IMMUNE PLANTS OF STRONG VIGOR.

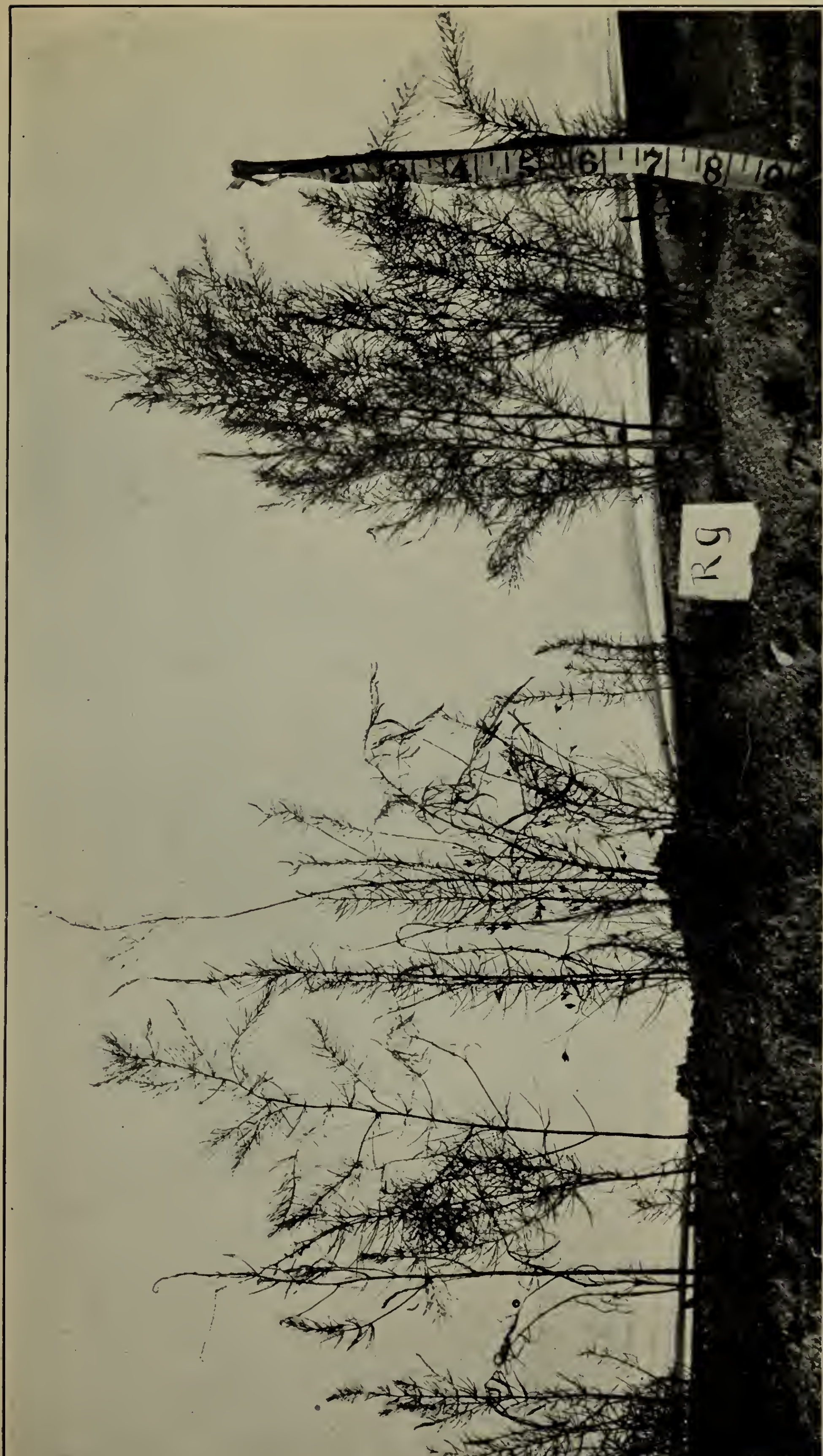
(Photograph taken September 25, 1911.)

PEDIGREE SEEDLINGS OF 1911 AFTER A SEVERE ATTACK OF RUST.



PEDIGREE SEEDLINGS OF 1911 AFTER A SEVERE ATTACK OF RUST, SHOWING VARIABLE RESISTANCE OF STANDARD GIANT ARGENTEUIL, ALL PLANTS SUFFERING FROM RUST.

(Photograph taken September 25, 1911.)

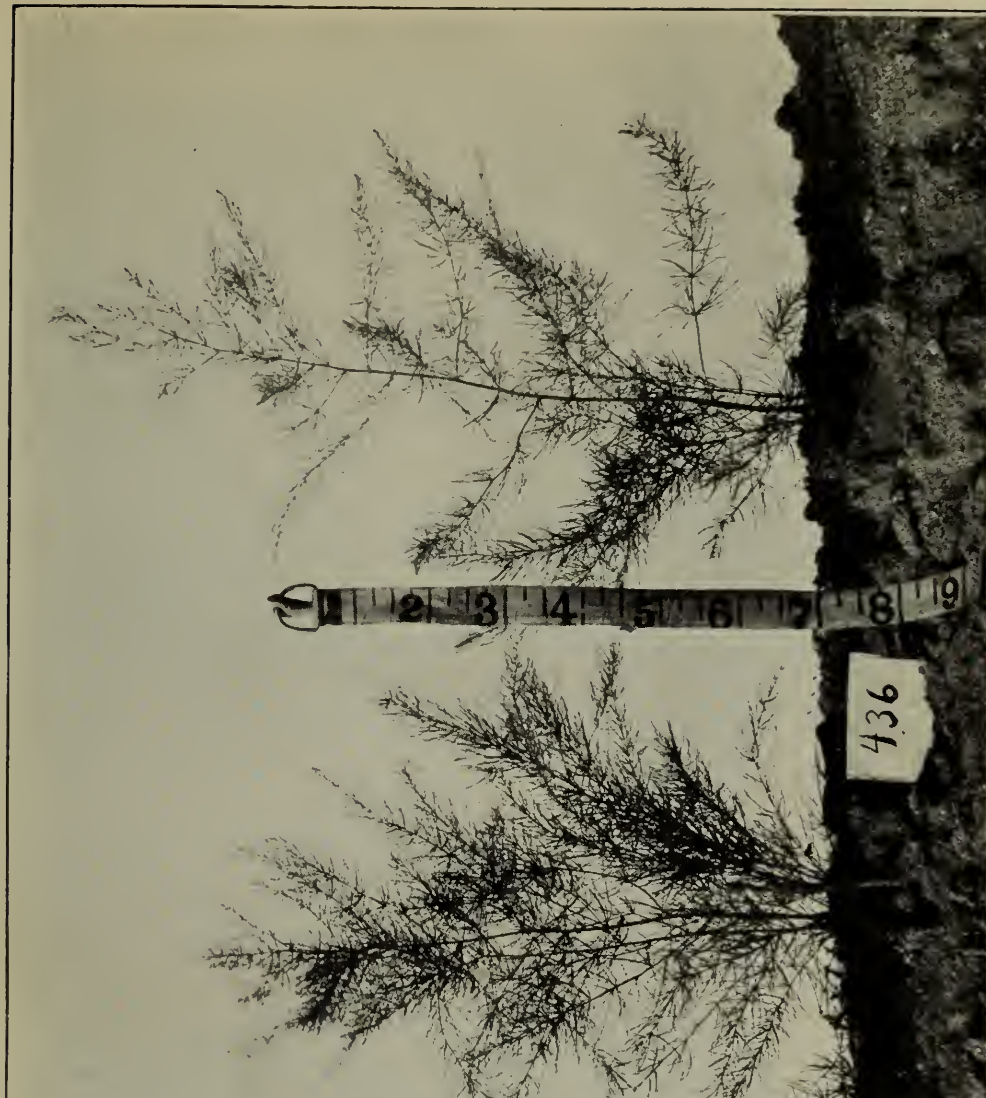


PEDIGREE SEEDLINGS OF 1911 AFTER A SEVERE ATTACK OF RUST, SHOWING PLANTS FROM STANDARD READING GIANT SEED, SOME NEARLY IMMUNE, OTHERS RUSTY.

(Photograph taken September 25, 1911.)

PEDIGREE SEEDLINGS OF 1911 AFTER A SEVERE ATTACK OF RUST, SHOWING THE EFFECT OF CROSSING A7-83 ON A FEMALE PLANT (A2-23) OF AVERAGE RESISTANCE.

(Photograph taken September 25, 1911.)



PEDIGREE SEEDLINGS OF 1911 AFTER A SEVERE ATTACK OF RUST, SHOWING PLANTS FROM OPEN-FERTILIZED SEED OF B32-39, USUALLY QUITE RESISTANT BUT LACKING IN VIGOR.

(Photograph taken September 25, 1911.)

FIG. 1.—SEEDLINGS AT THE SOUTH END OF A BED AT CONCORD, MASS., IN AUGUST, 1910, JUST BEGINNING TO RUST.

FIG. 2.—SEEDLINGS AT THE NORTH END OF THE BED SHOWN IN FIGURE 1 ON THE SAME DAY, SHOWING THE DESTRUCTION OF PLANTS CAUSED BY THEIR PROXIMITY TO A YOUNG BED ON WHICH CLUSTER CUPS DEVELOPED ABUNDANTLY.

EFFECT OF RUST ON ASPARAGUS SEEDLINGS.

Photographs do not show the dead color of plants injured by rust, but in Plate XIV the effect of the 1911 rust attack is shown on American-grown Argenteuil stock, and Plate XV shows American-grown Reading Giant stock in an adjoining row. There is no question that Reading Giant contains plants of greater resistance than any to be found in any lot of Argenteuil grown on the station grounds at Concord. Plates XIV and XV show this difference about as it occurs in the regular field growth of the two stocks. The seedlings from A7–83 progenies are superior to these standard strains in both rust resistance and vigor (Pl. XVI). B32–39 gives very resistant seedlings even when open fertilized with males of medium resistance (Pl. XVII). The small size of the seeds of this plant places the seedlings at a disadvantage, and the combination with A7–83 is needed to give vigor.

In the tests of 1911 no new progeny lots showed a resistance or vigor comparable with that of "Martha Washington." Most of the plants tested will be discarded, a few females being held for a further test. New selections from Reading Giant and from A7–83 progeny are included in the pollination work in 1912. Some of these new selections show such a high individual resistance that it is practically certain that some of the new combinations with A7–83 will show great resistance.

BUD PROPAGATION.

In order to increase the product of seed from individual plants it will be necessary to carry out some methods of vegetative propagation. Preliminary experiments to this end were undertaken in the greenhouse in 1910. Seedlings planted January, 1910, were separated when they had several shoots; the roots were divided more or less evenly and the plants repotted. Nearly all of them grew, and in about a month they were separated again. This was kept up until in January, 1911, two seedling plants were represented by 60 or more plants. When properly handled few plants die. In the fall of 1910 about half the crowns of No. 1 Washington, No. 2 Martha, B32–4, and A2–23 were dug up, shipped to Washington, and planted in the greenhouse. These crowns were split into several smaller clusters and planted in 12-inch pots. New shoots started, but on account of low temperature did not completely develop, and finally died back. When the pots were moved into a warmer house the divided crowns started growing again, and some of the plants have been divided a second time. This method of vegetative propagation will be necessary in breeding and seed-growing work.

PEDIGREE.

In pedigree breeding work the performance of the parent individuals is not important in itself, and is only of value as it shows the ability of the plant to transmit its good qualities to its offspring.

These plants selected for breeding purposes become valuable only as their progeny show uniformly high quality and yield. So when a pedigreed progeny shows a high commercial value its parent plants become of great importance. They should be increased as fast as possible by clonal propagation and should be isolated and allowed to produce as much seed as possible. It is now that records and history become important. Careful record should be kept of their original source, etc., and future development.

In carrying out the work on this breeding project and of private breeding work developing from it the following scheme will be used:

Number.—Each plant that proves of value as a breeding parent will be assigned a permanent serial number. These numbers will not be given to a plant until its progeny show it to be of value as a breeding parent. Its preliminary records will be kept under a temporary number used to mark its location in the testing plat.

Name.—Plants used to produce progenies for commercial planting will be given names as follows: The male plants will be given surnames as Washington, Wilson, Prescott, Wheeler, Moore, etc., the name assigned to one plant not to be duplicated in the future. Female plants will be named by assigning them different feminine names, as Martha, Mary, Edith, etc.

Progenies will take their commercial or trade names from the two parents. Thus the progeny of No. 2 Martha × No. 1 Washington will be known to the trade and growers as "Martha Washington"; No. 3 Edith × No. 1 Washington would be "Edith Washington"; No. 2 Martha × No. 4 Wheeler would be "Martha Wheeler." In this way each progeny would by its name indicate its parents.

Records.—In keeping pedigree records the loose-leaf record book will be used. A primary sheet for each parent admitted to registry will be used, giving its history, description, etc. The performance of the plants as shown by their progeny records will be filed under the female parent as secondary sheets. An abbreviated record of these progeny sheets will be filed under the male parents as secondary sheets to show the performance.

No. 1. Washington ♂.

Pedigree: ♀ unknown.
♂ unknown.

History: Original plant found in 1908, location A7–83. New American Concord-grown stock by Anson Wheeler. Marked as best male in type and rust resistance. Used in 1909 and 1910 in crossing work. In 1911 used as test male in all crossing work.

Progeny: Very resistant to rust and showing an added vigor above open-fertilized progeny no matter what female parent was used.

Propagation: Part of original parent dug up in 1910 for clonal propagation.

No. 2. Martha ♀.
Pedigree: ♀ unknown.
♂ unknown.
History: Original plant found in 1908, location B32–39. Reading Giant stock. Marked as best in rust resistance 1909; rather small type; used in 1909 and 1910 in crossing work. In 1911, under cage, crossed with No. 1.
Progeny: Open-fertilized lots of 1909 and 1910 better in resistance than any other open-fertilized lots tested. × No. 1 progeny best for resistance and type of any seedlings grown.
Propagation: Part of original parent dug up in 1910 for clonal propagation.

When plants from any named progeny develop as good breeding parents they will be assigned new names and handled as distinct parents, their history and pedigree being recorded on their original pedigree sheets.

When by vegetative propagation the original parent plants have increased so that different growers have lots of the same progeny, in offering them for sale the grower's name should accompany the progeny name for purposes of identification in case any error creeps in; as, Martha Washington (Frank Wheeler stock), Martha Washington (C. W. Prescott stock). The registry of new parents for breeding purposes should be through a central breeding organization, so that no duplication of names will occur. For the present this work can be done at the experimental station at Concord. These new progeny lots must be tested in competition with some standard progeny of known rust resistance and quality and their general value determined.

PLANS FOR DISTRIBUTION.

When sufficient stock of any progeny is obtained to warrant distribution to interested growers, plans will be made to plant the stock under conditions favorable to the satisfactory testing of these progenies for resistance to rust. The lots of seed or seedlings issued by the Department will, as far as possible, be sent to growers who will be in a position to aid in extending the cultivation of the rust-resistant strains.

SUGGESTIONS TO BREEDERS AND GROWERS.

In giving advice in regard to asparagus breeding at this time it must be remembered that our experiments are only just begun. Later results are liable to change the methods of procedure to be recommended, but the methods and practices at present followed are here outlined.

RUST RESISTANCE.

If rust is a factor in the region where the work is to be done, resistant varieties are of prime importance. In order to secure resistant selections rust must be present in abundance. Unless one can pick the one superior plant out of a thousand in point of rust resistance the work will be hard.

Late fall is the best time for making field selections, because at that season the rust will be developed sufficiently to have marked the nonresistant plants in the field so that they can be disregarded.

In providing rust for this work in New England it will usually be sufficient to leave an area of nonresistant plants in one corner of the field, preferably that from which the prevailing winds come. If there was plenty of black rust the preceding season, the spring stage will develop in sufficient abundance to provide rust for infection work later in the season. A bed of young asparagus not ready to cut for market is usually sufficient to provide a lodging place for the spring rust. Artificial inoculation has not been necessary at any time in our breeding fields.

ISOLATION.

After two mated plants have had their progeny tested and have proved their value as a breeding pair they will be dug up and propagated by crown division to secure a stock for breeding. This stock will be isolated and used only to grow seed.

Isolation will be secured by building an insect-proof cage over the field or by planting remote from other fields or wild plants, so that bees will not be able to carry in foreign pollen. The mesh of any cage will have to be small enough to keep out the small wild bees. One of the probable methods will be to grow the plants in the greenhouses in the winter. During the winter of 1910–11 in Washington we have been very successful in setting seed in the greenhouse by hand pollination. In making seed plantations a grower will not be limited to one female plant—any number may be planted with one male. Whenever it is desired to use two males a separate field must be used for each.

PROGENY BED.

In planting seed for a progeny test a uniform piece of good land is necessary. The presence of shade, such as overhanging trees, near-by buildings, etc., should be avoided. The bed should be set so as to be uniformly exposed to the attacks of rust from near-by infection plats. Any marked difference in moisture supply is apt to interfere with the test.

As it is not the intention in the progeny test to grow large plants, the custom at the Concord station has been to plant rather late in May so that all danger of frost and also of the first crop of beetles is past. About 10 feet of seedling row is sufficient for a fair test. Of course, many lots of seed will not plant so much as that, but it is a useless waste of space to take any more. Rows are first laid out with a line and then made about 2 inches deep with a hoe. The seed is sown by hand and covered with a rake. Skill in planting is acquired by experience, the intention being to drop about six seeds to the foot. A space of 18 inches between rows is ample to allow for passage and cultivation. The two things to judge in the first year are height and rust. The rust on the seedlings is closely correlated with the rust of the plant in future years and height is correlated with size and vigor. The first year progeny test will eventually be the main test of any plant's value in breeding work.

The use of a standard or uniform lot of seedlings as a check on rust infection is desirable, and where accurate results are expected is necessary. In our work up to 1912 we have used Reading Giant. Pedigree stock of good quality alternating with rusty stock will be planted hereafter as a double check.

VALUE OF BREEDING METHODS.

If asparagus growers ever hope to secure reasonably uniform strains of fixed type, the methods of commercial seed production will have to be changed from their present unscientific condition. With few exceptions no attention is now paid to the male parent and little effort is made to get good female plants, the process of seed selection consisting largely of going into a field that has made a good growth and harvesting seed stalks that have well-grown seed.

Mr. Frank Wheeler, of Concord, Mass., has for several years made a practice of selecting the best male and female plants in regard to type, vigor, yield, and rust resistance. These plants have been allowed to grow and bloom during the cutting season. The seed is saved from only those stems of the female plants that bloomed before the general field plants came into flower. These seed plants are the progeny of imported Argenteuil stock and produce a very desirable quality of seedlings.

In the spring of 1908 about 400 one-year plants of this strain were planted in comparison with a similar plat of a strain known locally as "Small" Argenteuil. The yields from these two plats were kept in 1910 and 1911, as shown in Table IV on page 26.

This difference in yield is apparently due to the difference in the strains in which the selection for large stalks by Mr. Wheeler has been an important factor. No apparent difference was noticed in the comparative rust resistance of the two lots, so that rust does not enter as a factor.

If the above striking difference exists through the simple selection methods used by Mr. Wheeler, would not other good farmers be justified in trying pedigree methods in growing seed? The above-mentioned strain is not pedigreed from either side, the parentage complex including about 20 individuals of each sex. Mr. Wheeler in 1910 and 1911 planted his lots of seed from each female in separate rows. The difference was so striking that in the future pedigree methods will receive more attention.

PROTECTION FROM BEETLES.[1]

One thing to be considered in seed production is the effect of the red or twelve-spotted asparagus beetle (*Crioceris 12-punctatus*), the larval stages of which live in and destroy the asparagus berries. This beetle proved a serious factor in the breeding work last year, and is liable to become worse as time goes on. The first specimens of this beetle found in Concord were discovered in the fall of 1908. The fall of 1910 showed nearly as many as of the ordinary species, *Crioceris asparagi*. Paper bags are not sufficient protection, as in several cases the berries under bags were destroyed. The beetles had either laid their eggs before the plants were bagged or else crawled up inside through the open spaces around the stems. Cages of 16-mesh wire fly screen keep out the red beetle but let in the smaller specimens of the common asparagus beetle. Both kinds may be kept out by the use of 18-mesh wire screen, which will be hereafter used.

PROTECTION OF NONIMMUNE FIELDS.

Spraying methods have been developed by different experiment-station workers in the past that if carefully followed by the grower will keep down the rust. The trouble in applying sprays and the high cost of their efficient application has kept many good growers from using them. Some farmers have gone out of the asparagus business while others have secured the best stock they could find and by careful methods have kept on. The high prices caused by increasing demand and lessening supply has made the profit in asparagus really higher than it was before the rust became known in the country.

It is now certain that by proper pedigree breeding work the whole question of noticeable rust injury in asparagus may be eliminated. At the same time the pedigree breeding work will make uniform and vigorous strains, thus greatly increasing the yield per acre. The elimination of rust as a factor in asparagus growing will render larger yields possible, so that the market price in many locations where rust now prevents adequate returns will fall within reach of the large body of consumers. At present in most regions asparagus is a luxury.

[1] For a full discussion of the two asparagus beetles and of the methods to be used for their control, the reader is referred to Circular 102 of the Bureau of Entomology, U. S. Department of Agriculture.

SUGGESTIONS FOR RUST PREVENTION.

Although the breeding work being carried on with asparagus will eventually lead to the control of rust in commercial plantings, several years must elapse before this result will become effective. Meanwhile, it is necessary to take all measures practicable to prevent the destruction of existing fields of asparagus by the rust. To this end the main factor is to keep the rust away from the fields in summer just as long as possible.

As pointed out by Smith and others, wild asparagus growing around the borders of the fields, along fences, ditches, etc., is one of the worst enemies of the grower. These wild plants act as infection centers and their influence can be easily traced later in the season when the cutting beds have grown up. During the summer of 1910 the writer made an examination of the fields near Concord just at the time the rust was coming on and in every case of infection was able to trace the cause to asparagus plants that had not been cut up to the close of the infection period of the spring rust (Pl. XVIII). When rust was found in a commercial field by following it up to the northwest, the direction from which the prevailing winds come, a young bed, an old neglected bed, or wild asparagus was found in every case and always with the remains of cluster-cup infections. Wild plants wherever found should be dug up and burned. New beds should be planted only at rare intervals of time and then if possible where they will be to windward of a cutting bed. Keep the seedlings out of the cutting bed, at least let none stay in at the time the bed is allowed to grow up after the cutting season. Allow no poor shoots to grow up in the cutting field. In other words, keep down every shoot of asparagus until the middle of June in the latitude of Boston and see that neighboring farmers do the same. In the fall the tops should be removed carefully from 1-year-old beds that are not to be cut the next year. This will in a large measure reduce the liability of infection from this source.

The writer does not recommend the removal of tops from a mature bed in the fall. The ordinary practice in the vicinity of Concord is to leave the bed undisturbed in the fall so that the tops will act as a winter cover and prevent the blowing of soil or snow. In the spring these tops are cut with a disk harrow. Fields in which this treatment had been used have been examined for spring rust after the bed had grown up at the end of the cutting season, but in no case have cluster cups been found. The Massachusetts station has at Concord a 3-acre fertilizer experimental plat on which plants have been infected during 1909, 1910, and 1911 from young beds near by that were not being cut. No cluster cups were found in this 3-acre bed except on plants left for breeding purposes.

SUMMARY.

Puccinia asparagi, the European asparagus rust, was discovered in America in 1896 and in the next six years spread over the asparagus-growing regions of the United States, causing great damage. In the Eastern States no successful remedy was found, although some strains were found to be more resistant than others. Among the resistant varieties were Argenteuil and Palmetto.

The Massachusetts Asparagus Growers' Association, organized in 1906 to obtain a resistant variety by breeding, secured the cooperation of the Massachusetts Agricultural Experiment Station and the United States Department of Agriculture in establishing experimental grounds for this work at Concord.

Previous work on the life history of the disease shows that the rust in all its stages occurs only on asparagus and that the uredo stage is the most injurious. The injury is due to the mechanical and physiological effect on the summer growth which prevents the storage of food supplies for the growth during the next cutting season.

A large number of strains from America and Europe have been collected and tested for rust resistance. No variety proving uniform or satisfactory, breeding work was undertaken to produce a stock that would be commercially immune. Some wild species have been imported from the Old World and one or more hybrids have been produced.

In making selections for rust resistance several acres of the best stock obtainable were used. From the different strains several hundred plants have been selected for pedigree testing after being subjected to attacks of rust.

Rust resistance in asparagus seems to be based upon structural differences. Vigor is not necessarily correlated with resistance.

Breeding work in asparagus is complicated by the fact that the species is diœcious, so that two parents must always be used in seed production. Hand pollination is used for pedigree work.

Progeny tests of select plants have been made each season since 1909. The rust resistance and vigor of these seedlings have determined the value of the breeding parents. The test male A7–83 and the test female B32–39 have given a very superior progeny, which has proved satisfactory as a "commercially immune" type. This progeny has been named and plans are under way for its production in quantity.

In carrying out the breeding work, studies have been made of the effect of the weight of seed on seedling vigor, the effect of seedling size on the plant in the field, etc. Correlations between size of plant, yield, rust resistance, etc., have been of value in carrying out the work.

Bud propagation of select breeding parents has been inaugurated to promote more extensive seed production.

Breeders and growers are advised to take up pedigree breeding to produce good strains and to use careful methods in keeping rust out of producing fields.

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Issued January 8, 1913.

U. S. DEPARTMENT OF AGRICULTURE.

BUREAU OF PLANT INDUSTRY—BULLETIN NO. 264.

B. T. GALLOWAY, *Chief of Bureau.*

THE PURPLING CHROMOGEN OF A HAWAIIAN DIOSCOREA.

BY

HARLEY HARRIS BARTLETT.

Chemical Biologist, Drug-Plant, Poisonous-Plant, Physiological, and Fermentation Investigations.

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LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry,
Office of the Chief,
Washington, D. C., September 10, 1912.

Sir: I have the honor to transmit herewith and to recommend for publication as Bulletin No. 264 of the series of this Bureau a manuscript by Mr. H. H. Bartlett, Chemical Biologist, entitled "The Purpling Chromogen of a Hawaiian Dioscorea." This paper, submitted by Dr. R. H. True, Physiologist in Charge of Drug-Plant, Poisonous-Plant, Physiological, and Fermentation Investigations, deals with a subject of great interest to both physiologists and geneticists—to the former because of the important function in the oxidizing mechanism of the plant cell, which has recently been ascribed to the vaguely characterized plant chromogens; to the latter because of an increasing need for the chemical identification of the "unit characters" for color in plants.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

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B. P. I.—776.

THE PURPLING CHROMOGEN OF A HAWAIIAN DIOSCOREA.

INTRODUCTION.

In 1903 the Office of Foreign Seed and Plant Introduction of the Bureau of Plant Industry received through Mr. Jared G. Smith, Special Agent of the Department of Agriculture in Charge of the Hawaii Agricultural Experiment Station, specimens of a Dioscorea under the native name "Hoi."[1] These were inventoried as Nos. 10311 and 10312 [2] and cultivated at the subtropical gardens at Miami and Gotha, Fla. Because of the confused state of the genus it is not now possible to assign a specific name to this Dioscorea.[3] Perhaps the taxonomy of the group to which it belongs may be treated in a future paper. For the present our plant may be conveniently referred to as the Hawaiian bitter yam. It has recently been offered to the horticultural trade by Childs [4] under the name of "air potato, or giant yam vine."

The name "air potato" has reference to large aerial tubers which develop in the axils of the leaves. Many species of Dioscorea have these tubers, but none exhibits them to greater perfection than the Hawaiian bitter yam. The tubers (shown in natural size in Pl. I) are propagative organs. In this country they are the sole means of reproducing the plant, for all the tubers originally imported seem to have been obtained from pistillate vines. However, the writer is informed by Dr. E. V. Wilcox, Special Agent in Charge of the Hawaii Agricultural Experiment Station at Honolulu, that in Hawaii both pistillate and staminate vines are found. Many yams

[1] The name "Hoi" seems to be of generic application to yams in Polynesia. Hillebrand, commenting on a species which he describes under the name *Dioscorea sativa*, says: "The yam, common in the forests of the lower zone, was cultivated for the supply of ships before the introduction of the potato, particularly on Kauai and Niihau. The species ranges westward over all the regions lying between the Hawaiian Islands and Africa, and its native name, 'Hoi,' follows it to Sumatra. The axillary buds are called 'alaala.'"—Flora of the Hawaiian Islands, p. 438.

[2] Bulletin 97, Bureau of Plant Industry, entitled "Seeds and Plants Imported during the Period from December, 1903, to December, 1905," p. 9. 1907.

[3] In Bulletin 97, Bureau of Plant Industry (loc. cit.), the name *Dioscorea divaricata* was used with a question mark. From this species, however, the plant described in this paper differs in many characters, of which the most striking is the radial rather than dorso-ventral disposition of the tissues of the aerial tuber.

[4] Childs's Combination Catalogue for 1910, p. 3 of cover.

which, so far as known, have only one sex are propagated exclusively by vegetative means.

There is a chromogen in the aerial tubers of the Hawaiian bitter yam which is of unusual interest in that it appears to be related to the class of ammonia-greening anthocyanins. It has been made the subject of considerable study in the hope that it might afford a clue to the better understanding of these compounds.

OCCURRENCE OF THE CHROMOGEN.

The freshly cut surface of an "air potato" is greenish white, but on exposure to the air it quickly becomes brown by the oxidation of a chromogen (probably a tannin) which has not been investigated. If the freshly cut surface is treated with ammonia, the natural greenish color is greatly intensified, especially in the region of the cambium immediately beneath the cortex.[1] Occasional tubers have pale-purplish instead of greenish flesh, and many greenish tubers become purplish in part when sprouting. If treated with ammonia these purplish tubers also color, just as the greenish ones do, to a somewhat intensified green. On the contrary, treatment with acids will not change the color of a greenish tuber to purplish. If, however, the juice is expressed from a greenish tuber or from the green tips of growing stems and acidified with acetic or hydrochloric acid, a pinkish or purplish color of considerable intensity develops. Nevertheless, the intensity of the green which resulted when a cut tuber was treated with alkali seemed to be out of proportion to the pink which resulted from acidifying the juice. Therefore, in addition to the pigment which was obviously one of the numerous compounds classed as ammonia-greening anthocyanins, a second substance was sought, which like the anthocyanin would give a green reaction with alkali, but unlike the anthocyanin would be colorless or nearly so in acid solution. After much experimenting, such a compound was isolated—a chromogen which in dilute neutral or acid solution is yellowish, which forms intensely wine-red solutions on oxidation with plant oxidases or weak inorganic oxidants and intensely green alkali salts both before and after oxidation.

ISOLATION OF THE CHROMOGEN.

As the result of several trials the following method is recommended for the isolation of the chromogen in a comparatively pure state.

[1] In regard to the anomalous morphology of the Dioscorea tubers, see the following:

Bucherer, Emil. Beiträge zur Morphologie und Anatomie der Dioscoreaceen. Bibliotheca Botanica, no. 16, 1889.

Dale, Elizabeth. On the Origin, Development, and Morphological Nature of the Aerial Tubers in Dioscorea Sativa. Annals of Botany, vol. 15, 1901, pp. 491–501.

Goebel, K. Die Knollen der Dioscoreen und die Wurzelträger der Selaginellen, Organe, welche zwischen Wurzeln und Sprossen stehen. Flora, vol. 95, 1905, pp. 167–212.

AERIAL TUBERS OF THE HAWAIIAN BITTER YAM.

(Natural size.)

The tubers are sliced into narrow strips and dried as expeditiously as possible at 60° C., or if plenty of material is available the tubers are pared and the parings only are dried. The dry material after having been ground is extracted at the room temperature, first with ether and then with a mixture of equal volumes of ether and alcohol. To the ether extract a small quantity of alcohol is added and the ether distilled off, leaving the chromogen in alcoholic solution. This is mixed with petroleum ether, and water is added to bring about a separation of a petroleum-ether layer (containing wax, etc.) from the alcoholic layer (containing the chromogen). The latter is added to the ether-alcohol extract.

The ether-alcohol extract is distilled until the residue is free from ether and considerably concentrated. It is then mixed with a cold, concentrated calcium-chlorid solution (or mixed with water and saturated with common salt) and shaken first with several portions of petroleum ether to remove various impurities, and then with acetic ether to remove the chromogen.[1] The chromogen in acetic-ether solution is then shaken with a large excess of concentrated aqueous lead-acetate solution. The copious gummy precipitate, most of which remains suspended in the acetic-ether layer, is filtered off on a Buchner funnel and washed with acetic ether. The acetic-ether filtrate, which is of a clear golden-yellow color, is now shaken successively with solutions of ferrous sulphate (strictly free from ferric salts and dissolved in recently boiled water) and bipotassium hydrogen phosphate and finally several times with distilled water. After filtering the solution it is poured into a large excess of petroleum ether. The chromogen is thrown down as a finely divided, white, opalescent precipitate, which speedily coalesces to form a soft, brown, resinous mass. In this condition the chromogen contains much acetic ether. It may be partially dried in a vacuum desiccator, then pulverized as much as possible, and dried at 78° C. in a vacuum drying oven in a slow stream of rarefied hydrogen.

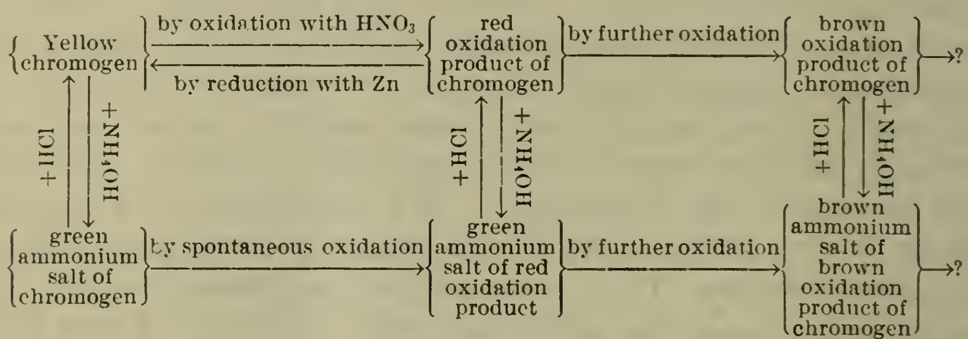
PHYSICAL AND CHEMICAL PROPERTIES.

The chromogen as obtained by the process outlined is a brown resinous compound soluble in alcohol, acetic ether, and chloroform, moderately soluble in ether, and insoluble in petroleum ether and water. Its melting point is not sharp, and it has not been possible

[1] The saline alcohol and water solution which remains after this operation soon changes from yellow, which color is imparted to it by a small residue of unextracted chromogen, to a fine purple. The purple compound is the same oxidation product of the chromogen which is mentioned on page 10 of this bulletin. Its formation in the presence of chlorids is no doubt analogous to the bluing of guaiac resin by chlorids, as described by Alsberg. (See Beiträge zur Kenntnis der Guajak-Reaktion, Schmiedeberg-Festschrift, 1908. Supplement, Archiv für Experimentelle Pathologie und Pharmakologie, pp. 39–53.) The oxidation product is precipitated by lead acetate, so that any small quantity which may contaminate the chromogen after salting out is removed in the next operation.

to obtain it in a crystalline condition. It is fairly stable in neutral solutions, and in alcoholic solution is unchanged by boiling with zinc dust under a reflux condenser. Prolonged heating in contact with the air, more particularly when a reducing substance, such as zinc, is not present, results in its destruction by oxidation. Acid solutions, which are of the same color as neutral solutions, slowly deposit insoluble brown compounds, with simultaneous destruction of the chromogen.

The chromogen is the first member of a series of acids. The other members are successive oxidation products of the chromogen. Each acid forms a series of salts. The relationship of these derivative compounds is as follows:



The oxidation of the chromogen to its red derivative and the formation of the intensely green ammonium salt of either the chromogen or its red derivative constitute the two characteristic color reactions which are mentioned on page 8.

The red oxidation product is best observed when the chromogen is dissolved in acetic ether and shaken with water containing a trace of fuming nitric acid or ferric chlorid. Under these conditions a beautiful wine-red acetic-ether solution of the oxidation product is obtained. If it is carefully washed by repeatedly shaking with distilled water the red solution may be preserved for several days, but not indefinitely, for oxidation will spontaneously proceed until the solution becomes brown by the formation of a further oxidation product. Even this brown compound does not represent the final stage of the oxidation of the chromogen, as a further change is indicated by the ultimate deposition from solution of a substance insoluble in acetic ether. The red stage in the oxidation of the chromogen is very easily overstepped, with the immediate formation of brown compounds, if the mineral oxidants used are too concentrated or if the chromogen is not protected from a too violent oxidation by being dissolved in a solvent, such as acetic ether or chloroform, which is immiscible with the solvent of the oxidant (water).

The chromogen may be oxidized, in aqueous solutions containing sufficient alcohol to prevent precipitation, through the agency of plant oxidases (see p. 12) or of halogen salts (see p. 9). Under such circumstances a purple solution is obtained. On this account the adjective "purpling" has been applied to the chromogen in the title of this paper. The purple compound which is formed in aqueous solution is identical with the red compound which is formed in acetic-ether solution. It may be completely removed from aqueous solutions by shaking with acetic ether. In water it is purple, in organic solvents red. The red compound is too unstable to be obtained in a solid state by the evaporation of its acetic-ether solution.

When the chromogen or one of its oxidation products is treated with an alkali a salt is formed. In striking contrast to the series of acids, the salts are all totally insoluble in acetic ether or chloroform, but very soluble in water. The salts of both the chromogen and its red oxidation product are deep green in aqueous solution. If a solution of the chromogen or of the red compound in acetic ether is floated on dilute ammonium hydroxid the corresponding ammonium salt is formed at the juncture of the two liquids. After the layers are shaken together the deep-green aqueous salt solution will settle out, leaving the acetic ether colorless. By immediately acidifying the mixture and shaking it the process may be reversed.

The ammonium salt of the chromogen in aqueous solution absorbs oxygen from the air with avidity, passing first, without visible change, to the similar salt of the red oxidation product. If the salt is acidified at this stage the red compound is obtained, and the chromogen itself may be recovered therefrom by reduction with zinc. On longer standing, however, the green salt solution becomes brown. If it is then acidified and shaken with acetic ether a brown acetic-ether solution is obtained, which to all appearances contains the same brown oxidation product of the chromogen that results when the chromogen is directly oxidized beyond the red stage by nitric acid.

Dr. P. G. Nutting, of the Bureau of Standards, has kindly photographed and absorption spectra of the chromogen, the wine-red oxidation product, and the two green alkali salts. From an examination of the negatives he has reported as follows:

The yellow chromogen solution, 1 : 2,000 in acetic ether, 21 millimeters thick, shows complete absorption of the ultra-violet. Transmission begins gradually at 0.39, just at the limit of the visible violet, and increases steadily to full value in the greenish blue at wave length 0.52.

The red oxidized chromogen, 1 : 2,000 in acetic ether, 21 millimeters thick, shows complete absorption of violet, blue, and green, gradually increasing transmission through the yellow and orange, and full transmission of the red. Transmission begins at wave length 0.55 in the greenish yellow and becomes full at 0.63 in the red.

The green ammonium salts, 1 : 4,000 and 1 : 40,000 in aqueous solution, showed absorption so nearly nonselective that no determinations were made upon them.

When the chromogen is oxidized by plant oxidases it need not be protected from the oxidant by solution in a solvent immiscible with water. Potato juice (oxidase), prepared by grinding potato parings and pressing through raw silk, actively oxidizes chromogen, which is added in alcoholic solution, first, to the wine-red compound and then to the more highly oxidized brown compound. The oxidation by potato juice is accompanied by measurable oxygen absorption from the air. The potato oxidase is much more active when oxygen is supplied by hydrogen peroxid. Horse-radish juice alone (peroxidase) fails to oxidize, even upon prolonged shaking, as is shown by the fact that the chromogen may be recovered unchanged in color, and by the additional fact that there is no measurable oxygen absorption. The tests for oxygen absorption were conducted with the apparatus designed and described by Bunzel.[1] Hydrogen peroxid alone does not oxidize the chromogen, but in the presence of hydrogen peroxid horse-radish juice is a powerful oxidant, carrying the oxidation almost instantaneously to the wine-red stage, at which it halts. The chromogen is as sensitive in its reactions to oxidases and peroxidases as guaiac resin.

Juice expressed from the Dioscorea tubers given rather unsatisfactory results as an oxidase solution because of its highly mucilaginous nature and because of the large quantity of easily oxidized tannin which it contains. Nevertheless, juice pressed from parings by a hydraulic press readily oxidized the chromogen in the presence of hydrogen peroxid to the same wine-red compound which was obtained when other plant juices or inorganic oxidants were used. The presence of hydrogen peroxid seemed to be essential to the reaction: that is, to use the terminology of Chodat, the tuber contains a peroxidase but no associated oxygenase. A moderately active oxidase solution was prepared from green tips of the stems of tubers which had sprouted in the laboratory by mashing with water and filtering. This solution gave (1) no reaction with chromogen alone, (2) an immediate darkening and eventually a deep-purple color with chromogen plus hydrogen peroxid, and (3) a more delayed but eventually pronounced purpling with hydrogen peroxid alone. In the second case, the purple-colored substance was of the usual wine-red color when shaken out into acetic ether. In the third case, the apparent indication that a chromogen was present in the juice led to a further investigation of the stems in the hope of finding in what form, if any, the chromogen of the tubers was translocated to the growing parts.

Several tubers were allowed to sprout in a dark chamber until the white etiolated shoots were 6 inches to a foot in length. These were cut into small pieces, discarding only the purple nodes and rudimentary leaves, and dropped into boiling acetic ether. The material thus

[1] Bunzel, H. H. The Measurement of the Oxidase Content of Plant Juices. Bulletin 238, Bureau of Plant Industry. 1912.

killed was ground in a mortar with the acetic ether and the solution filtered. The resulting pale-yellow solution was shaken with water and the two layers separately examined. The acetic-ether layer contained no chromogen which could be demonstrated by oxidation with dilute fuming nitric acid or by the formation of a characteristic alkali salt. The colorless water layer gave an immediate darkening (green passing quickly to brown) with ammonia, and became rose pink on the addition of acetic acid. The color obtained when the solution was treated with dilute fuming nitric acid was of the same intensity as that obtained with acetic acid, and it was therefore concluded that the substance giving the reactions was the water-soluble anthocyanin of the stems. The rose-pink compound formed by acidifying the solution could not be shaken out into organic solvents. Since the chromogen of the tuber is insoluble in water it would be surprising to find it in the stem. If, as seems likely, the chromogen is concerned with the formation of the water-soluble anthocyanin, the change to a soluble compound takes place in the tuber. In this connection it should be pointed out that no difference could be observed between the water-soluble anthocyanin of the stems and that of the tubers. (See p. 8.) The purpling of stem juice with hydrogen peroxid alone would seem to be due to the slight acidity of commercial solutions of this reagent.

EXISTENCE OF A SECOND CHROMOGEN.

It remains to call attention to a second chromogen which occurs in the tubers. The precipitate obtained when the ammonia-greening chromogen was treated with lead acetate (see p. 9) was dried and triturated with acetic ether slightly acidified with acetic acid. A small part of the precipitate was thereby decomposed. After filtration the filtrate was neutralized by shaking with bipotassium hydrogen phosphate and washed with water. Precipitation with lead acetate and recovery by suspension in acidified acetic ether were repeated, and the chromogen precipitated in a relatively pure condition by adding petroleum ether. It formed a brown resinous mass, similar in appearance to the ammonia-greening chromogen already described. Its yellow solution in acetic ether yielded a water-soluble purple ammonium salt on shaking with dilute ammonium hydroxid. This salt passed entirely into the ammoniacal aqueous layer, leaving the acetic ether colorless. On standing, it quickly oxidized to a brown compound, but if reacidified immediately after its formation the yellow unchanged chromogen could be recovered in the acetic ether. This ammonia-purpling chromogen gives an intensely red compound when oxidized by shaking its acetic-ether solution either with greatly diluted fuming nitric acid or with horse-radish peroxidase + hydrogen peroxid. The ammonium salt of the red oxidation product is a purple compound, insoluble in both water and acetic ether.

NOMENCLATURE.

It is proposed to designate the ammonia-greening chromogen of the Dioscorea tuber as rhodochlorogen. In so doing, the precedent set by Reinke[1] is followed. This author gave the name rhodogen (formed on the analogy of the word chromogen) to the colorless substance from which on oxidation he obtained the red coloring matter of beets. The word "rhodochlorogen" (from *ῥόδον*, rose, *χλωρός*, green, and *γενής*, producing) is formed with reference to the colors of the oxidation product and the alkali salts. It may be used as a generic designation for chromogens which appear to be related to the ammonia-greening anthocyanins, until the chemistry of these compounds is better understood. The ammonia-purpling chromogen of the Dioscorea tuber is hardly well enough established as a chemical individual to warrant applying even a temporary designation to it.

POSSIBLE RELATIONSHIP WITH THE ANTHOCYANINS.

In order to fix the characteristics of the two Dioscorea chromogens in mind, they are contrasted briefly in Table I before proceeding with a discussion of their possible relationship to the anthocyanins.

TABLE I.—*Comparison of two Dioscorea chromogens.*

	Rhodochlorogen.	Ammonia-purpling chromogen.
Solution in acetic ether	Yellow	Yellow.
Ammonium salt formed in acetic ether	Insoluble green precipitate	Purple, partly soluble.
Ammonium salt shaken into water	Deep-green solution	Purple solution.
Water solution of ammonium salt acidified after partial spontaneous oxidation and shaken with acetic ether.	Purple acetic-ether solution	Insoluble purple precipitate.
Oxidized by peroxidase + hydrogen peroxid in very dilute alcoholic solution.	Purple solution	Red-purple solution
Oxidized by nitric acid in acetic-ether solution.	Wine-red solution	Brownish red solution, eventually a purple precipitate.
Ammonium salt of oxidation product formed in acetic ether.	Insoluble green precipitate	Insoluble purple precipitate.
Ammonium salt of oxidation product in water.	Deep-green solution	Insoluble purple precipitate.

The similarity of these chromogens to the anthocyanins at once suggests itself. There is present in the stems and petioles of the bitter yam a water-soluble cell-sap color which belongs to the

[1] Reinke, J. Ein Beitrag zur Kenntnis leicht oxydirbaren Verbindungen des Pflanzenkörpers. Zeitschrift für Physiologische Chemie, vol. 6. 1882. p. 263. "* * * In der lebenden Pflanzenzelle leicht oxydirbare Substanzen vorhanden sind, welche begierig atmosphärischen Sauerstoff anziehen, und mit demselben Oxydationsprodukte bilden." In referring to the chromogen which produces the red coloring matter of beets, Reinke says. "Weil der Sauerstoff der atmosphärischen Luft diese Substanz zu einem roten Farbstoff zu oxydiren vermag, will ich dieselbe als Rhodogen bezeichnen * * *."

apex of the petioles (see fig. 1). It forms green salts with alkalis, ammonia-greening class of anthocyanins. It is of a very deep purple-red color and is particularly conspicuous in the enlarged base and which on standing spontaneously oxidize to brown compounds. It may be reduced by boiling with zinc dust to a yellow compound, from which the anthocyanin is regenerated by oxidation. (The oxidation of the yellow reduction product takes place spontaneously when its solution is exposed to the air.) This yellow reduction compound of the anthocyanin is strictly comparable to rhodochlorogen in that it forms green alkali salts. Rhodochlorogen oxidizes to a purple or red compound, depending upon the solvent. The yellow reduction product reoxidizes to purple or reddish purple anthocyanin. Thus far, the similarity of the two compounds is perfect. Just as the yellow rhodochlorogen and its purple or red oxidation product both form green salts, so the purple anthocyanin and its yellow reduction compound both form green salts. The two pairs of compounds are strikingly unlike only in their solubility relations, rhodochlorogen and its red oxidation compound being soluble in acetic ether or chloroform but not in water, whereas the anthocyanin and its yellow reduction compound are soluble in water but not in acetic ether or chloroform. The anthocyanin has not been isolated and is therefore not known to be a glucosid, but it seems probable, in view of Grafe's investigations, that the two water-soluble compounds are glucosids of the two which are not water soluble. Even if they are not related in this way, there is strong circumstantial evidence that the chromophoric nucleus in the molecules of rhodochlorogen and anthocyanin is identical and therefore that the two substances are in some way genetically related in the plant metabolism.

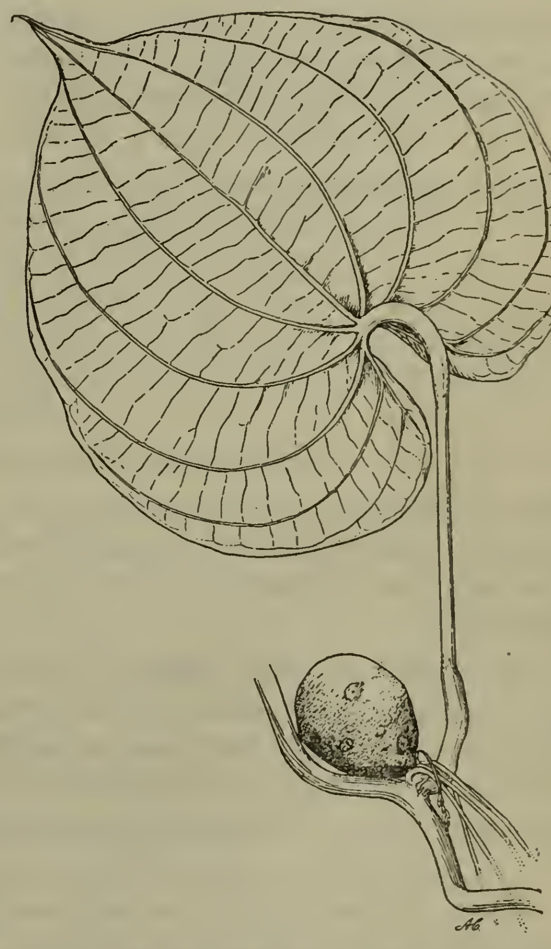


FIG. 1.—Leaf of Dioscorea showing petiole enlarged at base and apex, immature aerial tuber, and bases of racemes. (Reduced to one-half diameter.)

In this connection attention should be called to the fact that Grafe[1] has resolved the ammonia-greening anthocyanin of *Althaea rosea* into two components. One, a water-soluble sap color of the usual type, is a glucosid of the formula $C_{20}H_{30}O_{13}$. The other, insoluble in water but soluble in absolute alcohol, is not a gucosid. Its formula $C_{14}H_{16}O_{6}$ differs from that of the glucosid by a molecule of water and a molecule of glucose. In their color reactions the two components are similar, and there is little doubt that they contain the same chromophoric nucleus. The nonglucosid, which is insoluble in water, greatly resembles the red oxidation product of rhodochlorogen in that it may be reduced to a yellow compound which seems to be analogous to rhodochlorogen itself.[2]

Glan[3] had already shown before Grafe took up his work on the mallow pigments that the water-soluble anthocyanin of *Althaea rosea* was capable of reduction to a colorless compound (yellow if in sufficiently concentrated solution?) which again spontaneously reverted to the anthocyanin by oxygen absorption. In this respect it is similar to the Dioscorea anthocyanin. There seems to be a general parallelism between Grafe's compounds and those which occur in the Hawaiian yam. Not too much stress can be laid upon this parallelism, however, in view of the fact that rhodochlorogen has not been adequately studied either from a chemical or from a physiological standpoint, and its genetic relationship to a water-soluble anthocyanin, although in the highest degree probable, remains unproved.

In regard to the ammonia-purpling chromogen of Dioscorea tubers little can be said. It has been obtained in sufficient quantity for only a few tests and was probably not even approximately pure. It may be pointed out, however, that in its color reactions it resembles some of the nonammonia-greening anthocyanins.

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[1] Grafe, Viktor. Studien über das Anthokyan.—I. Sitzungsberichte der mathematisch-naturwissenschaftlichen Klasse der kaiserlichen Akademie der Wissenschaften, vol. 115, pt. 1, 1906, p. 975; II, vol. 118, pt. 1, 1909, p. 1033. Since this paper was submitted for publication (June 24, 1912) the third article of this series has reached the writer.

[2] "Bei der Reduktion mit Jodwasserstoffsäure entsteht eine gelbe Substanz, die durch nachfolgendes Schmelzen mit Aetzkali Protokatechusäure oder Brenzkatechin ergibt. Der Zusammenhang des Malvenanthokyans mit Gerbstoffen oder Substanzen der Xanthon-Flavongruppe wird dadurch und mit Rücksicht auf die Versuche anderer wahrscheinlich gemacht."—Grafe, Viktor, op. cit., II, vol. 118, pt. 1. 1909 p. 1044.

[3] Glan, Rudolf. Ueber den Farbstoff der schwarzen Malve (*Althaea rosea*). Inaugural Dissertation, Erlangen, 1892. "Der Farbstoff der Malvenblüten gibt mit Säuren eine rote, bei der Neutralization mit Basen eine blaue und mit überschussigen Basen eine grüne Farbung * * * Reduktionsmittel entfärben denselben, doch kann er durch Sauerstoffaufnahme noch regeneriert werden * * *. Die Einwirkung von verdünnter Schwefelsäure auf den Farbstoff der Malve leifert Dextrose und ein Spattungsprodukt, welches Acetylgruppen aufzunehmen vermag, mit Alkalien Brenzcatechin, zuletzt reichlich Protocatechusäure liefert Der Farbstoff hat Glycosidcharakter und darf als mit Dextrose combiniertes Protocatechusäurederivat aufgefasst werden."

WHELDALE'S THEORY OF ANTHOCYANIN FORMATION.

Miss Wheldale[1] has published during the last four years several papers on anthocyanin and flower pigmentation which have attracted wide attention. Since the work on the chromogens and anthocyanin of Dioscorea relates directly to the subject upon which she has been working, it will not be amiss to point out its bearing upon her views. In an article in the Journal of Genetics (1911) she has summarized her theory of anthocyanin formation in a series of propositions of which the following now concern us.

(1) The soluble pigments of flowering plants, collectively termed anthocyanin, are oxidation products of colorless chromogens of an aromatic nature which are present in the living tissues in combination with sugar as glucosids.

(2) The process of formation of the glucosid from chromogen and sugar is of the nature of a reversible enzyme action:

$$\text{Chromogen} + \text{sugar} \rightleftarrows \text{glucosid} + \text{water.}$$

(3) The chromogen can be oxidized to anthocyanin only after liberation from the glucosid, and the process of oxidation is carried out by one or more oxidizing enzymes:

$$\text{Chromogen} + \text{oxygen} = \text{anthocyanin.}$$

(4) From Nos. 2 and 3 we may deduce that the amount of free chromogen, and hence the quantity of pigment formed at any time in a tissue, is inversely proportional to the concentration of sugar and directly proportional to the concentration of glucosid in that tissue.

Inasmuch as the one water-soluble ammonia-greening anthocyanin of which we have reliable chemical information is itself a glucosid, it is obvious that Miss Wheldale's conclusion that water-soluble anthocyanins are formed by the oxidation of nonglucosidal chromogens is of dubious value as a general proposition. It becomes still more dubious when we consider that the only nonglucosidal ammonia-greening anthocyanin known (that of Althaea) is insoluble in water and that the only nonglucosidal chromogen which on oxidation gives a compound resembling an ammonia-greening anthocyanin (the rhodochlorogen of Dioscorea) yields oxidation products which do not dissolve in water. Furthermore, the regeneration of water-soluble anthocyanins from their yellow reduction products takes

[1] Wheldale, M. The Colours and Pigments of Flowers, with Special Reference to Genetics. Proceedings, Royal Society of London, ser. B, 1909, pp. 44–60.

——— On the Nature of Anthocyanin. Proceedings, Cambridge Philosophical Society, vol. 15, 1909, p. 137.

——— Plant Oxydases and the Chemical Interrelationships of Colour-Varieties. Progressus Rei Botanicae, vol. 3, 1910, pp. 457–473.

——— On the Formation of Anthocyanin. Journal of Genetics, vol. 1, 1911, pp. 133–158. Review by Molisch, Zeitschrift für Botanik, vol. 4, pp. 135–136. Review by Hagem, ibid., pp. 136–138. 1912.

place spontaneously in the presence of oxygen and does not require the agency of oxidizing enzymes. This is true not only of the water-soluble anthocyanins of Althaea and Dioscorea, but also of the similar ammonia-greening anthocyanin of red cabbage, the ordinary reactions of which are familiar to many chemists on account of its occasional use as an indicator. Of course, it is not impossible that the chromogen which is oxidized to anthocyanin in the plant cell is different from the artificially obtained reduction product of the same anthocyanin. The fact may be emphasized at this point, however, that rhodochlorogen gave an identical product with plant oxidases and with mineral oxidizing agents.

It is clear that too little is known regarding the physiological significance and chemistry of anthocyanins, which are chemically very diverse compounds, to warrant anyone in speculating on the mode of their formation in the plant cell. Miss Wheldale's speculations are too obviously without adequate experimental basis.

CONCLUSION.

A chromogen isolated from the aerial tubers of the Hawaiian bitter yam has been named "rhodochlorogen." It has possible chemical and physiological relationships with the ammonia-greening anthocyanin of that plant in that it forms green salts and oxidizes to a red compound which would itself pass from an anthocyanin if it were not insoluble in water. Many investigators have supposed that the anthocyanins were closely allied to the tannins. In this connection it is especially interesting that during the process of purification rhodochlorogen was separated from tannin by the use of lead acetate and ferrous sulphate. Lead acetate did not precipitate rhodochlorogen, but did precipitate its red oxidation derivative.

There is reason to believe that rhodochlorogen of Dioscorea contains the same chromophoric nucleus as the anthocyanin of the same plant; consequently, the reactions of rhodochlorogen afford evidence against Miss Wheldale's hypothesis that the so-called ammonia-greening anthocyanins are not autonomous compounds but merely mixtures of yellow flavones with ammonia-bluing anthocyanins, mixtures which would give green as a resultant color with alkalis.[1] Grafe[2] has already concluded that the ammonia-greening reaction

[1] "The pigment (anthocyanin) closely resembles the flavone in its general behavior toward chemical reagents except that the range of color reactions is different. The oxidized product gives a blue color with alkalis, whereas the chromogen, as previously stated, gives a yellow color. The result, therefore, of alkali action on the combination is green, due to the mixture of yellow and blue."—Wheldale, Progressus Rei Botanicae, vol. 3, 1910, p. 468.

[2] Die grüne Reaktion des Anthokyans, welche mit einem Überschuss an Alkali eintritt, dürfte entgegen der älteren Anschauung nicht oder wenigstens nicht in allen Anthokyanen auf die Anwesenheit von Gerbstoffen zurückzführen, sondern eine dem Anthokyan selbst eigentümliche Eigenschaft sein."—Grafe, Viktor, op. cit., I, vol. 115, pt. 1, 1906, p. 993.

of certain anthocyanins is not due to the admixture of tannins. This conclusion is confirmed by the work on the rhodochlorogen of Dioscorea.

The only yellow compounds known which oxidize to anthocyanin do so spontaneously, and the reaction does not involve the agency of oxidases. Although rhodochlorogen is easily oxidized by plant oxidases to its red oxidation derivative, there is as yet no evidence whatever that this process takes place in the plant cell. It seems more likely that rhodochlorogen is transformed first into a glucosid and that this oxidizes to anthocyanin than that the chromogen is directly oxidized in the plant cell. As yet, however, it is not known that rhodochlorogen has any further part in metabolism after it is laid down in the tuber. It may be nothing more than an end product.

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U. S. DEPARTMENT OF AGRICULTURE.

BUREAU OF PLANT INDUSTRY—BULLETIN NO. 265.

B. T. GALLOWAY, *Chief of Bureau.*

SOME FACTORS INFLUENCING THE EFFICIENCY OF BORDEAUX MIXTURE.

BY

LON A. HAWKINS,

Scientific Assistant, Fruit-Disease Investigations.

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LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry,
Office of the Chief,
Washington, D. C., September 10, 1912.

Sir: I have the honor to transmit herewith and to recommend for publication as Bulletin No. 265 of the series of this bureau a manuscript by Mr. Lon A. Hawkins, Scientific Assistant in Plant Pathology, entitled "Some Factors Influencing the Efficiency of Bordeaux Mixture."

This paper, submitted by Mr. M. B. Waite, Pathologist in Charge of Fruit-Disease Investigations, presents the results of investigations of various methods of preparation of Bordeaux mixture and of the effect of adding to the mixture different compounds designed to increase its adhesiveness.

The results obtained are of importance where this fungicide is used and should be of special interest to cranberry and grape growers.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

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SOME FACTORS INFLUENCING THE EFFICIENCY OF BORDEAUX MIXTURE.

INTRODUCTION.

The efficiency of Bordeaux mixture in preventing certain diseases which attack the young aerial portions of plants is dependent on several factors. Not the least among these is uniformity in the distribution of the copper compound throughout the liquid medium when the mixture is applied. That this is of importance is very apparent, for if the copper compound has settled out, even to a limited degree, part of the plant will receive a heavy coating of the fungicide, while other portions may receive none and thus be liable to infection by the fungus. Another important factor is adhesiveness, as it is obviously necessary for the fungicide to adhere to the susceptible portions of the plant if they are to be protected from fungous disease. With these requirements for efficiency in mind the questions naturally arise, By what methods can the most uniform distribution of the copper compound in the medium be obtained, and how can the adhesiveness of the mixture to the susceptible parts of the plants be increased? The present investigation deals with these two questions.

COMPOSITION OF BORDEAUX MIXTURE.

Bordeaux mixture is made up of copper sulphate and calcium hydroxid, and the rate of subsidence of the colloidal suspension of the precipitate which results from the interaction of these substances is partly dependent on the manner in which the two components are brought together. It is not necessary here to go into a detailed discussion of the chemical reactions that take place when copper sulphate and calcium hydroxid are brought together. They have been studied by Swingle,[1] Chester,[2] Sostegni,[3] Pickering,[4] and others, with various

[1] Swingle, Walter T. Bordeaux Mixture: Its Chemistry, Physical Properties, and Toxic Effects on Fungi and Algæ. Bulletin 9, Division of Vegetable Physiology and Pathology, U. S. Dept. of Agriculture. 1896.

[2] Chester, F. D. Copper Salts as Fungicides. Journal of Mycology, vol. 6, 1890, pp. 21–24.

[3] Sostegni, Livio. Sulla Composizione Chimica della Cosi detta Poltiglia Bordolese. Le Stazioni Sperimentali Agrarie Italiane, vol. 19, 1890, pp. 129–141.

[4] Pickering, Spencer U. Eleventh Report of the Woburn Experimental Fruit Farm. 1910.

conclusions as to the nature of the compounds formed. It is generally agreed that the insoluble copper compound of Bordeaux mixture, whether copper hydroxid, basic sulphate of copper, or both these compounds, is in colloidal suspension in a saturated or nearly saturated solution of calcium sulphate and calcium hydroxid.

PREPARATION OF BORDEAUX MIXTURE.

Different authors have recommended various methods for the preparation of Bordeaux mixture, with a view of obtaining the most economical and effective mixture. Millardet,[1] in describing the making of Bordeaux mixture for the first time, says:

> Dans 100 litres d'eau quelconque (de puits, de pluie, ou de rivière) on fait dissoudre 8 kilos de sulfate de cuivre du commerce. D'un autre côté, on fait, avec 30 litres d'eau et 15 kilos de chaux grasse, en pierres, un lait de chaux qu'on mélange à la solution de sulfate de cuivre.

This method of mixing, with the same formula, was adopted in America, having been first published by Scribner[2-3] in 1886. Two years later Scribner[4] recommended 4 pounds of copper sulphate and a like quantity of lime in 22 gallons of mixture, while Galloway[5] the same year recommended a formula of 6 pounds of copper sulphate and 4 pounds of stone lime to 22 gallons of water. Waite[6] in 1893 obtained good results in spraying for pear leaf-blight by using 6 pounds of copper sulphate to 50 gallons of water with just sufficient lime to react with the copper sulphate. In the same article this writer recommends the use of a stock solution of copper sulphate and a stock mixture of lime in the preparation of the fungicide. With these formulas, the method of preparation was to pour the calcium hydroxid into the copper-sulphate solution. In 1896 Galloway[7] recommended the use of two tubs, in which the copper sulphate and lime were separately diluted, each to half the volume of the Bordeaux mixture required. From these tubs the two solutions were poured simultaneously into a barrel. In the same article he recommended

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[1] Millardet, A. Journal d'Agriculture et d'Horticulture de la Gironda, May 1, 1885.

NOTE.—This publication was not at hand and the quotation given was taken from the same writer's paper, entitled "Sur l'histoire du traitement du mildiou par le sulfate de Cuivre," Journal d'Agriculture Pratique, vol. 49, pt. 2, 1885, pp. 801–805, in which Millardet quotes directly from his former paper in describing the method of preparing Bordeaux mixture.

[2] Scribner, F. Lamson. Report on the Mycological Section, in the Report of the Commissioner of Agriculture for 1886, p. 100.

[3] ——— Report on the Fungous Diseases of the Grapevine. Bulletin 2, Section of Plant Pathology, Botanical Division, U. S. Dept. of Agriculture, 1886, p. 16.

[4] ——— Fungicides or Remedies for Plant Diseases. Circular 5, Section of Vegetable Pathology, Botanical Division, U. S. Dept. of Agriculture, 1888.

[5] Galloway, B. T. Treatment of Black Rot of the Grape. Circular 6, Section of Vegetable Pathology, Botanical Division, U. S. Dept. of Agriculture, 1888, p. 2.

[6] Waite, M. B. Treatment of Pear Leaf-Blight in the Orchard. Journal of Mycology, 7, 1894, pp. 333–338.

[7] Galloway, B. T. Spraying for Fruit Diseases. Farmers' Bulletin 38, U. S. Dept. of Agriculture, 1896, p. 6.

6 pounds of copper sulphate and 4 pounds of lime to 50 gallons of mixture. This last method of mixing Bordeaux has been recommended by the investigators in the Department of Agriculture and most of the agricultural experiment-station workers in the United States since that time. Some of the experiment stations, however, recommend the pouring of one component into the other, as shown by the publication of Woods and Hanson,[1] Green, Selby, and Gossard,[2] and Smith and Smith.[3] Kelhofer[4] in an account of his investigations on the preparation of Bordeaux mixture says:

> Die grössten Niederschläge erzielen wir demnach bei langsamem (portionenweisem) Zusatz der Kupfervitriollösung zur Kalkmilch. Annähernd ebenso günstige Resultate werden erhalten, wenn man die Kalkmilch rasch zur Kupfervitriollösung giesst.

The copper sulphate and lime of Kelhofer's preparations were both diluted to the same volume. Two series of experiments were carried out, in one of which this volume was one-half that of the fungicide required and in the other one-fourth. Kelhofer[5–6] also added with good results small quantities of cane sugar to retard the rate of subsidence of the suspension. Kulisch[7] repeated some of Kelhofer's experiments with like results. Pickering,[8] in making common Bordeaux mixture, recommends the use of calcium hydroxid as dilute as possible to make the required quantity and the copper sulphate in concentrated solution. The copper sulphate is poured into the calcium hydroxid with very little stirring. An examination of the literature of this subject shows that the methods recommended for the preparation of a colloidal suspension of the copper compound which settles out slowly are rather varied. The problem of making a suspension which subsides slowly then resolves itself into testing the methods of mixing recommended by the different investigators to determine their comparative efficiency. Accordingly, to determine the effect on the rate of subsidence of the suspensions of some of these

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[1] Woods, Charles D., and Hansen, H. H. Paris Green Bordeaux Mixture. Bulletin 154, Maine Agricultural Experiment Station, April, 1908.

[2] Green, W. J., Selby, A. D., and Gossard, H. A. Spray Calendar. Bulletin 232, Ohio Agricultural Experiment Station, 1911.

[3] Smith, R E., and Smith, Elizabeth H. Bulletin 218, Agricultural Experiment Station of the University of California, 1911, p. 1185.

[4] Kelhofer, W. Versuch über die Herstellung der Bordeauxbrühe. Jahresbericht der Deutsch-Schweizerischen Versuchstation und Schule für Obst-Wein- und Gartenbau, vol. 8, 1897–98, p. 65.

[5] Kelhofer, W. Versuche über die Beeinflussung der Haltbarkeit der Bordeaubrühe durch Zusätze. Jahresbericht, der Deutsch-Schweizerischen Versuchstation und Schule für Obst-Wein- und Gartenbau, vol. 9, 1898–99, pp. 87–92.

[6] ——— Ueber einige Gesichtspunkte bei der Herstellung der Bordeauxbrühe. Zeitschrift für Pflanzenkrankheiten, vol. 18. Internationaler Phytopathologischer Dienst, vol. 1, no. 3, 1908, pp. 65–73.

[7] Kulisch, P. Die Darstellung haltbarer Kupferbrühen zur Bekämpfung der Peronospora. Zeitschrift für Pflanzenkrankheiten, vol. 21, 1911, pp. 382–384.

[8] Pickering, Spencer U. Op. cit., p. 56.

methods of preparing Bordeaux mixture the investigations described in the first part of this paper were planned and carried out.

EXPERIMENTS IN PREPARATION.

For the greater part of the investigation the copper sulphate and lime used were what is commonly known as chemically pure. Distilled water was used in these preparations. Later, a number of the series were repeated in order to approach commercial conditions as closely as possible, using a good grade of common stone lime, commercial copper sulphate, and tap water. The mixtures were prepared in glass-stoppered cylinders of 1-liter capacity graduated to divisions of 10 cubic centimeters. To prepare Bordeaux mixture by allowing the two diluted components to flow simultaneously into the container, two burettes of 1,000 cubic centimeters capacity were placed side by side, with the outlets connected by rubber tubes provided with pinch cocks to a single Y tube, the lower arm of which was so arranged as to project into the neck of the glass cylinder. The proper quantity of calcium hydroxid, made by slacking 3.75 grams of calcium oxid, was placed in one burette and diluted to half a liter, while in the other was placed a solution of the same volume, containing 5 grams of copper sulphate. A current of air was forced in at the bottom of the burette containing the calcium hydroxid to keep the precipitate from settling out. If the two pinchcocks were opened at the same time the milk of lime and the copper-sulphate solution flowed simultaneously into the graduated receiver, and by properly manipulating the pinchcock either component could be added to the other. This apparatus was used in preparing all mixtures in which both the lime and the copper sulphate were diluted to the same volume before bringing them together. The comparative volumes of the precipitates were determined by measuring the fall of the precipitate surface in the different mixtures during a given time interval, the graduations on the cylinders serving as convenient indexes for this purpose. The formula used in the preparation was usually that for 4–3–50 Bordeaux mixture (4 pounds of copper sulphate and 3 pounds of lime to 50 gallons of water), which is a standard formula for use on grapes and cranberries.

Effect of Different Methods of Mixing on the Rate of Subsidence of the Suspension.

The methods of mixing were as follows:

(1) Calcium hydroxid and copper sulphate were made up to 500 cubic centimeters each and allowed to flow simultaneously into the receiver.

(2) Calcium hydroxid and copper sulphate were made up to 500 cubic centimeters each and calcium hydroxid allowed to flow into the copper sulphate.

(3) Calcium hydroxid and copper sulphate were made up to 500 cubic centimeters each and copper sulphate allowed to flow into the calcium hydroxid.
(4) Calcium hydroxid was made up to 950 cubic centimeters and shaken; copper sulphate was made up to 50 cubic centimeters and poured into the calcium hydroxid.
(5) Copper sulphate was made up to 950 cubic centimeters; calcium hydroxid was made up to 50 cubic centimeters and added to the copper sulphate.
(6) Calcium hydroxid and copper sulphate were made up to 50 cubic centimeters each; the calcium hydroxid was added to the copper-sulphate solution and the mixture diluted to 1,000 cubic centimeters.

In addition to these the rate of subsidence of Woburn Bordeaux mixture, recommended by Pickering, which is made from clear lime water and copper-sulphate solution, was tested.

The results obtained in these tests were varied. Throughout the investigation, however, No. 1, the method of mixing described first by Galloway, was found to result in a suspension which subsided as slowly as any of the others, except the Woburn Bordeaux mixture. The results with No. 2 and No. 3 in most cases closely approached those with No. 1. With Nos. 4 and 5, in which one of the components in a concentrated form was poured into the other made up to nearly the volume of fungicide desired, about as good results were obtained as with No. 1, provided the mixtures were properly agitated.

When the two components were brought together in concentrated form, as in No. 6, the suspension settled out much more rapidly than with any of the other preparations. This method of making Bordeaux mixture seems therefore to be entirely unsatisfactory, as has been almost universally held by previous writers.

Effect of Varying Amounts of Agitation on the Subsidence of the Suspension.

Experiments were carried out to test the effects of varying amounts of agitation on the rapidity of subsidence of the suspension when prepared by adding one of the components in concentrated form to the other made up to nearly the quantity of solution required, as in Nos. 4 and 5. In agitating, the graduates were stoppered and shaken vigorously with an up-and-down motion, the amount of agitation being measured by the number of complete excursions made by the container during the shaking. As there was always a large air space above the liter of mixture in the graduated cylinder, a very thorough agitation could be easily obtained by this method.

EXPERIMENTS WITH CONCENTRATED LIME POURED INTO DILUTE COPPER SULPHATE.

In experiments with the concentrated lime poured into the dilute copper-sulphate solution, five mixtures were prepared: *A*, The standard solution, for purposes of comparison, was mixed by allowing the copper sulphate and calcium hydroxid, each diluted to 500 cubic centimeters, to flow simultaneously into the 1,000 cubic centimeter graduate. The others, *B* to *E*, were prepared by pouring the lime, made up to 50 cubic centimeters, into the copper-sulphate solution diluted to 950 cubic centimeters. *A* and *B* were each shaken 5 times; *C*, 15 times; *D*, 25 times; and *E*, 35 times. The preparations were then allowed to stand one hour, when the volume of the precipitate was read on the scale of the graduate. It was found that the pre-

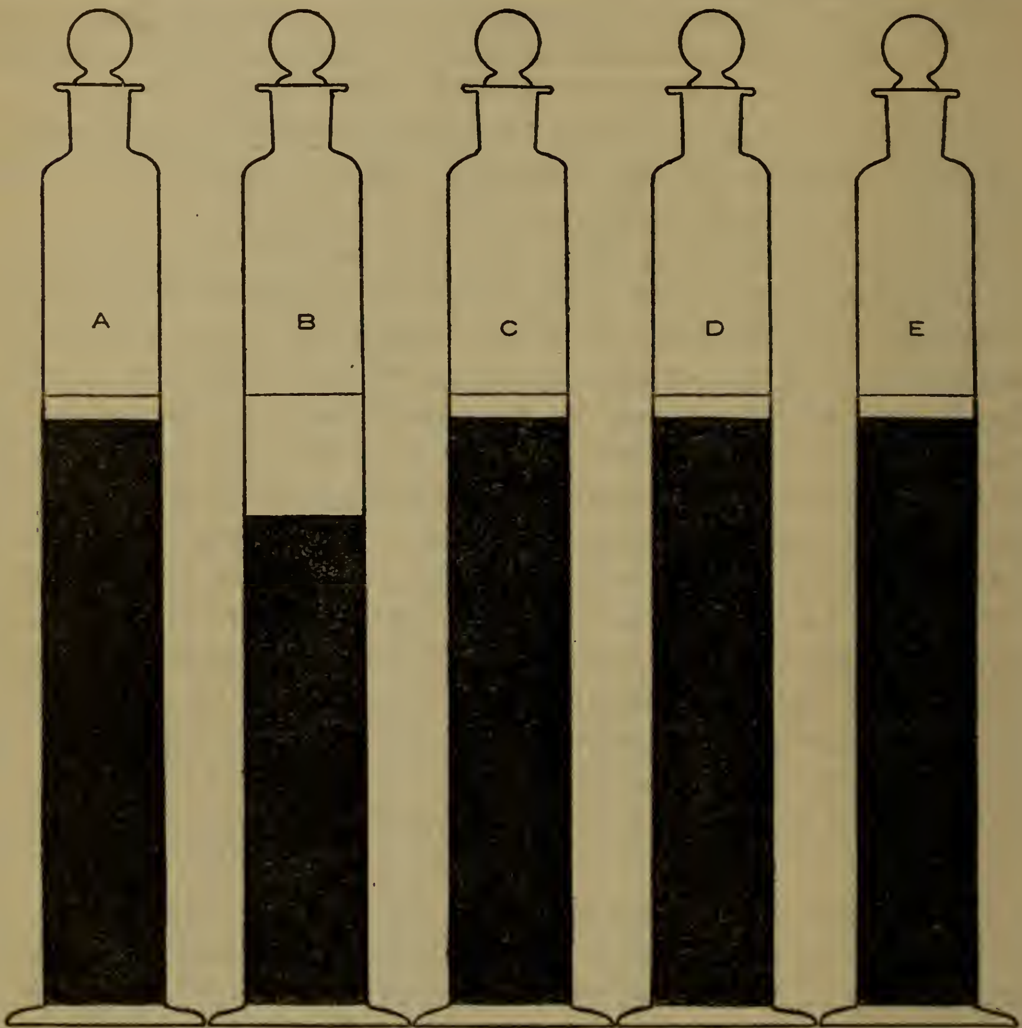

FIG. 1.—Diagram showing the effect of varying the amount of agitation of Bordeaux mixture when the concentrated lime is added to the diluted copper-sulphate solution. The shaded portion represents the precipitate after 1 hour. In *A*, the standard solution, the two components diluted to half the quantity were poured simultaneously into the container and shaken 5 times. In *B*, *C*, *D*, and *E*, the concentrated lime was poured into diluted copper-sulphate solution and shaken 5, 15, 25, and 35 times, respectively.

cipitate in *B*, shaken the same number of times as the control, settled much more rapidly than *A*, while in *C* to *E*, shaken from 15 to 35 times, the suspension subsided with about the same rapidity as in *A*. The experiments were repeated a number of times, with similar results. The average volume from three separate experiments is shown graphically in figure 1. A description of the treatment given the preparations and the results obtained are given in Table I.

TABLE I.—*Results of tests showing the effect of agitation of Bordeaux mixture when the concentrated lime is poured into the dilute copper-sulphate solution.*

Mixture used and method of preparation.	Times shaken.	Average volume of precipitate after 1 hour, as measured on scale of the graduate.
		Cubic centimeters.
A. Standard, both $Ca(OH)_2$ and $CuSO_4$ diluted to 500 cubic centimeters and allowed to flow simultaneously into receiver	5	970
B. $Ca(OH)_2$ diluted to 50 cubic centimeters and poured into $CuSO_4$ solution diluted to 950 cubic centimeters	5	805
C. Preparation same as *B*	15	970
D. Preparation same as *B*	25	970
E. Preparation same as *B*	35	970

EXPERIMENTS WITH CONCENTRATED COPPER SULPHATE POURED INTO DILUTE LIME.

In the series of experiments in which the copper-sulphate solution was made up to 50 cubic centimeters and then poured into the lime made up to 950 cubic centimeters the results were much the same as when the concentrated calcium hydroxid was poured into the dilute copper sulphate. The experiments are comparable in every way, since in both series the same quantities were used, the gradations in amount of agitation were the same, and the observations were made after the same length of time. The individual experiments were repeated a number of times, with practically the same results. An average of three, in which the preparations were made up with commercial copper sulphate and common stone lime, gave the results as shown in Table II.

TABLE II.—*Results of tests showing the effect of the agitation of Bordeaux mixture when the concentrated copper-sulphate solution is poured into the dilute lime.*

Mixture used and method of preparation.	Times shaken.	Average volume of precipitate after 1 hour, as measured on scale of the graduate.[1]
		Cubic centimeters.
A. Standard, both $Ca(OH)_2$ and $CuSO_4$ diluted to 500 cubic centimeters and allowed to flow simultaneously into receiver	5	972½
B. $CuSO_4$ diluted to 50 cubic centimeters and poured into the $Ca(OH)_2$ diluted to 950 cubic centimeters	5	703
C. Preparation same as *B*	15	836
D. Preparation same as *B*	25	916
E. Preparation same as *B*	35	900

[1] A graphic representation of these averages is shown in figure 2.

DISCUSSION OF EFFECTS OF AGITATION.

The foregoing experiments seem to show that the rate of subsidence of the suspension in Bordeaux mixture is not entirely dependent upon the manner in which the two components are brought together, but is also dependent to a certain extent on the amount of agitation the mixture receives. For example, if the mixtures, in which one of the components in high concentration is poured into the other diluted to nearly the required volume, are sufficiently agitated, the resulting suspension will subside about as slowly as the suspension in a mixture prepared by the standard method recommended by Galloway. (See figs. 1, *C*, *D*, and *E*, and 2, *D* and *E*.) On the other hand, if mixtures

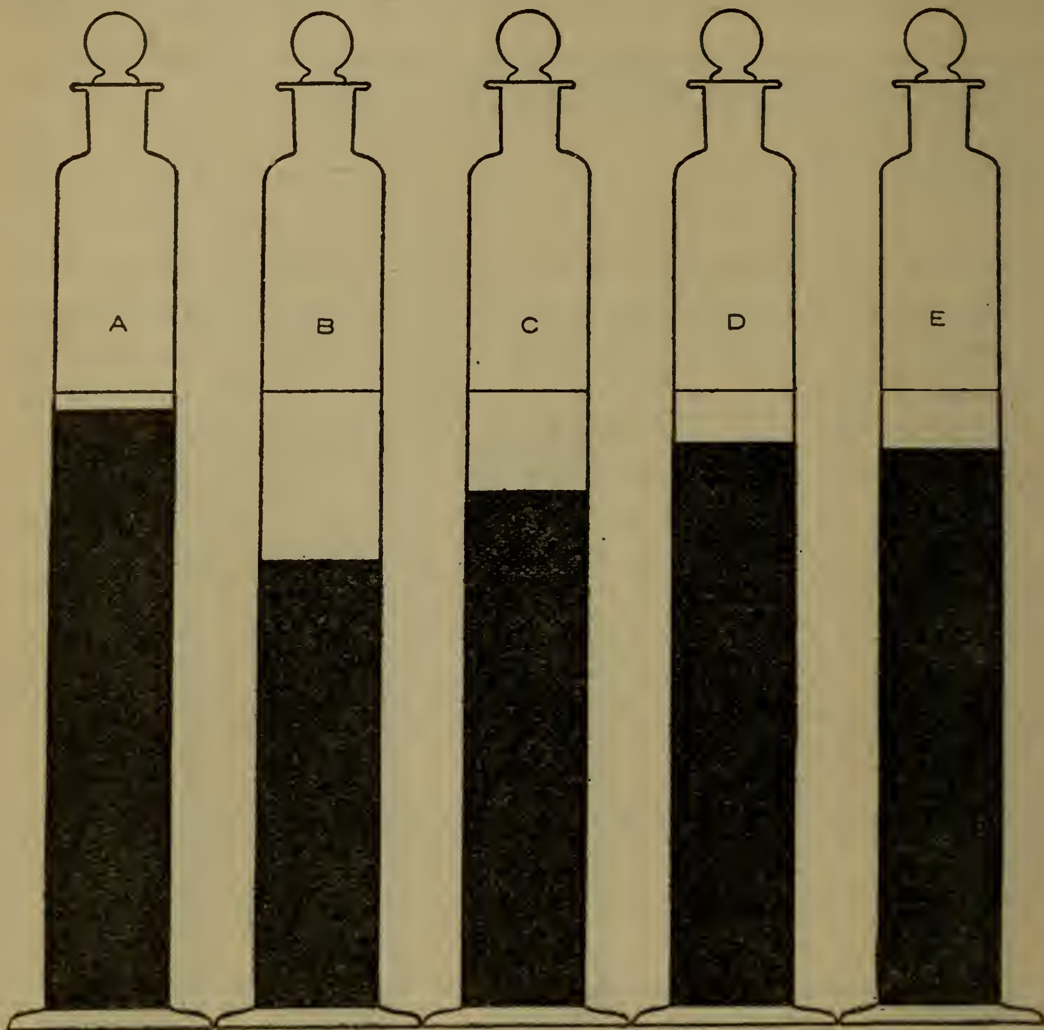

FIG. 2.—Diagram showing the effect of varying the amount of agitation of Bordeaux mixture when the concentrated copper-sulphate solution is added to the diluted lime. The shaded portion represents the precipitate after 1 hour. In *A*, the standard solution, the two components diluted to half the quantity required were poured simultaneously into the container and shaken 5 times; in *B*, *C*, *D*, and *E*, the concentrated copper-sulphate solution was poured into dilute lime and shaken 5, 15, 25, and 35 times, respectively.

prepared in this way receive only a small amount of agitation, the suspension subsides much more rapidly. (See figs. 1 *B*, and 2, *B*.) A probable explanation of this fact lies apparently in the structure of the precipitate formed when the two components of Bordeaux mixture are brought together. On this subject the work of Swingle[1] seems to be generally accepted.

[1] Swingle, Walter T. Bordeaux Mixture: Its Chemistry, Physical Properties, and Toxic Effects on Fungi and Algæ. Bulletin 9, Division of Vegetable Physiology and Pathology, U. S. Dept. of Agriculture. 1896, p. 13.

This writer has shown that when calcium hydroxid and a solution of copper sulphate are brought together small Traube cells are formed, composed of a precipitation membrane of the insoluble copper compound surrounding either a drop of calcium-hydroxid solution or a particle of the undissolved calcium hydroxid. It may be suggested that in the latter case the particles of calcium hydroxid remaining inside the precipitation membrane might weigh them down and make them settle out more rapidly than those in which no solid was present. If, however, the mixture were sharply agitated the precipitation membranes might be broken and the undissolved particles of lime could settle to the bottom without taking with them the precipitation membranes of the copper compound.

This explanation receives further support from the fact that when Bordeaux mixture is made by allowing the two dilute solutions to flow simultaneously into the container, the lime being well agitated meanwhile, the resulting suspension subsides much less rapidly than a mixture in which one of the components in high concentration is poured into the other one diluted, provided the mixtures receive only a small amount of agitation. In the case of the two components of the mixture, each diluted to half the required volume, the particles of calcium hydroxid would be suspended in a relatively large volume of liquid, and the precipitation membranes in the Bordeaux mixture made from this milk of lime should be smaller and contain less solid matter to weigh them down than if the concentrated calcium hydroxid were added in a pasty mass to the diluted copper-sulphate solution. On the other hand, the formation of thick, heavy precipitation membranes would be much less probable when the two diluted solutions were brought together than if the concentrated copper-sulphate solution were added to the diluted calcium hydroxid.

Pickering's Woburn Bordeaux mixture, as prepared in this investigation, also furnishes further evidence in support of this explanation. This mixture was prepared by adding a concentrated solution of copper sulphate to a saturated solution of calcium hydroxid which had been filtered. No undissolved calcium hydroxid and at most only a small quantity of undissolved calcium sulphate were in the mixture. Consequently, there being nothing to weigh down the precipitate, it should remain in suspension much better than the Bordeaux mixture made in the usual way. The result was as expected. The mixture was made up and stood on the table about two months, being agitated from time to time. The precipitate settled out very slowly after each agitation and never reached the stage at which it was impossible to bring it into relatively uniform suspension throughout the whole mixture. From these facts it seems fair to conclude that the slow subsidence of the suspension in the experiments in which one of the components in high concentration was added to the other which had been diluted to nearly the required volume was due to the breaking up of the precipitation membranes and the elimination, partially at least, of the solid calcium hydroxid and calcium sulphate from the suspended precipitate. Lutman,[1] in a recently published account of his investigations of Bordeaux mixture, reaches much the same conclusions in regard to the settling out of the precipitation membranes.

ADHERENCE OF BORDEAUX MIXTURE WITH AND WITHOUT ADDED ADHESIVES.

HISTORICAL REVIEW OF WORK ON ADHERENCE.

The fact that it is necessary for the fungicide to adhere to the surface of the plant in order to protect it from disease has led to many investigations of the comparative adhesiveness of different copper fungicides, both with and without added adhesives. The effects of added adhesives have been tested on potatoes by Girard;[2] on wheat and other cereals by Galloway;[3] on grapes by Perraud,[4] Guillon and Gouirand,[5] Cazeneuve,[6] Gastine,[7] Chuard and Porchet,[8] and others. General studies on the adhesiveness of fungicides have been made by Kelhofer,[9-10] and various other writers. Some of the compounds used as adhesives are soap, glue, cane sugar, dried blood, gelatin, and colophene. The compounds used in the present investigation to increase adhesiveness have all been tried by other investigators, with somewhat varied results. Sirrine[11] in his work on asparagus rust recommends the use of a rosin Bordeaux mixture,

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[1] Lutman, B. F. The Covering Power of the Precipitation Membranes of Bordeaux Mixture. Phytopathology, vol. 2, February, 1912, pp. 32–41.

[2] Girard, A. Recherches sur l'adhérence aux feuilles des plantes, et notamment aux feuilles de la pomme de terre, des composés cuivriques, destinés à combattre leur maladies. Comptes Rendus de l'Académe des Sciences, vol. 114, 1892, pp. 234–236.

[3] Galloway, B. T. Experiments with Treatment of Rusts of Wheat and Other Cereals. Journal of Mycology, vol. 7, 1893, pp. 195–226.

[4] Perraud, Joseph. Moynes d'augmenter l'adhérence des bouillies cupriques sur les raisins. Journal d'Agriculture Pratique, vol. 62, pt. 2, 1898, pp. 814–816.

[5] Guillon, G. M., and Gouirand, G. Sur l'adhérence de rouillies cupriques utilisées pour combattre les maladies cryptogamiques de la vigne. Comptes Rendus de l'Académie des Sciences, vol. 127, 1898, pp. 423–424.

[6] Cazeneuve, Paul. La bouillie bordelaise albumineuse. Revue de Viticulture, vol. 9, 1898, pp. 279–280.

[7] Gastine, G. Les préparations cupriques et leur adhérence. Bulletin Mensuel de l'Office de Renseignements Agricoles, vol. 5, 1906, pp. 595–603.

8 Chuard, E., and Porchet, F. L'adhérence des bouillies cupriques. Revue de Viticulture, vol. 24, 1905, pp. 33–37.

[9] Kelhofer, W. Ueber einige Gesichtspunkte bei der Herstellung der Bordeauxbrühe. Zeitschrift für Pflanzenkrankheiten, vol. 18. Internationaler Phytopathologischer Dienst, vol. 1, no. 3, 1908, pp. 65–73.

[10] ——— Ueber die Ausführung und die Ergebnisse von Haftfestigkeitsversuchen kupferhaltiger Bekämpfungsmittel gegen die Peronospora. Zeitschrift für Pflanzenkrankheiten, vol. 17, 1907, pp. 1–12.

[11] Sirrine, T. A. Spraying for Asparagus Rust. Bulletin 188, New York Agricultural Experiment Station, 1900.

prepared by adding to the Bordeaux mixture a soap made from rosin, fishoil, and potash. Chester[1] used a rosin Bordeaux mixture in spraying grapes and asparagus, and in referring to the grape-spraying experiments says "rosin soap added to Bordeaux mixture offered no advantage over the plain mixture." He, however, considered it of value in spraying asparagus. Shear[2] used rosin-fishoil soap with good effect in spraying cranberries. Wilson and Reddick[3] used a soap in spraying grapes in 1908. In a report of their work they say, "The different 'stickers,' such as rosin sal soda, or fishoil soap, are of no practical value." Rosin-fishoil soap was used in grape-spraying experiments carried on by the United States Department of Agriculture during the years 1907 and 1908[4] without definite results. Experiments were continued in 1909,[5] however, and considerable benefit was ascribed to its use. Iron sulphate has been recommended by Selby.[6] Glue was first used by Perraud,[7] and has since been employed from time to time by cranberry growers. From the investigations here mentioned it seemed probable that the adhesiveness of Bordeaux mixture on parts of certain plants might be increased by the addition of certain compounds, though from the varied results obtained by the different investigators the relative values of these adhesives were not definitely known. An investigation was accordingly undertaken to determine, if possible, the adhesiveness of Bordeaux mixture prepared according to several formulas, together with the value of certain adhesive compounds. It was also considered worth while to try to find some laboratory method for measuring the comparative adhesiveness of these mixtures.

EXPERIMENTS ON ADHESIVENESS.

Experiments with adhesives were carried on in connection with spraying experiments for the control of the black-rot of the grape near Vineland, N. J., in the vineyard of the Vineland Grape Juice Co., during the season of 1910, and at Paw Paw, Mich., in the vineyard of Mr. Roy L. Tuttle, in 1911.

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[1] Chester, F. D. Fungous Diseases in Delaware, Part II. Treatment of Certain Plant Diseases. Bulletin 63, Delaware College Agricultural Experiment Station, February, 1904.

[2] Shear, C. L. Cranberry Spraying Experiments in 1905. Bulletin 100, pt. 1, Bureau of Plant Industry, U. S. Dept. of Agriculture. 1906.

[3] Wilson, C. S., and Reddick, D. The Black-Rot of the Grape and Its Control. Bulletin 266, Cornell University Agricultural Experiment Station. 1909.

[4] Shear, C. L., Miles, George F., and Hawkins, Lon A. The Control of Black-rot of Grape. Bulletin 155, Bureau of Plant Industry, U. S. Dept. of Agriculture. 1909.

[5] Hawkins, Lon A. Grape-Spraying Experiments in Michigan in 1909. Circular 65. Bureau of Plant Industry, U. S. Dept. of Agriculture. 1910.

[6] Selby, A. D. Modifications of Bordeaux Mixture. Sixty-second Annual Report of Ohio State Board of Agriculture, 1907, pp. 896–898.

[7] Perraud, Joseph. Op. cit.

Experiments on Adhesiveness to Grape Leaves.

The mixtures used in the experiments on grape leaves were as follows:

- 4–3–50 Bordeaux mixture.
- 4–2–50 Bordeaux mixture.
- 4–3–50 Bordeaux mixture with 2 pounds of ferrous sulphate to 50 gallons.
- 4–3–50 Bordeaux mixture with 2 pounds of rosin-fishoil soap to 50 gallons.

All mixtures were made up by diluting the copper sulphate and calcium hydroxid each to half the volume of fungicide required, and allowing them to flow simultaneously into the tank of the sprayer. The adhesives were added as follows: The fresh solution of ferrous sulphate was added to the diluted solution of copper sulphate, as recommended by Selby; the required amount of soap was dissolved in a small quantity of water and poured into the Bordeaux mixture after it had been mixed.

A portion of a 5-year-old Concord vineyard, in which the vines were apparently uniform, was divided into four plats, each plat being sprayed with one of the mixtures mentioned. The fungicides were applied with a gasoline-power sprayer. Trailers, lines of hose with short extension rods attached so that the nozzles could be manipulated by hand, were used in all sprayings. Care was taken in the application of the fungicide to see that all the sprayed plats received as nearly as possible the same quantity of the fungicide at any one application and that it was applied under the same pressure. The plats were sprayed four times, and samples of the leaves were collected after the first application and both before and after each succeeding application. Other collections were made at intervals, as shown in Table III.

The method of collecting these samples was as follows: From each plat 40 or more leaves which appeared to represent the average condition of the foliage in the plat in regard to quantity and distribution of the fungicide upon the leaf surfaces were collected at the same time. These collections were immediately taken to the laboratory, the petioles removed, the leaves with holes in them or other imperfections discarded, and the outlines of 30 to 40 leaves traced on paper. The samples were then stored in large envelopes to dry. The tracings were filed away and the area of the leaves determined later by means of a planimeter or by the weighing method. By using this number of leaves for a sample and with this method of determining the area it was considered that the quantity of copper found on the leaves would be proportional, within a reasonable percentage of error, to the quantity of copper on the leaves in the entire plat.

Later the dried leaves of a sample were removed from the envelope, placed in a casserole, and treated with sulphuric acid to convert the copper on the leaves to sulphate. They were then ashed with a Bunsen burner. The ash was extracted with dilute sulphuric acid, thrown on a filter, and washed free from copper. The copper in the filtrate was determined electrolytically. The area of the leaves as found was multiplied by two to take into account both leaf surfaces, and the amount of metallic copper calculated per square meter of leaf surface. The results of the experiment are shown in Table III.

TABLE III.—*Results of tests showing the adhesiveness of Bordeaux mixture to grape leaves. The number of milligrams of metallic copper to a square meter of leaf surface is shown.*

Plat No.	Mixture.	Date of collection.											
		May 26.[1]	June 14.[2]	June 15.[3]	June 16.[4]	July 13.[5]	July 14.[6]	July 31.[7]	Aug. 1.[8]	Aug. 11.	Aug. 18.	Aug. 26.	Sept. 5.
1	Bordeaux mixture (4-3-50)	103.8	57.7	75.4	......	29.2	60.8	45.6	70.5	67.4	64.3	57.8	16.8
2	Bordeaux mixture (4-2-50)	142.0	......	128.4	64.0	37.1	59.6	37.4	68.0	64.6	63.1	58.4	35.8
3	Bordeaux mixture (4-3-50) + ferrous sulphate	191.1	130.4	121.1	87.7	29.1	68.6	29.3	75.8	64.2	55.3	50.1	40.9
4	Bordeaux mixture (4-3-50) + soap	264.0	149.8	143.6	90.9	64.3	65.7	23.7	81.8	80.4	76.3	62.3	46.6

[1] After first application.
[2] Before second application.
[3] After second application.
[4] Next day after a rain.
[5] Before third application.
[6] After third application.
[7] Before fourth application.
[8] After fourth application.

From Table III it is apparent that there was no considerable variation in the adhesiveness of the different mixtures throughout the season, though usually a little more copper per unit area was found on the samples of leaves from the plat sprayed with Bordeaux mixture to which the rosin-fishoil soap had been added. Immediately after the first application the increased adhesiveness of the rosin-fishoil soap mixture is very marked, but in samples collected after the third application a little more copper per unit area was found on samples from some of the other plats. On the whole, however, the addition of rosin-fishoil soap may be said to be of some slight benefit in increasing the adhesiveness of Bordeaux mixture to grape leaves.

It is apparent that in some cases the quantity of copper on the leaves after spraying was less than that found on the sample collected just before the same application, as before and after the second application in the plat sprayed with the ferrous-sulphate mixture. This is due to the fact that collections before spraying were made only of the leaves which had been sprayed in the previous application, while in collections made after an application leaves of all stages of growth were collected.

Some data on the effect of a rain on the adhesiveness of the spray mixture are given under the dates June 15 and 16 in Table III. The usual collection of leaves was made immediately after the second spraying, and about three hours after this application rain fell, the precipitation being 0.71 inch. Leaves were collected the following day, and from these samples the results under date of June 16 were obtained. The amount of copper on the leaves decreased 35 to 50 per cent by reason of the growth of the leaves during 18 hours and the removal of the copper by the rain. Data for plat 1 are unfortunately lacking.

No evidence is found in these experiments to verify Kelhofer's[1] suggestion that Bordeaux mixture with a large excess of lime adheres to the leaves for a longer time than the more nearly neutral mixture. The percentage of copper removed from the leaves in a given time after spraying with the 4–2–50 or 4–3–50 Bordeaux mixture varies only slightly. The growth of the leaves increasing the leaf area makes it impossible to secure accurate data on the quantity of copper actually removed from the leaves, but as the vines in these plats were the same to all appearances and the same method of sampling was used it is probable that the results are comparable.

Experiments on Adhesiveness to Grape Berries.

It was observed in the course of the preceding experiment that while all the mixtures adhered fairly well to the leaves there was considerable difference in their adhesiveness to the grape berry if the berry was covered with bloom. In spots on the berry from which the bloom had been removed, however, there seemed to be no marked difference in the quantity of fungicide present. When the grapes covered with this bloom were treated with Bordeaux mixture to which no adhesive compounds had been added the mixture seemed to run off immediately or round up into little droplets and hang at the lowest point of the fruit.[2] On the other hand, if the rosin-fishoil soap had been added, the mixture formed a film, completely covering the berry. From these observations it was concluded that the adherence of a fungicide to the leaves was no guaranty of its adherence to the fruit. In order to get accurate data upon the subject it was necessary to carry the experiments further, paying special attention to the fruit.

This investigation on the adhesiveness of the fungicide to the grape berries was carried on at Paw Paw, Mich., in 1911. More

[1] Kelhofer, W. Ueber einige Gesichtspunkte bei der Herstellung der Bordeauxbrühe, Zeitschrift für Pflanzenkrankheiten, vol. 18. Internationaler Phytopatholigischer Dienst, vol. 1, no. 3, 1908, pp. 65–73.

[2] Perraud, op. cit., p. 815, noticed this difference in the adhesiveness of Bordeaux mixture on the leaves and berries in his investigations and remarks, "La faculté d'adhérence des bouillies cupriques est beaucoup plus faible pour les raisins que pour les feuilles de vigne."

adhesive compounds were used in this experiment than in that with the leaves, and the concentration of some of the adhesives was varied. Eight plats of Concord grapes were sprayed four times in the experiment with the following mixtures:

Plat 1. 4–3–50 Bordeaux mixture.
2. 4–3–50 Bordeaux mixture, with 2 pounds of ferrous sulphate.
3. 4–2–50 Bordeaux mixture.
4. 4–2–50 Bordeaux mixture, with 2 pounds of rosin-fishoil soap.
5. 4–2–50 Bordeaux mixture, with 1 pound of rosin-fishoil soap.
6. 4–2–50 Bordeaux mixture, with 2 pounds of fishoil soap.
7. 4–2–50 Bordeaux mixture, with 2 pounds of ground glue.
8. 3–2–50 Bordeaux mixture, with 2 pounds of rosin-fishoil soap.

The soap and the ferrous-sulphate mixtures were prepared as in the experiment already described, and the glue (a dry, ground glue) was dissolved in a small quantity of hot water and added to the Bordeaux mixture. The mixture was prepared as in the preceding experiment.

The mixtures were applied with a steam sprayer, and, as in the work in 1910, care was taken to see that as nearly as possible the same quantity of mixture was applied under the same pressure to each plat. Collections of grape berries were made from each plat immediately after the last spraying, a similar system of sampling being employed as in the experiment with leaves the preceding year. In this case, however, duplicate samples were collected from each plat. Each sample consisted of about 500 grapes, which were cut from the pedicels, the diameter measured to tenths of a millimeter, and the berries stored to dry. The total area of the grape surfaces in each sample was calculated from the diameters of the individual grapes, as the berries were approximately spherical. Later, the copper on the grapes was determined, as in the preceding experiment with the leaves, and related to the square-meter grape surface for each sample. Through an accident in ashing, the data for one sample from plat 7 are lacking. A second collection was made five weeks after the first and treated in the same way. The quantity of copper on the individual samples was so small in this case that it was impracticable to attempt to compare the data from the different plats. Three milligrams was the greatest weight of copper found on any sample in this last series. The weight of copper on grapes immediately after the fourth application is shown in Table IV under "Weight of copper," etc.

SURFACE TENSIONS OF MIXTURES USED.

In the course of the experiment on the leaves it was found that the mixtures with the lowest surface tension seemed to adhere best to the fruit, i. e., the Bordeaux mixture to which the rosin-fishoil soap had been added, thereby lowering the surface tension, seemed to adhere much more tenaciously to the bloom-covered fruit than the mixture without the added adhesive. This is in accordance with the results of the work of Vermorel and Dantony[1] on the adhesiveness of certain fungicides to grape berries. These authors found in their investigations that the addition of soap to the mixture lowered the surface tension, and that the mixture with the lowest surface tension wet the grape berries best.

With a view to discovering any existing relation between the surface tension of the mixture and its adhesiveness to the bloom-covered grape, surface-tension measurements were made of all the mixtures used in the experiments in 1911. For these measurements a spring balance of the Joly type, provided with a loop of 36-gauge German-silver wire, was used. This loop consisted of a piece of wire with both ends bent down at right angles to the straight middle portion, which was about 5 centimeters long. When in use, the middle portion of the loop was parallel to the surface of the liquid, the ends being perpendicular to this surface and extending below it. To determine the surface tension of a mixture the loop was first weighed with the ends thrust into the liquid and the middle portion at a given height, usually about 8 millimeters above the surface. The loop was then dipped into the liquid and weighed again with a film of the liquid held in the loop. The difference in weight with and without this film was considered to be the weight of the film.

While taking these measurements the fungicide was contained in a crystallizing dish of convenient size on the stand of the apparatus. As the same loop was used in all measurements and at the same height above the surface of the liquid in each test, these weights should have the same relation to each other as the surface tensions of the liquids. The weights for samples of the Bordeaux mixtures used in spraying the different plats are given in column 4 of Table IV. The measurements are, of course, not of the highest degree of accuracy, yet they were made at nearly the same time, at the same temperature, and with all other conditions as nearly alike as possible, and should therefore be comparable.

If the increased adhesiveness of the fungicide to the bloom of the grape is due to the lowering of the surface tension of the liquid, as might seem to be the case from the results obtained in 1910 in this investigation and from the work of Vermorel and Dantony, the surface tensions should be inversely proportional to the adhesiveness of the Bordeaux mixture found by determining the copper on the grape berries. From Table IV it may readily be seen that such is not the case. No. 4, in which 2 pounds of rosin-fishoil soap were added to the mixture, adhered best and had the lowest surface tension, while in No. 5, where half the quantity of the same soap was

[1] Vermorel and Dantony. Sur les bouillies anticryptogamiques mouillantes. Reveu de Viticulture, vol. 35, 1911, pp. 493–494.

used and which had practically the same surface tension, the quantity of copper on the berries was about two-thirds that found in No. 4. No. 6, in which 2 pounds of fishoil soap was added to the Bordeaux mixture, had the next lowest surface tension, but only about one-fourth as much copper to the unit area was found on the berries as in No. 4. No. 7, the glue mixture, which had the highest surface tension of any of the mixtures to which adhesives were added, except No. 3, most nearly approached No. 4 in adhesiveness. As may be seen from Table IV, no increased adhesiveness of the fungicide resulted from the addition of ferrous sulphate. In the samples of grapes from the plat sprayed with this mixture, as well as those from the two plats on which Bordeaux mixtures without added adhesives were used, there was not sufficient copper present for accurate determination. The results of these experiments show clearly that the adhesiveness of the spray mixtures is not to be related directly to the depression of their surface tensions,[1] but that some other factor must be taken into account.

A Laboratory Method for Comparing the Efficiency of Added Adhesives.

In the method used for measuring the surface tension of the mixtures a surface was employed which was wet by the liquid, while the waxy coating of the grape under usual conditions is not wetted by ordinary Bordeaux mixture. Experiments to prove this point were made by weighing grapes covered with bloom and dipping them into Bordeaux mixture to which no adhesives had been added and again immediately weighing them. In hundreds of tests the weights before and after the immersion did not vary as much as a milligram. But when grapes were immersed in Bordeaux mixture to which 2 pounds of rosin-fishoil soap to 50 gallons of mixture had been added, a film of the liquid was formed over the surface in every case and the weight was increased 26 to 80 milligrams by the mixture adhering to the surface.

From these experiments a method of measuring the relation of the liquid to the bloom of the grape itself was suggested. Strips of grape skin 2 to 3 millimeters in width and 20 to 40 millimeters in length were cut from the surface of the grapes. When these were peeled off they immediately rolled up in a comparatively straight rod about a millimeter in diameter, with the external surface of skin on the outside. This rod was thrust into the mixture to be tested, which was contained in a crystallizing dish on a convenient stand, and the depression of the surface of the liquid next to the rod was measured by means of a horizontal microscope with a micrometer ocular.

[1] Some months after this manuscript was submitted for publication, an article by Vermorel and Dantony came to the writer's notice, in which they reach the conclusion that the wetting power of a mixture is not related directly to the lowering of the surface tension. (See Vermorel, V., and Dantony, E., "Tension superficielle et pouvoir mouillant des insecticides et fongicides," Revue de Viticulture, vol. 37, 1912, pp. 715–716.)

A series of such measurements of Bordeaux mixture with and without the added adhesives was made, and it was found that the measurements related in a rough way to the amount of copper found in the determination of the copper on the grapes. That is, the mixture which adheres best to the surface of the grape is depressed the least and that which adheres least is depressed the most. The other mix-

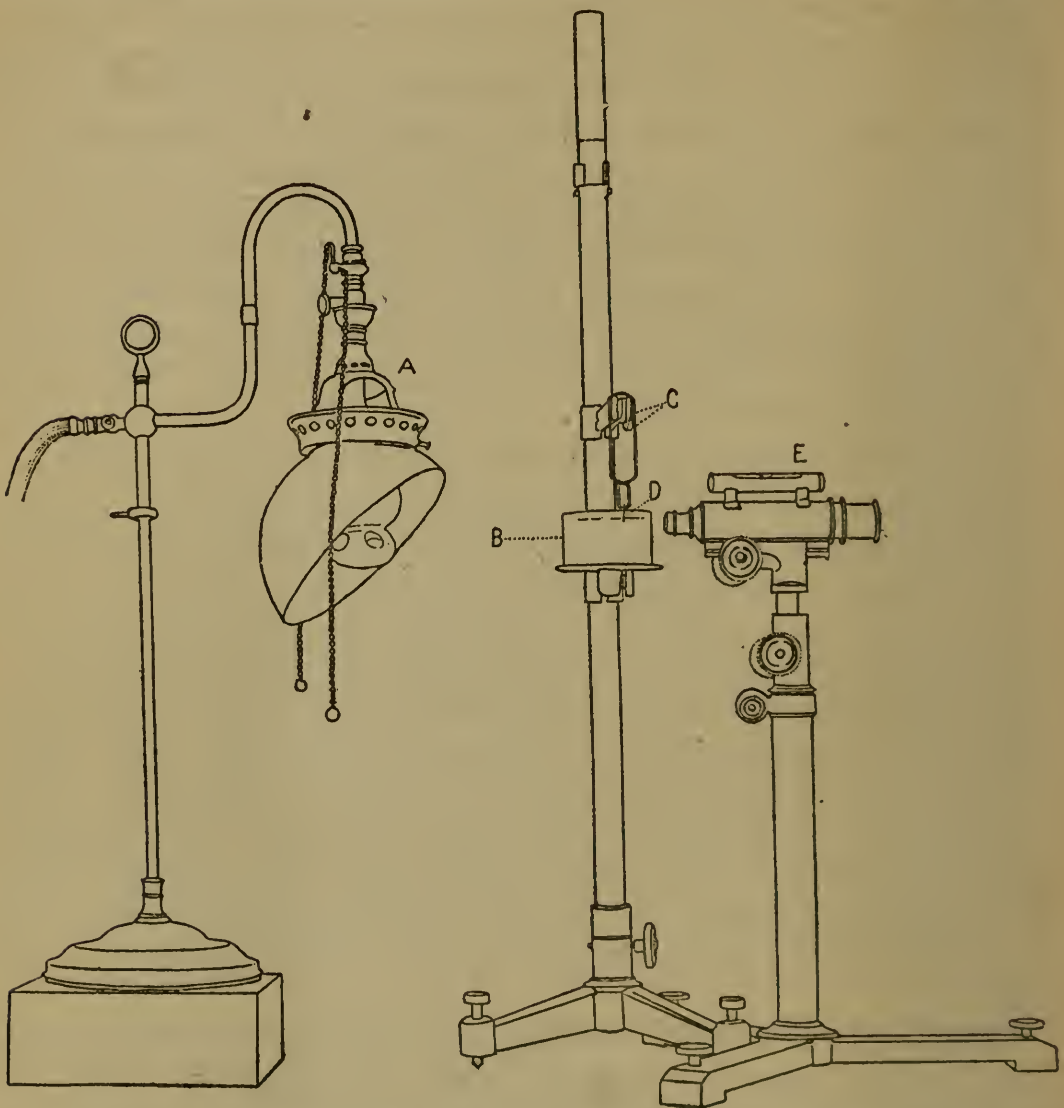

Fig. 3.—Sketch of apparatus used for measuring the depression of the surface films of Bordeaux mixture by the bloom of the grape. *A*, Source of light (Welsbach lamp); *B*, crystallizing dish containing mixture on apparatus stand; *C*, cover-glass forceps for holding roll of grape skin; *D*, roll of grape skin thrust partly below surface of liquid; *E*, horizontal microscope. The roll of grape skin *D* is placed so that the long axis is perpendicular to the liquid surface and the roll extends a little below this surface. The depression of the liquid surface next to the grape skin is then measured with the micrometer eyepiece of the horizontal microscope.

tures with added adhesives are grouped between these two extremes in the order of their value as determined by the amount of copper on the grapes. The method of measuring this depression is illustrated in figure 3. The average of a large number of measurements of Bordeaux mixture without added adhesives and with the four adhesives which proved of value are shown in Table IV under "Depression of surface of mixture." A diagram of the comparative depression of mixtures with the 2 pounds of rosin-fishoil soap and without additional adhesives is shown in figure 4. In making the

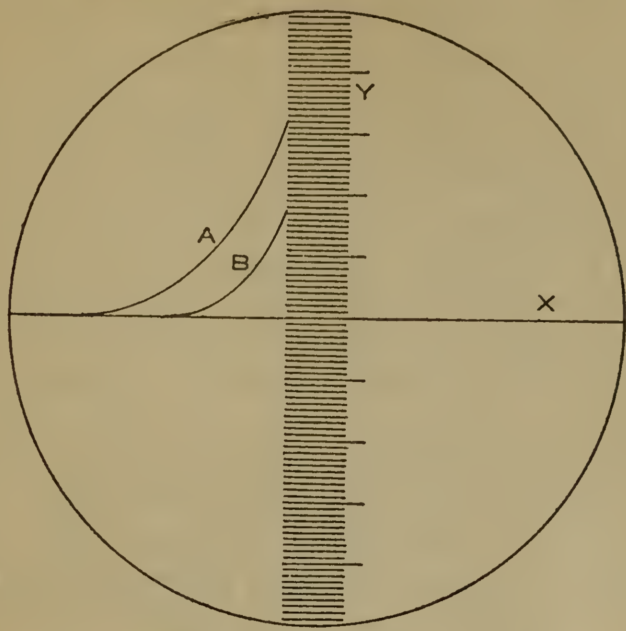

FIG. 4.—Diagram which compares the average depressions as seen in horizontal microscope of the surface films of Bordeaux mixture without added adhesive and Bordeaux mixture with 2 pounds of rosin-fishoil soap to 50 gallons of mixture. *X*, Median line of eyepiece micrometer which coincides with the surface of the mixture in crystallizing dish; *Y*, graduated scale of micrometer eyepiece; *A*, average depression of surface film of 4–2–50 Bordeaux mixture without added adhesives by the bloom of the grape; *B*, average depression of surface film of 4–2–50 Bordeaux mixture with 2 pounds of rosin-fishoil soap to 50 gallons of mixture.

measurements given in Table IV the mixture was always well agitated beforehand to stir up the precipitate and break up the film of calcium carbonate which formed on the surface.

TABLE IV.—*Results of tests showing the adhesiveness of Bordeaux mixture to grape berries alone and with various added adhesives, as shown by the quantity of copper on the berries, the comparative surface tension of various mixtures used as found by weight of a film of the mixture, and the depression of the surface of the mixture by the bloom of the grape.*

Plat No.	Mixture.	Weight of copper to a square meter of grape surface.			Weight of loop of mixture.	Depression of surface of mixture.
		Sample A.	Sample B.	Average.		
		Milligrams.	*Milligrams.*	*Milligrams.*	*Milligrams.*	*Millimeter.*
1	4–3–50 Bordeaux mixture	Trace.	Trace.	....	726	....
2	4–3–50 Bordeaux mixture with 2 pounds of ferrous sulphate	Trace.	Trace.	....	731	....
3	4–2–50 Bordeaux mixture	Trace.	Trace.	....	737	0.9442
4	4–2–50 Bordeaux mixture with 2 pounds of rosin-fishoil soap	24.5	30.3	27.4	499	.4980
5	4–2–50 Bordeaux mixture with 1 pound of rosin-fishoil soap	18.9	16.4	17.65	490	.6260
6	4–2–50 Bordeaux mixture with 2 pounds of fishoil soap	5.6	7.6	6.6	593	.6691
7	4–2–50 Bordeaux mixture with 2 pounds of ground glue	19.9	....	19.9	691	.6002
8	3–2–50 Bordeaux mixture with 2 pounds of rosin-fishoil soap	14.7	13.2	13.85	488	....

DISCUSSION OF THE RESULTS OF THE EXPERIMENTS.

The results of the experiments in preparing Bordeaux mixture detailed in the foregoing pages show that the rate of subsidence of the suspension is not dependent on the way in which the two components are brought together, provided one is dilute. It should be possible in practice to place the proper quantity of copper-sulphate solution or lime paste in the spray tank, dilute it with water to nearly the quantity of fungicide desired, and then to add the other component in a concentrated form. By vigorous agitation this should, according to the experiments described here, furnish a suspension which settles out as slowly as in any other common method of preparation. (See figs. 1 and 2 and Tables I and II.) This method of preparation should be useful in spraying cranberries. The conditions in this work are frequently such as to make impracticable the erection of raised platforms for mixing. It must be remembered that, with this method of mixing, good results were obtained only when the mixture was thoroughly agitated. The agitation was much more vigorous in the laboratory experiments than is usually obtained with the common barrel-pump outfit.

The addition of cane sugar to Bordeaux mixture to decrease the rate of subsidence, as Kelhofer and Kulisch suggested, was not tested. As these writers point out, it would probably be useful under certain conditions for persons using a small quantity of Bordeaux mixture from time to time. However, in commercial work in which Bordeaux mixture can be conveniently prepared and used immediately the addition of sugar would seem to entail an unnecessary expense. The method suggested by Pickering of preparing Bordeaux mixture by mixing the copper sulphate solution with a saturated solution of calcium hydroxid makes a suspension of the copper compound which settles out very slowly. It is doubtful whether this method would be practicable in the preparation of the large quantities of fungicides required for spraying in a commercial way. It would be impossible to prepare a neutral Bordeaux mixture containing three-fourths to 1 per cent of copper sulphate by this method, as a saturated solution of calcium hydroxid does not contain enough lime to react with the amount of copper salt in a solution of this concentration. The mixture might be prepared and the precipitate allowed to settle and enough of the supernatant liquid drawn off to leave a mixture of the proper concentration. This process would of course add greatly to the cost of spraying. It is possible that a smaller percentage of copper sulphate might be as effective as three-fourth to 1 per cent. From the evidence at hand, however, this conclusion would hardly be warranted.

Of the adhesive compounds added to Bordeaux mixture, the rosin-fishoil soap proved to be most effective on the grape berries—much more effective, in fact, than fishoil soap without the rosin. From this fact it seems probable that the adhesiveness is largely due to the rosin present. It is stated by various writers that the addition of a small quantity of soap to Bordeaux mixture could be of no particular benefit, as the soap would be precipitated as an insoluble calcium soap by the excess of calcium present in the mixture. Good results could therefore be expected only when a considerable quantity of soap was added. This, of course, may be true of certain kinds of soap, but in this investigation considerable benefit was derived from the addition of relatively small quantities of soap. Even the fishoil soap materially increased the adhesiveness over Bordeaux mixture without added adhesives.

In the treatment of the black-rot of the grape good results have been obtained in many cases by using Bordeaux mixture without added adhesives. When we consider this fact in connection with the evidence brought forth in the present investigation, that Bordeaux mixture without added adhesives does not adhere to the grape berry in any appreciable quantity, it seems probable that the protection is due to reducing the sources from which infection comes to the berry. By protecting the foliage from infection the possibility of secondary infection from the foliage to the fruit may be eliminated to a considerable extent. Covering the stems of bunches of grapes with the fungicide seems to be another means by which infection may be kept from the grapes. The writer has observed numerous instances of black-rot infection on bunches of grapes which had been bagged six weeks or more. In these instances spores probably washed down the stems in drops of water, as the only openings in the bags were immediately around the stems.

Though good results were obtained by the addition of glue to Bordeaux mixture, its cost (about 12 cents a pound) prohibits its use in commercial work in place of rosin-fishoil soap. When glue is added to alkaline Bordeaux mixture, part of the copper combines with the glue, forming a soluble compound bright purple in color. It is probable that much of the copper found on the grapes from this plat was in this form. As it is soluble in water, this protective covering might not remain on the berries as long as the insoluble precipitates in the mixtures with the soap.

It is difficult to see in just what way ferrous sulphate could be expected to influence the adhesiveness of Bordeaux mixture. On the addition of this compound to a solution of calcium hydroxid, ferrous hydroxid immediately precipitates out, and none of the ferrous sulphate remains in solution. It was found to be worthless as an adhesive for use on grapes.

In the plats sprayed with 3–2–50 Bordeaux mixture, to which was added 2 pounds of rosin-fishoil soap, a considerable quantity of copper was found on the grapes. This formula has given excellent practical result and should prove much more effective than the mixtures containing more copper but without the adhesive.

CONCLUSION.

It has been shown in these investigations that a Bordeaux mixture in which the suspension of the copper compound settles out slowly may be prepared by adding the concentrated calcium hydroxid to the diluted copper-sulphate solution or vice versa, provided the mixture is sufficiently agitated. Practically as good results were obtained with these methods of preparation as by diluting the two components in separate vessels and pouring them simultaneously into a third, as recommended by Galloway in 1896.

It is to be remembered that in preparing Bordeaux mixture, by pouring one of the components in concentrated form into the other diluted to nearly the required volume, the resulting mixture must be thoroughly agitated. The agitation necessary for preparing Bordeaux mixture with a low rate of subsidence by this method could hardly be obtained in practice except by means of a power outfit provided with a good agitator. This method of mixing is not designed to replace the old gravity method with its elevated platform, but offers a convenient substitute where for any reason the gravity method is impracticable.

In the experiments on the adhesiveness of certain Bordeaux mixtures and the relative value of certain adhesive compounds it was shown that by determining the quantity of copper retained on the leaves sprayed with the different mixtures the addition of rosin-fishoil soap slightly increases the adhesiveness of the mixture. In similar experiments on grape berries it was shown that the adhesiveness of the fungicide could be materially increased by the addition of certain adhesive compounds. Two pounds of rosin-fishoil soap to 50 gallons of spray mixture was the most valuable of any added adhesive, ground glue was second, 1 pound of rosin-fishoil soap to 50 gallons of mixture was third, and fishoil soap was fourth. Ferrous sulphate did not increase the adhesiveness of the Bordeaux mixture, as no appreciable quantity of copper adhered to the grape berries where the Bordeaux mixture to which the ferrous sulphate had been added was used. No appreciable quantity of copper was found on the grape berries from the plats sprayed with Bordeaux mixture without added adhesives. From the experiments on grape berries with adhesives it may be concluded that the use of an adhesive compound is necessary to make the fungicide adhere to the bloom-covered grapes. Two pounds of rosin-fishoil soap to 50 gallons of mixture gives the best results and is recommended as the most economical and efficient adhesive for use on grape berries. From the results obtained with 3–2–50 Bordeaux mixture, with the addition of soap, it seems probable that a mixture containing this quantity of copper sulphate would be effective when a good adhesive is used. A laboratory method of approximating the relative adhesiveness of these fungicides to grapes was developed.

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U. S. DEPARTMENT OF AGRICULTURE.

BUREAU OF PLANT INDUSTRY—BULLETIN NO. 266.

B. T. GALLOWAY, *Chief of Bureau.*

THE DESTRUCTION OF CELLULOSE BY BACTERIA AND FILAMENTOUS FUNGI.

BY

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AND

F. M. SCALES, *Assistant Soil Mycologist,*
Soil-Bacteriology and Plant-Nutrition
Investigations.

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LETTER OF TRANSMITTAL.

U. S. DEPARTMENT OF AGRICULTURE,
BUREAU OF PLANT INDUSTRY,
OFFICE OF THE CHIEF,
Washington, D. C., September 12, 1912.

SIR: I have the honor to transmit herewith and to recommend for publication as Bulletin No. 266 of the series of this Bureau a manuscript entitled "The Destruction of Cellulose by Bacteria and Filamentous Fungi." This paper was prepared by Messrs. I. G. McBeth, Physiologist, and F. M. Scales, Assistant Soil Mycologist, of the Office of Soil-Bacteriology and Plant-Nutrition Investigations, and has been submitted by the Physiologist in Charge with a view to publication.

New species of cellulose-destroying organisms are described, as well as special methods and new culture media adapted to their isolation and identification.

Respectfully,

B. T. GALLOWAY,
Chief of Bureau.

HON. JAMES WILSON,
Secretary of Agriculture.

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PREFATORY NOTE.

Of the phenomena relating to the art of agriculture none is more interesting or more vitally important than the formation of starch, sugar, cellulose, and similar compounds by green plants. The action of chlorophyll, as yet unexplained, by which plants utilize the energy of the sunlight to synthesize carbon dioxid and water into carbohydrates is logically the most fundamental question of plant physiology, for it is the enormous quantity of potential energy thus accumulated that directly or indirectly makes possible the continuance of all vital processes. Not only animal life which, generally speaking, is dependent upon plants for food supply, but even the successful growth of crop plants is largely controlled by the decomposing carbonaceous material in the soil.

Both in the Eastern and Western States the natural maintenance of the supply of available nitrogen is seldom considered when determining the most desirable system of farm management in any region, yet, as scientific research and experience in the field agree in showing, the soil itself may fix and render available to the crops considerable quantities of nitrogen, which is the highest priced of the plant foods if it be purchased as commercial fertilizer. Furthermore, as far as our experience extends, all of the fixation of atmospheric nitrogen in the soil is dependent upon the growth of microorganisms which must have large quantities of soluble carbon compounds for food. As by far the larger part of the carbonaceous material added to the soil as dried roots, stubble, green manure, etc., is cellulose, a substance which is unusually refractory and can not be used as such for food by nitrogen-fixing organisms, the biological phenomena which transform the cellulose to soluble compounds are obviously important. It is not only as a possible food supply for nitrogen-fixing organisms, however, that a soil requires a constantly replenished supply of cellulose. Its decomposition under proper soil conditions and in association with the decay of nitrogenous compounds makes possible the formation of the so-called soil humus. The beneficial effects of the presence of indefinite humic compounds upon the physical character and fertility of a soil are generally recognized throughout the agricultural regions of the United States.

The gradual processes of decay that are depended upon to maintain many of the factors of a soil's fertility are probably as complex as the microflora and fauna of the living soil itself, and it is chiefly through the discovery and comprehension of the essential biological phenomena relating to the growth of plants that permanent improvement in crop production can be made possible. The decomposition of cellulose is apparently one of the fundamental questions of the decay of organic material, and though a subject of research in foreign countries for many years, it has been but imperfectly understood. The investigations, therefore, of which this bulletin is a progress report, are regarded as of unusual importance.

KARL F. KELLERMAN,
Physiologist in Charge.

OFFICE OF SOIL-BACTERIOLOGY INVESTIGATIONS,
Washington, D. C., September 10, 1912.

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B. P. I.—779.

THE DESTRUCTION OF CELLULOSE BY BACTERIA AND FILAMENTOUS FUNGI.

INTRODUCTION.

The important functions of fission fungi is to dissolve and again place in circulation the complex organic substances which have ceased to live. Without their activity the cycle of change to which all organic matter is subject would come to a standstill and the food supply of plants would soon be depleted. It is well known that through the agency of micro-organisms all vegetable matter is gradually transformed into the complex mixtures ordinarily known as humus and that we are at least partially dependent upon the quantity and quality of the humus compounds for the fertility of the soil.

It is true that numerous chemical researches have added materially to our knowledge of these organic or humic compounds, but since the biological processes involved are the dominant factors in determining the manner in which complex organic substances are split up, a systematic study of the organisms which bring about the decomposition of vegetable matter and the formation of plant food is imperative.

Vegetable substances may be roughly divided into two great classes, nitrogenous and nonnitrogenous. In the decomposition of nitrogenous matter we are concerned chiefly with the fate of the nitrogen, a part of which seems to be invariably returned to the atmosphere. It is well known that this loss may be considerable. Fortunately, however, nature has provided a means of restoring this lost nitrogen through the activity of certain so-called nitrogen-fixing micro-organisms. A study of these organisms and the conditions under which they are able to fix nitrogen has shown that the process is controlled in large measure by the available supply of organic carbon.

On examining plant tissues we find a large percentage of the carbon content locked up in the celluloses; these are inert compounds which resist the attacks of the ordinary putrefactive bacteria and until broken down into simpler compounds are inaccessible to nitrogen-fixing bacteria. Little is known of the biological processes involved in the destruction of cellulose. It is true that many foreign investigators have studied cellulose ferments, but generally the work has been done in a tentative way, and no suitable methods for isolating these essential organisms have been worked out. Consequently, our ideas of the number and nature of the cellulose ferments have been very inadequate. Investigators have also devoted much attention to the products resulting from the fermentation of cellulose, but they apparently have been working with impure cultures and their conclusions are of doubtful value.

Believing that little progress could be made in the study of cellulose decomposition until satisfactory methods for isolating these organisms had been perfected, thus giving opportunity to learn something of their cultural characteristics, we have endeavored to work out methods to isolate and study them. The purpose of this bulletin is to review briefly the work of earlier investigators, point out the inadequancy of our present knowledge of cellulose fermentation, and set forth the results obtained from our own studies in the hope that they may be of value to other investigators.

HISTORICAL REVIEW OF INVESTIGATIONS OF THE DESTRUCTION OF CELLULOSE.[1]

BACTERIOLOGICAL AND CHEMICAL INVESTIGATIONS.

The fermentation of cellulose was first attributed to the activity of microorganisms by E. Mitscherlich in 1850. He noted that when slices of potato were immersed in water and held in a warm place the cellulose, which constitutes the main portion of the cell walls, was destroyed. First, the cells became separated from each other, and soon afterward the walls were broken down and the starchy material fell out. By filtering the solution and dropping in fresh potato the fermenting process was greatly accelerated. Microscopic examinations showed no trace of a mold growth, but Mitscherlich observed swarms of vibrios, which he believed to be the active agents of cellulose fermentation.

Four years later Haubner showed that it was impossible to recover from the feces more than 50 per cent of the crude fiber fed to ruminants. He obtained similar results with wood shavings which had been treated with acid and alkali, and also with thoroughly washed paper when fed with hay and bran to sheep. Haubner's work was soon confirmed by Henneberg and Stohmann. Through the experiments of Hofmeister, Zuntz, Knierem, Weiske, Lehmann, and others similar results were obtained with horses, sheep, goats, rabbits, etc. Although undertaken primarily to determine the nutritive value of crude fiber in foodstuffs, these investigations no doubt did much to stimulate later investigation which sought to determine the causative agent of cellulose fermentation.

[1] For bibliography, see pages 47 to 50.

In 1865 Trecul undertook a study of microorganisms in macerated plant tissues. He observed and described three forms, which he placed in a distinct genus, Amylobacter, and divided into three subgenera. This generic name was selected because these organisms stained blue with iodin. He believed that starch or cellulose favored the production of these bodies.

For our early knowledge of cellulose fermentation we are much indebted to the work of Popoff, who in 1875 first pointed out the connection between cellulose fermentation and the formation of methane. Methane had long been known to exist in sewers and marshes and had been found in fermenting horse manure by Reiset as early as 1856; however, no successful attempt had been made to determine the source of the gas. For experimental material Popoff used slime from the sewers of Strassburg. The material was mixed with sufficient water to make a thick solution, poured into large flasks, and preparations made to collect the gas over quicksilver. An analysis of the gas showed that considerable quantities of methane mixed with other gases had been produced. The optimum temperature for the gas formation was found to be 38° C. to 40° C.; at 45° C. the activity was much weakened, and at 50° C. it came to a standstill; lower temperatures were also shown to be very unfavorable. Popoff further showed that the fermentation process could be altered at will by the addition of antiseptics. The next step was to show that the formation of methane could result from the destruction of pure cellulose. With this end in view a quantity of pure Swedish filter paper was immersed in water and inoculated with a small quantity of slime known to contain the methane ferment. The paper was destroyed and a large quantity of gas was formed, which on examination proved to be a mixture of carbon dioxid, methane, hydrogen, and nitrogen. The gases collected during the first two weeks and again several weeks later were analyzed with the result shown in Table I.

TABLE I.—*Analyses of mixed gases formed by the decomposition of Swedish filter paper.*

Gases found in the mixture.	Collected at the end of—	
	Two weeks.	Several weeks.
	Per cent.	*Per cent.*
Carbon dioxid	25.70	34.07
Methane	14.42	37.12
Hydrogen	14.36	1.06
Nitrogen	45.52	27.75

It appears, therefore, that the quantity of hydrogen decreased with the duration of the experiment, while at the same time there was an increase in the formation of methane, so that in the end the quantities of carbon dioxid and the methane were about equal. In studying other substances Popoff found that a methane fermentation could be produced from gum arabic as well as from cellulose. He is of the opinion that the typical cellulose ferment gives rise to carbon dioxid and methane only, and that the presence of hydrogen in the gas is due to other fermentation processes.

In 1877 Van Tieghem, in working out the life history of the amylobacter of Trecul, found that it was motile, as Nylander had done twelve years before, and classified it as a bacillus. He further found that it was an anaerobic, cellulose-dissolving organism and that it grew readily in soluble starch and cellulose, first reducing them to dextrin and then converting the dextrin into glucose, which was fermented with the production of carbon dioxid, hydrogen, and an acid which inhibited the growth of the organism unless neutralized with calcium carbonate. No cytase was liberated in solution and the cellulose was dissolved only when in direct contact with the organism. He proved the cellulose-dissolving power of the organism to his own satisfaction by inoculation experiments in solutions containing macerated radish. However, he found that the organism did not act the same on all plant tissues; in a word, that *Bacillus amylobacter* could not attack all celluloses.

Results of studies on fermentation processes were published by Prazmowski in 1880. He described two species to which he attributed cellulose-fermenting properties and to which he gave the names *Clostridium polymyxa* and *Vibrio rugula*. The former was found to have only a weak fermentive power in dextrin solutions, but was extremely active in preparations of cooked potato and lupine seed; its activity on starch and cellulose is described as very vigorous. An analysis of the gas formed showed only hydrogen and carbon dioxid.

Vibrio rugula is of especial interest because the description given is so similar to that given later by Omelianski for his so-called hydrogen and methane ferments. In young cultures the rods were unusually thin, about 8 microns long, and showed a characteristic curved structure which made it easy to separate them from other species; later the rods became uniformly thicker, the end swelled up, and a round spore appeared. The young rods were actively motile and the organism was classed as an anaerobe. In infusions of plant tissue the organism was found to surround the cell walls, which were soon dissolved. Prazmowski also made a study of an organism which stained blue with iodin; to this he gave the name *Clostridium butyricum*, although he regarded it as synonymous with *Vibrion butyrique* Pasteur, *Amylobacter* Trecul, *Bacillus amylobacter* Van Tieghem, and *Bacterium navicula* Reinke and Berthold.

In an extensive series of experiments inaugurated in 1880 Tappeiner has given us interesting data on the disappearance of cellulose in the digestive tract of herbivorous animals. Incidentally he made a study of the compounds produced by cellulose fermentation. Pure cellulose in the form of cotton and filter paper was placed in flasks containing a rich nitrogenous solution. In one series of experiments a 1 per cent neutral flesh extract was poured into flasks; pure cellulose in the form of cotton was added, sterilized, and inoculated with a drop of material from the stomach. It was observed that the flesh-extract solution invariably resulted in a fermentation independent of the typical cellulose ferment; therefore, a check flask containing only the flesh extract was held under the same conditions as the flesh-extract cellulose flask. The result of one such experiment is shown in Table II.

TABLE II.—*Measurement of gas formed by the decomposition of cellulose and flesh extract and of flesh extract alone.*

Gas formed.	Flask 1, cellulose and flesh extract.	Flask 2, flesh extract (check).
	C. c.	*C. c.*
Carbon dioxid	191.00	10.10
Hydrogen	1.70	3.00
Nitrogen	10.40	8.60
Methane	88.30	4.20

The fatty acids found in flask 1 amounted to 1.6651 grams and consisted of 2.2 parts acetic acid to 1 part butyric acid, giving a carbon content of 0.7414 gram. The fatty acids found in flask 2 amounted to 1.005 grams and consisted of 2.1 parts acetic acid to 1 part butyric acid, giving a carbon content of 0.4918 gram. Subtracting the products found in flask 2 from those in flask 1 we have the products formed in the decomposition of cellulose, with a carbon content as follows: Carbon dioxid, 0.0966 gram; methane, 0.0436 gram; acids, 0.2496 gram. Total, 0.3898 gram.

In the fermentation of the cellulose 0.4165 gram of carbon was lost, being slightly more than the quantity recovered in the by-products.

A separate series of experiments was conducted with Nageli's solution [1] and 0.6 per cent asparagin. Flasks of 360 cubic-centimeter capacity were filled with this solution and 3.5107 grams of dry cotton added. The fermentation resulted in the formation of carbon dioxid, hydrogen, and nitrogen. Fatty acids, including acetic, propionic, and butyric, were also produced. In the control flask no gas was formed and only traces of fatty acids. It is noted in this experiment that

[1] Potassium phosphate (dibasic), 0.20 gram; magnesium sulphate, 0.04 gram; calcium chlorid, 0.02 gram; water, 100.00 c. c.

0.9447 gram of cellulose was destroyed without any methane formation, while in the previous experiment a considerable quantity of methane was produced even in the flesh-extract flask to which no cellulose was added.

The interesting series of experiments commenced by Hoppe-Seyler in 1881 did much to confirm the growing belief that cellulose is decomposed by a methane ferment. The main experiment is as follows: On December 2, 1881, 25.773 grams of pure filter paper were added to a 1,101 cubic-centimeter flask inoculated with a small quantity of slime and sufficient water added to bring the mixture up to about 700 cubic centimeters. The flask was protected from the light by a double layer of black paper and preparations were made to collect the gas over quicksilver. The flask was kept in this condition four years. In the early part of the experiment the gas formation was very active, toward the end of the year 1883 it became weaker, and in the second half of the year 1885 was scarcely apparent. The experiment was discontinued on December 6, 1885. Gas analyses to the number of 95 were made from time to time, the summary of which showed 3,281 cubic centimeters of carbon dioxid and 2,571 cubic centimeters of methane. An examination of the contents of the flask at the end of the experiment showed that 15 grams of cellulose had been consumed, and as no other appreciable quantity of by-products could be found the author concluded that cellulose is dissolved according to the following formula:

(1) The hydration of the cellulose with the formation of a hexose,

$$C_6H_{10}O_5+H_2O=C_6H_{12}O_6; \text{ and}$$

(2) the destruction of the carbohydrate with the formation of equal quantities of carbon dioxid and methane,

$$C_6H_{12}O_6=3CO_2+3CH_4$$

Hoppe-Seyler says in one part of his work that the formation of carbon dioxid took place only when he found in his solutions living bacteria which showed no difference from *Bacillus amylobacter* of Van Tieghem, and he is therefore of the opinion that the destruction of cellulose was due to the activity of this organism.

Gayon in 1883 and 1884 noted the presence of methane in fermenting manure, and from 1 cubic meter of this material well moistened with water and held at 35° C. he succeeded in obtaining as much as 100 liters of the gas in 24 hours. This fermentation he attributed to an extremely small anaerobic organism which was cultivable in nutrient solutions containing either straw or paper, in which it attacked the cellulose and liberated carbon dioxid and methane.

The extensive experiments of Deherain in 1883 and 1884 on the aerobic and anaerobic fermentation of straw and manure showed that in a pile of manure under natural conditions the gas liberated at the bottom of the pile is pure methane and carbon dioxid, while no methane is produced near the surface unless the manure is wet, when as much as 10 per cent of the gas produced may be methane. In hermetically sealed flasks the fermentation soon stopped, but on opening the flasks and resealing them the fermentation began anew, though it continued for but a short time. He concluded from this result that the methane ferment is not a strict anaerobe. Occasionally a fermentation produced hydrogen and carbon dioxid and gave a slightly acid reaction, due to the formation of butyric acid, while the pure methane fermentation was always neutral. A microscopic examination of the liquid of two such solutions showed in both cases numerous extremely fine rods, which were almost identical, and the butyric ferment in addition where hydrogen was produced. Deherain tried Pasteur's method of successive cultures, but did not reach an absolute conviction concerning the differences between the two organisms. A few drops of a manure infusion inoculated into dextrin and cane sugar solutions gave hydrogen, while a similar inoculum in a solution containing paper gave methane. A later experiment under the same conditions reversed these results in that 9 per cent of gas from the fermentation of cane sugar was found to be methane, while hydrogen was secured from the decomposition of paper. This evidence was thought sufficient to show the presence of two different anaerobic ferments, one hydrogen and the other methane. It sometimes happened in experiments with straw that an acid fermentation took place and that the dominating gas was methane. This production of acid he believed to be due to a fermentation of sugar, producing hydrogen and butyric acid, and that such fermentation was succeeded by the regular methane fermentation.

The results of a study of the fermentation of manure by Schloessing in 1889 showed that the anaerobic fermentation was much more active at 52° than at 42° C., and that methane was the predominating, if not the only combustible gas given off. Three years later he and his son carried on some experiments to determine what part bacteria play in the aerobic and anaerobic fermentation of manure at different temperatures. The aerobic work showed that no combustible gas was produced under these conditions and that the bacteria were very active up to 72.5° C., but that at 81° C. all action ceased; in an atmosphere of carbon dioxid he obtained a methane fermentation at 52° but none at 66° C. Once he obtained methane from cow manure and hydrogen from horse manure at 58° C. In one experiment 124.4 grams of fresh horse manure containing 76 per cent moisture were kept in an atmosphere of carbon dioxid for two months at 52° C., and in that time generated 4,217.5 cubic centimeters of carbon dioxid and 4,577.4 cubic centimeters of methane, which are equivalent to 4.72 grams of carbon. In the first 500 cubic centimeters of gas produced 15.8 cubic centimeters of hydrogen were also found. An analysis was made to determine the quantity of each element in the dry material of 124.4 grams of manure. The results are shown in Table III.

TABLE III.—*Analysis of 124.4 grams of horse manure (dry).*

Stage.	Carbon.	Hydrogen.	Oxygen.	Nitrogen.	Minerals.
	Grams.	*Grams.*	*Grams.*	*Grams.*	*Grams.*
Before fermentation	12.67	1.653	10.70	0.453	3.69
After fermentation	7.92	1.125	7.08	.392	3.79
Loss (−) or gain (+)	−4.75	−.528	−3.62	−.061	+.10

The loss of 4.75 grams of carbon corresponds very closely with the 4.72 grams found in the carbon dioxid and methane obtained during the fermentation and represents 37.5 per cent of the carbon in the fresh manure.

The methane fermentation of cellulose was likened to the alcoholic fermentation of sugars by Berthelot in 1889, in that it is determined by living agents, is accompanied by a fixation of the elements of water, and has a similar thermochemical mechanism. He represented the fermentation as taking place so that all the hydrogen of the water enters into one of the products (methane) while all the oxygen goes to form carbon dioxid. The total heat thus liberated was 41 calories, the products being gaseous.

The effect of alkalinity and aerobic or anaerobic conditions on the progress of fermentation and mode of decomposition of straw was investigated by Hebert in 1892. The importance of alkalinity in this fermentation was tested by adding solutions containing from 5 to 10 per cent of potassium carbonate, ammonium carbonate, and ammonium phosphate to dry powdered straw of known composition. This straw suspension was inoculated with several cubic centimeters of urine and incubated at 55° C. After four days the anaerobic flask containing 5 per cent of salts had produced the greatest quantity of gas, which was composed of equal parts of carbon dioxid and hydrogen, but a week later without any change in conditions this flask began producing methane. The predominance of either of the carbonates made no appreciable difference in the rapidity of fermentation, but an excess of ammonium carbonate gave hydrogen at first and methane six days later and an excess of potassium carbonate gave methane in the beginning. The composition of the straw before and after three months' fermentation is given in Table IV.

TABLE IV.—*Composition of straw before and after fermentation.*

Constituent.	Initial composition of straw.	Composition of straw after 3 months' fermentation.	
		Anaerobic fermentation.	Aerobic fermentation.
	Per cent.	*Per cent.*	*Per cent.*
Water	9.52	....	....
Nitrogenous materials	4.87	12.50	14.06
Chlorophyll materials	.93	1.36	.62
Reducing sugars	2.43	.00	.00
Dextrin	.05	.00	.00
Gums, tannins, acids	.60	1.26	1.60
Cellulose	28.25	23.30	25.65
Vasculose	28.03	22.70	28.25
Straw gum (into xylose)	20.00	26.00	19.00
Ash	7.15	12.80	12.80
Total	101.83	99.92	101.98
	Gram.	*Gram.*	*Gram.*
Weight of dry straw	0.4524	0.2645	0.2690
Weight of cellulose in the straw	.1412	.0615	.0689

The results show that in both fermentations the straw lost about half its weight and that this loss occurred principally in the three elements most abundant in straw, namely, cellulose, vasculose, and straw gum. The straw first lost all or part of the substances most easily attacked, as chlorophyll materials, gums, tannins, glucose, and dextrin, the higher carbohydrates, cellulose, and straw gum disappearing afterwards; the loss of the former amounting to as much as 50 per cent (7 or 8 grams). Finally the vasculose was partly dissolved in the solution and partly oxidized to carbon dioxid and water. The organisms appeared to work as vigorously under aerobic as under anaeorbic conditions, and Hebert was unable to decide from his figures what are the most favorable conditions for the destruction of cellulose.

Van Senus in 1890 published an extensive treatise on the decomposition of cellulose. He noted the rapid decomposition of cotton, pieces of bean, potato, etc., when inoculated with slime and held under favorable conditions, and attributed these decomposition processes to the joint activity of two organisms. One of them, *Bacillus amylobacter,* which he describes as a rod-shaped organism, 0.8 μ wide and 2 to 10 μ long, stains blue with iodine and forms spores when air is admitted, which then germinate only when air is excluded He showed that *B. amylobacter* in flesh-extract solution with cellulose (cotton, paper, crude fiber, etc.) under no conditions could ferment the cellulose; with sterilized beans, potatoes, etc., the walls were not broken down but only separated from each other, probably through the formation of the ferment pectase. The other organism, isolated from the intestines of the rabbit, is much smaller and, like *B. amylo-*

bacter, has no power to ferment cellulose in pure culture; however, when these organisms were grown in association a destruction of cellulose was secured. The reason for this result, in the opinion of this author, is the production of harmful products by the fermentation of the cellulose, and the presence of another organism is necessary to render these products less injurious. Van Senus is of the opinion that methane is not formed directly by the fermentation of cellulose and that the destruction of cellulose always results in the formation of hydrogen, carbon dioxid, and acetic acid, the action of the hydrogen upon the acetic acid reducing it to aldehyde, alcohol, and finally to methane.

In 1894 Omelianski began an investigation of bacterial cellulose destruction by inoculating a nutrient solution containing filter paper with slime from the River Neva, and incubating at 30° to 35° C. The paper was soon changed into a yellowish, transparent, gelatinous mass, which later disappeared, leaving only a slight residue. A long, extremely slender bacillus with a round polar spore was present in great numbers. In order to get a pure culture of this cellulose-fermenting organism, Omelianski employed Winogradski's "method of elective culture," selecting for this purpose a nutrient solution almost void of organic nitrogen and incubating the culture anaerobically, as the results obtained by Hoppe-Seyler had showed that these conditions were favorable for bringing about a predominance of the desired organism. After a sufficient number of transfers had been made this slender rod, *Bacillus fermentationis cellulosae*, was almost the only one present in the culture, and when inoculated into solutions containing cotton and cellulose of cabbage, turnip, and pith of the elder tree which had been precipitated from Schweitzer's reagent, it produced a vigorous fermentation with the liberation of hydrogen and carbon dioxid. A little methane generated in the first cultures was believed to be due to another organism which disappeared in transferring. A 300-cubic-centimeter flask was filled with a solution made as follows:

Potassium phosphate	1.0 gram.
Magnesium sulphate	.5 gram.
Ammonium sulphate	1.0 gram.
Sodium chlorid	Trace.
Distilled water	1,000.0 c.c.

To this solution were added 3.4743 grams of dry filter paper and 5.7698 grams of calcium carbonate. It was then inoculated with the cellulose ferment, purified by the elective culture method, and incubated 13 months at 35° C. During this time the volume of carbon dioxid in the total gas increased from 15 per cent at first to 98 per cent, and toward the end of the period dropped to 80 per cent. At the conclusion of the experiment an analysis of the solution showed the presence of 2.2402 grams of acetic and butyric acids, in the ratio of 1.7 to 1, and of gaseous products consisting of 0.9722 gram carbon dioxid and 0.0138 gram hydrogen, making the total weight of by-products produced 3.2262 grams. A loss in cellulose, amounting to 3.3471 grams, was noted, being somewhat greater than the total weights of the by-products found. Valerianic acid, higher alcohol, products giving the odor of decaying cheese, and dissolved hydrogen were not measured. These unmeasured products might account for the difference of 0.1209 gram between the cellulose added and the products obtained. According to these figures 70 per cent of the cellulose used is converted into fatty acids, while hardly 30 per cent is liberated as gas.

In 1904 Omelianski published his method for the separation of the two cellulose-destroying organisms, one called the "hydrogen bacillus," formerly *Bacillus fermentionis cellulosae*, and the other the "methane bacillus." The vegetative cells of the methane organism appeared to form spores more readily than those of the hydrogen bacillus, so to obtain a pure culture of the latter he first heated the inoculating material to 75° C. for 15 minutes in order to kill all the germinating methane organisms, and after several repetitions of this process his culture was apparently free from this organism. After five or six generations the surface of the paper was covered with a bacillus 4 to 8 μ long and 0.5 μ wide, the rods later reaching a length of 10 to 15 μ without gaining in thickness. They never formed chains and took the ordinary anilin stains readily, but would not color blue with iodin. Slightly curved or irregular spiral forms were abundant when fermentation was going on, and in older cultures the rods had a round polar spore 1.5 μ in diameter. When the paper had been destroyed there were many free spores and few rods in the solution. These spores were found to withstand a heat of 90° C. for 25 minutes, so the cultures were freed of nonspore formers in this way. After such heating there still persisted in his cultures a large bacillus with an oval polar spore, while another contaminating organism with a round polar spore and very much like the true cellulose ferment was occasionally present. The former grew readily on solid media like agar jelly, filter paper soaked in gummy salt solution, carrots, summer cabbages, turnips, and potatoes, either alone or in association with other bacteria, but the true hydrogen ferment did not grow on any of these media except that in one case some very small, yellow, liquefying, semitransparent colonies of the cellulose bacteria appeared on a potato plate; they were scarcely apparent without a magnifying glass, and this medium was evidently not a favorable one, as a heavy inoculation gave only a few colonies.

Omelianski asks, "Was the mother culture not absolutely pure or is the growth an association of bacteria?" Inoculation from these colonies gave fermentation of cellulose in only one case and that soon stopped. However, the organism from these colonies had all the morphological characteristics of the bacteria found in the fermenting paper, and he concludes that it was without doubt a pure culture of this bacillus, although it did not give a satisfactory fermentation.

A test to determine whether it was an associative action with the bacillus forming the oval spore gave negative results. The methane ferment was obtained by inoculating a flask containing the mineral salt solution, paper, and chalk with canal slime or fresh horse manure, and incubating anaerobically at 35° C. A microscopic examination of the paper showed that it was covered with an organism similar to the hydrogen bacillus, but thinner and more delicate in outline. The culture was purified by transferring and heating to kill nonspore formers until it presented a microscopically pure picture, appearing as a rod 5 μ long and 0.4 μ wide, with a round polar spore 1 μ in diameter. Morphologically these organisms might be classed as the same species, but physiologically they were very different, for one produced hydrogen and the other methane.

In later investigations Omelianski points out that methane may be produced not only from cellulose but from acetates, pentoses, pentosans, butyrates, lactates, and protein bodies, which, he believes, indicates that the number of reactions in nature which involve the formation of methane is no smaller, perhaps, than the fermentation processes leading to the evolution of hydrogen.

Experiments showing the destruction of the inner tissue of the turnip due to the parasitism of *Pseudomonas campestris* were reported by Smith in 1902. The leaves of the plant were inoculated with a pure culture of this bacterium. The disease moved downward, and sections of the root 52 days after inoculation showed the bacteria to be very abundant in the inner part, although the root was entirely white and sound externally. Cultures made from the diseased interior yielded only *P. campestris*. Carefully prepared sections showed all stages in the solution of the cell walls, from single cells or vessels occupied by the bacteria to cavities filling the place formerly occupied by hundreds of cells and filled with the bacteria and remnants of the cell walls.

Experiments by Van Iterson in 1904 have shown that the fermentation of cellulose may be caused by aerobic as well as by anaerobic bacteria. According to his results the anaerobic processes fall into two groups:

(1) Without the presence of nitrates the cellulose may undergo a hydrogen or methane fermentation.

(2) In the presence of nitrates the cellulose is destroyed by denitrifying bacteria according to the following formulæ:

$$5C_6H_{10}O_5+24KNO_3=24KHCO_3+12N_2+6CO_2+13H_2O.$$

$$C_6H_{10}O_5+8KNO_2=4KHCO_3+2K_2CO_3+4N_2+3H_2O.$$

The destruction of cellulose under aerobic conditions also falls into two classes:

(1) If the medium is slightly alkaline, certain aerobic bacteria will play the principal rôle.

(2) If the medium is acid, then the molds and higher fungi are the active agents of destruction.

In a simple nutrient solution containing dibasic potassium phosphate, potassium nitrate, and filter paper inoculated with a cubic centimeter of canal water and kept at 35° C., the process started in 6 days, and in about 15 days all of the nitrate added and all of the nitrite formed in the early stages of fermentation had disappeared. Analysis of the gas produced showed only carbon dioxid and nitrogen, with no trace of hydrogen or methane. Thus, the process here would seem to be entirely different from the processes resulting in the production of hydrogen or methane.

For the study of cellulose destruction by aerobic bacteria the following solution was prepared:

Tap water	100. 00 c. c.
Paper	2. 00 grams.
Ammonium chlorid [1]	. 10 gram.
Potassium phosphate (diabasic)	. 05 gram.
Calcium carbonate	2. 00 grams.

After inoculating with sewer slime the fermentation starts in five to six days, after which it goes on very rapidly. The fermenting paper was found to contain a variety of forms, among which was a very small bacillus frequently associated with a large micrococcus. The former was believed to be the active agent of fermentation and was given the name of *Bacillus ferruginus*. The micrococcus is described as having no cellulose-dissolving power in itself but as stimulating *B. ferruginus* to greater activity when associated with it.

Experiments by Macfadyen and Blaxall with thermophilic bacteria in 1899 showed that these organisms may be very active destroyers of cellulose. A nutrient solution containing pure filter paper was inoculated with a small quantity of soil and incubated at 60° C. The result was an active development of gas and odor, and in 10 to 14 days the filter paper was completely broken up. The experiments were repeated with filter paper and also with films of cellulose hydrate obtained from the solution of cotton fiber in the form of thiocarbonate.

[1] Potassium nitrite, potassium nitrate, peptone, or magnesium ammonium phosphate may replace the ammonium chlorid.

The following results were reported on the solutions which showed a disintegration of cellulose by the thermophilic organisms:

(1) No reduction CuO in original or after boiling with acid.

(2) No other evidence of any proximate products of resolution, i. e., carbohydrates of dimensions NC_6.

(3) On distillation 25 c. c. gave volatile acid=1 c. c. of normal NaOH, containing acetic and butyric acids. Residue gave traces only of furfurol on distillation with HCl (1.06 s. g.).

It appears that the destruction has been, for the most part, complete, probably to CO_2 and CH_4. On further investigation you may be able to get an intermediate stage or an organism acting less severely.

All the above results were brought about by mixtures of thermophilic bacteria occurring naturally in the soil, and the action appeared to be of a symbiotic nature. Their action resulted in a complete disintegration of filter paper, fibrous cellulose, and esparto cellulose.

Distaso in 1911 described an organism isolated from the intestinal flora of the chicken, to which he gave the name *Bacillus cellulosae desagregans,* because it was found to be capable of destroying cellulose. When cultivated in mineral solutions with pure cellulose (Berzelius filter paper) the paper is disintegrated, forming flocci or fibers; action never goes further and this author is not sure whether the filter paper is thoroughly transformed. The organism does not stain by Gram's method; forms oval subterminal spores; is aerobic in nature; grows in sugar gelatin; never gives off gas; grows well at 37° but not at 22° C.; produces no indol; grows only feebly on glucose; does not assimilate lactose, maltose, or saccharose, but transforms starch into glucose rapidly.

In 1911 Choukevitch in a study on the bacterial flora of the large intestine of the horse always obtained a fermentation of cellulose in Omelianski's nutrient solution when inoculated with several loopfuls of the intestinal contents. In the fermenting solutions a small organism (*Bacillus gasogenous*) was always found, which morphologically resembled the hydrogen and methane ferments described by Omelianski. Neither a pure culture of this organism nor any of the others which he isolated from the intestine of the horse was able to ferment cellulose.

The most recent contribution to our knowledge of cellulose destruction is that of Kellerman and McBeth, whose report is a preliminary one of work undertaken by the Office of Soil-Bacteriology Investigations. In this report special attention is given to that portion of Omelianski's work from which he concluded that cellulose undergoes either a hydrogen or methane fermentation. The impurity of Omelianski's cultures are discussed, and three new species of cellulose ferments isolated from his cultures (*Bacillus rossica*, *B. amylolyticus*, and *Bacterium flavigena*) are described.

INVESTIGATIONS WITH FILAMENTOUS FUNGI.

Early experiments with parasitic fungi indicated that many of these organisms were able to make their way into plant tissues by piercing the cell membrane. Such observations were made by Kühn in his study of blight-producing fungi, by the brothers Tulasne in their study of the rusts, and by De Bary in studies with Peronospora. Some years later Marshall Ward was able to watch the penetration of the cellulose walls of the lily bulb by parasitic fungi. The walls became swollen and evidently somewhat softened, which condition he believed to be due to the production of a ferment drop at the tip of the hyphæ. Miyoshi has recently observed a similar phenomenon with *Penicillium glaucum, Botrytis bassiani,* and *Botrytis tenula.* The power of fungi to destroy cellulose was also early suggested by Hartig in his studies on the destruction of woody tissues.

Later, the destruction of cellulose by fungi was observed in studies undertaken primarily to determine the causative agent of some common plant diseases. In this connection may be mentioned the work of DeBary with *Sclerotina libertiane,* Kissling in his biological studies of *Botrytis cinerea,* and Behrens with diseases of fruits.

The work of Van Iterson on the destruction of cellulose by fungi deserves special mention, as it gave the first indication of the extent of cellulose destruction by fungi. He was also the first to devise a method for isolating these organisms. To that purpose two sterile sheets of pure filter paper were placed in a Petri dish and moistened with the following solution:

Tap water	100.00 c. c.
Ammonium nitrate	.05 gram.
Potassium phosphate (monobasic)	.50 gram.

For inoculating material, soil or humus was used; however, the best results were obtained by exposing the dish to the air 12 hours and then cultivating at 24° C., taking care that the paper remained moist. After two or three weeks the paper was covered by a rich mold growth including a large number of species; among them were several species seldom found on malt gelatin, and on further study several of these were found to be active cellulose destroyers. The great abundance of these mold spores in the atmosphere was shown by the following experiment.

A Petri dish having a surface of 275 centimeters and containing filter paper moistened with the solution previously described was allowed to stand open in the garden 12 hours; 152 mold colonies developed, comprising 35 different species. It is evident from this experiment that large numbers of cellulose-destroying mold spores were floating in the air. The fungi found growing upon the paper were purified by means of malt gelatin. The destruction of cellulose by pure cultures was then studied by inoculating sterile nutrient solutions containing pure filter paper prepared as previously described. In this work the following 15 species were isolated and described: *Sordaria humicola* Oud., *Pyronema confluens* Tul., *Chaetomium Kunzeanum* Zopf., *Pyrenochaeta humicola* Oud., *Chaetomella horrida* Oud., *Trichocladium asperum* Harz., *Stachybotrys alternans* Oud., *Sporotrichum bombycinum* (Corda) Rabh., *Sporotrichum roseolum* Oud. and Beijer., *Sporotrichum griseolum* Oud., *Botrytis vulgaris* Fr., *Mycogone puccinioïdes* (Preuss) Sacc., *Stemphylium macrosporoïdeum* (B. en Br.) Sacc., *Cladosporium herbarum* (Pers.) Link., and *Epicco-cum purpurascens* Ehrenb.

Three years later the work of Appel appeared, which showed that certain forms of Fusarium can destroy cellulose with great rapidity. Ten grams of pure, dry filter paper were introduced into an Erlenmeyer flask, and 50 cubic centimeters of a nutrient solution containing potassium phosphate, magnesium sulphate, and potassium nitrate were added. An intensive cellulose fermentation developed, resulting in the destruction of 80 per cent of the paper in 14 days. Since all species of Fusarium can not use cellulose as a source of energy, the writer believes this fact can be made a valuable point in identifying the species of Fusarium.

In a series of experiments with a comparatively large number of molds Schellenberg showed that the destruction of the hemicellulose in plant tissues is sharply separated from the destruction of real cellulose, and also that molds act differently toward the hemicellulose of different plants. This difference in action is thought to be due to the differences in chemical composition of the plant tissues rather than to differences in solubility of the hemicelluloses. The results of this work are summarized in Table V.

TABLE V.—*Action of molds on real celluloses and hemicelluloses from various plants.*

Name of plant.	Cellulose from—		Hemicellulose from—				
	Cotton.	Hemp.	Molinia.	Lupinus.	Ruscus.	Phoenix.	Impatiens.
Mucor racemosus	−	−	+	−	−	−	−
Mucor neglectus	−	−	−	+	−	−	+
Mucor piriforme	−	−	−	+	−	−	+
Mucor globosus	−	−	+	+	−	−	+
Thamncidium elegans	−	−	−	+	−	−	+
Rhizopus nigricans	−	−	−	+	−	−	+
Penicillium glaucum (1)	−	−	−	−	−	−	+
Penicillium glaucum (2)	−	−	−	+	−	+	+
Sclerotinia fructigena	−	−	−	+	−	−	−
Sclerotinia cinerea	−	−	−	+	−	−	−
Botrytis cinerea	−	−	−	+	−	−	+
Botrytis vulgaris	−	−	+	+	−	−	+
Nectria cinnabarina	−	−	+	+	−	−	−
Cladosporium herbarum	−	−	+	+	−	−	+
Colletotrichum lindemuthianum	−	−	+	+	−	−	+
Trichothecium roseum	−	−	+	+	+	+	+

The recent work of Marshall Ward suggests the importance of Penicillium as a wood-destroying fungus. Spores of a pure culture of Penicillium were sown on sterile blocks of spruce wood; the mold grew freely and developed large quantities of spores on normal conidiophores. Sections of the wood showed that the hyphæ had entered the starch-bearing cells of the medullary rays of the sap-wood and consumed the whole of the starch. In cultures three months old the hyphæ were found deep in the woody tissue passing from tracheid to tracheid via the border pits. In conclusion Ward says:

> It certainly looks as if Penicillium may be a much more active organism in initiating and carrying on the destruction of wood than has hitherto been supposed, and that it is not merely a hanger-on or follower of more powerful wood-destroying fungi.

Bourquelot has shown the great versatility of Aspergillus in the production of enzymes, having found it capable of producing invertase, maltase, trehalase, emulsin, inulase, diastase, and trypsin, and Bertrand and Holderer have found that it also produces cellase. Ward suggests that Penicillium may be equally rich in the capacity for enzyme production.

Among the higher fungi Schornstein found that *Poria vaillanti*, *Polyporus vaporarius*, *Polyporus destructor*, *Coniophora cerebella*, and *Paxillus panuoides* are capable of destroying wood, which, as is well known, is largely composed of cellulose. *Polyporus destructor* quickly forms fruiting bodies and never entirely destroys the wood. *Murulius lacrymans* and *M. pulverulentus* appeared on wood soon after it had been built into position and entirely destroyed it.

Arzberger in an investigation of the fungus which causes root tubercles on Ceanothus and Eleagnus found that it belonged to the genus Frankia and secreted an enzyme that destroyed the cell walls.

METHODS AND CULTURE MEDIA EMPLOYED.

In taking up the study of cellulose fermentation the elective cultural method employed by Omelianski and the method of Van Iterson of using sheets of filter paper were tried under both aerobic and anaerobic conditions. Microscopic examinations of the cultures kept under anaerobic conditions showed the presence of organisms similar to the hydrogen and methane ferments of Omelianski. In young cultures these organisms appeared only in small numbers, but became very numerous as the decomposition process advanced. In cultures kept until the paper had been completely destroyed, the spores of this organism became extremely numerous and microscopically the cultures appeared to be almost pure, but the presence of many other species was easily demonstrated by plating on ordinary media. The cultures kept under aerobic conditions showed no organisms resembling the hydrogen or methane ferments which were so numerous in the anaerobic cultures, although the inoculum used was the same. The decomposition of the paper, however, was most rapid in the aerobic flasks. This was in accordance with Van Iterson's observations that cellulose may be rapidly decomposed by aerobic organisms. Plates from these cultures, like those grown anaerobically, showed the presence of several species of bacteria, even after numerous transfers. Our failure with these methods, together with Omelianski's admission of the impurity of his cultures after the most painstaking care to purify them, led us to believe that no accurate knowledge of cellulose fermentation could be obtained until a satisfactory plating medium had been secured.

In taking up the question of a suitable medium, a large number of nutrient solutions were first studied, including beef broth, decoctions of plant tissues, soil extracts, manure extracts, and numerous synthetic solutions. The following solution was finally adopted as giving the best results:

Potassium phosphate (dibasic)	1 gram.
Magnesium sulphate	1 gram.
Sodium carbonate	1 gram.
Ammonium sulphate	2 grams.
Calcium carbonate	2 grams.
Tap water	1,000 grams.

One hundred cubic centimeters of this solution is poured into a 200-cubic-centimeter Erlenmeyer flask and a single sheet of filter paper 10 centimeters in diameter folded so as to make a quarter circle is dropped into the solution. The paper should be in a loose fold, with the upper edge just beneath the surface of the solution. If the paper is not entirely immersed, the flask should be shaken until none of the paper is exposed. This precaution is necessary in order to keep down the mold growth which invariably occurs on the exposed paper. The flask is then plugged with cotton, sterilized, cooled, and inoculated with a very small quantity of the material under study and then gently shaken to insure an even distribution of the inoculum. The flasks are incubated at 30° C. until the fermentation process is well started, which generally takes from 5 to 10 days. The first signs of fermentation are usually indicated by the clouding of the solution, and a little later the paper takes on a dull, frayed appearance. A small quantity of the paper is now removed and placed in a control flask containing a small piece of sterile paper. If on agitation the paper from the inoculated flask is broken up more readily than that in the control the proper time for transfer has been reached. A small quantity of the paper is then removed by means of a small platinum spatula, placed in a second sterile flask, and the flask agitated until the paper used as an inoculum is well broken up and distributed. Since the object of this transfer is to increase the number of cellulose ferments and at the same time crowd out the foreign species, it is important that the transfer be made as soon as the cellulose ferments have reached a high stage of development and before the by-products have become sufficiently abundant to allow a rapid development of foreign species. After three or four transfers a small piece of the paper is introduced into a flask containing sterile water and agitated until completely broken up. The necessary dilutions are then prepared and cellulose agar plates poured and kept at 30° C. in a moist chamber, which keeps the agar moist an indefinite time and is indispensable with slow-developing colonies. A convenient type of moist chamber may be prepared by inverting a large beaker over a shallow dish filled with water.

In many instances the cellulose ferments have cleared a well-defined zone in five or six days, but others may require three weeks or longer; however, the rapidity with which the cellulose agar is cleared is not always a safe index of the vigor of the organism, as some species which dissolve cellulose very slowly on solid media are very active in cellulose solutions.

It is seldom possible to secure a pure culture from the first set of plates, as even where the cellulose-fermenting colony is well separated from all other apparent growth it is often found to contain two or more species. To obtain a pure culture, all colonies are replated on cellulose agar before further study, and the replating is continued until only one type of colony appears on the plates and slides prepared from these colonies show the presence of only one kind of organism. Since the colonies on cellulose agar do not ordinarily form raised growths, it is often necessary to lift out a small piece of the agar containing the colony by means of a small platinum spatula. This spatula is also used to macerate the inoculum when it is introduced into the dilution flask. If a soft agar is used, the maceration can be accomplished readily by rubbing the agar against the side of the flask just above the surface of the water. An even distribution can then be effected by shaking.

Four varieties of special culture media have been found useful in studying the cellulose ferments. Their composition and method of preparation will now be described.

CELLULOSE AGAR.

Prepare one liter of a dilute ammonium-hydroxid solution by adding 3 parts water to 10 parts ammonium hydroxid, sp. gr. 0.90. Add a slight excess of copper carbonate and shake vigorously, allow to stand overnight, and then siphon off the supernatant solution.

Add 15 grams of unwashed sheet filter paper and shake occasionally until the paper is dissolved. Dilute to 10 liters and add slowly a 1 to 5 solution of hydrochloric acid, with vigorous shaking until the precipitation of the cellulose is complete. Dilute to 20 liters, allow the cellulose to settle, and decant the supernatant liquid. Wash by repeated changes of water, adding hydrochloric acid each time until the copper color disappers; then wash with water alone until the solution is free from chlorin. Allow it to settle several days and decant off as much of the clear solution as possible. If the percentage of cellulose is still too low, a portion of the solution is centrifugalized to bring the cellulose content up to 1 per cent.

Cellulose solution		500 c. c.
Agar		10 grams.
Nutrient solution, composed of—		500 c. c.
Potassium phosphate (dibasic)	1 gram.	
Magnesium sulphate	1 gram.	
Sodium chlorid	1 gram.	
Ammonium sulphate	2 grams.	
Calcium carbonate	2 grams.	
Tap water	1, 000 c. c.	

STARCH AGAR.

To 800 cubic centimeters of boiling water add 10 grams of potato starch suspended in a little cold water. Concentrate by boiling to 500 cubic centimeters. This breaks up the starch grains and should give a nearly transparent starch solution.

Starch solution	500 c. c.
Agar	10 grams.
Nutrient solution (same as for cellulose agar)	500 c. c.

POTATO AGAR.

Pare, steam, and mash a quantity of potatoes. To 100 grams of mashed potato add 800 cubic centimeters of tap water and steam for one-half hour; filter through cotton.

Potato solution	500 c. c.
Agar	15 grams.
Nutrient solution (same as for cellulose agar)	500 c. c.

DEXTROSE AGAR.

Dextrose	10 grams.
Agar	15 grams.
Tap water	500 c. c.
Nutrient solution (same as for cellulose agar)	500 c. c.

THE OCCURRENCE AND GENERAL CHARACTERISTICS OF CELLULOSE-DESTROYING BACTERIA IN NATURE.

Preliminary examinations of sewer slime, of manures, and of the soils of the United States for cellulose-dissolving bacteria indicate that all decaying plant tissues are infested with these organisms and that numerous species occur, many of which have a wide distribution. Several species have been isolated again and again from widely separated regions. For instance, *Bacterium flavigena*, first isolated from Omelianski's hydrogen culture, has since been isolated from soils received from Hays, Kans., Arlington, Va., and Ponchatoula, La., while *Bacillus cytaseus* has been isolated from the soils of Jefferson, Me., Norfolk, Va., Rome, Ga., Alpena, Mich., and Riverside, Cal. All soil samples tested have been found to contain at least 1 species of cellulose-destroying bacteria, and as many as 3 species have been isolated from some. From the 12 soil samples examined 11 species have been isolated; in addition to these, 4 other species have been isolated from other sources, 1 of which belongs to the thermophile group. All soils examined have been found to contain thermophile bacteria capable of destroying cellulose very rapidly at 60° C. These organisms are especially abundant in manure heaps, and must play an important rôle in fermenting manures when high temperatures are reached.

In our investigations 15 species of cellulose-destroying bacteria have been isolated, all of which are morphologically and physiologically different from Omelianski's hydrogen and methane ferments. All of the organisms isolated grow upon ordinary gelatin, which in many cases is liquefied; however, gelatin is not a satisfactory medium for isolating these organisms, as it is in no way selective and some species soon lose their power to dissolve cellulose when grown upon it. Four species isolated from the soils of Utah grew more vigorously on gelatin than on any other solid medium, but after a few transfers on this medium the power to dissolve cellulose was entirely lost. This loss of cellulose-destroying power has also been noted with other species after several transfers on artificial media and is especially true of the thermophile organisms, which frequently lose their power to destroy cellulose after the first transfer on cellulose agar, although they continue to grow vigorously on ordinary media. All attempts to reestablish the normal activity of these organisms toward cellulose have been unsuccessful. No detailed descriptions of these species will be given, therefore, until their power to dissolve cellulose can be reestablished or they can be reisolated, and the descriptions written from cultures which act normally on cellulose agar.

After many trials to determine the rapidity of growth on the solid media used, we have found it advisable to give descriptions after 5, 10, and 15 days' incubation, the cellulose agar, potato slants, and gelatin stabs requiring the longer period. The quantity of acid produced by the different organisms in beef broth containing carbohydrates was determined according to the standard methods after incubation periods of 1, 2, and 3 days, but the quantities of acid produced during such short periods were too small to be of value; the titrations were therefore made after 6 and 12 days. The sugars, starch, and higher alcohols used were all broken down, with the production of more or less acid, with four exceptions: *Bacterium fimi* and *Bacterium liquatum* gave no acid in mannite, *Bacillus cytaseus* made no growth, and *Bacillus rossica*[1] gave an alkaline reaction. All except two of the organisms grew in litmus milk, most of which gave a faintly acid reaction in 2 days. Some of the species produced ammonia in Dunham's solution. In some cases nitrate was reduced to nitrite, but only one species carried the nitrite over to ammonia. Three of the organisms formed indol.

DESCRIPTIONS OF CELLULOSE-DESTROYING BACTERIA.

In describing the cultural characteristics of the cellulose-destroying bacteria our object has been to simplify the descriptions as much as possible by selecting media which would show the salient characteristics of the bacteria, and by using only those characters which remained fairly constant through three successive sets of cultures. In following this plan we have left out many data called for by the Society of American Bacteriologists, but the power to destroy cellulose is a test that places these organisms in a group by themselves, and it is believed the data given are sufficient for the identification of the organisms described. However, the descriptions are recognized as not entirely satisfactory, and as the study of the cultural characteristics of these and other cellulose-destroying organisms is continued it may be found advisable to add to the data now given.

BACTERIUM FIMI, N. SP.

(Pl. II, figs. 1, 2, and 3.)

I. *Morphology.*
  1. Vegetative cells from 24-hour cultures at 30° C.
     Beef agar, average length 0.9 μ, maximum length 1.5 μ, width 0.4 μ. Dextrose agar, average length 1.4 μ, maximum length 2.8 μ, width 0.6 μ.
  2. No endospores.
  3. Staining reactions: Methylene blue +, carbol fuchsin +, Gram −.

II. *Cultural features.*
  1. Agar strokes, 10 days. General characteristics: Glistening, smooth, moist growth, white to vitreous.
     Beef agar: Abundant, faint yellow.
     Potato agar: Abundant, iridescent.
     Dextrose agar: Moderate, iridescent.
     Starch agar: Abundant.
     Cellulose agar: Moderate.

[1] Described in Centralblatt für Bakteriologie [etc.], pt. 2, vol. 34, 1912, p. 492.

II. *Cultural features*—Continued.

2. Potato: After 15 days, moderate, glistening, smooth, cream colored; potato not colored.
3. Agar stab: Growth best at top, echinulate.
4. Gelatin stab: After 15 days liquefaction infundibuliform, becoming saccate in 50 days.
5. Beef broth: Slight clouding.
6. Litmus milk: Faintly acid in 2 days.
7. Plate cultures—

Cellulose agar, 15 days.

Form: Surface and bottom, round; embedded, lenticular or irregular.

Size: Surface and bottom, 1 to 4 mm.; embedded 0.7 to 1.8 mm. on major axis.

Enzymic zone: 0.3 to 1.6 mm.

Elevation: Raised.

Topography: Smooth.

Consistency: Slimy.

Chromogenesis: Surface and bottom, reflected light, gray with yellowish or whitish nucleus; transmitted light, brown. At angle of 45° by transmitted light bottom colonies show interior of colony bluish or iridescent and white ring around border. Embedded, reflected light, white; transmitted light, opaque.

Internal structure: Surface and bottom granular with opaque to translucent granular nucleus, and frequently having finely granular ring at border; embedded, opaque, often with numerous outgrowths.

Edge: Entire to undulate.

Potato agar, 5 days.

Form: Surface and bottom, round; embedded, lenticular to triangular.

Size: Surface and bottom, 1 to 6 mm.; embedded, 0.5 to 1.5 mm.

Elevation: Convex.

Topography: Smooth.

Consistency: Slimy.

Chromogenesis: Surface, reflected light, glistening grayish white; transmitted light, opaque or translucent brownish gray, often with opaque nucleus. Embedded, reflected light, white; transmitted light, opaque. Bottom, reflected light, gray; transmitted light, light brown, at 45° bluish and often iridescent.

Internal structure: Surface, homogenous, opaque or finely granular, often with lenticular or round nucleus; embedded, opaque, sometimes with translucent irregular, finely granular outgrowths; bottom, homogenous, finely granular, frequently with granular nucleus.

Edge: Entire.

Beef agar, 5 days.

Form: Surface and bottom, round; embedded, lenticular.

Size: Surface and bottom, 1 to 4 mm.; embedded, 0.6 to 1.5 mm. on major axis.

Elevation: Convex.

Topography: Smooth.

Consistency: Slimy.

Chromogenesis: surface and bottom, reflected light, yellowish or grayish white; transmitted light, translucent brown; at 45° iridescent ring at border. Embedded, reflected light, yellowish or grayish white; transmitted light, opaque.

Odor: None.

II. *Cultural features*—Continued.

7. Plate cultures—Continued.

Beef agar, 5 days—Continued.

Internal structure: Surface, granular, often with lenticular nucleus and finely granular and hyaline at edge; embedded, opaque; bottom, granular, sometimes with nucleus.

Edge: Entire.

Starch agar, 5 days.

Form: No surface colonies; embedded, lenticular to irregularly round; bottom, round.

Size: Embedded, 0.4 to 1.2 mm.; bottom, 1 to 1.5 mm.

Enzymic zone: 1.7 to 3 mm.

Elevation: No surface growth, but agar raised by colonies just below surface.

Chromogenesis: Embedded, reflected light, white; transmitted light, opaque. Bottom, reflected light, opalescent or white; transmitted light, barely translucent, dark gray.

Internal structure: Embedded, opaque and often irregular with outgrowths; bottom, granular, generally becoming finely granular at edge, and usually with lenticular nucleus.

Edge: Entire.

Dextrose agar, 5 days.

Form: Surface and bottom, round; embedded, lenticular.

Size: Surface and bottom, 0.8 to 1.2 mm.; embedded, 0.4 to 1 mm.

Elevation: Convex.

Topography: Smooth.

Consistency: Slimy.

Chromogenesis: Surface, reflected light, grayish white, generally with white nucleus; transmitted light, translucent brownish gray. Embedded, reflected light, white; transmitted light, opaque. Bottom, reflected light, gray; transmitted light, smoky brown.

Internal structure: Surface and bottom, finely granular, usually with round or lenticular nucleus; embedded, opaque, granular at edge.

Edge: Entire.

III. *Physical and biochemical features.*

1. Peptone water—

Test for—	Addition to peptone water.						
	Dextrose.	Lactose.	Saccharose.	Maltose.	Glycerin.	Mannite.	Starch.
Gas production	0	0	0	0	0	0	0
Acid production, 6 days	1.42	.70	1.60	1.45	.53	0	1.48
Acid production, 12 days	1.60	.90	1.63	1.43	.76	0	1.62

2. Dunham's: Ammonia trace.
3. Nitrate broth: Nitrite +; ammonia −.
4. Peptone-nitrite solution: Indol +.

BACTERIUM LIQUATUM, N. SP.

(Pl. I, figs. 1, 2, and 3.)

I. *Morphology.*

1. Vegetative cells from 24-hour cultures at 30° C. Beef agar, average length 1.7 μ, maximum length 2.6 μ, width 0.4 μ. Potato agar, average length 0.8 μ, maximum length 1.5 μ, width 0.3 μ.
2. No endospores.
3. Staining reactions: Methylene blue +; carbol fuchsin +; Gram −.

II. *Cultural features.*

1. Agar strokes, 10 days. General characteristics: Glistening, smooth, moist growths.
   Beef agar: Abundant, raised, grayish yellow; agar whitened.
   Potato agar: Abundant, flat, watery, pale grayish yellow.
   Dextrose agar: Scant, flat, watery, vitreous to pale yellow.
   Starch agar: Moderate, flat, vitreous to pale yellow.
   Cellulose agar: Moderate, convex, vitreous to pale yellow.
2. Potato: After 15 days growth abundant, glistening, smooth, brilliant canary yellow; potato not colored.
3. Agar stab: Growth best at top, papillate.
4. Gelatin stab: After 15 days liquefaction nápiform; after 50 days stratiform with liquefied gelatin present.
5. Beef broth: Moderate clouding, scant compact sediment.
6. Litmus milk: Faintly acid in two days.
7. Plate cultures—

Cellulose agar, 15 days.

Form: Surface and bottom, round; embedded, lenticular to irregularly round.

Size: Surface, 1 mm.; embedded, 1 mm. on major axis; bottom, 1.5 mm.

Enzymic zone: 0.4 to 0.75 mm. wide and slightly depressed.

Elevation: Raised or umbilicate. Embedded colonies just below surface give umbonate appearance due to depression of enzymic zone.

Topography: Smooth.

Consistency: Soft.

Chromogenesis: Surface, embedded and bottom, reflected light, faint yellowish gray; surface and bottom, transmitted light, translucent gray generally with opaque nucleus, surface sometimes showing opaque ring at edge; embedded, opaque.

Internal structure: Surface and bottom, granular, generally with round or lenticular nucleus; embedded, opaque, sometimes conglomerate.

Edge: Surface and bottom, irregular, finely granular; embedded, entire.

Starch agar, 5 days.

Form: Round.

Size: Embedded and bottom, 0.5 to 2 mm.

Enzymic zone: 1 mm.

Elevation: No surface colonies. Medium may be raised by embedded colonies just below surface.

Chromogenesis: Embedded, reflected light, white, opaque. Bottom, reflected light, light gray with whitish gray nucleus; transmitted light, gray with opaque nucleus or like embedded.

Internal structure: Embedded and bottom, central area opaque, becoming coarsely granular near edge.

Edge: Embedded, irregular, granular; bottom, blending with medium or irregularly granular.

Beef agar, 5 days.

Form: Surface and bottom, round; embedded, lenticular to round.

Size: Surface, 1.5 to 3 mm.; embedded, 0.5 to 1 mm.; bottom, 1 to 1.5 mm.

Elevation: Convex to pulvinate.

Topography: Smooth.

Consistency: Slightly viscid.

II. *Cultural features*—Continued.

7. Plate cultures—Continued.

Beef agar, 5 days—Continued.

Chromogenesis: Surface, reflected light, glistening, sebaceous; transmitted light, translucent brown, some with vitreous edge. Embedded, reflected light, sebaceous; transmitted light, opaque. Bottom, reflected light, gray; transmitted light, translucent brown.

Odor: None.

Internal structure: Surface, coarsely granular, generally showing irregular granular nucleus; embedded, opaque, often showing outgrowths in all directions; bottom, granular, with a granular nucleus.

Edge: Entire.

Potato agar, 5 days.

Form: Surface and bottom, round; embedded, lenticular.

Size: Surface, 1 to 5 mm.; embedded, 0.5 to 1.5 mm. on major axis; bottom, 1 mm., sometimes spreading to 15 mm.

Elevation: Convex.

Topography: Smooth.

Consistency: Watery.

Chromogenesis: Surface, reflected light, glistening, opalescent; transmitted light, edge bluish and iridescent. Embedded, reflected light, cream color; transmitted light, opaque. Bottom, reflected light, gray; transmitted light, vitreous, sometimes with a brownish central area.

Internal structure: Surface, granular, often almost hyaline, generally showing nucleus; embedded, granular, sometimes showing numerous outgrowths; bottom, finely granular, generally with granular nucleus and often having grumose center.

Edge: Entire.

Dextrose agar, 5 days.

Form: Surface and bottom, round; embedded, lenticular.

Size: Surface and bottom, 1 to 2 mm.; embedded, 0.5 to 0.75 mm.

Elevation: Convex.

Topography: Smooth.

Consistency: Watery.

Chromogenesis: Surface, reflected light, vitreous to gray, with whitish gray central area often showing white nucleus; transmitted light, translucent light brownish gray, with vitreous edge and opaque nucleus. Embedded, reflected light, grayish white; transmitted light, opaque; bottom, like surface, no nucleus.

Internal structure: Surface, finely granular, usually with opaque lenticular nucleus; embedded, opaque, but showing granular at edge; bottom, finely granular with small granular nucleus.

Edge: Entire.

III. *Physical and biochemical features.*

1. Peptone water—

Test for—	Addition to peptone water.						
	Dextrose.	Lactose.	Saccharose.	Maltose.	Glycerin.	Mannite.	Starch.
Gas production	0	0	0	0	0	0	0
Acid production, 6 days	1.20	.50	1.32	1.16	.08	0	1.38
Acid production, 12 days	1.27	.98	1.33	1.16	.23	0	1.40

II. *Physical and biochemical features*—Continued.

2. Dunham's: Ammonia +.
3. Nitrate broth: Nitrites +; ammonia −.
4. Peptone-nitrite solution: Indol +.

BACILLUS BIBULUS, N. SP.

(Pl. II, figs. 4, 5, and 6, and Pl. IV, figs. 1 and 2.)

I. *Morphology.*

1. Vegtative cells from 24-hour cultures at 30° C. Beef agar, average length 1.3 μ, maximum length 2.0 μ, width 0.4 μ. Dextrose agar, average length 0.8 μ, maximum length 1.4 μ, width 0.4 μ.
2. No endospores.
3. Staining reactions: Methylene blue +; carbol fuchsin +; Gram −.

II. *Cultural features.*

1. Agar strokes, 10 days. General characteristics: Glistening, smooth, moist, raised or convex growth.
   Beef agar: Abundant, grayish yellow.
   Potato agar: Abundant, grayish yellow.
   Dextrose agar: Scant, white.
   Starch agar: Scant, vitreous to gray.
   Cellulose agar: Moderate, yellowish.
2. Potato: After 15 days' growth abundant, glistening, smooth, brilliant canary yellow. Potato not colored.
3. Agar stab: Growth best at top, papillate.
4. Gelatin stab: After 15 days' line of puncture filiform, later echinulate; liquefaction crateriform. After 50 days deeply crateriform, no liquefied gelatin present.
5. Beef broth: Slight clouding, scant compact sediment.
6. Litmus milk: Faintly acid in two days.
7. Plate cultures—

   Cellulose agar, 15 days.

   Form: Surface and bottom, round; embedded, round to irregularly round.
   Size: Surface and bottom, 0.5 to 0.8 mm.; embedded, 0.3 to 0.5 mm.
   Enzymic zone: 0.3 mm. in some cases.
   Elevation: Convex.
   Topography: Smooth.
   Consistency: Soft.
   Chromogenesis: Surface and bottom, reflected and transmitted light, opalescent, usually with grayish white opaque nucleus. Embedded, reflected light, grayish or yellowish white; transmitted light, opaque.
   Internal structure: Surface granular, sometimes with clouded radial areas extending to edge of colony or with a grumose center; embedded, granular, may show lenticular mother growth with numerous outgrowths; bottom, granular, often with roundish granular nucleus, and may also be clouded.
   Edge: Irregular and granular.

   Starch agar, 5 days.

   Form: No surface growth; embedded, irregularly round to round; bottom, round.
   Size: Embedded and bottom, 0.3 to 2.5 mm.
   Enzymic zone: 1 to 2.5 mm.
   Elevation: Medium slightly raised by colonies just under the surface.
   Chromogenesis: Embedded and bottom, reflected light, white; transmitted light, translucent gray to opaque.

II. *Cultural features*—Continued.

7. Plate cultures—Continued.

Starch agar, 5 days—Continued.

Internal structure: Embedded, narrow finely granular zone around edge, remainder opaque; bottom, finely granular zone at edge wider than embedded; remainder opaque.

Edge: Embedded, entire to irregularly granular; bottom, blending with medium.

Dextrose agar, 5 days.

Form: Surface and bottom, round; embedded, lenticular.

Size: Surface and embedded, 0.4 to 0.6 mm.; bottom, 0.5 to 1 mm.

Elevation: Convex.

Topography: Smooth.

Consistency: Slimy.

Chromogenesis: Surface, reflected light, white to faint yellowish gray; transmitted light, barely translucent, dark brown. Embedded, reflected light, like surface; transmitted light, opaque; bottom, reflected light, opalescent.

Internal structure: Surface, finely granular, with an opaque round or granular nucleus; embedded, opaque; bottom, finely granular, sometimes with nucleus.

Edge: Entire.

Beef agar, 5 days.

Form: Surface and bottom, round; embedded, lenticular.

Size: Surface and bottom, 1 to 2 mm.; embedded, 0.5 to 1 mm. on major axis.

Elevation: Convex.

Topography: Smooth.

Consistency: Soft.

Chromogenesis: Surface, reflected light, light yellow; transmitted light, translucent brownish yellow, may have opaque nucleus. Embedded reflected light, yellow to yellowish gray; transmitted light, opaque. Bottom, reflected light, gray; transmitted light, translucent smoky brown.

Internal structure: Surface, granular, with dark round or lenticular nucleus, sometimes with hyalin edge; embedded, granular, occasionally with outgrowths in one or two planes; bottom, granular, usually with small granular nucleus, and often with edgelike surface.

Odor: None.

Edge: Entire.

Potato agar, 5 days.

Form: Surface and bottom, round; embedded, lenticular.

Size: Surface and bottom, 0.5 to 1.5 mm.; embedded, 0.5 to 0.8 mm. on major axis.

Elevation: Pulvinate.

Topography: Smooth.

Consistency: Soft.

Chromogenesis: Surface, reflected light, yellowish gray; transmitted light, barely translucent brown. Embedded, reflected light, yellow; translucent light, opaque. Bottom, reflected light, gray; transmitted light, translucent brown.

Internal structure: Surface, finely granular, generally with round or lenticular opaque or granular nucleus, often with hyalin edge; embedded, opaque; bottom, like surface except nucleus, which if present is small, round, and granular.

Edge: Entire.

III. *Physical and biochemical features.*

1. Peptone water—

Test for—	Addition to peptone water.						
	Dextrose.	Lactose.	Saccharose.	Maltose.	Glycerin.	Mannite.	Starch.
Gas production	0	0	0	0	0	0	0
Acid production, 6 days	1.75	1.20	1.57	1.22	.15	.75	1.90
Acid production, 12 days	1.85	1.28	1.50	1.47	.35	1.20	2.07

2. Dunham's: Ammonia +.
3. Nitrate broth: Nitrites −; ammonia −.
4. Peptone-nitrite solution: Indol, trace.

PSEUDOMONAS SUBCRETUS, N. SP.

(Pl. I, figs. 4, 5, and 6.)

I. *Morphology.*

1. Vegetative cells: Starch agar, 24 hours at 30° C., average length 1.2 μ. Maximum length 1.4 μ, width 0.3 μ. Beef agar, 48 hours at 30° C. (no growth in 24 hours), average length 1.4 μ, maximum length 3.0 μ, width 0.4 μ.
2. No endospores.
3. Staining reactions: Methylene blue +; carbol fuchsin +; gram −.

II. *Cultural features.*

1. Agar strokes, 10 days. General characteristics; glistening, smooth, moist, vitreous to faint yellow.
   Beef agar: Moderate, flat.
   Potato agar: No growth.
   Dextrose sugar: Scant.
   Starch agar: Moderate.
   Cellulose agar: No surface growth. Moderate, generally faint yellow growth in medium, area of growth sunken.
2. Potato: After 15 days' growth scanty, concave due to slight liquefaction of potato, white to faint yellow. Potato bleached around growth.
3. Agar stab: Growth best at top, papillate.
4. Gelatin stab: After 15 days filiform, no liquefaction.
5. Beef broth: No growth.
6. Litmus milk: No growth.
7. Plate cultures—
   Cellulose agar, 15 days.
      Form: Round.
      Size: Average 3 mm.
      Enzymic zone: Enzyme acts within the colony; older colonies may show narrow clear zone about 0.3 mm. wide.
      Elevation: Medium concave, no surface growth.
      Chromogenesis: Reflected light, transparent gray or yellowish gray; transmitted light, smoky brown.
      Internal structure: Central area clouded light brown, then a zone like the medium but less dense, sometimes surrounded by a denser border line which may be broken.
      Edge: Entire to undulate.
   Starch agar, 5 days.
      Form: Round.
      Size: Surface and bottom, 0.3 to 1.5 mm.; embedded, 0.5 to 0.7 mm.
      Enzymic zone: 2 to 4 mm.
      Elevation: Convex.

II. *Cultural features*—Continued.

7. Plate cultures—Continued.

Starch agar, 5 days—Continued.

Topography: Smooth.

Consistency: Very soft.

Chromogenesis: Surface, reflected light, yellowish white; transmitted light, opaque nucleus surrounded by narrow or wide translucent yellow zone. Embedded, reflected light, yellowish white; transmitted light, opaque. Bottom, reflected light, gray with grayish white nucleus; transmitted light, translucent yellowish gray with opaque nucleus.

Internal structure: Surface, granular brownish nucleus surrounded by finely granular light-brown zone often almost hyalin at edge; embedded, opaque with narrow, finely granular edge; bottom, opaque or nearly opaque, brown, granular central area surrounded by light-brown granular zone, often with narrow lighter ring at edge, the latter often finely granular.

Edge: Surface and bottom, entire; embedded, irregularly granular or shading off into medium.

Dextrose agar, 5 days.

Form: Surface and bottom, round; embedded, lenticular.

Size: Surface and bottom, 1 to 1.3 mm.; embedded, 0.4 mm.

Elevation: Convex.

Topography: Smooth.

Consistency: Soft.

Chromogenesis: Surface, reflected light, yellowish white; transmitted light, translucent light brown. Embedded, reflected light, yellowish white; transmitted, opaque. Bottom, reflected light, gray; transmitted light, translucent light brown.

Internal structure: Surface, finely granular, generally with small granular nucleus. Embedded, coarsely granular; bottom, central granular area usually becoming finely granular around the edge and having small granular nucleus.

Edge: Entire.

Beef agar, 5 days.

Form: Irregularly round.

Size: Surface and bottom, 0.7 to 1.4 mm.; embedded, 0.4 to 0.7 mm.

Elevation: Convex, often arising from depression in agar.

Topography: Smooth.

Consistency: Soft.

Odor: None.

Chromogenesis: Surface, reflected light, faint yellowish gray; transmitted light, translucent light brown. Embedded, reflected light, faint yellowish gray; transmitted light, opaque. Bottom, transmitted light, light brown generally with small opaque nucleus and ring near edge.

Internal structure: Surface, finely granular, with round or oval dark-brown barely translucent nucleus; sometimes growth so dense nucleus can not be distinguished. Embedded, large central area opaque or occasionally translucent with narrow, translucent, finely granular edge. Bottom, granular, usually almost opaque at center.

Edge: Surface and bottom, entire or granular and blending with medium; embedded, entire.

Potato agar, 5 days.

No growth.

III. *Physical and biochemical features.*

1. Peptone water—

Test for—	Addition to peptone water.						
	Dextrose.	Lactose.	Saccharose.	Maltose.	Glycerin.	Mannite.	Starch.
Gas production	0	0	0	0	0	0	0
Acid production, 6 days	.35	.22	.05	.52	0	0	.60
Acid production, 12 days	.57	.53	.05	.52	0	0	.60

2. Dunham's: Ammonia −.
3. Nitrate broth: Nitrite trace; ammonia −.
4. Peptone-nitrite solution: Indol −.

BACILLUS CYTASEUS, N. SP.

(Pl. III, figs. 1, 2, 3, 4, and 5, and Pl. IV, figs. 3 and 4.)

I. *Morphology.*

1. Vegetative cells from 24-hour cultures at 30° C. Cellulose agar, average length 1.8 μ, maximum length 3.4 μ, width 0.2 μ. Potato agar, average length 2.7 μ, maximum length, 5.0 μ, width 0.5 μ.
2. Endospores: Form elliptical; limits of size, 1.4 μ by 0.7 to 2.6 μ by 1 μ; size of majority, 2μ by 0.9 μ. Spores withstand 80° C. for 15 minutes.
3. Involution forms on potato agar in 2 days at 30° C.
4. Staining reactions: Methylene blue +; carbol fuchsin +; gram −.

II. *Cultural features.*

1. Agar strokes, 10 days. General characteristics: Flat, vitreous to faint gray growth.
   Beef agar. No growth.
   Potato agar: Abundant, glistening.
   Dextrose agar: Abundant, dry, with oily luster.
   Starch agar: Abundant, glistening.
   Cellulose agar: No raised growth. Moderate growth under surface with shallow, clear stratum below.
2. Potato: After 15 days no growth.
3. Agar stab: Echinulate.
4. Gelatin stab: After 15 days very scant growth on surface with few isolated beads just below the surface.
5. Beef broth: No growth.
6. Litmus milk: No growth.
7. Plate cultures—
   Cellulose agar, 15 days.
      Form: Surface and bottom, irregularly round; embedded, round.
      Size: Surface, 1.5 to 5 mm.; embedded, 0.5 to 0.8 mm.; bottom, 12 to 25 mm.
      Enzymic zone: 0.5 to 1 mm.
      Elevation: Flat.
      Topography: Smooth.
      Consistency: Soft.
      Chromogenesis: Surface, reflected light, light gray; transmitted light, transparent gray. Embedded, reflected light, gray, often with narrow white border and very small nucleuslike border; transmitted light, translucent brownish gray with small, almost opaque nucleus. Bottom, reflected and transmitted light, vitreous.

II. *Cultural features*—Continued.
7. Plate cultures—Continued.
Cellulose agar, 15 days—Continued.
Internal structure: Surface, homogeneous, granular, with small raised nucleus; embedded, granular, often may be coarsely granular around edge; bottom granular, generally no nucleus, and often with faint, irregular, clouded areas.
Edge: Surface, lobate; embedded, regular and granular; bottom, regular and granular, or blending with medium.
Potato agar, 5 days.
Form: Surface and bottom, irregular; embedded, lenticular.
Size: Surface and bottom, 6 to 30 mm.; embedded, 0.2 to 0.4 mm. on major axis.
Elevation: Flat.
Topography: Beaded.
Consistency: Soft.
Chromogenesis: Surface and bottom, vitreous, often with whitish gray edge; embedded, reflected light, white; transmitted light, opaque.
Internal structure: Surface, a spreading colony of granular beads becoming denser and fusing at edge; embedded, granular; bottom, spreading, granular.
Edge: Surface, irregular, beaded; embedded, entire; bottom, irregular often ameboid.
Starch agar, 5 days.
Form: No surface growth; embedded, round; bottom, round or irregularly round.
Size: Embedded, 1 to 7 mm.; bottom, 3 to 10 mm.
Enzymic zone: 2 to 5 mm.
Chromogenesis: Embedded, reflected light, whitish gray, generally with concentric whitish rings near border; transmitted light, translucent, faint, brownish gray. Bottom, reflected light, faint gray or vitreous; transmitted light, faint brown or vitreous.
Internal structure: Embedded, granular, generally more coarsely granular at edge, often with rings near border; bottom, finely granular, with growth generally denser around edge and often with small granular nucleus.
Edge: Embedded, irregularly granular; bottom, granular, sometimes auriculate to fimbriate.
Dextrose agar, 5 days.
Punctiform colonies, growth not typical.
Beef agar, 5 days.
Colonies very small, growth not typical.
III. *Physical and biochemical features.*
1. Peptone water—

Test for	Addition to peptone water.						
	Dextrose.	Lactose.	Saccharose.	Maltose.	Glycerin.	Mannite.	Starch.
Gas production	0	0	0	0	0	0	0
Acid production, 6 days	0	0	0	0	0	0	0
Acid production, 12 days	0	0	0	0	0	0	0

2. Dunham's: Ammonia –.
3. Nitrate broth: Nitrites –; ammonia –.
4. Peptone-nitrite solution: Indol –.

CELLULOSE DESTRUCTION BY FILAMENTOUS FUNGI.

From early studies of cellulose fermentation in our laboratory of soil bacteriology it became apparent that many species of filamentous fungi are capable of destroying cellulose very rapidly. This was first indicated by a series of experiments designed primarily to test the cellulose-dissolving power of various soils and also to study the value of cellulose as a source of energy for nitrogen-fixing organisms. Pure filter paper was cut into small squares, approximately 2 millimeters, and evenly distributed through the soil; 200-gram samples of soil were used, to which 2 per cent of paper was added; as much water as the soil would hold without becoming sticky was then added, and after a thorough mixing the samples were held at 30° C. The small pieces of paper soon became covered with mold growths, including many species. In plating out from these soils on cellulose agar after 30 days' incubation, in one instance as many as 200,000,000 mold colonies were obtained from a gram of dry soil. The check sample at the beginning of the experiment gave less than 20,000 mold colonies, and after 30 days' incubation the number had increased to 100,000. Thus, it appears that the sample to which cellulose had been added contained 2,000 times as many mold spores capable of growing on cellulose agar as the check sample. It is therefore obvious that cellulose either directly or indirectly stimulated the development of molds in the soil.

This stimulation might be explained on the theory that the molds were able to utilize the by-products of cellulose decomposition caused by the activity of bacteria; but on examining the mold colonies on the cellulose-agar plates it was found that about 40 per cent of them had cleared up a well-defined zone around the colony, thus showing their cellulose-dissolving power, while a careful search failed to reveal a single cellulose-dissolving bacterial colony. It therefore appears that the great development in the mold flora was due, in part at least, to the presence of numerous species of cellulose-dissolving molds; but as an increase in the number of noncellulose-dissolving molds was also found, the conclusion was reached that a great many species of molds which were able to use the by-products produced by the fermentation of the cellulose were also present. The failure to obtain cellulose-destroying bacteria from this soil by direct plating does not mean that these organisms were not present in considerable numbers in this soil. Owing to the large number of mold colonies developing on the plates it was necessary to prepare plates from very high dilutions, and as the mold colonies ordinarily develop more readily than the bacterial colonies the detection of the latter was uncertain, even when present in considerable numbers. In fact, the presence of cellulose-dissolving bacteria was easily demonstrated by dropping a small quantity of the soil into a nutrient solution containing filter paper and handling after the manner previously described for isolating cellulose-destroying bacteria.

A second experiment was planned to determine the presence of the cellulose-dissolving mold spores in the atmosphere. Ordinary Petri dishes containing sterile cellulose agar were exposed to the laboratory air for five minutes and then incubated at 30° C. for six days. From 8 to 25 mold colonies developed, a high percentage of which cleared a well-defined zone about the colony, and some destroyed the cellulose with such rapidity that the entire plate of cellulose agar was cleared up at the end of the sixth day.

The power of *Penicillium africanum*, *P. pinophilum*, an unnamed new species of Penicillium, and *Aspergillus fumigatus* to dissolve different forms of cellulose, was also tested. These species had previously shown a rapid destruction of the precipitated cellulose in the regular cellulose agar. Cellulose in the form of cotton, precipitated filter paper, rye straw, cherry-wood shavings, and cedar-wood shavings was placed in large Petri dishes and moistened with the regular cellulose nutrient solution. The experiment was continued three months at room temperature, care being taken to keep all the material under study well moistened. At the end of the experiment the differences shown in Table VI were observed.

TABLE VI.—*Action of Penicillium and Aspergillus on different forms of cellulose.*

Material tested.	Penicillium africanum.	Penicillium pinophilum.	Penicillium not named.	Aspergillus fumigatus.
Precipitated cellulose.	Good growth; cellulose dissolved.	Good growth; cellulose dissolved.	Good growth; cellulose dissolved.	Good growth; cellulose dissolved.
Cotton	....do	....do	....do	Do.
Rye straw	Good growth; cellulose not attacked.	Poor growth; cellulose not attacked.	Good growth; cellulose not attacked.	Good growth; cellulose not attacked.
Cherry shavings	Poor growth; cellulose not attacked.	No growth	Poor growth; cellulose not attacked.	Poor growth; cellulose not attacked.
Cedar shavings	....do	Poor growth; cellulose not attacked.	No growth	Do.

The cotton was reduced to a transparent gelatinous mass and was stained a reddish brown color by all molds except by *Penicillium* n. sp. (not named). Although the molds made a good growth on rye straw they did not appear to attack the walls of the cells nor did they show any action on the wood shavings from cherry or cedar.

With the exception of the Penicillium and Aspergillus forms, only a few of the species isolated have been identified. For the identification of the species named below we are indebted to Dr. Charles Thom, Mycologist in Charge of Cheese Investigations, Bureau of Animal Industry, United States Department of Agriculture.

Penicillium expansum (Link) Thom.
Penicillium pinophilum Hedg.
Aspergillus nidulans Eidam.
Aspergillus fumigatus Fres.
Aspergillus flavus Link.
Gliocladium viride Matr.
Trichoderma lignorum (Tode) Harz.
Penicillium africanum Doebelt.
Penicillium n. sp.
Penicillium n. sp.

Dr. Thom has also kindly furnished us a set of his Penicillium cultures described in his bulletin.[1] These have been tested for their cellulose-dissolving power, and the following species have been found to destroy cellulose more or less rapidly:

Penicillium claviforme Bainier. Destruction strong.
pinophilum Hedg. Destruction weak.
luteum Zukal. Destruction strong.
Penicillium roseum Link. Destruction medium.
stoloniferum Thom. Destruction strong.
rugulosum Thom. Destruction strong.

Mrs. Flora W. Patterson, Mycologist of the Bureau of Plant Industry, has identified the following species:

Cephalothecium roseum Cda.
Haplographium echinatum (Riv.) Sacc.
Sporotrichum radicicolum A. Zimm.
Sporotrichum thebaicum Ehrenb.
Sporotrichum sporulosum Sacc.
Fusarium sp. undet.

Contrary to the general belief that soil fungi are largely confined to acid soils, the writers have found that the cellulose-dissolving forms multiply with great rapidity in alkaline soils when cellulose in the form of filter paper was added.

While only preliminary studies of cellulose destruction in our soils have been made, the great variety and vigor of the cellulose-dissolving fungi lead to the conclusion that cellulose destruction in soil is due in a large measure to the activity of these organisms, and it is certain that their importance in this connection has been greatly underestimated.

PRODUCTS RESULTING FROM THE DESTRUCTION OF CELLULOSE BY BACTERIA.

In view of the fact that the cellulose added to the soil represents a large amount of potential energy, the value of which depends upon the nature of the compounds formed in its destruction, it becomes quite interesting to inquire into the nature of these substances. If we are to accept the conclusions of earlier investigators, a large part of the energy contained in the cellulose is invariably lost in the form of gaseous products; however, the failure of any of the cellulose-dissolving bacteria studied in our laboratory to produce gaseous products from cellulose or sugar solutions in which they made a

[1] Thom, Charles. Cultural Studies of Species of Penicillium. Bulletin 118, Bureau of Animal Industry, U. S. Dept. of Agriculture. 1910.

luxuriant growth would seem to indicate that no gas is formed by pure cultures of the cellulose ferments.

Under normal conditions the compounds formed by the cellulose ferments will of course be seized upon by a host of other microorganisms and split up into simpler compounds. In some sandy soils the destruction may be extremely rapid and complete, resulting in the formation of little or no humus; under such conditions the carbon content of the cellulose is no doubt liberated as carbon dioxid, and under other conditions a large percentage of the carbon may be lost as methane. In either case the loss is believed to be due to secondary fermentations induced by organisms acting upon the by-products of the cellulose ferments.

It is well known that many fermentation processes may result in the formation of carbon dioxid and hydrogen and that such fermentations may be induced by many species of bacteria. In our investigations of the by-products of cellulose fermentation many experiments have been conducted to determine the nature of the gaseous products formed by mixed cultures of cellulose ferments. In a large series of flasks the sterile nutrient solution containing a quantity of pure filter paper was inoculated with pure cellulose ferments; a disintegration of the paper always occurred without any gas formation. The flasks were then divided into two series. The first series was left uncontaminated, while each flask of the second series was inoculated with a very small quantity of soil, manure, or sewage slime. In 48 hours after contamination all of the flasks of the second series had produced small gas bubbles; the gas formation continued from two to four weeks, during which time 100 cubic centimeters or more of gas was collected. None of the flasks of the uncontaminated series showed any gas, although the disintegration of the paper was almost complete. An analysis of the gas formed showed it to consist of a mixture of carbon dioxid and hydrogen.

In no case has a methane fermentation been secured from pure filter paper in the cellulose nutrient solution, but only from fermenting solutions rich in nitrogenous organic matter, and then only when the solution has been held under strict anaerobic conditions. A quantity of fresh horse manure was placed in a 2-liter filter flask, the flask filled with tap water, and the air exhausted to produce strictly anaerobic conditions. The flask was then incubated at 37° C. for 16 days before any gas formation was evident, but when once started the formation was very rapid and about 40 cubic centimeters of gas was produced a day. An analysis of the gas produced during the first seven days showed a large percentage of carbon dioxid and hydrogen with a little nitrogen; a later analysis, however, gave a large percentage of carbon dioxid, methane, and only a little hydrogen and nitrogen. The experiment was repeated at 55° C. The gas formation started in 24 hours and continued vigorously for 6 days, after which it ceased, although a considerable quantity of cellulose remained undissolved. An analysis of the gas showed it to consist of carbon dioxid, hydrogen, and nitrogen.

Considerable time has been spent in an effort to isolate the organisms giving rise to the gaseous products in the cellulose solutions, and while several organisms have been isolated which form gas in sugar solutions none has been discovered which will form gas from cellulose when grown in association with the cellulose ferments. Future investigations, however, will no doubt reveal many organisms, which have this power. Such investigations can be more intelligently carried on when we know the nature of the compounds formed by the cellulose ferments. While it has been possible to make only preliminary examinations of these compounds from rather small quantities of solutions, sufficient data have been secured to warrant the conclusion that the principal by-products of some species of cellulose-dissolving bacteria consist of formic and acetic acid, while with other species only traces of fatty acids are formed. None of the solutions examined have shown any trace of aldehydes, ketones, alcohols, or carbohydrates capable of reducing Fehling's solution. Since the by-products of cellulose decomposition apparently vary with the species of bacteria causing such decomposition, an accurate knowledge of the compounds formed will no doubt be of much value in specific classification of these organisms.

CONCLUSIONS.

(1) Neither the elective culture method of Omelianski nor the filter-paper sheet method of Van Iterson is satisfactory for isolating the cellulose-dissolving bacteria, but these organisms may be isolated readily by means of a selective medium such as cellulose agar.

(2) Cellulose-destroying bacteria and cellulose-destroying molds are universally present in cultivated soils.

(3) The cellulose-destroying bacteria are facultative in nature but destroy cellulose most rapidly in the presence of air.

(4) The cellulose-dissolving bacteria isolated by the writers are morphologically and physiclogically different from the hydrogen and methane ferments of Omelianski.

(5) Some species of cellulose-destroying bacteria, including the thermophiles, lose their power to destroy cellulose very rapidly on artificial media.

(6) Thermophilic cellulose-destroying bacteria are well distributed in nature and are extremely active agents in the destruction of cellulose under favorable temperature conditions.

(7) Filamentous fungi play a much more important rôle in the destruction of cellulose in soils, especially in alkaline soils, than has hitherto been supposed.

(8) The cellulose-destroying molds act differently toward different kinds of cellulose.

(9) The gaseous products attributed to the cellulose ferments by earlier investigators are due to secondary fermentations induced by other organisms.

(10) The principal products formed by some species of cellulose-dissolving bacteria consist of the lower fatty acids; with other species only traces of fatty acids are formed. No aldehydes, ketones, alcohols, or reducing sugars were produced by any of the species examined.

(11) An accurate knowledge of the by-products formed by different species of cellulose-destroying bacteria will aid in a specific classification of these organisms.

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DESCRIPTION OF PLATES.[1]

Plate I. Colonies and vegetative cells of *Bacterium liquatum* and *Pseudomonas subcretus*. Fig. 1.—*Bacterium liquatum*. Cellulose-agar plate, 15 days at 30° C. Normal size. Fig. 2.—*Bacterium liquatum*. Starch-agar plate,[2] 5 days at 30° C. Normal size. Fig. 3.—*Bacterium liquatum*. Vegetative cells from 24-hour culture on beef agar, aqueous fuchsin stain. Magnification 1,000. Fig. 4.—*Pseudomonas subcretus*. Vegetative cells from 24-hour culture on starch agar, carbol fuchsin stain. Magnification 1,000. Fig. 5.—*Pseudomonas subcretus*. Cellulose-agar plate, 15 days at 30° C. Normal size. Fig. 6.—*Pseudomonas subcretus*. Starch-agar plate, 5 days at 30° C. Normal size.

Plate II. Colonies and vegetative cells of *Bacterium fimi* and *Bacillus bibulus*. Fig. 1.—*Bacterium fimi*. Cellulose-agar plate, 15 days at 30° C. Normal size. Fig. 2.—*Bacterium fimi*. Starch-agar plate, 5 days at 30° C. Normal size. Fig. 3.—*Bacterium fimi*. Vegetative cells from 24-hour culture on beef agar, carbol fuchsin stain. Magnification 1,000. Fig. 4.—*Bacillus bibulus*. Vegetative cells from 24-hour culture on beef agar, carbol fuchsin stain. Magnification 1,000. Fig. 5.—*Bacillus bibulus*. Cellulose-agar plate, 15 days at 30° C. Normal size. Fig. 6.—*Bacillus bibulus*. Starch-agar plate, 5 days at 30° C. Normal size.

Plate III. Colonies, spores, vegetative cells, and involution forms of *Bacillus cytaseus*. Fig. 1.—Cellulose-agar plate, 15 days at 30° C. Normal size. Fig. 2.—Starch-agar plate, 5 days at 30° C. Normal size. Fig. 3.—Spores from fermenting filter paper, 9 days at 30° C., aqueous fuchsin stain. Magnification 1,000. Fig. 4.—Vegetative cells from 24-hour culture on potato agar at 30° C., carbol fuchsin stain. Magnification 1,000. Fig. 5.—Involution forms from 48-hour culture on potato agar, carbol fuchsin stain. Magnification 1,000.

Plate IV. Vegetative cells of *Bacillus bibulus* and *Bacillus cytaseus*, showing flagella. Figs. 1 and 2.—*Bacillus bibulus*. Vegetative cells from 24-hour culture on beef agar, Dr. Hugh Williams's flagella stain. Magnification 1,000. Figs. 3 and 4.—*Bacillus cytaseus*. Vegetative cells from 24-hour culture on potato agar, Dr. Hugh Williams's flagella stain. Magnification 1,000.

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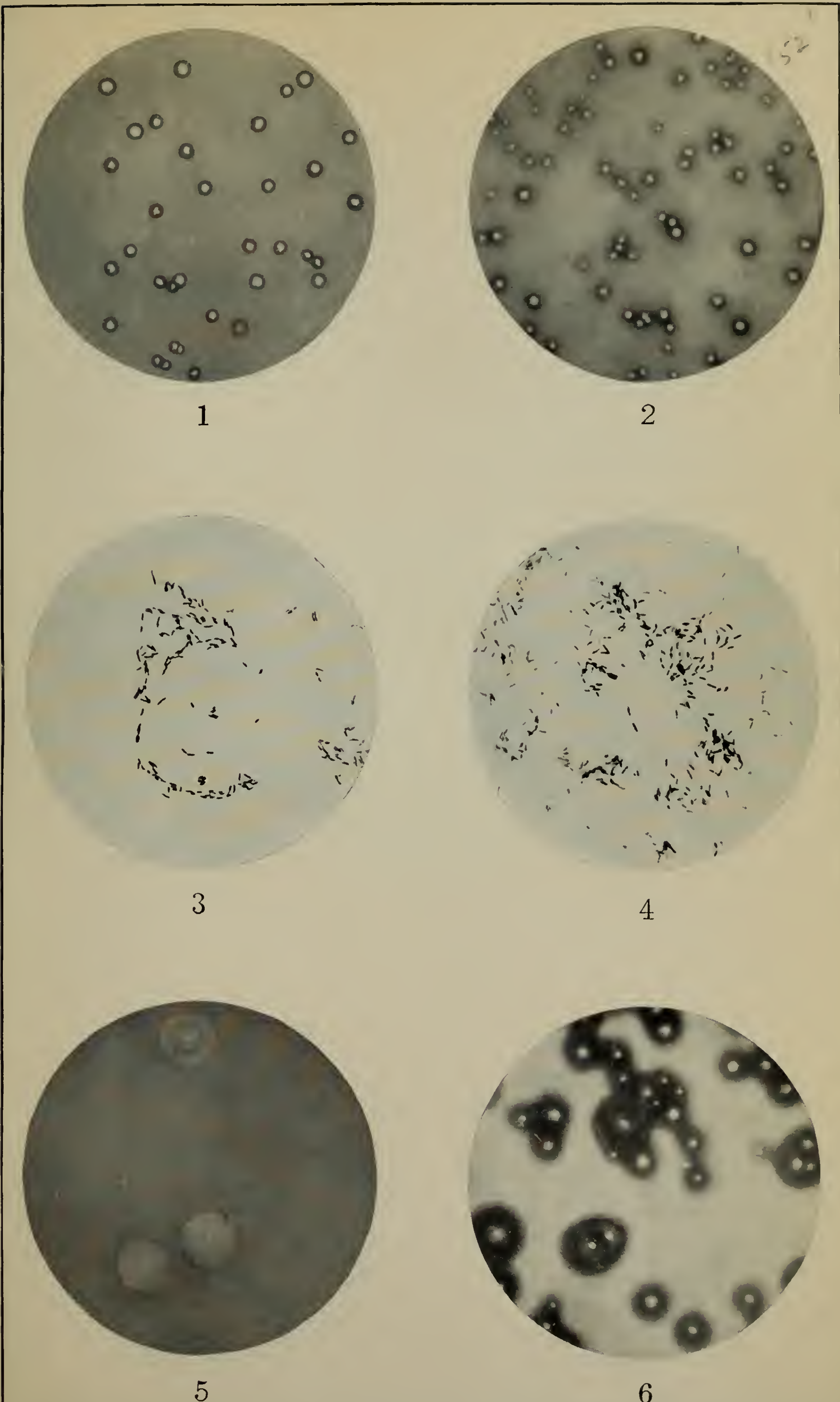
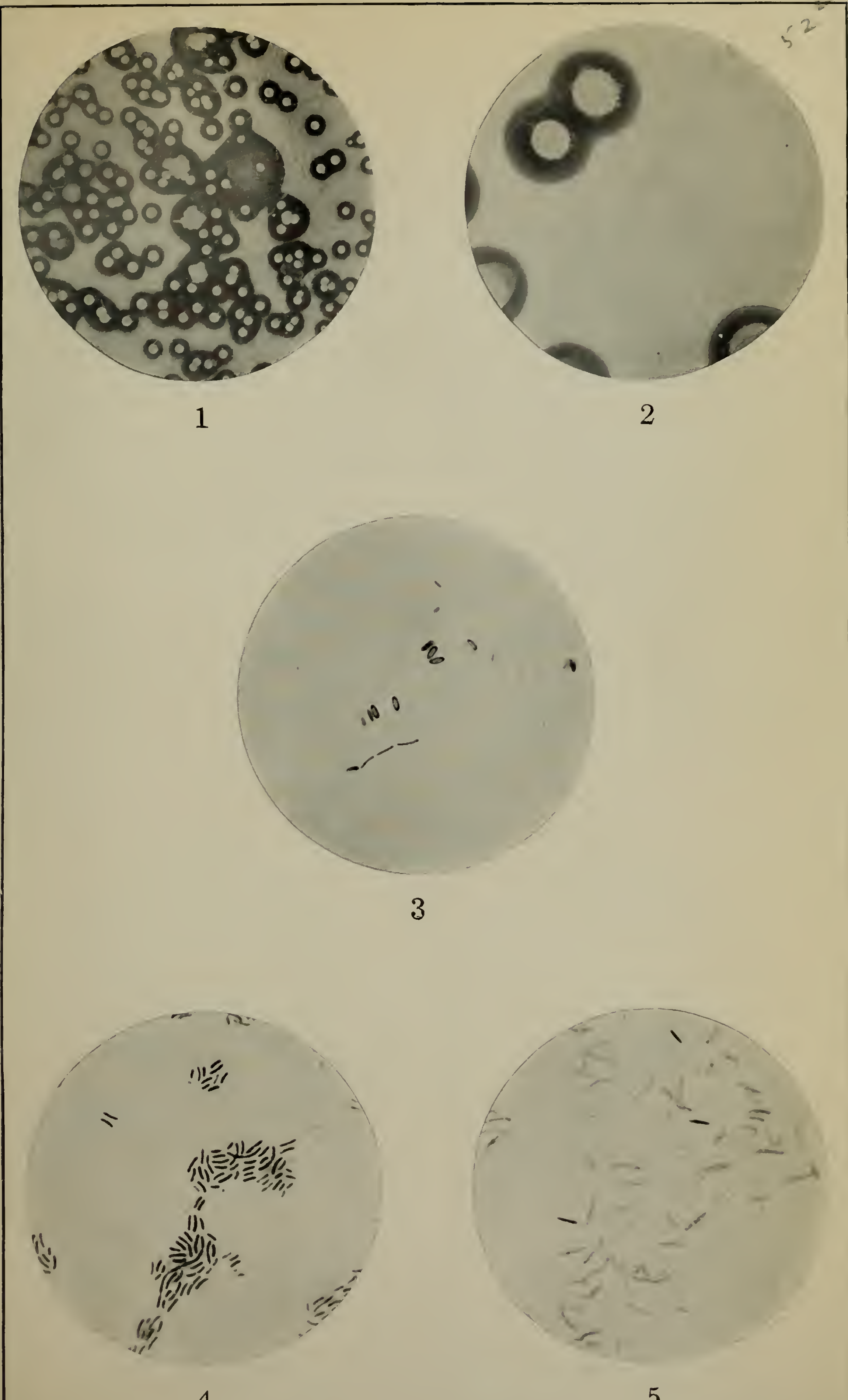
[1] The photomicrographs and photographs here reproduced were made by Mr. F. L. Goll, of the Office of Soil-Bacteriology and Plant-Nutrition Investigations.

[2] A small quantity of 95 per cent alcohol was poured over the surface of all the starch-agar plates to bring out the enzymic zone.



COLONIES AND VEGETATIVE CELLS OF BACTERIUM LIQUATUM AND PSEUDOMONAS SUBCRETUS.

COLONIES AND VEGETATIVE CELLS OF BACTERIUM FIMI AND BACILLUS BIBULUS.



COLONIES, SPORES, VEGETATIVE CELLS AND INVOLUTION FORMS OF BACILLUS CYTASEUS.

VEGETATIVE CELLS OF BACILLUS BIBULUS AND BACILLUS CYTASEUS, SHOWING FLAGELLA.

Issued February 21, 1913.

U. S. DEPARTMENT OF AGRICULTURE.

BUREAU OF PLANT INDUSTRY—BULLETIN NO. 267.

B. T. GALLOWAY, *Chief of Bureau.*

NONPERENNIAL MEDICAGOS:

THE AGRONOMIC VALUE AND BOTANICAL RELATIONSHIP OF THE SPECIES.

BY

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AND

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LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry,
Office of the Chief,
Washington, D. C., September 19, 1912.

Sir: I have the honor to transmit and to recommend for publication as Bulletin No. 267 of the series of this Bureau the accompanying manuscript entitled "Nonperennial Medicagos: The Agronomic Value and Botanical Relationship of the Species," prepared by Messrs. Roland McKee, of the Office of Forage-Crop Investigations, and P. L. Ricker, of the Office of Taxonomic and Range Investigations. The botanical descriptions have been prepared by Mr. Ricker and the remainder of the bulletin by Mr. McKee.

This paper gives the results of an investigation of a large number of species usually known as bur clovers, mostly secured with the assistance of the Office of Seed and Plant Introduction. These species have been extensively tested in California and to a less extent in the Southern States in order to ascertain their agronomic possibilities in comparison with three species already grown in the United States.

The increasing importance of these plants for pasturage and green-manuring purposes throughout the areas mentioned makes the paper a timely contribution to our knowledge of the possibilities of this group of forage plants.

The illustrations for Plates V to XV, inclusive, are from photographs by Mr. E. L. Crandall, except the drawings of seeds on Plates VI and IX, which were made by Prof. F. H. Hillman.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

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NONPERENNIAL MEDICAGOS: THE AGRONOMIC VALUE AND BOTANICAL RELATIONSHIP OF THE SPECIES.

INTRODUCTION.

The genus Medicago, as commonly accepted by botanists, includes about 7 perennial species, with about 16 subspecies, of which alfalfa is the best known and most important, and about 37 annual species, with about 80 subspecies, one of which, yellow trefoil (*Medicago lupulina*), has also a biennial or possibly perennial form. The duration of several—at least three—species is uncertain. There is considerable difference of opinion among botanists as to the number of annual species, mostly known as bur clovers. In 1873 Urban [1] recognized 39 such species, with 64 subspecies, since which time 3 other species and 17 additional subspecies have been described.

In this paper agronomic and botanical notes are given concerning 14 species and 9 subspecies which have been studied for two to five years.

Three species are more or less cultivated or established in the United States, namely, toothed bur clover (*Medicago hispida*) and its subspecies, principally on the Pacific coast; spotted bur clover (*M. arabica*), mainly in the Cotton States and in California; and yellow trefoil, or black medic (*M. lupulina*), more or less abundant throughout the United States. By far the greatest amount of agronomic information at hand concerns these three species, and the desirability of utilizing any of the other species will depend largely on whether they exhibit any points of superiority.

All the annual medicagos grow under natural conditions as winter annuals, and under cultivation they succeed best when planted in the fall. Yellow trefoil is the only hardy species; other species can be successfully grown only where the winters are not too cold.

In the various sections where bur clovers grow somewhat extensively most of the plants are usually of one species or subspecies. In California toothed bur clover (*Medicago hispida denticulata*) is most widely distributed. *Medicago arabica*, *M. hispida confinis*, and *M.*

[1] Verhandlungen des Botanischen Vereins der Provinz Brandenburg, bd. 15, 1873, pp. 1–85, pls. 1–2.

hispida apiculata are also found in that State, but to a more limited extent. The wide distribution of *Medicago hispida denticulata* in California is partially explained by its natural adaptation, but perhaps more by the fact that it is the most widely introduced species, whether intentionally as pure seed for sowing for pasturage or green manuring or unintentionally as a mixture with other seed. *Medicago hispida*, *M. hispida apiculata*, and *M. hispida confinis* were in all probability introduced into California along with *M. hispida denticulata*, with which they are found nearly everywhere, but in lesser quantity.

Spotted bur clover (*Medicago arabica*) is apparently of more recent introduction into California than *M. hispida denticulata* and is far less widely distributed in that State. On the creek pasture lands on the Bidwell ranch at Chico, *M. arabica* is more often found than *M. hispida denticulata*. To judge from the quantity there, it was perhaps first introduced at this point and has been distributed thence to various parts of the Sacramento Valley, where it is found in small areas. According to Mrs. Katherine Brandegee, as reported by Mr. Willis L. Jepson,[1] *Medicago arabica* is almost as common as *M. hispida denticulata* in San Francisco County. *Medicago arabica* is the commonest species throughout the South Atlantic and Gulf Coast States east of the Mississippi River and succeeds exceptionally well throughout this section. It can stand lower winter temperatures than the toothed bur clovers (the *M. hispida* group), and for this reason is better adapted to this section, in which the toothed bur clovers more often winterkill. It is practically the only species used for pasturage or green manuring in the Southern States. *Medicago hispida denticulata* and *M. arabica* succeed well in Texas, the former species being the more generally distributed.[2]

Yellow trefoil (*Medicago lupulina*) occurs throughout the greater part of the United States, and on account of its hardiness is adapted to sections farther north than either *M. arabica* or *M. hispida* and its forms.

SOIL AND MOISTURE REQUIREMENTS.

Toothed bur clover and spotted bur clover succeed under varied conditions as to moisture, soil, etc. In California, as well as in the South, they grow on all types of soil from nearly pure gravel to heavy adobe. They do better on the heavier loam soils, but will grow in almost any soil containing sufficient moisture. They make a fair growth even under rather arid conditions. In the dry foothill pasture lands of California the toothed bur clover makes a valuable

[1] Jepson, W. L. Flora of Western Middle California, 1901, p. 313.
[2] Bulletin 108, Texas Agricultural Experiment Station. 1908.

addition to the native pasturage, and in the dry-land pastures of the valleys it is generally distributed and does well. In different parts of Texas it is found growing along the roadsides and in waste places where the conditions are more or less severe. It will stand a small percentage of alkali. In California it is found on slightly alkaline soils, but not on soils heavily charged with salts. While fairly well-drained lands are the most desirable, spotted bur clover and toothed bur clover produce good crops on moist lands. On California adobe lands, which are sometimes poorly drained and lose their moisture slowly, all three species do exceptionally well. Where there is excessive moisture the crop matures later and remains green far into the summer. While not particularly adapted to shade, both the spotted and toothed bur clovers grow vigorously among the timber along streams. Observation indicates that *Medicago arabica* is better adapted to shady conditions than *M. hispida*.

Yellow trefoil, or black medic, is somewhat notorious, from the fact that its seed has frequently been used to adulterate alfalfa seed. Nevertheless, the plant has agricultural merit not only as forage, but perhaps even more as a winter cover and green-manure crop when used in the same manner as crimson clover. The seed is usually cheaper than that of crimson clover and the plant more hardy. At the Arlington Experimental Farm the two plants mixed gave excellent results, and yellow trefoil alone compares very favorably with crimson clover alone.

VALUE FOR PASTURAGE.

The general characteristics of spotted bur clover and toothed bur clover make them especially valuable for pasturage. They have high feeding value, spread readily, and make satisfactory growth under varied soil conditions. In the pasture lands of the South, as well as on the Pacific coast, they have spread very rapidly after being once introduced. The tendency of part of the seed to carry over in the soil for several years before germination insures against extermination by failure to develop seed in any year—from whatever cause, such as overpasturing or unfavorable weather. Whether the seed germinates or carries over is apparently a matter of depth in planting. Viable seed sown too deep will not germinate until it is brought nearer the surface. Spotted bur clover and toothed bur clover both contain what is known as hard seed that probably will not germinate the first season, even if other conditions are favorable. Such seed carries over until the second or third year.

Most bur clovers are admirably provided with means for natural dissemination. The spiny burs of some species readily adhere to various animals and in this way are carried long distances. Burs without spines are distributed by other means. It is commonly observed in California that orchards fertilized with manure from corrals produce a heavy growth of bur clover, evidently from seed that passed through the animals undigested and that still retained its power of germination.

In a mixture with grasses, bur clovers are excellent pasturage and make considerable growth of green feed during the winter and in early spring before the grasses start. Green bur clover will often produce bloat in cattle, and care should be taken when first turning them into such fields.

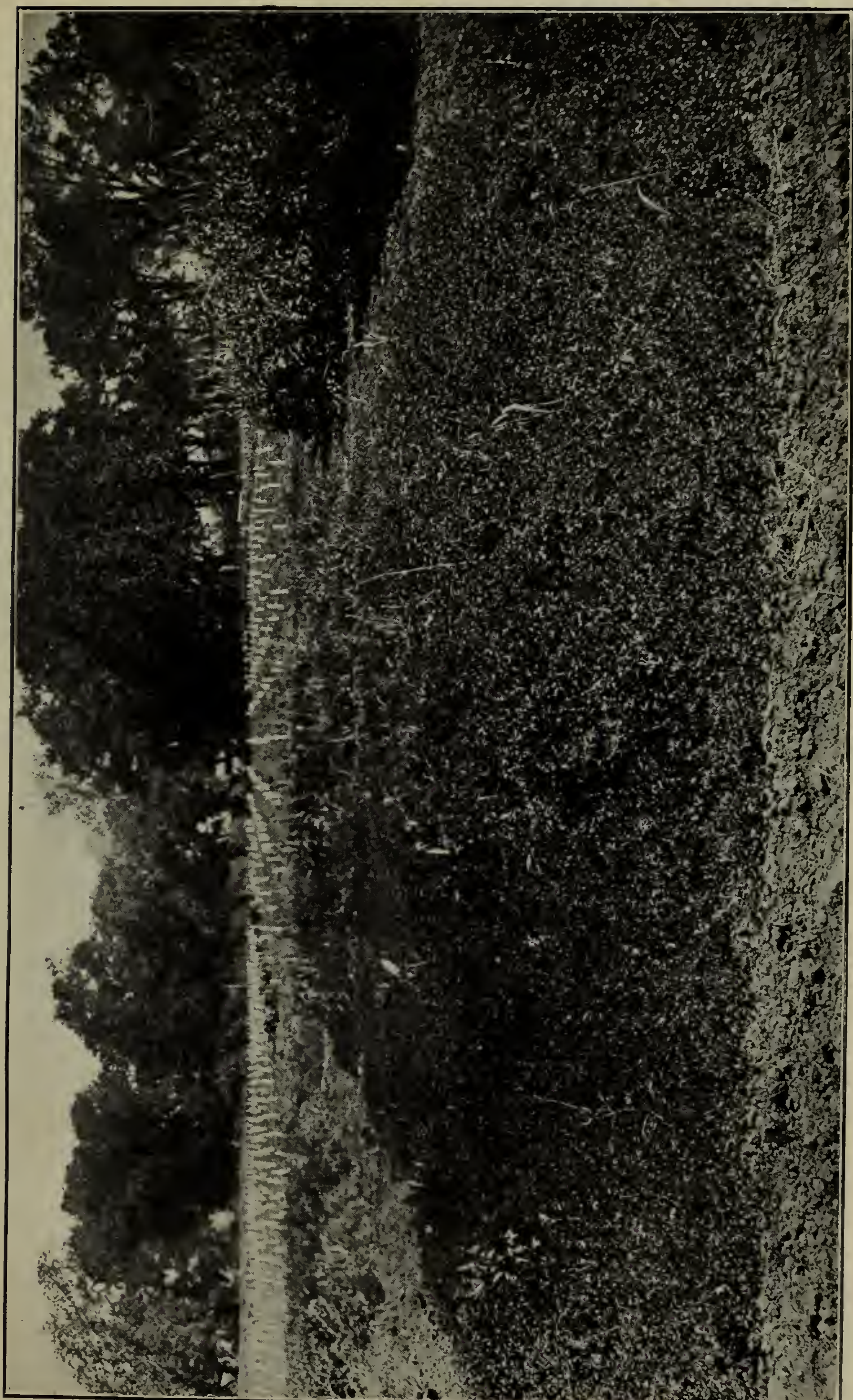
A heavy crop of pods and seed afford a large quantity of valuable feed. In fact, it is the seed and pods that constitute the greater part of the feeding value of this crop when in the dry state. Their feeding value is high, and stock fed on them fatten rapidly. Pastures containing large quantities of matured burs of the common bur clovers are especially desirable for fattening sheep. The time at which the burs are available adds importance to their feeding value, especially in California, where the dry season usually continues from May until November. Here the valley pastures are wholly dry by midsummer, and dry pasturage is all that is available. The bur clovers, being mature, possess their greatest feeding value at such time, but if not then fed are available later. The burs are mostly eaten dry, but those with heavy spines are much more readily eaten after rains have softened them.

The spiny species or subspecies of bur clovers are objectionable as sheep pasturage on account of the burs getting into the wool. For this reason the spineless forms are preferable. *Medicago hispida confinis* is a spineless form of toothed bur clover and *M. arabica inermis* is a new spineless form of spotted bur clover. Button clover (*M. orbicularis*) is another very promising species (Pls. I and II), which, on account of its large spineless burs and heavy yields of seed, is superior to the more common spotted or toothed bur clovers wherever it will make an equally good growth.

SECURING AND MAINTAINING A STAND IN PASTURES.

The seeding of pasture lands to bur clover on the Pacific coast is a very simple process. The seed, either hulled or in the bur, is scattered over the land and, without further attention, is left to itself. Hulled seed will generally give a better stand than seed in the bur and is to be preferred when bought. Seed in the bur is also more likely to contain undesirable weed seeds.

In the Southern States a stand is not so readily secured, owing in part to the fact that nodule-forming bacteria which supply nitrogen to the plant are not present, or at least do not develop readily, except



A HEAVY GROWTH OF BUTTON CLOVER (MEDICAGO ORBICULARIS) AT CHICO, CAL.

STEMS OF BUTTON CLOVER (MEDICAGO ORBICULARIS), SHOWING APPEARANCE CHARACTERISTIC OF THIS SPECIES.

(One-half natural size.)

in the more favored and better prepared soils. It is therefore necessary to supply inoculation before a good growth can be secured.

In California seeding may be done at any time during the summer, or in the fall before the winter rains begin. In the Southern States summer seeding is not advisable, on account of rains, and seed should be sown in the early fall, so that it will start and continue growth during winter. About the first of September is perhaps the best time for seeding in most sections. When bur clover is once seeded it persists indefinitely. Species with spiny burs, having the advantage of this means of distribution, are perhaps more persistent than species with smooth burs. Spiny burs are less readily eaten by animals. Thus protected, many of them drop or are knocked from the plants, are trampled into the ground, and thus reseed themselves. When not trampled into the ground, the seeds of species with small burs seem to germinate and take root much more readily than those of species with large burs. Large smooth-podded sorts must be pastured lightly or stock kept off in the spring until the seed has matured, else the immature seed is largely eaten, digested, and lost.

PALATABILITY.

As pasturage or hay, spotted bur clover and toothed bur clover are not as readily eaten by most kinds of stock as ordinary grass and hay. Especially in the green state, they possess a slightly disagreeable taste, which at first makes them somewhat unpalatable. The taste is not a serious drawback, as stock soon become accustomed to it, and the green plants of all three species are readily eaten where other feed is not abundant. Whether it is owing to a difference in the palatability of the different species has not been definitely determined, but comparison indicates that in the green state most animals apparently prefer *Medicago orbicularis* to *M. hispida denticulata.*

VALUE FOR HAY.

The bur clovers can not be considered as among the best crops for hay. They are somewhat unpalatable, and their decumbent habit of growth makes them difficult to handle alone. If grown with oats or other small grain for support, they can be handled with sufficient ease to be sometimes profitably utilized in this way. The fact that there are better crops for hay will always limit the use of the bur clovers for this purpose. In California, in seasons when the hay crop is short, considerable quantities of mixed bur clover and wild-oat hay are put on the market, but such hay is considered inferior by the trade and sells at a lower price than grain hay. Table I gives the chemical analyses of bur clover in comparison with alfalfa.

TABLE I.—*Average percentage composition of bur clover and alfalfa hay.*

Kind of hay.	Number of analyses.	Water.	Ash.	Protein.	Crude fiber.	Nitrogen-free extract.	Ether extract.
Bur clover[1]	3	10.11	7.27	12.60	26.37	41.14	3.16
Alfalfa[2]	21	8.4	7.4	14.3	25.0	42.7	2.2

[1] Annual Report, California Agricultural Experiment Station, 1894–5, p. 147; 1896–7, p. 113.
[2] Westgate, J. M. Alfalfa. Farmers' Bulletin 339, U. S. Dept. of Agriculture, 1908, p. 28.

VALUE FOR COVER AND GREEN-MANURE CROPS.

Spotted bur clover is now used to a considerable extent in the cotton-growing States as a winter cover crop, and its popularity for this purpose is increasing. Over much of this area it has given better results than crimson clover. Owing to the scarcity of seed of the spotted bur clover, that of toothed bur clover is frequently sown; but the results clearly show that toothed bur clover is less hardy and in severe winters is destroyed. Toothed bur clover is commonly used as a cover crop in California orchards, as when once well established it volunteers from year to year. In China it is often used as a cover crop on rice fields, and the results of preliminary tests indicate that it will be excellent for this purpose on rice lands in Louisiana and Texas.

Yellow trefoil has proved itself an excellent winter cover crop in Virginia and its wide distribution leads to the belief that it has greater merit than has been heretofore realized.

Inoculation is of paramount importance in attaining a satisfactory stand with any of these species.

SOURCES OF SEED.

Bur clover is little grown in the United States as a seed crop. Spotted bur clover has been grown to some extent for seed in the Southern States, but usually only a small acreage is so handled. In California considerable quantities of seed of toothed bur clover are secured with the crops of small grain, among which it grows naturally as a weed. The seeds of bur clover ripen at nearly the same season as the grain and are of necessity harvested with it. The raising of grain on large areas by individual farmers, as in California, necessitates delay in harvesting much of it, and thus favors the development and ripening of a much larger quantity of the bur clover than would otherwise mature. The bur clovers ripen a little later than wheat or barley, and if these grain crops are cut when first ripe, little bur clover seed is secured. The use of a combined harvester and thrasher in harvesting and thrashing small grain is especially favorable to the saving of bur clover seed, as in this way a minimum number of burs is knocked from the vines.

Bur clover seed is also obtained as a by-product from wool waste. Sheep running in pastures get the burs entangled in the wool and the seeds are thus carried to the mills to be separated as a waste product. Wool from Argentina, South America, where bur clovers are abundant, contains quantities of both toothed bur clover and spotted bur clover. This wool is shipped to the woolen mills, where the bur clover is generally taken out as a by-product. Southern European grazing sections are also sources of bur clover seed, which is carried in wool as from other countries and separated at the woolen mills.

GROWING FOR SEED.

In growing a crop of bur clover for seed several difficulties are encountered. The prostrate growth made by the plants, the failure of the burs to mature all at the same time, and their tendency to drop very easily from the stem as soon as ripe make the harvesting of seed difficult. To grow bur clover as a seed crop on a large scale is most practical in sections having a continuous dry summer. Rains in summer are apt to cause the seeds to germinate in the burs, making them more difficult to handle; but where such rains occur it is both practicable and advisable to raise seed in small quantity for one's own use.

The fact that the greater part of the commercial bur clover seed is *Medicago hispida* and its forms, with little *M. arabica*, makes it almost necessary for the farmer in the Southern States to grow his own seed at the present time. As has been stated, *M. arabica*, according to our present knowledge, is the best species for the South. Before seeding, the land should be put in as good condition as possible by plowing and harrowing, and if the seed is to be harvested by any method such as sweeping, the field should be run over with a float or roller to leave a smooth surface in order to facilitate the harvest. If a drill is used to sow the seed the ground should be especially well firmed.

TIME OF SEEDING.

In sections having a mild winter climate, bur clover should be sown in the fall. In California, where dry weather prevails throughout the summer, the seed may be sown at any time before the fall rains begin. Where summer rains occur, as in the Southern States, the planting should be delayed until the first of September. In dry sections, where it is desirable to start the seed in the fall with irrigation, the planting should be done about the first of October. The object is to sow the seed so late that a subsequent irrigation will not be necessary. Summer seeding in the South is not advised, as the young plants starting at that time are liable to suffer from drought, and where a heavy growth is made the plants tend to mature and die rather than continue growth through the winter. When seed in the bur is used, earlier planting may be practiced than when hulled seed is used. Germination will be delayed on account of the protection afforded by the burs, and the result is the same as a later planting of hulled seed.

METHOD AND RATE OF SEEDING.

The clean seed may be sown broadcast or by using an ordinary grain drill with press-wheel attachment. Special care should be taken to cover the seed thinly. The drill should be used only on well-firmed soil, as otherwise the seed will be planted too deep. The press-wheel attachment is necessary for the best success when a drill is used. In general, broadcast seeding will perhaps be found the most satisfactory and is the only method that can be employed when the seed is sown in the burs. A light harrowing is all that is necessary to cover hulled seed sown broadcast and will usually cover seed sown in the bur. When the land is left with light furrow markings, such as are made by a large-toothed harrow, seed not covered by the harrow at the time of seeding will fall into these small furrow depressions and be covered by the washing of subsequent rains. Good stands have been secured by this method without covering the seed at all at the time of sowing, and it probably will be found satisfactory in sections where a continuous rainy season occurs.

INOCULATION.

In the Southern States inoculation of the land is necessary to grow clover for the first time. In California the soil apparently is in most places already inoculated. The best method of inoculation is perhaps to mix a small amount of soil from an old bur-clover field with the seed, whether hulled or in the bur. The quantity of soil used need be only a mere dusting. Sowing seed in the bur seems also to insure inoculation, and for this reason it is commonly practiced in the Gulf Coast States. Open and loamy soils are most easily inoculated, and it is recommended that to establish bur clovers on a place an old garden patch or other well-prepared and manured piece of land be selected.

HARVESTING AND THRASHING.

The limited work that has been done with bur clovers has not entirely demonstrated the best method of harvesting the seed but has at least indicated the difficulties to be overcome and has suggested improvements on methods used. Several processes have been tried. The combined harvester and thrasher has been used in an attempt to cut and thrash the crop direct from the field at one operation, as small grains are harvested in the West. The seed of bur clover ripens continuously through a long period; hence, a large quantity of unripe burs are harvested even if the crop is cut when the yield is at its maximum. The green burs and accompanying green portions of the stems, which are gathered with the ripe burs and seed, contain much moisture and without special drying are likely to heat before they can be taken to a huller and the seed separated.

If the bur clover is sown with a grain crop which is allowed to ripen thoroughly, the difficulty just mentioned is largely overcome, but the yield of seed by this method is small, because the burs drop from the plant so easily as soon as ripe. It is also necessary to let the grain crop become overripe in order to allow the bur clover, which matures a little later, to develop its maximum yield of seed. The use of a common self-rake reaper has been suggested but not yet tried. The idea is to cut the crop when a maximum amount of seed is ripe and then to handle it as the seed crop of red clover or alfalfa is handled. The use of the reaper should reduce the loss of pods to the minimum possible with ordinary farm machinery, but whether the method is practicable remains to be demonstrated. The readiness with which the burs drop from the plants will make this method difficult at best, but by operating at a favorable time, as on a cloudy day or early in the morning, the loss of burs will be reduced to a minimum.

The idea of air suction has been tried in an attempt to overcome the difficulty occasioned by the burs dropping from the vines. This process has been tried by Mr. R. W. Jessup, of Oakland, Cal., who reports it only a partial success. A power suction machine was used and 20 acres of burs harvested. The vines were allowed to become thoroughly dry and were then cut with an ordinary mower and raked into windrows. The ground was thus left comparatively clean and in shape for the suction machine to operate. In the process of mowing and raking, all the burs were knocked from the vines, so that a maximum yield was obtained. By this method a quantity of other substances, such as sticks and small stones, were gathered with the burs and were very objectionable on account of the damage to the cylinder in the process of hulling. To overcome this objection a special device for cleaning foreign substances from the burs as they were harvested was used in connection with the suction machine in 1911. This device made the machine more satisfactory, but the method at best is somewhat expensive. With a heavy yield of seed the expense is reduced.

A method of harvesting first employed in the West and South is to allow the seed to ripen thoroughly and then to cut the vines with an ordinary mowing machine and rake them into windrows. The burs are then swept together with large barn brooms and hauled from the field. The burs gathered in this manner are mixed with more or less gravel and other foreign substances, which must be removed before the seed can be satisfactorily hulled or used in the bur. This separation is accomplished by the use of handbarrow screens and an ordinary fanning mill regulated to blow the burs over; or, if running water is handy, a quicker and more satisfactory method is to throw the burs into the water. All heavy substances sink, and the burs and lighter substances are dipped from the stream. To facilitate this method of separation the channel of the stream should be narrowed in the shape of an open V, which generally aids in collecting the cleaned burs. To dip the burs from the water a large handbarrow, with a bottom made of wire netting, has been found very satisfactory. The burs are spread on canvas to dry, after which they are ready for the huller.

All bur clovers, whether with large hard burs, like *Medicago turbinata*, or small soft burs, like *M. hispida denticulata*, or large soft burs, like *M. orbicularis*, are successfully hulled with an ordinary clover huller.

YIELD OF SEED.

Few data as to the actual seed yield of the various species of bur clovers are available. Table II gives the results of tests made at Chico, Cal.

During the winter of 1907–8 seasonal conditions were rather unfavorable to the production of heavy yields of seed, and the figures given (Table II) are undoubtedly somewhat lower than may be expected in a more favorable year. In the test referred to, the seed was sown in the fall before the winter rains began, and the crop was allowed to develop under natural seasonal conditions, without irrigation.

During the winter of 1908–9 *Medicago orbicularis*, *M. hispida nigra*, and *M. hispida confinis* were again grown for seed in one-twentieth acre plats. In this test the seed was sown early in October, with irrigation at time of seeding only. A good winter growth was thus insured. The yield of seed in the irrigated plats (Table II) was considerably greater than in the nonirrigated plats, which amounted to little.

During the winter of 1909–10 *Medicago orbicularis*, *M. scutellata*, *M. hispida confinis*, *M. hispida nigra*, *M. turbinata*, and *M. hispida terebellum* were again grown in one-twentieth acre plats. They were sown in October, 1909. The plats were irrigated before seeding only. The seed yields are not entirely comparable, on account of variation in the stands germinated. The plat of *M. hispida terebellum* had a very poor stand, and those of *M. turbinata*, *M. hispida nigra*, and *M. orbicularis microcarpa* (No. 7738) were thin. *M. orbicularis* (No. 10725), *M. hispida confinis*, and *M. scutellata* had good stands.

The large seed yields of *Medicago orbicularis*, which is a very promising species, have been very consistent through the four years.

Hulled seed of bur clover weighs about the same as alfalfa seed—60 pounds to the bushel.

TABLE II.—*Yields of seed per acre at Chico, Cal.*

S. P. I. No.[1]	Species tested.	Yield of hulled seed per acre.			
		1908	1909	1910	1911
		Pounds.	*Pounds.*	*Pounds.*	*Pounds.*
10725	Medicago orbicularis	860	790	1,160	947
7738	Medicago orbicularis microcarpa	520	....	300	....
16879	Medicago hispida terebellum	390	....	40	....
26077	Medicago scutellata	240	....	950	....
26076	Medicago turbinata	230	....	180	....
26071	Medicago hispida nigra	470	440	160	....
26070	Medicago hispida confinis	320	375	350	....
	Medicago hispida denticulata	450	....	....	....

[1] Seed and Plant Introduction number.

RELATION OF WEIGHT OF SEED TO VOLUME AND WEIGHT OF BURS.

The weight of seed in a given volume of burs varies considerably, mainly owing to differences in bulkiness of the burs in the several species. These variations are due not entirely but largely to differences in length of spines. Species with long spines have less seed in a given volume than spineless forms, especially when closely related types are compared.

The weight of seed in a given weight of burs also varies somewhat in the different species, mainly owing to differences in the texture of the burs. The harder types of burs have the smaller percentages of seed. Table III shows that the weight of seed in a bushel of burs in the different species varies from 1.75 pounds in *Medicago arabica* to 4.66 pounds in *M. turbinata;* and that the weight of seed in 100 pounds of burs varies from 20.89 pounds in *M. turbinata* to 33.78 pounds in *M. hispida denticulata.*

RELATION OF WEIGHT OF BURS TO THEIR VOLUME.

The great differences in the spines and in the texture of the burs make decided differences in the weight of burs in a given volume in the several species. A given volume of a species having very short or no spines or of those with hard burs is much heavier than of species having long spines and soft burs. Considerable variation in weight is caused by packing the burs, especially of the spiny species, and for this reason the weights can be only approximated. Table III shows that the weight of a bushel of burs in different species varies from 6 pounds in *Medicago arabica* to 22 pounds in *M. turbinata.*

TABLE III.—*Relation of weight of burs and seed to volume.*

S. P. I. No.	Species tested.	Weight of 1 bushel of burs.	Weight of seed.	
			In 1 bushel of burs.	In 100 pounds of burs.
		Pounds.	*Pounds.*	*Pounds.*
26077	Medicago scutellata	14	3.83	27.38
10725	Medicago orbicularis	8	2.66	33.5
26076	Medicago turbinata	22.33	4.66	20.89
26071	Medicago hispida nigra	8	2	25
9749	Medicago ciliaris	8.66	2.16	25
16879	Medicago hispida terebellum	11.99	3.18	26.39
	Medicago denticulata	6.16	2.08	33.78
	Medicago arabica	6	1.75	29.16

RESEMBLANCE TO ALFALFA SEED.

The seeds of a number of species of bur clover resemble alfalfa seed very closely. The most common are the *Medicago hispida* group, *M. lupulina*, and *M. arabica*, the seeds of which are of lighter yellow color, lacking the rich, greenish yellow shade of alfalfa; and all are uniformly larger except *M. lupulina*, which is somewhat smaller and is the only species in which the difference in size is readily noticeable. *Medicago arabica* is further distinguished by having a small, well-developed projection at the end of the hilum.

VITALITY OF SEED.

As mentioned elsewhere, bur clover seed retains its vitality for a very long time. Seed three years old will generally show delayed germination, but it is only after several years that the percentage of germination is noticeably decreased. (Table IV.) Not only does seed retain its power of germination when kept under dry conditions, as in a seed room, but it will carry over in the soil for a number of years in the same way.

TABLE IV.—*Germination tests of Medicago arabica.*[1]

S. P. I. No.	Age of seed.	Duration of test.	Germination of unclipped seed.	Hard seed.	Good seed.	Germination of clipped seed.
	Years.	*Days.*	*Per cent.*	*Per cent.*	*Per cent.*	*Per cent.*
16213	6	12	20	60	80	77
21550	4	12	30	65	95	90
23661	3	7	26	53	79	77
25878	2	7	19	76	95	94

[1] Results obtained by the Seed Laboratory, Bureau of Plant Industry.

It has been observed in California, in orchards kept free from all growth during the spring months and no seed allowed to develop, that a good stand of the toothed bur clover occurs each year for four or five years after the last crop of seed was allowed to mature in the orchard.

Plantings of a number of species of bur clover were made in pots so that this point could be more definitely observed and showed that it was a common occurrence for seed to carry until the second year before germinating, even when the depth of planting and other conditions were favorable for growth.

INSECT ENEMIES.

The clover-seed chalcis fly (*Bruchophagus funebris* How.) which attacks red clover[1] and alfalfa[2] is also common in bur clover. The small flylike insect lays its eggs in the ovules; the larvæ develop in the seed and reach maturity by the time the seed is ripe. The amount of seed thus destroyed at Chico, Cal., is considerable, especially in that maturing late. Of the early-maturing seed perhaps 10 per cent is destroyed, while the loss of late seed may be as high as 75 per cent. All species tested are subject to its attacks, some more severely than others. No practical way of controlling this pest seems to be known.

DIFFERENCES IN THE BURS OF DIFFERENT SPECIES.

The pods, or burs, of the different species of annual medicagos differ very much in size, form, and with regard to the spines (Pl. III). They also vary widely in weight and texture.

In such species as *Medicago orbicularis* and *M. scutellata* the burs are very large and spineless, being decidedly flattened in *M. orbicularis* and nearly spherical in *M. scutellata*. The pods of both are soft and somewhat papery. In *Medicago ciliaris* and *M. echinus* the burs are very large and have heavy spines. The spines are erect in the former and decidedly appressed in the latter species. The texture of the bur tends to be hard in *M. ciliaris* and a little less so in *M. echinus*. The general form in both species is oval. In *M. turbinata* the bur is large, oval, very hard (in the most common type), and has a few short tuberclelike spines. *Medicago rigidula* and *M. murex* are somewhat similar to *M. turbinata*, but they are smaller and commonly have longer spines. Of species with smaller burs, some, as *M. hispida denticulata* and *M. arabica*, have spines, and some, as *M. hispida confinis*, are without spines. All variations between these types are found, and there are many other forms which mark botanical characteristics peculiar to definite species.

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[1] Circular 69, Bureau of Entomology, U. S. Dept. of Agriculture, 1906, p. 7.
[2] Farmers' Bulletin 339, U. S. Dept. of Agriculture, 1908, p. 41.

DESIRABILITY OF A BUR WITHOUT SPINES.

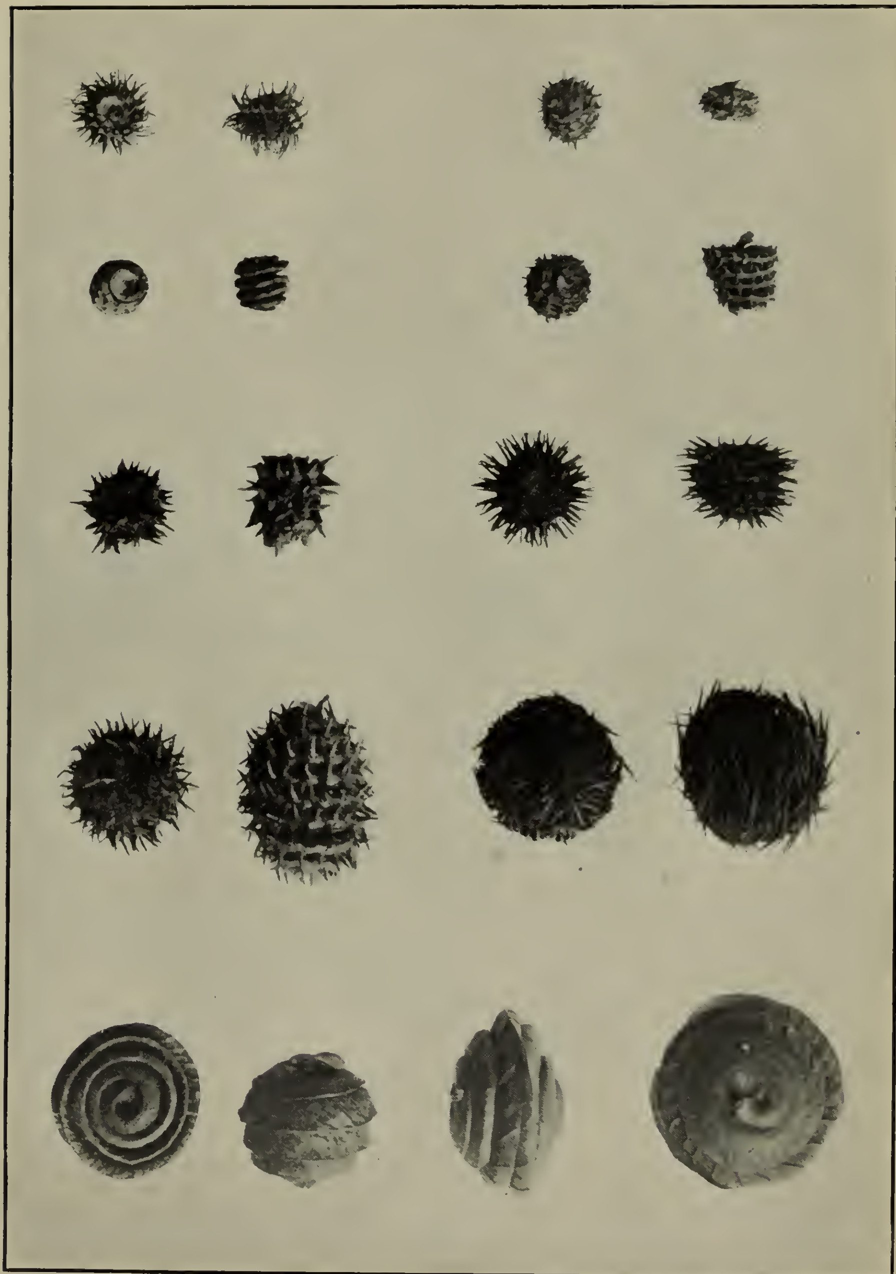
For various reasons a bur without spines is more desirable than one with spines. Spineless burs do not catch in the wool of sheep, an objectionable feature of ordinary bur clovers. On the other hand, they may be objected to on account of being deprived of this means of distribution, as the smooth burs will not hang to stock to be carried about and are a little harder to maintain in pasture, especially the larger podded varieties. Furthermore, the spineless burs are more readily eaten by stock. As already explained (p. 10), the weight of evidence favors the smooth bur.

STUDIES RELATING TO VARIATION IN THE BURS OF DIFFERENT SPECIES.

Since the fall of 1908 a large collection of bur clovers has been used in a study to determine to what extent the burs of the various species and subspecies vary from their normal type. This collection comprises 202 selections, and includes 20 species and subspecies.

It may be well to mention here a few difficulties encountered in the work. Like the seeds of many other legumes, fresh seeds of bur clover do not germinate readily. A common experience has been that the entire lot of seed of a selection failed to germinate. Some difficulty has been found in so protecting the plantings as to be perfectly sure that mice had not carried seed from one selection to another. To get soil absolutely free from bur clover seed is somewhat difficult where bur clover is naturally abundant, and it has necessitated extra care. The first year all the soil used was sifted through screens sufficiently fine to exclude any bur clover seed that it might contain. In the second and subsequent years the soil used was taken from 4 feet below the surface of the ground, at which depth it was found to be free from all germinable seed. The latter method is satisfactory at Chico, Cal., where the work has been carried on. Plants grown in soil from a depth of 4 feet have been found to make a growth quite as good as plants grown in soil taken at the surface.

In order that the comparison between the original burs selected and the burs of their progeny might be as accurate as possible, burs of each selection from which seed was taken for planting (or burs as nearly like the type as could be found) were saved for future comparison. In the descriptive records of the plants the following points were observed: (1) Pubescence, (2) leaf markings, (3) size and color of leaflets, (4) size and number of flowers, (5) size, color, and shape of stems, and (6) general notes. Besides descriptive notes, typical burs produced in each season were saved, together with herbarium specimens of most of them, so that comparison of any variation in the progeny from the original selections could be noted.



PODS OF TEN SPECIES OF MEDICAGO.

Top row, *M. arabica* and *M. hispida denticulata;* second row, *M. hispida confinis* and *M. hispida terebellum;* third row, *M. muricata* and *M. hispida nigra;* fourth row, *M. ciliaris* and *M. echinus;* bottom row, *M. scutellata* and *M. orbicularis.* (Natural size.)

BRANCH OF MEDICAGO MUREX (NO. 0147), SHOWING VARIATION IN CHARACTER OF PODS.

The plants have been grown each year in 7-inch pots. The plantings have been made each year in the fall, and the first year (1908–9) the plants were carried through the winter in a cool greenhouse. The second year (1909–10) the plants were carried through the winter in an ordinary lath house, and the third year (1910–11) in a glass-covered lath house. Every year in the spring the pots were plunged in soil to their entire depth and allowed to remain in the lath house until the seed was mature.

VARIATION IN MEDICAGO MUREX.

From plantings made in the fall of 1908 a very marked variation occurred in two selections (F. C. I. No. 0147 and S. P. I.[1] No. 16875) which had been received under the names *Medicago polycarpa* and *M. murex*, respectively. A single plant of each produced burs varying not only in shape, but ranging from spineless to forms having many medium-sized stout spines (Pl. IV). The variation with regard to spines was as great as could be. F. C. I. No. 0147 and S. P. I. No. 16875, which had been received under different specific names, proved to be identical, both being *M. murex.* In their first season and in subsequent years the plants of these two numbers were noted as being identical in general growth, size, shape, color, and markings of stems, leaves, and flowers.

In the fall of 1909 plantings were again made as in 1908, but this time seed from the progeny of the 1908 plantings was used. Burs of F. C. I. No. 0147 and S. P. I. No. 16875, both smooth and spiny and also including intermediate forms, were planted. Each series represented all types of burs taken from a single plant. The resulting plants from the varying types of burs were all alike, and they were like the plants of the year before. The following spring (1910) these plants fruited, and all the early burs were spiny and essentially like the spiny forms produced in the spring of 1909. To see whether adverse conditions would produce such a variation in the burs as occurred in 1909, pots containing the plants were lifted out of the ground in which they had been plunged, to cause them to dry out more readily. Abnormal or spineless burs began to form at once on the plants thus lifted. A week later a hot spell of a few days' duration dried all the plants severely, and spineless burs began to develop also on the plants not lifted. From that time on, hot weather prevailed and varying burs continued to form. (See Pl. X, fig. 1.)

In the fall of 1910 plantings were made again as in previous years. In the spring of 1911 pots were lifted as before, with the same general results. The burs without spines produced in 1911 were not as well developed as in 1909, but showed the same general variations.

[1] F. C. I. is an abbreviation for Forage-Crop Investigations; S. P. I., for Foreign Seed and Plant Introduction.

VARIATION IN MEDICAGO CILIARIS.

In the spring of 1910, at the time of lifting the pots of *Medicago murex*, pots of *M. ciliaris* were also lifted, to note the effect on burs of this species. Up to this time all the burs developed had been normal. In two or three days after lifting the pots the burs began to show elongation. Burs subsequently produced were decidedly elongated, and the spines were much shorter than normal. In the pots not lifted the plants continued to produce normal burs until severely checked by a hot spell some time later, after which abnormal burs were produced on all the plants. (See Pl. XI, fig. 1.) In the spring of 1911, pots of this species were again lifted as in the previous year, with the same general results.

VARIATION IN MEDICAGO MURICATA.

The pots of *Medicago muricata* were not lifted as were those of *M. murex* and *M. ciliaris*, but in the seasons of 1909, 1910, and 1911 it was noted that the burs formed late in the season varied greatly with regard to spines. In some instances the burs were nearly smooth and varied from this type to nearly normal burs. The burs formed earlier in the season were always normal and spiny. Plantings of this species had been made in the open field in the fall of 1910 and were observed to determine the extent of variation under field conditions. The results were practically the same as with the pot-grown plants. The burs which developed early were normal and with spines, while those formed late in the season varied from nearly smooth to nearly normal. (See Pl. IX, fig. 2.)

VARIATION IN OTHER SPECIES.

Besides *Medicago murex*, *M. ciliaris*, and *M. muricata* the following species were planted in the fall of 1908 and the two succeeding years: *M. scutellata*, *M. orbicularis*, *M. orbicularis microcarpa*, *M. orbicularis marginata*, *M. hispida*, *M. hispida nigra*, *M. hispida confinis*. *M. hispida apiculata*, *M. hispida denticulata*, *M. hispida terebellum*, *M. echinus*, *M. turbinata*, *M. tuberculata*, *M. rugosa*, *M. lupulina*, *M. radiata*, *M. arabica*, and *M. intertexta*.

Within each of these species, types and variants were selected from a general lot of burs and the seed planted, to note variations in the progeny. In none of these species was there the marked variation of burs on single plants noted in *Medicago murex* and *M. muricata*, but in every species the burs on individual plants varied somewhat in size and the spines varied more or less in length. In *M. hispida* and its subspecies the variation was so great that the smallest burs produced in the different subspecies were smaller than the typical burs of the nearest related subspecies having a smaller bur. (Pl. V, fig. 1.) In all cases the number of burs of the progeny that varied from the type of the species or subspecies was much less than the number of those that were typical.

It has been noted that burs of the same species vary in color, ranging from very dark or almost black to straw color. General observations had indicated that the dark color might be due to moisture in contact with the burs before and after ripening. That the dark colors were due to such contact was demonstrated very clearly on a large scale in the spring of 1911 at Chico, Cal. Until the burs were ripening the weather had remained clear and dry, and the burs developed were all light colored. Then a light rain fell during one night, and the next day all the burs that were fully developed and ripening (which included the greater part of the crop) turned black. No other rain fell, and all the burs that matured later were light colored and remained so.

The following species growing in the open at Chico at the time of the rain referred to were noted as showing a similar change in color: *Medicago orbicularis*, *M. orbicularis marginata*, *M. ciliaris*, *M. scutellata*, *M. turbinata*, *M. muricata*, *M. tuberculata*, *M. rigidula*, *M. hispida*, *M. hispida confinis*, *M. hispida apiculata*, *M. hispida terebellum*, *M. hispida nigra*, and *M. arabica*. The discoloration in *M. arabica* was not as marked as in the others.

To test this phenomenon artificially, burs of *Medicago orbicularis* that had developed without becoming wet or discolored were dampened and left over night; the next morning all had become dark, with the exception of a few burs that were mature and dry. It would seem from these observations that the dark-colored burs in all species are probably those that have come in contact with moisture during the period of their ripening and before they are fully mature. The color of the burs can not be used as a character on which to base botanical subspecies.

VARIETAL STRAINS

To determine whether varietal strains exist in different species or subspecies, a number of selections were made representing types with regard to length of spines and size and form of burs. Two types of burs of *Medicago scutellata* were selected and planted. In one type the bur was so coiled as to have a definite truncate end, while in the other the bur was more nearly conical. All the progeny of the truncated type produced truncated burs, and by far the greater part of the progeny of the conical burs produced conical burs and only a few tended to be truncated, as shown by the bulk of burs harvested.

A large number of burs of *Medicago orbicularis* and *M. orbicularis microcarpa* of various types were selected—double convex, convex on one side only, and burs open (or loosely coiled, as in *M. orbicularis marginata*). The seed from the selected burs was grown in the field in plats. Most of the burs from the progeny plants were like the type planted, as shown by the bulk of burs harvested.

In the *Medicago hispida* group, burs representing the different sizes and lengths of spines were planted. In a number of cases they reproduced true to the type selected. *M. hispida confinis* produced two types, the bur of one having 2½ to 3 turns and the other 4 to 4½ turns. Another type approaching that of *M. hispida confinis*, but with spines sufficiently developed to throw it out of that group, reproduced true to type. In the other groups of *M. hispida* the types were not so definitely marked, but a few showed variations. The other species showed no definite varietal strains, but the work was not extended enough to say that they do not exist. Thus far the work indicates that there exist definite varietal strains within at least several of the species and subspecies, and that these may be grown as pure strains by selection. On account of the variations that may occur within the different species, a type or variety can not be defined by the appearance of the burs in a bulk lot of seed. As already explained, an individual plant may produce burs that are as different as the burs of two subspecies, but when grown under normal conditions most of the burs on individual plants will be true to type.

POLLINATION IN THE VARIOUS SPECIES.

The flowers of the various species of bur clovers are similar in form but differ somewhat in size, in the number borne in a cluster, and in the details of the explosive mechanism. Seven of the species studied have a tripping mechanism similar to alfalfa, so that after tripping the stigma is exposed—*Medicago scutellata*, *M. rugosa*, *M. turbinata*, *M. muricata*, *M. rigidula*, *M. ciliaris*, and *M. echinus*. In *M. echinus* the flowers are in clusters of six. The other species have the flowers in clusters of two. *Medicago turbinata* often appears to have single flowers in a place, because one of each pair usually dies in the bud or withers shortly after it opens and fails to develop a pod.

All the bur clovers studied except *Medicago echinus* seem to be readily self-fertile. The various species have been grown in a greenhouse where little, if any, cross-pollination was probable, and with the exception of *M. echinus* all set pods freely. Alfalfa plants growing in the greenhouse beside the bur clovers set no seed except when artificially tripped. A number of flowers of *M. echinus* were tripped by means of a toothpick to determine the effect on seed setting. These

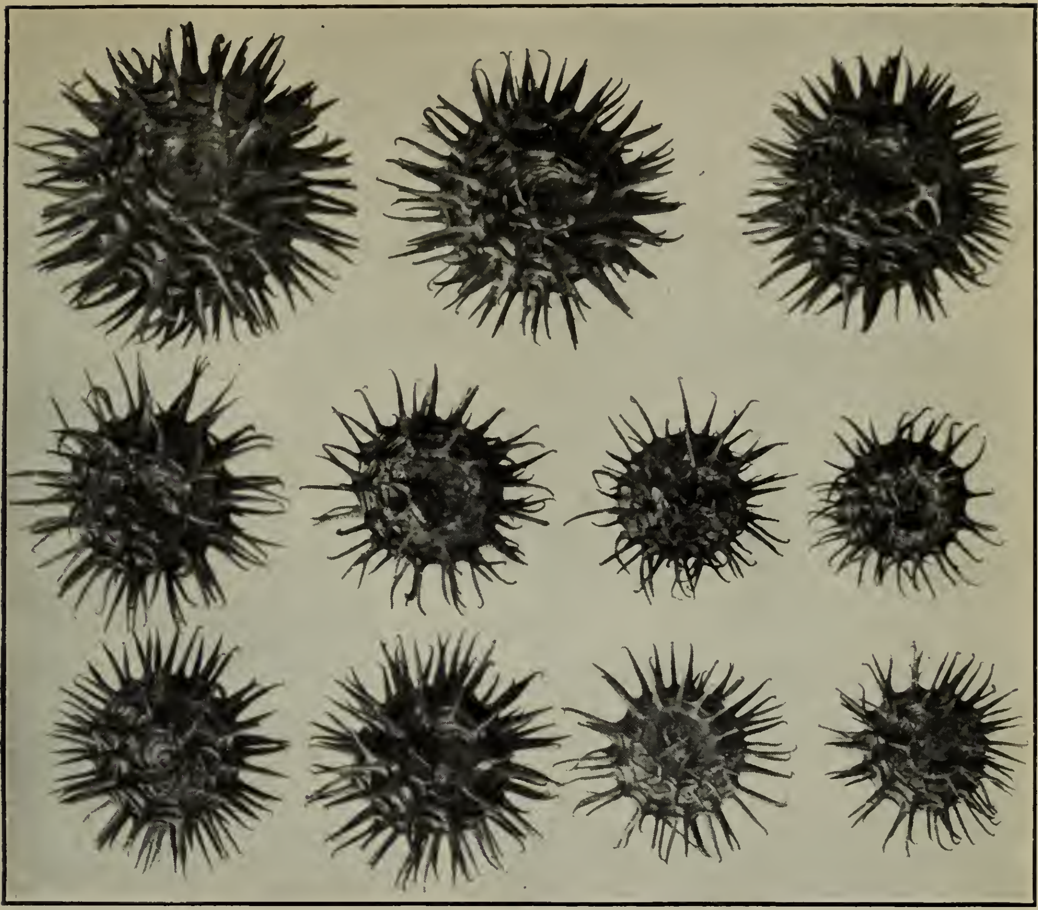


FIG. 1.—ROWS OF BURS FROM SINGLE PLANTS OF MEDICAGO, SHOWING VARIATION IN SIZE.

Upper row, *M. hispida nigra;* middle and lower rows, *M. hispida denticulata.* (Enlarged 2¾ diameters.)

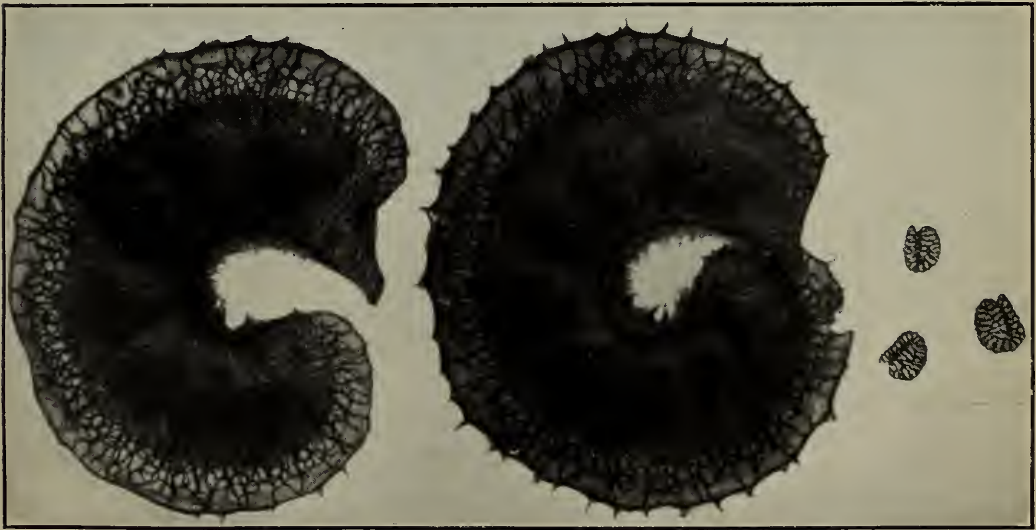


FIG. 2.—PODS AND SEEDS OF MEDICAGO RADIATA, SHOWING SEED AND VENATION OF PODS.

(Enlarged 2 diameters.)

FIG. 1.—PODS AND SEEDS OF MEDICAGO LUPULINA, SHOWING VENATION OF PODS AND PROMINENCE ON THE SEED AT THE TIP OF THE RADICLE.

(Enlarged 9 diameters.)

FIG. 2.—PODS AND SEEDS OF MEDICAGO SCUTELLATA, SHOWING VENATION AND WINDINGS OF PODS AND CHARACTERISTICALLY NOTCHED SEEDS.

(Enlarged 2 diameters.)

flowers, as well as the alfalfa flowers, set a number of pods, but the exact percentage was not determined. Other flowers were tripped and cross-fertilized with pollen from another plant of *M. echinus*, with the apparent result that more pods were set from these flowers than from those tripped but not cross-pollinated.

GENERAL CLASSIFICATION OF THE SPECIES.

The nonperennial species of Medicago here considered may for convenience be divided into six groups. The most of the species in each group are so nearly alike in flower and leaf characters that they are distinguished with certainty only when the pods have matured.

The first group contains a single species, *Medicago radiata*, the leaflets of which are mostly rather small and the stems somewhat woody, procumbent to erect. The pods are large, flattened, kidney shaped, and have a row of short, simple or sometimes forked spines along the back. The seeds have the surface somewhat convoluted and the radicle as long as the seed.

The second group contains one species, *Medicago lupulina*, having stems slightly procumbent or suberect in habit, with rather small, strigose-veined leaflets and small kidney-shaped pods.

The third group contains three species, *Medicago orbicularis*, *M. scutellata*, and *M. rugosa.* The stems in this group, as in the following three groups, are more procumbent and, unless the stand is thick, have a tendency to become trailing. The pods are rather large, disk shaped, consist of several thin spiral windings, are of a papery texture, and are without spines.

The fourth group contains four species, *Medicago rigidula*, *M. turbinata*, *M. tuberculata*, and *M. murex.* The pods of these species are harder in texture, more closely wound, of an oval form, and vary from smooth to tubercular and spiny.

The fifth group contains two species, *Medicago ciliaris* and *M. echinus*, which have rather large, closely wound, oval pods, with windings the edges of which are thickly covered with interlocking spines.

The sixth group contains two species, *Medicago arabica* and *M. hispida*, which have smaller, somewhat short cylindrical pods, with windings the edges of which are usually covered with more or less erect spines. Forms of both species are found without spines.

In the last four groups the flower and leaf characters are so nearly alike that the species can be distinguished with certainty only by the mature pods.

The seeds vary from 2 to 6 mm. (one-sixteenth to one-fourth inch) in length, and in the species here considered are yellow or greenish yellow in all but three species, *Medicago ciliaris* and *M. echinus* having black seeds and *M. orbicularis* having yellowish brown seeds. In all but two species here considered the seeds are kidney shaped and smooth, the exceptions being *M. radiata*, in which they are oval in outline with a convoluted surface, and *M. orbicularis*, in which they are obovoid and the surface papillose.

Many of the species upon which these studies are based were originally obtained in Algeria by Mr. C. S. Scofield in 1901. *Medicago orbicularis*, S. P. I. No. 10725, is also from Algeria, secured in 1902 by Mr. T. H. Kearney. The others are mainly from botanical gardens, especially that at Madrid, Spain, and a few are from miscellaneous sources.

DISTRIBUTION AND DESCRIPTION OF SPECIES.

The genus Medicago is at present widely distributed over southern Europe, western Asia, northern Africa, and the adjacent islands. Its northern limit seems to be southern Scotland, Sweden, and Siberia. A few species have become naturalized in recent years in Abyssinia, South Africa, and Chile. In the United States about 60 species and subspecies have been introduced for experimental purposes since 1898 through the Office of Seed and Plant Introduction. Some of these importations may become naturalized. Prior to 1898 five species had become well established along the eastern, western, and southern coasts of the United States. These species and two others more recently introduced, are gradually working their way inland, but their progress is slow. At present but two species are established in the Central States north of Texas.

MEDICAGO RADIATA L.

(Pl. V, fig. 2.)

Stems decumbent, pubescent, 10 to 30 cm. (4 to 12 inches) long; leaflets obovate to cuneate, downy to villous on both sides, 2 to 6 mm. (one-sixteenth to three-sixteenths inch) wide, 3 to 10 mm. (one-eighth to three-eighths inch) long, rounded, and toothed at the apex, the base entire; leafstalks equaling or twice the length of the leaves, pubescent, the stalk of the terminal leaflet five times longer than the lateral; stipules awl shaped, pubescent, entire, 2 to 4 mm. (one-sixteenth to one-eighth inch) long; flowers usually axillary, in clusters of two, 2½ to 3mm. (three thirty-seconds to one-eighth inch) long, the stigma not exposed when tripped, the peduncles and calyx pubescent; pods papery, brownish when ripe, sickle shaped to circular, 15 to 25 mm. (one-half to 1 inch) long, 7 to 10 mm. (one-fourth to three-eighths inch) wide, glabrous, netted veined, with a row of simple, sometimes forked spines about 1 mm. (one-sixteenth inch) in length along the outer side, and an irregularly toothed, membranous margin along the inner side, 5 to 6 seeded; seeds oval, flattened, light to yellowish brown, 2 to 2½ mm. (one-sixteenth to three thirty-seconds inch) long, surface convoluted, the radicle as long as the seed.

Distribution: Spain to Persia.

This species was received from Madrid, Spain, and Karlsruhe, Germany, under S. P. I. Nos. 9746 and 16266, respectively. It has been tested only at Chico, Cal. It makes little growth compared with toothed or spotted bur clover, and yields little seed.

MEDICAGO LUPULINA L. (YELLOW TREFOIL).

(Pl. VI, fig. 1.)

Stems four angled, pubescent to nearly glabrous, 10 to 80 cm. (4 to 32 inches) long, decumbent; leafstalks, 2 to 10 cm. (three-fourths to 4 inches) long, pubescent, the leaflets oval to broadly obovate or even obcordate, sometimes wedge shaped at the base, pubescent on both sides, 3 to 12 mm. (one-eighth to one-half inch) wide, 6 to 20 mm. (one-fourth to three-fourths inch) long, the base entire. stalk of the terminal leaflet 3 to 5 times longer than the lateral; stipules rather large, broad, and few toothed at the base; flowers very small, 1½ to 2 mm. (one-sixteenth inch) long, in rather close, oval, or oblong heads of 10 to 40 flowers, the stigma not exposed when tripped; pod kidney shaped, about 2 mm. (one-sixteenth inch) in diameter, netted veined, minutely pubescent, blackish when mature, one seeded; seed 1½ to 2 mm. (about one-sixteenth inch) long, yellow or greenish yellow.

Distribution: Spain to Scotland, east to Persia, and probably widely introduced into every civilized country.

This species is a semierect, leafy plant, usually biennial, but with annual and perennial forms. It occurs spontaneously both in the Pacific Coast States and in the Eastern States. It is hardy at least as far north as central New York and, in addition to its use as pasturage, is of promise as a green-manure crop. It is being tested in comparison with crimson clover in the Atlantic States, and while its actual yield of forage and green manure is not equal to crimson clover, the occasional high price of the seed of crimson clover makes this species potentially important. At the Arlington Experimental Farm, Va., it made a growth varying from 12 to 26 inches in height, depending on the character of the soil. It will grow on stiff clay soils somewhat too poor for the successful growth of alfalfa or red clover. It has been recommended as a constituent of lawn mixtures, since it remains green during rather severe drought. At Chico, Cal., it has been grown under F. C. I. No. 0268, seed received from the Botanical Gardens of Madrid, Spain, and also under S. P. I. No. 4340, from Naples, Italy. In hot weather it made considerably better growth than the other bur clovers under test. It is used to some extent in European pastures and is ordinarily regarded as being inferior to clover and alfalfa. Its use in the past to adulterate alfalfa and clover seed has caused it to be classed as a weed, but it is not troublesome in this respect, since it is readily eradicated by ordinary tillage. Its presence in uncultivated lands is not objectionable, but usually advantageous. Its short life makes it compare unfavorably with alfalfa, and its small size makes it less valuable than such plants as crimson clover or red clover, with which it must ordinarily compete in agricultural use.

MEDICAGO ORBICULARIS (L.) ALL. (BUTTON CLOVER).

(Pl. VII, fig. 1.)

Stems procumbent, 10 to 80 cm. (4 to 32 inches) long, sparingly pubescent; leaflets oval to obovate, sometimes truncate at the base, sparingly pubescent on both sides, up to 15 mm. (five-eighths inch) wide and 20 mm. (seven-eighths inch) long, rounded at the apex, the margin toothed nearly to the base, the leafstalks 1 to 10 cm. (three-eighths to 4 inches) long; stipules with slender teeth about 2 to 3 mm. (one-sixteenth to one-eighth inch) long; flowers 4 to 5 mm. (about three-sixteenths inch) long, in pairs, on an axillary peduncle, the stigma slightly exposed when tripped; pods papery, straw colored, netted veined, 18 to 20 mm. (about three-fourths inch) in diameter, twisted spirally into 4 to 6 thin, flattened turns, the margin often recurved, the central winding largest, the others gradually decreasing in size; seed yellowish brown, obovoid, flattened, about 2½ to 3 mm. (one-eighth inch) long, the surface minutely papillose, the radicle as long as the seed.

Distribution: France, Spain, and Algeria; thence east to Persia. In the United States it has been reported growing spontaneously only in Alabama and southern California.

This species was received from Algeria and from Brunswick, Germany, under S. P. I. Nos. 10725 and 16876, respectively. It has been tested most extensively at Chico and other places in California, but has also been tried in several localities in the South Atlantic and Gulf Coast States. It promises well for pastures in California, but has not been sufficiently tested in the Southern States to determine its value there. In general its growth is about like the spotted and toothed bur clovers, but in California it yields very much more seed than either of these species. Its large spineless burs and heavy yields of seed make it superior to common species for pasturage. It matures earlier and is affected much less by the clover-seed chalcis than the toothed or spotted bur clovers.

MEDICAGO ORBICULARIS MICROCARPA ROUY AND FOUC.

(Pl. VII, fig. 2.)

This subspecies differs from typical *Medicago orbicularis* only in having uniformly smaller pods, varying from 8 to 12 mm. (one-fourth to one-half inch) in diameter, having the same season of growth.

Distribution: Same as for the species.

This importation was from near Oued Smaar, Algeria, under S. P. I. No. 7738. Tested in California and in the Southern States in comparison with the species it shows no difference in agronomic value.

MEDICAGO ORBICULARIS MARGINATA (WILLD.) BENTH.

(Pl. VII, fig. 2.)

This subspecies differs from typical *Medicago orbicularis* in having pods with looser windings, all of the same diameter, and the margins always straight, never recurved.

Distribution: Same as for the species.

This importation was from Karlsruhe, Germany, under S. P. I. No. 16265, and has been tested at Chico, Cal. It makes less growth than the species proper, yields very much less, and therefore is of much less value.

MEDICAGO SCUTELLATA (L.) WILLD. (SNAIL CLOVER).

(Pl. VI, fig. 2.)

Plants densely glandular pubescent throughout; stems procumbent, 10 to 75 cm. (4 to 30 inches) long; leaves oval, oblong, or obovate, rarely broadly cuneate at the base; leaflets up to 15 mm. (five-eighths inch) wide, and 25 mm.

FIG. 1.—PODS AND SEEDS OF MEDICAGO ORBICULARIS, SHOWING VENATION AND WINDINGS OF PODS AND MARKINGS OF SEED COATS.

(Enlarged 2 diameters.)

FIG. 2.—PODS OF TWO SUBSPECIES OF MEDICAGO ORBICULARIS, MARGINATA (UPPER ROW) AND MICROCARPA (LOWER ROW), SHOWING VARIATION IN WINDINGS AND SIZE.

(Enlarged 2 diameters.)

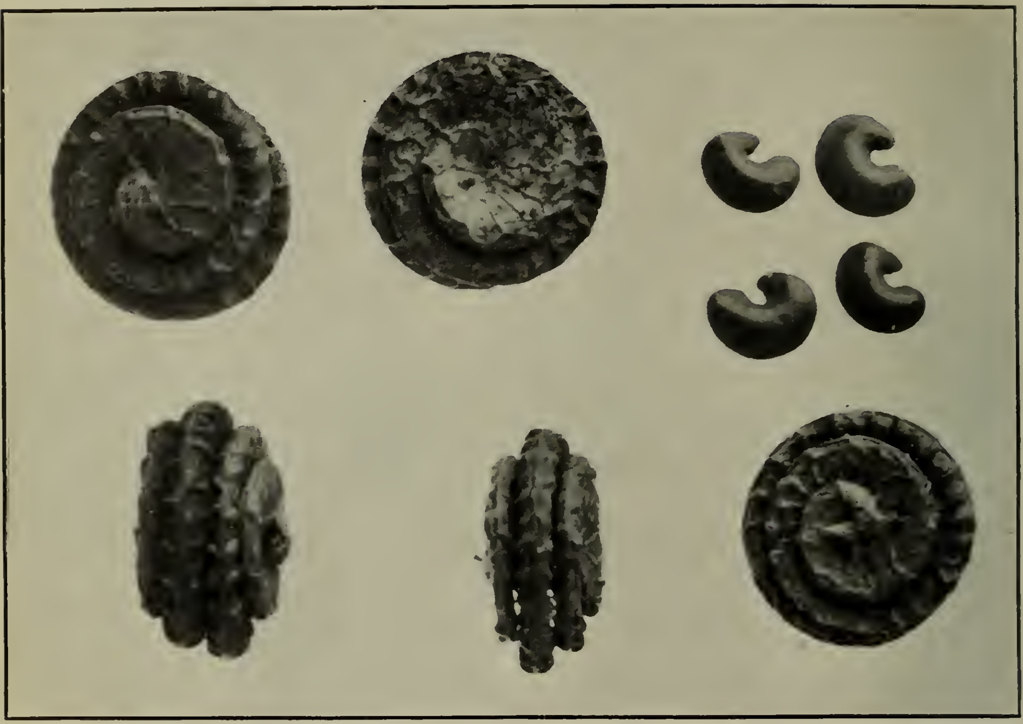


FIG. 1.—PODS AND SEEDS OF MEDICAGO RUGOSA, SHOWING VENATION AND WINDINGS OF PODS AND CHARACTERISTICALLY NOTCHED SEEDS.

(Enlarged 2 diameters.)

FIG. 2.—PODS AND SEEDS OF MEDICAGO TUBERCULATA, SHOWING VARIATION OF WINDINGS AND SHORT TUBERCULAR SPINES OF PODS AND CHARACTERISTIC NOTCHES IN SEEDS.

(Enlarged 2 diameters.)

(1 inch) long, coarsely and sharply toothed nearly to the base, the apex obtuse to acute, the leafstalk rarely much longer than the leaf; stipules up to 4 mm. (three-sixteenths inch) wide, and 10 mm. (three-eighths inch) long, sparingly toothed; flowers about 7 mm. (one-fourth inch) long, in clusters of two, the stigma exposed when tripped; pods 12 to 15 mm. (one-half to five-eighths inch) in diameter, straw colored, netted veined, with 5 to 8 thin, spiral, cup-shaped windings; seed kidney shaped, yellow, 5 to 5½ mm. (about three-sixteenths inch) long, the radicle half the length of the seed.

Distribution: Spain and Algeria to Asia Minor.

This species was received under S. P. I. Nos. 9747 from Madrid, Spain; 16267 from Karlsruhe, Germany; and 16877 from Brunswick, Germany. It has been grown at Chico and other points in California and at several places in the Southern States. It matures earlier than any of the other species tested, and makes less growth than the common spotted and toothed bur clovers. In comparison with others it makes a heavy yield of seed under California conditions, but has done little in tests in the Southern States. On account of its large smooth burs, good yield of seed, and early maturity it is valuable for pasturage in California. The seeds are little attacked by the clover-seed chalcis.

MEDICAGO RUGOSA DESR.

(Pl. VIII, fig. 1.)

Plants sparingly glandular pubescent throughout, except on the upper surface of the leaves; stems decumbent, 10 to 60 cm. (4 to 24 inches) long; leaflets up to 15 mm. (five-eighths inch) wide and 20 mm. (seven-eighths inch) long, broadly truncate and entire at the base, the apex rounded or retuse, sharply toothed, the leafstalk up to 5 cm. (2 inches) long; flowers about 5 mm. (three-eighths inch) long, in clusters of two, the stigma slightly exposed when tripped; pods 7 to 8 mm. (one-fourth to five-sixteenths inch) in diameter, the windings 2½ to 4, somewhat inflated, with conspicuous, radiating, marginal striæ; seed kidney shaped, yellow, 3 to 4 mm. (about one-eighth inch) long.

Distribution: Syria, Mesopotamia, and Palestine.

This species was received from Madrid, Spain, under S. P. I. No. 19442. It has been tried only at Chico, Cal., makes much less growth than the spotted or toothed bur clovers, and yields considerably less seed. Its season is practically the same as the spotted or toothed bur clovers. It is little attacked by the clover-seed chalcis. The absence of spines makes it desirable for further testing.

MEDICAGO TUBERCULATA (RETZ.) WILLD.

(Pl. VIII, fig. 2.)

Stem procumbent, 10 to 60 cm. (4 to 24 inches) long, sparingly villous, with brownish hairs; leaflets pubescent on both sides, obovate to ovate, up to 15 mm. (five-eighths inch) wide, and 24 mm. (three-fourths inch) long, the apex rounded to acute, closely toothed, the base truncate, the terminal pedicels three to five times longer than the lateral; stipules not deeply toothed; flowers about 5 mm. (three-sixteenths inch) long, in clusters of 5, the stigma not exposed when tripped; pod 6 to 7½ (one-fourth to five-sixteenths inch) in diameter, twisted spirally into 4 to 5 turns, a few radiating veins near the center of the turn surrounded by a smooth band bordered by a vein parallel to the dorsal suture. The parallel vein and dorsal suture are connected by numerous radiating veins, these are usually swollen at the base when mature. Seed kidney shaped, yellow, about 4 mm. (one-eighth inch) long, radicle nearly half as long as the seed.

Distribution: France to Algeria; thence east to Syria.

This species was received from the Royal Botanic Gardens, Dublin, under F. C. I. No. 9229 and S. P. I. No. 17783. It has been grown only at Chico, Cal. Its season is practically the same as the spotted or toothed bur clovers. but it makes much less growth and yields little seed. It is worthy of further testing on account of its spineless burs.

MEDICAGO TURBINATA (L.) ALL.

(Pl. IX, fig. 1.)

Plant more or less densely pubescent throughout; stems procumbent, 10 to 60 cm. (4 to 24 inches) long; leaflets up to 16 mm. (five-eighths inch) wide and 25 mm. (1 inch) long, the leafstalk not much longer than the leaf. the stalk of the terminal leaflet five to eight times longer than the lateral; stipules not deeply toothed; flowers about 5 mm. (three-sixteenths inch) long, single, or rarely in clusters of two, the stigma exposed when tripped; pod 7 to 8 mm. (one-fourth to five-sixteenths inch) in diameter, twisted spirally into about 5 windings, which are smooth, woody, and sparingly covered with short, stiff, tubercular-pointed spines; seed kidney shaped, light yellow, 5 to 6 mm. (three-sixteenths to one-fourth inch) long. Pods without spines sometimes appear in dry seasons.

Distribution: Portugal and Algeria to Asia Minor.

This importation is represented by S. P. I. Nos. 19447 and 19449, both probably from the Madrid Botanical Gardens. It has been grown at Chico and other places in California. Its season of maturing is about the same as or a little later than toothed bur clover, but its growth is not as great, and its hard, woody, spiny pod makes it less desirable for pasture. It is little affected by the clover-seed chalcis.

MEDICAGO MURICATA (L.) ALL.

(Pl. IX, fig. 2.)

Plant pubescent throughout; stems procumbent, 10 to 50 cm. (4 to 20 inches) long; leaflets up to 14 mm. (nine-sixteenths inch) wide and 18 mm. (three-fourths inch) long, the leafstalks about as long as the leaves, the stalk of the terminal leaflet five to eight times longer than the lateral; stipules not deeply toothed; flowers in twos, about 5 mm. long, the stigma exposed when tripped; pod 6 to 7 mm. (about one-fourth inch) in diameter, spirally twisted into five to six windings, the lateral veins parallel to the dorsal suture, bearing nearly opposite, stiff, sharp spines as long as the diameter of the windings, the edges of the windings partly obscured by the slightly interlocking prickles; seed about 3 mm. (one-eighth inch) long, yellow, somewhat kidney shaped or angular along the back, the radicle half as long as the seed and the end turned up so as to form a small beak near the hilum.

Distribution: Canary Islands, both shores of the Mediterranean, and east to Syria and Egypt.

This species was received from Madrid, Spain, under S. P. I. No. 9743. It has been grown only at Chico, Cal. The season of maturing is the same as toothed bur clover, but it makes much less growth. Its yield of seed is light, and its hard burs, with sharp, heavy spines, make it undesirable for pasturage.

MEDICAGO MUREX (L.) ALL.

(Pl. X, fig. 1.)

Stems procumbent, glabrous, 10 to 50 cm. (4 to 20 inches) long; leaflets glabrous above, pubescent beneath, the leafstalk about as long as the leaf, the stalk of the terminal leaflet about 5 times longer than the lateral; stipules

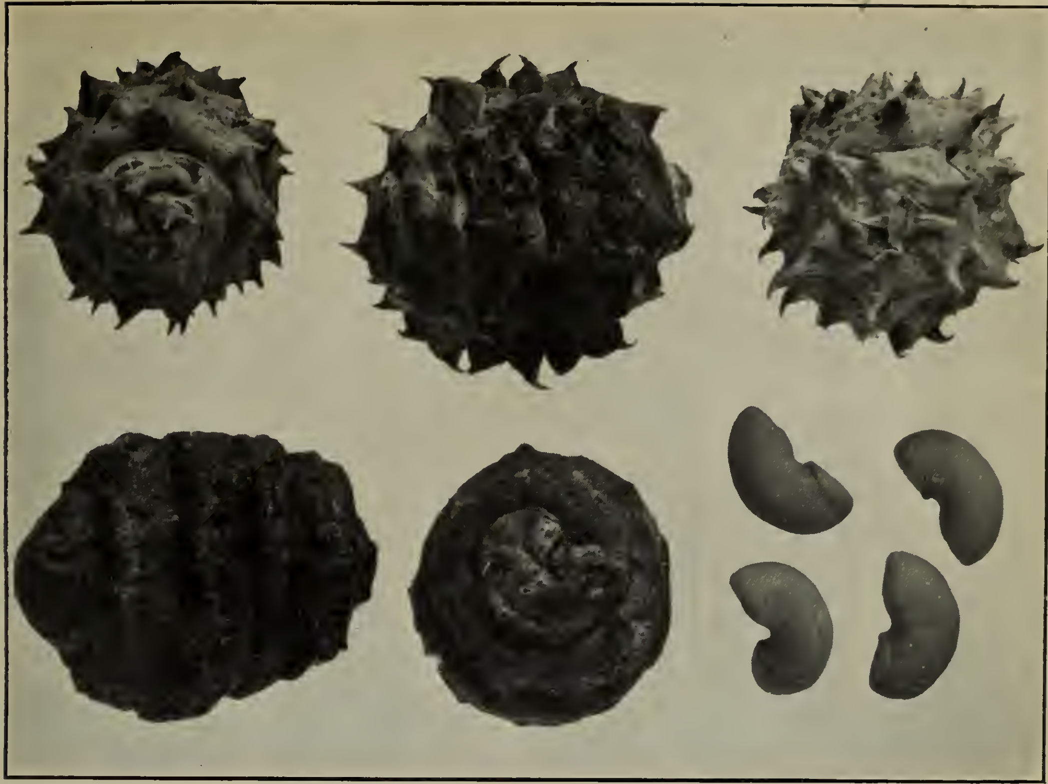


FIG. 1.—PODS AND SEEDS OF MEDICAGO TURBINATA, SHOWING WINDINGS OF PODS, SPINY AND SPINELESS PODS, AND SEEDS STRAIGHT AND NOTCHED ON THE UNDER SIDE.

(Enlarged 2 diameters.)

FIG. 2.—PODS AND SEEDS OF MEDICAGO MURICATA, SHOWING WINDINGS OF PODS, SPINY AND NEARLY SPINELESS PODS, AND ANGULAR SEEDS WITH PROMINENCE AT THE TIP OF THE RADICLE.

(Enlarged 2 diameters.)

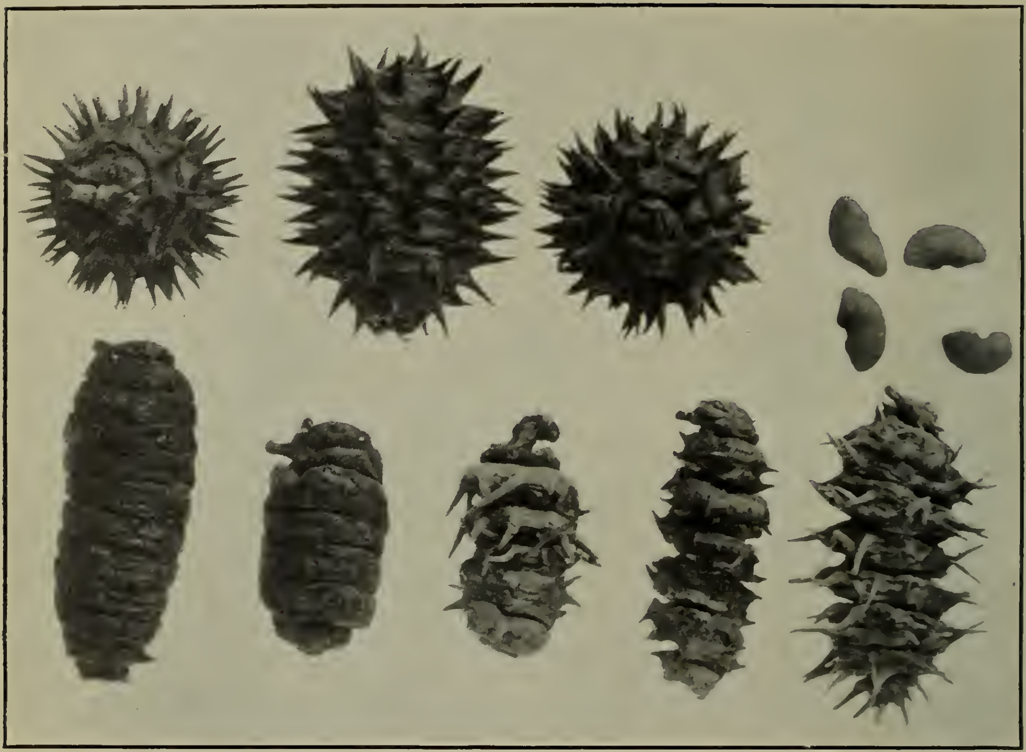


FIG. 1.—PODS AND SEEDS OF MEDICAGO MUREX, SHOWING VARIATION OF PODS OFTEN FOUND ON ONE PLANT.

(Enlarged 2 diameters.)

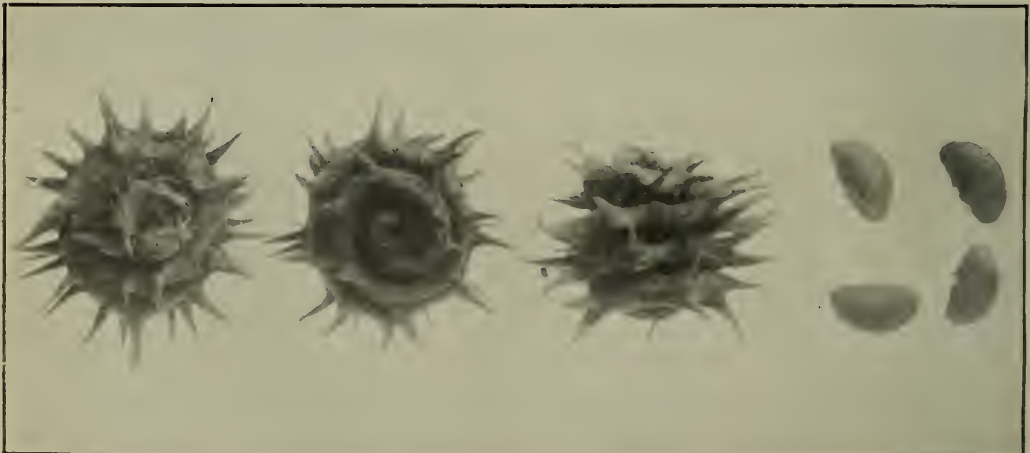


FIG. 2.—PODS AND SEEDS OF MEDICAGO RIGIDULA, SHOWING WINDINGS OF PODS, VENATION OF PODS HIDDEN BY PUBESCENCE, AND PROMINENCE AT THE TIP OF THE RADICLE.

(Enlarged 2 diameters.)

rather deeply toothed, the teeth 3 to 4 mm. (one-eighth inch) long; flowers in twos, about 4 mm. (three-sixteenths inch) long, the stigma not exposed when tripped. The pods are very similar in size and appearance to those of the preceding species, but have 7 to 9 turns, and the spines are more erect and do not interlock. Pods with few or no spines are produced in dry weather. The seeds are kidney shaped, about 3 to 4 mm. (one-eighth inch) long, and the radicle is nearly half the length of the seed.

Distribution: France, Italy, Turkey, and Algeria.

This species is represented by F. C. I. No. 0147, from Jamaica, and S. P. I. No. 16875, from Brunswick, Germany. It has been tested only at Chico, Cal. It has practically the same season as toothed bur clover, but makes much less growth. Its spiny burs make it objectionable for pasturage.

MEDICAGO RIGIDULA (L.) DESR.

(Pl. X, fig. 2.)

Plants pubescent throughout; stems procumbent, 10 to 50 cm. (4 to 20 inches) long; leaflets up to 12 mm. (one-half inch) wide and 24 mm. (1 inch) long, the leafstalk often equaling the leaves, but mostly shorter, the stalk of the terminal leaflet 3 to 4 times longer than the lateral; stipules not deeply toothed; flowers in twos, about 5 mm. (three-sixteenths inch) long, the stigma exposed when tripped; pods 7 to 8 mm. (about five-sixteenths inch) in diameter, similar to the preceding species, the windings not so thick and covered with a fine pubescence, the spines somewhat hooked at the tips; seed about 4 mm. (five thirty-seconds inch) long, yellow, kidney shaped, the radicle about half the length of the seed, the tips slightly raised.

Distribution: France and Spain; thence east to the Caucasus, Persia, and Egypt.

This species was received from Madrid, Spain, under F. C. I. Nos. 0373 and 0377, and from Strasburg, Germany, as S. P. I. No. 16288. It has been tested only at Chico, Cal. It makes much less growth than the toothed or spotted bur clovers and yields little seed. Its comparatively hard pod and stiff spines make it less desirable for pasturage.

MEDICAGO CILIARIS (L.) ALL.

(Pl. XI, fig. 1.)

Stems decumbent, glabrous, 10 to 100 cm. (4 to 40 inches) long; leaflets up to 17 mm. (five-eighths inch) wide and 30 mm. (1¼ inches) long, pubescent beneath, the leafstalks slightly pubescent, about equaling the leaves, the stalk of the terminal leaflet 3 to 5 times longer than the lateral; stipules not deeply toothed; flowers about 7 mm. (five-sixteenths inch) long, in twos, the stigma exposed when tripped; pod pubescent, 7 to 12 mm. (five-sixteenths to one-half inch) in diameter, 10 to 22 mm. (three-eighths to seven-eighths inch) long, 7 to 10 spiral windings thickly covered with stiff, somewhat interlocking spines about 2 to 3 mm. (one-eighth inch) long; seed 5 to 6 mm. (three-sixteenths to one-fourth inch) long, kidney shaped, black.

Distribution: France to Madeira; thence east to Asia Minor and Mesopotamia.

This species includes S. P. I. Nos. 7742, from Oued Smaar, Algeria, and 9747 and 19435, from Madrid, Spain. It has been tested at Chico and other points in California. It has practically the same season as toothed or spotted bur clover and makes equally good growth. Its very spiny pods make it less desirable for pasturage, though it produces a comparatively good crop of seed.

MEDICAGO ECHINUS DC. (CALVARY CLOVER).[1]

(Pl. XI, fig. 2.)

Stems procumbent, glabrous, 10 to 100 cm. (4 to 40 inches) long; leaflets up to 15 mm. (five-eighths inch) wide and 24 mm. (1 inch) long, pubescent beneath or only along the midrib, usually marked with a small reddish spot in the center of each leaflet, the leafstalk about as long as the leaf, the stalk of the terminal leaflet 3 to 5 times larger than the lateral; stipules not deeply toothed; flowers about 7 mm. (five-sixteenths inch) long, in clusters of 6, the stigma exposed when tripped; pod ovoid to spheroid, glabrous, 10 to 15 mm. (three-eighths to five-eighths inch) wide, 15 to 20 mm. (five-eighths to three-fourths inch) long, 7 to 9 spiral windings, thickly covered with slender, rigid, closely interlocking spines 5 to 7 mm. (about one-fourth inch) long; seed 5 to 6 mm. (three-sixteenths to one-fourth inch) long, kidney shaped, black.

Limited observation indicates that it is essential that the stigma be tripped as in alfalfa before the seed will set, and that crossing is advantageous to seed setting.[2]

Distribution: Spain and the Canary Islands; thence east to Italy and Algeria.

This species was selected from S. P. I. No. 7742, from near Oued Smaar, Algeria. It has been tested only at Chico, Cal. It has practically the same season as the toothed and spotted bur clovers and makes equally as good growth. Its very spiny pods make it less desirable for pasturage, though it produces a comparatively good crop of seed.

The subspecies *Medicago echinus variegata* (Urban) Ricker (*M. intertexta echinus variegata* Urban) differs from typical *M. echinus* in having a large, triangular, dark-reddish spot extending from the base to near the middle of the leaflet. The stems are less decumbent than those of the typical form, and the season of maturing is a little later. The name is a new trinomial.

Distribution: Same as for the species.

This importation was received from Brunswick, Germany, under S. P. I. No. 16874, and has the same agronomic value as the species. It perhaps can be used to advantage in California as an ornamental plant for winter border or bedding work.

MEDICAGO ARABICA (L.) ALL. (SPOTTED BUR CLOVER).

(Pl. XII, fig. 1.)

Stems procumbent, pubescent, 10 to 100 cm. (4 to 40 inches) long; leaflets up to 22 mm. (seven-eighths inch) wide, 27 mm. (1$\frac{1}{16}$ inches) long, pubescent beneath, a dark-red spot in the center of each leaflet, the leafstalk often 4 to 5 times longer than the leaf, the stalk of the terminal leaflet not more than 2 to 3 times longer than the lateral; stipules not deeply toothed; flowers 4 to 5 mm. (about three-sixteenths inch) long, in clusters of 5 to 10; pods 3½ to 5 mm. (one-eighth to three-sixteenths inch) in diameter, rather soft, twisted into 3 to 5 spiral windings, the edges bearing numerous interlocking grooved spines about as long as the width of a winding, the veins inconspicuous; seed about 2½ mm. (three thirty-seconds inch) long, kidney shaped, the radicle somewhat

[1] Also called "crown of thorns," the name being derived from the suggested resemblance of one of the windings (Pl. XI, fig. 2).

[2] Proceedings, Cambridge Philosophical Society, vol. 8, 1894, pp. 142–143. Bulletins, Kansas Agricultural Experiment Station: No. 151, 1907, p. 101; No. 155, 1908, p. 319. Circular 24, Bureau of Plant Industry, 1909, p. 8.

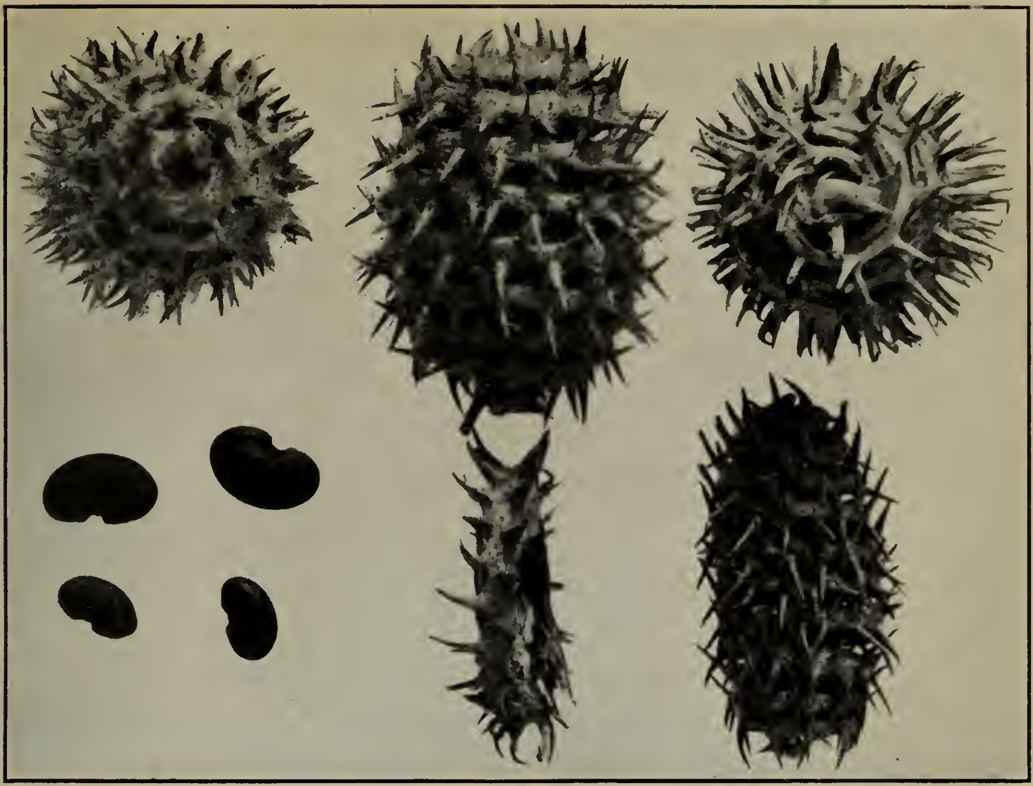


FIG. 1.—PODS AND SEEDS OF MEDICAGO CILIARIS, SHOWING VARIATION IN SHAPE OF PODS, CHARACTER OF WINDINGS, HAIRY SPINES, AND NOTCHED SEED.

(Enlarged 2 diameters.)

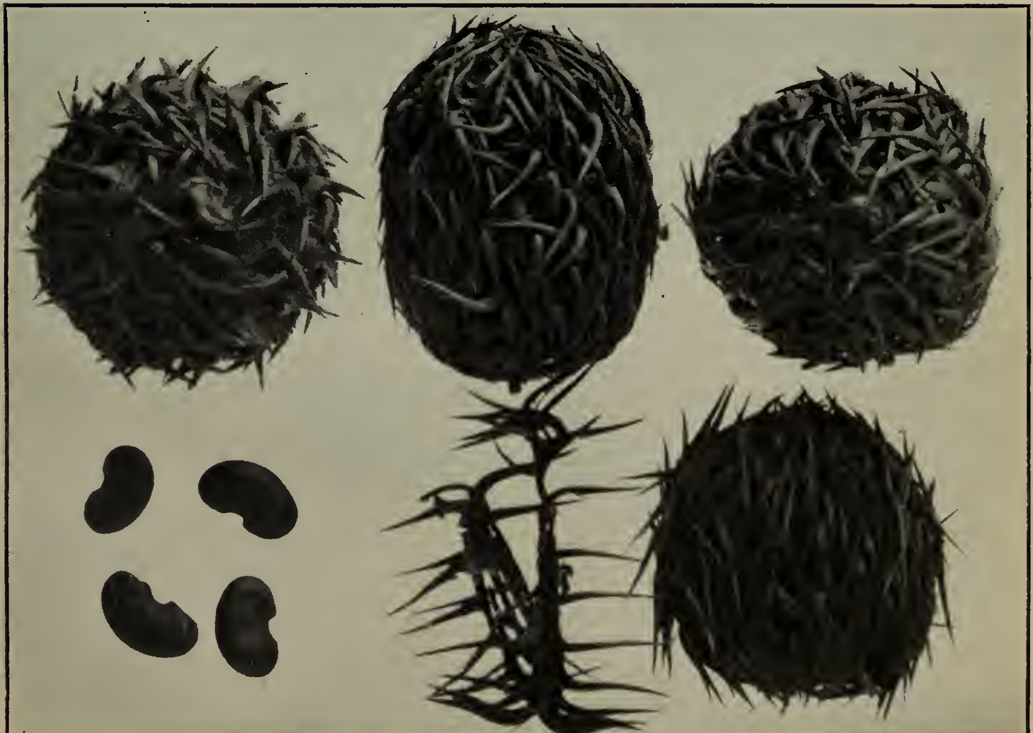


FIG. 2.—PODS AND SEEDS OF MEDICAGO ECHINUS, SHOWING INTERLOCKING SPINES, A SINGLE WINDING (LIKE A CROWN OF THORNS), AND NOTCHED SEED.

(Enlarged 2 diameters.)

FIG. 1.—PODS AND SEEDS OF MEDICAGO ARABICA (UPPER ROW) AND ITS SUBSPECIES INERMIS (LOWER ROW), SHOWING CHARACTER OF WINDINGS, LACK OF VENATION, AND PROMINENCE AT THE TIP OF THE RADICLE.

(Enlarged 2 diameters.)

FIG. 2.—PODS AND SEEDS OF MEDICAGO HISPIDA (UPPER ROW) AND ITS SUBSPECIES NIGRA (LOWER ROW), SHOWING VENATION OF PODS, DIFFERENCE IN NUMBER OF WINDINGS, LENGTH OF SPINES, AND SHAPE AND SIZE OF SEEDS.

(Enlarged 2 diameters.)

more than half the length of the seed, the end slightly raised, forming a small beak near the hilum.

Distribution: Ireland to Algeria; thence east to Asia Minor. Established throughout the Atlantic, Gulf, and California coasts and extending rapidly into the interior States. It is reported from Colorado.

This has been extensively tested in the South Atlantic and Gulf Coast States under S. P. I. Nos. 16213, 21550, 23661, and 25878, all from American sources. It is common throughout this region, and on account of its hardiness has proved the best bur clover for the region designated. It also does well in California, where the spiny form is common. Its season for maturing is medium as compared with other species. It is usually in bloom from early until late spring. Wherever it does well it affords a large amount of pasturage both in the green and the dry state.

MEDICAGO ARABICA INERMIS RICKER (NEW SUBSPECIES).

(Pl. XII, fig. 1.)

This subspecies differs from typical *Medicago arabica* only in the absence of spines on the pod. Many seeds of this subspecies have been planted at Chico. Cal. In every instance they have produced pods true to type and without spines.

Distribution: Seed received without definite statement of locality (S. P. I. No. 23284) from Mr. José D. Husbands, of Santiago, Chile. The type specimen grown from these seeds and collected by Mr. Roland McKee at Chico, Cal., June 1, 1910, has been deposited in the United States National Herbarium and a duplicate in the Economic Herbarium of the United States Department of Agriculture.

This importation has been grown only at Chico, Cal. In growth and season it is the same as typical *Medicago arabica*. On account of its spineless pods it gives promise of being especially serviceable in the Middle Atlantic and Gulf Coast States, where spotted bur clover in particular does well.

MEDICAGO HISPIDA GAERTN. (TOOTHED BUR CLOVER).

(Synonym, *M. hispida lappacea* (Desr.) Urban.)

(Pl. XII, fig. 2.)

Stems decumbent, glabrous, 10 to 110 cm. (4 to 44 inches) long, and up to 3 mm. (one-eighth inch) in diameter; leaflets broadly obovate to obcordate, glabrous above, sparingly pubescent beneath, up to 22 mm. (seven-eighths inch) wide and 28 mm. (1⅛ inches) long, often containing very small scattered whitish and dark-red spots, which disappear with age or drying, the apex finely toothed and emarginate, the base interruptedly toothed to entire, the stalk of the lateral leaflets very short, the terminal leaflet about five times longer than the lateral; stipules with linear teeth up to 3 mm. (one-eighth inch) long; flowers 4 to 5 mm. (one-eighth to three-sixteenths inch) long, in loose clusters of 6 to 9 on axillary peduncles, the stigma not exposed when tripped; pods netted veined, 7 to 10 mm. (one-fourth to three-eighths inch) in diameter, twisted spirally into 1½ to 4 windings with a conspicuous vein on each side of the winding parallel to the dorsal suture, and a double row of nearly opposite, stiff, erect, or slightly divergent spines connecting the dorsal suture and its parallel vein, the length of the spines being from one-half to full width of the windings; seed from light to brownish yellow, about 3 mm. (one-eighth inch) long, kidney shaped, the radicle half the length of the seed.

Distribution: The species and all its subspecies are natives of the northern Mediterranean regions, and are now found throughout the region from Spain to southern Germany and east to Central Asia and India. The species has been introduced extensively in Chile and Argentina and is becoming widely distributed in California.

This species was received under F. C. I. No. 0301 from Cambridge, England, and under S. P. I. No. 9736 from Madrid, Spain. It occurs commonly in California, and has been grown in comparative tests at Chico and other places in that State. It makes a good growth and is one of the best of the bur clovers in that respect. Its season for maturing is medium as compared with the other species, being in bloom in California from March until June, though in damp places it is found in bloom throughout the year. It is one of the forms of toothed bur clover already discussed (p. 8). Where it does well it affords a large amount of pasturage, either green or dry. The seed of this species and all its subspecies are badly attacked by the clover-seed chalcis. On account of its spiny burs it is less desirable than the spineless forms for pasturage.

MEDICAGO HISPIDA CONFINIS (KOCH) BURNAT.

(Pl. XIII, fig. 1.)

This subspecies differs from typical *Medicago hispida* in the general absence of leaf markings, in having rarely a small spot at the base of the leaf, and in having 1½ to 3½ windings and no spines. Short veins connecting the dorsal suture and lateral veins replace the spines.

Distribution: Same as for the species.

This importation was received under F. C. I. No. 0309 from Berlin, Germany, and under S. P. I. No. 9737 from Madrid, Spain. It has been tested at Chico and other places in California and in several localities in the Southern States. It has the same season as the species proper. It makes a good growth and is one of the best of the bur clovers in this respect. Its pods are spineless and, for this reason, it is especially desirable for pasturage. This form of the toothed bur clover is more fully discussed elsewhere (p. 20).

MEDICAGO HISPIDA RETICULATA (BENTH.) URBAN.

(Pl. XIII, fig. 1.)

This subspecies differs from *Medicago hispida confinis* only in having a pod with five windings. It has been occasionally found as a mixture with other species tested at Chico, Cal. Its growth and season are the same as the typical *M. hispida*. On account of its spineless bur it is desirable for pasturage.

Distribution: Same as for the species proper.

MEDICAGO HISPIDA APICULATA (WILLD.) URBAN.

(Pl. XIII. fig. 1.)

This subspecies differs from *Medicago hispida reticulata* only in the presence of short spines about as long as the thickness of the windings of the pod. It was received under F. C. I. Nos. 0266 and 0372 from Madrid, Spain, and under S. P. I. Nos. 16873 from Brunswick, Germany, and 19431 and 19434 from Madrid, Spain. It has been tested at Chico and other places in California. Its growth and season are the same as the species. It is preferable for pasturage to the more spiny form.

Distribution: Same as for the species proper.

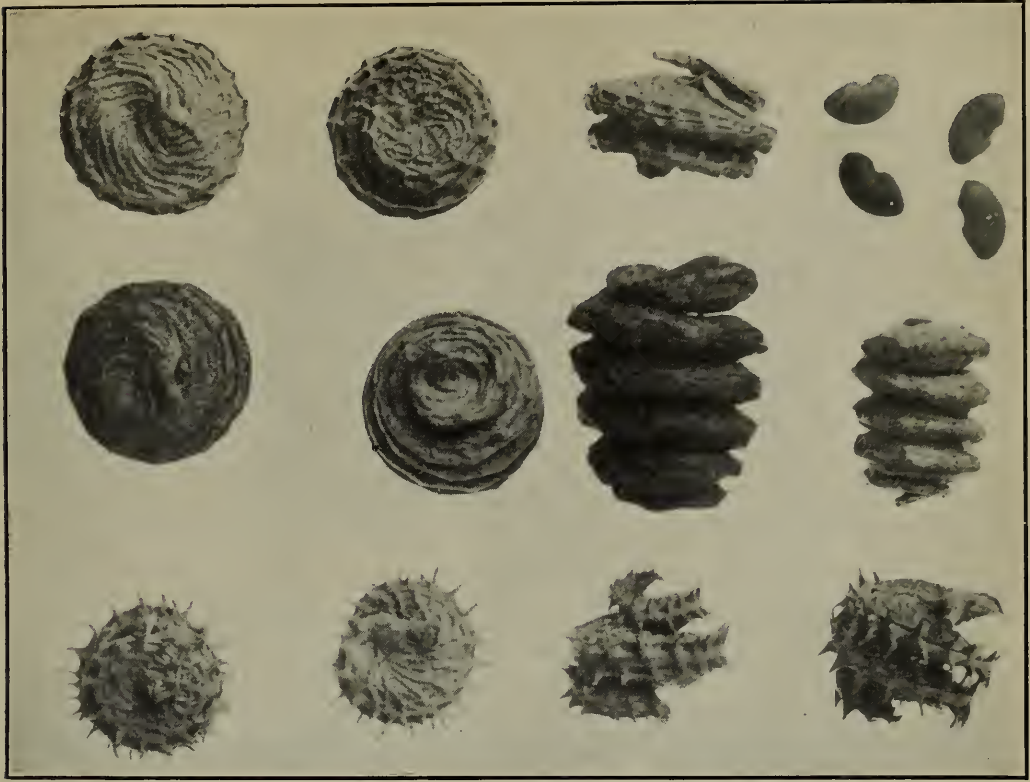


FIG. 1.—PODS OF THREE SUBSPECIES OF MEDICAGO HISPIDA, SHOWING DIFFERENCES IN WINDINGS, VENATION, ABSENCE OF SPINES, ETC.; ALSO SLIGHTLY NOTCHED SEEDS OF ONE SUBSPECIES.

Upper row, *M. hispida confinis;* middle row, *M. hispida reticulata;* lower row, *M. hispida apiculata.* (Enlarged 2 diameters.)

FIG. 2.—PODS AND SEEDS OF TWO SUBSPECIES OF MEDICAGO HISPIDA, SHOWING DIFFERENCES IN WINDINGS, IN LENGTH OF SPINES, AND IN SIZE AND SHAPE OF SEEDS.

Upper row, *M. hispida denticulata;* lower row, *M. hispida terebellum.* (Enlarged 2 diameters.)

MEDICAGO HISPIDA DENTICULATA (WILLD.) URBAN.

(Pl. XIII, fig. 2.)

This subspecies differs from *Medicago hispida apiculata* in having longer spines, about as long as half the width of the windings of the pod. This form has been received from various sources under F. C. I. Nos. 0149, 0151, 0262, 0271, 0272, 0273, 0280, 0374, 0382, 0884, and S. P. I. Nos. 19444, 19450, 19452, 19453, 19455, 20715, 22649, 24596.

This form is the most common of the toothed bur clovers already discussed. Its growth and season are the same as the species. On account of its spiny burs it is less desirable for pasturage than the spineless forms.

Distribution: New Brunswick to Florida; thence west to California and Washington.

MEDICAGO HISPIDA NIGRA (WILLD.) BURNAT.

(Pl. XII, fig. 2.)

This subspecies differs from typical *Medicago hispida* in having 4 to 6 windings of the pod and stout rigid spines equaling or exceeding the width of the windings.

Distribution: Same as for the species.

This form was received under F. C. I. Nos. 0264, 0269, 0379, and S. P. I. Nos. 9739, 19439, 19448, 26072. All were originally from the Botanic Gardens at Madrid, Spain.

This importation has been tested at Chico and other places in California and at several localities in the Southern States. Its growth and season are the same as for the species. On account of its very spiny burs it is less desirable for pasturage than the spineless forms.

MEDICAGO HISPIDA TEREBELLUM (WILLD.) URBAN.

(Pl. XIII, fig. 2.)

This subspecies differs from *Medicago hispida nigra* in having pods with more compact windings, the spines being absent or reduced to rudiments. The pod is comparatively large and somewhat harder than in the other subspecies. It was received under F. C. I. No. 0274 from Madrid, Spain, and under S. P. I. No. 16879 from Brunswick, Germany, and under Nos. 19446 and 19456, probably also from Madrid. It has been tested at Chico and other places in California and at several localities in the Southern States. Its growth and season are the same as for the species proper. On account of the large size of the pod and the absence of spines, this form is desirable for pasturage. Nos. 0274 and 19446 have matured earlier than the other numbers tested and may be of greater value, as in a dry season late-maturing varieties do not yield as well.

Distribution: Same as for the species.

FURTHER WORK PLANNED.

The comparative testing of the bur clovers is being continued with the species discussed in this bulletin, together with a number of others more recently introduced. The newer introductions will be tested at various stations throughout the bur-clover sections of the United States, to determine their relative agronomic value in comparison with the spotted and toothed bur clovers.

It is the intention to bring together as complete a collection of species as possible. Species and subspecies not discussed in this bulletin are shown in the list that follows. Species marked with a star (*) have been recently introduced for this work. Attempts are being made to obtain the others.

List of nonperennial species and subspecies of Medicago not discussed in this bulletin.

Medicago aschersoniana Urban.
**M. blancheana* Boiss.
M. bonarotiana Arcang.
**M. carstiensis* Wulf.
**M. coronata* (L.) Desr.
M. daghestanica Rupr.
**M. disciformis* DC.
M. galilaea Bois.
M. globosa Presl.
M. granatensis Willd.
M. hispida microdon (Ehrenb.)
**M. hispida reticulata* (Benth.) Urban.
**M. intertexta* (L.) Mill.
M. intertexta decandollei (Trin.) Urban.
M. intertexta panormitana (Trin.) Urban.
**M. laciniata* (L.) Mill.
**M. litoralis* Rhode.
M. litoralis breviseta DC.
M. litoralis pentacycla Urban.
M litoralis tricycla (DC.) Urban.
**M. minima* (L.) Grufb.
**M. murex sorrentini* (Tin.) Urban.
M. muricoleptis Tineo.
M. noeana Boiss.
M. obscura Retz.
**M. obscura helix* (Willd.) Urban.
M. obscura lenticularis (Desr.) Urban.
M. obscura muricata (Willd.) Urban.
M. obscura tornata (Willd.) Urban.
M. orbicularioides Cand.
M. orbicularis applanata (Willd.) A. and Gr.
M. orbicularis biancae (Tod.) Urban.
M. orbicularis canescens Urban.
M. orbicularis glandulosa Urban.
M. pironae Visiani.
**M. praecox* DC.
M. rigidula cinerascens (Jord.) Urban.
M. rigidula eriocarpa Rouy and Fouc.
M. rigidula morisiana (Jord.) Rouy and Fouc.
M. rigidula timeroyi Boreau.
**M. rotata* Boiss.
M. rugosa incisa (Moris) Urban.
**M. soleirolii* Duby.
M. tenoreana Ser.
**M. tuberculata* (Moris) Urban.
M. tuberculata aculeata Moris.
M. tuberculata apiculata (Bast.) Urban.
M. tuberculata chiotica Urban.
M. turbinata inermis Aschers.
M. turbinata neglecta (Guss.) Urban.
M. turbinata olivaeformis (Guss.) Urban.
M. truncatula Gaertn.
M. truncatula breviaculeata (Moris) Urban.
M. truncatula longeaculeata (Moris) Urban.
M. truncatula tentaculata (Willd.) Urban.

SUMMARY.

The nonperennial species of Medicago consist principally of bur clovers, mostly annual plants native to the Mediterranean region.

Spotted bur clover (*Medicago arabica*), toothed bur clover (*M. hispida denticulata*), and yellow trefoil (*M. lupulina*) are the only species now widely distributed in the United States.

Spotted bur clover is the species best suited to and most commonly grown in the Middle Atlantic and Gulf Coast States.

Toothed bur clover is the most common bur clover in California, but spotted bur clover does equally well there.

Yellow trefoil is quite generally distributed throughout the United States and makes good growth in practically all sections in which it occurs.

Yellow trefoil promises to be of value for green manuring, not only in sections of the eastern United States where crimson clover is grown, but especially farther north.

The bur clovers are adapted for general use only in sections having a very mild winter climate, such as the Southern and Pacific Coast States.

Toothed bur clover, spotted bur clover, and yellow trefoil are suited to varied conditions with regard to soil and moisture.

The stronger growing bur clovers make good pasturage and green-manuring crops.

The bur clovers sometimes cause bloat in cattle when fed in the green state.

Yellow trefoil, toothed bur clover, and spotted bur clover seem to be somewhat unpalatable to stock not used to them.

The feeding value of bur clovers is good, as indicated by general experience and also by chemical analyses.

Bur clovers without spines are the most desirable for pasturage.

Medicago hispida confinis is a spineless form of toothed bur clover that is especially desirable for this reason.

Medicago orbicularis is one of the more recently introduced species that has large spineless burs and is very promising for pasturage in California.

Medicago arabica inermis is a new subspecies that has a spineless bur and is promising for use in sections where spotted bur clover does well.

Fall seeding of bur clover is necessary for the best results in all sections having mild winters.

In the Southeastern and Gulf Coast States the first of September is usually about the right time for seeding.

In California seeding may be done any time during September or October, but if the land is irrigated before sowing October is best.

In the Eastern and Gulf Coast States it is necessary for the best results to inoculate the soil in which bur clover is grown for the first time, but in the Pacific Coast States inoculation is not necessary.

One hundred pounds of burs of either spotted or toothed bur clover contain 25 to 30 pounds of seed.

In seeding any of the species of bur clover 15 pounds of seed (hulled) per acre should be sown when a thick stand is desired, either in pastures or cultivated fields.

To handle bur clovers as a seed crop is somewhat expensive, but not impracticable.

The farm machinery used in harvesting grain and hay crops, although not suited for handling bur clover, can be used to some extent.

Most bur clover seed on the market at the present time is obtained as a waste product from woolen mills, where it has been carried in the wool, and from screenings of small grains, with which it grows as a weed in California.

Twenty-three species and subspecies of bur clover have been studied in connection with the results here presented.

The plants of the various species are very similar in habit of growth and appearance of stems and leaves.

The burs of the species differ more or less, and their botanical classification is based largely on these differences.

Most of the species and subspecies studied are made up of definite types or forms which may be selected and grown as pure strains.

Environmental conditions may cause a wide variation in the burs of an individual plant, and must be taken into consideration in the identification of species.

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B. T. GALLOWAY, *Chief of Bureau.*

TOBACCO MARKETING IN THE UNITED STATES.

BY

E. H. MATHEWSON,

Crop Technologist, Tobacco Investigations.

WASHINGTON:
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LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry,
Office of the Chief,
Washington, D. C., September 14, 1912.

Sir: I have the honor to transmit herewith a manuscript entitled "Tobacco Marketing in the United States," by Mr. E. H. Mathewson, Crop Technologist, Office of Tobacco Investigations, and to recommend that it be published as a bulletin of the series of this Bureau.

It is believed that the information which is here brought together in a systematic and complete form will be of value to the tobacco interests of the country.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

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TOBACCO MARKETING IN THE UNITED STATES.

INTRODUCTION.

Tobacco is a commodity with somewhat inexact and varying standards of value. Every year, and perhaps every day, tobacco changes hands at prices varying from a few cents to several dollars a pound. Points as to difference in quality can be predicated with considerable detail and apparent precision on paper, but in judging the sample in hand it is discovered that a real standard does not exist, except in the varying ideas and opinions of men in the trade and in relation to the supply and demand at the time. The tobacco itself changes in appearance slightly, according to the conditions under which it is examined, and the ideas of the men themselves fluctuate more or less without other than personal reasons. In the low grades selling for a few cents a pound the differences of judgment concerning standards of quality is of but little importance as it affects sales and prices; but in the higher grades, with the much more refined and subtle differentiations to be considered, the divergence of opinions as to their proper values, even among experienced men, becomes increasingly great. Of course, very real differences in quality exist according to the general standards set up by the trade at the time, and men are able to recognize these points of difference along broad lines with a surprising degree of accuracy, even considering that it is so completely a matter of judgment.

Under such circumstances it was to be expected in the early days of the industry that misunderstandings and claims of misrepresentation, pretended, fancied, or real, would be common between buyers and sellers. If tobacco was sold by sample it was easy for the buyer to claim that the tobacco was not up to the sample, and the grower was prone to believe that the buyer had willfully and dishonestly failed to pay the proper value for his particular crop.

The tobacco industry was of such paramount importance in the early development of Virginia and Maryland that those colonies soon took cognizance of these unsatisfactory features and passed laws from time to time regulating the inspection and sale of tobacco. The purposes of these laws was to encourage foreign trade by better assuring the integrity of the sample and the keeping quality of the tobacco, by controlling the relations between buyers, sellers, and middlemen in the interests of equity and fair dealing, and by the satisfactory and expeditious settlement of questions in dispute.

Both of these colonies in very early times established official warehouses where planters could bring their tobacco for storage and sale. The system of the official sampling of tobacco in hogsheads by sworn State officers also came into existence at a comparatively early date. This official State inspection was generally compulsory in the case of tobacco for shipment out of the colonies, and sometimes also attempts were made to arbitrarily fix the price of the different hogsheads according to the sample as drawn.

The buyers often were only transitory foreign residents, which was an additional factor adding to the chance of misunderstanding and lack of good will between buyers and sellers and was an added reason for the necessity of legal regulation of the tobacco trade between buyers and sellers.

Thus, we have a conception of the fundamental causes that led up to the establishment of an elaborate system of warehousing, inspection, and auction sales under rigorous State supervision, which does not exist for any other agricultural commodity with which the writer is familiar.

State inspections are now, however, generally done away with, and the tobacco trade is more largely self-regulated, but the influence of this close State supervision as practiced in the old days is still clearly discernible, and even now all the older tobacco-growing States have quite elaborate statutes in force regulating the inspection, sale, and warehousing of tobacco.

THE MARYLAND OR BALTIMORE SYSTEM OF MARKETING.

THE BALTIMORE MARKET.

Of all the systems of marketing tobacco, that existing in Maryland, or rather Baltimore, since that is the only central market in the State, has undergone less change from the methods followed in the early days than that of any other of the old tobacco States, and, indeed, the methods now followed are practically what they always have been. State warehouses, State inspections (although no longer compulsory), and State guarantee of the samples are all still in force in the Baltimore market.

In earlier times State-inspection houses were located here and there at a number of convenient points throughout the producing territory. These, however, were abandoned one at a time, and the trade became more and more centered in Baltimore, and finally became entirely centered there.

A system of five large State storage and inspection houses resulted, each presided over by an independent inspector appointed by the governor. With a shrinkage in business, however, the number actually in operation was finally reduced to three. Under the system of having independent inspectors for each house there grew up much rivalry and competition for business in the effort to make each house self-sustaining. Under this system the tendency was to give a better sample than the contents of the hogshead warranted. To counteract this evil the Maryland Legislature remodeled its inspection law in 1898, providing, among other changes, for the appointment of a single chief inspector, with deputy inspectors for all State houses, all to be appointed by the governor for biennial periods and placed under heavy bonds for the proper discharge of their duties. This is the law as it stands to-day.

The more important provisions of the law, so far as general rules of procedure are concerned, are substantially as follows: The hogsheads are numbered and inspected in the regular order of their receipt. The gross weight of the tobacco and hogshead is taken first; then the hogshead is stripped from the tobacco and the hogshead weighed separately. The tobacco is then inspected by drawing samples from at least six different places. This sample is sealed with wax and appropriately marked with the number of the hogshead, date, and inspector's name. No charge is made to sellers of Maryland-grown tobacco for either storage or inspection. The buyer, however, covers the expense of the service to the State by paying a fee of $2 per hogshead upon the removal of the tobacco and an additional storage fee of 15 cents per month, if in storage more than six months after purchase.

Tobacco is not sold at auction on the Baltimore market. There are a number of commission firms, to one of which the farmer usually consigns his tobacco. On inspection the sealed sworn samples are taken in charge by the commission merchant and removed to his office. The sale of the hogshead of tobacco is thereafter a matter of private bargaining between the commission merchant and the buyer, although the grower sometimes sets a minimum price for the commission merchant to ask. When the market is active it is customary for the buyers on the Baltimore market to make the rounds from commission house to commission house, looking at the samples and making offers on such hogsheads as they think they can use. For this service the commission merchant charges the seller a fee of $1.50 per hogshead, independently of its weight or the price it brings. In addition to the price of the tobacco, however, he collects from the buyer $1 per hogshead, which he turns over to the farmer as reimbursement for the value of the hogshead itself.

In case of dispute as to the correctness of any sample furnished by the inspector under the seal of the State, the controversy under certain limitations is referred to a board of arbitration consisting of three disinterested persons. One of the members of this board is selected by the inspector, and one by the claimant, and if after consultation these two can not agree, they are to select the third member. Under the State law this board has power to summon and examine witnesses and to assess such reclamation or damages as it deems just. This reclamation allowance is paid by the inspector from the general tobacco fund, and it is not charged against either the grower or the seller.

The average net weight of a hogshead of tobacco as sold on the Baltimore market is about 700 pounds, and those weighing more than 850 pounds or less than 600 pounds are likely to be discriminated against by buyers. In former days hogsheads were packed heavier, but the lighter packed ones are preferable because of less danger from injury by bruising. If too light, however, the export freight rate, which is charged by the hogshead, becomes relatively greater per pound.

The main features of the Baltimore system of selling tobacco are as outlined, but there are, of course, numerous details covering the laws or customs as to reinspections, cooperage, false packing, damaged tobacco, etc., which it hardly seems necessary to consider here. The striking feature of the Baltimore system is the paternalism of the State and the consequent low charges to the seller on that market.

From the "outage" fee of $2 per hogshead, paid on removal from the warehouse, and other minor sources of income, such as storage, collected for so many years, together with a heavy insurance indemnity resulting from the destruction by fire of two inspection houses several years ago, the State has accumulated a large surplus fund, amounting to several hundred thousand dollars. Out of this fund the State has recently built a fine new storage and inspection warehouse. This building is six stories in height, of fireproof construction, and will store at one time from 15,000 to 18,000 hogsheads of tobacco, or about two-thirds of the annual production of the State.

Until 1878 all tobacco inspected in Maryland, including the eastern Ohio type, was inspected in the State inspection warehouses, and it had generally been considered that the Maryland law made such official inspection compulsory. At that time, however, the question was raised as to the jurisdiction of the State over the inspection of tobacco produced in other States. Therefore, in 1878, a private warehouse was established at Locust Point, catering particularly to the shipments from the eastern Ohio district. The same charges and the same warehouse and inspection rules prevailed at Locust Point as at the State-inspection warehouses, but the Locust Point warehouse had railroad terminals as well as wharfage, which saved drayage. This was an advantage, particularly to the inland shipments, and it soon became the custom to store and inspect all of the eastern Ohio types at this private warehouse. Later, the compulsory features of the remodeled law were changed so as to permit the warehouse inspection of Maryland-grown tobacco by other than State inspectors. Since 1898, therefore, a considerable proportion of the Maryland crops has been inspected in this private warehouse in addition to all the eastern Ohio shipments.

In Table I are shown the total receipts of tobacco at both the State and private warehouses for the 15-year period from 1895 to 1909, inclusive. The figures are computed from the report of the board of directors of the Baltimore Leaf-Tobacco Association.

TABLE I.—*Receipts of Maryland and eastern Ohio tobacco at Baltimore, Md., for the 15 years from 1895 to 1909, inclusive.*

Year.	Maryland tobacco.			Eastern Ohio tobacco—private warehouse.	Total, Maryland and eastern Ohio tobacco.	Reclamations.[1]
	State warehouse.	Private warehouse.	Total.			
	Hhds.	*Hhds.*	*Hhds.*	*Hhds.*	*Hhds.*	
1895	28,085	----------	28,085	6,899	34,984	$722.73
1896	29,623	6	29,629	4,360	33,989	1,202.43
1897	34,869	6	34,875	8,043	42,918	2,101.75
1898	30,822	5,470	36,292	5,970	42.262	1,203.99
1899	26,622	4,862	31,484	7,224	38,708	717.00
1900	27,122	7,448	34,570	3,453	37,023	957.77
1901	23,776	7,662	31,438	4,443	35,881	704.89
1902	24,593	10,069	34,662	4,818	39,480	622.31
1903	23,500	11,535	35,035	5,016	40,051	938.86
1904	28,052	8,224	36,276	4,231	40,507	678.40
1905	25,080	8,597	33,677	4,919	38,596	590.05
1906	22,805	7,977	30,782	6,507	37,289	433.43
1907	15,637	5,690	21,363	4,149	25,512	677.56
1908	19,831	7,149	26,980	2,218	29,198	1,025.51
1909	19,100	7,972	27,072	1,988	29,060	1,324.39

[1] Includes only Maryland tobacco, but the reclamations on the eastern Ohio type were very small.

The total number of hogsheads of Maryland tobacco received in this period was 472,220, an average of 31,481 yearly. The reclamations amounted to $13,602.10, an average of $906.81 yearly. The value of the average hogshead of Maryland tobacco in this period has been probably something less than $50. The value of the reclamations, therefore, are equal to the value of only 18 or 20 hogsheads of tobacco annually, which is in the ratio of about 1 to 1,500. Of course, there were reclamations in a greater proportion than 1 hogshead in every 1,500, because probably in no case would more than a fraction of the total value of the hogshead be allowed in the reclamation. Reclamation is not usually resorted to except in extreme cases. As a matter of fact, mixed and careless, though not necessarily dishonest, packing is much more prevalent than it should be and is a constant source of complaint from the buyers.

So-called local or home buyers are scattered through the growing sections. They buy scattered crops here and there at a round or average price and ship to Baltimore, expecting to make a profit by the transactions.

In 1910 the French Government changed somewhat its system of buying Maryland tobacco. Purchases are now made direct through agents, instead of letting the contract to the best competitive bidder for the major part of the requirements, as heretofore.

Maryland tobacco is naturally of a dry and gumless nature. Conditioning for shipment abroad is done by the growers themselves before putting their tobacco on the Baltimore market. A large proportion of the crop, therefore, is not marketed till nearly a year after it is grown, in order that it may pass through the spring sweat and be in good keeping order. It is packed dry in the hogsheads so as to avoid any chance of damage or darkening in color from fermentation. Nearly all tobacco growers put together their own hogsheads from the rough staves obtained from the local sawmill or from secondhand hogshead material purchased from manufacturers in Baltimore, and every tobacco farm has its screw or lever press for prizing the tobacco into hogsheads.

As already stated, the present Maryland system of marketing tobacco presents a very good picture of the general method as followed in the early days in nearly all of the older tobacco States. As such it is of considerable historical as well as present commercial interest.

BALTIMORE AS A TOBACCO-TRADE CENTER.

The importance of Baltimore as a center of the tobacco trade is far from being fully comprehended because of the selling of the Maryland and eastern Ohio export crops there. It is also a very important tobacco-manufacturing center, and is one of the three important centers from which the bulk of our exports of leaf tobacco is finally cleared. In volume of exports it is second only to New York. A considerable proportion of both southern and western leaf is exported through Baltimore in addition to nearly all of the Baltimore types. In the fiscal year ended June 30, 1908, there were 93,279,562 pounds of leaf tobacco and 3,166,117 pounds of stem and trimmings cleared for export from the port of Baltimore.

As a manufacturing center Baltimore ranks high. There are more little cigars manufactured here than in any other center, and there are also large factories engaged in the manufacture of various other forms of tobacco products. In 1909 the total consumption of raw material in the form of leaf, stems, and scrap amounted to over 25,000,000 pounds.

Combining the exports with the quantity manufactured we have a total of about 120,000,000 pounds, representing with reasonable accuracy the total annual movement of tobacco through Baltimore. This places this city very close to the lead among our centers of tobacco trade in total movement, and it is probably surpassed only by New York and Louisville.

THE LOOSE-LEAF TOBACCO AUCTION-SALES SYSTEM.

THE MARKETS OF VIRGINIA, NORTH CAROLINA, AND SOUTH CAROLINA.

From colonial times until 1877 the sale of leaf tobacco in Virginia was based on a system of compulsory State inspection by samplers appointed by the governor. These samplers were appointed to the numerous warehouses located at the various central market points in the State. Tobacco for export was subject to seizure if found in transit out of the State without the seal of inspection. By a State law passed in 1844 tobacco that came from the West—that is, from Kentucky, Tennessee, or Ohio—and which was inspected in Virginia was branded "Western," as tobacco from Maryland was branded "Maryland," in order to prevent, so far as possible, their sale as Virginia leaf, which was in greater favor in both the domestic and the foreign markets.

This compulsory system of State inspection, although of great advantage in the colonial period, when the export trade was in its infancy, had long since outgrown its usefulness and had become so contaminated with politics in the appointment of the samplers that it was no longer satisfactory to the tobacco trade. The farmers, however, feared a change to a private inspection system, and several vigorous legislative campaigns were necessary before the final overthrow of the system was accomplished in 1877 by the aid of the legislature.

The law as revised still provided for the appointment of State samplers and for the official inspection of all prized hogshead tobacco belonging to farmers, except as removed from the package and sold loose, but under somewhat modified restrictions permitting private inspections. Although the modified laws for the State inspection of tobacco still stand on the statute books of Virginia, they are as a matter of fact practically a dead letter, because private inspection has completely superseded State inspection of prized tobacco.

Meanwhile the practice of selling tobacco loose at auction had started on a small scale in some of the market towns, particularly of southern Virginia and North Carolina. The act of 1877 gave an added impetus to this method of selling by changing the law so as to permit the sale of loose tobacco on the warehouse floor without inspection. So far as the sales of loose tobacco are concerned the Virginia law merely requires the weigher to take a prescribed form of oath to keep the warehouse scales properly standardized and to properly weigh the tobacco. The warehouseman must issue a statement to the seller covering in detail all charges connected with the sale of the tobacco.

Another agency working for the rapid spread of this system is the manifest preference of the largest manufacturers and exporters to purchase leaf tobacco by it rather than by the hogshead and inspection method.

At the present time, so far as the first-hand sale of tobacco from farmers is concerned, the loose-leaf tobacco system of selling at public auction is almost universal in the tobacco districts of Virginia, North Carolina, and South Carolina.

The system has been brought to a high state of perfection and efficiency and in its general convenience both to buyers and sellers surpasses any other method of marketing tobacco or other farm product that has come under the writer's observation.

Sales warehouses are located in nearly all the centers of any importance in the tobacco-growing sections of Virginia, North Carolina, and South Carolina. Most of the important producing counties have at least one tobacco-market town, and in some cases there are several markets in a county. The most important exception to this is in the few counties producing tobacco north of the James River in Virginia. Except Richmond, there is no tobacco market in Virginia north of the James.

Most of the larger towns have from two to four sales warehouses each, and in Danville, Va., which is a very large market, there are generally six and in some seasons more warehouses open for business. The sales warehouses are often substantial brick structures with a great expanse of floor space, sometimes covering from 25,000 to 30,000 square feet in area, with as few posts or other obstructions as possible. The roof construction, covering such large expanses without center supports, is a feat of considerable engineering skill. An exterior view of the great Acree warehouse in Danville, Va., one of the largest and most substantial of these loose-leaf auction warehouses, is shown in figure 1. In front are some of the farmers' tobacco wagons lined up after being unloaded.

Competition for business among warehouses is very keen, and conveniences and facilities for both sellers and buyers are as complete as possible with a view of attracting patronage. The effective lighting of the larger sales floor is accomplished by skylights and side windows. Usually the warehouse is so arranged that farmers can drive inside and unload. Bunks and lounging rooms for the farmers and stables for the horses are generally provided.

In some sections, as, for example, in the eastern North Carolina markets, there are basements for grading and tying tobacco, so that farmers can have this work done for them at a reasonable price if they so desire. Generally, however, the farmers themselves grade and tie up the tobacco in small hands at home, then pack the different grades one after the other neatly in the wagon bed, and cover the load with sailcloth, bed blankets, or some other protection from the weather.

At the warehouse each grade is carefully and neatly piled on a warehouse truck, carried to the scales and weighed, and a ticket is attached showing the owner's name, the number of the lot, the number of piles or grades in the lot, and the weight of the pile. In addition there are blank spaces left for filling in at the time of sale, showing the buyer's name and the price. Stubs are usually provided

Fig. 1.—Loose-leaf tobacco auction warehouse, Danville, Va.

also so that these data may be entered on each part, one to be retained by the warehouseman and the other to go to the purchaser. Each grade is then placed on the floor in rows, usually allowing about 18 inches each way between piles.

On days when the sales are heavy there will often be from 500 to 1,000 or more piles on the floor at a time. The piles are sometimes very small, often weighing less than 50 pounds, but sometimes reaching 1,000 pounds or more. They will usually average 150 to 200 pounds each.

The ringing of the warehouse bell gives notice of the beginning of the sale, which proceeds rapidly, generally at the rate of from 150 to 200 piles per hour. The scene is an interesting one, enhanced by the quick, snappy crying of bids by the auctioneer, with an occasional joke thrown in and a moment of merriment, the entreaties and pleadings for a better bid on the part of the sales manager, and the rapid though sometimes silent bidding by a nod or wink on the part of the buyers.

The crowd moves slowly along from pile to pile, the ticket marker immediately putting down the price, the buyer's name, and private marks indicating the grade. Close behind the ticket marker follows a clerk, who calculates the value of the pile and places it on a slip containing the grower's name and the number of the pile.

In the larger markets, after the sales have continued about half an hour, these calculations are carried to the office, where other clerks immediately begin computing the gross and net proceeds, and the

FIG. 2.—Interior of a loose-leaf tobacco auction warehouse, Danville, Va., during a sale.

payment to the seller, generally in cash, is often begun long before the sale on the floor is finished and before the warehouseman has received the purchase price from the buyer. In figure 2 is shown a section of the floor of Acree's warehouse in Danville, Va., during the progress of a sale, which is going on at the farther corner of the room.

The wagons of the buyers are on hand soon after the sale starts, and the removal of the purchases immediately begins. Large shallow baskets are used, which are piled high on the wagons, tier on tier, to be hauled at once to the buyers' handling houses. The piles of tobacco are frequently nearly all removed by the time the sales are over, and are entirely removed soon afterwards, or at least before the day is ended.

ADVANTAGES OF THE LOOSE-LEAF TOBACCO AUCTION SYSTEM.

A number of advantages of the loose-leaf tobacco auction system, both to seller and buyer, are apparent.

The seller is conveniently brought into contact at once with the buyer under as fair conditions as it is possible to create, so far as a mere system of selling is concerned. He can see for himself what his tobacco brings as compared with the general market for similar grades. The sale is prompt—within the day after he arrives with his tobacco—and his money is ready immediately in cash or its equivalent. If he is not satisfied with the prices obtained, he may reject the sale within a reasonable time limit, and he can then immediately reoffer his tobacco for sale or he may take it home, or to some other market, or to another warehouse, without charge of any kind.

From the buyer's standpoint there are also important advantages connected with the loose-leaf tobacco system. He can look over carefully, although rather hurriedly, all the tobacco, not merely a sample, that he buys. In order that tobacco may keep free from damage by mold, etc., either during storage prior to resale or manufacture or during shipment to distant countries, it must be thoroughly dried out and conditioned—that is, put into safe keeping order. When sold in hogsheads at first hand, the tobacco comes to the buyer in all sorts of conditions, good and bad, and some of it must be redried, a matter to be determined by the purchaser. All loose tobacco is sold in soft condition, the buyer taking the responsibility of redrying and putting it in safe and uniform keeping order, which is the most satisfactory way.

Of course there are drawbacks connected with this method of selling, and it does not give satisfaction in every case, either to the seller or to the buyer. On the whole, however, it is the most generally satisfactory method of sale yet devised, and it seems to be able to hold its ground against the hogshead and inspection method wherever introduced, so far as first-hand sales from farmers are concerned. Under this system there is no necessity for official inspection or sampling. The warehouse proprietors in conducting the sales merely employ an auctioneer and such other clerks, weighers, bookkeepers, and laborers as are necessary. For this they make a charge to the seller, which in the case of the larger markets usually consists of three items, about as follows: (1) An unloading and weighing fee of 10 cents per 100 pounds; (2) an auction or selling fee at the rate of 15 cents for each pile of 100 pounds or less and 25 cents for each pile of more than 100 pounds; (3) a commission of 2½ per cent on the gross proceeds. Some of the markets charge 3 per cent commission and some have no commission charges whatever. In small markets the charges are usually less in the aggregate than in the larger markets, as an extra inducement to come to the smaller market. Other inducements to patronize the smaller markets are those of convenience and shorter haul for the immediately surrounding territory and local pride in building up the business interests of the home town. Buying interests, however, are usually more fully represented in the larger markets.

The selling charge for a 1,000-pound load of tobacco divided into five grades bringing an average of 10 cents per pound, based on the charges just enumerated, would be as shown below. These charges are those in effect in most of the "flue-cured" tobacco markets and are the maximum allowed by the State law of North Carolina.

Weighing, at 10 cents per hundred pounds	$1.00
Auction fee, at 25 cents per pile	1.25
Commission, 2½ per cent on $100	2.50
Total	4.75

These charges are considerably higher than they would be if the same tobacco had been sold in a hogshead by sample, either at the present scale of prices or those in force under the old State system of inspection; but there are offsetting advantages, such as lack of expense for hogsheads and the obtaining of immediate returns. Furthermore, the tobacco is sold in soft order instead of more or less dry, which means an increased selling weight because of the higher moisture content.

ORGANIZATION OF THE LOOSE-LEAF TOBACCO TRADE.

In practically all of the larger loose-leaf tobacco markets the members of the tobacco trade, including warehousemen, leaf-tobacco dealers, and manufacturers, are organized into trade organizations, which have established market rules or regulations in the interests of the trade as a whole and in the interests of fair dealing and the avoidance or settlement of disputes between members.

The supervisor of sales is usually one of the more important officers of the trade organization. It is this officer's duty to see that the rules of the organization are carried out, particularly as regards the running of the sales, such as looking after the correctness of scales, the proper spacing of the piles on the floor, the number of piles sold per hour, and the rotation of sales from warehouse to warehouse.

The arbitration committee in its function of settling disputes between members also has most important duties.

The privilege of bidding on or purchasing tobacco at the sales is usually restricted to members of the organization. The minimum raise between bids is generally the subject of regulation, as, for example, a minimum of 10 cents a bid up to $6 per 100 pounds; 25 cents from $6 to $15; 50 cents from $15 to $25; and $1 a bid over $25 per 100 pounds.

In most of the larger markets there are from two to four or more warehouses open for business. These take turns by mutual agreement with the trade organization as to the order in which the sales shall be held. The first sale of the day is the most popular with the farmers, and the schedule is so arranged as to alternate in regular order among the different warehouses on different days. At the large markets it is necessary to have two sales going on simultaneously, each with its corps of buyers, etc., in order to complete the day's work. On the Danville market the sales are run in triplicate through much of the season.

Competition among warehousemen in the larger market centers for the farmers' patronage has been very keen and expensive. This has resulted in the warehousemen pooling or consolidating their interests in some of the larger markets and placing the management of all the warehouses in town under a joint management. Danville and Lynchburg, Va., are notable examples of this consolidation of interest and management.

On the other hand, selling charges to the farmer are rather high, usually amounting to more than 4 per cent of the total value of the crop, and the profits of the warehousemen have been liberal. This has led in some instances to a movement toward the joint ownership and management of the warehouses by the farmers themselves.

The most notable and successful movement of this kind is that of the Farmers' Consolidated Warehouse Co., of Greenville, N. C. This company has established branches in many of the other important new belt markets, including Wilson, Kinston, and Williamston in North Carolina and Mullins in South Carolina. It has also invaded the western field in Maysville, Ky., with the establishment of a loose-leaf market at that point in the fall of 1909. The farmers of the locality in each case subscribed to a majority of the stock.

DANVILLE, ONE OF THE LARGEST LOOSE-LEAF TOBACCO MARKETS AND CENTERS OF THE LEAF-TOBACCO TRADE.

With the development in the production of flue-cured tobacco since the Civil War, Danville forged rapidly to the front as the preeminent market for the sale of bright leaf, and since the eighties of the past century for many years ranked as the largest loose-leaf tobacco market of the world. The first-hand sales at one time ran over 40,000,000 pounds of loose-leaf tobacco yearly. Other large markets have also rapidly sprung up in the flue-cured tobacco section, with which Danville has had to share patronage, although this has not as yet by any means endangered its preeminence in the seaboard States as a leaf center. The annual sales of loose-leaf tobacco now run in the neighborhood of 35,000,000 pounds.

The tobacco trade of Danville had become of considerable importance early in the nineteenth century, and warehouses were established for the inspection of tobacco under the State system. The panic of 1837, however, struck the town with disastrous effect and its tobacco trade went to pieces, not to be revived until just before the breaking out of the Civil War. It was the custom to ship the tobacco produced in this section to the Lynchburg, Petersburg, and Richmond markets.

In 1858 interest had greatly revived, and Neal & Graves erected a large wooden building for the sale of leaf tobacco at auction, known as Neal's warehouse. The opening of this warehouse is generally recognized as the beginning of the Danville tobacco market upon its present basis. In 1860 the legislature established an official inspection of tobacco at this warehouse. The sales were practically all of loose tobacco, and the venture proved successful. The growth of Danville as a city has been intimately connected with its growth as a tobacco market. In 1850 the population of Danville was 1,760; in 1860 it was 3,500. The tobacco trade of Danville was, of course, greatly set back during the Civil War period, but it immediately revived after the war was over. The rapid development of the tobacco trade also meant the rapid development of Danville as a business center. In 1900 the population of Danville was 16,520.

The following record of the yearly sales of loose tobacco on the warehouse floor from 1869, the earliest date at which reliable records are available, to 1909 will be of interest as illustrating the growth of the greatest loose-leaf tobacco market in the world.[1] The figures are taken from the annual report of Mr. A. B. Carrington, president of the Danville Tobacco Association.

TABLE II.—*Record of the yearly sales of loose-leaf tobacco at auction on the Danville (Va.) market from 1869 to 1909, inclusive.*

Year.	Quantity sold.	Average price per 100 pounds.	Year.	Quantity sold.	Average price per 100 pounds.
	Pounds.			*Pounds.*	
1869	10,621,557	$12.25	1890	40,099,289	$11.95
1870	13,191,406	12.00	1891	37,707,180	8.87
1871	14,065,639	12.34	1892	39,001,755	8.23
1872	15,827,846	11.64	1893	42,050,141	6.46
1873	16,147,715	13.47	1894	39,206,789	7.96
1874	14,679,421	20.45	1895	40,160,999	7.79
1875	23,466,413	13.32	1896	46,693,654	6.46
1876	16,624,296	12.23	1897	49,464,741	7.81
1877	27,698,125	8.80	1898	48,939,542	6.61
1878	26,851,900	11.91	1899	50,229,083	6.74
1879	33,151,247	11.39	1900	37,134,068	7.38
1880	30,552,738	10.86	1901	33,685,062	10.25
1881	25,572,536	9.81	1902	46,710,547	8.77
1882	35,503,121	13.45	1903	42,908,499	7.94
1883	27,548,014	13.07	1904	38,029,050	8.55
1884	37,017,904	13.54	1905	34,976,623	9.42
1885	40,352,942	9.41	1906	36,040,017	9.11
1886	29,343,728	8.65	1907	35,567,464	11.42
1887	31,269,257	10.60	1908	39,062,004	9.77
1888	28,803,846	8.75	1909	35,327,614	10.62
1889	24,925,076	13.22			

[1] Since 1910 the loose-leaf sales in the Lexington, Ky., market have equaled or exceeded those at Danville.

This table shows that the high-water mark in the sales on the Danville market was reached in 1899, when more than 50,000,000 pounds were sold. The decline, however, does not indicate a decline in the crop produced. The real reason is found in the development of a large number of small markets throughout the producing territory. Formerly, the Danville sales included much tobacco shipped from distant points. Now, however, this business is much smaller and the sales represent almost entirely tobacco brought in on wagons from the territory immediately surrounding the city and within hauling distance, usually not exceeding 30 miles.

A marked change has been noted in late years in regard to the time of marketing tobacco. Farmers formerly delayed marketing heavily until after the Christmas holidays. Recently, however, there has been noted a tendency to market early, and the bulk of the crop is now often marketed in the first few months of the selling season. In 1908 more than a fourth of the entire crop marketed at Danville was sold in the month of October.

In the New Belt the crop is sold earlier and more rapidly than in the Old Belt. In South Carolina the crop is harvested in July and sales begin immediately; August and September are usually the heavy months for sales and the market is practically closed by November 1.

Table III shows the monthly sales on the Danville market for the years 1876, 1890, and 1908, illustrative of this change in time of selling the crop. The data were kindly furnished by Dibrell Bros., Danville, Va.

TABLE III.—*Record of the monthly sales, with average price realized, for tobacco sold on the Danville (Va.) market in 1876, 1890, and 1908.*

Month.	Sales.					
	Crop of 1876.		Crop of 1890.		Crop of 1908.	
	Quantity.	Average price.	Quantity.	Average price.	Quantity.	Average price.
	Pounds.		*Pounds.*		*Pounds.*	
October	900,902	$18.01	4,155,750	$11.62	10,107,125	$9.77
November	959,711	11.79	3,883,935	10.56	6,401,013	9.77
December	107,655	10.80	1,488,763	10.01	4,719,917	9.89
January	696,448	12.16	2,019,562	12.05	4,813,471	10.41
February	1,176,839	14.35	5,479,977	11.62	4,815,537	10.36
March	1,457,988	12.43	4,669,455	11.88	3,761,626	9.62
April	1,808,872	12.08	4,551,081	12.59	1,770,595	9.11
May	2,562,190	10.75	3,368,621	13.68	785,733	8.68
June	1,960,338	12.30	3,199,692	12.35	327,503	8.31
July	1,634,759	11.04	3,605,113	13.04	(1)	(1)
August	2,044,124	11.94	2,477,723	11.84	[1] 613,259	[1] 5.46
September	1,221,470	11.58	1,399,617	11.08	[1] 916,224	[1] 8.75
Total	16,624,296	12.23	40,099,289	11.95	39,062,004	9.77

[1] Market closed in July. The sales year now begins on Aug. 1 instead of Oct. 1, in response to this change in time of selling. August and September sales in this column should stand really at the top, representing the first sales of the 1908 crop, consisting mostly of primings, and thus accounting for the low average price for these months.

The commanding importance of Danville in the flue-cured leaf trade is not adequately measured merely by the pounds of leaf sold on its warehouse floors. Danville is the one great central receiving and distributing point for all types of bright leaf as produced in the Piedmont or western portion of the flue-cured belt, the great middle district in which Danville is situated, and the New-Belt Coastal-Plain section of eastern North Carolina and South Carolina. In Danville are located the greatest array of sales warehouses, redrying plants, stemmeries, and establishments of tobacco-leaf dealers and commission merchants of any market in the flue-cured tobacco belt.

The total movement of tobacco through the Danville market is greatly in excess of the sales of loose tobacco. Aside from the leaf sold through the auction warehouses, from ten to fifteen million pounds of leaf come into this market yearly through the purchases of dealers or through consignments to them from other markets. This brings the total volume of the leaf trade of Danville to upward of 50,000,000 pounds per annum. We have spoken of Danville as one of the largest loose-leaf tobacco markets in the country. There are other markets that handle a much larger volume of leaf than Danville, notably Louisville, Ky., whose annual sales generally run more than 100,000,000 pounds. A great but uncertain portion of this larger trade of other markets consists of dealers' tobacco, i. e., tobacco bought up speculatively by country buyers who travel through the growing districts picking up individual crops here and there. If the truth could be accurately determined, it might be found that Danville would be a leading market not only in the sales of loose tobacco but also in first-hand sales from farmers. Practically all the tobacco sold loose at the Danville warehouses is first-hand sales direct from farmers.

OTHER LOOSE-LEAF TOBACCO MARKETS OF VIRGINIA, NORTH CAROLINA, AND SOUTH CAROLINA.

There were 23 loose-leaf tobacco markets in operation in Virginia in the sales year 1908–9. One of these, the Richmond market, sold "sun cured" tobacco almost exclusively, while another, Amelia, sold the olive-stemming type exclusively. Twelve markets sold tobacco of the dark-fired type, either chiefly or exclusively, and nine were principally or entirely flue-cured or bright tobacco markets.

In North Carolina there were in the same year 45 loose-leaf tobacco markets which sold the flue-cured type of leaf, 20 being in the Old-Belt and 25 in the New-Belt section.

In South Carolina during the same period there were 12 markets in operation, all selling the New-Belt flue-cured type of leaf.

A complete list of these markets is given in Table IV, with the amount of their first-hand sales for producers and the average price for all sales per hundred pounds. The returns for the South Carolina markets are for the year 1909, based on the reports made under the law to the commissioner of agriculture of that State. Those for Virginia and North Carolina are for the 1908 crop, based on the returns to the respective commissioners of agriculture of those States under the State laws. The price averages for each market in Virginia and North Carolina were obtained from the different warehousemen, as determined by the actual sales records kept from day to day, and were furnished through the courtesy of Dibrell Bros., Danville, Va.

TABLE IV.—*Record of the sales of leaf tobacco and the average prices obtained at the loose-leaf tobacco auction markets of Virginia, North Carolina, and South Carolina for the years specified.*

VIRGINIA DARK-TOBACCO MARKETS SELLING 1908 CROP.

Market.	Principal type of tobacco sold first hand.	Quantity sold first hand.	Average price per 100 pounds.
		Pounds.	
Richmond	Sun cured	9,371,576	$8.06
Amelia	Olive stemming	829,070	8.50
Lynchburg	Dark fired	20,178,700	7.20
Petersburg	do	6,340,260	7.14
Drakes Branch	do	5,219,020	7.15
Farmville [1]	do	4,184,208	7.02
Bedford City	do	3,796,476	7.00
Blackstone	do	2,456,357	7.30
Brookneal	do	1,904,147	6.73
Cumberland	do	1,125,000	6.80
Appomattox	do	687,190	6.75
Keysville	do	400,000	6.75
Lawrenceville	do	51,698	6.00
Total		56,543,402	7.32

VIRGINIA OLD-BELT BRIGHT-TOBACCO MARKETS SELLING 1908 CROP.

Market.	Principal type of tobacco sold first hand.	Quantity sold first hand.	Average price per 100 pounds.
Danville	Flue cured	34,348,914	$9.80
South Boston	do	15,556,807	9.38
Martinsville	do	3,251,408	9.50
South Hill	do	2,932,979	8.75
Rockymount	do	2,498,485	8.00
Chase City	do	2,411,994	8.50
Clarksville	do	1,964,780	8.25
Kenbridge	do	1,217,503	8.00
Virgilina	do	821,939	7.64
Chatham	do	764,508	9.25
Total		65,769,317	9.50

NORTH CAROLINA OLD-BELT BRIGHT-TOBACCO MARKETS SELLING 1908 CROP.

Market.	Principal type of tobacco sold first hand.	Quantity sold first hand.	Average price per 100 pounds.
Winston-Salem	Flue cured	20,939,200	$10.23
Oxford	do	7,546,003	10.87
Durham	do	6,645,670	12.40
Reidsville	do	5,797,438	9.46
Henderson	do	5,279,709	11.28
Roxboro	do	4,760,322	10.41
Mount Airy	do	4,375,125	10.00
Louisburg	do	3,498,376	9.85
Stoneville	do	2,214,698	9.00
Warrenton	do	2,148,148	9.08

[1] Farmville sales only partially reported; should have been over 7,000,000 pounds.

TABLE IV.—*Record of the sales of leaf tobacco, etc.*—Continued.

NORTH CAROLINA OLD-BELT BRIGHT-TOBACCO MARKETS SELLING 1908 CROP—Con.

Market.	Type.	Quantity sold first hand.	Average price per 100 pounds.
		Pounds.	
Creedmore	Flue cured	1,732,500	$15.50
Madison	do	1,636,691	9.00
Greensboro	do	1,140,811	9.04
Burlington	do	1,071,558	9.00
Apex	do	953,944	15.63
Youngsville	do	848,891	10.38
Raleigh	do	497,429	11.00
Statesville	do	286,101	8.50
Leaksville	do	225,718	10.00
Pilotmountain	do	205,456	10.00
Total		71,785,788	10.00

NORTH CAROLINA NEW-BELT BRIGHT-TOBACCO MARKETS SELLING 1908 CROP.

Market.	Type.	Quantity sold first hand.	Average price per 100 pounds.
Wilson	Flue cured	16,435,712	$9.84
Greenville	do	10,530,935	9.07
Kinston	do	9,298,021	8.55
Rocky Mount	do	9,183,146	9.99
Farmville	do	2,880,603	8.58
Goldsboro	do	2,458,458	8.52
Smithfield	do	2,150,618	8.58
La Grange	do	2,113,604	7.72
Fair Bluff	do	1,707,774	8.19
Robersonville	do	1,563,500	8.50
Williamston	do	1,488,352	8.18
Fairmont	do	1,397,168	8.64
Richlands	do	1,385,470	7.50
Snow Hill	do	1,284,728	7.25
Ayden	do	1,181,533	7.25
Zebulon	do	950,641	7.25
Ahoskie	do	783,218	7.40
Enfield	do	721,793	7.75
Wendell	do	665,969	7.25
Warsaw	do	607,174	7.25
Lumberton	do	454,003	8.50
Clinton	do	504,090	7.25
Dunn	do	349,592	7.15
Fuquay Springs	do	230,291	7.25
Clarkton	do	106,124	8.50
Total		70,433,517	9.10

SOUTH CAROLINA NEW-BELT BRIGHT-TOBACCO MARKETS SELLING 1909 CROP.

Market.	Type.	Quantity sold first hand.	Average price per 100 pounds.	Total value.
		Pounds.		
Mullins	Flue cured	6,805,899	$6.89	$468,982.85
Darlington	do	4,740,707	7.94	377,045.43
Lake City	do	4,448,299	7.97	354,770.61
Timmonsville	do	4,045,721	7.68	310,905.23
Florence	do	2,129,779	7.84	166,971.30
Marion	do	1,818,237	6.54	118,966.96
Nichols	do	1,445,980	6.66	96,380.91
Kingstree	do	1,416,466	7.67	108,749.24
Manning	do	1,164,497	6.89	80,316.77
Loris	do	1,078,480	5.45	58,774.47
Dillon	do	1,049,961	5.91	62,068.04
Latta	do	934,782	7.00	65,428.83
Conway	do	741,711	6.17	45,791.69
Total		31,820,519	7.27	2,315,152.33

Of the 13 markets operating in South Carolina in 1909, 7 (Mullins, Nichols, Loris, Dillon, Latta, Marion, and Conway) were in the section east of the Pedee River. The combined total of their sales amounted to 13,875,050 pounds, selling at an average of $6 per 100 pounds, while the 6 markets on the west side of the Pedee sold 17,945,469 pounds, at an average of $7.78 per 100 pounds. Practically no tobacco is carried across the Pedee by farmers, so that the sales of the markets on either side of the river closely represent the tobacco produced on that side. The markets of the western side of the river, however, do not always make a higher average over those on the eastern side. As previously stated, the land on the eastern side is, on the whole, sandier than that on the western side. The 1909 crop season was particularly unfavorable to sandy-land tobacco, as compared with that grown on stiffer land. In some seasons this condition is reversed.

Sometimes proximity to a body of land producing a type of leaf above the average in quality will have a material effect in raising the average price of a market. The averages of Lake City and Darlington, S. C., can both be explained in this way. The Creedmoor (N. C.) market is another striking illustration of this point. Creedmoor draws largely on the famous Dutchville section of Granville County for the tobacco sold in its warehouses.

The South Carolina law requires that each warehouseman report the grade of tobacco sold, together with the price. This grade, of course, can only be approximated, as the warehousemen have no way of knowing the grade except as a matter of judgment within broad lines. As reported, however, the 1909 sales of the South Carolina markets, which may be of interest, were divided as shown in Table V.

TABLE V.—*Record of the sales of tobacco on the loose-leaf tobacco market of South Carolina by grade and price for the year 1909.*

Grade.	Quantity.	Average price per 100 pounds.
	Pounds.	
Wrappers	158,386	$12.05
Stripping leaf	2,425,101	8.06
Leaf	24,101,811	7.44
Fillers	513,746	7.19
Common and medium leaf	1,212,801	6.00
Smokers and loose	1,077,141	5.79
Lugs and primings	2,322,518	4.52
Scrap	8,997	1.06
Total	31,820,501	7.27

Evidently most of the warehousemen were inclined to put all the leaf grades into one class, as leaf. The returns as a whole show entire lack of a system in reporting the grades.

The sales by months in the South Carolina markets, 1908 crop, were as follows:

Month	Pounds.
July	4, 529, 833
August	12, 870. 419
September	10, 140, 375
October	3, 976, 807
November	303. 085
Total	31. 820, 519

It is not to be supposed, of course, that these lists of markets are identical from year to year. The larger markets are generally fixtures, but the smaller ones are coming and going from year to year under the influence of various local causes.

SUMMARY OF SALES IN THE LOOSE-LEAF BRIGHT-TOBACCO MARKETS FOR TWELVE YEARS.

The sales of tobacco in the markets of the Old-Belt sections of Virginia and North Carolina and in those of the New-Belt sections of eastern North Carolina and South Carolina as a whole for the 12 years, from 1898 to 1909, inclusive, are shown in Table VI. The figures were kindly furnished by Dibrell Bros., of Danville, Va., from the returns obtained from warehousemen in all the markets, based on actual daily transactions. However, they include resales, so in order to get at the first-hand sales with approximate accuracy, they should be reduced by about 10 per cent. The correction is unnecessary, of course, in connection with the price averages.

TABLE VI.—*Sales of tobacco with the average price obtained on the Old-Belt and New-Belt flue-cured markets for 1898 to 1909, inclusive.*

Crop.	Market.	Sales.	Average price per 100 pounds.
		Pounds.	
1898	Old Belt (Virginia and North Carolina)	116,000,000	$6.50
1898	New Belt (eastern North Carolina)	47,000,000	6.75
1898	New Belt (South Carolina)	17,000,000	6.75
	Total	180,000,000	
1899	Old Belt (Virginia and North Carolina)	133,500,000	6.50
1899	New Belt (eastern North Carolina)	69,500,000	6.50
1899	New Belt (South Carolina)	25,000,000	6.50
	Total	228,000,000	
1900	Old Belt (Virginia and North Carolina)	104,000,000	7.25
1900	New Belt (eastern North Carolina)	58,500,000	7.75
1900	New Belt (South Carolina)	19,500,000	8.00
	Total	182,000,000	
1901	Old Belt (Virginia and North Carolina)	100,000,000	9.50
1901	New Belt (eastern North Carolina)	56,000,000	9.50
1901	New Belt (South Carolina)	17,000,000	9.50
	Total	173,000,000	

TABLE VI.—*Sales of tobacco with the average price obtained, etc.*—Continued.

Crop.	Market.	Sales.	Average price per 100 pounds.
		Pounds.	
1902	Old Belt (Virginia and North Carolina)	145,000,000	$9.00
1902	New Belt (eastern North Carolina)	101,000,000	10.50
1902	New Belt (South Carolina)	33,000,000	10.75
	Total	279,000,000	
1903	Old Belt (Virginia and North Carolina)	135,000,000	6.75
1903	New Belt (eastern North Carolina)	100,000,000	6.50
1903	New Belt (South Carolina)	42,000,000	5.25
	Total	277,000,000	
1904	Old Belt (Virginia and North Carolina)	124,500,000	8.50
1904	New Belt (eastern North Carolina)	52,000,000	8.85
1904	New Belt (South Carolina)	14,000,000	7.83
	Total	190,500,000	
1905	Old Belt (Virginia and North Carolina)	127,441,803	9.04
1905	New Belt (eastern North Carolina)	57,409,992	8.00
1905	New Belt (South Carolina)	18,372,475	7.50
	Total	203,224,270	
1906	Old Belt (Virginia and North Carolina)	127,358,877	8.81
1906	New Belt (eastern North Carolina)	49,538,582	9.75
1906	New Belt (South Carolina)	10,650,200	9.50
	Total	187,547,659	
1907	Old Belt (Virginia and North Carolina)	121,559,498	10.50
1907	New Belt (eastern North Carolina)	64,761,829	11.00
1907	New Belt (South Carolina)	22,424,520	9.60
	Total	208,745,847	
1908	Old Belt (Virginia and North Carolina)	148,007,216	9.80
1908	New Belt (eastern North Carolina)	79,793,020	9.10
1908	New Belt (South Carolina)	28,024,035	8.91
	Total	255,824,271	
1909	Old Belt (Virginia markets)	75,211,373	10.62
1909	Old Belt (North Carolina markets)	73,046,811	
1909	New Belt (eastern North Carolina)	84,581,682	8.29
1909	New Belt (South Carolina)	38,992,004	7.27
	Total	271,831,870	

SOME HISTORIC VIRGINIA TOBACCO TRADE AND MANUFACTURING CENTERS.

RICHMOND.

Up to the time of the Civil War, Richmond, Va., was preeminent in the manufacture of tobacco and as a center of the general leaf-tobacco trade. In later years, however, the scepter in both these lines has shifted to other cities, owing to a number of economic causes more or less well understood. This has been due, primarily, not to any diminished tobacco trade in Richmond, but rather to an outstripping rate of growth in other centers. Richmond still ranks high as a manufacturing city, and her leaf trade, while no longer preeminent in point of volume, is still very large. In point of variety and comprehensiveness of types handled, including all types of southern and western leaf, this city still may be regarded as occupying a commanding position among the most important leaf markets of the country.

As a market for the first-hand sale of leaf tobacco from farmers Richmond is easily beaten by a number of markets in Virginia and North Carolina. The total annual sales of first-hand tobacco amount to about 12,000,000 pounds. About 65 per cent of this class of sales is "sun cured," Richmond being practically the only market for this type. The other 35 per cent is divided between the dark types, principally olive stemming, flue cured, and Burley.

Richmond, although to a lesser extent than formerly, is still the center of a considerable leaf trade in inspected tobacco, which is shipped in hogsheads and consigned to commission merchants. This tobacco is sampled by sworn inspectors and sold at private sale by a method similar to that pursued at Baltimore, except that the warehousing and inspecting is done by a private board of trade rather than by regular State officials, subject, of course, to certain legal regulations in the interest of fair dealing. The general trade of dealers and commission merchants for the past few years has amounted to 15,000 to 20,000 hogsheads of tobacco yearly, including all types of flue-cured, dark-manufacturing, and export leaf, Burley, etc.

Prior to the concentration of the business, both domestic and foreign, into a comparatively few hands, the general dealers' trade in tobacco was much greater than now, and in proportion to the total trade in leaf tobacco the change is, of course, vastly more striking. Richmond in the old days was the natural center for this class of trade. Her leaf dealers had buying agents scattered through the growing districts in the upcountry markets, as they were called, in addition to which there was an army of large and small dealers and speculators buying on their own account. Much of the tobacco from these sources found its way to Richmond storage houses, having been consigned to commission merchants and dealers for sale. With few exceptions, the manufacturers purchased their leaf supplies from the dealers.

Now, however, this is greatly changed. The larger manufacturing concerns have their own extensive leaf departments for the purchase and handling of leaf direct from farmers without recourse to middlemen. Several of the most important régie Governments have also managed their leaf purchases in this country so as to concentrate them in a few hands, and the general brokerage trade in the régie types of tobacco is comparatively small. In the aggregate, however, the remnant of the commission and brokerage business between country buyers and manufacturers and foreign importers is still large, and Richmond ranks near or at the top in volume and variety of this class of trade.

A number of the resident buyers representing some of the most important foreign purchasers make their headquarters in Richmond. There are large stemming and rehandling plants and storage warehouses for leaf tobacco, and the city is also a very important center for the manufacturing of cigarettes, little cigars, cheroots, and a great variety of popular brands of cut-plug smoking tobaccos. Considerable flat plug and twist is manufactured there from the sun-cured and flue-cured types of leaf. Richmond is the largest center for the manufacture of cut-plug smoking tobacco, which constitutes the city's most important tobacco product. Burley tobacco is principally used in this class of goods, and it is estimated that nearly 75 per cent of the leaf tobacco manufactured in Richmond is Burley.

In 1909 Richmond factories used 16,125,590 pounds of stemmed and 8,903,189 pounds of unstemmed leaf, a total of more than 25,000,000 pounds, and produced 215,552,869 cigars, 1,525,138,650 cigarettes and little cigars, and 24,276,657 pounds of manufactured tobacco. Contemplating all phases of this tobacco trade, especially if its varied and comprehensive nature be given proper consideration, we find that Richmond still stands well to the front as a leading tobacco-trade center.

Mr. John L. Wingo, president of the Richmond Tobacco Trade, in his annual report of October 8, 1907, stated that, as estimated from the sources of information at his command, the total tobacco trade of Richmond through her factories, warehouses, and leaf plants amounts to about 70,000,000 pounds a year. The receipts of tobacco for the preceding year in public-bonded warehouses were 19,700 hogsheads and the deliveries were 19,459 hogsheads.

The tobacco trade of Richmond is organized under charter, with rules and by-laws for the control and best interests of the trade. An official inspector, who is sworn and bonded, is elected annually by the members of the tobacco-trade organization. Special rules have been made governing sales, reclamations, storage, nesting, reinspection, damage, etc. Until recent years the trade organization maintained an exchange, where hogshead tobacco was sold by sample. At the set time on sales days each dealer would appear at the exchange with his samples. Sales were conducted both privately and at auction. When sold at auction the samples were conveniently placed on a table, where the buyers could look them over and make their bids. The exchange feature, however, was discontinued about 1905, and inspected tobacco is now sold entirely at private sale in the dealer's office. Nearly all farmers' tobacco, however, is sold loose at auction on the warehouse floor.

Beginning in the winter of 1909–10 regular days for the sale of loose Burley at auction have been established in Richmond. The tobacco was derived largely from the scattered crops of Burley in

Virginia and from shipments made by West Virginia growers. About a million pounds of Burley were thus sold loose in Richmond during the first season and over 3,000,000 pounds the second season to the numerous Burley manufacturers in the city, who have given the market every possible encouragement.

LYNCHBURG AND PETERSBURG AS MARKET CENTERS.

Under the older régime Lynchburg and Petersburg were important market centers, particularly for the sale of first-hand farmers' tobacco. In this respect they considerably outranked Richmond itself. The general brokerage trade, however, while important in both cities, was not nearly so large and diversified as that of Richmond.

Before the war a large percentage of the air and coal cured manufacturing types as produced in what is now the older portion of the flue-cured territory was marketed through these centers, hauled over the roads, many times a hundred miles or more, in wagons, or more often by rolling the hogshead itself by means of shafts attached to an axle driven through the center of the hogshead. Several neighbors with their entire families would frequently club together for this market trip, taking along provisions and cooking utensils and making it a holiday excursion of mirthfulness and pleasure.

With the development of the numerous leaf-tobacco markets throughout the flue-cured tobacco sections since 1870, the trade in leaf at these points has been confined almost exclusively to the local type as produced in the territory immediately tributary. The producing territory tributary to Lynchburg is considerably larger than that of Petersburg, and its leaf trade has, therefore, been better maintained, though not as large as in the seventies and eighties of the last century, when it was at its maximum. Lynchburg at present is the largest market in Virginia for the sale of dark tobacco. The quantity of leaf sold at first hand from farmers on both these markets in 1908 will be found on page 27.

Both Lynchburg and Petersburg were also important manufacturing centers and each had a large number of factories engaged principally in the production of plug chewing tobacco. The manufacturing interests of both cities are still considerable, although the manufactures of Petersburg have been much better maintained than those of Lynchburg because of the location there of important manufacturing plants engaged principally in the manufacture of plug tobacco for export. Since 1906 an export cigarette factory also has been located there. However, this factory was closed down temporarily in 1910 because of a very great increase in the duty levied on all forms of tobacco, including cigarettes, by India, to which country the product of the factory at that time was almost exclusively exported.

In total volume of exports of manufactured tobacco Petersburg far outranks any other center, and, in fact, exceeds all other centers combined. Some of the factories there are devoted exclusively to the manufacture of tobacco for export.

The total tobacco product from the factories of Petersburg in 1909 was 5,634,064 pounds of plug, 356,000,000 cigarettes, and 16,048,172 cigars. Of this, all the cigarettes and nearly all the plug—that is, 5,593,175 pounds—were exported.

DEVELOPMENT OF THE WESTERN MARKETS.

STATE REGULATIONS OF INSPECTIONS IN THE WESTERN MARKETS.

The early inspection laws of Louisiana provided that the inspectors should declare the quality of the tobacco as first, second, or third grade, and should brand it upon the head of the hogshead. Since western tobacco was exported via New Orleans and passed through its inspections, the early inspection laws of the Western States generally provided for a similar declaration of quality by the local inspectors. Before statehood had been attained the Virginia inspection laws applied in Kentucky, and those of North Carolina, which were similar to the Virginia laws, applied in Tennessee. After independence and statehood had been achieved these laws, in basis at least, were adopted in the new States, but were frequently revised and modified as occasion demanded.

In 1798 the Kentucky Legislature reduced the provisions of several previous acts to a single one. By this act tobacco could be exported only after its inspection in hogsheads. Inspections were established at one or more points in the counties of Garrard, Mercer, Madison, Fayette, Clark, Nelson, Washington, Mason, Bourbon, Franklin, Jefferson (Louisville), Logan, Bracken, Warren, and Campbell. Warehouses were privately owned, but the owners received rent according to the quantity of tobacco inspected. In general, the provisions covering the inspections were all handed down from the Virginia statutes.

In 1810 the law prohibiting the exportation of tobacco without inspection was repealed in regard to tobacco in the hands of growers, and in 1825 that part applying to tobacco in the hands of dealers was also repealed. In 1820, following the Louisiana statute, the legislature provided for the classification of tobacco into three grades. In 1860 a law was passed whereby the city council of Louisville could establish a private warehouse system in that city, the inspectors to be appointed by the governor, with alternates appointed by the city council to act in the absence of the regular appointees. In 1864 the alternate inspectors were appointed by the proprietors of the warehouses instead of by the city council. In

1865 the legislature provided that the governor should appoint for the four warehouses then in operation in Louisville four inspectors, who should draw and tie the samples and seal them with wax, the inspectors to alternate at the different warehouses. In the early seventies the change to an entirely private system of inspection upon essentially the same basis as exists to-day took place.

In Tennessee the legislation concerning tobacco inspections was very similar to that in Kentucky. In 1843 the Tennessee Legislature memorialized the Louisiana Legislature urging a change in the inspection laws to the sample basis, similar to the Virginia method, claiming that the declaration of grades merely as a basis for determining the price gave but little encouragement to planters to produce quality rather than quantity. Among the earliest inspection and shipping points in Tennessee were Carthage, Nashville, and Clarksville. The acts of 1817, 1840, 1843, and 1858 provided for the appointment of inspectors by the county courts. By the law of 1858 a county court could either build or rent warehouses; and if the court failed to appoint inspectors, the governor of the State could appoint them. None of these Tennessee laws, however, was compulsory in its nature. The Civil War caused the temporary closing of all the Tennessee markets. The foundation of the present Tennessee inspection laws was laid in the act of 1870, by which the warehouse keeper is appointed inspector under a $5,000 bond to the State. The hogshead is broken in four places and the sample sealed with wax.

In Missouri a compulsory State warehouse and inspection system was provided for at St. Louis in the act of 1843, although considerable tobacco had been handled there for several years previous to that date. The keeper of the State warehouse was appointed by the governor, as also were two of the three members of the board of inspectors, who were to hold office for two years. The third inspector, who could act only in case of the disagreement of the appointees of the governor, was appointed by the county court of St. Louis. Two to four breaks in the hogshead were to be made, and 10 pounds was to be deducted for the weight of the sample. In 1871 the law provided that only one inspector should be appointed by the governor.

Tobacco-inspection laws were enacted in Indiana as early as 1843, providing for the appointment of inspectors by the board of county commissioners, who were to declare the quality by grades, as was the practice in other Western States and at New Orleans. By the law of 1857 the inspectors were to be appointed by the judge of the circuit court, and auction sales were not permitted unless the tobacco had been sampled by such inspectors. Evansville was the important tobacco warehouse and inspection point in Indiana. In Ohio the first inspection laws were passed in 1826, but they were not compulsory. The act of 1856 provided for the appointment of inspectors by the judge of the probate court. Cincinnati, quite naturally, early became the most important center of tobacco trade for southern Ohio and central and northern Kentucky.

Notwithstanding all this early legislation, however, there is nothing to indicate that the warehousing or inspection of tobacco at any of these new western centers of trade amounted to much before about 1830 or 1840. In fact, with two or three exceptions, notably at Clarksville, Tenn., and at St. Louis, Mo., no large quantity of tobacco was inspected or warehoused at any of these centers up to the outbreak of the Civil War, which completely closed the New Orleans and Tennessee markets, with the result that other western centers, particularly Louisville and Cincinnati, expanded very rapidly at that time. The foundation, permitting the expansion at that time, however, had been laid by the development of such transportation facilities as steamboats, roads, canals, and railways.

EARLY PERIOD BEFORE THE CIVIL WAR.

In a previous bulletin[1] we have seen that tobacco culture in the great new region west of the Alleghenies had its origin as a commercial industry among the pioneers from the tobacco-growing sections of Virginia, North Carolina, and Maryland. Indeed, up to the time of the Revolutionary War the present States of Ohio, Kentucky, and Tennessee were really but the undeveloped western portions of the States of Virginia and North Carolina. Before the Revolutionary War this westward pioneering movement was confined largely to backwoodsmen, explorers, hunters, and outlaws.

The Cumberland Gap, in what is now southwestern Virginia, formed the main gateway in crossing the Alleghenies to the new western country. Only a short distance beyond this famous pass in eastern Tennessee lay the rugged but beautiful and fertile valleys of the French Broad, Holston, Watauga, and Nolichucky Rivers. Some of the earliest western settlements were made in this section of eastern Tennessee, and the pioneers no doubt raised tobacco very early for their own use. The difficulties of transportation, however, were such that commercial shipments in any volume could not be seriously considered.

Farther west, in the present State of Kentucky, some of the earliest settlements were made in the beautiful rich country of the Kentucky River, Lexington being one of the first. It is probable that the infant beginnings in producing tobacco on a commercial scale

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[1] Bulletin 244, Bureau of Plant Industry, entitled "The Export and Manufacturing Tobaccos of the United States, with Brief Reference to the Cigar Types," 1912.

for export in this new country took place in this fertile section. Killebrew (Tenth Census Report) makes a note indicating that small shipments of tobacco were made from the Lexington section as early as 1783 or 1785. About this time also the first mention is made in the acts of the Virginia Assembly of the establishment of State inspections for tobacco in this new section.

It was, however, considerably later than this that the production and shipment of tobaccos from the West became of sufficient importance to command attention. In the period immediately succeeding the Revolution settlers, attracted by the great quantities of cheap, fertile land, began to move into this new country in rapidly increasing numbers.

Much of eastern Kentucky and Tennessee, particularly Kentucky, was hilly, and it all was heavily timbered. Farther on in the central and western sections of these States the lay of the land was better for agricultural purposes. Large areas were free from timber growth, although heavily sodded with native grasses. It thus readily supported domestic live stock, harbored great quantities of large game useful for food purposes, such as buffalo and deer, and it was comparatively easy to bring large fields into cultivation without first laboriously clearing the land of a heavy growth of timber.

The early settlements for both farming and trade were quite generally located at favorable points along or in the vicinity of the navigable portions of the numerous streams, particularly the Cumberland, Ohio, Tennessee, Kentucky, and Green Rivers, and many were pushed farther on up the Missouri or the Mississippi.

By 1810 the development of tobacco growing was beginning to be of real importance in a number of sections, particularly along the Cumberland, Green, Kentucky, and Ohio Rivers. In this year there were two small tobacco factories at Lexington, Ky., and mention is made of the beginning of trade in tobacco at Louisville. In 1819 two establishments at Louisville were engaged in putting up strips and other manufactured tobacco for local consumption. In selling this tobacco, produced almost entirely by settlers from the East, at first thought it would seem natural that it should be sold in the eastern markets, such as Richmond, Petersburg, and Baltimore. However, the difficulties of transportation upstream and over the mountains were entirely unfavorable to this course. In those days, before the advent of railways, the tobacco could much easier be floated downstream via the Mississippi River and its numerous navigable tributaries, which extend into all parts of the producing territory, reaching ocean navigation at the then already flourishing French commercial city, New Orleans. New Orleans was the natural southern outlet for the many trading posts of the great Mississippi waterway and its tributaries, particularly those established by the French many years earlier along their important trade highway from the Gulf to the St. Lawrence.

NEW ORLEANS THE PRINCIPAL EXPORT POINT FOR WESTERN TOBACCO.

The exportation of tobacco from New Orleans had been going on in a small way for many years. From the earliest settlement the Louisiana planters had introduced the cultivation of tobacco for trade purposes with their home country, France. Produced on the rich Mississippi alluvium, however, this Louisiana tobacco was coarse and strong and the quality poor. As early as 1785, probably, the first small shipments of Kentucky tobacco reached New Orleans, and by 1810 the trade had begun to be of real importance. The inferior Louisiana tobacco was unable to compete with the steadily increasing supplies of better quality from the Ohio and Cumberland River districts, and its cultivation was soon almost completely abandoned.

In these earliest days a number of planters would combine and ship their hogsheads of tobacco to New Orleans on flatboats in charge of an agent called a "freighter," whose duty it was to look after their interests in the transportation and sale of the tobacco and to purchase supplies with the proceeds. The tobacco trade of New Orleans increased until the annual inspections and shipments amounted to more than 100,000 hogsheads annually in the years immediately preceding the Civil War.

The consignment of tobacco to New Orleans factors direct by planters, however, necessarily entailed a long wait, often several months, before returns could be had, which delay, of course, was often unsatisfactory. The distance was great and communication so slow that complaints were very difficult of satisfactory adjustment. While most of the tobacco ultimately made its way to New Orleans, considerable trading in tobacco by local merchants at many of the more important river settlements naturally sprang up. This sort of trading, in the nature of advances to farmers or of speculation by merchants, in its earliest days was unsystematically carried on, and its beginnings are clouded in obscurity.

IMPORTANCE OF THE DEVELOPMENT OF MEANS OF TRANSPORTATION.

In 1811 the first steamboat appeared on the waters of the Ohio and Mississippi Rivers. The subsequent rapid increase in steam navigation was, of course, of much assistance to the general river trade, including that in tobacco.

In 1818 the Cumberland road to Wheeling on the Upper Ohio was completed and proved not only a great aid to settlers who were pouring in in ever-increasing numbers from Virginia and Maryland, but was also useful in fostering a rapidly increasing wagon trade with the eastern cities, particularly Philadelphia, Baltimore, and Richmond. On the return trips tobacco was one of the most important items.

In 1825 the Erie Canal, which opened up a fairly convenient northern trade route to New York, was completed. The Miami River Canal, completed from the Great Lakes to Cincinnati in 1842, opened up an all-water route to the seaboard at New York, which greatly helped general trade with the East, and particularly the tobacco trade of Cincinnati.

By 1860 the Middle West was also well served by railroad communication with all the important eastern seaboard cities. Up to the time of the Civil War, however, New Orleans received the larger proportion of western tobacco, except such as was used for home manufacturing, an industry already becoming of considerable importance in a number of local centers, particularly Louisville, St. Louis, and Cincinnati. However, much of this tobacco had first passed through the hands of local traders in the smaller primary markets which had sprung up at strategic points in or near the producing territory, such as St. Louis, Clarksville, Louisville, and Cincinnati. The representatives of the European régie Governments and the foreign agents of the great German and English tobacco markets were also located principally at New Orleans.

BEGINNINGS OF THE MORE IMPORTANT MARKET CENTERS.

The tobacco trade of St. Louis, which soon became the most important western market except New Orleans, began about 1830 and was soon placed under a system of State inspection similar to those of Virginia and Maryland. Much tobacco was received from Kentucky and Indiana, as well as from the producing territory in Missouri that is directly tributary. As early as 1850 the sales on the St. Louis market were about 20,000 hogsheads annually.

At Clarksville, Tenn., the center of one of the most important growing districts, some small factories were established as early as 1835, which put up dried leaf and strips principally for the English market. This business increased, and by 1844 the Clarksville tobacco market had become regularly established and was a great convenience to farmers who prized their own tobacco. In 1856 (selling the 1855 crop) the sales at Clarksville reached the highest figure prior to the Civil War, i. e., 18,000 hogsheads.

Louisville did not become a market of great importance till the early sixties, when it began to increase its tobacco trade enormously, the cause being the closing of both the Clarksville and New Orleans markets, which were within the limits of the active war territory, and the opening up in 1860 of the Louisville & Nashville Railroad, which passed through much of the most important tobacco territory. Mention has already been made of the beginning of the tobacco trade in Louisville as early as 1810. The first strictly tobacco warehouse (Booth's) seems to have been built in Louisville in 1825. During 1826, 1,100 hogsheads were sold from this warehouse at an average price of $2.67 per 100 pounds. In 1858 the Louisville tobacco trade had expanded to about 8,000 hogsheads a year.

Cincinnati was a natural market point for some of the earliest producing districts of Kentucky and Ohio. It is probable that there was more or less trade in tobacco at Cincinnati almost from the founding of the settlement. The records of the Cincinnati Board of Trade show receipts of 6,078 hogsheads and 655 boxes and bales as early as 1845. Official inspections and break sales, however, were not inaugurated there until 1853, when Mr. R. K. Love, formerly of Baltimore, established the Cincinnati warehouse. The inspector was elected by the popular vote of the city, and his duties and the rules for conducting the warehouses were specified in quite an elaborate city ordinance. In 1856 this method of electing inspectors was changed by State law, and they were thereafter appointed by the judges of the probate court.

In 1860 the receipts of tobacco at Cincinnati were 14,480 hogsheads. As with Louisville, the closing of the New Orleans and Clarksville markets by the war was a great stimulus to the tobacco trade of Cincinnati. Its real development and maintenance in later years, however, depended more directly upon the introduction of White Burley, as will be more fully described later in these pages.

Evansville, Ind., was also quite an important inspection market for a number of years. It drew its trade largely from the southern Indiana tobacco district, which in earlier times was of much greater importance than now. Some tobacco from conveniently located parts of Kentucky was also marketed at Evansville.

According to Killebrew, Nashville had begun to handle considerable tobacco as early as 1835. It dealt principally in the types produced in the upper Cumberland district, with which it was in direct and easy communication by means of the Cumberland River. It also secured most of the tobacco produced south of Nashville, in Williamson and Maury Counties, and a little from Robertson County and other parts of the Clarksville and Paducah districts most adjacent. Some strips were also put up there, and the total trade in tobacco in Nashville, at its maximum prior to the Civil War, amounted to about 7,000 or 8,000 hogsheads annually. As with Clarksville, the breaking out of the war put a stop to the tobacco trade of Nashville, and it was not reestablished until some years after the cessation of hostilities.

Paducah is also one of the older markets. It was established as a break market in the early fifties, but became of little importance until the Civil War.

Henderson and Owensboro, Ky., though for many years important centers of trade in the leaf produced in the territory tributary to them, known as the Henderson, or "stemming," and the lower Green River districts, respectively, have never, except for brief periods, developed into regular inspection or break markets. The districts which they serve along the lower Ohio were not settled as early and did not start the commercial production of tobacco as soon as the central counties of the State along the Kentucky River, or the southern counties, particularly Logan and Barren. Killebrew states, however, that shipments of tobacco were made to Henderson from the tributary territories as early as 1835, and that stemmeries were established there at that time.

DEVELOPMENT OF THE WESTERN MARKETS SINCE THE CIVIL WAR.

CHANGE OF TRADE FROM NEW ORLEANS TO NEW YORK.

Mention has already been made of the marked change in the market conditions in the West by the outbreak of the Civil War. New Orleans was put under an effective blockade and the continuance of the tobacco trade there was no longer possible. The foreign representatives and the tobacco trade of New Orleans as a whole changed their headquarters to New York, which had already begun to make some progress in trade under the influence of the better and more satisfactory transportation facilities which had become available. For the most part this trade never returned to New Orleans, and New York became at once the great leaf market of the country for export tobacco, and, indeed, it is to this day the principal export point, although it has now become of but little importance so far as its leaf trade is concerned. The foreign representatives have made a still further change and now reside and buy for the most part at the primary western markets, as near as possible to the sections where the tobacco is produced. This exodus of foreign buyers, dealers, and brokers from New York went on at a rapid rate in the early eighties, and their head offices are now located principally at Louisville for the western types and at Baltimore and Richmond for the eastern types of tobacco. The great bulk of the tobacco that was formerly inspected or resold at New York now merely passes through that port in transit to foreign countries or is billed direct to the large tobacco factories in and tributary to that city.

There is quite a volume of leaf that continues to pass through New Orleans in transit to foreign countries, particularly to Germany and England, and it is in this respect the third largest export point for tobacco in the country, New York standing first by a large margin, and Baltimore second.

GROWTH OF THE LOUISVILLE MARKET.

Reference has already been made to the rapid growth of the Louisville market as a result of the Civil War and the almost coincident opening up of the Louisville and Nashville Railroad. The war closed both the Clarksville and the Nashville markets, and most of the tobacco which had formerly moved through those points, as well as a portion of that which went to New Orleans direct, came to Louisville. Sales at Louisville, which in 1857 were 8,000 hogsheads, rose in 1862 to 29,500 and in 1865 to 63,000 hogsheads, when, for the first time, Louisville ranked as the largest tobacco market of the country.

After the war the Clarksville and Nashville markets were reopened, and in 1869 a market was started at Hopkinsville, Ky. There was no material increase in the quantity of tobacco handled in Louisville from 1865 until the early eighties and, indeed, the receipts were often substantially less. This market did not receive any considerable quantity of the new White Burley tobacco until some years after its introduction. It was first produced in the territory more directly tributary to the Cincinnati market, in Brown County, Ohio, and Mason County, Ky., from which points it gradually spread as its popularity increased. In 1879, the first year that separate figures are available, out of total receipts of 48,852 hogsheads on the Louisville market, 9,684 hogsheads were Burley. As the production of Burley spread toward the West, more and more of it tended to move to Louisville, until in 1883 came the first big year in this type, when of the total receipts of 74,866 hogsheads more than 50 per cent was Burley, and it has ever since held the most prominent place among the leaf sales at Louisville.

In 1885 Louisville's receipts of all types of leaf for the first time exceeded 100,000 hogsheads, the actual number being 108,821 hogsheads, 40,000 hogsheads more than at any other market in the country, and since then her prestige in the leaf trade in point of the volume of receipts and sales has been easily supreme. Since 1885 the receipts of tobacco at Louisville have averaged about 100,000 hogsheads, often considerably exceeding it.

GROWTH OF THE CINCINNATI MARKET.

The Cincinnati market was also greatly helped by the Civil War, though, perhaps, less extensively than the Louisville market. The prominence of Cincinnati as a tobacco market has depended largely on the popularity of White Burley, which at first was produced exclusively, and always principally, in the territory naturally tributary to Cincinnati. By the early seventies the production of Burley had increased to such an extent as materially to expand the receipts and sales of tobacco at Cincinnati. In 1872 the receipts were 24,198 hogsheads and by 1878 they had risen to 38,999 hogsheads. By this time the great value of Burley for plug fillers began to be fully recognized, which constituted the greatest stimulus to the production of this type. The acreage spread with remarkable rapidity during the eighties, until the great crop of 1888 swelled the receipts of Burley at Cincinnati to 90,000 hogsheads in 1889, the maximum receipts for that city. In 1890 the receipts declined under the influence of very low prices and greatly curtailed planting to 56,070 hogsheads, and until 1903 ranged from about 50,000 to 80,000 hogsheads. Since 1904 new factors have been introduced into the situation, which have more seriously affected Cincinnati than any other market; that is, the marketing of tobacco through the Burley growers' organization, which had its principal strength in the Cincinnati territory, and the increased country buying by the larger manufacturing interests, either at the grower's farm or on the floor of the loose-leaf tobacco auction houses at Lexington and elsewhere.

It will, perhaps, be found interesting to observe the sources from which the Burley receipts at Cincinnati were derived, say, in 1882, and again in 1902, twenty years later. It has been the custom in this market to divide into districts the producing territory from which it drew, giving each the name of the principal county in the district or some other suitable and suggestive title as to its origin. An approximate distribution of the receipts for the years mentioned is shown in Table VII.

TABLE VII.—*Receipts of Burley tobacco on the Cincinnati market by districts in 1882 and in 1902.*

Districts.	1882	1902
	Hhds.	*Hhds.*
Brown County, Ohio	10,824	10,586
Mason County, Ky	14,436	17,644
Pendleton County, Ky	10,701	12,786
Owen County, Ky	8,053	12,317
The bluegrass counties, Kentucky	2,279	9,008
Southern Kentucky counties	80	....
Eastern Ohio district	1,530	1
Indiana and Illinois	429	54
West Virginia	846	1,493
Tennessee, Missouri, etc	79	23
Total	49,257	63,910

The point to be noted in this table is that nearly all of the increase in this 20-year period has been in the Kentucky portion of the Burley district, particularly in the bluegrass counties.

The territory included within these districts is distributed about as follows:

Brown County district of Ohio:
 Brown, Adams, Clermont, and Scioto Counties.
Mason County district of Kentucky:
 Mason, Bracken, Lewis, Fleming, Nicholas, and Robertson Counties.
Pendleton County district of Kentucky:
 Pendleton, Harrison, Grant, Boone, Kenton, and Campbell Counties.
Owen County district of Kentucky:
 Owen, Gallatin, Carroll, Scott, Franklin, Henry, Trimble, and Oldham Counties.
Bluegrass district of Kentucky:
 Bourbon, Fayette, Woodford, Clark, Montgomery, Bath, Jessamine, Anderson, Shelby, Boyle, Garrard, Madison, Mercer, Nelson, Spencer, Washington, and Bullitt Counties.

The total production of all of these groups of counties is, of course, much greater than is shown in the Cincinnati receipts. Large quantities of tobacco from this territory go to Louisville or are bought in the country directly from farmers by manufacturers and others and are never recorded in the receipts of any market.

More recently these subdivisions of the Burley-producing territory in Kentucky into districts have been largely lost sight of, except to distinguish in a general way the bluegrass section from the other regular producing territory.

CIGAR-LEAF BREAKS IN CINCINNATI.

An interesting feature of the Cincinnati market is the fact that since some time before the Civil War, when cigar leaf first began to be raised in the Miami Valley and to some extent in other tributary sections, that city has always been the center of a large trade in cigar leaf in boxes of about 300 pounds net weight each, this tobacco being inspected in the regular way and sold from the sample at auction the same as Burley. Cincinnati is the only regular cigar-leaf auction sale and inspection market in the United States. At present these sales consist mostly of odds and ends or damaged leaf from the cigar districts of Ohio and Wisconsin. The sales of seed leaf at the Cincinnati breaks have generally ranged from 5,000 to 10,000 cases annually. Figure 3 shows a view of one of the cigar-leaf breaks just prior to the beginning of the auction sale.

GROWTH OF THE CLARKSVILLE MARKET.

The Clarksville market was reopened immediately after the Civil War and its trade reached the maximum of development in 1887, after the great crop of 1886, when the receipts were 41,000 hogsheads, or fully 60,000,000 pounds, of tobacco. The large crops of

1886 and 1888 were followed by several years of depression in the production of dark tobacco, and the receipts at Clarksville have since averaged about 20,000 to 30,000 hogsheads yearly. Although the Clarksville market has fallen considerably behind both Louisville and Cincinnati since the war in the gross quantity of tobacco handled, it has, nevertheless, easily retained its preeminence as the largest and most important distinctively dark-tobacco market in the country, generally outranking by 10,000 to 15,000 hogsheads any other dark-tobacco market of the country.

Among the other six distinctively dark-tobacco auction break and inspection markets which have been in operation in the West since

FIG. 3.—Interior of a cigar-tobacco auction warehouse, Cincinnati, Ohio, before a sale. The official sample on which the sale is made lies on the top of each case.

the Civil War, St. Louis was as important as any, even including Clarksville, up to about 1879, and the receipts ran from about 15,000 to 20,000 hogsheads yearly. The largest crop of tobacco ever raised in Missouri was in 1876, when the crop was 43,000,000 pounds. Because of the heavy production at this period of dark tobacco of a type similar to that produced in Missouri, together with the rapid inroads which Burley was making in competition with the dark western manufacturing type and the consequent very low prices for dark tobacco, the production in Missouri rapidly declined until, in 1879, only three years later, the crop was only 12,015,657 pounds, and a portion of that had been changed to Burley. The receipts of St. Louis were drawn principally from the Missouri territory, and its volume of trade in tobacco leaf, of course, also suffered a coincident decline. The Missouri crop of 1880 was estimated to be about 50 per cent Burley, and it continued thereafter to fluctuate greatly between the two types, Burley and dark, the total crop generally not exceeding 10,000,000 to 12,000,000 pounds and dwindling rapidly, until in the early years of the present century, with large yields elsewhere, accompanied by very low prices, the production in Missouri practically ceased. For several years prior to 1907 there were probably less than a million pounds of tobacco annually produced in the entire State.

With the destruction of the Pepper warehouse by fire in 1898 the last of the public warehouses for the open sale of tobacco in St. Louis came to an end. The receipts had been averaging only about 5,000 to 8,000 hogsheads since 1889.

Evansville, drawing its receipts of tobacco principally from the southern Indiana district, was never a very important market. During its best days, in the seventies, when the southern Indiana crops were largest, the receipts ran about 5,000 hogsheads of tobacco yearly. Stemmeries were located at Evansville, in addition to those locally scattered through the district at such points as Rockport, Dale, and Booneville. Following the depression resulting from the great crops of the late eighties the receipts at Evansville in 1889 were but 1,210 hogsheads. This was the last year of the market, and the remaining warehousemen removed their headquarters to Louisville.

The Nashville market, reopened in 1871, did not regain its trade rapidly after the Civil War. It was, indeed, not until 1876 that receipts, following the large crop of 1875, reached the size they had obtained before the war. In 1876 they amounted to 9,000 hogsheads, which proved the high figure for the market. In 1881 the receipts declined to 2,707 hogsheads, but reached 8,000 hogsheads in 1889. From that time the market has continued to dwindle, owing to a decreased production in the upper Cumberland district, together with diversion of shipments, principally to Louisville. Since 1896 the receipts have not exceeded 2,000 hogsheads, except in one year, 1899. At the end of the sales season of 1900 the warehouses were closed and have not since been reopened.

The Paducah market for a number of years after the war ranked quite close to St. Louis and Clarksville and in one year, 1876, outranked either of them by more than 4,000 hogsheads, with receipts of 20,834 hogsheads. The receipts declined to 8,377 hogsheads, however, in 1879. Until very recently they have since varied generally from 12,000 to 20,000 hogsheads, reaching their maximum in 1889, since which they have gradually declined. In 1894, in common with other dark-tobacco markets, Paducah's receipts were very small, but this market regained its position and in 1895 and 1896 the receipts amounted to over 16,000 hogsheads in each year. These figures have not again been reached, however, and since 1897 Hopkinsville has outranked Paducah as the most important dark-tobacco market except Clarksville.

The Mayfield (Ky.) market, established as an auction and inspection market in 1876 and located more centrally in the heart of a great producing section, cut heavily into Paducah's territory, and since 1897 the receipts at Paducah have generally been only about 8,000 to 12,000 hogsheads of tobacco annually. Since 1899 the receipts of tobacco at Mayfield have generally exceeded those of Paducah by about 2,000 hogsheads annually.

Hopkinsville, Ky., situated, like Mayfield, in the center of a great producing territory, was not established as an auction and inspection market until after the Civil War. Besides Mayfield it is the only important inspection market of the country not situated on navigable water. Both Hopkinsville and Mayfield were established after the railroads became the more important means of conveyance in shipping tobacco and facilities for shipment by water were no longer essential to the success of a market. Hopkinsville entered the field as an open-break market in 1869. Owing to its favorable situation in relation to the producing territory its development was quite rapid. By 1896 its receipts had risen to 21,525 hogsheads, the highest ever attained. In 1897 the receipts were again high (19,950 hogsheads), but they have not since exceeded 15,000 hogsheads except in 1899, when there was a total of 15,930 hogsheads. The receipts have generally ranged from 10,000 to 14,000 hogsheads, Hopkinsville ranking next to Clarksville among the purely dark-tobacco markets of the West.

CAUSES FOR THE DECLINE IN RECEIPTS SINCE THE LATE EIGHTIES.

In looking over the foregoing figures, covering the receipts at the various western markets, it will be noticed that the maximum receipts in most cases were reached by the larger markets in the late eighties. For example, the maximum receipts of the Louisville and Clarksville markets were reached in 1887 and at Cincinnati in 1889. The lack of future growth or the actual decline which was experienced was not due in any substantial degree to a cessation in the expansion of the production of tobacco, nor to the splitting up of the trade among an increased number of markets. On the other hand, there has been an actual decrease in the number of markets operated. St. Louis, Nashville, and Evansville have all been closed as tobacco markets during the period. The Louisville market, being the largest, has been the best maintained, apparently having profited somewhat by the closing down or reductions of other markets, but even this help has hardly been sufficient to make good the losses from other causes. As it is, the receipts at Louisville and Cincinnati probably would not have been so well maintained were it not the general custom of the warehousemen at these points to keep up the business by actually buying through agents or at least financing the buying of loose tobacco from farmers, packing it in hogsheads, and bringing it to their warehouses for sale.

The western crop, particularly Burley, is much larger now than in the eighties of the last century. The chief reason this has not been reflected in increased market receipts is the direct buying from farmers by the great domestic and foreign manufacturing interests. The movement to make purchases direct from farmers at the barn or from the wagon in practically all of the towns of any size throughout the producing territory was begun in the late eighties and has continued to increase to the present day. But little of the very large volume of tobacco bought in this way by ultimate consumers, of course, ever appears in the regular statements of receipts, inspections, or sales of any market. Since 1905, also, the receipts of all of these markets through the regular trade channels have been greatly affected by the handling and sale of large quantities of tobacco through farmers' pooling organizations.

PRESENT STATUS OF THE WESTERN MARKETS.

DEVELOPMENT OF THE DIRECT-BUYING TENDENCY.

As already noted, the tendency of large manufacturers and the régie Governments to make the bulk of their purchases of tobacco leaf direct from farmers in the country had begun to have its effect in reducing the volume of trade through the open-break markets in the early nineties of the last century.

The maximum receipts in the combined markets of the West were reached in 1889, following the great crop of 1888. These receipts were divided among the nine markets of the West then in operation as follows:

TABLE VIII.—*Receipts of tobacco in the western markets in 1889.*

Market.	Hogsheads.	Market.	Hogsheads.
Louisville	107,994	Nashville	8,074
Cincinnati	90,091	St. Louis	7,104
Clarksville	40,436	Evansville	1,210
Paducah	23,087		
Mayfield	11,182	Total	299,367
Hopkinsville	10,189		

These combined receipts of nearly 300,000 hogsheads represented about 375,000,000 pounds of tobacco and indicate, therefore, that at that time practically all of the tobacco produced in the West passed through the regular channels of trade in these markets. In fact, a careful estimate of the tobacco crop of the State of Kentucky for the preceding year, 1888, by districts and counties, made by Mr. E. T. Girard, a broker of Louisville, gives a total production for Kentucky of 283,200,591 pounds, divided by districts as shown in Table IX.

TABLE IX.—*Estimated production of tobacco in Kentucky, by districts, in 1888.*

District.	Pounds.
Paducah, or western (in Kentucky)	37,690,420
Clarksville (in Kentucky)	44,473,470
Henderson and lower Green River	69,009,422
Upper Green River	29,937,294
Upper Cumberland	1,968,884
Mountain and scattered	844,249
Total dark types	183,923,739
Burley (Kentucky)	99,276,842
Total crop in Kentucky	283,200,581

An examination of Table IX brings out the interesting fact that the tobacco produced in the dark districts of Kentucky in 1888 was approximately equal in quantity to that of the present time. It is a reasonable inference, therefore, that the other dark-tobacco sections of the West were producing about the same quantity as at present. The total Burley crop of the year 1888 was currently estimated by the trade at about 125,000,000 pounds. In order to obtain the total western production of tobacco in that year, therefore, there should be added to the Kentucky production from Tennessee about 48.000.000 pounds of dark tobacco; from Missouri about 12,000,000 pounds of dark tobacco; from Indiana about 7,000,000 pounds of dark tobacco: and from Ohio, Indiana, West Virginia, etc., about 25,000,000 pounds of Burley, which makes the total western crop of 1888, exclusive of Kentucky, about 92,000,000 pounds.

This quantity checks very closely with the receipts, 299,367 hogsheads, at the western markets then open and shows that at that time the country buying by manufacturers and exporters was of small proportions. An approximate estimate of the increase in country buying may be obtained by comparing these figures with those of, say, 1903, when the total production of tobacco in the West was around 425,000,000 pounds, including an unusually large crop of all the dark types.

The total market receipts of the six western markets in 1904, when most of the 1903 crop was sold, are shown in Table X.

TABLE X.—*Receipts of tobacco in the western markets in 1904.*

Market.	Hogsheads.	Market.	Hogsheads.
Cincinnati	21,022	Paducah	8,690
Louisville	84,104	Mayfield	10,772
Clarksville	21,220		
Hopkinsville	14,930	Total	160,738

These 160,738 hogsheads probably represented 200,000,000 to 225,-000,000 pounds of leaf, or approximately half of the total production of the year before, the other 50 per cent having evidently been bought in the country direct from the farmers by the large manufacturers and exporters.

INFLUENCE OF THE POOLING MOVEMENT ON THE LARGER MARKETS.

By 1906 another influence, adverse to the regular markets, began to make itself felt. In the early years of this decade the prices of leaf tobacco were very low, and in 1904 producers of the dark types received only about 4 to 5 cents per pound, while the cost of production, on the other hand, was increasing, owing to the higher cost of labor and general farm supplies. As early as 1901 agitation by farmers to reduce the acreage and for pooling the crop in the interest of higher prices had begun, and the very low prices of 1904 caused this agitation to become exceedingly active in all sections of the western tobacco territory.

The Dark District Planters' Association (the predecessor of the Planters' Protective Association) was organized, and as a result of the pooling of the crop, combined with a decreased production, the receipts at all the western dark-tobacco markets fell markedly in 1905, and, with a rapid spread of the pooling movement, to very low figures in 1906.

Likewise, in the Burley district the movement for pooling and reduction of acreage, begun in 1902 or earlier, gained headway and, chiefly as a result of the efforts of the growers' organization, the acreage planted in 1908 in the principal Burley sections of Ohio and Kentucky fell to only about 18 per cent of the normal. The crops of this period were handled largely through pooling.

In 1907 and 1908 the effect of this pooling movement in Burley was felt with peculiar force in Cincinnati, and in 1909 the receipts there fell to almost nominal proportions compared with the trade of former years.

By 1906 such pooling movements of farmers had become established in nearly all the regular producing sections of the West, including the "Stemming" and Green River districts and portions of the one-sucker territory, particularly in the Bowling Green section and in southern Indiana.

Nearly all of this pooled tobacco was sold direct to final purchasers without passing through the regular trade channels. The common method has been to "rehandle" and prize into hogsheads in shipping condition before offering for sale from samples drawn for the purpose, generally by the association's own samplers and not by the regular board of trade samplers. The combined effect of this pooling movement, supplemented by a considerable amount of direct country buying, was still further to reduce the proportion of tobacco passing through the regular trade channels, although much of the pooled holdings represented tobacco that ordinarily would have been bought in the country.

The receipts of the western markets, which in 1889 were approximately 300,000 hogsheads, representing, as we have seen, practically the entire production in the previous year, fell in 1909, notwithstanding a larger aggregate production, to but slightly more than one-third of this number, 108,389 hogsheads, which was about one-third of the total production of the export and manufacturing types produced in the West.

BUYING AT THE FARM FOR LOOSE DELIVERY.

These selling organizations have, for obvious reasons, generally had their greatest strength in the sections producing the best tobacco and in some cases have controlled nearly all the tobacco produced in those neighborhoods. This has naturally led to a limitation of the amount of country buying, particularly by the method of riding through the country from farm to farm and purchasing the tobacco at the barn by grades or at a round price, according to whether the tobacco was already stripped and assorted or not. This method of buying was general throughout all the tobacco regions of the West until the pooling movement gathered force, but more recently it has been largely abandoned.

LOOSE-LEAF TOBACCO SALES FROM THE WAGON ON THE STREET.

In the eastern tobacco districts practically all the tobacco is sold in piles on the warehouse floor at auction, each pile representing a grade as assorted by the grower. In this system the buyers bid on and purchase only such piles as they have use for, either prospective or immediate.

In the West also a large proportion of the tobacco produced is sold by farmers in a loose condition, but for the most part on a materially different plan.

The common practice at present in the dark-tobacco districts with such tobacco as is not pooled is for the farmer to grade his tobacco like the Virginia grower, load it on the wagon loose, and haul it to some convenient receiving point. Instead, however, of unloading it at an auction warehouse, he bargains privately in the street with the buyers without official inspection or sampling or selling charges of any kind. In this method of selling, the offers of the buyers may constitute a species of bidding, but the formal auctioneering of the load is not customary. After the sale is arranged the grower delivers the load directly to the leaf-tobacco warehouse of the purchaser. A view taken in the Mayfield market of loads of tobacco exhibited for sale in this manner is shown in figure 4.

Of course, the tobacco may have been bargained for by contract before it left the barn, but in any case the sale and delivery are consummated in the same individual and private way.

Purchasing tobacco in crop lots in this way naturally and frequently involves the purchase of grades not desired or useful to the purchasers. In reassorting and matching up the grades received from different growers these undesired grades must be segregated and disposed of to some other purchaser, either directly and privately or through consignment to some warehouse or commission merchant for public or private offering through the regular channels of the trade after inspection and sampling.

Fig. 4.—Loaded wagons, showing the common method of delivering loose tobacco, either for private or auction sale, in nearly all export and manufacturing tobacco districts.

In the Green River and Henderson districts most of the pooled tobacco is delivered loose to the purchasers at the various receiving points of the districts. An association grader is stationed at each designated receiving point and declares the grade and consequently the price which the buyers must pay. In these districts until recently much of the tobacco has been stemmed by dealers before shipment, and heavy prizing would be a disadvantage. Tobacco intended for stemming is usually tied in much larger "hands" than other types of dark tobacco. In both the Green River and Henderson districts, however, a portion of the pooled holdings has been "rehandled" (dried to proper keeping order and more or less re-sorted and matched into uniform grades) and prized into hogsheads in shipping condition.

THE OWENSBORO LOOSE-LEAF TOBACCO AUCTION SYSTEM.

For a number of years there has been in operation at Owensboro, and also for a short time at Henderson, a warehouse for the sale of loose tobacco at auction by sample. In this method of procedure the farmer drives up to the warehouse, the inspector draws a sample of each grade, usually three (leaf, lugs, and trash), and takes these samples into the sales warehouse, where they are displayed on a table, and at an appointed time the load is sold at auction to the highest bidder. The grower then drives to the leaf warehouse of the purchaser and delivers the tobacco. The charge of the auction warehouse for this service is $1 a load.

LOOSE-LEAF TOBACCO AUCTION MARKETS IN THE WEST.

The method of selling loose tobacco at auction from samples drawn from the load as it remains on the wagon in the street, as practiced at Owensboro, is, of course, quite different from the Virginia auction method of selling the loose leaf, when the entire bulk of tobacco is piled on the floor and sold directly after an examination of the pile as a whole as the sale proceeds, without the necessity of special official inspection and sampling.

The Virginia method, while seemingly the more nearly perfect, involves greater expense, as well as some obvious advantages. The tobacco must be unloaded from the wagon and piled on the warehouse floor before the sale and again reloaded and transported on the wagons of the buyer. Furthermore, a large expanse of warehouse space is necessary for piling such large quantities of tobacco as often come to market in a single day after a good stripping season has intervened. In nearly all sections farmers handle tobacco, i. e., strip, assort, tie, and prepare for market in a natural, or damp, season. It happens, therefore, in the marketing of tobacco that farmers are nearly all marketing somewhat simultaneously at irregular intervals, following damp spells of weather during the winter and spring months. Thus, hundreds of loads of tobacco are often brought to market each day for several successive days in many of the larger markets, and it is necessary to have extensive floor space to handle the tobacco offered.

During the past decade, however, a number of trials of the loose-leaf tobacco auction system upon approximately the Virginia plan have been made in the West.

In December, 1901, a loose-leaf tobacco auction-sales warehouse, based on the Virginia plan, was established at Clarksville, Tenn., and in December, 1902, similar loose-leaf sales were inaugurated at Hopkinsville, Ky. Again, two years later, in January, 1905, the experiment of loose-leaf auction sales in the Burley district was initiated at Lexington. The success of this system of selling in the West has been greatest in the Burley district, and since 1908 the loose markets have rapidly multiplied in that section. extending also into the upper Green River section of the dark one-sucker district, with large sales warehouses at Glasgow and Bowling Green. In figure 5 a scene in the warehouse district of Lexington, Ky., is presented, showing an accumulation of wagons waiting for a chance to unload, the warehouse floor space being insufficient to hold all the tobacco at a single sale.

The selling charges in the western loose-leaf tobacco markets are on the average somewhat lower than in the Virginia and North Carolina markets. Instead of three items of charge, only two are made. The so-called auction fee is omitted. The charges at Lexington, with which those of the other western markets are substantially uniform, are 15 cents per hundredweight and 2 per cent commission.

Fig. 5.—Wagons waiting to unload during a congestion in the loose-leaf tobacco market, Lexington, Ky.

The warehouses. particularly as established at Lexington, are fully equal in size and construction to any that can be found in the East. Some of them have concrete floors and walls, and the immense roof expanse is often of steel trusses and the framework of real architectural interest.

THE LEXINGTON MARKET.

Lexington, Ky., has already become one of the great loose-leaf tobacco auction markets of the country. In the season 1909–10, selling the 1909 crop, the sales of leaf at Lexington in these auction houses were fully 20,000,000 pounds. Only Danville, Va., and Winston, N. C. exceeded Lexington in the volume of such loose sales. Many members of the trade confidently predict that it will be only a few years before Lexington will outdistance all markets, even including Danville, and become the greatest loose-leaf tobacco auction market of the country.[1] The railroads have modified their freight charges so that tobacco can be transported loose for a considerable distance at reasonable rates.

FIG. 6.—Exterior of a loose-leaf tobacco auction warehouse, Lexington, Ky.

In the sales season of 1909–10 six large warehouses were in operation at Lexington and a seventh was constructed for 1910–11. A number of dealers and commission merchants have established headquarters there. The trade has been organized under the title of the

FIG. 7.—Interior of a large tobacco auction warehouse, Lexington, Ky., during an off season.

Lexington Tobacco Association for its better control and supervision in the best interests of all. Figure 6 shows an exterior and figure 7 an interior view of one of these large loose-leaf tobacco warehouses at Lexington.

[1] See footnote on p. 20.

SUCCESS OF THE LOOSE-LEAF TOBACCO AUCTION SYSTEM.

The loose-leaf tobacco auction system of first-hand sales now seems to have a firm foothold in the West, particularly in the Burley district. Such markets, on the Virginia plan, are already in operation or in contemplation in Lexington, Danville, Maysville, Frankfort, Springfield, Carrollton, Glasgow, Bowling Green, Hopkinsville, and Paducah, Ky.; Madison, Ind.; Clarksville, Tenn.; Huntington, W. Va.; St. Joseph, Mo., and elsewhere.

DECLINE OF AUCTION SALES OF TOBACCO IN HOGSHEADS.

Accompanying the development of very unusual conditions, which have beset the old-established trade methods of the western tobacco markets, assisted no doubt by generally advancing prices, there has been a marked tendency to handle an increasing proportion of the inspected hogshead tobacco by private sale rather than at public auction. All of the regular markets have exhibited this tendency strongly and, indeed, with the exception of Louisville, Cincinnati, and Clarksville, the public auction sales of hogshead tobacco may be said to be almost a thing of the past in the markets of the West, just as they have already passed out in the eastern markets. Even at Clarksville hogshead tobacco auction sales have become almost nominal in volume, and in Cincinnati for 1909 such public offerings amounted to only 5,381 hogsheads, as against 70,355 hogsheads so offered in 1906. In addition, however, to the 5,381 hogsheads sold publicly in 1909 there were 12,821 disposed of privately.

In Louisville the same tendency is also distinctly shown. The annual trade reports issued by the Louisville Leaf-Tobacco Exchange are not published in such form as to make possible a distinction between the auction and private sales. Weekly reports are issued in this form, however, and this tendency for an increasing preponderance of private rather than auction sales is distinctly shown in these reports. It is a matter of common knowledge also that for many years nearly all the Green River tobacco handled in Louisville has been sold privately.

LOUISVILLE AND CINCINNATI THE ONLY DISTINCTIVE HOGSHEAD INSPECTION AND AUCTION MARKETS.

To sum up the main features concerning the present position of the western tobacco markets, we may note that Louisville and Cincinnati are the only remaining distinctive hogshead inspection and auction markets.

Except the relatively unimportant cigar-leaf breaks, Cincinnati deals only in Burley leaf. Its final position as a leaf market, therefore, seems to hinge largely upon two main factors, the continuance of the pooling movement in the Burley district and the permanent success and spread of the loose-leaf tobacco auction system in the country market centers.

Louisville is the one great central market of the West where all types of western leaf, Burley, dark-fired, Green River, one-sucker tobacco, etc., are dealt in. The changed market conditions as they exist have seemed to accentuate its distinctiveness as the one great public hogshead market of the West of the clearing-house type. Figures 8, 9, and 10 show familiar market scenes in Louisville, illustrating characteristic stages in conducting the inspection and auction sale of tobacco in the West.

FIG. 8.—Breaking a hogshead and drawing a sample at a tobacco inspection, Louisville, Ky.

Neither Louisville nor Cincinnati is situated directly in the important tobacco-producing territory, and the trade of these centers is entirely in hogshead tobacco.

In every other western market a large part of the trade consists in loose-leaf tobacco purchased directly from farmers, after which the leaf is rehandled and prized into hogsheads, and a portion of it may then appear among the hogshead receipts of the same or some other market for storage, inspection, and sale.

CLARKSVILLE, TENN., THE MOST IMPORTANT DARK-TOBACCO MARKET.

Clarksville, Tenn., continues to be by far the largest distinctive dark-tobacco market, and in total volume of leaf handled, counting none twice, it probably now ranks ahead of Cincinnati and next to Louisville. More than 30,000,000 pounds of leaf are handled there annually, a large part of which is received loose. Although its hogshead auction-break sales have shrunk to insignificant proportions, it remains, nevertheless, a very important hogshead storage and inspection market. Sales however, are generally conducted privately. From time immemorial Clarksville has been justly proud of the distinction, both at home and abroad, of representing the highest grades of dark leaf produced in the West. This high-grade tobacco, however, was drawn largely from the adjoining county of Robertson, Tenn., and the southern part of Logan County, Ky., particularly along the course of the Red River and its tributaries. In this section is produced the largest percentage of really fine dark leaf to be found anywhere in the West, such as the fine, rich tobacco suitable for wrappers for plug, the fine leafy Austrian and Italian cigar-wrapper grades, and the fat, rich but fine German and English spinning leaf.

FIG. 9.—An auction-break sale, Louisville, Ky. The sale is based on the official sample shown on the top of each hogshead of tobacco.

THE SPRINGFIELD, TENN., MARKET.

Prior to 1904 Springfield, the county seat and natural trade center of Robertson County, Tenn., was merely a receiving and rehandling point, and the tobacco received after being prized was shipped elsewhere, principally to Clarksville, for inspection and sale.

In 1904, however, the Planters' Protective Association established at Springfield one of its more important inspection and selling agencies, and Springfield to-day has become a really important inspection market, ranking probably next to Clarksville in the volume of its hogshead trade, and because of the fine grade of leaf produced in the territory which it serves outranks Clarksville and all other dark-tobacco markets of the West in the average quality of its offering.

Over 5,000 hogsheads of the 1904 crop and about 14,000 hogsheads of the 1908 crop were handled at Springfield. Four large storage warehouses have been constructed there for the accommodation of the trade.

FIG. 10.—Recoopering hogsheads of tobacco after inspection, sampling, and sale, Louisville, Ky.

THE PADUCAH, KY., MARKET.

Paducah is an important western dark-tobacco market, standing, historically at least, as the leading market center for the Paducah or western district. In the total quantity of leaf handled, however, it is generally equaled and sometimes surpassed by the Mayfield, Ky., market, in the same district but in the adjoining county of Graves. Paducah, however, remains more of a center for hogshead tobacco shipped from all points of the district, while Mayfield is more distinctly a loose-leaf tobacco market and its receipts are more local in origin. The total receipts of tobacco of all classes at Paducah amounted in the trade year 1909–10 to about 17,000,000 pounds, of which about 10,000,000 pounds came in loose and 7,000,000 pounds in hogsheads.

OTHER IMPORTANT MARKET POINTS.

As already noted, Mayfield and Hopkinsville, although handling a large quantity of tobacco, are now principally loose-leaf tobacco receiving and rehandling centers rather than important hogshead markets, as are also Owensboro and Henderson.

Next to Clarksville, but some distance behind in total amount of tobacco trade, there are six market centers, all handling about the same gross volume of business. They are Owensboro, Henderson, Springfield, Hopkinsville, Paducah, and Mayfield, each handling approximately 15,000,000 to 20,000,000 pounds or more of leaf annually. There are no other western dark-tobacco markets that handle as much as 10,000,000 pounds of leaf yearly, although there are many small receiving and handling points doing a strictly local business, scattered here and there throughout the entire producing territory. Some of these minor local centers, however, do quite a large business in the aggregate, amounting annually in many cases to 5,000,000 pounds or more of leaf.

IMPORTANT RECEIVING POINTS IN THE ONE-SUCKER DISTRICT.

Perhaps special mention should be made of the leading receiving centers of the one-sucker territory in Kentucky. Glasgow, Bowling Green, and Scottsville are the principal points, each handling, respectively, about 6,000,000, 4,000,000, and 3,000,000 pounds of tobacco annually. Practically all the leaf tobacco however, of this one-sucker district, except such as goes directly into the manufacturer's or exporter's hands, or such as is controlled and sold by the growers' pooling organization, is sent to Louisville for inspection and sale.

TRADE ORGANIZATION AND MARKET REGULATION IN THE WESTERN MARKETS.

When the first inspections were established in the western markets the method followed in nearly all cases was to have the inspectors appointed by some public agency, as, for example, the city council, the city or county courts, the mayor of the city, the governor of the State, or, as in Cincinnati, by the direct vote of the people of the city. Statutes were enacted for regulating the trade in the interests of fair dealing between the members of the trade and farmers, particularly for the purpose of giving the inspection a better standing abroad than any system of private inspection and regulation would have done at that time. The laws of Virginia and Maryland naturally served as the basis of the earlier warehouse and inspection laws of the West.

In the decade 1850 to 1860 nearly all the larger inspection markets had begun to feel the need of trade organization, and such organizations were established in the more important of these markets during that period.

As in Virginia, it was not many years, particularly in that strained period of readjustment of the Government in the States of the South immediately succeeding the close of the Civil War, before the public appointment of tobacco inspectors became such a mere political plum, with but little regard to the fitness of the appointees, as to render the system exceedingly unsatisfactory to the trade both at home and abroad.

In the early seventies this dissatisfaction with the State inspection system had reached such a point as to result generally in the taking over of this function by the organized trade, with sufficient changes in the existing statutes where it was necessary to render this action legal. Under this board of trade system of inspection the general plan was to elect the inspectors by the vote of the individual members of the entire organized body. This trade organization now became in effect the guarantor of the integrity of the inspection and sample, and its inspectors were placed under bond for the faithful performance of their duties.

This new system of the semipublic nature in turn has sometimes fallen into disfavor, particularly among growers, who have claimed they were not given sufficient consideration and guarantee of fair treatment by the board of trade system, and on several occasions, particularly after periods of low prices, there have been vigorous movements in nearly every interested State to reestablish official State inspections on a compulsory basis. Thus far, however, the organized trade has possessed sufficient influence to prevent the enactment of such measures.

The general plan of organization and the rules under which these chartered tobacco-trade bodies in the western markets operate are quite uniform in their essential points. A brief general description of the Louisville trade organization, or tobacco exchange, as it is called, perhaps will be sufficient to give a general idea of the way in which the tobacco trade is conducted in the western markets.

ORGANIZATION OF THE LOUISVILLE TOBACCO TRADE.

Membership in the Louisville Leaf-Tobacco Exchange is obtained by vote of the members and by purchasing a share of stock in the exchange and the payment of annual dues.

The members are divided into buyers and warehousemen, the class of membership being designated on the certificate of stock. An interesting point in the organization of the tobacco trade of Louisville and most other western markets is that on nearly every question the buying interests and the warehouse interests, that is, the selling interests, have equal votes. Thus, the executive committee has three members who are warehousemen and three who are buyers. The members of this committee are elected, in turn, by a two-thirds vote of the warehousemen and a two-thirds vote of the buyers. Similarly, the committee on arbitration and the committee on by-laws consist of six members each, three of whom are buyers and three warehousemen, but in the first-named committee the buyers elect the warehouse members and the warehousemen elect the buyers on the committee. In the committee on by-laws each side elects its own members. The quotations committee, consisting of two buyers and two warehousemen, with the secretary of the exchange, is appointed by the president.

On the first Monday in December of each year a joint committee on inspection is elected, as follows: Each warehouse member is represented by one member of his firm and a corresponding number of buyers are elected by the buyers. On the second Monday in December this joint committee elects an inspector to serve for four years to succeed one of the four inspectors whose term will expire that year. For any neglect of duty this joint committee of inspections may remove an inspector at any time by a majority vote and elect his successor.

The important committee on reclamations consists of eight members, four of whom are buyers and four warehousemen. The warehouse members of this committee are elected by the warehousemen and the buyers are elected by the buyers. Two warehousemen and one buyer constitute a quorum on this committee, and when the buyer and one warehouseman shall agree in adjudging a case coming before it on any given day their decision is final. If these two fail to agree, however, they refer the question to the president of the exchange, whose decision is final.

Claims on tobacco shipped to points in the United States are not considered after six months from the date of inspection nor those to foreign countries after seven months from the date of inspection. The inspectors and their sureties are jointly liable for such reclamation as the committee may allow. In making a claim the claimant deposits $1 with the committee. If the claim is allowed, the inspectors and owners of the tobacco are assessed $1 in excess of the amount of damage allowed to be reimbursed to the claimant. To substantiate reclamations the original sample, properly sealed and tied, together with a sworn resample of the same hogshead, must be returned to Louisville. If the hogshead complained of is in Louisville, the inspector must be notified before the tobacco has been taken from the hogshead and he must promptly examine such hogshead.

Inspectors have the right to prosecute claims against the sellers of any tobacco against which damage has been assessed.

In sampling tobaccos no uniform method is prescribed, but the hogshead is generally broken in at least three places and a sample not to exceed 10 pounds in weight is obtained. Usually about 12 or 16 hands are taken. The layers of the sample are tied in the same order in which the tobacco stands in the hogshead. When a hogshead of tobacco is damaged in any way, the character of such damage is marked on the tag in ink. On the tag there also appears in ink the packer's name, the number of the hogshead from which the sample was taken, the date of inspection, and the gross and net weight. The tag and sample are tied with strong cord and sealed with wax, using the registered seal of the exchange.

In selling tobacco at auction the hogshead must be inspected on the day of sale and the sample placed on top of the open bulk of the hogshead in full view of the buyers.

The tobacco is weighed before sampling and after sale, and on the Louisville market the warehouseman collects from the buyer by first weight and settles with the seller at 10 pounds less than this weight. A gain on the part of the warehousemen of over 5 pounds in sampling, however, is not allowed. The buyer, of course, gets the sample.

In selling tobacco at auction the time consumed on each hogshead must not exceed one minute. The minimum bid on tobacco up to $6 is 5 cents per 100 pounds; up to $10, 10 cents per 100 pounds, except to round up; and over $10, 25 cents per 100 pounds; and over $20, 50 cents per 100 pounds.

An owner may reject the sale of any hogshead of tobacco by serving notice on the buyer of such rejection within two hours after the closing of the sale, but the buyer in turn may proceed to reject an equal number of hogsheads within two hours after the receipt of such notice, unless the owner accepts or rejects each hogshead at the time it is knocked out.

Warehousemen must furnish good storage for the tobacco, the president of the exchange appointing two members, a buyer and a warehouseman, who, acting unanimously, have authority to condemn premises unsuitable for tobacco storage. After a sale the warehousemen must recooper each hogshead into first-class shipping condition.

WAREHOUSE FEES.

The fees collected by the warehousemen are not strictly uniform in all the markets. The usual charges, however, are either $1.50 or $2 per hogshead and 1 per cent commission to the seller and an outage fee of $2 per hogshead to the buyer. The Louisville rate to the seller is $1.50 per hogshead and 1 per cent commission. Storage is free to the shipper for 4 months and to the buyer for 15 days. After four months the seller is charged 40 cents per month or fraction thereof for storage, in addition, of course, to such general charges as insurance, freight, drayage, cooperage, etc. If the sale of a hogshead of tobacco is rejected, $1.50 is charged unless the hogshead is removed, when the rejection fee is $2.50 per hogshead and storage is charged from the date of receipt.

Reduced selling fees are charged buyers who are members of the exchange on tobacco purchased from members of the exchange. Thus buyers may resell tobacco at Louisville within 30 days for $1 per hogshead or for $1.50 per hogshead after 30 days.

CONSOLIDATION OF WAREHOUSE INTERESTS.

At the present time there are in Louisville 12 warehouses in active operation and a number of others once active but now used only for storage purposes. Of these 12 actively operated 8 are under joint ownership, though operated separately under the control of the Louisville Warehouse Co. One of these 12 is not even a member of the exchange.

In Cincinnati likewise, although there are five houses in actual operation, four of them are consolidated under the ownership of the Cincinnati Warehouse Co. In 1890, in fact, a movement was reported looking toward the combination of both Louisville and Cincinnati under single ownership and control.

PRIVATE WAREHOUSE INSPECTIONS.

In the last few years, owing perhaps to the general disturbance within the regular channels due to changing conditions, the trade bodies in some instances have been allowed to fall somewhat into decay, and except as a matter of custom exert very little influence over the trade.

In some of the markets the inspectors, instead of being appointed and controlled by the trade organizations, are simply employees of the warehouse company, and the warehouse, acting without outside regulation by the organized trade, adjusts its own claims for reclamation.

In Tennessee the law specifically provides for the appointment of warehousemen as inspectors under bond to the State.

SUMMARY OF THE RECEIPTS AT THE IMPORTANT HOGSHEAD-TOBACCO MARKETS FOR 10 YEARS.

In the case of the Mayfield, Paducah, Hopkinsville, and Clarksville markets the figures in Table XI of the receipts of hogshead tobacco are not to be taken as indicating anything like their total general leaf and brokerage trade. A considerable proportion of the trade of these markets, and the larger portion in some cases, is in loose tobacco bought by manufacturers or exporters direct from the farmers and does not appear in the regular market receipts of any market, as already explained. Some of the figures in Table XI were obtained from the compilation made by the Western Tobacco Journal.

TABLE XI.—*Receipts of leaf tobacco in the principal hogshead markets of the United States from 1900 to 1909, inclusive.*

Market.	1900	1901	1902	1903	1904	1905	1906	1907	1908	1909
	Hogsheads.	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*
Louisville	106,827	123,279	124,213	80,051	84,104	100,335	105,973	107,525	97,099	75,190
Cincinnati	56,070	60,318	51,638	52,093	21,022	45,419	55,380	37,317	38,103	11,702
Clarksville	20,501	22,322	21,791	20,843	21,220	22,980	9,847	11,533	8,559	17,322
Hopkinsville	14,165	12,465	11,975	11,350	14,930	9,715	5,450	4,655	6,585	1,875
Springfield	....	....	....	....	....	4,500	7,500	8,700	9,000	14,300
Paducah	9,987	7,273	8,697	11,000	8,690	5,996	5,381	6,311	4,011	1,100
Mayfield	12,518	9,780	10,594	7,995	10,770	8,039	5,481	4,569	3,100	1,200
Richmond	27,663	21,522	20,096	21,150	17,487	23,330	20,404	19,636	21,640	25,113
Baltimore	38,023	35,881	39,480	40,051	40,734	38,563	37,055	25,594	28,189	28,883
Total	285,754	292,840	288,484	244,533	218,957	258,877	252,471	225,840	216,286	176,785

TABLE XII.—*Inventory of the stock of tobacco on hand on Jan. 1 in the principal hogshead-tobacco markets for each of the 10 years from Jan. 1, 1901, to Jan. 1, 1910, inclusive.*

Market.	1901	1902	1903	1904	1905	1906	1907	1908	1909	1910
	Hogsheads.	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*
Louisville	13,031	15,627	12,266	11,796	18,001	16,215	17,906	27,078	23,978	15,737
Cincinnati	9,391	12,287	10,084	8,781	6,238	9,080	8,955	11,278	14,234	4,312
Clarksville	2,662	531	1,533	3,348	1,049	3,560	579	329	3,780	740
Hopkinsville	3,124	808	381	2,435	1,392	1,671	374	210	2,000	100
Springfield [1]	--------	--------	--------	--------	--------	--------	--------	--------	--------	--------
Paducah	824	221	1,211	2,149	976	517	251	376	6,500	225
Mayfield	1,597	222	1,024	825	291	727	144	--------	1,400	423
Richmond	12,020	9,839	9,526	9,326	8,230	10,659	13,065	12,506	13,799	17,181
Baltimore	5,425	5,521	4,989	7,159	5,240	4,925	6,111	3,300	3,891	3,065
Total	58,074	47,056	41,414	45,819	41,417	47,354	47,385	55,077	69,582	41,783

[1] No record.

The total stocks on hand on these principal markets have averaged, according to the figures in Table XII, not far from 50,000 hogsheads of tobacco on January 1 of each of the 10 years specified. This, of course, does not include that stored privately in dealers' hands or stored privately in the hands of manufacturers. The latter figure would reach a very large total, as it is customary for manufacturers to carry from one to two years' stocks, and many of the larger firms own private storage plants.

These stocks held privately by manufacturers amount probably to at least 400,000,000 pounds at any given time, and often, no doubt, to very much more than this, against something like 60,000,000 to 80,000,000 pounds generally held in the public warehouses of the regular markets.

The Louisville tobacco trade is well organized, with a salaried secretary, and publishes very complete annual statistics of the transactions on that market, including the classification of sales. Since Louisville is the largest and most important hogshead-tobacco market in the country, the Louisville leaf-trade statistics are given in Table XIII for each of the past 10 years. They throw considerable light on certain trade features not before mentioned, such as differentiation between offerings, sales, rejections, reinspections, etc.

TABLE XIII.—*Statistics of the Louisville market for 10 years, from the report of the secretary of the Louisville Tobacco Exchange.*

Item.	1900	1901	1902	1903	1904	1905	1906	1907	1908	1909
	Hogsheads.	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*	*Hogsheads.*
Offerings	145,339	156,788	164,696	104,241	107,007	140,112	143,784	125,673	118,612	95,967
Rejections	29,123	28,851	32,159	17,989	12,967	18,831	14,986	13,559	9,341	8,770
Actual sales	116,216	127,937	132,537	86,252	94,040	121,281	128,798	112,114	109,271	87,197
Original inspection	105,046	130,693	112,966	87,656	82,875	96,200	118,014	98,290	71,289	66,494
Receipts	106,827	123,279	124,213	80,051	84,104	100,335	105,973	107,525	97,099	75,190
Total stock Dec. 31	13,031	15,627	12,266	11,796	18,001	16,215	17,906	27,078	23,978	15,737
Unsold stock Dec. 31	9,612	12,733	9,920	10,622	14,835	10,717	12,580	22,306	14,898	9,446
Unsold Burley	3,803	9,160	7,832	1,869	1,158	2,096	3,688	9,767	12,676	2,233
Unsold dark	5,809	3,573	2,063	2,353	6,934	2,296	1,870	1,243	1,915	4,484
Unsold Green River	....	....	25	6,400	6,743	6,325	7,022	11,296	307	2,729
Classification of sales:										
Burley	99,969	116,094	120,733	70,279	43,554	86,512	90,345	71,484	77,547	51,752
Dark	45,370	40,694	43,963	33,962	63,453	53,600	53,439	54,189	41,065	44,215
Rejections:										
Burley	21,375	21,984	24,132	11,726	4,963	13,326	12,075	10,344	5,481	3,609
Dark	7,748	6,867	8,027	6,263	8,004	5,505	2,911	3,215	3,860	5,161

There is a small error in the foregoing figures, since they fail to show the full volume of the tobacco-leaf trade of Louisville. Of the 12 warehouses in operation in the city in recent years, one is not a member of the exchange and its transactions are not included in the report.

METHOD OF ESTIMATING THE AVERAGE ANNUAL PRODUCTION OF TOBACCO IN THE UNITED STATES BASED ON STATISTICS OF THE TREASURY DEPARTMENT.

On their face, the fully itemized records of the offices of Internal Revenue and Customs of the Treasury Department, specifying with substantial accuracy the quantity of raw tobacco imported and exported and the quantity that is consumed in manufacture, should furnish the data for estimating the actual farm production of tobacco in the United States on a very reliable basis.

It would appear as though a statement of the quantity of raw tobacco used in manufacture plus the exports and less the imports of raw tobacco for a year or for a series of years would be equal to the farm production of tobacco in the country.

There are, however, a number of important sources of error to which production figures derived in this way would be subject, in so far as they purported to serve as a check on the actual weights of barn-cured tobacco reported by farmers.

The factors of importance producing this error are the following:

(1) The shrinkage to which leaf tobacco is subject while it is in the hands of dealers or manufacturers because of the redrying to put it in good keeping order and while it remains in storage. This shrinkage in weight from the time the unstemmed leaf tobacco leaves the farmers' hands till it enters the manufactory or is cleared for export ranges from 5 to 20 per cent, according to the type of tobacco and its moisture content at the time of purchase from the farmer.

(2) In the case of leaf tobacco which is stemmed before it is weighed in the manufacturing plant or is declared for export, there is an additional loss due to the weight of stem removed. The loss in weight to which stemmed tobacco has been subjected, including the drying as well as the stemming, is estimated to be from 28 to 35 per cent.

(3) There is also considerable tobacco not recorded, which is consumed in its natural state locally by producers, dealers, farm hands, laborers, and others, destroyed by fire, etc. This source of error all told is believed to amount to about 1 per cent of the total production, and, while small in respect to the whole, it is of considerable importance in the aggregate.

The total of these three sources of error makes so large an error as to render an uncorrected compilation of the statistics of the Treasury Department of but little service in estimating the farm production. The question which at once presents itself is to what extent and with what degree of accuracy can corrections be introduced into the recorded figures to make them of such service. As a matter of fact, this seems comparatively easy to do with approximate accuracy and to such a degree, the writer believes, as to render them the most accurate basis for estimating the average annual crop of tobacco produced in the United States for any period of years, preferably not less than three.

The shrinkage of leaf tobacco in redrying or stemming and in storage after it leaves the farmers' hands is a matter of regular estimate by leaf dealers and manufacturers who make direct purchases of leaf, and these estimates are accurate to a very small percentage of error where a sufficient number of pounds for each of the different types produced in the country are involved. The actual figures, of course, vary somewhat for the different types of tobacco, but a general average correction applicable to all types would doubtless give an average result of sufficient accuracy for general purposes.

The most important missing item in the figures of the Treasury Department is the fact that exports of stemmed and unstemmed leaf are not reported separately, but are classified together under one head. It so happens, however, that a very large proportion of the stemmed tobacco exported from this country goes to the United Kingdom, and the trade reports of the United Kingdom separate the stemmed leaf from the unstemmed, so that an approximately correct statement is available as to the quantity of stemmed leaf tobacco which is included in our export figures.

Production estimates based on compilation of these figures of the Treasury Department, with proper corrections, of course, would not give much of an idea of the size of the crop for any particular year. The exports of leaf tobacco are generally made during the year succeeding its production, but stocks of tobacco for domestic manufacture are generally held from one to two years in order that the tobacco may improve and mellow with age before it is manufactured. To be of value the compilation, as suggested, must represent the average of figures covering three to five years, and as such they should give an accurate average estimate of the rate of production for a period of three to four years, dating back one year from those figures from which the compilations were made.

Such a compilation, based on the statistics of the Treasury Department for the three years 1907 to 1909, inclusive, is presented below.

In considering the quantity of tobacco consumed in producing the so-called manufactured forms of tobacco, namely, plug, twist, fine-cut smoking tobacco, snuff, and cigarettes, and the quantity of exported leaf, we have to deal almost entirely with the so-called export and manufacturing types. We will assume that, as a general average, these types shrink in redrying and handling about 10 per cent if unstemmed and about 30 per cent if stemmed. In the cigar types the shrinkage is probably somewhat greater and will be calculated at 15 per cent for the unstemmed leaf. The correction to an approximate equivalent of unstemmed but dry leaf is made by the office of Internal Revenue in the case of the stemmed leaf used in the manufacture of cigars and cigarettes, so this factor does not need to be considered here.

We will first consider the leaf used in producing manufactured tobacco and cigarettes and for export. In the 3-year period, 1907 to 1909, inclusive, there were consumed in manufactured tobacco and cigarettes 283,550,157 pounds of stemmed leaf, 631,736,223 pounds of unstemmed leaf, and 99,785,876 pounds of scrap. The exports of domestic leaf for the same period amounted to 957,080,408 pounds. The import statistics of Great Britain show that at least 147,145,070 pounds of this latter total was stemmed leaf. We also export some little stemmed leaf to countries other than Great Britain. It would be well within the facts, therefore, to assume that not less than 150,-000,000 pounds of stemmed leaf were exported during the 3-year period. Combining this with the stemmed leaf reported by the office of Internal Revenue, we have a total of 433,550,157 pounds of stemmed leaf of the export and manufacturing type to which the 30 per cent correction should be applied. We find this to have been equivalent to 619,357,367 pounds of unstemmed leaf, based on farmers' weights.

The scrap tobacco reported by the office of Internal Revenue consisted largely of cigar clippings and other material, much of which perhaps had already been corrected for loss of stem. For our purposes, therefore, it may be classed with the unstemmed leaf, to be corrected for loss simply for shrinkage from drying and age. Making this correction of 10 per cent, we find that this total of 1,689,328,514 pounds of unstemmed leaf of the export and manufacturing types used in domestic manufacture and exported to have been equivalent to 1,877,031,682 pounds of tobacco at farm weights. The farm weight of the combined total, stemmed and unstemmed, manufactured, and exported, was, therefore, during the 3-year period about 2,496,389,049 pounds.

The quantity of unstemmed leaf consumed in the manufacture of large and small cigars during the three years amounted to 414,636,193 pounds. Allowing for a shrinkage of 15 per cent this was equivalent to 490,160,227 pounds, farmers' weight.

The export factories under the supervision of the Customs Service consumed 25,462,093 pounds of leaf, not included in the foregoing estimates. It is uncertain how much of this was stemmed weight, but as the quantity involved is small, it is not very material. It is, however, subject to the 10 per cent correction for shrinkage in drying. Making this correction we find this to have been equivalent to 28,291,214 pounds.

Combining these three totals we have a grand total of leaf consumed in manufacture and export, reduced to farmers' weights of unstemmed leaf, as follows:

	Pounds.
Export and manufacturing types exported and reported by office of Internal Revenue	2,496,389,049
Consumed in cigars	490,160,227
Consumed in export factories reported by the customs office	28,291,214
Total for three years	3,014,840,490

From this total must now be deducted the quantity of leaf imported for consumption during the 3-year period, including that from Porto Rico.[1] This imported leaf, however, has been included in the tobacco to which the foregoing corrections to a farm-weight basis were applied. In subtracting the imports as recorded by the Cus-

[1] The deduction of imported leaf from Porto Rico was for fiscal-year periods; all other figures were for calendar-year periods.

toms Service, therefore, the figures must also first be raised to a farm-weight basis. About four-fifths of this imported leaf was of the cigar type and the remainder of the Turkish type. A compromise figure of, say, 14 per cent instead of either 10 or 15 per cent, as used in correcting the manufacturing types and cigar types, would give only a small error either way.

The imports for consumption, including those from Porto Rico, during the 3-year period amounted to 119,062,219 pounds. Making the correction for the estimated shrinkage of 14 per cent, we find the computed farm weight of this imported leaf to be 138,569,051 pounds.

Subtracting this quantity from the total consumed and exported, as shown, we have a total of 2,876,271,439 pounds.

Adding to this the 1 per cent estimated to have been consumed on the farm, destroyed by fire, and otherwise unaccounted for, we have a grand total, corrected to an approximate unstemmed farm-weight basis, of 2,905,324,686 pounds of domestic tobacco consumed in the United States and exported during the three calendar years 1907, 1908, and 1909. Dividing this total by 3, we have 968,441,562 pounds as the indicated average annual production of leaf tobacco in farm weights in the continental United States within the approximate period, say from 1906 to 1908.

These computations have been submitted mostly as an illustration of the method by which the official Treasury records might be utilized in estimating the average annual production of tobacco in the United States on a farm-weight basis. Possibly there are still some important sources of error in the method that have failed to receive proper consideration. The percentage basis upon which the corrections were based, 10 per cent for unstemmed leaf of the export and manufacturing types, 15 per cent for the cigar types, and 30 per cent for the stemmed leaf of the export and manufacturing types, are, as stated, assumed figures of a somewhat arbitrary character.

Issued April 24, 1913.

U. S. DEPARTMENT OF AGRICULTURE.

BUREAU OF PLANT INDUSTRY—BULLETIN NO. 269.

B. T. GALLOWAY, *Chief of Bureau.*

EXPERIMENTS IN WHEAT BREEDING:

EXPERIMENTAL ERROR IN THE NURSERY AND VARIATION IN NITROGEN AND YIELD.

BY

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LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry,
Office of the Chief,
Washington, D. C., *October 16, 1912.*

Sir: I have the honor to transmit herewith and to recommend for publication as Bulletin No. 269 of the series of this Bureau the accompanying paper entitled "Experiments in Wheat Breeding: Experimental Error in the Nursery and Variation in Nitrogen and Yield," by Prof. E. G. Montgomery. This paper contains the results of special experiments in wheat breeding conducted by the Nebraska Agricultural Experiment Station in cooperation with the Office of Cereal Investigations of this Bureau, during the years 1905 to 1910, inclusive. In part, the work is a continuation of that recorded in Bureau of Plant Industry Bulletin No. 78, by Dr. T. L. Lyon, under whose direction the experiments were conducted from 1902 to 1906, inclusive. From 1907 to 1910, inclusive, the work was under the charge of Prof. Montgomery, experimental agronomist of the Nebraska experiment station, who has since become professor of farm crops in the College of Agriculture at Cornell University.

The paper is concerned chiefly with the nature and extent of experimental error in the wheat nursery in connection with breeding experiments in the inheritance of nitrogen content and yield in wheat plants. The standardization of agronomic experiments has been receiving much attention in recent years and is regarded as of fundamental importance in agronomic research. The results contained in this paper are presented as a contribution to this subject as well as to the improvement of wheat.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

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EXPERIMENTS IN WHEAT BREEDING: EXPERIMENTAL ERROR IN THE NURSERY AND VARIATION IN NITROGEN AND YIELD.

I.—EXPERIMENTAL ERROR IN THE NURSERY AND VARIATION IN NITROGEN CONTENT.

INTRODUCTION.

The investigation of the variation of plants of winter wheat in relative nitrogen content when grown under field or nursery conditions was begun by Dr. T. L. Lyon, formerly agronomist of the Nebraska Agricultural Experiment Station, in collaboration with the Bureau of Plant Industry of the United States Department of Agriculture. His results were published as a bulletin of that bureau.[1] Since 1907 the investigation has been continued by the writer and his assistants.[2]

One of the striking features of the data obtained by Dr. Lyon was the variation in nitrogen content of the kernels from different plants of wheat grown under apparently similar conditions. For example, 800 spikes of Turkey wheat were selected and half of each spike analyzed for proteid nitrogen, the lowest having only 1.12 per cent while the highest contained 4.95 per cent.

In 1903, 288 plants which were the progeny from 119 of the spikes analyzing above 3 per cent proteid nitrogen in 1902 were analyzed and found to vary from 1.20 per cent to 5.85 per cent in nitrogen content. In most of the families only a single plant was selected for analysis, but in the remainder two to six plants were selected. Even where all the plants were grown from a single parent the variation was quite as great.

[1] Lyon, T. L. Improving the Quality of Wheat. Bulletin 78, Bureau of Plant Industry. 1905.

[2] The writer wishes to acknowledge with thanks the assistance of a number of men who have contributed to the production of these data. Dr. T. L. Lyon, now of Cornell University, planted the first wheat nursery in 1902 and conducted the work until 1906, being assisted by Prof. Alvin Keyser, now of the Colorado Agricultural Experiment Station. They left an excellent set of records, from which data previous to 1906 have been prepared (Table II). Mr. L. L. Zook, now of the Bureau of Plant Industry, assisted with the work in 1907 and 1908, as did Mr. Erwin Hopt in 1908 and 1909. Prof. T. A. Kiesselbach had charge of the records during the seasons of 1909 and 1910 and has prepared much of the tabular matter for publication. The chemical analysis has been under the direction of Dr. F. J. Alway, who devised a rapid method especially for this work.

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Dr. Lyon noted this variation, as follows:

For instance, the plants numbered 21205 to 21212, all of which come from the same parent, vary from 2.16 to 5.23 per cent in proteid nitrogen content, while plants 69805 and 69806 vary from 5.82 to 1.66 per cent in this constituent.[1]

In addition to the 119 "highs" preserved in 1903, progeny were analyzed also from 20 "mediums" and "lows." When these data were summarized it seemed that there had been some tendency to transmit the character, as shown in Table I.

TABLE I.—*Results of a study of transmission of nitrogen content in wheat kernels in 1902 and 1903.*[2]

Range in percentage of proteid nitrogen.	1902		1903	
	Number of analyses averaged.	Proteid nitrogen in kernels.	Number of analyses averaged.	Proteid nitrogen in kernels.
		Per cent.		*Per cent.*
1 to 3	20	1.85	70	2.59
3 and over	119	3.39	288	2.92

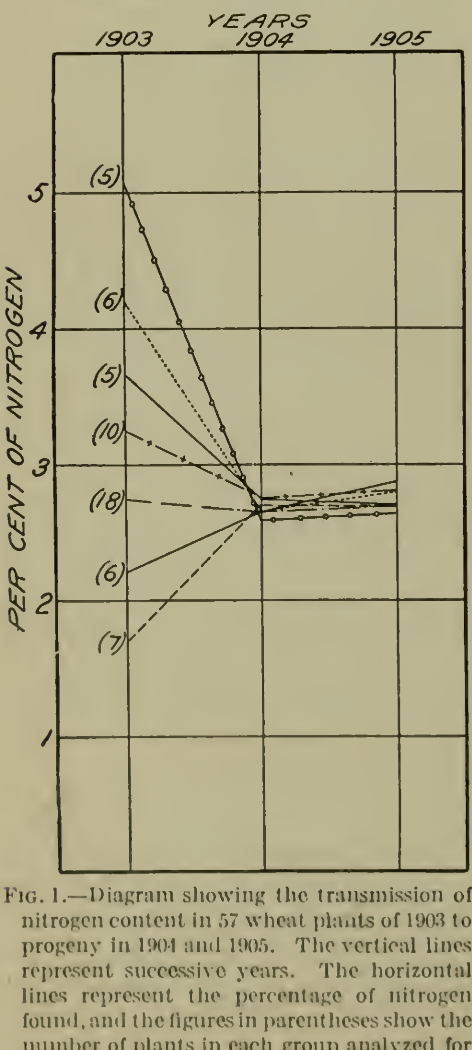

FIG. 1.—Diagram showing the transmission of nitrogen content in 57 wheat plants of 1903 to progeny in 1904 and 1905. The vertical lines represent successive years. The horizontal lines represent the percentage of nitrogen found, and the figures in parentheses show the number of plants in each group analyzed for nitrogen content in 1903.

Summaries of the results obtained in 1904 and 1905 show very little tendency to transmit this character.

In 1906, after four years of selection of extremely high fluctuates, and later, after two more years of selection, by taking a composite sample of all the progeny of a plant it was found that no gain had been made in the nitrogen content of the crop.

In Table II is a summary of data obtained in the years 1903 to 1905 from 57 of the original plants. Figure 1 is a graphic presentation of the same data, in which the horizontal lines represent the percentage of nitrogen, the vertical lines represent successive years, and the figures in parentheses show the numbers of plants in each of the seven groups analyzed for nitrogen content in 1903.

[1] Lyon, T. L. Op. cit., p. 99.
[2] Lyon, T. L. Op. cit., table 26, p. 98.

TABLE II.—*Nitrogen content of 57 wheat plants in 1903 and of their progeny in 1904 and 1905, arranged in groups according to the percentage of nitrogen.*

Record of 57 plants harvested in 1903.					Record of progeny plants in 1904.					Record of progeny plants in 1905.				
	Kernels per plant.		Nitrogen in kernels.			Kernels per plant.		Nitrogen in kernels.			Kernels per plant.		Nitrogen in kernels.	
Number of analyses.	Average number.	Weight.	Per cent.	Weight.	Number of analyses.	Average number.	Weight.	Per cent.	Weight.	Number of analyses.	Average number.	Weight.	Per cent.	Weight.
		Gms.		*Gms.*			*Gms.*		*Gms.*			*Gms.*		*Gms.*
7.....	555	10.82	1.68	0.1709	35....	641	9.12	2.69	0.2491	112...	1,052	17.71	2.68	0.4606
6.....	654	14.66	2.20	.3040	35....	790	12.63	2.63	.3262	99....	1,040	19.13	2.84	.5207
18....	601	11.26	2.74	.3068	90....	745	11.42	2.64	.2985	285...	1,165	21.44	2.69	.5716
10....	338	5.87	3.27	.1919	53....	694	10.59	2.74	.2947	180...	1,057	19.49	2.82	.5486
5.....	306	6.44	3.68	.2431	23....	723	10.56	2.74	.2875	51....	1,093	23.16	2.71	.6154
6.....	366	6.62	4.25	.2837	30....	779	11.54	2.65	.3137	98....	1,245	22.02	2.80	.6153
5.....	274	5.68	5.13	.2773	25....	716	10.56	2.59	.2710	81....	1,183	20.12	2.64	.5317

It is difficult to explain why such great variations exist when there seems to be little or no tendency to transmit them. It seems apparent that the variations must be due to differences in environment. Since the ordinary factors of environment, as sunlight, warmth, moisture, and apparent fertility of the soil, are constant for all plants under our nursery conditions, we must conclude that there are factors profoundly influencing the growth of plants beyond the ordinary range of observation.

Figure 2 shows the general appearance of the centgener nursery, each centgener containing 100 plants 6 inches apart each way. Figure 3 shows a single centgener just after growth has started in the spring, about 40 per cent of the plants having winterkilled. The great variation in the size of the remaining plants is probably due to the effect of environment and is not hereditary. This environmental variation is usually noted even in centgeners where most plants have survived and is often interpreted as indicating real hereditary differences.

A number of interesting problems are suggested. Why should one plant, growing under practically the same environment as another, collect from the soil two or three times as much nitrogen? Or why should two plants yielding different quantities of grain collect the same quantities of nitrogen? Table III illustrates these variations.

TABLE III.—*Data from six wheat plants, showing irregular variation in yield and in nitrogen content of grain.*

Plant No.	Kernels.		Nitrogen in kernels.		Plant No.	Kernels.		Nitrogen in kernels.	
	Number.	Weight.	Per cent.	Total weight.		Number.	Weight.	Per cent.	Total weight.
		Grams.		*Grams.*			*Grams.*		*Grams.*
21107.....	1,058	22.879	2.45	0.5605	23905.....	776	18.507	3.57	0.6607
21108.....	1,030	16.679	2.59	.4324	23907.....	1,167	23.018	2.86	.6583
21109.....	927	16.026	1.74	.2789	24005.....	1,495	30.064	2.19	.6584

The three plants, Nos. 21107 to 21109, are from the same mother growing in a single centgener, probably less than 2 feet apart, yet the actual grams of nitrogen gathered differ more than 100 per

FIG. 2.—Centgener nursery, Nebraska Agricultural Experiment Station. Each centgener contains 100 plants.

cent. This difference is not inherited, as these plants rarely transmit this quality. It therefore seems hard to explain on a difference in the root development or in the functioning parts of the plant. As plants growing only 6 inches apart commonly exhibit such differences, it can not be ascribed to a difference in soil solution. Difference in vigor of growth is not a satisfactory explanation, as plants Nos. 23905, 23907, and 24005 illustrate. These three plants under uniform conditions yielded different quantities of grain, yet the heaviest yielder produced no more nitrogen than the lowest. Such differences are not only common among plants from the same centgener, but quite marked variations are also noted between centgeners from the same mother plant.

Some of the results obtained from the study of this problem are shown in figures 4 and 5. Figure 4 is a plat of a single centgener (1907), with the plants 6 inches apart each way, making the entire area 5 feet square. All plants in this centgener are from the same parent. Each square represents a plant. Where no figures occur the plant was missing. The upper number shows the percentage of nitrogen, the central number represents the kernels borne by the plant, and the lower number the weight in grams of the good kernels. Each plant was harvested separately, the kernels counted and weighed, and the percentage of nitrogen determined. Two wave lines indicate plants analyzing above 3 per cent nitrogen and one wave line those

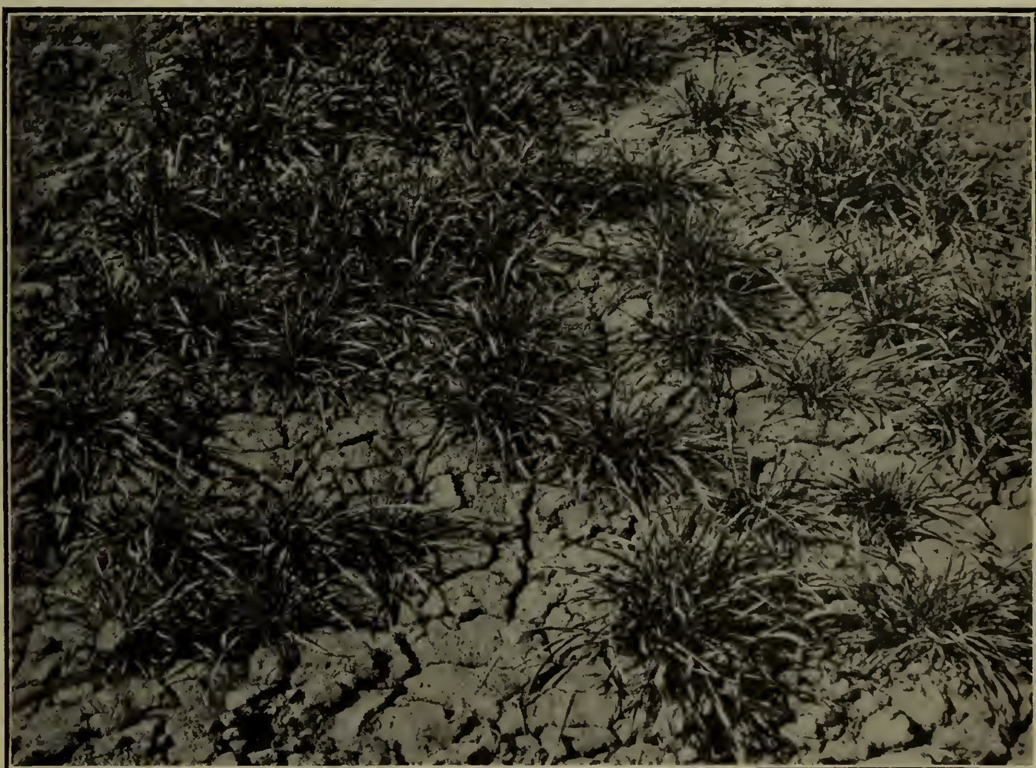

FIG. 3.—Wheat centgener just after growth has started in spring, showing about 40 per cent of the plants winterkilled. Note the great variation in size of the remaining plants.

analyzing between 2.8 and 3 per cent. A tendency to group is noted. Those containing between 2.56 and 2.80 per cent are not marked. One straight line indicates plants with between 2.55 and 2.40 per cent of nitrogen; two straight lines, less than 2.40 per cent.

Figure 5 shows a section of the wheat nursery in 1907. The small squares represent centgeners 5 feet square and the heavy lines outline family groups; that is, all the plants and centgeners within a heavy line came from the same original plant. The percentage of nitrogen was obtained by taking a composite sample from all plants on the centgener. Variation is quite marked, although there is some tendency for certain families to run high or low; as, for example, family 339.

An illustration of the irregularities in number and weight of kernels, in percentage of nitrogen, and in total yield of nitrogen per plant is afforded in Table IV, pedigree records of two families. The wide variations were supposed at first to represent natural fluctuations which would be in some degree transmitted, but the selection of these high fluctuations has had no apparent effect in modifying the

2.99 682 17.8	2.32 429 13.0	2.46 450 14.0	2.24 385 12.8	2.21 247 6.80	2.32 299 8.86	2.52 501 13.4	2.52 361 10.0	×	2.55 445 15.7
3.04 225 5.8	2.79 500 15.5	2.52 481 13.4	2.41 284 8.1	2.60 507 13.8	2.60 401 11.1	2.68 347 9.8	2.63 549 15.0	2.52 411 11.8	2.72 513 12.5
2.86 535 14.3	2.60 633 16.7	2.56 657 18.6	2.56 562 14.5	2.53 409 9.8	2.50 417 10.1	2.56 227 5.3	2.75 543 14.3	2.82 298 7.0	2.56 681 17.5
3.06 325 4.9	2 73 767 18.7	×	2.50 225 5.0	2.29 410 11.0	2.56 284 8.0	2.63 390 9.4	2.66 378 9.3	2.83 466 10.4	3.09 423 8.9
2.62 957 25.2	2.59 715 18.2	×	2.49 464 10.8	2.45 322 8.9	2.75 406 9.7	2.69 392 9.5	3.16 218 3.8	3.10 352 7.6	3.58 144 2.8
×	×	2.89 535 11.1	2.65 141 3.1	2.42 363 8.5	2.90 251 5.2	2.96 228 4.8	3.09 319 6.6	3.20 490 6.0	3.38 421 9.5
2.75 719 22.2	2.46 382 10.3	2.55 421 11.9	2.41 155 4.1	2.71 457 5.3	×	2.86 425 8.9	2.60 379 7.4	3.10 295 6.3	×
2.59 290 7.1	2.60 600 18.8	2.28 425 10.4	2.28 329 9.0	2.66 470 11.1	2.76 239 5 5	2.77 362 4.8	3.07 194 3.3	3.27 510 10.8	2.90 368 6.5
×	2.15 292 7.4	2.39 325 8.7	2.63 460 17.4	2.52 661 16.8	×	2.80 641 14.4	2.96 466 11.1	2.89 488 11.8	×
×	2.66 832 22.9	2.18 258 7.6	2.48 716 19.6	2.58 576 14.4	2.60 510 12.1	2.80 540 14.6	2.82 339 7.0	2.58 313 8.1	3.11 395 7.5

FIG. 4.—Wheat centgener of 100 plants, showing variations in yield of grain and of nitrogen in 1907. Upper figures, percentage of nitrogen content; middle figures, number of kernels produced; lower figures, weight of good kernels in grams. The various underscorings of the upper figures indicate five groups having successively higher nitrogen content as follows: (1) Figures underscored with two straight lines lie between 2.15 and 2.40 per cent; (2) those underscored with one straight line lie between 2.41 and 2.55 per cent; (3) those not underscored lie between 2.56 and 2.59 per cent; (4) those underscored with one wave line lie between 2.80 and 3 per cent; (5) those underscored with two wave lines lie above 3 per cent.

nitrogen content of plants in a family, as there always seemed to be a mean content for each family, to which the types returned.

As examples of variation, note that No. 35809 in family 42 has a low yield of nitrogen, yet the yield of nitrogen found in its progeny is practically equal to that of other members of the family. In family 48 (Table IV) note that in 1903 the three plants selected analyzed 3.82, 4.43, and 5.48 per cent of nitrogen, respectively, while the family as a whole contained 3.53 per cent. The progeny of these plants returned to normal in percentage and total yield of nitrogen, except No. 21909, in which the yield of grain was above the average. Just why these wide fluctuations occur when every precaution is taken to grow the plants under uniform conditions is not very apparent.

In 1908 a more thorough investigation of this point was made. Twenty-nine plants from the 1907 crop were all selected from a single centgener, and therefore all came from a single plant in 1906. From each of these 29 plants a centgener was planted, and also a row 14 feet long. The 29 centgeners were planted side by side, also the 29 rows. At harvest time all the plants in each of the 10 adjacent

2.35	2.66	2.65	2.93	3.04	2.89	2.73	2.56	2.59	2.89	2.77	2.62	2.55	2.77	2.71	2.55	2.89	2.48	2.77
313	314	314	328	328	381	381	385	385	405	405	417	417	522	522	556	556	3	3
2.46	2.60	2 72	3.00	2.45	2.72	2.75	2.71	2.72	2.50	2.83	2.63	2.58	2.93	2.99	2.52	2.52	2.68	3.02
313	314	314	328	328	381	381	385	385	391	405	417	417	522	522	556	556	3	3
2.56	2.62	2.69	2.93	2.92	2.57	2.82	2.53	2.62	2.53	3.00	2.53	2.49	2.93	3.11	2.59	2.68	2.50	2.36
313	314	314	317	338	381	385	385	385	391	405	417	417	522	522	556	556	3	3
2.58	2.60	2.63	2.73	3.17	2.83	2 53	2.53	2.76	2.49	2.89	2.59	2.63	2.66	2.95	2.55	2.66	2.55	3.03
313	314	316	317	339	381	385	385	385	391	405	417	417	522	522	556	556	632	3
2.34	2.69	2.84	2.60	3.16	2.80	3.02	2.66	2.93	3.06	2.87	2.75	2.71	2.89	2 92	2.39	2.63	2.62	2.96
313	314	316	317	339	381	385	385	390	391	407	417	417	522	555	556	556	632	3
2.72	2.76	2.97	2.87	3.17	2.89	2.49	2.80	2.71	2.48	2.89	2 60	2.68	2.89	2.68	2.92	2.69	2.68	2.95
313	314	316	317	339	381	385	385	390	391	407	417	417	522	555	556	556	632	3
2.46	2.95	2.93	2.90	3.09	2.79	2.60	2.89	2.75	2 77	2.82	3.00	2.77	3.00	2 83	2.83	2.83	2.56	2.83
313	314	316	317	339	381	385	385	390	391	407	408	417	522	555	556	556	632	3
2.62	2.80	2.89	2 73	3.13	2.45	2.63	2.56	2.76	2.68	2.92	2.84	2.71	3.03	2.97	2.59	2.79	2.62	2.89
313	314	316	317	332	379	385	385	390	391	408	408	425	522	555	555	556	556	3
2.85	2.59	2.76	2.63	2.58	2.87	2.73	2.68	2.60	2.68	2.69	2.89	2.66	2.72	3.02	2.73	2.76	2.69	2.68
313	314	316	317	377	379	385	385	390	391	408	408	425	425	555	555	556	556	3
2.87	2.44	2.86	2.63	2.46	2.82	2.73	2.63	2.86	2.66	2.59	2.83	2.52	2.63	2.77	2.73	2.68	2.71	2.66
314	314	316	317	377	379	385	385	390	391	408	408	425	425	555	555	556	556	3
2.87	2.80	3.00	2.71	2.79	2.34	2.66	2.49	2.65	2.68	2.79	2.87	2.90	2.62	3.16	2.84	2.72	1.76	2.68
314	314	316	317	377	379	385	385	390	391	408	408	425	425	555	555	556	556	3
2.77	2.58	2.96	2.72	2.63	3.20	2.73	2.73	2.72	2.77	2.84	3.02	2.66	2.65	2.79	2.90	2.62	2.11	2.73
314	314	316	317	377	377	385	385	390	390	408	408	425	425	555	555	556	556	3

FIG. 5.—Diagram of a portion of the wheat nursery in 1907, showing variations in nitrogen content in centgeners and families. Each square represents a single centgener, and each area within heavy lines shows the centgeners belonging to a single family. The upper figures represent the percentage of nitrogen; the lower figures are the family numbers.

centgeners were harvested in order and analyzed. The 10 duplicate rows were grown in a manner similar to field conditions; that is, sown at the rate of 5 pecks to the acre and the rows 8 inches apart. The plan was to see whether the same sort of variation would be found among plants under field conditions. To secure a uniform sample from the rows, 7 plants were harvested from each foot of row, 98 plants being harvested and analyzed from each row. In the centgeners the results were similar to those obtained in 1907. The plants in the rows, being planted close together, yielded only about one-tenth as much grain per plant, but the variation in yield and in percentage of nitrogen per plant was even greater than in the centgeners.

TABLE IV.—*Nitrogen content and yield of grain from families Nos. 42 and 48, from 1902 to 1905, inclusive.*

Original plant in 1902.		Record of progeny plants in 1903.					Record of progeny plants in 1904.					Record of progeny plants in 1905.				
Plant No.	Nitrogen in kernels.	Plant No.	Kernels per plant.		Nitrogen in kernels.		Plant No.	Kernels per plant.		Nitrogen in kernels.		Plant No.	Kernels per plant.		Nitrogen in kernels.	
			Number.	Weight.	Per cent.	Weight.		Number.	Weight.	Per cent.	Weight.		Number.	Weight.	Per cent.	Weight.
	Per cent.			*Grams.*		*Grams.*			*Grams.*		*Grams.*			*Grams.*		*Grams.*
							35805	693	11.4281	2.63	0.3006	8005	843	13.600	2.60	0.3536
												8006	602	10.496	2.69	.2823
												8007	1,200	19.279	2.81	.5415
												8008	1,640	34.290	2.91	.9978
												8009	1,355	23.360	2.75	.6424
												Average.	1,128.0	20.205	2.75	.5635
21301 (family 42)......	3.05	21305	312	6.2514	2.67	0.1669	35806	406	10.2617	2.48	.2545	8105	924	17.356	2.72	.4721
		21306	226	4.1516	2.90	.1204						8106	1,421	14.093	2.74	.3861
		21307	142	2.5691	3.04	.0781						8107	1,152	19.864	2.94	.5840
		21308	354	5.3080	3.45	.2004						8108	1,727	37.730	2.84	1.0715
		Average.	258.7	4.6950	3.02	.1414						8109	1,501	30.185	2.81	.8482
												Average.	1,345.0	23.8456	2.81	.6724
							35807	538	8.4050	2.79	.2345	8405	950	21.365	2.83	.6005
							35808	739	8.4660	3.25	.2751	8406	715	15.219	2.87	.4368
							35809	571	10.945	1.26	.1272	8407	1,091	22.852	2.65	.6056
												8408	1,218	26.530	2.62	.6951
												8409	1,215	29.783	2.67	.7952
							Average.	589.4	9.7311	2.48	.2377	Average.	1,037.8	23.1498	2.73	.6266

		21905	791	14.3111	2.64	.3778	38205	761	10.7984	2.52	.2721					
		21906	408	10.4800	3.18	.3340	38206	502	7.4332	2.26	.1719					
		21907	158	2.9243	3.35	.0980	38207	597	8.3134	2.30	.1912					
		21908	173	3.5574	3.82	.1359	38208	803	12.0218	2.32	.2789					
							38209	1,145	16.8862	2.38	.3971					
												10905	1,520	21.824	2.79	.6089
							Average.	761.6	11.0906	2.36	.2622	10906	1,253	27.854	2.80	.7799
												10907	1,805	41.999	2.96	1.2432
							38305	979	13.3663	2.57	.3435	10908	1,693	38.614	2.66	1.0271
												10909	1,218	20.737	2.65	.5495
												Average.	1,497.8	30.2056	2.77	.8417
							38306	1,000	14.2411	2.54	.3617	11105	680	10.185	2.97	.3025
												11106	1,449	26.542	2.85	.7654
												11107	2,075	43.237	2.83	1.2236
							38307	739	11.2404	2.85	.3203	11108	1,077	14.590	2.78	.4056
21901 (family 48).....	3.17	21909	525	12.1819	4.43	.5389						11109	1,902	24.726	2.64	.6528
												Average.	1,436.6	23.856	2.31	.6682
												11305	1,106	16.700	2.82	.4709
												11306	1,179	24.940	2.47	.6160
							38308	1,175	16.6916	2.50	.4173	11307	1,845	14.722	2.88	.4241
							38309	975	13.5982	2.46	.3345	11308	1,845	45.275	2.84	1.2858
												11309	868	15.644	2.58	.4036
							Average.	973.6	13.8275	2.58	.3555	Average.	1,160.2	23.4562	2.72	.6401
							38605	704	11.0436	2.34	.2584					
							38606	956	14.0310	2.61	.3662					
		21910	144	3.2348			38607	652	8.2336	2.77	.2281					
		21911	383	8.4593	5.48	.4636	38608	1,130	16.8243	2.30	.3870					
		21912	510	9.7236	2.31	.2246	38609	563	7.2238	2.64	.1907					
		21913	492	10.1925	3.01	.3062	Average.	801.0	11.4713	2.53	.2861					
		Average.	398.2	8.3406	3.53	.3100										

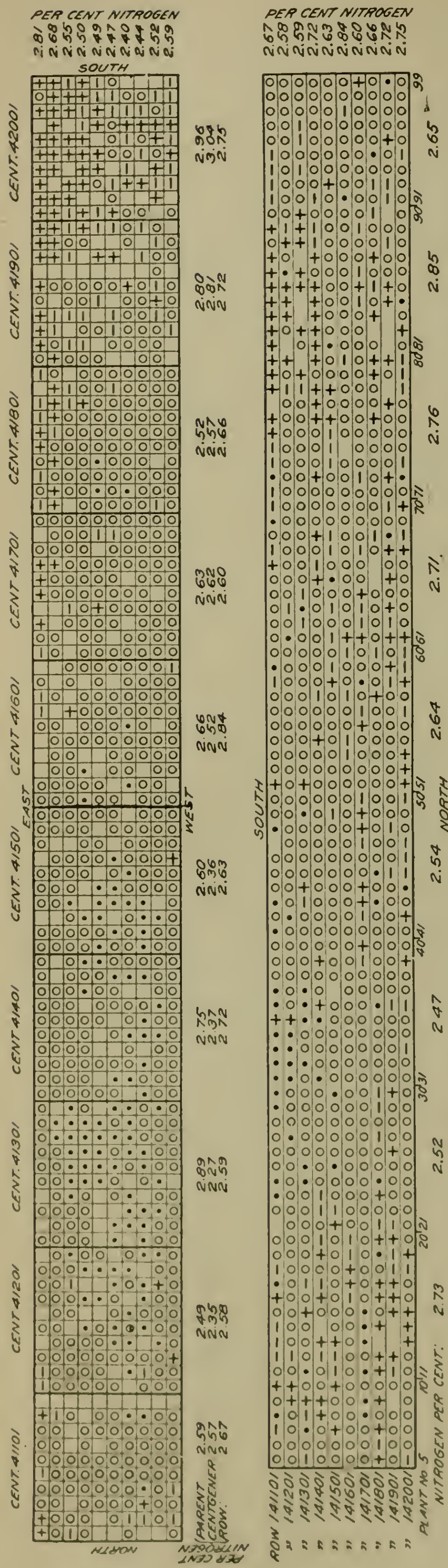

FIG. 6.—Diagram of 10 centgeners and 10 corresponding rows, showing variations in nitrogen content for individual plants, for each centgener, for each corresponding row, and also for their parents. The symbols used are as follows: Blank spaces indicate plants missing; dots, plants analyzing below 2.2 per cent nitrogen; circles, plants analyzing between 2.2 and 2.8 per cent; dashes, plants analyzing between 2.8 and 3 per cent; crosses, plants analyzing, 3 per cent and over.

Figure 6 is a graphic illustration of the centgener and row plats. The soil on which these plats were located was thought to be very uniform, and the appearance of the crop on the various plats was also uniform. The average per cent of nitrogen in the 10 centgeners varied from 2.27 (No. 41301) to 3.04 (No. 42001). To show the location of high and low nitrogen plants in each centgener the method of plotting shown in the figure was used. Where plants were missing the space is left blank; where a plant analyzed less than 2.20 per cent in nitrogen a dot (.) is used; from 2.20 to 2.80 per cent the circle is employed, denoting intermediates; from 2.80 to 3 per cent a dash (—) is used; and all plants analyzing above 3 per cent are marked by a cross (+). The "highs" and "lows" tend to segregate, as in 1907. Also there is a progressive increase toward one end of the series. The 10 duplicate rows are plotted in a similar manner. The rows were more uniform in composition than the centgeners, but they show a variation in composition from 2.58 to 2.84 per cent of nitrogen. Also there seems to be no consistent relation between the rows and the corresponding centgeners, or the parent plants. The variation in both centgeners and rows seems to be due to local effects, and does not appear to be hereditary. We may eliminate any possibility of hereditary effect by adding together the short rows in the centgeners which come end to end, thus making 10 long rows with each centgener equally represented. The results of such composite analyses are shown at the right of the centgener plat, indicating a variation ranging from 2.40 to 2.81 in percentage of nitrogen in the 10 rows. In the same way we may divide the 10 original rows into blocks having each row equally represented. Variation in the 10 blocks thus formed is shown, at the bottom of figure 6, to be from 2.47 to 2.85 per cent of nitrogen.

RELATION OF YIELD OF GRAIN TO NITROGEN CONTENT.

Since some centgeners yield more grain than others, the 29 centgeners from the same parent plant, of which the 10 centgeners just considered were a part, were arranged according to percentage of nitrogen and summarized in groups of 5 centgeners. This summary (Table V) shows some relation between yield of grain per centgener and nitrogen content, the yield varying inversely with the nitrogen content, but when the 29 corresponding rows were arranged in the same way no such relation was shown.

TABLE V.—*Nitrogen content of 29 centgeners and corresponding rows from family No. 831 in 1908.*

Arranged according to nitrogen in centgeners.							Arranged according to nitrogen in rows.						
Number of centgeners.	Centgener.			Corresponding row.		Nitrogen in parent plants.	Number of rows.	Row.		Corresponding centgener.			Nitrogen in parent plants.
	Nitrogen.	Total yield.	Average yield per plant.	Nitrogen.	Total yield.			Nitrogen.	Total yield.	Nitrogen.	Total yield.	Average yield per plant.	
	P. ct.	*Grams.*	*Grams.*	*P. ct.*	*Grams.*	*Per cent.*		*P. ct.*	*Grms.*	*P. ct.*	*Grms.*	*Grms.*	*P. ct.*
5......	2.83	616	7.19	2.67	194	2.69	5......	2.78	189	2.63	652	7.52	2.65
5......	2.63	614	7.13	2.66	200	2.68	5......	2.72	196	2.57	763	9.03	2.73
5......	2.56	851	9.60	2.65	173	2.63	5......	2.65	194	2.52	817	9.14	2.72
5......	2.52	767	9.01	2.57	179	2.66	5......	2.60	195	2.57	789	11.13	2.62
5......	2.44	799	9.01	2.52	189	2.71	5......	2.55	188	2.55	777	8.72	2.64
4......	2.34	916	10.50	2.64	209	2.68	4......	2.35	176	2.50	729	8.50	2.68

TABLE VI.—*Nitrogen content and yield of grain from 180 wheat plants, arranged in inverse order of percentage of nitrogen, in groups of 10.*

NINETY PLANTS FROM CENTGENER NO. 41801.

Plant No.	Nitrogen.	Yield.	Plant No.	Nitrogen.	Yield.	Plant No.	Nitrogen.	Yield.
	Per ct.	*Grams.*		*Per ct.*	*Grams.*		*Per ct.*	*Grams.*
52	3.44	1.55	64	2.62	11.11	27	2.43	7.49
79	3.22	.60	83	2.62	16.23	47	2.43	7.11
61	3.21	7.86	93	2.62	19.49	59	2.43	6.60
81	3.19	4.36	9	2.61	9.16	78	2.43	7.39
91	3.19	6.45	51	2.61	1.20	26	2.41	7.48
41	3.17	7.92	31	2.60	7.00	63	2.41	14.83
22	3.13	4.89	67	2.60	3.11	20	2.40	10.37
12	3.00	13.42	8	2.58	8.22	35	2.39	8:82
6	2.98	10.65	16	2.58	6.45	48	2.39	1.44
61	2.98	7.54	54	2.58	6.76	69	2.39	8.40
Average	3.15	6.52	Average	2.60	8.87	Average	2.41	7.99
71	2.92	2.46	56	2.58	6.61	68	2.37	8.00
90	2.92	10.52	60	2.57	10.56	96	2.37	6.07
80	2.89	9.14	73	2.57	15.31	55	2.36	8.30
33	2.85	7.69	76	2.56	1.34	24	2.34	11.07
72	2.82	11.75	32	2.53	8.42	45	2.30	9.59
87	2.82	.45	95	2.51	12.25	25	2.29	13.16
23	2.79	11.46	14	2.50	17.52	58	2.29	8.19
42	2.78	6.82	18	2.48	11.22	28	2.27	7.90
75	2.77	7.59	49	2.48	6.15	36	2.27	8.60
97	2.77	4.44	50	2.48	4.69	44	2.27	6.85
Average	2.83	7.43	Average	2.52	9.41	Average	2.31	8.77
11	2.71	11.47	86	2.48	10.01	39	2.26	8.66
74	2.70	6.83	34	2.47	21.10	29	2.25	9.85
21	2.68	10.52	43	2.47	11.18	30	2.23	9.17
85	2.68	6.32	70	2.47	13.25	46	2.22	7.24
15	2.65	5.55	98	2.47	6.83	37	2.20	10.01
53	2.65	14.26	40	2.46	12.08	57	2.20	14.78
84	2.65	7.45	77	2.46	8.27	89	2.20	13.50
88	2.65	5.21	99	2.46	12.81	17	2.11	10.31
94	2.65	16.94	10	2.44	2.93	19	2.11	10.40
7	2.62	9.17	5	2.43	16.20	38	2.09	6.90
Average	2.66	9.47	Average	2.46	11.47	Average	2.19	10.08

NINETY PLANTS FROM ROW NO. 141801.

Plant No.	Nitrogen.	Yield.	Plant No.	Nitrogen.	Yield.	Plant No.	Nitrogen.	Yield.
79	3.31	0.84	88	2.77	0.56	25	2.54	0.87
7	3.29	.26	8	2.76	1.69	31	2.54	1.54
57	3.25	.30	18	2.76	1.08	40	2.54	1.07
11	3.20	.35	99	2.76	3.41	72	2.53	1.72
9	3.19	.77	49	2.75	1.29	73	2.53	2.42
76	3.18	.44	46	2.74	2.51	77	2.52	.10
80	3.18	.79	89	2.74	1.30	62	2.51	.66
5	3.17	1.94	15	2.71	.53	33	2.48	1.62
87	3.10	.33	38	2.69	.63	35	2.48	4.32
14	3.09	.96	10	2.67	.18	69	2.47	2.00
Average	3.20	.70	Average	2.73	1.32	Average	2.51	1.63
83	3.09	.30	50	2.67	.65	81	2.47	1.46
16	3.08	.65	32	2.66	1.01	95	2.47	2.07
17	3.07	.79	64	2.66	1.21	37	2.45	.60
20	3.05	.69	93	2.66	1.29	39	2.45	.81
23	3.02	2.37	97	2.65	7.76	98	2.44	7.10
12	3.00	.77	47	2.64	.70	41	2.43	2.33
21	2.99	2.27	78	2.64	.45	42	2.39	2.74
27	2.96	1.59	50	2.63	.74	44	2.39	2.22
85	2.90	1.28	70	2.62	.57	58	2.39	.86
90	2.88	2.02	51	2.61	2.01	52	2.35	1.12
Average	3.00	1.27	Average	2.64	1.64	Average	2.42	2.13
68	2.87	1.42	74	2.61	1.07	71	2.33	1.39
30	2.84	2.34	26	2.60	1.18	91	2.30	.81
29	2.82	1.61	54	2.60	.10	61	2.29	.20
6	2.80	.52	63	2.60	.94	48	2.25	1.17
69	2.80	.24	34	2.58	.99	24	2.18	.21
55	2.79	.88	56	2.58	.38	43	2.14	1.31
84	2.79	1.48	86	2.58	.90	36	1.99	1.13
22	2.77	.65	96	2.58	2.50	45	1.87	1.07
53	2.77	1.92	28	2.57	2.16	94	1.37	.79
75	2.77	.52	67	2.56	2.31	19	1.33	.47
Average	2.80	1.14	Average	2.59	1.25	Average	2.00	.85

TABLE VI.—*Nitrogen content and yield of grain from 180 wheat plants, arranged in inverse order of percentage of nitrogen, in groups of 10*—Continued.

SUMMARY OF AVERAGES.

Group No.	Groups from centgener No. 41801.		Groups from row No. 141801.		Group No.	Groups from centgener No. 41801.		Groups from row No. 141801.	
	Nitrogen.	Grain per plant.	Nitrogen.	Grain per plant.		Nitrogen.	Grain per plant.	Nitrogen.	Grain per plant.
	Per cent.	*Grams.*	*Per cent.*	*Grams.*		*Per cent.*	*Grams.*	*Per cent.*	*Grams.*
1	3.15	6.52	3.20	0.70	6	2.46	11.47	2.59	1.25
2	2.83	7.43	3.00	1.27	7	2.41	7.99	2.51	1.63
3	2.66	9.47	2.80	1.14	8	2.31	8.77	2.42	2.13
4	2.60	8.87	2.73	1.32	9	2.19	10.08	2.00	.85
5	2.52	9.41	2.64	1.64					

The fluctuation in yield between plants within a centgener or row commonly varies as much as 500 per cent. To note whether this wide variation bore any relation to nitrogen content, the results from all the plants in centgener No. 41801 and from its corresponding row were tabulated according to nitrogen content (Table VI). While the summary shows a marked variation in nitrogen content, there is no corresponding change in yield, although there is a slight irregular tendency for yield to increase as nitrogen decreases.

While the foregoing data deal almost entirely with variations under nursery conditions, it would be interesting to know whether such local variations also occur under field conditions. That individual plants vary in this way is without doubt true, as the records of the row plats just cited show. To determine whether local variations were found in field plats, two sets of data were secured in the following manner:

In one of the field plats a drill row 224 feet long was selected and divided into 2-foot sections. The soil was of average fertility and uniformity. The results are shown in Table VIII.

Also a plat of land 77 by 88 feet with a 5-foot margin outside of this was sown to Turkey winter wheat, using a drill 5.5 feet wide. A very uniform stand and quite uniform growth were secured. The plat would have yielded about 30 bushels per acre. At harvest time it was divided into blocks 5.5 feet square, making 224 such blocks. A composite sample was made from the harvest of each block and each sample was analyzed for total nitrogen. Figure 7 is a diagram of this plat, showing the yield of grain and percentage of nitrogen in each block. The same variation is here found that has been noted heretofore in the centgeners and nursery rows. For example, in the first series of 16 blocks, Nos. 2 and 13 average 1.74 per cent of nitrogen, while Nos. 3 and 8 average 2.07 per cent. This variation seems large in view of the fact that each block has an average of 600 to 800 plants. In this case the variation must be due to some local soil condition. When different numbers of blocks are grouped, the resulting areas are of considerable size, as illustrated by figure 8. The plats shown at *B* are 11 by 22 feet in area and each was sampled 8 times, yet they show a variation in nitrogen content ranging from 1.81 to 1.97 per cent.

REDUCING THE EXPERIMENTAL ERROR.

So great is the fluctuation in individual plants, due to variation in environment, that there would seem to be no hope of improving the percentage of nitrogen through the continuous selection of high fluctuates. No cumulative results can be expected where the error is larger than the expected variation.

1.80 224	1.83 208	2.00 192	1.91 176	1.90 160	1.89 144	1.79 128	1.75 112	2.03 96	1.83 80	2.18 64	1.93 48	1.77 32	1.86 16
1.80 223	2.07 207	1.77 191	1.90 175	1.70 159	1.79 143	1.90 127	2.04 111	1.95 95	1.83 79	2.06 63	1.76 47	1.86 31	1.79 15
1.93 222	1.96 206	1.83 190	1.92 174	1.69 158	1.90 142	1.80 126	1.89 110	1.83 94	1.85 78	2.00 62	2.13 46	1.82 30	1.83 14
1.89 221	1.96 205	1.92 189	1.86 173	1.79 157	1.86 141	1.79 125	1.94 109	1.92 93	1.80 77	1.97 61	2.00 45	1.87 29	1.73 13
2.00 220	2.01 204	1.89 188	1.77 172	1.97 156	1.85 140	1.97 124	2.10 108	1.99 92	1.83 76	2.00 60	1.92 44	1.79 28	1.89 12
1.96 219	1.96 203	2.00 187	1.82 171	1.93 155	1.82 139	1.87 123	1.87 107	1.92 91	1.99 75	1.87 59	1.83 43	1.92 27	1.96 11
1.89 218	2.11 202	1.99 186	1.87 170	1.86 154	1.84 138	2.06 122	1.90 106	1.90 90	1.82 74	1.81 58	1.97 42	1.79 26	1.89 10
2.03 217	1.86 201	1.80 185	1.86 169	2.06 153	1.72 137	1.86 121	1.72 105	2.07 89	1.82 73	1.84 57	1.97 41	1.96 25	2.01 9
1.83 216	1.82 200	1.82 184	1.75 168	1.77 152	1.72 136	1.90 120	1.83 104	1.90 88	1.83 72	1.90 56	1.85 40	1.76 .24	2.07 8
1.87 215	2.14 199	1.96 183	1.87 167	1.97 151	1.90 135	1.90 119	2.13 103	1.80 87	1.83 71	1.90 55	2.06 39	1.94 23	1.87 7
1.90 214	1.94 198	1.94 182	1.77 166	1.89 150	1.86 134	1.82 118	1.87 102	1.80 86	1.84 70	1.87 54	2.04 38	1.94 22	1.89 6
1.94 213	1.76 197	1.96 181	1.99 165	1.87 149	2.04 133	1.93 117	1.77 101	1.74 85	1.89 69	1.93 53	1.96 37	2.04 21	1.97 5
1.83 212	1.99 196	1.97 180	2.08 164	1.99 148	1.96 132	2.15 116	1.82 100	1.78 84	1.83 68	1.98 52	1.89 36	1.85 20	1.87 4
1.85 211	1.87 195	1.85 179	1.82 163	1.92 147	1.89 131	2.13 115	1.82 99	1.73 83	1.83 67	1.96 51	2.04 35	1.86 19	2.08 3
2.10 210	1.83 194	1.85 178	1.96 162	2.01 146	1.92 130	1.68 114	1.89 98	1.85 82	1.85 66	1.83 50	1.85 34	2.07 18	1.75 2
1.93 209	1.86 193	1.93 177	1.87 161	1.90 145	1.86 129	1.99 113	1.89 97	1.83 81	1.82 65	1.96 49	1.99 33	1.99 17	2.06 1

FIG. 7.—Diagram of plat of Turkey wheat containing 224 blocks (each 5.5 feet square), showing the location of each block (lower figures) and variations in the percentage of nitrogen in the grain (upper figures).

The problem of finding a high-nitrogen wheat seems to resolve itself into the isolation of pure strains and the comparison of these for nitrogen content. To effect this isolation and comparison, a method must be found that will reduce error to the minimum. Replication according to some systematic method seems the most practical way. A number of examples have been worked up from data at hand to illustrate the effect of replication in reducing error.

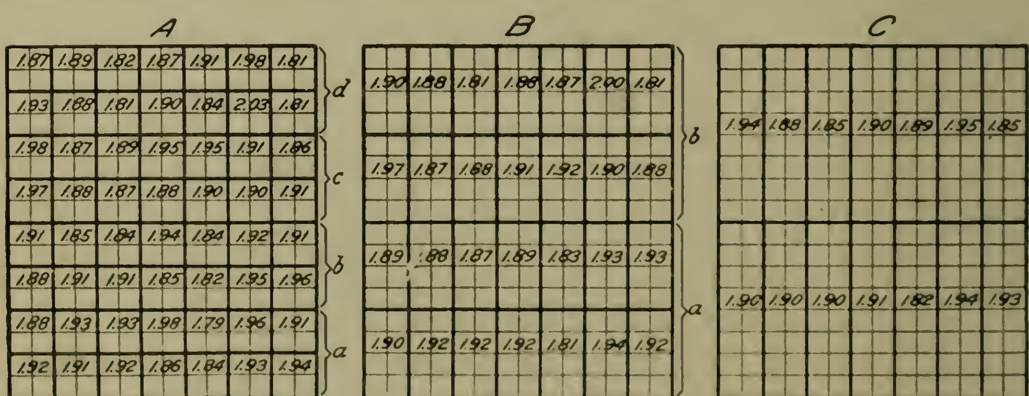

FIG. 8.—Diagrams of plats of Turkey wheat, showing the arrangement of 224 blocks (each 5.5 feet square) when combined in groups of adjacent blocks, with average nitrogen content for each group: *A*, Groups of 4; *B*, groups of 8; *C*, groups of 16.

REPLICATION OF SINGLE PLANTS.

In the first case the 840 centgener plants from the same parent, heretofore referred to (fig. 6), were grouped in various ways. Starting with the first plant and taking every forty-second plant thereafter gave a composite group of 20 plants. Taking the second and every forty-second thereafter in the same way gave a second group, and so on in the same manner until 42 groups of 20 plants each had been made. In a similar manner 21 groups of 40 plants each were formed. Also, each of the 10 centgeners, being 10 plants square, was formed into 10 rows and the rows numbered 1 to 10. Each row would have 10 plants if the stand were perfect, but in this case it averaged only 8.4 plants per row. By combining the first rows, second rows, etc., in the 10 centgeners, 10 groups were made of 10 rows, or 84 plants each. The results of the above combinations are shown in Table VII. Where 20 plants, uniformly distributed, were combined, the variation in nitrogen content was from 2.40 to 2.67 per cent. Where 40 plants were combined, the variation ranged from 2.47 to 2.60 per cent, but the 10 groups of 10 rows each varied only from 2.49 to 2.59 per cent. Just what should constitute the limits of error in any case will depend on the minimum limit of the variations which are to be detected. In this case 0.1 per cent of nitrogen might be considered such a limit.

TABLE VII.—*Nitrogen content of 90 plants of Turkey wheat from 1 centgener and of 840 plants variously combined into groups to show deviation from mean.*

90 single plants in 1 centgener (united when having same nitrogen content).						840 single plants variously combined into groups.							
						Combination 1.—Four groups, each composed of every 42d plant in 10 centgeners, or 20 plants.				Combination 2. — Two groups, each composed of every 21st plant in 10 centgeners, or 40 plants.		Combination 3.—A group composed of every 10th row in 10 centgeners or 10 rows, equaling 84 plants.	
Nitrogen.	Frequency.	Deviation.	Nitrogen.	Frequency.	Deviation.	Nitrogen.	Deviation.	Nitrogen.	Deviation.	Nitrogen.	Deviation.	Nitrogen.	Deviation.
P. ct.			*Per ct.*			*Per ct.*		*Per ct.*		*Per ct.*		*Per ct.*	
2.09	1	−0.482	2.58	4	+0.008	2.47	−0.043	2.44	−0.089	2.48	−0.044	2.49	−0.042
2.11	2	− .462	2.60	3	+ .028	2.47	− .043	2.51	− .019	2.49	− .034	2.51	− .022
2.20	3	− .372	2.61	2	+ .038	2.48	− .033	2.51	− .019	2.50	− .024	2.52	− .012
2.22	1	− .352	2.62	4	+ .048	2.50	− .013	2.51	− .019	2.52	− .004	2.52	− .012
2.23	1	− .342	2.65	5	+ .078	2.51	− .003	2.51	− .019	2.52	− .004	2.53	− .002
2.25	1	− .322	2.68	2	+ .108	2.51	− .003	2.51	− .019	2.53	+ .006	2.53	− .002
2.26	1	− .312	2.70	1	+ .128	2.53	+ .017	2.52	+ .009	2.53	+ .006	2.53	− .002
2.27	3	− .302	2.71	1	+ .138	2.54	+ .027	2.59	+ .061	2.53	+ .006	2.54	+ .008
2.29	2	− .282	2.77	2	+ .198	2.54	+ .027	2.59	+ .061	2.55	+ .036	2.56	+ .028
2.30	1	− .272	2.78	1	+ .208	2.58	+ .067	2.60	+ .071	2.59	+ .066	2.59	+ .058
2.34	1	− .232	2.79	1	+ .218								
2.36	1	− .212	2.82	2	+ .248	2.513	.0276	2.529	.0386	2.524	.023	2.532	.0188
2.37	3	− .202	2.85	1	+ .278								
2.39	3	− .182	2.89	1	+ .318	2.46	− .092	2.40	− .128	2.47	− .07		
2.40	1	− .172	2.92	2	+ .348	2.51	− .042	2.48	− .048	2.48	− .06		
2.41	2	− .162	2.98	2	+ .408	2.52	− .032	2.50	− .028	2.48	− .06		
2.43	5	− .142	3.00	1	+ .428	2.52	− .032	2.52	− .008	2.50	− .04		
2.44	1	− .132	3.13	1	+ .558	2.54	− .012	2.53	+ .002	2.53	− .01		
2.46	3	− .112	3.17	1	+ .598	2.54	− .012	2.54	+ .012	2.53	− .01		
2.47	4	− .102	3.19	2	+ .618	2.54	− .012	2.54	+ .012	2.54	.00		
2.48	2	− .092	3.21	1	+ .638	2.58	+ .028	2.56	+ .032	2.54	.00		
2.50	1	− .072	3.22	1	+ .648	2.64	+ .088	2.56	+ .032	2.56	+ .02		
2.51	1	− .062	3.44	1	+ .868	2.67	+ .118	2.57	+ .042	2.60	+ .06		
2.53	1	− .042						2.57	+ .042	2.61	+ .07		
2.56	1	− .012	2.572		.214	2.552	.0468	2.58	+ .052				
2.57	2	− .002								2.54	.0364		
								2.528	.0365				

Where the plants were repeated 20 and 40 times the error was almost what we might reasonably expect the actual variation in pure strains to be, but where 10 centgener rows, or 84 plants, were combined the extreme error was within bounds. The data would indicate that single plants would have to be replicated nearly 100 times to bring the variation within the limits of error.

REPLICATION OF 2-FOOT ROWS.

Table VIII illustrates the variation to be expected by replicating 2-foot rows. For these data a single 220-foot drill row in the general wheat field was divided into 2-foot sections and a composite sample made of each section. The sections were combined in two different ways. The first combination was composed of every twenty-second section, making 22 groups of 5 sections each, and the second combination was composed of every eleventh section, making 11 groups of 10 sections each.

Here, again, the extremes are rather wide, but if these are excluded the results would be called satisfactory. If a comparison of pure strains of wheat was being made under similar conditions, it would be necessary to take for further trial the entire best half of the strains tested in order to be within the limit of error. (See p. 30 and fig. 9.)

Table VIII.—*Nitrogen content of 110 2-foot sections of drill row of Turkey wheat, arranged in order of percentage of nitrogen and also in groups of 5 and 10, to show deviation from mean.*

Single 2-foot rows (those with same nitrogen content united).						Five rows in a group composed of every 22d row.		Ten rows in a group composed of every 11th row.	
Nitrogen.	Frequency.	Deviation.	Nitrogen.	Frequency.	Deviation.	Nitrogen.	Deviation.	Nitrogen.	Deviation.
Per cent.			*Per cent.*			*Per cent.*		*Per cent.*	
1.76	1	−0.271	2.04	5	+0.009	1.91	−0.109	1.96	−0.069
1.79	1	− .241	2.05	2	+ .019	1.96	− .059	1.98	− .049
1.81	1	− .221	2.06	4	+ .029	2.01	− .009	1.98	− .049
1.85	1	− .181	2.07	5	+ .039	2.01	− .009	2.00	− .029
1.86	2	− .171	2.08	4	+ .049	2.01	− .009	2.02	− .009
1.87	1	− .161	2.09	3	+ .059	2.02	+ .001	2.03	+ .001
1.88	1	− .151	2.10	6	+ .069	2.02	+ .001	2.04	+ .011
1.89	1	− .141	2.11	1	+ .079	2.03	+ .011	2.06	+ .031
1.90	1	− .131	2.12	2	+ .089	2.06	+ .041	2.06	+ .031
1.91	1	− .121	2.13	2	+ .099	2.09	+ .071	2.06	+ .031
1.92	7	− .111	2.14	3	+ .109	2.09	+ .071	2.13	+ .101
1.93	5	− .101	2.15	1	+ .119				
1.95	5	− .081	2.16	1	+ .129	2.019	.0355	2.029	.0374
1.96	3	− .071	2.17	1	+ .139				
1.97	2	− .061	2.19	2	+ .159	1.96	− .077		
1.98	5	− .051	2.20	3	+ .169	1.98	− .057		
1.99	2	− .041	2.26	4	+ .229	1.99	− .047		
2.00	4	− .031	2.28	1	+ .249	1.99	− .047		
2.01	7	− .021	2.37	1	+ .339	2.03	− .007		
2.02	2	− .011				2.03	− .007		
2.03	6	− .001	2.031		+ .093	2.04	+ .003		
						2.05	+ .013		
						2.08	+ .043		
						2.09	+ .053		
						2.17	+ .133		
						2.037	.0443		

REPLICATION OF 16-FOOT ROWS.

Since 16-foot rows are frequently used as test plats, a determination was made of the variation in a series of these. One hundred check plats from the 1909 field nursery were grouped by fives and tens in the same manner as in the two cases just cited. The results are shown in Table IX. The coefficient of variability and also the extreme variation are less than in the cases just considered.

TABLE IX.—*Nitrogen content of 100 16-foot rows of Turkey wheat, all from the same seed, arranged singly in order of percentage of nitrogen and also in groups of 5 and 10, to show deviation from mean.*

Single 16-foot rows (those with same nitrogen content united).						Five rows in a group composed of every 20th row.		Ten rows in a group composed of every 10th row.	
Nitrogen.	Frequency.	Deviation.	Nitrogen.	Frequency.	Deviation.	Nitrogen.	Deviation.	Nitrogen.	Deviation.
Per cent.			*Per cent.*			*Per cent.*		*Per cent.*	
1.82	1	−0.316	2.18	6	+0.044	2.09	−0.056	2.10	−0.037
1.86	2	− .276	2.20	1	+ .064	2.11	− .036	2.11	− .027
1.87	1	− .266	2.21	1	+ .074	2.13	− .016	2.12	− .017
1.89	1	− .246	2.22	4	+ .084	2.13	− .016	2.13	− .007
1.92	2	− .216	2.23	1	+ .094	2.13	− .016	2.14	+ .003
1.93	2	− .206	2.25	2	+ .114	2.14	− .006	2.14	+ .003
1.94	1	− .196	2.27	2	+ .134	2.16	+ .014	2.15	+ .013
1.96	3	− .176	2.28	2	+ .144	2.17	+ .024	2.15	+ .013
1.97	2	− .166	2.29	3	+ .154	2.19	+ .044	2.15	+ .013
1.99	1	− .146	2.31	3	+ .174	2.21	+ .064	2.18	+ .043
2.00	3	− .136	2.34	2	+ .204				
2.01	1	− .126	2.35	1	+ .214	2.146	.0292	2.137	.0176
2.03	5	− .106	2.36	1	+ .224				
2.04	3	− .096	2.38	1	+ .244	2.07	− .058		
2.06	6	− .076	2.42	1	+ .284	2.08	− .048		
2.07	2	− .066	2.43	1	+ .294	2.09	− .038		
2.08	6	− .056	2.45	1	+ .314	2.09	− .038		
2.10	3	− .036	2.48	1	+ .344	2.10	− .028		
2.11	2	− .026	2.50	1	+ .364	2.13	+ .002		
2.13	7	− .006	2.52	1	+ .384	2.17	+ .042		
2.14	2	+ .004				2.18	+ .052		
2.15	4	+ .014	2.136		.1178	2.18	+ .052		
2.17	4	+ .034				2.19	+ .062		
						2.128	.0420		

Where the 16-foot rows were repeated 10 times the extreme difference in nitrogen content was only 0.08 per cent. To further test this question, 500 rows, each 16 feet in length, were planted in the fall of 1909 under quite uniform conditions. These rows were harvested and combined in groups of 5, 10, 15, and 20, as in the previous case. The results are summarized at the bottom of Table X. When the rows are repeated only 5 times the error is too wide for satisfactory results, but when repeated 10 times the error is small enough for experimental purposes. Repeating 15 and 20 times gave only a small further reduction in variation.

A point of interest is the fact that in 1909, where rows were repeated 5 or 10 times, the experimental error was less than it was in 1910. In fact, there is no way of establishing a set rule as to the number of repetitions necessary, since the experimental error is influenced by all the factors affecting the growth of plants, such as soil fertility, climate, or insects. In order to know what this error is in a particular case it would be advisable to grow a sufficient number of check plats in each system of plats to determine the error by actual test.

Table X is a summary of results with the systematic repetition of plants and rows. The column under "Coefficient of variability" shows that repeating 20 single plants in a systematic way has given about as great accuracy in determining nitrogen content as 2-foot rows repeated 10 times or 16-foot rows repeated 5 times. For determining comparative nitrogen content, repeating single plants 20 to 40 times in a systematic method seems to give quite satisfactory results.

TABLE X.—*Summary showing degree of error due to variation in environment, according to several methods of comparison.*

Classification.	Number of groups.	Mean nitrogen content.	Extreme variation.	Average deviation.	Standard deviation.	Coefficient of variability.
		Per cent.				*Per cent.*
90 single plants (Table VII)	90	2.572	2.09–3.44	0.2140	0.279	10.85
840 single plants (Table VII):						
Every 42d plant, 20 plants in a group	a 10	2.513	2.47–2.58	.0276	.0335	1.33
	b 10	2.552	2.46–2.67	.0468	.0592	2.32
	c 10	2.529	2.44–2.60	.0386	.0473	1.87
	d 12	2.528	2.40–2.58	.0365	.0484	1.91
Average	10.5	2.531	2.44–2.61	.0374	.0471	1.86
Every 21st plant, 40 plants in a group	a 10	2.524	2.48–2.59	.0230	.0307	1.22
	b 11	2.540	2.47–2.61	.0364	.0455	1.17
Average	10.5	2.532	2.47–2.60	.0297	.0381	1.50
As 10 centgeners, every 10th centgener row, 10 rows in a group	10	2.532	2.49–2.59	.0188	.0260	1.03
110 2-foot rows, single rows (Table VIII)	110	2.031	1.76–2.37	.093	.1087	5.35
Every 22d row, 5 rows in a group	a 11	2.019	1.91–2.09	.0355	.0499	2.47
	b 11	2.037	1.96–2.17	.0443	.0574	2.82
Average	11	2.028	1.94–2.13	.0399	.0537	2.65
Every 11th row, 10 rows in a group	11	2.029	1.96–2.13	.0374	.0464	2.28
100 16-foot rows (Table IX)	100	2.136	1.82–2.52	.1178	.1492	6.98
Every 20th row, 5 rows in a group	a 10	2.146	2.09–2.21	.0292	.0347	1.62
	b 10	2.128	2.07–2.19	.0420	.0451	2.12
Average	10	2.137	2.08–2.20	.0356	.0399	1.87
Every 10th row, 10 rows in a group	10	2.137	2.10–2.18	.0176	.0219	1.02
500 16-foot rows (in 1910):						
Single rows		1.905	1.68–2.28	.0859	.1080	5.67
Every 100th row, 5 rows in a group	a 25	1.904	1.81–1.99	.0393	.0469	2.46
	b 25	1.917	1.84–2.02	.0376	.0469	2.45
	c 25	1.908	1.81–2.03	.0403	.0502	2.63
	d 25	1.890	1.81–1.98	.0388	.0488	2.42
Average		1.905	1.82–2.00	.0379	.0474	2.49
Every 50th row, 10 rows in a group	a 25	1.908	1.83–1.96	.0273	.0329	1.72
	b 25	1.902	1.83–1.96	.0286	.0343	1.80
Every 33d row, 15 rows in a group		1.905	1.86–1.96	.0235	.0276	1.45
Every 25th row, 20 rows in a group		1.905	1.85–1.96	.0190	.0242	1.27

THE SMALL-BLOCK TEST.

Figure 7 (p. 22) illustrates the method of making the small-block test and also shows the percentage of nitrogen in the grain from each block. Table XI shows the result of repeating these blocks 4, 8, and 16 times in a systematic method, i. e., taking every fifty-sixth, twenty-eighth, or fourteenth block (fig. 8). The experimental error varied inversely with the number of repetitions, but was only within the limit of error when the repetition was 16 times.

TABLE XI.—*Nitrogen content of Turkey wheat grown in 224 block plats (each 5.5 feet square) in 1909 and 1910.*

SYSTEMATICALLY REPEATED TO FORM GROUPS OF 4, 8, AND 16 BLOCKS.

Four blocks in a group composed of every 56th block.				Eight blocks in a group composed of every 28th block.		Sixteen blocks in a group composed of every 14th block.	
Nitrogen.	Deviation.	Nitrogen.	Deviation.	Nitrogen.	Deviation.	Nitrogen.	Deviation.
Per cent.		*Per cent.*		*Per cent.*		*Per cent.*	
1.78	−0.128	1.81	−0.115	1.82	−0.096	1.84	−0.06
1.84	− .068	1.85	− .075	1.87	− .046	1.86	− .04
1.85	− .058	1.87	− .055	1.88	− .036	1.87	− .03
1.88	− .028	1.87	− .055	1.90	− .016	1.87	− .03
1.89	− .018	1.90	− .025	1.90	− .016	1.89	− .01
1.90	− .008	1.93	+ .005	1.90	− .016	1.90	+ .00
1.91	+ .002	1.93	+ .005	1.92	+ .004	1.90	+ .00
1.91	+ .002	1.96	+ .035	1.92	+ .004	1.91	+ .01
1.93	+ .022	1.96	+ .035	1.94	+ .024	1.91	+ .01
1.93	+ .022	1.96	+ .035	1.94	+ .024	1.92	+ .02
1.94	+ .032	1.96	+ .035	1.94	+ .024	1.92	+ .02
1.95	+ .042	1.97	+ .045	1.95	+ .034	1.93	+ .03
1.98	+ .072	1.97	+ .045	1.95	+ .034	1.94	+ .04
2.02	+ .112	2.01	+ .085	1.99	+ .074	1.94	+ .04
Av'ge..1.908	.0439	1.925	.0464	1.916	.032	1.90	.024
1.83	− .041	1.80	− .094	1.83	− .051		
1.83	− .041	1.86	− .034	1.85	− .031		
1.83	− .041	1.87	− .024	1.86	− .021		
1.84	− .031	1.87	− .024	1.86	− .021		
1.84	− .031	1.87	− .024	1.86	− .021		
1.85	− .021	1.88	− .014	1.87	− .011		
1.85	− .021	1.89	− .004	1.88	− .001		
1.86	− .011	1.90	+ .006	1.88	− .001		
1.87	− .001	1.90	+ .006	1.88	− .001		
1.87	− .001	1.90	+ .006	1.89	+ .011		
1.88	+ .009	1.90	+ .006	1.90	+ .021		
1.94	+ .069	1.90	+ .006	1.92	+ .041		
1.95	+ .079	1.94	+ .046	1.92	+ .041		
1.95	+ .079	2.04	+ .146	1.93	+ .051		
Av'ge..1.871	.034	1.894	.0314	1.881	.0231		

Table XI.—*Nitrogen content of Turkey wheat grown in 224 block plats (each 5.5 feet square) in 1909 and 1910*—Continued.

Combined in Groups of 4, 8, and 16 Adjacent Blocks, to Show the Effect of Size of Plat on Variability.

Four sets of 14 groups, with 4 adjacent blocks in each group.						Two sets of 14 groups, with 8 adjacent blocks in each group.			One set of 14 groups, with 16 adjacent blocks in each group.		
Nitrogen.	Deviation.	Standard deviation.	Nitrogen.	Deviation.	Standard deviation.	Nitrogen.	Deviation.	Standard deviation.	Nitrogen.	Deviation.	Standard deviation.
Per cent.			*Per ct.*			*Per ct.*			*Per ct.*		
1.79	−0.117	....	1.86	−0.05	....	1.81	−0.086	....	1.82	−0.077	....
1.84	− .067	....	1.87	− .04	....	1.83	− .066	....	1.85	− .047	....
1.86	− .047	....	1.87	− .04	....	1.87	− .026	....	1.85	− .047	....
1.88	− .027	....	1.88	− .03	....	1.88	− .016	....	1.88	− .017	....
1.91	+ .003	....	1.88	− .03	....	1.89	− .006	....	1.89	− .007	....
1.91	+ .003	....	1.89	− .02	....	1.89	− .006	....	1.90	+ .003	....
1.92	+ .013	....	1.90	− .01	....	1.90	+ .004	....	1.90	+ .003	....
1.92	+ .013	....	1.90	− .01	....	1.92	+ .024	....	1.90	+ .003	....
1.93	+ .023	....	1.91	0	....	1.92	+ .024	....	1.90	+ .003	....
1.93	+ .023	....	1.91	0	....	1.92	+ .024	....	1.91	+ .013	....
1.93	+ .023	....	1.95	+ .04	....	1.92	+ .024	....	1.93	+ .033	....
1.94	+ .033	....	1.95	+ .04	....	1.93	+ .034	....	1.94	+ .043	....
1.96	+ .053	....	1.97	+ .06	....	1.93	+ .034	....	1.94	+ .043	....
1.98	+ .073	....	1.98	+ .07	....	1.94	+ .044	....	1.95	+ .053	....
Average..1.907	.037	0.048	1.91	.0314	0.0376	1.896	.0299	0.0372	1.897	.028	0.0363
1.82	− .072	....	....	....	....	1.81	− .081	....			
1.84	− .052	....	1.81	− .072	....	1.81	− .081	....			
1.84	− .052	....	1.81	− .072	....	1.87	− .021	....			
1.85	− .042	....	1.81	− .072	....	1.87	− .021	....			
1.85	− .042	....	1.82	− .062	....	1.88	− .011	....			
1.88	− .012	....	1.84	− .042	....	1.88	− .011	....			
.....	....	....	1.87	− .012	....	....	....	....			
1.91	+ .018	....	1.87	− .012	....	1.88	− .011	....			
1.91	+ .018	....	1.88	− .002	....	1.88	− .011	....			
1.91	+ .018	....	1.89	+ .008	....	1.90	+ .009	....			
1.91	+ .018	....	1.90	+ .018	....	1.90	+ .009	....			
1.92	+ .028	....	1.91	+ .028	....	1.91	+ .019	....			
1.94	+ .048	....	1.93	+ .048	....	1.92	+ .029	....			
1.95	+ .058	....	1.98	+ .098	....	1.97	+ .079	....			
1.96	+ .068	....	2.03	+ .148	....	2.00	+ .109	....			
Average..1.892	.0390	.0436	1.882	.0496	.0636	1.891	.0358	.0492			

TABLE XI.—*Nitrogen content of Turkey wheat grown in 224 block plats (each 5.5 feet square) in 1909 and 1910*—Continued.

SUMMARY SHOWING EXPERIMENTAL ERROR WHEN BLOCKS ARE ASSEMBLED IN VARIOUS WAYS..

Classification.	Number of groups.	Season of 1909. Mean nitrogen content.	Season of 1909. Extreme variation.	Season of 1909. Average deviation.	Season of 1909. Standard deviation.	Season of 1909. Coefficient of variability.	Season of 1910. Mean nitrogen content.	Season of 1910. Extreme variation.	Season of 1910. Average deviation.	Season of 1910. Standard deviation.	Season of 1910. Coefficient of variability.
		P. ct.				*P. ct.*	*P. ct.*				*P. ct.*
Systematically repeated..	1	1.898	1.68–2.18	0.0786	0.0981	5.17	1.866	1.66–2.24	0.0692	0.8987	4.81
Every 56th block, 4 blocks in a group...	a 14	1.908	1.78–2.02	.0439	.0579	3.03	1.852	1.81–1.89	.0250	.0278	1.50
	b 14	1.871	1.83–1.95	.0340	.0425	2.27	1.850	1.79–1.91	.0300	.0342	1.85
	c 14	1.925	1.81–2.01	.0464	.0546	2.84	1.871	1.81–1.92	.0259	.0316	1.69
	d 14	1.894	1.80–2.04	.0314	.0503	2.61	1.885	1.82–2.04	.0386	.0532	2.82
Average..........	14	1.90	1.80–2.00	.0389	.0513	2.69	1.865	1.81–1.94	.0299	.0347	1.96
Every 28th block, 8 blocks in a group...	a 14	1.916	1.82–1.99	.0320	.0438	2.29	1.863	1.83–1.89	.0143	.0167	.89
	b 14	1.881	1.83–1.93	.0231	.0287	1.53	1.869	1.83–1.91	.0184	.0226	1.21
Average..........	14	1.90	1.82–1.96	.0275	.0362	1.91	1.866	1.83–1.90	.0163	.0196	1.05
Every 14th block, 16 blocks in a group...	14	1.90	1.84–1.94	.0240	.0295	1.55	1.864	1.84–1.88	.0123	.0133	.72
Adjacent groups: Four blocks in a group	a 14	1.907	1.79–1.98	.0370	.0480	2.52	1.929	1.80–2.02	.0503	.0612	3.17
	b 14	1.892	1.82–1.96	.0390	.0436	2.31	1.871	1.79–1.96	.0400	.0484	2.59
	c 14	1.910	1.86–1.98	.0314	.0376	1.93	1.841	1.78–1.92	.0301	.0368	2.00
	d 14	1.882	1.81–2.03	.0496	.0636	3.31	1.817	1.77–1.89	.0261	.0319	1.76
Average..........	14	1.894	1.82–1.99	.0442	.0482	2.54	1.865	1.78–1.95	.0366	.0446	2.38
Eight blocks in a group	a 14	1.896	1.81–1.94	.0299	.0372	1.96	1.901	1.82–1.98	.0444	.0504	2.65
	b 14	1.891	1.81–2.00	.0358	.0492	2.60	1.829	1.79–1.89	.0263	.0299	1.63
Average..........	14	1.894	1.81–1.97	.0328	.0432	2.28	1.865	1.80–1.93	.0353	.0401	2.14
Sixteen blocks in a group...............	14	1.897	1.82–1.95	.028	.0363	1.91	1.866	1.80–1.96	.0403	.0464	2.49

In 1910 the same set of blocks was again in wheat, and they were again grouped in the same way, exactly the same plats being combined each year. The experimental error was less in 1910 than in 1909, and repeating the blocks 8 times gave total variations ranging from 1.83 to 1.90 per cent, which is well within the experimental limit. It appears that repeating the plats in this particular case 8 times in 1910 gave as good results as repeating 16 times in 1909; in fact, repeating 4 times would have been almost as satisfactory. In the case of the 16-foot rows just described the error was least in 1909, while the reverse was true in regard to the error in the blocks. This result again illustrates the point, made in discussing the row tests, that sufficient check plats must be used in order to know the experimental error in any particular case.

INCREASING THE SIZE OF PLAT.

Table XI also illustrates the effect of increasing the size of the plat. While such increase ought theoretically to reduce error from the standpoint of increasing the number of plants and including a larger number of local soil variations, yet an equal number of large plats reach over into new territory and include new causes for variation. To secure a practical illustration the 224 small blocks were combined into several series of larger plats by adding together adjacent plats. This method is illustrated in figure 8 (p. 22), and the statistical data are given in Table XI, a study of which shows that increasing the size above four adjacent blocks does not decrease the variability. In comparing systematic replication (Table X) with increase in size of plat it will be seen that the former constantly decreases variation, and would so continue to infinity, while the latter would not be controlled by such a law. It has been noted that the degree of variability was not the same when similar data were collected from different fields or in different years.

Variation is not a constant factor even where conditions are quite uniform, as is illustrated by the four sets of 14 groups composed of four adjacent plats. The fluctuation of extremes is almost twice as great in the fourth set as in the third.

The foregoing data can not be taken as a strict comparison of the different methods, as the data in each case were secured under somewhat different conditions. They are mainly valuable in illustrating the expected variation when different methods of comparison are used. It is evident that, whatever the method used, a single plat or duplicate plats can not be relied on for determining the actual nitrogen content of a strain or variety of wheat. The plats must be repeated 5 to 15 times, depending on uniformity of conditions and accuracy of results desired. In addition, at least a few series of check plats should be included in order to determine the experimental error as a guide to accuracy of results.

Our experience so far indicates that the simplest and most accurate method is to use 16-foot rows, replicated 10 times, with a check plat every 5 or 10 rows.

THE LIMIT OF EXPERIMENTAL ERROR.

The above examples give some indication of the experimental error to be expected by the different methods. Since the experimental error depends upon the variation in environmental conditions, it is possible that conditions might be found so ideal that there would be practically no experimental error; also that under other conditions it might be greater than in the cases just cited. In all cases the experimental error should be determined by the use of check plats. With this factor known it will be possible to decide on some plan of selection. Figure 9 illustrates an ideal case where the experimental error is known. Suppose that in 10 strains of wheat being tested by the row method with check plats, the highest should average 3 per cent of nitrogen and the lowest 2 per cent of nitrogen, the remainder being distributed between. Suppose a series of checks, repeated in the same way as the tested strains, showed a variation of 2.2 per cent to 2.6 per cent, with a mean of 2.4 per cent. This would give an experimental error of 0.2 per cent; that is, a certain strain might be 0.2 per cent higher than it should be or 0.2 per cent lower than it should be. Let the line *ab* indicate the variation in nitrogen content obtained in the 10 strains under test. If the experimental error equaled 0.2 per cent, then No. 1 might equal either 3.2 per cent or 2.8 per cent, and in the same way a strain analyzing 2.6 per cent might possibly be either 2.8 per cent or 2.4 per cent. In other words, the strain analyzing 2.6 per cent might be just as good as the one analyzing 3 per cent. Therefore, if the experimental error has been determined, the rule would be to double the error and subtract this sum from the highest variant. The remainder after subtraction would represent the nitrogen content below which all strains could be discarded without danger of discarding a high-nitrogen strain. (The same method applies to the use of experimental error in selecting for yield.) In the case illustrated in figure 9, all strains analyzing above 2.6 per cent must be selected for further test to be sure that one of the best is not being left. If the experimental error equals one-half the real variations in strains compared, then no selection can be made, but all the strains must be retested.

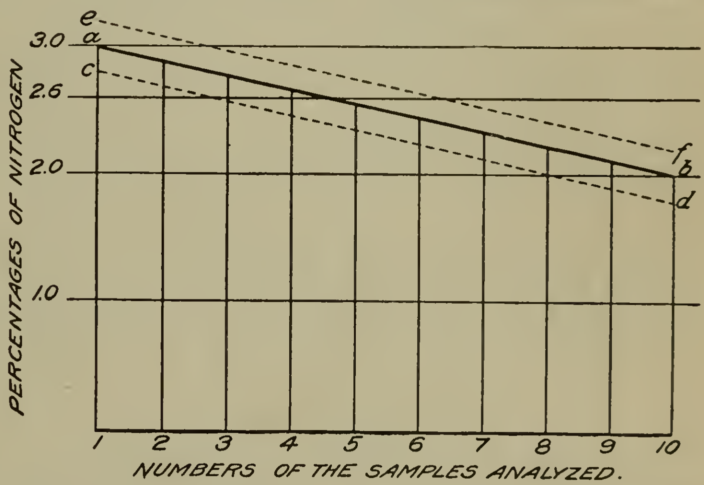

FIG. 9.—Diagram showing the method of selection for nitrogen content when the experimental error is known.

SUMMARY.

(1) Wheat plants growing under field conditions or nursery conditions show great variation in nitrogen content. This variation, however, does not seem to be inherited but is apparently due to local variation in environment and is therefore not capable of transmission.

(2) Centgeners, rows, and small plats vary almost as much as individual plants, owing to local variation in environment.

(3) The most practical way of overcoming this variation is by replicating the plats a sufficient number of times to reduce the error to less than one-half the real variation.

(4) To bring the experimental error within proper bounds, single plants should be repeated 40 or more times, 16-foot rows 5 to 10 times, and blocks 5.5 feet square 8 to 16 times. No data are given for centgeners, but the variation in centgeners is about the same as in the blocks.

(5) In order to eliminate the undesirable strains, the experimental error must be less than one-half the real or expected variation.

(6) The easiest and most practical method of growing strains to compare for nitrogen content is to plant in rows 12 to 16 feet in length and repeat 10 times in different parts of the field. Several series of check plats should also be inserted.

II.—EXPERIMENTAL ERROR IN THE NURSERY AND VARIATION IN YIELD

INTRODUCTION.

A very large share of cereal breeding to-day consists in the separation of pure strains from what we call our ordinary varieties of cereals. In dealing with new hybrids the separation involves the selection of the most desirable strains after types have been fixed. We are mainly concerned for the present in finding the best-yielding strains. This necessitates the finding of a method by which comparative field tests can be made rapidly in large numbers.

The method of comparing strains in "centgeners" first came into general use some 10 years ago, and later the "row" method was evolved. At present the Nebraska Agricultural Experiment Station is experimenting with a small block similar in size to the centgener, but sown at the ordinary rate of seeding.

A number of sources of error in all these methods are due to unexpected variations in soil and climate. It is the purpose of this paper to discuss some of these sources of error and to suggest methods of correction.

VARIATION IN YIELD FROM CHECK ROWS.

In our row-breeding work we use every fifth row as a check plat. All check plats are from the same seed and sown in the same way. A great variation is found in these check rows even when conditions appear quite uniform. Table XII shows the yield of 47 consecutive check rows in one of the 1909 series. These rows were 14 feet long and each was hand planted with 400 seeds of Turkey wheat. They were not quite as uniform as we sometimes have them, owing to dry weather in the spring, although the appearance of the plats was uniform enough at harvest time. One object in presenting the data here, however, is to illustrate the effect of repeating check plats on correction of error. All things being equal, the yields of the 47 plats should have been the same. But all factors can never be equal, so in row-breeding work, owing to unequal environment, we must expect a wide degree of error. The only practical way so far suggested to overcome this error is to repeat the plats, according to some systematic method, enough times to equalize variations in soil or climatic effects. If the plats are repeated only a few times there is still danger of a large error due to the chance combination of plats unduly high or low. This is illustrated by data given in the lower half of Table XII, where all the above check rows have been brought together in groups of six each (except group *h*), taking every eighth plat to form a group, to show the chance of error in repeating a series of strains six times.

TABLE XII.—*Yield of 47 14-foot check plats of Turkey wheat in 1909.*

CHECK PLATS IN CONSECUTIVE ORDER.

Grams.	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Grams.*
205	239	162	165	182	205	216	230
170	240	204	279	265	270	278	310
202	167	200	238	285	265	222	238
247	216	225	284	255	265	279	155
304	272	226	237	245	150	268	186
209	310	278	347	168	306	239	

CHECK PLATS SYSTEMATICALLY REPEATED TO FORM EIGHT GROUPS.

Group *a*.	Group *b*.	Group *c*.	Group *d*.	Group *e*.	Group *f*.	Group *g*.	Group *h*.
Grams.	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Grams.*
205	170	202	247	304	209	239	240
167	216	272	310	162	204	200	225
226	278	165	280	238	284	237	347
182	265	285	255	245	168	205	270
265	265	150	306	216	278	222	280
268	239	230	310	238	155	186	....
219	239	217	285	234	216	215	272

Groups arranged in order of average yield of included plats: *g*, 215; *f*, 216; *c*, 217; *a*, 219; *e*, 234; *b*, 239; *h*, 272; *d*, 285. Average, 237.

Groups *a* and *d* illustrate the chance grouping of high-yielding and low-yielding plats. With only 5 plats out of the 47 yielding above 300 grams, three of them by chance fell into group *d* while a number below normal fell into group *a*. The average yields of the 8 groups varied from 215 grams to 285 grams. This variation indicates the amount of experimental error to be expected under similar conditions. Since the experimental error is larger than the real variation expected in different strains of wheat, results obtained under such circumstances would not be reliable. Until our system of testing will show actual differences of 10 per cent in yield, it can not be of much value in comparing the yielding value of different strains.

VARIATION IN YIELD FROM REPEATED ROWS.

In order to make a more careful test of the accuracy of replicating row tests, 100 rows of Kherson oats were planted, each row being 12.5 feet in length and containing 500 seeds. The plat chosen for this test was quite uniform and the appearance of the plat at harvest was very satisfactory. Three-fourths of the rows yielded from 225 to 275 grams each, with a few much higher and a few very low. Table XIII shows these rows arranged in consecutive order as they were in the nursery, with the yield of each row and the average yield of groups of five adjacent rows.

We may get some idea of how replication corrects error by assembling these rows into series as in the comparative test of strains. These data are shown and also summarized in Table XIII. In series 1 the yields of 5 adjacent rows are averaged (as rows 1 to 5, 6 to 10, etc.). In series 2 every twentieth row is taken (as rows 1–21–41–61–81, etc.) and averaged. In series 3 every tenth row is taken, making 10 repetitions; and in series 4 every fifth row, making 20 repetitions.

TABLE XIII.—*Yield of thrashed grain from 100 rows of Kherson oats.*

BY ROWS AND GROUPS OF FIVE ADJACENT ROWS IN CONSECUTIVE ORDER.

Row No.	Yield of grain.		Row No.	Yield of grain.		Row No.	Yield of grain.		Row No.	Yield of grain.	
	Actual.	Mean of 5 rows.		Actual.	Mean of 5 rows.		Actual.	Mean of 5 rows.		Actual.	Mean of 5 rows.
	Grams.	*Grams.*		*Grams.*	*Grams.*		*Grams.*	*Grams.*		*Grams.*	*Grams.*
1	228		26	252		51	244		76	233	
2	273		27	248		52	261		77	247	
3	255	236	28	206	246	53	258	250	78	211	225
4	224		29	280		54	244		79	237	
5	204		30	247		55	245		80	200	
6	205		31	257		56	248		81	248	
7	230		32	240		57	229		82	200	
8	260	224	33	233	235	58	211	221	83	227	222
9	195		34	230		59	227		84	220	
10	233		35	217		60	188		85	214	
11	257		36	226		61	248		86	184	
12	243		37	190		62	257		87	233	
13	250	248	38	222	220	63	270	263	88	244	234
14	230		39	261		64	264		89	241	
15	261		40	200		65	275		90	270	
16	235		41	246		66	300		91	241	
17	273		42	249		67	151		92	174	
18	273	255	43	276	254	68	153	216	93	247	242
19	263		44	237		69	205		94	266	
20	230		45	261		70	272		95	281	
21	244		46	267		71	194		96	240	
22	245		47	259		72	274		97	232	
23	209	234	48	252	249	73	242	235	98	228	247
24	247		49	251		74	235		99	290	
25	224		50	218		75	229		100	245	

SYSTEMATICALLY GROUPED IN VARIOUS WAYS TO SHOW EFFECT OF REPLICATION ON REDUCTION OF VARIATION IN YIELD.

Series 1.—Groups of 5 adjacent rows.				Series 2.—Groups of 5 rows, taking every 20th row.				Series 3.—Groups of 10 rows, taking every 10th row.		Series 4.—Groups of 20 rows, taking every 5th row.	
Yield.	Deviation.	Yield.	Deviation.	Yield.	Deviation.	Yield.	Deviation.	Yield.	Deviation.	Yield.	Deviation.
Grams.	*Grams.*	*Grams.*	*Grams*	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Grams.*
216	−21.8	236	− 1.8	212	−25.8	239	+ 1.2	226	−11.8	234	−3.8
220	−17.8	242	+ 4.2	223	−14.8	231	+ 3.2	229	− 8.8	235	−2.8
221	−16.8	246	+ 8.2	224	−13.8	242	+ 4.2	230	− 7.8	238	+ .2
222	−15.8	247	+ 9.2	229	− 8.8	242	+ 4.2	237	− .8	241	+3.2
224	−13.8	248	+10.2	234	− 3.8	243	+ 5.2	238	+ .2	241	+3.2
225	−12.8	249	+11.2	234	− 3.8	246	+ 8.2	239	+ 1.2		
234	− 3.8	250	+12.2	234	− 3.8	246	+ 8.2	241	+ 3.2		
234	− 3.8	254	+16.2	236	− 1.8	248	+10.2	243	+ 5.2		
235	− 2.8	255	+17.2	237	− .8	245	+17.2	245	+ 7.2		
235	− 2.8	263	+25.2	238	+ .2	256	+18.2	250	+12.2		
Average...		237.8	11.4	....	....	237.8	7.9	237.8	5.8	237.8	2.6

TABLE XIII.—*Yield of thrashed grain from 100 rows of Kherson oats*—Continued.

SUMMARY SHOWING COMPARISON OF GROUPINGS.

Number of rows in a group.	Average deviation from mean yield when rows are—		Coefficient of variability when rows are—		Variation of extremes in yield when rows are—	
	Adjacent.	Distributed.	Adjacent.	Distributed.	Adjacent.	Distributed.
	Grams.	*Grams.*	*Per cent.*	*Per cent.*	*Grams.*	*Grams.*
1	21.26	21.26	11.5	11.5	151 to 300	151 to 300
5	11.38	7.87	5.50	4.8	216 to 263	212 to 256
10	7.60	5.84	3.66	3.0	227 to 251	226 to 250
20	3.46	2.60	1.56	1.7	234 to 243	234 to 241

There is less variation when the five rows are distributed throughout the plat than when adjacent; also the variation decreases as the number of repetitions increases. Evidently in the case under consideration it would be necessary to repeat about 20 times in order to obtain comparable data.

To secure more data on this point, 500 row plats of Turkey wheat were planted in the autumn of 1909. The rows were each 16 feet in length, all planted uniformly and with the same seed in soil of average fertility and in good tilth. At harvest the rows appeared to be uniform in character and would have yielded about 30 bushels to the acre. Table XIV summarizes the results.

TABLE XIV.—*Yield of grain from 500 16-foot rows of Turkey wheat, systematically repeated in various ways to show experimental error.*

Classification.	Number of groups.	Mean yield.	Extreme variation.	Average deviation.	Standard deviation.	Coefficient of variability.
		Grams.	*Grams.*	*Grams.*	*Grams.*	*Per cent.*
Single rows	500	250.7	156 –403	28.67	35.85	14.34
Every 100th row, 5 rows in a group	a 25	242.0	219.6–277.8	13.3	15.76	6.51
	b 25	256.0	233.6–281.4	11.7	14.11	5.51
	c 25	247.3	226.4–272.6	11.9	13.03	5.27
	d 25	256.6	228.4–282.4	11.8	14.85	5.79
Average		250.5	227.0–278.5	12.17	14.44	5.77
Every 50th row, 10 rows in a group	a 25	245.1	224.1–270.7	10	12.18	4.97
	b 25	256.3	242.0–276.6	8	9.69	3.78
Average		250.7	233.0–273.6	9	10.93	4.37
Every 33d row, 15 rows in a group	33	250.2	235.5–273.9	6.2	7.95	3.18
Every 25th row, 20 rows in a group	25	250.7	234.3–269.6	7.4	9.28	3.70

In this case the experimental error was much higher than with the 100 rows of oats (Table XIII) and did not reduce so rapidly by repetition. This may be due to the fact that the 500 rows of wheat covered a greater area than the 100 rows of oats, thus having more causes for variation. It is probable that the greater the number of strains to be compared the more replications will be necessary because of the larger area they will cover. However, when repeated 15 times, the average deviation is about half as much as when repeated 5 times, but replicating 20 times did not make a further improvement. It will be seen that the extreme variation is rather wide. It would be impossible to make a direct comparison of yield between strains or varieties tested under conditions where the variation of extremes is so large. It would be necessary in such case to select a rather large percentage of the high-yielding varieties and continue the test with them for some years. The method of selection will be hereafter explained.

WEST

671 209	657 210	703 211	755 212	760 213	686 214	592 215	739 216	732 217	710 218	753 219	680 220	680 221	677 222	795 223	723 224
658 193	713 194	613 195	632 196	667 197	645 108	660 199	768 200	786 201	768 202	666 203	843 204	795 205	763 206	716 207	741 208
657 177	671 178	623 179	715 180	543 181	613 182	640 183	798 184	759 185	764 186	995 187	793 188	936 189	755 190	792 191	838 192
642 161	680 162	654 163	673 164	760 165	709 166	682 167	724 168	774 169	860 170	787 171	725 172	664 173	851 174	690 175	770 176
735 145	580 146	620 147	675 148	765 149	742 150	772 151	698 152	652 153	661 154	768 155	777 156	745 157	768 158	851 159	719 160
575 129	598 130	705 131	642 132	704 133	643 134	650 135	572 136	752 137	740 138	863 139	680 140	722 141	723 142	703 143	756 144
727 113	633 114	615 115	685 116	662 117	639 118	657 119	608 120	620 121	624 122	745 123	764 124	703 125	752 126	788 127	682 128
572 97	373 98	560 99	645 100	692 101	644 102	632 103	574 104	606 105	648 106	806 107	791 108	629 109	650 110	679 111	588 112
580 81	425 82	732 83	730 84	706 85	732 86	736 87	655 88	673 89	793 90	765 91	576 92	609 93	568 94	728 95	620 96
588 65	526 66	596 67	777 68	776 69	779 70	721 71	728 72	604 73	742 74	665 75	621 76	611 77	623 78	646 79	617 80
617 49	683 50	726 51	835 52	668 53	664 54	691 55	770 56	775 57	685 58	723 59	583 60	580 61	395 62	511 63	653 64
602 33	662 34	640 35	700 36	650 37	655 38	563 39	600 40	730 41	690 42	713 43	530 44	568 45	410 46	478 47	636 48
665 17	736 18	630 19	598 20	895 21	592 22	593 23	659 24	718 25	705 26	667 27	585 28	560 29	655 30	633 31	733 32
609 1	706 2	790 3	678 4	695 5	715 6	622 7	658 8	597 9	632 10	713 11	585 12	657 13	495 14	618 15	652 16

EAST

FIG. 10.—Diagram of plats of Turkey wheat, showing the arrangement of 224 blocks (each 5.5 feet square) and the yield of grain (in grams) for each block.

VARIATION IN YIELD FROM SMALL BLOCKS.

For this experiment 224 blocks, each 5.5 feet square, were laid out in a plat 14 blocks one way by 16 the other. The blocks were all sown to Turkey winter wheat in the fall of 1908. To sow these the drill, 5.5 feet wide, was driven straight across the first series of 14 blocks, the boundaries of the blocks being later established. Each series was sown in the same way and no path or space was allowed between the blocks. Figure 10 is a diagram of the blocks, the yield in grams being marked in each block. A large variation in yield was found in the different blocks, amounting to 100 per cent in the extreme cases, although the appearance at harvest time was fairly uniform. These plats have been combined in various ways in order to study the yields. The results are shown in Table XV.

EFFECT OF REPETITION IN REDUCING ERROR.

The first part of Table XV shows the 224 blocks combined in various ways, namely, 56 groups of 4 blocks each when every fifty-sixth block was taken, 28 groups of 8 blocks each when every twenty-eighth block was taken, and 14 groups of 16 blocks each when every fourteenth block was taken. The 56 groups of 4 blocks each were divided into 4 sets of 14 each and the 28 groups of 8 into 2 sets of 14 each. This division into sets was for the purpose of having an equal number of blocks for comparison in each case. We find illustrations of a combination of high-yielding plats and low-yielding plats such as were noted in Table XII. For example,

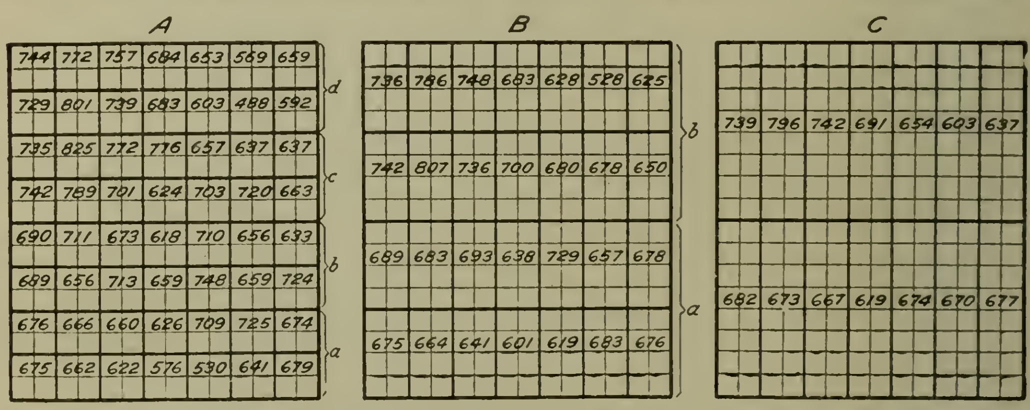

FIG. 11.—Diagrams of plats of Turkey wheat, showing the arrangement of 224 blocks (each 5.5 feet square) when combined in groups of adjacent blocks, with the average yield for each group: *A*, Groups of 4; *B*, groups of 8; *C*, groups of 16.

when the blocks are repeated 4 times the range in yield varies from an average of 595.2 grams per block to an average of 786.5 grams per block, a difference of 32 per cent. If varieties were being tested by the same system, this variation would be more than we might expect in the yields of different kinds.

When the blocks are repeated 8 times the average variation ranges from 627.5 to 717.8 grams, a difference of 90 grams, or 14 per cent. By repeating 16 times the extreme variation is reduced to 47 grams, or 7 per cent. However, with the exception of the extremes, the variation here is small. The question now to consider is the minimum number of blocks which will insure comparable results. If comparable results are to be secured the first season, the blocks should be repeated 15 to 20 times. If it were desirable to carry the strains for a period of 3 years, repeating 8 to 10 times would probably be sufficient, since this would give a total of 24 to 30 blocks for the 3 years.

RELATION OF SIZE OF PLAT TO VARIATION.

It is very desirable in plant-breeding work to determine the minimum size of plat that it is practicable to use, since with hundreds of strains to try each year it would be impossible to handle them in large plats. Taking the above series of 224 small blocks, the adjacent blocks could be combined to give a continuous series of larger and larger blocks. Figure 11 shows how these combinations were made and Table XV gives statistical results, showing also in a summary for two years the comparative effect of increasing the size of the block and of repeating small blocks.

TABLE XV.—*Yield of Turkey wheat grown in 224 block plats (each 5.5 feet square) in 1909 and 1910.*

SYSTEMATICALLY REPEATED TO FORM GROUPS OF 4, 8, AND 16 BLOCKS.

Classification.	Number in group.	Season of 1909. Mean yield per block.	Season of 1909. Extreme variations.	Season of 1909. Average deviation.	Season of 1909. Standard deviation.	Season of 1909. Coefficient of variability.	Season of 1910. Mean yield per block.	Season of 1910. Extreme variations.	Season of 1910. Average deviation.	Season of 1910. Standard deviation.	Season of 1910. Coefficient of variability.
						P. ct.					*P. ct.*
Single blocks	224	680.38	373 –995	67.23	81.98	13	463.71	300 –809	47.62	62.62	13.50
Every 56th block, 4 blocks in a group.	a 14	668.82	613.25–721.25	29.33	35.79	5.35	468.23	441.75–502.75	15.80	17.88	3.82
	b 14	689.96	645.75–786.50	32.99	39.80	5.77	467.09	412.25–504.25	15.36	21.87	4.68
	c 14	680.95	595.25–728.00	21.71	30.09	4.42	450.59	397.50–490.50	21.52	26.34	5.84
	d 14	683.36	627 –744.75	24.36	32.09	4.69	468.93	403.75–581.50	35.12	44.23	9.43
Average	......	680.77	620.32–745.12	27.09	34.44	5.05	463.71	413.81–515.80	21.95	27.58	5.94
Every 28th block, 8 blocks in a group.	a 14	674.20	627.50–704.88	18.04	21.63	3.21	459.41	441.38–472.50	8.46	9.65	2.10
	b 14	686.66	660.38–717.88	16.45	18.62	2.71	468.01	433.38–515.38	15.28	19.34	4.13
Average	......	680.43	643.94–211.38	17.25	20.13	2.96	463.71	437.38–493.94	11.87	14.43	3.11
Every 14th block, 16 blocks in a group.	14	680.43	653.25–700.38	7.24	10.41	1.53	463.72	450.86–490.56	6.47	9.60	2.07

COMBINED IN GROUPS OF 4, 8, AND 16 ADJACENT BLOCKS, TO SHOW THE EFFECT OF SIZE OF PLAT ON VARIABILITY.

Classification.	Number in group.	Season of 1909. Mean yield per block.	Season of 1909. Extreme variations.	Season of 1909. Average deviation.	Season of 1909. Standard deviation.	Season of 1909. Coefficient of variability.	Season of 1910. Mean yield per block.	Season of 1910. Extreme variations.	Season of 1910. Average deviation.	Season of 1910. Standard deviation.	Season of 1910. Coefficient of variability.
Four adjacent blocks in a group.	a 14	652	529.75–725.25	37.36	49.08	7.53	443.18	387.25–497	26.76	30.93	6.98
	b 14	681.48	617.75–748.25	30.84	35.75	5.25	465.30	389.50–576	45.62	33.86	11.57
	c 14	713.61	624.50–825	53.14	61.46	8.47	504.64	413.75–587.50	31.84	41.90	8.30
	d 14	676.78	488.25–801.50	70.96	85.80	12.67	441.71	386.75–493.25	26.37	31.49	7.13
Average	......	680.97	565.06–775.00	48.07	58.02	8.48	463.71	394.31–538.44	32.65	39.34	8.49
Eight adjacent blocks in a group.	a 14	666	636.13–829	24.57	31.57	4.74	454.21	398.62–510.25	27.86	37.97	8.36
	b 14	695	586.50–706.75	55.93	70.30	10.11	473.18	894.12–545	31.55	40.01	8.45
Average	......	680	611.31–767.87	40.25	50.93	7.42	463.69	396.37–527.62	29.70	38.99	8.40
Sixteen adjacent blocks in a group.	14	680	603.75–797.06	35.43	48.87	7.20	463.71	406.69–509.38	21.67	26.43	5.70

TABLE XV.—*Yield of Turkey wheat grown in 224 block plats (each 5.5 feet square) in 1909 and 1910*—Continued.

SUMMARY BASED ON THE AVERAGE OF BOTH SEASONS, SHOWING THE EFFECT ON COEFFICIENT OF VARIABILITY OF INCREASING THE SIZE OF PLAT AS COMPARED WITH DISTRIBUTING THE SAME AREA BY A SYSTEMATIC METHOD.

Number of blocks combined in each group.	Number of groups averaged.	Coefficient of variability.		
		Plats increased in size, blocks adjacent.	Plats distributed systematically.	Four adjacent blocks combined and combination distributed 4 times=16 blocks.
1	224	13.25	13.25	3.42
4	56	8.48	5.44	
8	28	7.91	3.03	
16	14	6.45	1.80	

SUMMARY BASED ON THE YIELDS OF 1909, ARRANGED TO SHOW THE RELATION BETWEEN SIZE OF PLAT AND AVERAGE DEVIATION.

Shape of plat.	Number of blocks in plat.	Total number of plats.	Average deviation.	Average deviation when plats are made up by systematic method.	
				Every—	Per cent.
			Per cent.		
1	1	224	9.76	....	....
1 by 2	2	112	8.57	....	....
1 by 4	4	56	7.16	56th block	5.05
2 by 2	4	56	7.38		
2 by 4	8	28	6.05	28th block	2.96
1 by 8	8	28	6.08		
2 by 8	16	14	5.30	14th block	1.53
4 by 8	28	8	5.37	....	....
7 by 8	56	4	5.29	....	....

Starting with a coefficient of variability of 13.25 per cent, it is decreased to 8.48 when the block is made 4 times as large, to 7.91 per cent when 8 times as large, and to 6.45 per cent when increased 16 times in size. Table XV gives the result of repeating the same number of plats equal distances apart. Here we see that where the plats are repeated 16 times the average variability for the two years rapidly decreases to 1.8 per cent.

It might appear from a study of the first part of this table that if the size of the plat were constantly increased the variability would be constantly reduced. However, increasing the size of the plat beyond a certain point does not continue to remove the cause of variability, namely, variation in soil. The last part of the table, which contains the data for 1909, is arranged to show the effect of increasing the size of the plat. It indicates a rapid decrease in variability up to plats 16 blocks in size, but no decrease in the next two cases. While acre plats are probably less variable than tenth-acre plats and tenth-acre plats less variable than hundredth-acre plats, yet plats of this size are too variable for direct comparison and they are much too large for practical plant-breeding work. On the other hand, repeating the plats in a systematic way constantly removes the cause of variation as the number of repetitions increases. It then appears that the most practical method of removing error is to repeat series of small blocks a rather large number of times.

CONSTANCY OF VARIATION ON THE SAME PLATS.

Table XV gives statistical data for 1909 and 1910 on the same blocks arranged in the same way both years. Figure 12 shows the average yield of each section and the average percentage of nitrogen in the two years for sections *a*, *b*, *c*, and *d*. The yield per block varied about the same for the two years, being highest in section *c* in both seasons. The variation in nitrogen was not as regular, section *c* being highest in nitrogen in 1909, while in 1910 there was a small but regular increase in nitrogen from *d* to *a*. The second part of figure 12 shows the coefficient of variability in both yield and percentage of nitrogen when the small blocks are combined in sets of four (Table XV). Section *a* was highest in variability of both yield and nitrogen content in 1909 and low in 1910. Section *b* was low in variation in yield in 1909 and high in 1910. This would indicate that different seasons do not affect equally all parts of the plat, and illustrates the difficulty of "standardizing" plats by the system of sowing all plats to one crop for a season in order to determine relative yield.

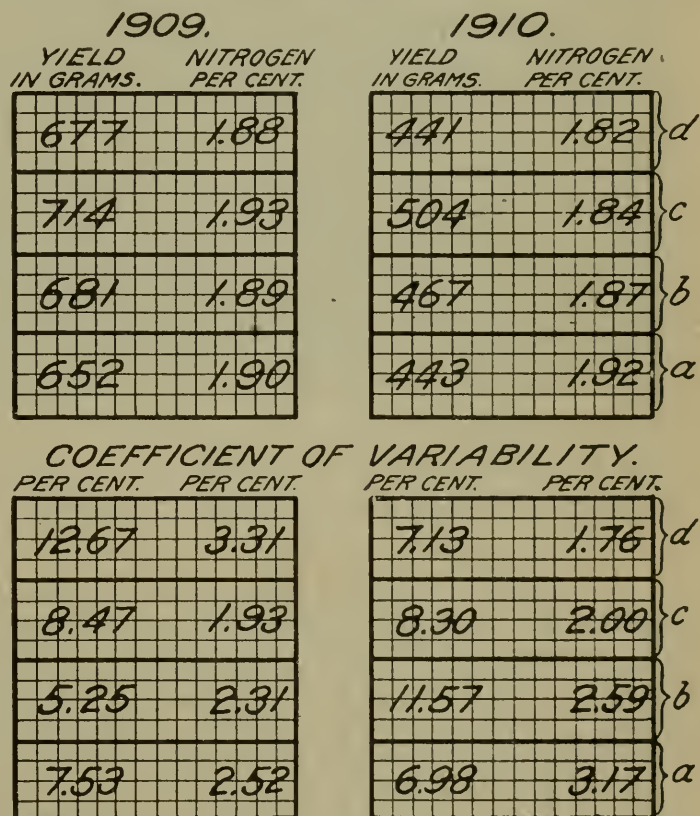

FIG. 12.—Diagrams showing Turkey wheat grown in 224 blocks, combined in four groups (Table XV, *a*, *b*, *c*, *d*) of 56 adjacent blocks to show variations in yield and nitrogen content in 1909 and 1910.

VARIATION IN YIELDS FROM CENTGENER PLATS.

The centgener method consists of planting 100 plants in a centgener, 6 inches apart each way, making blocks 5 feet square. In 1908, 178 centgener blocks were compared for variability with an equal number of duplicate row plats 16 feet in length. The variability was found to be practically the same. Other data confirm this conclusion, although under unfavorable conditions centgener plats are quite variable, owing to the fact that the individual plants are so far apart that the missing plants are not compensated for by the tillering of neighbors, as is the case where the planting is at the normal rate.

ALTERNATING CHECK ROWS AS A MEANS OF OBTAINING COMPARATIVE YIELDS.

In order to test the value of the method of alternating check rows, the 500 rows before referred to (pp. 36–37) were used as a basis for data. It was assumed that every odd-numbered row would represent a check row, while every even-numbered row would represent a strain being compared with a check row. Thus, row No. 2 would be considered a strain to compare with rows 1 and 3 as checks. It is apparent from data heretofore presented that the error would be too great if only a single row were compared with its adjacent checks. For example, there are numerous cases in the 500-row plats where the even-numbered row would be 20 to 30 per cent higher or lower in yield than the average of the adjacent odd-numbered row plats. Table XVI shows the result of averaging five odd rows and the adjacent five even rows, i. e., rows 1, 3, 5, 7, and 9 are averaged to compare with rows 2, 4, 6, 8, and 10. In the first 10 rows, for example, the five odd rows averaged 235.6 grams per row and the five even rows 226.4 grams per row, or 9.2 grams less than their checks. In the next block the even rows yielded 9 grams more than the checks. Out of the 50 cases here cited the extremes vary from −26.6 to +32.8, with an average deviation of 10.14 grams or 4 per cent. In selective work it is the unusually high variants that are sought after, but with an experimental error greater than the expected variation they would be difficult to locate. Table XVI also shows the result of dividing the 500 rows into blocks of 20 and 40 rows and comparing the yield of odd and even rows in each case. While in most cases the average deviation is small, yet there are a number of quite wide variations. For example, when 50 series of five odd rows are compared with five even rows, 17 series, or about one-third, show a deviation greater than 5 per cent of the mean; when 25 series of 10 odd rows are compared with 10 even rows, 6 series, or about one-fourth, show greater than 5 per cent average deviation; and when 12 series of 20 odd rows are compared with 20 even rows, none show 5 per cent deviation.

EFFECT OF INCREASING LENGTH OF ROW.

To increase the length of the row will decrease the error in about the same way as to increase the size of the block. In the 500-row plats just discussed most of the rows were in series and end to end,

FIG. 1.—HEAD-TO-ROW NURSERY, IN WHICH 25 GRAINS FROM A SINGLE HEAD ARE PLANTED IN A ROW 20 INCHES LONG.

The second year the seed from each 20-inch row is planted in a 16-foot row.

FIG. 2.—ROW-PLAT NURSERY, IN WHICH THE ROWS ARE 16 FEET IN LENGTH WITH A 4-FOOT ALLEY ADJACENT, THUS MAKING THE BEDS 20 FEET IN WIDTH.

These beds are slightly rounded, to give perfect drainage.

FIG. 1.—INCREASE PLATS OF ONE-THIRTIETH ACRE EACH.
Selected strains from the nursery are tested in these plats for 3 years.

FIG. 2.—INCREASE PLATS HARVESTED AND READY TO THRASH.
The plats in this field averaged 60 bushels per acre.

with only a narrow alley a few inches in width between the ends. By adding together the rows end to end, longer rows could be made. (Pl. I.) A total of 84 rows 64 feet in length was made in this manner, and the yields calculated. Table XVI gives the variability in the original 500 rows, each 16 feet long, in comparison with the same rows when combined into lengths of 64 feet. By increasing the length four times the deviation and variability are reduced not quite one-half. The longer rows are also less variable than blocks of five adjacent 16-foot rows, but more variable than five rows distributed in a systematic way throughout the plat.

The best length of row to use must be determined by circumstances. If sufficient uniform land is available and it is more convenient to make long rows, to do so would lessen the number of repetitions of plats necessary to reduce the error within proper limits, but it would always take a larger area to secure the same degree of accuracy with the long rows.

TABLE XVI.—*Yield, in grams, of Turkey wheat grown during the season of 1910 in 500 rows, each 16 feet in length.*

ARRANGED BY ODD AND EVEN ROWS AND AVERAGED IN GROUPS OF TEN.[1]

Rows 1 to 100.			Rows 101 to 200.			Rows 201 to 300.			Rows 301 to 400.			Rows 401 to 500.		
Odd.	Even.	Difference.	Odd.	Even.	Difference.	Odd.	Even.	Difference.	Odd.	Even.	Difference.	Odd.	Even.	Difference.
205	212		246	244		258	253		259	292		278	230	
226	222		235	298		233	241		317	340		278	288	
245	231		230	245		262	219		314	285		277	280	
262	238		248	226		182	220		223	219		307	300	
240	229		266	231		246	239		238	262		283	295	
235.6	226.4	−9.2	245	248.8	3.8	236.2	234.4	−1.8	270.2	279.6	9.4	284.6	278.6	−6
180	183		228	226		224	218		198	223		268	250	
214	255		242	266		222	261		251	287		258	250	
256	220		220	227		256	234		223	204		253	250	
216	234		213	235		229	241		209	223		242	239	
237	256		225	245		292	198		229	194		189	287	
220.6	229.6	9	225.6	239.8	14.2	244.6	230.4	−14.2	222	226.2	4.2	242	255.2	13.2
214	231		271	212		235	229		216	216		284	276	
235	208		235	238		235	271		222	204		200	217	
284	316		247	227		235	212		245	236		316	285	
275	240		247	255		221	242		231	194		194	243	
272	228		267	296		273	269		200	218		363	343	
256	244.6	−11.4	253.4	245.6	−7.8	239.8	244.6	4.8	222.8	213.6	−9.2	271.4	272.8	1.4
272	239		269	259		219	255		228	200		248	294	
275	215		250	289		270	280		227	228		380	338	
224	237		278	246		275	191		218	208		326	287	
220	240		222	256		282	249		242	257		265	315	
182	263		308	276		268	237		221	230		314	290	
234.6	238.8	4.2	265.4	265.2	−.2	262.8	242.4	−20.4	227.2	224.6	−2.6	306.6	304.8	−1.8
156	252		297	280		213	246		215	252		328	273	
227	263		230	310		255	269		269	210		324	315	
254	224		323	263		235	242		171	228		300	279	
226	188		317	261		247	268		230	207		252	249	
246	255		305	268		303	242		222	200		331	273	
221.8	236.4	14.6	294.4	276.4	−18	250.6	253.4	2.8	221.4	219.4	−2	307	277.8	−29.2

[1] Average difference in yield between odd and even rows, grams, 10.4; per cent, 4.

TABLE XVI.—*Yield, in grams, of Turkey wheat grown during the season of 1910 in 500 rows, each 16 feet in length*—Continued.

ARRANGED BY ODD AND EVEN ROWS AND AVERAGED IN GROUPS OF TEN—Continued.

Rows 1 to 100.			Rows 101 to 200.			Rows 201 to 300.			Rows 301 to 400.			Rows 401 to 500.		
Odd.	Even.	Difference.	Odd.	Even.	Difference.	Odd.	Even.	Difference.	Odd.	Even.	Difference.	Odd.	Even.	Difference.
227	247		275	249		257	225		282	246		224	303	
230	285		241	269		190	204		269	240		349	320	
263	281		285	245		227	250		250	230		272	283	
255	255		249	222		222	174		233	268		291	331	
280	280		246	223		208	216		213	200		208	271	
251	269.6	18.6	259.2	241.6	−17.6	220.8	213.8	− 7	249.4	236.8	−12.6	268.8	301.6	32.8
275	237		232	228		229	187		200	207		269	293	
264	277		200	228		200	215		219	233		293	327	
268	227		282	242		238	238		225	207		257	263	
204	218		227	239		242	202		205	230		254	244	
237	202		246	222		190	245		243	216		250	311	
249.6	232.2	−17.4	237.4	231.8	− 5.6	223.8	217.4	− 6.4	218.4	218.6	.2	264.6	287.6	23
235	229		266	287		218	212		250	208		286	243	
204	210		351	312		212	184		276	254		297	403	
257	312		265	300		234	267		235	245		316	285	
294	295		322	313		211	213		243	255		297	239	
313	315		293	285		232	284		257	281		211	247	
260.6	272.2	11.6	299.4	299.4	0	221.4	232	10.6	252.2	248.6	− 3.6	281.4	283.4	2
233	265		248	273		230	219		298	316		208	219	
276	214		233	243		236	228		261	208		255	255	
327	300		280	261		290	314		225	248		243	289	
312	316		259	263		208	195		233	226		247	275	
370	290		248	272		225	219		205	214		249	266	
303.6	277	−26.6	253.6	262.4	8.8	237.8	235	− 2.8	244.4	242.4	− 2	240.4	260.8	20.4
271	236		241	250		232	211		250	230		217	215	
251	246		266	254		185	257		254	211		235	285	
269	251		310	263		242	242		205	269		252	235	
217	242		245	245		279	270		237	216		222	289	
298	308		275	269		265	270		242	192		271	288	
261.2	256.6	− 4.6	267.4	256.2	−11.2	240.6	250	9.4	237.6	223.6	−14	239.4	262.4	23

SUMMARY OF ODD AND EVEN ROWS ARRANGED IN BLOCKS OF 20 AND 40 ROWS TO SHOW DEVIATION.

The 10 odd and 10 even rows from each adjacent 20 grouped together.						The 20 odd and 20 even rows from each adjacent 40 grouped together.		
Odd.	Even.	Difference.	Odd.	Even.	Difference.	Odd.	Even.	Difference.
228.1	228.0	− 0.1	222.6	224.7	+ 2.1	236.7	234.8	− 1.9
245.3	241.7	− 3.6	239.2	242.5	+ 3.3	245.7	252.6	+ 6.9
236.4	253.0	+16.6	246.1	252.9	+ 6.8	258.8	255.5	− 3.3
255.1	252.2	− 2.9	225.0	219.1	− 5.9	268.1	257.2	−10.9
282.4	266.8	−15.6	235.4	228.1	− 7.3	264.4	263.9	− .5
235.3	244.3	+ 9.0	235.3	233.6	− 1.7	245.8	237.9	− 7.9
259.4	255.4	− 4.0	241.0	233.0	− 8.0	229.1	229.1	0
276.8	259.0	−17.8	263.3	266.9	+ 3.6	242.6	247.7	+ 5.1
268.4	268.6	+ .2	271.0	288.8	+17.8	230.2	223.6	− 6.6
260.5	259.3	− 1.2	287.9	289.7	+ 1.8	238.1	233.3	− 4.8
240.4	232.4	− 8.0	273.0	285.5	+12.5	267.1	277.8	+10.7
251.3	243.5	− 7.8	239.9	261.6	+21.7	280.4	287.6	+ 7.2
235.7	233.6	− 2.1						
			Average difference.		7.256	Average difference.		5.483

SUMMARY SHOWING RESULT OF COMBINING 16-FOOT ROWS IN VARIOUS WAYS.

Classification.	Extreme variation.	Standard deviation.	Coefficient of variability.	Classification.	Extreme variation.	Standard deviation.	Coefficient of variability.
			Per cent.				*Per cent.*
500 single rows 16 feet long	156–403	35.85	14.33	5 adjacent rows 16 feet long	212–317	26.11	10.38
84 rows 64 feet long (4 times length of above)	196–284	21.43	8.8	5 rows 16 feet long distributed (every 100 rows)	227–278	14.44	5.77

Table XVII shows a comparative summary, based on the data herein reviewed, from which it appears that to repeat the 5.5-foot square blocks, in series, will reduce the error at the greatest rate, while to repeat the 16-foot rows will give the next most rapid reduction. However, the method of alternating 5 odd rows with 5 even rows gave about as good results as to repeat 10 rows, and the system of alternate planting with check rows would sometimes be desirable.

Table XVII.—*Summary showing coefficients of variability under various systems of arranging block plats and row plats.*

Kind of plats and arrangement.	Number of plats combined.					Remarks.
	1	5	10	15	20	
Blocks 5.5 feet square (Table XV):						
Adjacent	13	8	7	6.4	....	Variation will not continue to decrease.
Repeated	13	5	2.5	1.8	....	Variation decreases to infinity.
Rows 16 feet long (Table XVI):						
Adjacent	14	11	7	5	3.5	Variation does not continue to decrease.
Repeated	14	5	4	3.2	3.7	Variation decreases to infinity.
Alternated	14	....	4	3.6	3	Do.

To correct by check plats is sometimes uncertain, as pointed out hereafter, and therefore the advantage seems to be with the method of systematic repetition of rows or blocks. The repetition of the square blocks 10 times gave a higher degree of accuracy than repeating rows even 15 and 20 times, and it seems probable that further experimenting is likely to show the block system to be the most accurate for close comparisons. It also does away with the competition which no doubt takes place between adjacent rows, and also makes note taking easier by giving a mass effect, such as is secured under field conditions. To increase the size of plat will also reduce error, as shown by Table XV, and on the basis of these data a plat, about three times the size of the 5.5-foot blocks reported on, is being tested. This block is 4.2 by 16 feet in size, and indications are that with this block the variation will be reduced about one-half, as compared with the 5.5-foot block.

INFLUENCE OF RATE OF PLANTING ON YIELD.

For several years in planting our row plats care was taken to plant the same number of seeds in each row. In case of wheat, 400 kernels usually were planted to the 16-foot row, which was equivalent to the normal rate of seeding, or 5 pecks per acre. However, in 1908 and 1909 counts of the number of plants harvested were made of many rows at harvest time. In the row plats of wheat where the rate of seeding was about normal it was found that only about 60 to 80 plants were harvested to every 100 kernels planted. A variation of 20 per cent in stand was not apparent to the observer because all plats would appear to have an equally good stand at harvest time. This loss of plants was due to many causes, such as winterkilling, insects, and accidents, but chief among the causes appeared to be the normal competition of plants. For example, certain plants were weak or were slow in starting spring growth; the stronger plants would quickly outdistance them, causing at least a certain percentage of the weaker plants to perish as a result of competition. It seemed doubtful, however, whether this difference in plants harvested had a marked effect on yield. For example, here and at many experiment stations tests have been made with sowing wheat at various rates, from 4 pecks to 10 pecks per acre. The difference in yield is never large. Doubling the amount of seed sown, from 4 to 8 pecks per acre, does not double the yield, and in many cases does not affect it at all. This is due to the tillering power of the plant, which is able in this way to compensate for the difference in number of plants.

To make a test of the rate of planting under row-plat conditions, a series of plats was planted with Red Rustproof oats in the spring of 1910. Each row plat was 16 feet in length. There were 5 plats in the series, planted at the rate of 400, 500, 600, 700, and 800 grains per plat. The series was repeated 20 times, making 100 rows in all. The results are shown in Table XVIII.

TABLE XVIII.—*Results of rate-of-seeding test on 100 16-foot rows of Red Rustproof oats.*

Statement of averages.	Number of grains sown per row.				
	400	500	600	700	800
Number of plats averaged	20	20	20	19	20
Average yield per row ... grams	197	213	215	215	234

The normal rate of seeding would have been about 600 grains per row. It appears from the data that a slight variation in rate of planting, as 25 or 50 grains more or less than normal, would not affect the results.

To further test the effect of rate of seeding, a series of 60 blocks was laid out, each block being 5 drill rows wide and 16 feet in length. As the drill rows were 10 inches apart, this made the plats each 4.2 by 16 feet. The blocks were planted with a small drill devised for the purpose. Five rates of seeding were used, namely, 42.6, 49.1, 55.9, 65.4, and 73.3 grams per block. The series was repeated 12 times, giving 12 blocks of each rate for averaging. The results are shown in Table XIX.

TABLE XIX.—*Results of rate-of-seeding test on 60 block plats of Kherson oats.*

Statement of averages.	Weight of seed sown (grams).				
	42.6	49.1	55.9	65.4	73.3
Number of blocks averaged	12	12	12	12	12
Average yield per block ....grams..	1,069	1,101	1,151	1,149	1,156

The normal rate of seeding would be about 60 grams per block, and it is apparent from the data that a variation of 10 or even 20 per cent above or below normal in the weight of seed used would not have a marked effect on the yield.

In view of the data just presented, it would seem not to be necessary to actually count the number of seeds to be planted in each small plat, providing some other quicker means can be found of obtaining approximate accuracy. If the seed is first carefully fanned, scoured, and screened to one size, equal volumes will usually not vary more than a small percentage in number of kernels. Also equal weights will have approximately the same number of kernels, provided the seed has first been carefully prepared by fanning and screening to a uniform size.

EFFECT OF COMPETITION BETWEEN ADJACENT ROWS.

In 1908 it was observed that a certain strain of early wheat in a series of row plats made a very poor appearance at harvest time, while the same strain planted in centgeners made a much better comparative showing. Apparently the larger and faster growing strains on each side, the rows being only 8 inches apart, exercised some competitive effect. This effect of competition has been noted for two years since. Also in certain variety tests of oats, grown in row plats 10 inches apart, the same effect was noted. Exact data can not be given on this point, as the results from the series of plats planted in 1909 and in 1910 for this purpose were seriously impaired by unfavorable conditions; but Table XVIII, giving results from adjacent row plats sown at different rates, shows that the 800-seed rate made a marked increase over the 700-seed rate, while in a similar series of blocks (Table XIX), sown at the same rate, this marked increase was not noted. Since the 800-seed row was always adjacent to the 400-seed row, it may have had some advantage on this account. Danger from this source can probably be avoided if care is taken to plant only similar varieties in adjacent rows. Where the block plat is used this source of error is eliminated.

VARIATION IN PURE STRAINS AND RELATION OF DATA IN CENTGENER NURSERY AND IN FIELD PLATS

ISOLATION OF PURE STRAINS.

In 1902, Dr. T. L. Lyon, now of Cornell University, planted 800 heads of Turkey winter wheat in a centgener nursery, arranging to keep a record of the progeny of each head. The heads were numbered from 1 to 800 and these original numbers are still retained as family numbers. The original plan was to select for increased nitrogen content and yield, discarding those families which did not show high averages in both these respects, and to practice continuous selection of individual plants from among those families that were promising. A considerable number of the families were discarded each year, until at the end of harvest in 1906 only 47 of the original 800 were retained for further work. At this time the writer took up the work and the practice of continuous selection was discontinued as it began to be apparent that the isolation of pure strains was a more promising way of obtaining results. The 47 pure strains were put in field plats to test for yield. Complete records, however, can be given on only 24 pure strains, as all but 26 were dropped in 1908 for lack of space, and the nursery data are incomplete on 2 strains out of the 26. These strains are shown in field plats in Plate II. Table XX gives the average results for four years, both in the centgener nursery and in the field plats. By "centgener" is meant the method of planting 100 seeds from a single plant in a square plat, the plants 6 inches apart each way.

TABLE XX.—*Relations of certain characters of 24 strains of Turkey wheat grown in nursery and in field and tested during 4-year periods.*

RANKED IN GROUPS OF FIVE IN ORDER OF YIELD IN THE FIELD.

Family No. (The numbers in italic indicate the five highest yielders.)	Data from centgener nursery (average for four years, 1906–1909).							Data from field plats (average for four years, 1907–1910).[1]		
	Nitrogen content.	Yield per— Plant.	Yield per— Centgener, 1906–1908.	Average weight of kernels, 1903–1906.	Strength of straw.	Fruiting period.	Number of centgeners averaged.	Nitrogen content.	Yield.	Number of plats averaged.
	P. ct.	*Grams.*	*Gms.*	*Grams.*	*P. ct.*	*Days.*		*P. ct.*	*Bush.*	
48	2.66	13.38	764	0.02192	80	36	22	2.60	40.75	9
287	2.52	12.38	704	.01864	81	35	23	2.49	40.59	12
42	2.68	11.58	622	.02105	63	35	23	2.48	39.90	5
312	2.61	12.27	646	.02344	68	36	31	2.55	39.21	9
425	2.70	9.83	612	.01824	67	35	35	2.56	39.18	8
Total or average	2.63	11.89	670	.02058	72	35	134	2.54	39.93	43
556	2.72	12.36	593	.02267	55	36	38	2.54	38.86	5
225	2.53	11.12	623	.01984	65	35	29	2.55	38.78	10
215	2.63	11.14	605	.02064	70	34	48	2.54	38.64	8
47	2.60	12.26	664	.02013	63	36	123	2.51	38.50	39
3	2.70	11.01	620	.02168	63	34	18	2.53	38.48	23
Total or average	2.64	11.58	621	.02099	63	35	256	2.53	38.65	85

[1] Averages of check plats: Nitrogen content, 2.61 per cent; yield, 35.18 bushels.

TABLE XX.—*Relations of certain characters of 24 strains of Turkey wheat grown in nursery and in field and tested during 4-year periods*—Continued.

RANKED IN GROUPS OF FIVE IN ORDER OF YIELD IN THE FIELD—Continued.

Family No. (The numbers in italic indicate the five highest yielders.)	Data from centgener nursery (average for four years, 1906–1909).							Data from field plats (average for four years, 1907–1910).		
	Nitrogen content.	Yield per— Plant.	Yield per— Centgener, 1906–1908.	Average weight of kernels, 1903–1906.	Strength of straw.	Fruiting period.	Number of centgeners averaged.	Nitrogen content.	Yield.	Number of plats averaged.
	P. ct.	*Grams.*	*Gms.*	*Grams.*	*P. ct.*	*Days.*		*P. ct.*	*Bush.*	
391	2.70	10.93	510	.02185	64	34	28	2.51	37.96	5
221	2.63	12.46	647	.02295	64	34	59	2.59	36.57	12
313	2.50	11.95	597	.02118	68	36	114	2.53	36.27	5
377	2.81	11.05	563	.02016	60	34	20	2.70	36.13	5
206	2.59	11.08	558	.01889	65	34	26	2.58	36.12	5
Total or average	2.65	11.49	575	.02111	64	34	247	2.58	36.61	32
314	2.68	9.70	560	.02020	66	34	52	2.63	35.22	8
168	2.63	10.70	512	.02247	63	34	23	2.60	34.84	8
526	2.76	10.03	614	.02029	66	34	18	2.73	34.20	5
379	2.84	10.79	620	.02057	62	24	20	2.71	33.56	5
216	2.52	12.11	594	.02108	66	35	43	2.48	33.46	8
Total or average	2.69	10.67	580	.02089	65	34	156	2.63	34.26	34
209	2.49	11.38	570	.01915	82	34	37	2.45	32.93	8
37	2.87	11.27	526	.02270	54	34	48	2.75	32.58	5
2	2.56	9.86	548	.02180	58	36	33	2.55	31.88	8
328	2.62	11.89	508	.02522	62	33	30	2.73	28.88	5
Total or average	2.63	11.10	538	.02228	64	34	148	2.62	31.56	26

SUMMARY OF RESULTS, ARRANGED IN GROUPS OF FIVE STRAINS AND RANKED IN VARIOUS WAYS.

CENTGENER TESTS.										
In order of nitrogen content:										
37, 379, 377, 526, 556	2.80	11.10	583	0.02128	59	34	144	2.69	35.07	25
425, 391, 3, 314, *42*	2.69	10.61	585	.02060	65	34	156	2.54	38.15	49
48, 221, 168, 215, 328	2.63	11.91	607	.02224	68	34	182	2.61	35.94	42
312, 47, 206, 2, 225	2.58	11.32	608	.02082	64	35	242	2.55	36.90	71
287, 216, 313, 209	2.51	11.95	616	.02001	74	35	217	2.49	35.81	33
In order of strength of straw:										
209, *287*, *48*, 215, *312*	2.58	12.11	658	.02076	76	35	161	2.53	38.42	46
313, *425*, 314, 526, 216	2.63	10.72	595	.02020	67	35	262	2.59	35.67	34
206, 225, 391, 221, 3	2.63	11.32	592	.02104	64	34	160	2.55	37.58	55
42, 168, 47, 379, 328	2.67	11.44	585	.02189	63	34	219	2.61	35.14	62
377, 2, 556, 37	2.74	11.13	558	.02183	57	35	139	2.63	34.86	23
In order of yield per plant:										
48, 221, *287*, 556, *312*	2.63	12.57	671	.02192	70	35	173	2.55	39.20	47
47, 216, 313, 328, *42*	2.58	11.96	597	.02173	64	35	333	2.55	35.40	62
209, 37, 215, 225, 206	2.62	11.20	576	.02024	67	34	188	2.57	35.81	36
377, 526, 3, 391, 379	2.76	10.76	585	.02091	63	34	104	2.64	36.07	43
168, 2, *425*, 314	2.64	10.02	558	.02068	63	35	143	2.58	35.28	32
In order of yield per centgener:										
48, *287*, 47, 221, *312*	2.60	12.55	685	.02041	71	35	258	2.55	39.12	81
225, *42*, 3, 379, 526	2.70	10.91	620	.02068	64	34	108	2.60	36.98	48
425, 215, 313, 216, 556	2.61	11.48	600	.02076	65	35	278	2.53	37.28	34
209, 377, 314, 206, 2	2.63	10.61	560	.02040	66	34	168	2.58	34.46	34
37, 168, 391, 328	2.70	11.20	514	.02306	61	34	129	2.65	33.56	23
FIELD-PLAT TESTS.										
In order of nitrogen content:										
37, 526, 328, 379, 377	2.78	11.01	566	.02179	61	34	136	2.72	33.07	25
314, *48*, 168, 221, 206	2.64	11.46	608	.02129	68	34	182	2.60	36.70	42
425, 2, 225, *312*, 556	2.62	11.09	604	.02120	63	36	166	2.55	37.58	40
215, 3, 313, 391, 47	2.63	11.46	604	.02120	66	35	331	2.52	37.97	80
287, *42*, 216, 209	2.55	11.86	622	.01998	73	35	126	2.47	36.72	33
In order of yield per acre:										
48, *287*, *42*, *312*, *425*	2.63	11.89	670	.02058	72	35	134	2.54	39.93	43
556, 225, 215, 47, 3	2.64	11.58	621	.02099	63	35	256	2.53	38.65	85
391, 221, 313, 377, 206	2.65	11.49	575	.02111	64	34	247	2.58	36.61	32
314, 168, 526, 379, 216	2.69	10.67	580	.02089	65	34	156	2.63	34.26	34
209, 37, 2, 328	2.63	11.10	538	.02228	64	34	148	2.62	31.56	26

To sum up, the 24 pure strains have varied in the centgeners from 2.49 to 2.87 in per cent of nitrogen, from 54 to 82 per cent in strength of straw, from 9.70 to 13.38 grams in yield per plant, and from 508 to 764 grams in yield per centgener. They also have shown a variation

FIG. 13.—Field plats of pure strains and check plats of original seed of Turkey wheat, 1910. The upper numerals are family numbers; the lower, 4-year average yields. Two of the poorest yielders out of 26 strains came adjacent to two of the best. The difference in yield would not have been suspected from the appearance of the plats.

FIG. 14.—Wheat nursery plats, showing variations in winterkilling. Pure strains were alternated with the original Turkey wheat from which the strains were isolated. The original was mostly winterkilled while many of the select strains withstood the winter well.

in average weight of kernel ranging from 0.01824 to 0.02522 gram. In the field plats the percentage of nitrogen varied from 2.45 to 2.75 and the yield per acre from 28.8 to 40.7 bushels—a difference of about 12 bushels. Some of these plats and the check plats noted below are shown in figure 13. It is interesting to note that the check plats of original unselected Turkey winter wheat averaged 35.18 bushels per acre, or about halfway between the highest and lowest pure strains. More strains surpass the check in yield than fall below it, but this is probably because a large percentage of the poor-yielding strains were discarded after the first field test in 1907. It appears that neither the original selection of the 800 heads, nor the discarding of centgeners in the nursery, nor the continuous selection of high-yielding plants within the centgeners had any effect on eliminating the poor yielders. There was a marked difference in the appearance of the pure strains, some

Fig. 15.—Field plats, showing variations in winterkilling between two pure strains of Turkey wheat. Strain No. 377 is shown at the right and No. 102 at the left; No. 377 withstood the winter almost perfectly.

having short grains and others long grains. They also varied in color, lodging, and general appearance in the field, both in fall growth and spring growth. Figures 14, 15, 16, and 17 illustrate these variations better than they can be described.

Table XX also shows the data from Table XV grouped in series of 5, and arranged in various ways to illustrate relationships. The principal considerations in this work were the improvement of wheat in nitrogen and yield. Records were kept of many characters of the plant in the nursery, but evidence points to six that are of interest, namely, (1) nitrogen content, (2) yield per plant, (3) yield per centgener, (4) weight of kernel, (5) strength of straw, and (6) length of fruiting period.

PERCENTAGE OF NITROGEN.

The percentage of nitrogen is in inverse ratio to strength of straw and length of fruiting period, but has no direct relation to other characters. It is transmitted in the field plats as indicated in the summary of Table XX. A striking example of this conclusion is seen in a comparison of families Nos. 209 and 37, Table XX. These families represent the two extremes in percentage of nitrogen and strength of straw, with an inverse relation, but are nearly the same in all other characters.

From 1903 to 1906, records were kept of individual plants selected from the nursery. When these plants were classified according to

FIG. 16.—Increase rows of Turkey wheat, showing variations in the time of heading in different strains, each from a single plant. Four rows of each strain are grown.

percentage of nitrogen or size of kernel, regardless of the family from which they came, there was a marked inverse relation, the percentage of nitrogen increasing as the size of kernel decreased. It seems probable, however, that the individual plants having small kernels may have suffered some degree of arrested development, since this relation disappears when the pure strains are so classified. Percentage of nitrogen and yield per acre in field plats vary inversely.

STRENGTH OF STRAW.

Strength of straw varies inversely with percentage of nitrogen and directly with yield per acre and yield per centgener.

FIG. 1.—TYPE OF ROAD GRADER OR DRAG USED IN GRADING A NURSERY INTO BEDS 20 FEET WIDE TO AFFORD UNIFORM DRAINAGE.

FIG. 2.—GRAINS OF TURKEY WHEAT, SHOWING VARIATION IN APPEARANCE.

Nos. 51 and 60 are typical kernels from two pure strains and represent the shortest and longest kerneled types out of 80 strains. No. 76 is a hard, vitreous kernel, somewhat approaching the durum wheat in type. No. 75 is a soft wheat. The plants of this strain are typical Turkey in appearance but the grain is larger and almost white. Notwithstanding the white color, this strain was the highest in nitrogen content of 80 strains in 1910.

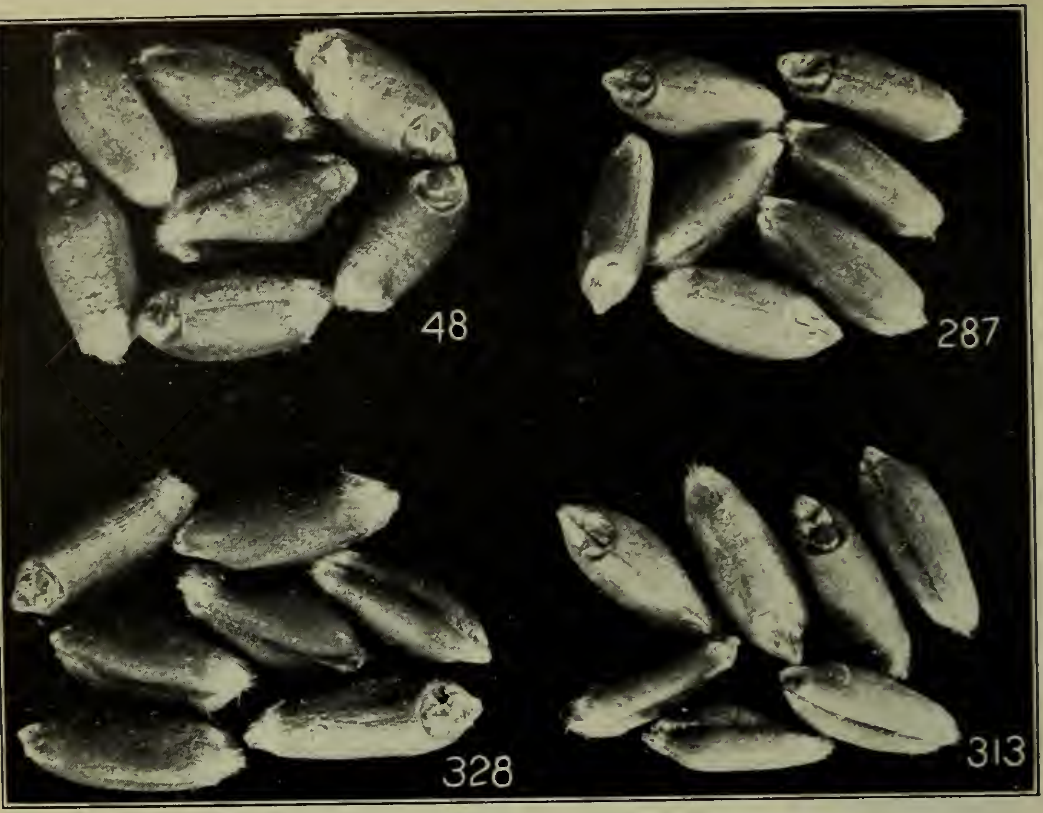

FIG. 1.—REPRESENTATIVE KERNELS FROM 4 STRAINS OF TURKEY WHEAT, SELECTED TO SHOW VARIATION IN APPEARANCE.

No. 48 is a large plump-kerneled strain, while No. 287 has a rather small kernel. No. 328 has a large dark-colored kernel, while No. 313 has a decided yellow color and is long and pointed.

FIG. 2.—REPRESENTATIVE KERNELS FROM 4 STRAINS OF TURKEY WHEAT, SELECTED FROM A SERIES OF 80 STRAINS TO SHOW VARIATION IN QUALITY.

On the basis of a perfect wheat, grading 100, these strains grade as follows: No. 77 grades 50, No. 51 grades 70, No. 27 grades 80, and No. 42 grades 95.

YIELD PER PLANT.

The yield per plant shows some correlation to yield per acre and yield per centgener, but this correlation is not high, as it is only in the first class that the correlation is marked. One of the best-yielding strains (No. 425) had a small plant yield in the nursery.

YIELD PER CENTGENER.

The yield per centgener shows a high correlation with yield per acre and strength of straw, but not a close relation to other characters.

SIZE OF KERNEL.

The size of kernel (Pl. III, fig. 2) appears to have no fixed relationships; as a character of a pure strain it seems to be independent of other characters. An example of this is shown in Table XX. Families

FIG. 17.—Field plats of Turkey wheat, showing variations in stiffness of straw in two strains. Each strain originated from a single plant.

Nos. 287 (Pl. IV, fig. 1) and 425 have small kernels, but they are among the best in yield, while No. 328 (Pl. IV, fig. 1) is poorest in yield, but has the largest kernel. Nos. 48 and 287 are the best yielders out of the 26 strains (Table XX), averaging 40.7 and 40.6 bushels per acre, respectively, in a four-year test. No. 48 has a large, plump kernel, while No. 287 has a rather small kernel. No. 328 has averaged 28.9 bushels under the same conditions, yet this strain has a large, dark-colored kernel. Our records do not seem to show a relation between the appearance of the berry and the yield. No. 313 has averaged 36.3 bushels per acre, but the kernel has a decided yellow color, and is long and pointed in shape, approaching a rye grain in type.

QUALITY OF KERNEL.

As already noted, there does not seem to be a definite relation between the appearance of the berry and the yield of the strains.

Plate IV, figure 2, illustrates four strains of Turkey wheat selected from a series of eight strains to show variation in quality. On the basis of a perfect wheat grading 100 these strains grade as follows: No. 77 grades 50, No. 51 grades 70, No. 27 grades 80, and No. 42 grades 95. These grades indicate the variation in quality found in pure strains and show the great possibility of improving quality (fig. 18).

To sum up, high yield in the field is associated with high yield per centgener and strong straw, has a slight relation to size of plant, no relation to size of berry, and varies inversely with percentage of

FIG. 18.—Cereal laboratory, showing the method of taking notes on quality. Comparisons of 80 strains of Turkey wheat are being made. There were 10 plats of each strain, making 800 in all, but the 10 samples of each strain are arranged together. Notes are taken on each sample separately, then an average is made of the results. To facilitate note taking, a set of "standard samples" representing different qualities is kept in long, 2-ounce vials. A set of these vials is plunged into the sample, and by comparison very accurate data are obtained.

nitrogen. A high or a low nitrogen content as indicated in the nursery gives correlated results in the field. High nitrogen content is antagonistic to high yield. However, an occasional strain, such as No. 48, combines a fair percentage of nitrogen with high yield.

SUPERIORITY OF STRAIN.

From the point of view of yield per acre there would seem to be little choice among the five best strains (Table XX), but in some ways No. 48 is outstanding in desirable qualities. With a high yield per acre, it still is above the average in nitrogen content, has a strong straw and a large berry of good appearance. It is also very winter resistant, as was shown in the severe winter of 1909–10, when it came through with much less winterkilling than standard varieties, such as Big Frame and ordinary Turkey wheat.

COMPARISON OF ROWS, CENTGENERS, BLOCKS, AND FIELD PLATS.

In 1909–10 the 26 strains of Turkey wheat were sown in field plats and duplicated in rows, centgeners, and blocks, but this portion of the nursery was winterkilled. However, very good results were obtained with 11 varieties of oats sown in the spring of 1910 in all four ways. The field plats were one-fifteenth of an acre in size and

Fig. 19.—Block nursery, showing blocks 4.2 by 16 feet in size. The beds are slightly elevated, as in the row nursery.

were repeated three times. Each variety was repeated 10 times in centgeners, rows, and blocks. The centgeners were each 5 feet square and contained 100 plants 6 inches apart each way. The rows were 12 feet in length and the grain sown in them at the rate of 10 pecks per acre, the usual rate of seeding in this region. The blocks were each 4.2 by 16 feet, or 5 drill rows wide (fig. 19), and sown at the usual rate of seeding. The results summarized in Table XXI show a high degree of correlation between the yield of the field plats and the rows and blocks, but practically no correlation in the case of centgeners. Except for the Lincoln oat, which yielded exceptionally high in the nursery, the correlation in the rows and blocks would be very high.

TABLE XXI.— *Yields of grain from 11 varieties of oats grown in field plats, centgeners, rows, and blocks.*

[The field plats were repeated 3 times, the others 10 times.]

Variety.	Average yield of thrashed grain.			
	Field plats, per acre.	Cent-geners.	Rows.	Blocks.
	Bushels.	*Grams.*	*Grams.*	*Grams.*
Burt	60.7	729.2	238.2	1,196.8
Texas Red	60.6	622.9	206.9	1,047.2
Average	60.65	676.05	222.55	1,122.0
Swedish Select (01)	55.8	584.3	213.5	865.8
University No. 6	55.3	572.8	178.0	867.1
No. 855	55.2	473.7	167.0	809.8
Average	54.77	543.6	186.16	847.56
American Banner	51.9	550.9	170.4	800.8
Lincoln	51.8	654.4	207.3	1,071.5
No. 1174	51.0	601.3	184.2	842.5
Average	51.56	602.2	187.3	904.93
Average without the Lincoln oat	....	576.1	177.3	821.6
Swedish Select (09)	50.3	560.4	170.8	791.1
No. 611	49.8	581.1	174.3	760.4
No. 70	49.3	470.3	159.6	776.7
Average	49.83	537.26	168.23	776.26

It is possible that the lack of correlation with the centgeners in the last three groups is due to the fact that the lower yielding ones were rather late, coarse-strawed, and rank-growing strains. Under the thin planting of the centgeners every plant had opportunity to develop to full size, which probably gave a slight advantage to the later and coarser types, which was not apparent under the usual rate of seeding. Table XX, giving results for 24 strains of Turkey wheat, shows a perfect correlation between yield of centgener and field plats, wherein the wheat strains were all similar, there being no great difference in time of maturity or habit of growth. This suggests that when similar strains are being compared data on the average yield of centgeners would be reliable, but that in the case of dissimilar varieties, especially of large ones and small ones, the results would not be comparable with those obtained under field conditions.

COST OF PLANTING AND HARVESTING CENTGENERS, ROWS, AND BLOCKS.

When a large number of strains is being tested in rows or blocks and the series is repeated ten to twenty times, the question of time and relative cost becomes one of importance. Rapidity of planting and harvesting is probably the matter of first consideration, as it is desirable to have these operations performed in the shortest possible time in order to secure comparable conditions.

Table XXII is a summary showing the relative number of plats of each kind that can be planted in a day by one gang of men, based on our experience. All preliminary work, such as preparing the land, sorting and labeling the seed, preparing stakes, etc., is done beforehand. However, there is usually time for preliminary work, so that

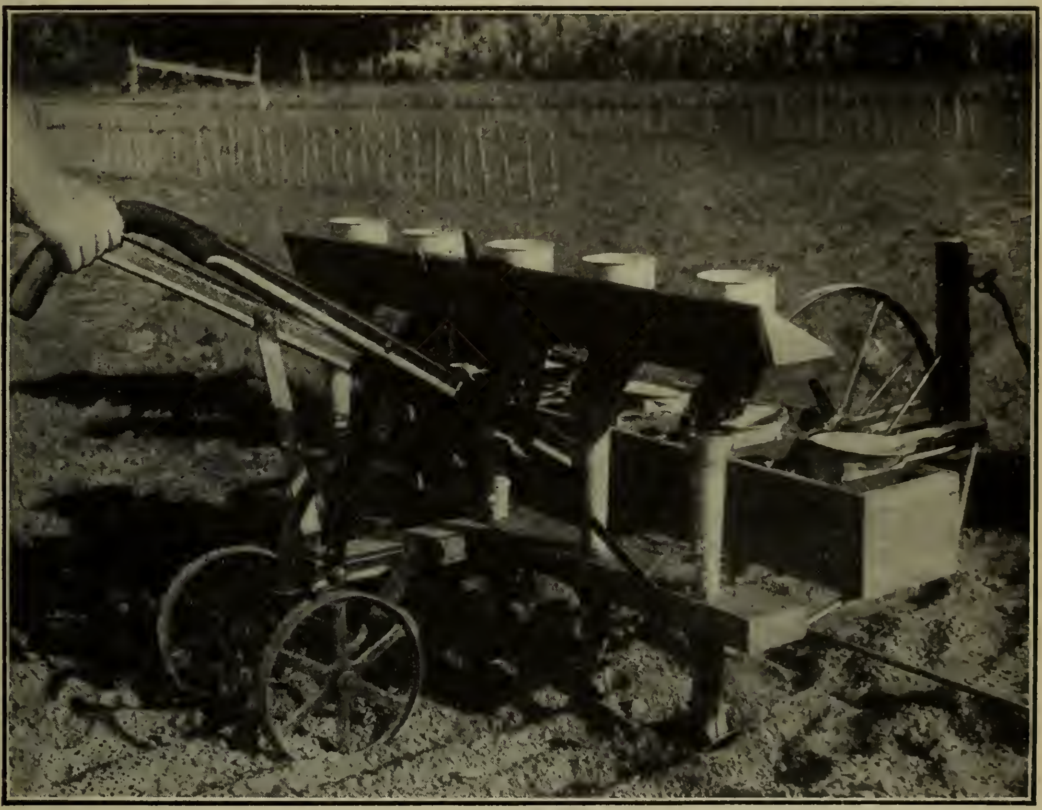

FIG. 20.—Five-row nursery drill used for planting row plats and block plats. Four or five men with this drill will easily plant 500 row plats in a day, where not more than 100 row plats could be planted by hand. The work of the drill is also more uniform and satisfactory than planting by hand.

the method of planting that permits the greatest rapidity has some advantage. The planting is done with a nursery drill (fig. 20), the harvesting with hand sickles (fig. 21), and the thrashing with a small power machine.

TABLE XXII.—*Comparative number of plats of different types that can be planted or harvested in 10 hours.*

Kind of plat.	Men in gang.	Number of plats covered in 10 hours.		
		Planted.	Harvested.	Thrashed, weighed, etc.
Centgener, using centgener planter	5	100	250	500
16-foot row plat, row made with hoe or spade and planted by hand	5	100	500	750
16-foot row plat, planted with nursery row drill	5	500	500	750
Block 4.2 by 16 feet, planted with nursery row drill	5	150	200	250

USE OF CHECK PLATS.

In nursery work where plats are repeated 10 or more times the checks will not be needed as a means of correcting error. Their principal use here is to determine the degree of experimental error. For example, in 1910 we had in one series 80 strains of Turkey wheat. The series was repeated 10 times, making 800 rows in all, but every fifth row was a check, making 200 check rows. To determine the experimental error, the 200 check plats are grouped in sets of 10, taking 1 plat from each series, the same as in grouping the strains. This method gives 20 sets of 10 checks each, and the range of experimental error to be allowed for is at once apparent.

FIG. 21.—Row plats at harvest time. Each plat is harvested with a hand sickle and the stake tied into the bundle. The bundles from the row plats that are to be kept pure for seed should be covered with cheese cloth or paper bags.

Another important use of checks is as a standard by which to judge progress, and this judgment can be formed very well if the original stock from which selections were made is used for checks.

Figure 22 illustrates the method of selection when the experimental error is determined by the use of checks. Let the figure illustrate an ideal case where any number of strains are being compared for yield, the yield varying from 200 to 320 pounds. Let line *eh* represent this variation. A series of check plats shows a variation ranging from 240 to 280, represented by line *cj*. The mean yield of the checks is

260, with an extreme error of 20 above or below the mean. The limits of error for the line *eh* are shown by the lines *dg* and *fi*. To be sure that a certain strain was better than the check plats its yield must fall outside the line *cl* or be better than 280. In the same way two strains can not be compared without making an allowance of 40. For example, 320 is not surely better than 280, since they both might equal 300, but we are sure 320 is better than anything less than 280. If the extreme variation of checks should equal the variation in strains or varieties, then selection would not be possible. On the other hand, if the error in checks is zero, direct comparison between strains could be made, and small differences would be significant. This illustration emphasizes the importance of knowing the probable experimental error and having it well within the limits of expected variation if results are to be secured by selection.

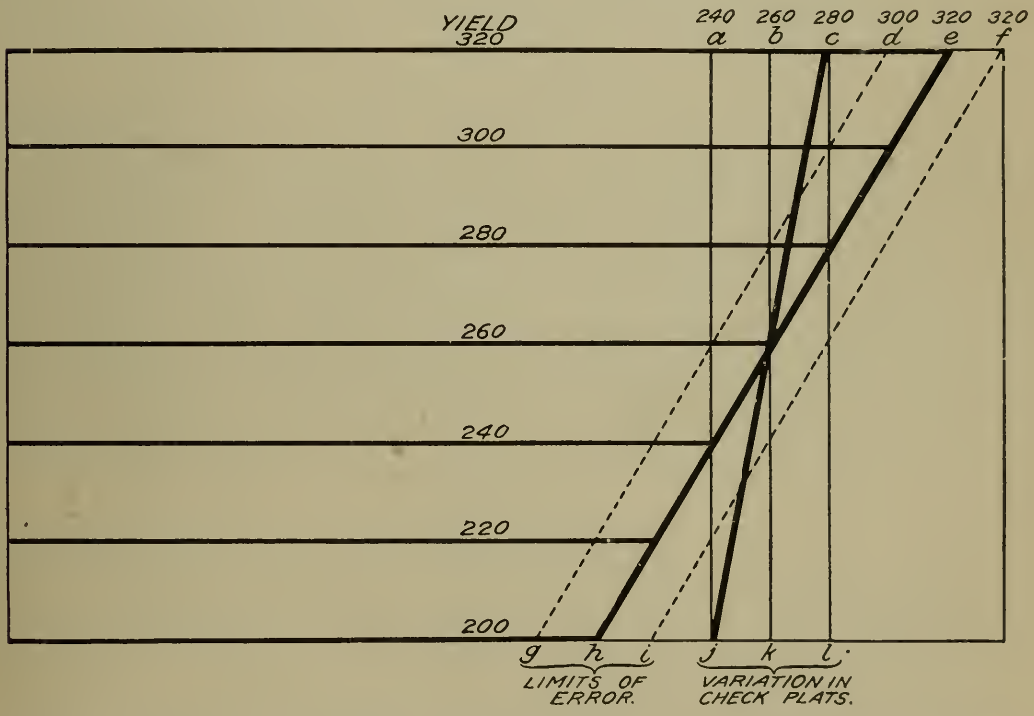

FIG. 22.—Diagram showing the method of selection for yield when the experimental error is known.

ERROR IN CHECK PLATS.

By reference to Table XVI it is seen that where odd and even numbered (alternate) row plats are checked against each other there is a certain experimental error. If this error should be large, it would be unsafe to correct by the checks. For example, if the yield of a certain check variety equals 100, but in series A its yield, due to error, should be 105, and in series B only 95, a correction of yield by these checks would introduce an error of 10 per cent. It might happen that the strain used for checks would be the most variable strain in use in some particular season, though reliable at other times. For this reason care should be taken to ascertain the experimental error in checks before using them for correction, else a greater error may be introduced by their use than by discarding them.

OTHER PRECAUTIONS AGAINST ERROR.

ACCIDENTAL INJURY TO PLATS.

A problem which always confronts the experimenter at harvest time is what to do with plats injured from some cause, such as the damage of burrowing animals, being partly washed out by floods, etc. Where a plat is obviously injured it should be discarded. To allow for accidents we have adopted the plan of planting one or two extra series to be discarded or used, as is needful, at harvest time. In this way plats accidentally injured may be discarded without disturbing the regular number in the series.

UNEQUAL DRAINAGE.

Unequal effects of winter injury are partly due to unequal distribution of moisture in the soil. Wherever there is a depression run-off water may collect and increase the soil moisture. In some cases, as in a very dry spring, these slight depressions may have an advantage, while at other times these low places suffer from winterkilling. Perfect drainage is very important and can be easily secured by laying off the nursery in slightly elevated beds about 20 feet in width. This bedding can be quickly accomplished by the use of a road grader or road drag (Pl. III, fig. 1).

SUMMARY.

(1) When 14-foot or 16-foot row plats are used as checks or sown in duplicate, great variation is found in yield, owing to natural unequal effects of the environment. When the row plats are repeated only five or six times, the extreme error will still be large, owing to the chance combination of high or low variants.

(2) Systematic repetition constantly reduces error as the number of repetitions increases, but with 16-foot row plats 10 to 20 repetitions must be made, depending on the degree of accuracy desired.

(3) It is probable that the greater the number of strains to be compared the more repetitions will be necessary, because of the greater area they will cover.

(4) Small blocks, 5.5 feet square, give results similar to those of the row plats, except that the reduction of experimental error is somewhat greater as a result of repetition. Blocks repeated 8 to 10 times give results apparently about as accurate as rows repeated 15 to 20 times.

(5) To increase the size of the block, up to a certain limit, rapidly decreases variability; but error can not be indefinitely decreased by continuing to increase the size of the plat, as it can be by repetition.

(6) Variability is not constant from year to year on the same plats.

(7) To alternate with check rows gives a high degree of accuracy, with a few extreme variations, when as high as 10 or more checks are used. The total number of plats required for the same degree of accuracy, however, is greater by this method than by systematic repetition.

(8) To increase the length of the row 4 times decreases deviation about one-half.

(9) By increasing the length of the row or the size of the block, the number of repetitions necessary is decreased, but the total area required to secure the same accuracy is increased. An excellent size, where land is plenty, would be 2 to 4 rods in length for rows, and 5 by 16 feet in area for blocks.

(10) The rate of planting, within certain wide limits, has little influence on yield.

(11) There is some competition between adjacent rows, especially when varieties very different in habit of growth are planted side by side. The use of blocks does away with this source of error.

(12) Pure strains differ in a very marked way in most important characters. High yield in the field is associated with high yield in the nursery plats, and strong straw has a slight relation to size of plant, no relation to size of berry, and varies inversely with the percentage of nitrogen.

(13) Block plats and row plats at the usual rates of seeding will probably correlate more closely with results in field plats than in plats where the plants are spaced, as in centgeners.

(14) Where error is corrected by the system of repetition of plats, check plats should be used for the purpose of determining the experimental error. When the variation in checks equals the variation in strains, no possible selection can be made.

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BUREAU OF PLANT INDUSTRY—BULLETIN NO. 270.

B. T. GALLOWAY, *Chief of Bureau.*

CONTRIBUTIONS TO THE STUDY OF MAIZE DETERIORATION.

BIOCHEMICAL AND TOXICOLOGICAL INVESTIGATIONS OF PENICILLIUM PUBERULUM AND PENICILLIUM STOLONIFERUM.

BY

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BUREAU OF PLANT INDUSTRY.

DRUG-PLANT, POISONOUS-PLANT, PHYSIOLOGICAL, AND FERMENTATION INVESTIGATIONS.

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LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry,
Office of the Chief,
Washington, D. C., September 25, 1912.

Sir: I have the honor to transmit herewith and to recommend for publication as Bulletin No. 270 of the series of this Bureau the accompanying manuscript entitled "Contributions to the Study of Maize Deterioration. Biochemical and Toxicological Investigations of Penicillium Puberulum and Penicillium Stoloniferum." The paper was prepared by Dr. Carl L. Alsberg and Mr. Otis F. Black, Chemical Biologists in the Office of Drug-Plant, Poisonous-Plant, Physiological, and Fermentation Investigations, and has been submitted by Dr. R. H. True, Physiologist in Charge, with a view to its publication.

The results of technical laboratory studies of organisms occurring in deteriorated maize, (1) *Penicillium puberulum* Bainier and (2) *Penicillium stoloniferum* Thom, are here presented, demonstrating that these organisms have specific physiological properties. One of these molds is shown to develop toxic substances in maize. Owing to the serious problems now grouping themselves about this important American farm crop, it is believed that the results of this investigation constitute a timely contribution to our information on the subject of the deterioration of maize.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

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CONTRIBUTIONS TO THE STUDY OF MAIZE DETERIORATION.

BIOCHEMICAL AND TOXICOLOGICAL INVESTIGATIONS OF PENICILLIUM PUBERULUM AND PENICILLIUM STOLONIFERUM.

INTRODUCTION.

Whether molds or the products of their growth have an injurious effect on animals is a question which has not yet been conclusively settled. The literature contains many records of alleged intoxications due to these fungi. Certain diseases of men and domesticated animals have been attributed to this cause. Though the solution of this problem is obviously urgent, few serious attempts have been made to identify chemically the alleged toxic substances. The present paper is such a chemical study. Incidental observations on the metabolism of molds have been made and have been recorded because they have a general biological interest and because they may prove useful in characterizing different species physiologically.

The difference of opinion concerning the toxicity of Penicillium is probably due not merely to the fact that the earlier investigators studied accidental mixtures of organisms under varying and undefined conditions,[1] but also that complex substrata like corn, wheat, and bread were used for the growth of the organisms. Consequently it is impossible to know whether any of the different substances found were derived from the substratum or were produced by the

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Monselise, G. Ricerche chimico-tossicologiche intorno ad alcuni campioni di mais per lo studio della pellagra, Mondovi, 1881, 58 p. (Cited by Gosio.)

organisms. Thus Selmi[1] thought acrolein or a condensation product of acrolein with ammonia was produced, while Lombroso and Dupré,[1] Lombroso and Erba,[1] Brugnatelli and Zenoni,[1] Pelloggio,[1] and Coeytaux[2] found alkaloids. Though some of these investigators examined the maize or other material employed as substratum, before the development of the organisms, only Lussana and Ciotto[3] give sufficient details to inspire confidence in the adequacy of the controls. They found alkaloids in both moldy and sound maize and wheat. In the investigation of maize in progress in this laboratory, of which this bulletin is in part a report, cholin, betain, and bases unidentified as yet have been isolated from sound maize.

The first investigator to use pure cultures in a simple culture medium was Gosio,[4] who used Raulin's solution. Under these conditions Penicillium endowed the culture medium with the power to react like a phenol with weak aqueous ferric-chlorid solutions. A similar observation on *Aspergillus niger* had been made many years before by Raulin,[5] who found that in the absence of iron this fungus has a similar effect on the culture medium. Raulin attributed the ferric-chlorid reaction present under these conditions to the formation of sulphocyanid. This observation was apparently unknown to Gosio. Recently Javillier and Sauton[5] have confirmed Raulin's observations, but have doubted that the reaction was due to sulphocyanid. They found, furthermore, that the reaction fails when the organism is grown in the absence of both iron and zinc. Moreover, Raciborski[6] also obtained the ferric-chlorid reaction with a number of fungi. He concluded that molds may secrete a variety of aromatic substances, for the culture medium may give Millon's reaction and the diazo reaction of Griess. He also found that substances reducing Fehling's solution, ammoniacal silver solution, and ammonium vanadate were formed. He regarded them all as probably products of the protein metabolism of the fungus.

Gosio found further that by administering the culture medium to mice, to rats, to guinea pigs, to rabbits, to cats, or to dogs, symptoms resembling phenol poisoning could be produced. The culture media

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[1] Op. cit.

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[4] Gosio, B. Ricerche batteriologiche e chimiche sulle alterazioni del mais. Rivista d'Igiene e Sanità Pubblica, ann. 7, p. 825–849, 869–888, 1896.

[5] Cited by Javillier, M., and Sauton, B. Le fer est-il indispensable à la formation des conidies de l'Aspergillus niger? Comptes Rendus de l'Académie des Sciences [Paris], t. 153, p. 1177–1180, 1911.

[6] Raciborski, M. Über die Assimilation der Stickstoffverbindungen durch Pilze. Bulletin International de l'Académie des Sciences de Cracovie. Classe des Sciences Mathématiques et Naturelles, ann. 1906, p. 733–770, 1907.

presented other properties characteristic of phenols. Hence, Gosio concluded that phenols were produced by the molds and that the toxicity of the culture media was due to their presence. By means of ether he even succeeded in isolating from the culture medium a small quantity of a crystalline substance giving the ferric-chlorid color reaction. The substance was slightly soluble in cold water, though freely soluble in hot water and in most organic solvents. It failed to react with Fehling's solution or with phenylhydrazin. It contained 62.2 per cent carbon, 6.34 per cent hydrogen, and 28.45 per cent oxygen, corresponding to the empirical formula $C_9H_{10}O_3$. In alcoholic solution ferric chlorid produced an intense blue coloration. In spite of the high melting point, 143° to 144° C., and the failure of Millon's reaction, Gosio regarded the substance as probably *p*-hydro-cumaric acid. Moreover, he presented evidence for the existence in the culture medium of other unisolated phenolic substances. As a result of these researches Gosio proposed ferric chlorid as a reagent for the detection of moldy maize, but stated that its usefulness might be limited to the detection of the molds producing phenols.[1] These discoveries, in the main, were confirmed by others.[2]

Unfortunately, none of the investigations on green Penicillium can be repeated because the organism used was inadequately described. Undoubtedly the different investigators employed different species, for nearly all of them recognized the fact that the quantities of Gosio's toxic phenolic substances varied for different strains. None of them attributed these variations to the use of distinct species, though Ceni and Besta [3] did distinguish between toxic and nontoxic forms without, however, adequately differentiating them.

Di Pietro [4] first showed toxicity to be limited to certain distinct species of Penicillium. From moldy maize he isolated *Penicillium brevicaule*, *P. candidum*, and *P. glaucum*; but stated that "*P. glaucum*" did not mean *P. glaucum* Link, because six quite distinct green species were isolated, none of which is described in Saccardo's Sylloge. Of

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[1] Black, O. F., and Alsberg, C. L. The determination of the deterioration of maize, with incidental reference to pellagra. U. S. Department of Agriculture, Bureau of Plant Industry, Bulletin 199, p. 27, 1910.

[2] Gosio, B., and Ferrati, E. Sull' azione fisiologica dei veleni del mais invaso da alcuni ifomiceti. Rivista d'Igiene e Sanità Pubblica, ann. 7, p. 961–981, 1896.

Antonini, G., and Ferrati, E. Sulla tossicità del mais invaso da "Penicillium glaucum." Archivio di Psichiatria, Scienze Penali ed Antropologia Criminale, v. 24 (s. 2, v. 8), p. 581–585, 1903.

Ceni, Carlo. Le proprietà tossiche di alcuni ifomiceti in rapporto colle stagioni e col ciclo annale dell'endemia pellagrosa. Comunicazione preliminare. Rivista Pellagrologica Italiana, v. 4, p. 89.

——— Idem. Seconda nota preventiva. Comunicazione fatta al III Congresso della Società Italiana di Patologia tentosa in Roma, 26-28 Aprile, 1905. Rivista Pellagrologica Italiana, v. 5, p. 184.

[3] Ceni, Carlo, and Besta, Carlo. I Penicilli nell' etiologia e patogenesi della pellagra. Rivista Sperimentale di Freniatria e Medicina Legale della Alienazioni Mentali, v. 29, p. 741–815, 1903.

——— Die pathogenen Eigenschaften des Aspergillus niger mit Bezug auf die Genesė der Pellagra. Beiträge zur Pathologischen Anatomie und zur Allgemeinen Pathologie, Bd. 37, p, 578–589, 1905.

[4] Di Pietro, Melchiorre. Sui veleni di alcune muffe. Annali d'Igiene Sperimentale, v. 12 (n. s.), p. 314–365, 1902.

——— Intorno al "Penicillio tossico." Rivista Pellagrologica Italiana, v. 3, p. 221, 1903.

all the organisms isolated from maize by Di Pietro only one of the six species of "*P. glaucum*" proved to be toxic. The published description of the organism [1] was not accessible in Washington.

Gosio [2] replied to these papers of Di Pietro, stating that he had previously pointed out [3] that different strains vary greatly in their power to produce toxic substances. Though he studied Di Pietro's organism he refused to acknowledge that Di Pietro's observations had any further significance and was inclined to think that *Penicillium glaucum,* like pathogenic bacteria, varies in virulence.

Di Pietro believed that the toxic principle was a glucosid,[4] although his reasons were not clearly stated, for this toxic principle was not obtained in a state of purity. All that showed glucosidic properties was the reduction of Fehling's solution by the extracts and the diminution of toxicity by heating with hydrochloric acid. Though Gosio's substances were excreted into the medium and those of Di Pietro were confined to the spores, both substances gave a ferric-chlorid color reaction.

Sturli [5] also found that an organism isolated from polenta or maize mush from Bukowina did not render the medium toxic, while the mycelium was very toxic. He believed it premature to draw any conclusions concerning the chemical properties of the toxic substances, calling the toxic material "indifferent substances."

Possibly the descriptions of all of the green species of Penicillium used in previous investigations are inadequate because the medical bacteriologist is usually insufficiently acquainted with molds.[6] The identification of the species can be intrusted only to the skilled mycologist.[7] One of the consequences of the study of molds by mycologists has been that Thom has found it necessary to disregard the species *Penicillium glaucum* Link, because Link's description is applicable to many forms.[8] Hence the identity of many of the molds used hitherto in toxicological investigations can no longer be established. Di Pietro was therefore right in his belief that a

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[1] Di Pietro, Melchiorre. Studio morfologico e biologico sul Penicillium glaucum (var. tossica) Teramo. 1904.

[2] Gosio, B. Per l'etiologia della pellagra. Rivista Pellagrologica Italiana, v. 3, p. 177, 1903.

[3] Gosio, B. Ricerche batteriologiche e chimiche sulle alterazioni del mais. Rivista d'Igiene e Sanità Pubblica, ann. 7, p. 825–849, 869–888, 1896.

[4] Di Pietro, Melchiorre. Glucosidi di elevato potere tossico trovato nelle spore del Penicillium glaucum. Rivista Pellagrologica Italiana, v. 2, p. 63, 1902.

[5] Sturli, Adriano. Ueber ein in Schimmelpilzen (Penicillium glaucum) vorkommendes Gift. Wiener Klinische Wochenschrift, Jahrg., p. 711–714, 1908.

[6] Tiraboschi, C. Studi sugli ifomiceti parassiti del granturco guasto. Atti del Terzo Congresso Pellagrologico Italiano, Milano, 24-25-26, Septembre, 1906, p. 126. 1907.

[7] Nikitinsky, Jacob. Über die Beeinflussung der Entwicklung einiger Schimmelpilze durch ihre Stoffwechselprodukte. Jahrbücher für Wissenschaftliche Botanik, Bd. 40, p. 3, footnote, 1904.

Wehmer, C. Notiz über Rhizopus-Arten. Berichte der Deutschen Botanischen Gesellschaft, Bd. 28, p. 547–549, 1911.

[8] Thom, Charles. Cultural studies of species of Penicillium. U. S. Department of Agriculture, Bureau of Animal Industry, Bulletin 118, 1910.

number of green species exist. Moreover Dox[1] has shown that biochemically the different species vary and has asserted that many of the conflicting statements in the literature, especially those concerning the proteolytic enzyms, may be explained on the assumption that different organisms were used under the same name. Nikitinsky[2] also suspected that *Penicillium glaucum* and *P. griseum* comprised a number of physiological varieties because of their varying power to form or to withstand acid.

For this investigation the genus Penicillium was chosen, because it is well systematized in the recent monograph of Thom.[3] Five species inhabiting spoiled maize in the United States were obtained from Dr. Thom and cultivated separately by Dr. Erwin F. Smith. Without such help this investigation could hardly have been undertaken. Seventy-five to one hundred grams of air-dry, white corn meal were sterilized, usually in the autoclave, in 1-liter Erlenmeyer flasks, with enough water to form a layer of the mixture over the bottom about 3.75 centimeters thick. These flasks were observed for several days, to detect inadequate sterilization, when the sterile flasks were inoculated by Dr. Smith with spores of fresh cultures. The cultures were allowed to develop at room temperature in diffused light for 34 days, from April 12 to May 16, 1910. All developed spores.

To determine toxicity each culture was digested 16 hours with 500 cubic centimeters of 95 per cent alcohol. The extract was filtered and all alcohol from 50 cubic centimeters, corresponding to 6 to 10 grams of meal, evaporated on the steam bath. The residue was filtered, yielding usually about 1 cubic centimeter of an acid solution. One-half of this filtrate was then injected subcutaneously into mice weighing 20 grams each. The extract from one of the cultures caused death in about 10 hours; another in 7 hours; the others had no harmful effect. Neither the convulsions nor the spasms described by Gosio and Ferrati[4] were observed. The mice seemed merely very sick and died quietly. When the dosage was larger, initial convulsions with paralysis of the fore limbs were produced. These symptoms soon passed away and the mice died as did those that had received smaller dosages. The fatal dose for both cultures was more than ten times that of Gosio's cultures, so that neither is likely to have been identical with his culture.

As a control, a culture flask containing corn-meal mush in the same quantity was left uninoculated. After standing sterile during the

[1] Dox, A. W. The intracellular enzyms of Penicillium and Aspergillus, with special reference to those of Penicillium camemberti. U. S. Department of Agriculture, Bureau of Animal Industry, Bulletin 120, p. 36, 1910.

[2] Nikitinsky, J. Op. cit.

[3] Thom, Charles. Op. cit.

[4] Gosio, B., and Ferrati, E. Sull' azione fisiologica dei veleni del mais invaso da alcuni ifomiceti. Rivista d'Igiene e Sanità Pubblica, ann. 7, p. 961-981, 1896.

growth of the organisms in the other flasks it was, simultaneously with them, extracted by the same proceeding and tested in the same way for toxicity. It did not produce any symptoms in the mice. Consequently, the effects observed were the result of the growth of the molds.

Because the toxic substances are believed by Gosio to be identical with those giving color reactions with ferric chlorid the cultures were tested with this reagent. Fifty cubic centimeters of the alcoholic extracts were neutralized and evaporated nearly to dryness on the steam bath. The residues were then extracted with water weakly acidulated with hydrochloric acid and filtered. The clear filtrate was extracted with a relatively large volume of ether and the ethereal extract evaporated to dryness. The residue from the ether was extracted with water before testing with a very weak solution of ferric chlorid. In one case a weak but distinctly reddish brown color was obtained. One culture gave no color. Two gave a slight brown coloration. Another culture, after the evaporation of the alcohol, gave a brick-red, watery extract, which, on neutralization, turned to a deep-claret color. Therefore, in this culture, it was impossible by this method to detect phenols, if present. The organism producing this dye is the one referred to by Thom [1] as having been found by Prof. F. D. Heald on maize and as being either identical with or very closely related to *Penicillium purpurogenum* O. Stoll. It has been possible to isolate the dyestuff and its description is reserved for a future publication.

The culture which gave on extraction the brownish red coloration with ferric chlorid was the one that killed the mice in 10 hours. It was identified as *Penicillium puberulum* Bainier, and a detailed study of this species is here given.

PENICILLIUM PUBERULUM.

Penicillium puberulum Bainier was originally isolated from corn by Prof. F. D. Heald, then of Lincoln, Nebr., now of Austin, Tex. The culture obtained from Dr. Thom is No. 45 of his collection, and had been grown on a 3 per cent sugar decoction of beans. Dr. Thom states that if it is not identical with *P. puberulum* of Bainier [2] it is very closely related to it. He has very kindly furnished the following description:

Colonies in Czapek's solution agar, dull bluish green to green, velvety, spreading slowly upon the substratum, zonate at the margin in older colonies, with an outer band of submerged mycelium; mycelium yellowish or greenish to tan below (as viewed from below); agar uncolored; odor weak but noticeable; conidiophores

[1] Thom, Charles. Op. cit., p. 36.

[2] Bainier, G. Sur dix espèces nouvelles de Penicillium et sur le genre G. graphiopsis. Bulletin, Société Mycologique de France, t. 23, 1907, p. 16. 1908.

FLASK SHOWING TWELVE DAYS' GROWTH OF PENICILLIUM PUBERULUM ON RAULIN'S MEDIUM.

mostly arising separately from the submerged hyphæ, short—up to 100 μ long by 3.5 μ diameter, crowded, with walls more or less rough with delicate warts or granules or smooth; conidial fructifications becoming more or less loosely columnar masses 20 to 30 μ long and consisting of the main conidiophore, a primary branch or verticil of branches, secondary verticils of branchlets, all swollen or enlarged at apex, bearing verticils of conidiferous cells 8.5 by 2 to 2.5 μ, producing very long chains of conidia; conidia elliptical to globose, 3 to 3.5 μ diameter when ripe, smooth, swelling in germination to 5 to 8 μ and germinating by one or two tubes 3 μ in diameter—when two tubes are present these are separated by 180°—appear at the points of contact of the conidia in the chains (where the walls are thinner); colonies liquefy simple gelatin, producing a trace of brown color in the liquid and an alkaline reaction to litmus, and become dark or smoky in color in media without sugar; with lactose gelatin no acid is produced; in rich substrata under humid conditions colonies become overgrown with white floccose sterile mycelium after the conidial areas are matured; colonies grown readily in all common media. Collected by Prof. F. D. Heald in Nebraska upon decaying Indian corn (*Zea mays*).

Note.—In measuring conidiophores I measure from the point of origin at the surface of the substratum to the lowest branch of the fruit. Simple gelatin here means 15 per cent of gelatin in distilled water.

Plate I shows a culture of *Penicillium puberulum* Bainier after 12 days' growth on Raulin's medium.

In investigating the toxic properties of this mold, the injurious effects were found due not to the acidity [1] of the extracts of the culture, as prepared above, but to some specific substance. This is shown in the following experiment:

Fifty cubic centimeters of the alcoholic extract of the corn-meal cultures were carefully neutralized with sodium carbonate before concentration. The solution thus obtained was not toxic. As loss of toxicity might be due either to neutralization of acid or to destruction of the toxic substance by heat in neutral solution, fresh extracts were tested by injecting the ethereal extract neutralized after concentrating. The mice died as before, showing that the toxicity was due to some toxic material stable only in acid solution.

PENICILLIC ACID FROM PENICILLIUM PUBERULUM.

Cultures of *Penicillium puberulum* Bainier were grown about a month on Raulin's medium. At the end of this period the medium was still acid, but far less toxic than the extracts prepared from the cultures on corn-meal mush. However, the medium gave the color reaction with ferric chlorid. Extracts of the mycelium were neither toxic nor did they give the ferric-chlorid reaction. The substance responsible for this reaction did not occur in the mycelium. It was extracted from the medium with chloroform and crystallized on concentration of the solvent. Recrystallized from hot water it forms large, transparent, biaxial, monoclinic or triclinic, rhombic crystals

[1] Black, O. F., and Alsberg, C. L. The determination of the deterioration of maize, with incidental reference to pellagra. U. S. Department of Agriculture, Bureau of Plant Industry, Bulletin 199, p. 30, 1910.

with good cleavage normal to an axis. The substance burns without ash, contains no nitrogen, and is not optically active; but it is acid to litmus and phenolphthalein and decomposes carbonates at ordinary temperatures.

For purposes of analysis the material was purified with bone black and twice recrystallized from hot water. Recrystallization from alcohol, water, ether, benzol, or chloroform and precipitation from ethereal solution by petroleum ether failed to remove traces of a yellowish color or to change the melting point.

The crystals from water have a melting point of 64° to 65° C., uncorrected, and effloresce rapidly in the air. The anhydrous substance is a white powder, melting sharply at 86° to 87° C., uncorrected, without decomposition. The water of crystallization was determined by pressing the crystals dry between filter paper, weighing rapidly, and drying to constant weight at room temperature under diminished pressure, with sulphuric acid as dehydrating agent. Heated to 50° to 60° C. the material suffered no further loss in weight. The other analyses made were the determinations of the content of carbon and hydrogen, of the magnesium content of the salt, and the titration of the free acid with standard alkali. The last two determinations furnished data for the calculation of the molecular weight. The substance was dried for these analyses in the manner above given. The analytical data follow:

TABLE I.—*Analyses of penicillic acid.*

Weight of substance (grams).	CO_2 (grams).	H_2O (grams).	C (per cent).	H (per cent).	Water of crystallization.		$MgSO_4$ (grams).	Mg (per cent).	N/10KOH (c. c).
					Grams.	Per cent.			
0.2268 (anhydrous substance)	0.4649	0.1165	55.90	5.71					
0.2017 (anhydrous substance)	.4135	.1081	55.91	5.68					
0.2215 (anhydrous substance)									12.97
0.3460 (crystals from water)					0.0577	16.6			
0.2664 (crystals from water)					.0475	17.8			
0.2238 (anhydrous magnesium salt)							0.0750	6.78	
0.1923 (anhydrous magnesium salt)							.0639	6.72	
Average			55.90	5.69		17.2		6.75	

Calculated for $C_8H_{10}O_4$: carbon, 56.6 per cent; hydrogen, 5.89 per cent.
Found .... carbon, 55.90 per cent; hydrogen, 5.69 per cent.

Calculated for $C_8H_{10}O_4$, $2H_2O$: water, 17.48 per cent.
Found .... water, 17.20 per cent.

A molecular weight determination by the elevation of the boiling point in chloroform solution gave the results shown in Table II.

TABLE II.—*Ebullioscopic determination of molecular weight of penicillic acid.*

Weight of substance (grams).	Weight of solvent (grams).	Rise of boiling point (°C.).	Molecular weight.
0.1802....	26.24	0.170	148
.2091....	26.24	.191	153
.2349....	26.24	.177	177
Average..	....	....	162

Molecular weight calculated for $C_8H_{10}O_4$........................	170
Molecular weight found from titration........................	170.4
Molecular weight found from magnesium content of salt.........	168.3
Molecular weight found by boiling-point elevation.............	162

From the data at hand the formula $C_8H_{10}O_4$ may be provisionally assigned to this substance. It has not been possible to identify it with any known substance, nor has it as yet been possible to determine its constitution. Tentatively, the name penicillic acid is suggested for it.

Penicillic acid is soluble in alcohol, ether, benzol, and chloroform, but insoluble in petroleum ether. In cold water it is soluble in the proportion of about 2 to 100, but readily soluble in hot water. From its aqueous solutions it can be extracted most readily by chloroform. Olive oil dissolves it but slightly and does not extract it from aqueous solution. It decomposes carbonates. It has a somewhat salty, slightly bitter-sweet taste, and is somewhat irritating to mucous membranes. It produces a rather persistent sensation of burning at the fauces. It absorbs bromin without forming an insoluble compound; reduces Fehling's solution when heated, and ammoniacal silver at room temperature. It is fairly resistant to acids, for it may be heated with weak acid without undergoing decomposition, and it is not charred by concentrated sulphuric acid. It is, however, very sensitive to alkali. Fixed alkalis turn the solution yellow. Dilute ammonia gradually converts it into a deep-red dye, resembling that obtained from orcin by ammonia. The color with ammonia is so similar to that of the dyestuff obtained from *Penicillium purpurogenum*[1] that similarity in constitution between the two substances is at once suggested.

The magnesium salt forms transparent, rapidly efflorescent plates, drying to a white powder. It is very soluble in water and alcohol and is not readily precipitated from its alcoholic solution by ether.

[1] Cf. supra, p. 12.

The salt was prepared by digesting an aqueous solution of the pure substance on the water bath with magnesium carbonate, filtering, and concentrating to dryness in a desiccator. It contains about 30 per cent of water of crystallization. Because of its great solubility it was not further purified for analysis, but dried to constant weight in vacuo under sulphuric acid at room temperature. The salts of calcium, barium, sodium, ammonium, potassium, and copper are also very soluble in water. Their solutions dry down to varnishes. The silver salt is not stable. The lead salt is insoluble in water and amorphous.

Aqueous solutions of penicillic acid are not changed by very dilute ferric chlorid till after some time, when they gradually turn brownish red. Either it is not a simple phenol and must first be oxidized, or the action of the ferric chlorid is inhibited for a time by the reducing power of the substance. The reaction is not so delicate as that with ammonia. The formation of the color with ferric chlorid is very like that obtained by the oxidation of β-oxybutyric acid by means of weak peroxid of hydrogen containing traces of ferric chlorid.[1] Penicillic acid also gives the color under these conditions. The reaction carried out with the aid of peroxid of hydrogen is very much more delicate than that without the peroxid of hydrogen. Alcoholic solutions do not give a color with ferric chlorid. Aqueous solutions give no color with calcium hypochlorite. Nickel and cobalt chlorid give no color reaction.

Because of the observations of Brugnatelli and Zenoni[2] on the occurrence of strychninlike alkaloids in moldy corn, reactions characteristic for strychnin were tried. (1) A few crystals treated with concentrated sulphuric acid gave only a faint yellow color. On adding a small crystal of bichromate of potassium to the mixture a weak green color from reduced bichromate developed. The reaction was therefore negative. (2) A few crystals were treated with a little concentrated nitric acid. No color developed. On warming, a yellow color appeared, which, as the acid was evaporated, turned pale orange. After cooling, an orange-colored sticky residue remained, which when treated with ammonia water turned a little darker orange. Hence, both these tests for strychnin proved negative.

If an aqueous solution of the substance be boiled under a reflux condenser with barium hydrate, barium carbonate is the only insoluble product formed. If the solution be then distilled after acidifying, a small quantity of delicate needles with an odor akin to cumarin and vanillin may be separated from the distillate by extracting with

[1] Black, O. F. The detection and quantitative determination of β-oxybutyric acid in the urine. Journal of Biological Chemistry, v. 5, p. 207-210, 1908.

[2] Brugnatelli, T., and Zenoni, E. Di un alcaloide che si trova nella melica guasta e nel pane di mais ammuffito. Reale Istituto Lombardo di Scienze e Lettere, Rendiconti, s. 2, v. 9, p. 293-297, 1876.

ether. They gave no color with ferric chlorid. Gosio obtained extracts with an odor of cumarin from cultures of Aspergillus.[1]

The dry crystals heated with concentrated hydrochloric acid form a yellow oil, showing a tendency to crystallize when chilled. The oil is insoluble in water and with ferric chlorid gives a dark-red color at once, not weakly and gradually like the mother substance.

Lieberman's reaction produced a beautiful carmine-red coloration. Millon's reaction is negative. In performing Piria's reaction the substance when dissolved in warm, concentrated sulphuric acid turned yellow, then red; and when diluted with water gave a heavy, amorphous-red precipitate, which was not obtained crystalline from alcoholic solutions. This is evidently an insoluble sulphonic acid. It was removed by filtration and the filtrate neutralized with barium carbonate. The barium sulphate was removed and the clear filtrate tested with a little weak ferric chlorid. A dirty red color developed, quite different from the fine violet color obtained with tyrosin. It is doubtful whether this test has the significance of Piria's reaction, since a similar color is obtained after heating penicillic acid with dilute sulphuric or hydrochloric acid without subsequently neutralizing.

By the method suggested by Schotten and Baumann no crystalline benzoyl derivative was obtained. No crystalline oxime could be prepared. A small quantity of a crystalline derivative was obtained by heating with acetic anhydrid. Crystalline ethyl ester was also prepared by heating in absolute alcohol with a little dry hydrochloric acid. Heating an aqueous solution with phenylhydrazin hydrochlorid and sodium acetate caused decomposition, as shown by the evolution of gas. Lemon-yellow prisms were formed, which were recrystallized from hot alcohol until a constant melting point, 171° C., uncorrected, without decomposition, was obtained.

The determination of carbon and hydrogen in these yellow crystals by combustion gave the following results:

Weight of substance burned.	0.2103 gram.	
Weight of CO_2	.5655 gram;	C, 73.34 per cent.
Weight of H_2O	.1429 gram;	H, 7.56 per cent.

From these figures no formula could be calculated which might be derived from the original substance. It is probably a compound formed by the condensation of phenylhydrazin with a decomposition product. With methylphenylhydrazin nothing crystalline could be obtained. With an excess of nitrophenylhydrazin and acetic acid a red crystalline substance separates, which was not further studied.

Penicillic acid seems to be a substance not hitherto described. Hydroxyphenylglycolic acid would have a similar formula. It is

[1] Gosio, B. Mutamento de chimismo ifomicetico in rapporto all' alta e bassa fermentazione. Atti del Quarto Congresso Pellagrologico Italiano, Udine, 23-24-25 Septembre, 1909, p. 65. 1910.

not known and would in all probability give Millon's reaction. Certain of the lichen acids found hitherto only in lichens and the substance of Gosio cited above have similar empirical formulæ and properties. Not only have some of the lichen acids empirical formulæ resembling penicillic acid, but penicillic acid resembles them in its chemical behavior. Lichen acids are stable in acid solution, but are often decomposed by alkalis to form dyestuffs, as is also the case with penicillic acid. Some of the dyestuffs in common use, like orseille and litmus, are formed in this way. Most of them are decomposed by hot barium hydrate. Some of them are bitter and irritating, often much more so than penicillic acid. Some are derivatives of phenolic substances like orcin or divarin. The fact that penicillic acid forms a sulphonic acid insoluble in water and yields a dye when subjected to Lieberman's reaction also indicates that it contains a phenol nucleus. Lichen acids of this constitution turn red when treated with ammonia, like penicillic acid. Many of them give color reactions with ferric chlorid. The evidence thus far at hand decidedly indicates that penicillic acid may eventually prove to belong to this class of compounds.[1]

However, aliphatic formulæ are possible. The propyl esters of diacetylcarbonic, glyoxylpropionic, and acetylpyruvic acids would have very similar formulæ and, presumably, at least some of the characteristics of penicillic acid. Should penicillic acid ultimately prove to be a substance of this general type it would still in some ways resemble certain of the lichen acids; for example, vulpinic acid, which is an inner anhydrid of diphenylketipinnic acid.

If penicillic acid is related to the lichen acids its formation by the fungus is not strange, since lichens are symbiotic growths composed of fungi and algæ. These observations would then acquire biological interest, since they indicate that lichen acids are produced by the metabolism of the fungous part of the symbiotic lichen rather than by that of the alga. Tobler[2] concluded that lichen acids are solely the product of symbiosis, because a number of fungi from lichens grown separately without algæ formed no lichen acids. Since the formation of penicillic acid is so very dependent upon a number of conditions, it may well be that Tobler did not find lichen acids because he did not happen to employ the necessary environmental conditions.

The penicillic acid described in this paper is not the substance obtained by Gosio, as shown by the fact that the latter gives a dif-

[1] Hesse, O. Beitrag zur Kenntnis der Flechten und ihrer charakteristischen Bestandteile. Journal für Praktische Chemie, n. F., Bd. 83, p. 22-96, 1911.

[2] Tobler, F. Das physiologische Gleichgewicht von Pilz und Alge in den Flechten. Berichte der Deutschen Botanischen Gesellschaft, Bd. 27, p. 421-427, 1909.

——— Zur Ernährungsphysiologie der Flechten. Berichte der Deutschen Botanischen Gesellschaft, Bd. 29, p. 3-12, 1911.

ferent color with ferric chlorid, that it is less soluble in cold water, and that it has a higher melting point, properties in which it resembles lichen acids even more than penicillic acid does.

Nevertheless, the properties and formulæ of the two substances are so similar that some genetic relationship between them is very probable. This possibility gains likelihood from the fact, demonstrated by Gosio, that in addition to the substance isolated by him other similar ones were present giving different colors with ferric chlorid. Moreover, in his cultures the ferric-chlorid reaction varied both in intensity and quality of color. The extract of only 5 to 10 cubic centimeters of culture fluid of some cultures gave a reaction with ferric chlorid, while others required 50 to 100 cubic centimeters.

Evidence of the occurrence of other substances was encountered in the course of this investigation. Some of the culture fluid which had been exhausted by extraction with chloroform was dehydrated by treating with plaster of Paris. The dry powdered mixture was extracted with chloroform and the chloroform evaporated. The residue with an odor like cresol consisted of a few yellowish crystals in a reddish oil. The material was somewhat toxic.

Other evidences in the same direction consist of observations made upon corn spoiled naturally by unidentified green molds and giving positive ferric-chlorid reactions. In every sample encountered in this laboratory the reaction appeared at once, not gradually, as with penicillic acid. The few attempts made to isolate penicillic acid from some of these samples failed, although the acid could readily be isolated from corn inoculated with *Penicillium puberulum*. Undoubtedly in the samples substances other than penicillic acid were responsible for the ferric-chlorid reaction.

Another indication of the presence in the culture fluids of unidentified substances is the considerable acidity of the culture fluids. Four hundred and eighty-five cubic centimeters of a four weeks' culture required 92.15 cubic centimeters n/10 sodium hydroxid for neutralization. From 485 cubic centimeters of this culture 0.2 of a gram of penicillic acid was isolated, equivalent to 11.1 cubic centimeters of n/10 alkali. The acidity of fresh Raulin's solution is due entirely to the tartaric acid, but at the end of growth no tartaric acid could be isolated. On the assumption that all the tartaric acid had been consumed and that all the penicillic acid had been extracted, a considerable quantity of acid remained undetermined.

Furthermore, there are great differences in the metabolism of the organism used by Gosio and of *Penicillium puberulum*. The former produced acetaldehyde, a little acetic acid, glycerin, and succinic acid;[1] the latter produced no more than traces of these substances.

[1] Gosio, B. Ricerche batteriologiche e chimiche sulle alterazione del mais. Rivista d'Igiene e Sanità Pubblica, ann. 7, p. 874, 1896.

Finally, a conclusive argument is the fact that, as shown below, it has been possible to isolate in quantity from another species, *P. stoloniferum* Thom, a new acid more nearly resembling Gosio's substance than penicillic acid does. These facts do not support Paladino-Blandini's [1] suggestion that the toxic material formed by a large number of fungi is identical.

Di Pietro's toxic principle also differs from penicillic acid in its insolubility in water and in the fact that with ferric chlorid it gave a grass-green, "verda-erba," color instead of a reddish one.[2] Di Pietro [3] believed the toxic principle to be a glucosid. His evidence for this view, as above stated, seems to have been based on the fact that the reducing power of his toxic extracts was lost by boiling in acid, but was not lost in neutral solution. But this is also true of penicillic acid, though there is no question of its being a glucosid. If it contained a carbohydrate it would char with concentrated sulphuric acid and would in all probability be optically active. It is not necessary for a substance to be a glucosid in order to have reducing power. Many lichen acids, for example, have this power. In another important way Di Pietro's extracts resemble penicillic acid. He says that the "glucosid" itself gives the reaction with ferric chlorid only very weakly and gradually. This is exactly the behavior of penicillic acid. If, however, says Di Pietro, its sugar be split off by hydrolysis an aromatic acid is set free, giving the ferric-chlorid reaction at once. Penicillic acid behaves in quite the same way when heated with mineral acid. It is not necessary to assume that either substance is a glucosid. Treatment with acid might merely saponify an ester. It is not necessary to assume, as does Di Pietro, that the substance giving the color is present in three forms, as free aromatic acid, as a salt of the acid, giving the reaction only after acidifying, and as a glucosid. There are so many resemblances between Di Pietro's extracts and penicillic acid that the question involuntarily suggests itself whether his extracts were not mixtures of an unknown very toxic substance and penicillic acid or some very closely related substance. Di Pietro's "glucosid," Gosio's substance, and penicillic acid equally resemble lichen acids. It does not necessarily follow that the very toxic substance or substances of the extract were responsible for the "glucosid" or ferric-chlorid reactions.

CONDITIONS OF PENICILLIC-ACID FORMATION.

On Raulin's medium penicillic acid seems to accumulate only at very definite oxygen tensions. Both a full and a very scanty supply

[1] Paladino-Blandini, A. Tossici di ifomiceti. Archivio di Farmacologia Sperimentale e Scienze Affini, v. 5, p. 606–664, 1906.

[2] Di Pietro, Melchiorre. Intorno al "Penicillio tossico." Rivista Pellagrologica Italiana, v. 3, p. 221, 1903.

[3] Di Pietro, Melchiorre. Glucosidi di elevato potere tossico trovato nelle spore del Penicillium glaucum. Rivista Pellagrologica Italiana, v. 2, p. 63, 1902.

of air are unfavorable. When to insure good aeration the organism was grown in rectangular quart bottles, known in the trade as "Long Blakes," turned on their sides and containing about 200 cubic centimeters of medium, no penicillic acid could be found except traces in cultures less than a week old. Under these conditions [1] the exposed surface of the medium is relatively great and the aeration is good because the neck of the bottle is only about 1 centimeter above the surface. It was hoped that under these conditions the growth of mycelium would be more rapid and the yield of penicillic acid greater than in the 2-liter Erlenmeyer flasks, in which the same amount of medium offers far less surface and the carbonic acid must diffuse upward more than 20 centimeters before escaping from the tightly plugged neck of the flask. Growth was actually better in the bottles, but the yields of penicillic acid were insignificant. Perhaps the good aeration accounts for the latter phenomenon, since penicillic acid is easily oxidized and since even in the Erlenmeyer flasks the yields were poor when they were very loosely plugged with cotton. On the other hand, too poor a supply of air also prevents the formation of penicillic acid, as was shown when the supply was almost completely cut off by tying thin rubber sheeting over the necks of flasks and bottles. This deficiency of yield may be due to the fact that, even after a month, growth is scanty and without spore formation, though it is somewhat better in the bottles than in the flasks. The culture medium remained white and transparent. When the seal was removed from one of the bottles, growth and spore formation became rapid. Within 24 hours the medium turned quite dark. After six days a very small quantity of penicillic acid could be isolated—rather more than was obtained from the 6-day control bottle that had never been sealed. The flasks and bottles that remained sealed failed to develop more than a trace of penicillic acid or of alcohol, even after two months, though more mycelium gradually developed.

This influence of aeration on the quality of the metabolism of Penicillium has not hitherto been noted. It furnishes a further explanation for some of the discordant results obtained with Penicillium. It shows, moreover, that the ferric-chlorid reaction when negative need not, as some have thought, give reliable information concerning the toxicity of a fungus. Under special conditions of oxygen supply, the reaction may become positive. Species of Penicillium which have not been found to give the reaction [2] will have to be reinvestigated under definite conditions of oxygen tension.

From media which are alkaline or become so, penicillic acid can not be isolated, perhaps because it is sensitive to alkali. Raulin's medium

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[1] This method was very kindly suggested by Dr. Hideyo Noguchi, of the Rockefeller Institute of New York.

[2] Cf. supra, p.12.

is such when deprived of tartaric acid, or when it contains an equivalent amount of sodium acetate instead of tartaric acid, or when it contains 1 per cent of Witte's peptone as the source of nitrogen. Tartaric acid itself is unessential, since succinic acid may be substituted for it without changing the quantity of penicillic acid formed. The failure to find penicillic acid on peptone medium is not in harmony with the observation of Paladino-Blandini[1] that toxic phenolic substances are most abundantly formed on media containing nitrogen in the form of peptone.

The form in which nitrogen is offered may also be of influence, since when tyrosin or leucin were the only source of nitrogen no penicillic acid was detected. As these experiments were performed before the effect of the air supply was known, they are not conclusive. At any rate the absence of the acid from the tyrosin culture is remarkable, as penicillic acid is probably also an aromatic compound. The suggestion advanced by Audenino,[2] that some of the phenolic substances found in spoiled maize may be derived from the phenylalanin of the zein, does not receive support from these experiments on tyrosin.

On corn-meal mush the organism grew faster and fruited sooner than on Raulin's medium. Penicillic acid was formed, though the quantities extracted were decidedly smaller than those from Raulin's medium, perhaps because of the greater difficulties of purification. The alcoholic extract of the mush on evaporation left a red oil, like that reported by Lombroso.[3] From the water extract of the oil the acid crystallized on concentrating and cooling. These cultures gave but a weak or even negative ferric-chlorid reaction, showing that in detecting moldiness the unmodified reaction of Gosio is either not delicate enough or is inhibited by other substances present.

It is therefore evident that the amount of penicillic acid formed varies greatly with the conditions of growth. From 700 cubic centimeters of medium from the 2-liter, tightly plugged, Erlenmeyer flasks small quantities were separable at the end of the first week, about 0.1 gram at the end of the second, 0.2 gram the third, and 0.25 gram at the end of the fourth week, making the final concentration about 1 to 2,000. The method of separation probably does not isolate all the substance present.

TOXICITY OF PENICILLIC ACID.

Penicillic acid contains more oxygen than the material isolated by Gosio. This may explain why the former is toxic and the latter is

[1] Paladino-Blandini, A. Tossici di ifomiceti. Archivio di Farmacologia Sperimentale e Scienze Affini, v. 5, p. 606–664, 1906.

[2] Audenino, E. Etiologia della pellagra. Atti del Quarto Congresso Pellagrologico Italiano, Udine, 23-24-25 Septembre, 1909, p. 41. 1910.

[3] Lombroso, Cesare, and Erba, Carlo. Sulle sostanze stricniche e narcotiche del mais guasto. Reale Istituto Lombardo, Rendiconti, s. 2, v. 9, p. 133–147, 1876.

not, for it is a well-known fact that the introduction of hydroxyl into the benzol ring increases toxicity.[1] Benzol and phenol are good examples of this phenomenon.

The toxicity of penicillic acid is shown by the following experiments: When 7 milligrams of penicillic acid, dissolved in 0.5 cubic centimeter of physiological salt solution, are injected subcutaneously into a mouse of about 20 grams weight, convulsions and paralysis, particularly of the forelegs, develop very quickly, so that the head and forward portion of the body fall down on the ground while the hind quarters are still struggling and kicking. After a while these violent symptoms wear off and the animal is quiet; usually it assumes a sitting posture, with fur ruffled, the vessels of the ears congested, muzzle twitching, and diaphragm contracting spasmodically, so that the mouse has the appearance of retching. The animal might be regarded as recovering. However, death usually comes quietly in 5 to 12 hours. Apparently the symptoms are identical with those produced by the extracts of the corn cultures. A smaller dose (5 milligrams) causes the initial symptoms of convulsions and paralysis, but the animal usually recovers. Death is not due to acid action, for the same symptoms follow the injection of the same dose in solutions neutralized with sodium hydroxid. Moreover, the injection of equivalent quantities of hydrochloric or sulphuric acid is without serious effect. The fatal dose for mice is therefore about 0.3 to 0.35 gram per kilogram when injected subcutaneously.

With a stomach tube a guinea pig of 525 grams weight was fed 10 cubic centimeters of a 2 per cent solution of penicillic acid in warm normal saline solution. In the course of half an hour the animal became weak, particularly in the hind legs, and lay prone on the abdomen. When turned on the back he remained in that position. An hour later he began to recover. When returned to the cage he sat quietly without eating. The following day the weight had dropped to 495 grams, and on the second day to 470 grams. On the third day the weight rose to 485 grams and the animal recovered.

Another guinea pig of 410 grams weight received subcutaneously in the region of the spinal column 1.8 cubic centimeters of 2 per cent penicillic acid in warm normal saline solution. At first the effects were not marked. The animal remained quiet with fur bristling. Then a little weakness of the hind limbs developed. A second injection of 2 cubic centimeters was made 4½ hours later beneath the skin of the abdomen. In both injections an appreciable amount of the injected liquid was lost by oozing through the needle puncture. The animal was observed for 2½ hours after the second injection. No change in its condition was noted. The following morning it was found dead. Autopsy revealed nothing but rather extensive serous

[1] Fränkel, Sigmund. Die Arzneimittel-Synthese, Aufl. 2, Berlin, 1906, p. 53.

effusions at the sites of injection. There was neither pus nor other sign of severe inflammation. The right heart was dilated, the left ventricle strongly contracted. In two injections a third guinea pig, a female of 400 grams weight, received under the skin of the abdomen 0.1 gram of penicillic acid neutralized with sodium carbonate and dissolved in 1.5 cubic centimeters of normal saline solution. As soon as returned to the cage it began to feed, but an hour later was quiet and not very responsive to stimuli. It remained in this condition for an hour longer, when slight dyspnœa and signs of weakness appeared. An hour later the legs were weak, for the abdomen was allowed to rest on the ground. It still responded to stimuli, however, and did not remain on its back when turned over. Half an hour later its head was also resting on the ground. At the end of the fourth hour the weakness was so great it fell over on its side from time to time, but was able to right itself. Four and a half hours after the injection the animal sank over on its side and died without struggling. The autopsy showed that one of the injections had been made into the region of the functioning mammary gland. There was at this place a hemorrhagic effusion about an inch in diameter. There was a little serous exudation. The site of the other injection showed merely a little serous exudation. The animal was in an early state of pregnancy. Nothing else significant could be found.

Twelve milligrams of penicillic acid were dissolved in 1 cubic centimeter of normal saline solution by neutralizing with sodium carbonate. This was injected into the dorsal lymph sac of a frog of 45 grams weight. Though the frog was watched for 6 hours no symptoms were observed. Nevertheless, the following morning the animal was dead. The only thing noted on autopsy was that the heart was in systole.

A second frog of 35 grams weight received into the dorsal lymph sac 30 milligrams of penicillic acid dissolved with sodium carbonate. He soon began to get weak, without any other marked symptoms, and in 45 minutes had ceased breathing and was quite unresponsive. The body was at once opened. The auricles were still beating vigorously, but the ventricle was quiet in complete systole. It exhibited, only occasionally, slight fibrillation in the neighborhood of the auriculo-ventricular junction. This soon ceased, and 10 minutes later the auricles ceased pulsating.

Penicillic acid is also toxic to tadpoles. In a solution of 1 to 5,000 the animals seemed uncomfortable and restless as compared with the controls. There seemed to be considerable irritation, for the water became clouded with secreted mucus. At the end of 24 hours the animals were still in good condition and, when transferred to the aquarium, continued to live. In a solution of 1 to 2,000 similar phenomena were observed, but after an hour there were occasional convulsive lashing movements. In the third hour the animals seemed to get weaker and the respiration became weak and shallow. At the end of 4 hours the animals no longer responded to stimuli, the respiration had almost ceased, and the animals did not recover when placed in the aquarium. In a solution of 1 to 1,000 the irritation and the lashing movements were more pronounced. For 2 hours there were fairly vigorous responses to stimuli; in 3 hours the animals were dead. The toxic effect was not observed when the tadpoles were placed in solutions of the neutral sodium salt. There was very little irritation, and they survived in solutions as strong as 1 to 1,000. For tadpoles the acidity of penicillic acid seems to be the chief factor in the toxic action, for in solutions of sulphuric and hydrochloric acid of a strength of 1 to 1,000 the animals were killed in 18 minutes. Moreover, in n/200 sulphuric acid, which has an acidity equivalent to that of a 1 to 1,000 solution of penicillic acid, the tadpoles died in an hour. These observations do not militate against the toxicity of penicillic acid, for it is probable that it is not to any considerable extent absorbed, because only minute traces, as has been shown, can be extracted from water by ether or by oil. The partition coefficient is too much in favor of water, and Overton [1] has shown that substances like these enter the organism very slowly. All these factors are still more effective in the case of solutions of the salt, which is quite insoluble in oils and most organic solvents. This is true in many other cases where the free acid is itself toxic aside from its acidity, but the salt quite harmless. Thus, Clark,[2] in whose paper the older literature is cited, states that the toxicity of acetic acid to Penicillium and certain other molds is due almost entirely to the un-ionized molecule CH_3COOH. Recently this fact has been rediscovered by Reichel.[3] It was found in this investigation that even in neutralized solutions of salicylic acid of 1 to 1,000 tadpoles are unaffected. Finally Gosio and Ferrati [4] state that neutralization of the toxic extracts prepared from their Penicillium cultures renders them much less toxic.

From all these data it appears that the toxicity of penicillic acid is of the same general magnitude as that of some of the phenols and phenol acids. According to Duplay and Maurice Cazin [5] the lethal dose of phenol for mice is 0.296 gram per kilogram. This is nearly as large

[1] Overton, E. Studien über die Narkose, zugleich ein Beitrag zur allgemeinen Pharmakologie, Jena, 1901, 195 p.

[2] Clark, J. F. On the toxic effect of deleterious agents on the germination and development of certain filamentous fungi. Botanical Gazette, v. 28, p. 319, 1899.

[3] Reichel, J. Ueber das Verhalten von Penicillium gegenüber der Essigsäure und ihren Salzen. Biochemische Zeitschrift, Bd. 30, p. 152–159, 1911.

[4] Gosio, B., and Ferrati, E. Sull' azione fisiologica dei veleni del mais invaso da alcuni ifomiceti. Rivista d'Igiene e Sanità Pubblica, ann. 7, p. 961–981, 1896.

[5] Duplay, Simon, and Cazin, Maurice. De l'action de l'acide phénique sur les animaux. Comptes Rendus de l'Académie des Sciences [Paris], t. 112, p. 627–630, 1891.

as the lethal dose of penicillic acid. For guinea pigs and rabbits the lethal dose of phenol is about twice as much. For guinea pigs the lethal dose of penicillic acid seems to be smaller.

The data of Duplay and Cazin agree quite well with the findings of Paul Binet, who determined the lethal dose for rats at 0.5 to 0.6 gram and for guinea pigs at 0.55 gram per kilogram.[1] Gosio and Ferrati[2] state that 5 to 10 milligrams of phenol or resorcin and 15 milligrams of salicylic acid subcutaneously are fatal to mice.

Granting that penicillic acid is related to lichen acids, it becomes a matter of interest to note that certain lichen acids have been found toxic. Kobert[3] has found that vulpinic acid, a yellow substance found in *Evernia vulpina; Xantoria parietina*, and some other lichens, is a protoplasmic poison. He found both vulpinic and pinastric acid very toxic to frogs, and vulpinic and pinastric acids toxic to higher animals, while chrysophyscin is harmless. Zopf[4] found none of them except vulpinic acid injurious to orthoptera, arachnids, or helix. He does not think the lichen acids protect the lichen from attacks by animals, a view previously expressed by Bachmann[5] and Zukal.[6] Stahl,[7] however, believes that Zopf is only partly right.

Penicillic acid was found to resemble the crystalline substance of Gosio and the lichen acid, vulpinic acid, studied by Kobert,[8] in having some antiseptic power, as shown by its action on pure cultures of baker's yeast and of *Bacillus coli*. Yeast was chosen because it flourishes when the reaction is acid. *B. coli* was selected because it forms acid from sugar and is not harmed by considerable variations of the environment. The culture was kindly furnished by Dr. Rogers, of the Dairy Division of the Bureau of Animal Industry, being No. 22 f. g. of his collection. It had been isolated originally in the Hygienic Laboratory of the Public Health and Marine-Hospital Service.

Yeast.—Six flasks, provided with a trap containing sulphuric acid to prevent moisture losses, received each 25 cubic centimeters of sterile Raulin's medium. Three received in addition enough crystalline penicillic acid to make the concentration 1 to 1,000. All

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[1] Binet, Paul. Toxicologie comparée des phénols. Revue Médicale de la Suisse Romande, ann. 15, p. 567, 631, 1895.

Prevost, and Binet, Paul. Travaux du Laboratoire de Thérap., Genf, 1896, p. 143. Cited in Fränkel, Sigmund, Die Arzneimittel-Synthese, Aufl. 2, Berlin, 1906, p. 55.

[2] Gosio, B., and Ferrati, E. Op. cit.

[3] Kobert, R. Über Giftstoffe der Flechten. Sitzungsberichte der Naturforscher-Gesellschaft. Dorpat, Bd. 10, p. 165, 1892.

[4] Zopf, W. Zur Biologischen Bedeutung der Flechtensäuren. Biologisches Centralblatt, Bd. 16. p. 592-610, 1896.

[5] Bachmann, E. Ueber nicht-krystallisirte Flechtenstoffe, ein Beitrag zur Chemie und Anatomie der Flechten. Jahrbücher für Wissenschaftliche Botanik, Bd. 21, p. 17, 1890.

[6] Zukal, H. Morphologische und biologische Untersuchungen über die Flechten. Sitzungsberichte der Mathematisch-Naturwissenschaftliche Classe der Kaiserlichen Akademie der Wissenschaften [Wien], Abt. 1, Bd. 104, p. 529-574, 1303-1395, 3 pl., 1895.

[7] Stahl, Ernst. Die Schutzmittel der Flechten gegen Tierfrass. Festschrift zum Siebzigsten Geburtstage von Ernst Haeckel, Jena, 1904, p. 357. (Denkschriften der Medizinisch-Naturwissenschaftlichen Gesellschaft zu Jena, Bd. 11.)

[8] Kobert, R. Op. cit.

six were then inoculated with equal quantities of a suspension of the yeast and weighed. At intervals a pair of these flasks was heated to boiling and dry air drawn through them. They were cooled and weighed. The loss represents chiefly carbonic acid. The experiment was repeated, using a concentration of penicillic acid of 1 to 500. The results obtained are shown in Table III.

TABLE III.—*Loss in weight of flasks of Raulin's medium treated with various quantities of penicillic acid and inoculated with yeast.*

Time.	Loss in weight (grams) of flasks inoculated with yeast.			
	Proportion of penicillic acid 1 to 1,000.	Control (untreated).	Proportion of penicillic acid 1 to 500.	Control (untreated).
First day	0.4507	0.4537	0.3117	0.3566
Second day	.4820	.5482	.3535	.5308
Third day	.5293	.5720	.5550	.5755

In the control flasks no sugar was present after the third day. It was impossible to test the others because of the presence of penicillic acid, which is not destroyed by yeast but may be recovered almost quantitatively.

While these experiments do not disclose a powerful antifermentative action, still in both some decided effect is indisputable. While the yeast cells were not killed, there is an unmistakable retardation in the rate of fermentation. As far as could be judged by the naked eye, the multiplication of the cells was partly inhibited. At the end of the experiments there seemed to be much more yeast in the control flasks than in those containing penicillic acid. Whether the gross appearance corresponds to the facts will have to be determined by cell counts. It is not likely that these effects are due to the slight increase in acidity.

Bacillus coli.—For the culture medium 1 per cent acid bouillon was used. Series of test tubes were charged with 8 cubic centimeters. To the control tubes 1 cubic centimeter of normal salt solution was added. To the other tubes 1 cubic centimeter of a neutral solution of the sodium salt of penicillic acid was added of such a strength that the final concentration was 1 to 500. Both sets of tubes were sterilized for 20 minutes on three successive days in an Arnold sterilizer. A very dilute suspension of active bacteria was prepared and 1 cubic centimeter measured into each tube. The tubes were then incubated for 20 hours. When examined it was found that in the tubes containing penicillic acid there was very slight growth, whereas in the control tubes growth was abundant. The difference was so apparent that it was unnecessary to pour plates. As the neutral sodium salt was used in this experiment there was no increased acidity, an objection which might be urged against the yeast experiments.

These experiments show that penicillic acid exerts some, though weak, antifermentative action. Stahl [1] has pointed out that the antiseptic power of vulpinic acid may be of considerable use to the lichen *Evernia vulpina* in protecting it from micro-organisms. One might advance a similar hypothesis for the secretion of penicillic acid. While it is likely that execretory products of one kind or another have this effect, their usefulness must be regarded as purely accidental and secondary, though it is not impossible that they play a part in selection. The theory that the formation of alcohol in the course of fermentation is for the purpose of suppressing competitors [2] must in this form be condemned.[3] One might as well maintain that the formation of carbonic acid is for the purpose of furnishing the yeast cell with a means of locomotion or for the purpose of creating the optimum acidity for the action of invertase.[4] It is true that alcohol and carbonic acid do these things for the yeast cell, but the phenomena are merely concomitant. The yeast experiments given above may indicate that penicillic acid has such a secondary effect, particularly when the oxygen supply to the organism is insufficient.

These observations on the antifermentative action of penicillic acid may furnish an explanation of the observation of Nikitinsky [5] that yeast will not grow on a medium on which Penicillium has been sown one or two days before, and vice versa. This is not due to the exhaustion of the medium, for replacing the sugar consumed does not alter the results. On the whole, Nikitinsky is inclined to regard the acids formed by the first organism as the agent inhibiting the growth of the second. He draws this conclusion because acids are the only inhibiting substances known to be formed. The discovery of penicillic acid renders it possible that some of these inhibitions are due to the formation of specific substances. They may be very generally formed, for Nikitinsky found the same inhibiting effect with other pairs of organisms than yeast and Penicillium.

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[1] Stahl, E. Op. cit., p. 374.

[2] Wortmann, Julius. Tätigkeit der Station in Bezug auf die Untersuchung und Behandlung kranker Weine. Bericht der Königl. Lehranstalt für Wein-, Obst-, und Gartenbau zu Geisenheim, p. 136, 1902.

Delbrück, M. Der physiologische Zustand der Zelle und seine Bedeutung für die Technologie der Gärungsgewerbe. Wochenschrift für Brauerei, Jahrg. 23, p. 513-516, 1906.

——— and Schönfeld, F. System der natürlichen Hefereinzucht, Berlin, 1903, p. 10.

Jost, Ludwig. Vorlesungen über Pflanzenphysiologie, Jena, 1904, p. 258.

Kienitz-Gerloff, Felix. Bakterien und Hefen, etc., Berlin, 1904, p. 35.

[3] Bierberg, W. Die biologisch-ökologische Theorie der Gärung. Centralblatt für Bakteriologie, [etc.], Abt. 2, Bd. 26, p. 187-189, 1910.

——— Alkohol- und Essigsäuretoleranz der Bakterien und die Wortmannsche biologische Gärungstheorie. Idem, Abt. 2, Bd. 24, p. 432-435, 1909.

[4] Hudson, Dr. C. S. Personal communication.

[5] Nikitinsky, J. Ueber die Beeinflussung der Entwickelung einiger Schimmelpilze durch ihre Stoffwechselprodukte. Jahrbücher für Wissenschaftliche Botanik, Bd. 40, p. 66, 1904.

It is not possible, however, to reach any definite opinion concerning his experiments, because they are complicated by the fact that he does not state how the medium was sterilized after the growth of the first organism. If sterilized with heat, it is possible that inhibiting substances were injured or volatilized.

A detailed study of the pharmacological action of penicillic acid on warm-blooded animals was not made, because to procure enough material would have unduly delayed publication. Such a study is reserved for a future paper. A single experiment indicating the effect on the blood pressure was performed, using a 6-kilogram dog in morphine-ether anæsthesia. The blood pressure in the carotid was recorded upon a kymograph in the usual way. In the course of 12 minutes 23 cubic centimeters of a 2 per cent solution of penicillic acid in physiological salt solution were introduced into the saphenous vein. Somewhat before the end of the injection the blood pressure began to fall and ultimately reached 65 per cent of the original value. Then it gradually recovered, until at the end of 10 minutes it had returned to its original value. During the stage of depression the heartbeat was slightly more rapid, but it also recovered.

It is a matter of considerable interest to compare the toxicity of penicillic acid with that of the extracts obtained by various investigators from various fungi. The only experiments strictly comparable with those on penicillic acid just recorded are those with Gosio's pure substance, in which no toxic effect was obtained. All other recorded experiments deal with impure mixtures. The extracts obtained by Gosio, Di Pietro, Sturli, and others were far more toxic than penicillic acid. This is particularly true of Di Pietro's "glucosid." One and a half milligrams of his impure extract caused within a quarter of an hour intense acute symptoms in a guinea pig of 300 grams.

Toxic preparations were obtained by Gosio's student, Paladino-Blandini,[1] from the toxic variety of *Penicillium glaucum* used by Gosio ("Var. ipertossico, Gosio"), from Oospora, Rhizopus, and Aspergillus. One does not, on carefully examining his protocols, get the impression that any of the organisms were very poisonous. This may have been due to the fact that his culture media were often alkaline or that he concentrated the media on the steam bath before testing for toxicity. If any of the organisms produced substances like penicillic acid these might have been destroyed either by the alkalinity or by the heating, for, as has been shown, penicillic acid is unstable under these conditions. Paladino-Blandini makes this point himself for the poison of *P. glaucum*, which he found to be ther-

[1] Paladino-Blandini, A. Tossici di ifomiceti. Archivio di Farmacologia Sperimentale e Scienze Affini, v. 5, p. 606–644, 1906.

molabile and volatile. Obviously in these experiments he was not dealing with *P. puberulum*.

Paladino-Blandini used these observations to explain some of the results of Ceni,[1] who claimed that the production of toxic or phenolic substances by various molds fluctuated with the season, being greatest in the spring and fall. Since Ceni always injected inspissated culture fluid or extracts, Paladino-Blandini suggested that the heating probably destroyed or volatilized the toxic substances, so that conditions were not constant. Di Pietro, too, was unable to confirm Ceni, for his organism was equally toxic all the year round. In the present investigation these results were checked by determining the amount of penicillic acid formed under like conditions at different times of the year. About the same yield of this acid was obtained in every month of the year. Against these observations must be set the statements of Otto [2] that extracts of Aspergillus cultures are toxic from April to October.

PHYSIOLOGICAL STUDIES OF PENICILLIUM PUBERULUM.

In order to obtain, if possible, better yields of penicillic acid, the organism was grown upon Raulin's medium, variously modified. The other conditions of growth remained the same as those previously used to procure penicillic acid.

The culture media employed may be classed in two groups: Those in which some component of Raulin's medium was replaced by an equivalent amount of another substance, and those in which some substance was added to Raulin's medium. The first group comprised the following substitutions:

Succinic acid for tartaric.

Sodium acetate for tartaric acid.

One and a half per cent ethyl alcohol for cane sugar and tartaric acid.

Leucin instead of nitric acid and ammonia. The amount of leucin was calculated to contain an equivalent amount of nitrogen. The leucin was pure and prepared by hydrolyzing plant protein, esterifying the resulting amino acids, subjecting them to fractional distillation, and saponifying.[3]

Tyrosin instead of nitric acid and ammonia. The tyrosin was prepared in the usual way by hydrolyzing horn with strong mineral acids.

One-tenth per cent amyl alcohol for cane sugar and tartaric acid.

Witte's peptone for nitric acid and ammonia.

The second group comprised the following:

Addition of 0.05 per cent amyl alcohol to Raulin's medium.

Addition of 0.01 per cent amyl alcohol to Raulin's medium.

Addition of 0.1 per cent amyl alcohol to Raulin's medium.

Most of the results have been collected in Table IV.

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[1] Ceni, Carlo. Le proprietà tossiche di alcuni ifomiceti in rapporto colle stagioni e col ciclo annale dell' endemia pellagrosa. Comunicazione preliminare. Rivista Pellagrologica Italiana, v. 4, p. 89.

[2] Otto, M. Ueber die Giftwirkungen einiger Stämme von Aspergillus fumigatus und Penicillium glaucum nebst einigen Bemerkungen über Pellagra. Zeitschrift für Klinische Medizin, Bd. 59, p. 332–333, 1906.

[3] This preparation was very kindly furnished by Dr. I. K. Phelps, of the Bureau of Chemistry.

TABLE IV.—*Observations on the physiology of Penicillum puberulum.*

Medium.	Age of culture.[1]	Appearance of spores.[2]	Color of medium.	Weight of mycelium.	Volatile products.	Ethyl alcohol.	Acidity of 10 c. c. of medium.	Penicillic acid.	Remarks.
	Days.	*Day.*		*Grams.*					
Raulin's medium.	30–40	6th to 10th	Brown, dark, transparent.	8. Some already autolyzed.	No aldehyde; traces of acid, none with insoluble Ag. salts.	0.1 per cent	1.1 c. c. n/10 alkali.	About 0.2 gram in 700 c. c.	
Succinic acid	31	About the 6th.	do	4.75	Traces of aldehyde and acetic acid.	Present	Not determined.	Present	
Sodium acetate.	31	10th traces, 15th abundant.	Darkening delayed.	Not weighed	Not studied	Not studied	Alkaline	Absent	Spore formation much delayed;[3] culture medium pale.
Ethyl alcohol 1.5 per cent.	70	About the 60th	Pale	Very slight	do	do	Not studied	Not tested	Spores did not germinate; after inoculation with mycelium very slow and scanty growth.
Witte's peptone.	25	9th	Very dark	Not weighed	Absent	Absent	Alkaline	Trace	A trace of an alkaloid, soluble in chloroform, present.
Leucin	33	8th	Light brown	7.7	No amyl alcohol; volatile acid-reducing Ag. present.	Present	0.6 c. c. n/10 alkali.	Absent	Very abundant spore formation.
Tyrosin	38	5th	do	4.25	No tyrol or tyrosol; volatile acid-reducing Ag. present.	Not studied	2.7 c. c. n/10 alkali.	do	Rapid, but very flat, thin, and delicate growth; strong musty odor.
Amyl alcohol:									
0.1 per cent									No growth.
0.05 per cent		6th to 10th	As in plain Raulin's medium.	Not weighed	Amyl alcohol present.	Present	Acid	Present	Growth good.
0.01 per cent		do	do	do	do	do	do	do	Do.
0.1 per cent									Almost no growth.

[1] The age refers to the interval between inoculation and chemical examination.
[2] The first tinge of green coloration visible to the naked eye was taken as a sign of spore formation. Many colorless or a few colored spores may have been formed sooner.
[3] Hasselbring (Botanical Gazette, v. 45, p. 170, 1908) reported abundant spore formation with potassium acetate as the only source of carbon.

A number of other facts were observed which have not been incorporated in the table. Thus it was found that in the cultures on Raulin's medium the tartaric acid soon disappeared. At least none could be isolated as the acid potassium salt. Indeed, the tartaric acid seems to be attacked at the beginning more rapidly than the sugar. Pfeffer [1] has reported analogous observations upon Aspergillus. Reichel[2] found very recently that Penicillium when grown in the presence of acetic acid begins to destroy the acid until the unfavorable acidity is reduced. A concentration of acetic acid equivalent to the tartaric acid of Raulin's medium was found to inhibit the development of *Penicillium puberulum.* That is why in one of the experiments of Table IV sodium acetate was used. There seems to be a general tendency for molds to reduce the acidity of a very acid medium. This may be done by destroying the acid, or, if the acid can not be attacked, by neutralizing it with ammonia when this can be formed by deamidization.[3] At any rate in the present instance the sugar disappeared more slowly than the tartaric acid. By the end of the fourth week, however, the medium was no longer optically active[4] or fermentable. Because penicillic acid renders the solution antiseptic the fermentation test is not in this instance reliable. Nevertheless the medium reduces Fehling's solution, 25 cubic centimeters yielding 78.1 milligrams of cuprous oxid.[5] The reduction is caused by penicillic acid.

Alcohol was determined by taking the specific gravity of the rectified distillate. The cultures contained 0.1 per cent as early as the end of the first week. The low concentration of alcohol found might not be the result of a scanty alcohol formation, but of the further oxidation of the alcohol formed, since alcohol is a good food for Penicillium.[6] Indeed, it has been said that "P. glaucum" does not produce alcohol at all.[7] Dox states that alcohol of a concentration not over 0.1 per cent is produced only when the air supply is insufficient.[8] All cultures of *Penicillium puberulum* tested contained alcohol, even those grown in the flat bottles above described, in which the aeration was certainly good.

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[1] Pfeffer, W. Ueber die Election organischer Nährstoffe. Jahrbücher für Wissenschaftliche Botanik, Bd. 28, p. 205-268, 1895.

[2] Reichel, J. Ueber das Verhalten von Penicillium gegenüber der Essigsäure und ihren Salzen. Biochemische Zeitschrift, Bd. 30, p. 152-159, 1911.

[3] Butkewitsch, Wl. Umwandlung der Eiweissstoffe durch die niederen Pilze im Zusammenhange mit einigen Bedingungen ihrer Entwickelung. Jahrbücher für Wissenschaftliche Botanik, Bd. 38, p. 198, 1902.

[4] The determinations were very kindly made by Dr. C. S. Hudson, of the Bureau of Chemistry.

[5] This determination was very kindly made by Dr. H. Hasselbring, of the Bureau of Plant Industry.

[6] Hasselbring, Heinrich. The carbon assimilation of Penicillium. Botanical Gazette, v. 45, p. 176-193, 1908.

[7] Brefeld, Oscar. Ueber Gährung. III. Vorkommen und Verbreitung der Alkoholgährung im Pflanzenreiche. Landwirthschaftliche Jahrbücher, Bd. 5. p. 315, 1876.

[8] Dox, A. W. The intracellular enzyms of Penicillium and Aspergillus, with special reference to those of Penicillium camemberti. U. S. Department of Agriculture, Bureau of Animal Industry, Bulletin 120, p. 33. 1910.

No glycerin could be detected in the culture fluid when 200 cubic centimeters, rendered weakly alkaline with sodium carbonate, were concentrated to a sirup, the sirup extracted with alcohol, and the alcoholic extract after evaporation tested with the borax bead.

Though the Raulin medium cultures remained distinctly acid to litmus, no nonvolatile acid other than penicillic acid could be extracted with sulphuric ether, petroleum ether, or acetic ether, even after acidifying with phosphoric acid. No insoluble lead, copper, calcium, barium, or zinc salt could be obtained. Great care was taken to detect oxalic acid, but in young cultures none could be found. Even in old cultures none could be detected by the ordinary method of extraction with ether. A small amount was isolated in the following manner: Seven hundred cubic centimeters of culture medium about two months old were concentrated to a sirup, acidified with phosphoric acid, and mixed with clean sand and plaster of Paris. When the plaster had set, the mass was ground and extracted with ether in a Soxhlet extractor. Oxalic acid, identified by its melting point, crystallized from the extract, which also contained other material, as shown by the evolution of gas and the odor of nitrous oxid. Apparently some nitric acid passed from the medium into the extract and there caused decomposition. The extract contained a substance soluble in chloroform and giving a bright-green color with ferric chlorid. An alkaline solution of penicillic acid when concentrated to a sirup does not yield oxalic acid, but it does give a green color with ferric chlorid. Fumaric acid was absent. As already indicated, the culture medium contained unidentified substances. This was further shown by the fact that by the method of Griess, using sodium nitrite, sulphanilic acid, and acetic acid, an azo dye of a beautiful carmine color was produced. This reaction was obtained by Raciborski[1] with a number of fungi.

Only very small quantities of volatile material other than alcohol were detected in the culture medium. For the purposes of this examination a culture medium from which the penicillic acid had been removed as thoroughly as possible with chloroform to avoid obtaining its decomposition products, was used. The medium was then distilled and the distillate extracted, first with chloroform and then with ether. The residue from the chloroform consisted of a few small white crystals with a melting point of 112° C. The residue from the ether consisted of a few fine hairlike crystals. None of the crystals gave the ferric-chlorid reaction. Distillation of this liquid with mineral acid yielded no other products. Distillation with

[1] Raciborski, M. Über die Assimilation der Stickstoffverbindungen durch Pilze. Bulletin International de l'Académie des Sciences de Cracovie. Classe des Sciences Mathématiques et Naturelles, ann. 1906. p. 733-770. 1907.

alkali yielded a few white crystals obtained by extracting the distillate with ether.

All these experiments refer to the unmodified Raulin's medium. The mycelium was also studied. From mycelium of varying age grown under different conditions no toxic material could be extracted with boiling alcohol. An oily, waxy residue remained after the alcohol was removed on the steam bath. When this residue was extracted with water and the extract injected subcutaneously into mice no serious toxic effects were observed.

Mannitol has long been known as a constituent of Penicillium.[1] Trehalose and trehalum, or substances resembling them, have also been described. It is possible that in fungi some investigators may have mistaken trehalum for starch or glycogen.[2] Cramer [3] found that by treating the spores of Penicillium with boiling water no carbohydrate material precipitable by alcohol passed into the extract. When, however, the spores were exhausted with ether before the extraction, a carbohydrate was obtained giving a deep blue color with iodin. This carbohydrate, Cramer thought, resembled hemicellulose, but it may have been similar to trehalum. Moreover, a direct relationship between mannitol, trehalose, and trehalum has been demonstrated, trehalose being formed only at certain stages of growth, while later only mannitol occurs.[4]

Mannitol was readily detected in the mycelium of *Penicillium puberulum* by extracting the mycelium dried in air with boiling alcohol. On cooling, sweet, fine, white, silky needles separated, soluble in water and alcohol, insoluble in chloroform, and with a melting point of 162° to 163° C., uncorrected. Cholesterol reactions were negative. They did not reduce Fehling's solution, though they did so after oxidation with nitric acid. They rotated polarized light slightly to the left.

In the mycelium dried in air neither trehalose nor trehalum could be detected. When fresh mycelium was immersed in boiling alcohol, as soon as removed from the culture flask and the boiling extract filtered, no trehalose separated on cooling. This extracted mycelium, boiled with water and filtered hot, gave, on cooling, a small quantity of gummy material which iodin colored intensely violet and which was not easily inverted by hot dilute hydrochloric acid. This substance is plainly trehalum, in no way mistakable for glycogen or the more readily soluble and easily inverted starch.

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[1] Zopf, Wilhelm. Die Pilze, Breslau, 1890, p. 125.

[2] Lippmann, E. O. von. Die Chemie der Zuckerarten, Aufl. 3, Halbbd. 2, Braunschweig, 1904, p. 1429–1430.

[3] Cramer, E. Die Zusammensetzung der Sporen von Penicillium glaucum und ihre Beziehung zu der Widerstandsfähigkeit derselben gegen äussere Einflüsse. Archiv für Hygiene, Bd. 20, p. 197–210, 1894.

[4] Lippmann, E. O. von. Op. cit., p. 1427.

Both the culture fluid and the mycelium were examined for oxidizing enzyms. The former contains an abundance of catalase, though no oxidase detectable by guajac, aloïn, or benzidin. A very faint peroxidase reaction was found, due perhaps to the presence of chlorids.[1] The statement of Loew [2] that filtered *Penicillium glaucum* cultures contain only catalase is therefore amply confirmed. Fresh and air-dry mycelia were ground in a mortar with distilled water and allowed to digest at room temperature for several hours. The extract, filtered through paper, contained far more catalase than the culture medium, but neither oxidase nor peroxidase could be detected by the color tests. In performing these tests great care was taken to vary the reaction, for this has been shown to influence these tests greatly.[3]

The mycelium was then tested for oxidizing power by the method of oxygen absorption as developed in this laboratory by Dr. H. H. Bunzel.[4] The air-dry mycelium was ground and the dry powder obtained used directly in the oxidase apparatus in the presence of pyrogallol and of tyrosin. No oxygen absorption was observed. To make certain that neither the drying nor the acid of the medium was accountable for the negative results, the organism was grown on a medium containing monosodium phosphate and disodium phosphate, which, as shown by Henderson and Webster,[5] remains neutral. This medium was Raulin's solution, to which was added 5 per cent of a mixture of two parts Na_2HPO_4 and one part NaH_2PO_4. On this medium few spores developed in 10 days. The mycelium remained colorless. The medium contained alcohol and only traces of penicillic acid. Ten grams of perfectly fresh mycelium of 12 days' growth were ground with clean sand and then transferred to the absorption flask, together with 4 cubic centimeters of 10 per cent pyrogallol. This concentration of pyrogallol was selected because of its antiseptic action. Under these conditions no oxygen absorption was observed for more than an hour. Absorption then gradually began. This phenomenon is still under investigation.

Some of the results obtained with the variously modified Raulin's medium require further comment.

In the distillate from the succinic-acid culture aldehyde was detected by the power to reduce ammoniacal silver, and acetic acid by the formation of the ethyl ester with its characteristic odor.

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[1] Alsberg, C. L. Beiträge zur Kenntnis der Guajak-Reaktion. Archiv für Experimentelle Pathologie und Pharmakologie, Festschrift Schmiedeberg, Supplementband, p. 39, 1908.

[2] Loew, Oscar. Catalase, a new enzym of general occurrence, with special reference to the tobacco plant. U. S. Department of Agriculture, Report 68, 1901.

[3] Alsberg, C. L. Op. cit.

[4] Bunzel, H. H. The measurement of the oxidase content of plant juices. U. S. Department of Agriculture, Bureau of Plant Industry, Bulletin 238, 1912.

[5] Henderson, L. J., and Webster, H. B. The preservation of neutrality in culture media with the aid of phosphates. Journal of Medical Research, v. 16, p. 1–5, 1907.

The observation made on the 1.5 per cent ethyl-alcohol culture that mycelium failed to develop from spores, whereas inoculation with pieces of mycelium resulted in growth, is in harmony with the statement of Duclaux [1] that while alcohol restrains or arrests the germination of mold spores it is utilized almost as abundantly as sugar by the adult plant. These observations were confirmed by Clark.[2] The mycelium of *Penicillium puberulum* developed very slowly from mycelium inoculation on 1.5 per cent alcohol. In the course of a few weeks a delicate, thin, green growth spread over the surface of the medium. Except for the green color it had the appearance of the scum of lead oxid that forms on the surface of molten lead exposed to the air.

The peptone cultures turned exceedingly dark. When the alkaline medium was extracted with chloroform, the residue of the extract consisted of a little oil which was for the greater part soluble in acid. The acid solution gave a decided precipitate with Meyer's reagent for alkaloids. The small quantity available was not toxic to mice. When the medium was acidified before extraction a little nontoxic oil passed into the chloroform. By extracting with warm water no penicillic acid was obtained. However, on long standing with ferric chlorid the extract developed a faint rose color.

The purpose of the leucin and tyrosin cultures was to ascertain whether *Penicillium puberulum* is able to deamidize amino acids as yeast does. If this were the case amyl alcohol would have been found in leucin cultures and tyrosol or tyrol in tyrosin cultures.[3] Amyl alcohol was sought by the method of Beckmann,[4] and tyrol and tyrosol by the method of Ehrlich.[5] Raciborski [6] first observed differences in the manner in which different molds decompose tyrosin. He found that *P. glaucum* grown on tyrosin agar produced a substance reducing silver. Since both leucin and tyrosin are destroyed by *P. puberulum*, it is evident that either their decomposition differs from that to which these amino acids are subjected by yeast, or else amyl alcohol, tyrol, and tyrosol are merely intermediary products.

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[1] Duclaux, E. Sur la nutrition intracellulaire. Annales de l'Institut Pasteur, ann. 3, p. 97-112, 1889.

[2] Clark, J. F. On the toxic effect of deleterious agents on the germination and development of certain filamentous fungi. Botanical Gazette, v. 28, p. 385, 1899.

[3] After the completion of these experiments, Ehrlich and Jacobsen reported that *Penicillium glaucum* is able to decompose amino acids to simpler compounds of lower molecular weights. Still more recently, Herzog and Saladin published similar results. See Ehrlich, Felix, and Jacobsen, K. A., Über die Umwandlung von Aminosäuren in Oxysäuren durch Schimmelpilze. Berichte der Deutschen Chemischen Gesellschaft, Jahrg. 44, p. 888-897, 1911; Herzog, R. O., and Saladin, O., Über das Verhalten einiger Pilze gegen Aminosäuren, Zeitschrift für Physiologische Chemie, Bd. 73, p. 302-307, 1911.

[4] Beckmann, Ernst. Zur Bestimmung des Fuselölgehaltes alkoholischer Flüssigkeiten. Zeitschrift für Untersuchung der Nahrungs- und Genussmittel, Bd. 10, p. 143-152, 1905.

[5] Ehrlich, Felix. Über die Vergährung des Tyrosins zu *p*-oxyphenyl-äthylalkohol (Tyrosol). Berichte der Deutschen Chemischen Gesellschaft, Jahrg. 44, p. 139-146, 1911.

[6] Raciborski, M. Über die Assimilation der Stickstoffverbindungen durch Pilze. Bulletin International de l'Académie des Sciences de Cracovie. Classe des Sciences Mathématiques et Naturelles, ann. 1906, p. 767. 1907.

That the latter is probably the case is indicated by the fact that penicillic acid is formed abundantly only under conditions of imperfect aeration. Perhaps under these conditions tyrosin and leucin would be less completely oxidized. It certainly is significant that yeast which grows anaerobically produces tyrosol or *p*-oxyphenyl alcohol from tyrosin, a substance which resembles penicillic acid in bitter, toxic,[1] and certain chemical properties. Another observation of Raciborski[2] suggests a different explanation of the results. He found that *Aspergillus niger* in a condition of carbon hunger decomposes tyrosin in a different way than when it is plentifully supplied with sugar. It is possible that since *P. puberulum* was allowed to grow more than a month it was, during the latter period of its growth, in a state of carbon starvation because all the sugar had been consumed.

The amyl-alcohol cultures were designed to learn whether this alcohol could be oxidized by *P. puberulum.* It was thought that in this way some indication might be given showing whether it was an intermediary product in the decomposition of leucin. Unfortunately, as inspection of Table IV shows, amyl alcohol is so poisonous that the question can not be definitely settled in this way. The percentage tolerated is so small that the quantity can not be determined with sufficient accuracy. It may, however, be stated that there is no evidence that amyl alcohol is oxidized by *P. puberulum,* since after several weeks' growth it had not disappeared from cultures containing as little as 0.05 per cent. In these cultures it was separated by the method of Beckmann, the characteristic odor being recognized in the final extract. These observations probably have little bearing on the question of the intermediary formation of amyl alcohol from leucin. As far as they indicate anything they are not in favor of it.

In both leucin and tyrosin cultures an appreciable amount of volatile acid was found, possibly formic acid, since the silver salt was rapidly reduced. Because of this property the acid from the leucin culture was not studied. That from the tyrosin culture formed a crystalline barium salt. The quantity was too small for analysis.

The tyrosin culture presented a number of peculiarities. While the musty odor of the growth on ordinary Raulin's medium was perceptible only when the mycelium was held close to the nose, that on tyrosin had a typical and extremely musty odor which permeated the room when the flask was opened. The odor of ordinary cultures became more perceptible when they were distilled, since the odoriferous

[1] Ehrlich, Felix. Über die chemischen Vorgänge des pflanzlichen Eiweissstoffwechsels und ihre Bedeutung für die alkoholische Gärung und andere pflanzenphysiologische Prozesse. Landwirthschaftliche Jahrbücher, Bd. 38, Ergänzungsbd. 5, p. 306–307, 1909.

[2] Raciborski, M. Op. cit., ann. 1906, p. 765. 1907.

principle seemed to accumulate in the distillate. Thom [1] observed that the production of odors varied greatly not merely from species to species, but also in some individual species according to the culture medium. It would be interesting to investigate whether in the present case the production of the odorous principle was actually dependent upon the presence of an aromatic compound or upon some other condition. Such factors as these may be involved in the production of the flavor of cheese.

The rate of growth was very much more rapid at first than in the controls, in spite of the fact that little more than half the tyrosin was consumed. Moreover, spore formation began before the fifth day and was very abundant. In less than a week the entire surface of the liquid was covered with mycelium uniformly green with spores. It was more delicate and less stiff and woody than that of the controls. It was smooth and even, and lay flat on the surface, whereas in the controls it was convoluted and twisted so that some of it was pushed below the surface, resulting in the formation of new mycelium above. In the tyrosin the growth, after the surface had once been covered, seemed less bulky, although the old mycelium was gradually overgrown with new mycelium.

Certain of the facts here recorded have some theoretical interest. This is particularly true of the peculiarities of growth of the tyrosin culture and of the absence of easily detectable oxidizing enzyms in all the cultures tested.

The peculiarities of the tyrosin culture which are of interest in this connection are the thinness of the mycelium and precocity of the spore formation. Tyrosin contains the aromatic ring. Perhaps a large amount of aromatic derivatives is required so that spore formation can not take place until the organism has had time to manufacture these aromatic compounds from the sugar and other straight-chain carbon compounds offered. When an assimilable aromatic compound is offered, this latent period is perhaps bridged over. Certainly, on the tyrosin the spore formation is at least as rapid as on corn-meal mush, which, in its proteins, contains an abundance of aromatic compounds. The importance of various amino acids for microbic growth has recently been brought out in studies on the cultivation of the leprosy bacillus.[2] Another explanation may, however, be based on the assumption that spore formation is rapid in an exhausted or unfavorable medium. That this is actually true is still an open question, though Tiraboschi [3] presents

[1] Thom, Charles. Cultural studies of species of Penicillium. U. S. Department of Agriculture, Bureau of Animal Industry, Bulletin 118, p. 90, 1910.

[2] Duval, C. W. Cultivation of the leprosy bacillus from the human tissues, with special reference to the amino acids as culture media. Journal of Experimental Medicine, v. 13, p. 365–373, 1911.

[3] Tiraboschi, C. Studi sugli ifomiceti parassiti del granturco guasto. Atti del Terzo Congresso Pellagrologico Italiano, Milano, 24-25-26 Septembre, 1906, p. 138. 1907.

evidence for this view. If this hypothesis be accepted it would be necessary to assume that tyrosin is an unfavorable source of nitrogen, either because of its chemical properties or because of its insolubility. That its chemical properties are the cause seems hardly likely, since leucin, which also contains amino nitrogen, gives such an abundant growth, although Raciborski [1] states that tyrosin is a much poorer source of nitrogen for *Aspergillus niger* than ammonia. That its insolubility is the cause is quite possible. Tyrosin is so little soluble that the amount in solution at any moment would be very much less than in the unmodified Raulin's medium, for most of the tyrosin contained in the culture had crystallized out on the bottom of the flask, at least 5 centimeters below the mycelium. Hence, it is possible that the rapid sporification was a sign of nitrogen starvation, which is the more likely, as the culture was very little handled or agitated. Under these conditions diffusion is very slow indeed, particularly for substances of great molecular weight like tyrosin. That diffusion does not keep pace with consumption of material is evident if any flask culture which has remained undisturbed for some days be examined. If such a culture be held between the eye and the light and gently rotated, convection currents can be seen near the mycelium, showing that the specific gravity of the liquid in immediate contact with the mycelium is very much less than that of the deeper layer. It may therefore be that after the first few days the mycelium was in relative nitrogen starvation. This hypothesis would account for the rapid initial and slow subsequent growth.

A perusal of the literature on the metabolism of the fungi shows that this question of the rate of diffusion has not received adequate attention. If the organism is grown on a thick layer of culture fluid the rate of diffusion of the dissolved substances is undoubtedly an important factor. A substance of small molecular weight will diffuse upward more rapidly to replace that consumed than one of great molecular weight. Thus the substance of small molecular weight might appear to be a better food relatively than it really is. The same logic applies to metabolic products. When these products are substances like alcohol, of light specific gravity, they will accumulate at the surface, particularly if the viscosity of the culture fluid is great, owing to the presence of much sugar or peptone. If, then, the alcohol concentration be determined for the whole fluid the values obtained may be spurious. Actually the mycelium may have been in contact with a much higher concentration of alcohol. This may be one of the reasons why the crop of mycelium for a given amount of sugar is said to be greater when the organism is grown on a thin

[1] Raciborski, M. Über die Assimilation der Stickstoffverbindungen durch Pilze. Bulletin International de l'Académie des Sciences de Cracovie. Classe des Sciences Mathématiques et Naturelles, ann. 1906, p. 764. 1907.

layer with great surface than when grown on a thick layer of culture fluid with smaller surface.[1] The largest yields were obtained in this way.

The absence of easily detectable oxidase is significant in connection with the discovery of Euler and Bolin that the oxidase of alfalfa (*Medicago sativa*) is a mixture of the calcium salts of simple oxy-acids.[2] It is believed by some that many molds do not require calcium.[3] It is tempting to imagine a connection between the absence of easily detectable oxidase, the observation of Euler and Bolin, and the absence of calcium.

GENERAL CONSIDERATIONS.

Since it has been definitely shown in the present paper that a distinct species of Penicillium produces a substance of moderate toxicity, the question very naturally arises, has it any pathological significance? At present it can only be said that it is too early to answer this question. All that can be done is to discuss the possibility and to indicate further work to be done.

In acute intoxications, alleged to be due to molds, penicillic acid alone can hardly be of significance. The lethal subcutaneous dose for mice, as has been shown, is about 0.3 gram per kilogram of body weight. Assuming the same susceptibility for the average man of about 70 kilos, the dangerous dose would probably be about 21 grams. Hence, an acute intoxication from penicillic acid would require that an inconceivably great quantity of moldy food be consumed in a single day. Even herbivorous animals could hardly eat enough moldy fodder in a day to be acutely affected by penicillic acid. It is quite out of the question that in the natural course of events penicillic acid is likely to produce acute intoxication.

However, the results of this investigation of *Penicillium puberulum* indicate the possibility of acute intoxication by moldy food. As already stated, the different species of Penicillium differ radically in their biochemical behavior. If there is so much difference in the ordinary products of metabolism, it is altogether likely that a series of toxic substances may be produced by different species. Some of these substances might very well be far more toxic than penicillic acid. Indeed, Italian investigators have shown this contingency to be very probable. Gosio, Di Pietro, and Sturli have obtained from pure cultures of Penicillium toxic extracts far more poisonous than

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[1] Nikitinsky, J. Ueber die Beeinflussung der Entwickelung einiger Schimmelpilze durch ihre Stoffwechselprodukte. Jahrbücher für Wissenschaftliche Botanik, Bd. 40, p. 43, 1904.

Raciborski, M. Op. cit., p. 733–734.

[2] Euler, H., and Bolin, I. Über die chemische Zusammensetzung und die biologische Rolle einer Oxydase. Zeitschrift für Physikalische Chemie, Bd. 69, p. 187–202, 1909.

[3] Loew, Oscar. Über die Giftwirkung von oxalsauren Salzen und die physiologische Funktion des Calciums. Biochemische Zeitschrift, Bd. 38, p. 226–243, 1912.

Robert, Mlle. Influence du calcium sur le développement et la composition minérale de l'Aspergillus niger. Comptes Rendus de l'Académie des Sciences [Paris], t. 153, p. 1175–1177, 1911.

anything hitherto obtained in the new work herein recorded. Because none of these investigators have isolated the toxic principle in a state of purity their researches have not been given the serious consideration that is their due. It will be the task of this laboratory to extend these investigations to other species of Penicillium in the hope that other toxic substances, perhaps more active than penicillic acid, may be isolated.

In the matter of chronic intoxication the situation is quite different. Continued use of moldy food containing penicillic acid might produce symptoms. The quantity of badly spoiled corn-meal mush which a man would consume at a single meal might contain as much as 0.1 to 0.5 gram of penicillic acid. As this acid has a toxicity of the same order of magnitude as phenol, resorcin, or salicylic acid, and as such substances are believed by many to be undesirable as food preservatives, it seems reasonable to demand that great care be exercised in eliminating moldy corn from the diet. Owing to the difficulty of procuring material, it has not been possible to conduct long-continued feeding experiments. Therefore it is impossible to say whether penicillic acid has cumulative action. Should it prove to have such action chronic intoxication might be brought about by comparatively small doses. For this reason it is very desirable to learn the constitution of penicillic acid in order to be able to make it synthetically. This is the most promising way to obtain larger quantities of it.

While the finding of penicillic acid indicates that the relation of moldy corn to pellagra[1] deserves renewed attention, this discovery does not materially strengthen the maize theory of the etiology of pellagra. Penicillic acid itself is not sufficiently toxic. It is quite possible that penicillic acid or a closely related substance may have been responsible for the toxic effects following the administration of "pellagrozein," the poison obtained from spoiled maize, with which, according to the experiments of Lombroso,[2] the disease could be produced artificially. "Pellagrozein" itself Lombroso did not regard as anything but a mixture. He believed it contained two alkaloids, which accounted for the toxic action. Neither alkaloid has ever been obtained in a state of purity, so that it is impossible to form any definite opinion about them. Indeed, other investigators have not been able to find alkaloids at all in spoiled maize.[3] It is quite

[1] Cf. Marie, A., Pellagra, authorized translation from the French, by C. H. Lavinder and J. W. Babcock, Columbia, S. C., 1910.

[2] Lombroso, Cesare, and Erba, Carlo. Sulle sostanze stricniche e narcotiche del mais guasto. Reale Istituto Lombardo, Rendiconti, s. 2, v. 9, p. 133–147, 1876.

[3] Monselise, G. Ricerche chimico-tossicologiche intorno ad alcuni campioni di mais per la studio della pellagra, Mondovi, 1881, 58 p. (Cited by Gosio.)

Selmi, Antonio. Delle alterazioni alle quali soggiace il granturco (Zea mais) e specialmente di quello che ingenera la pellagra. Atti della R. Accademia dei Lincei, s. 3, Memorie della Classe di Scienze Fisiche, Matematiche e Naturali, v. 1, dispensa 2, p. 1099–1141, 1877.

Di Pietro, Melchiorre. Sui veleni di alcune muffe. Annali d'Igiene Sperimentale, v. 12 (n. s.), p. 314–365, 1902.

possible that these alkaloids were either normal constituents of maize or ptomainelike bases, as was pointed out by Pelloggio,[1] for the investigators who found alkaloids seem to have often allowed the maize to spoil to an extreme degree. In one case the maize was actually allowed to rot until it stank.[2] If the maize used contained either penicillic acid or some similar substance the method of preparation was such that these substances would have passed into the "pellagrozein," which is even less toxic than penicillic acid. The lethal subcutaneous dose varied from 1.5 grams per kilogram for frogs to 7 to 10 grams for cats.[3] Hence, it is not impossible that "pellagrozein" contains substances of this type. Whatever evidence there is for the relation between "pellagrozein" and pellagra would apply equally well to penicillic acid. The discussion of this question lies beyond the scope of the present paper.

PENICILLIUM STOLONIFERUM.

In a former publication [4] it was stated that most of the samples of spoiled American maize examined in this laboratory failed to give Gosio's [5] phenol test with ferric chlorid. In this respect American spoiled maize seems to differ from that found in Italy, where the ferric-chlorid reaction is regarded as a reliable test for the deterioration of maize.[6]

Since the publication of the above-cited studies of the deterioration of maize the test has been improved in this laboratory so that in its new modification it is more delicate. The procedure as now conducted consists in extracting 50 grams of ground corn or meal in a stoppered flask, with sufficient chloroform to cover the mass. After two hours the chloroform extract is filtered off and concentrated to a bulk of 10 to 15 cubic centimeters. This concentrate is transferred to a small separatory funnel or test tube and covered with 5 cubic centimeters of water containing a trace of ferric chlorid. If substances like penicillic acid are present, the characteristic color develops in the aqueous layer.

[1] Pelloggio, Pietro. Materia reagente quale alcaloide, trovata nell' estratto del mais guasto preparato dall' Erba. Reale Istituto Lombardo, Rendiconti, s. 2, v. 9, p. 118–121, 1876.

[2] Lombroso, Cesare, and Erba, Carlo. Loc. cit.

Biffi, S. Sulla nota del prof. Cesare Lombroso: I veleni del mais e la pellagra. Reale Istituto Lombardo di Scienze e Lettere, Rendiconti, s. 2, v. 9, p. 282–288, 1876.

[3] Lombroso Cesare. I veleni del mais e la loro applicazione all' igiene ed alla terapia. Rivista Clinica di Bologna, s. 2, ann. 7, p. 109–112, 1877.

—— and Erba, Carlo. Op. cit.

[4] Black, O. F., and Alsberg, C. L. The determination of the deterioration of maize, with incidental reference to pellagra. U. S. Department of Agriculture, Bureau of Plant Industry, Bulletin 199, 1910.

[5] Gosio, B. Ricerche batteriologiche e chimiche sulle alterazioni del mais. Rivista d' Igiene e Sanità Pubblica, ann. 7, p. 825–849, 869–888, 1896.

[6] Gosio, B. Alterazioni del granturco e loro profilassi. Italy, Direzione generale dell' Agricoltura, Annali di Agricoltura, no. 261, 1909.

When the tests are conducted in this way the number of samples of obviously deteriorated maize showing the reaction is greater than when the unmodified Gosio test is employed. Nevertheless, a positive result seems to be less frequent in American maize than in Italian maize. Moreover, the colors obtained with American spoiled maize have always been found to be red or red brown, while in Italy tests of spoiled corn are most commonly described as showing violet, blue, purple, and greenish tints. None of these tints have been encountered in American maize in this laboratory.

Since this sharp difference apparently exists between American and Italian deteriorated maize, it is desirable to compare samples of Italian spoiled maize with American ones. Opportunity to make this comparison was offered by Dr. C. H. Lavinder, of the Public Health Service, who while on a visit to Italy very kindly secured samples of condemned maize.

From one of these samples of maize Dr. E. F. Smith, of this Bureau, isolated two species of Penicillium. One of these species was identified by Dr. Charles Thom, of the Storrs Agricultural Experiment Station, as *P. stoloniferum* Thom.[1]

This organism when grown on Raulin's medium gives the very strong and characteristically violet ferric-chlorid reaction of Gosio. It is certainly a remarkable fact that the first sample of spoiled Italian corn examined gave the violet color described by Italian authors, whereas no American sample has been found giving a similar tint.

It was therefore decided to isolate, if possible, the substance responsible for the ferric-chlorid reaction. For this purpose the organism from Italian spoiled corn was grown in "Long Blake" bottles on Czapek's medium and on Raulin's medium in the manner above described. It was found that the organism grew more rapidly upon Raulin's medium. Therefore, for the preparation of material Raulin's medium only was used.

The substance responsible for the ferric-chlorid reaction was isolated by the following procedure: The culture fluid and the mycelium were transferred to an evaporating dish and rendered weakly alkaline with sodium carbonate. The contents of the dish were then heated to boiling and filtered hot. The mycelium remaining on the filter was thoroughly expressed. The mass was then again extracted with water rendered weakly alkaline with sodium carbonate. The combined extracts were evaporated to a small bulk over a free flame and filtered hot. To the clear filtrate a slight excess of hydrochloric acid was added. An abundant precipitate

[1] Thom, Charles. Cultural studies of species of Penicillium. U. S. Department of Agriculture, Bureau of Animal Industry, Bulletin 118, 1910.

was produced, which consisted of a mixture of needle clusters and amorphous material. The precipitate was separated by filtration and washed with cold water. After drying spontaneously it was extracted with hot toluene and the hot extract filtered. Only the crystalline portion of the precipitate dissolved. The amorphous dark-brown material which remained on the filter was discarded, for it did not give a color reaction with ferric chlorid. On cooling and evaporating, the toluene extract spontaneously precipitated in the form of needles, the material giving the ferric-chlorid reaction. These needles, which were still slightly colored, were finally obtained white either by decolorizing with boneblack in hot toluene solution or by dissolving in alcohol and adding alcoholic potassium hydroxid to form the potassium salt, which is insoluble in alcohol. This salt was then washed free from color with alcohol. From the potassium salt the free acid was recovered in the form of white needles by dissolving the salt in water and precipitating with hydrochloric acid.

The substance thus obtained consists of white needles with a melting point of 140° C., uncorrected. The name mycophenolic acid is provisionally suggested for it. It is almost insoluble in water, but freely soluble in alcohol, in ether, and in chloroform. It is somewhat less soluble in benzene, only moderately soluble in cold toluene, and very soluble in hot toluene. With ferric chlorid it gives a violet color in aqueous solution, though its solubility in water is not sufficient to render the color intense. In alcoholic solution it gives a bright-green color with ferric chlorid. It does not react with Millon's reagent. It does not give Lieberman's reaction and could not be diazotized. It does not reduce Fehling's solution nor ammoniacal silver nitrate. It is fairly resistant to sodium, ammonium, and potassium hydrates and to hydrochloric, sulphuric, and acetic acids, being unaffected by boiling in 10 per cent solutions of any of these reagents. It does not contain water of crystallization. Its salts of potassium and sodium are very soluble in water. The salt of potassium is soluble in dilute alcohol, but insoluble in absolute alcohol. The salt of sodium is soluble in absolute alcohol, but may be precipitated in crystalline form by adding ether. The salt of barium is only very slightly soluble in water and forms clusters of minute needles. The copper, lead, and silver salts are amorphous and insoluble in water. In characterization of the substance the facts collected in Table V were ascertained by analysis of the free acid, by titration of the alcoholic solution of the free acid with n/10 sodium hydroxid, and by the determination of the barium content of the salt on ignition in platinum with sulphuric acid.

TABLE V.—*Analyses of mycophenolic acid.*

Weight of substance.	CO_2.	H_2O.	C.	H.	$BaSO_4$	Ba.	N/10NaOH.
Gram.	*Gram.*	*Gram.*	*Per cent.*	*Per cent.*	*Gram.*	*Per cent.*	*C. c.*
0.2316	0.5419	0.1315	63.81	6.30			
.2044	.4770	.1161	63.64	6.31			
.2494					0.1256	29.65	
.1990							11.53
Average..			63.725	6.305			

Calculated for $C_{17}H_{20}O_6$: carbon, 63.74 per cent; hydrogen, 6.25 per cent.
Found..................... carbon, 63.72 per cent; hydrogen, 6.30 per cent.

Calculated for Ba $(C_{17}H_{18}O_6)$: barium, 29.15 per cent.
Found..................... barium, 30.09 per cent.

A molecular weight determination by the elevation of the boiling point in chloroform solution gave the results shown in Table VI.

TABLE VI.—*Ebullioscopic determination of the molecular weight of mycophenolic acid.*

Weight of substance.	Weight of solvent.	Rise of boiling point.	Molecular weight.
Gram.	*Grams.*	*Degree C.*	
0.1641	30.32	0.065	308
.1578	30.32	.060	321
Average..			314.5

Molecular weight calculated for $C_{17}H_{20}O_6$........................ 320
Molecular weight found from titration........................ 345.4
Molecular weight found from barium content of salt.............. 328
Molecular weight found from boiling-point elevation.............. 314.5

The formula $C_{17}H_{20}O_6$ may therefore be assigned to mycophenolic acid. It does not readily decompose carbonates at ordinary temperatures. It is apparently a dibasic acid, or, at any rate, combines with two atoms of a monovalent base. Whether the base combines entirely with carboxyl groups or with phenol groups has not been determined.

The acid seems to form two series of salts. Presumptive evidence on this point was obtained by the following experiments: Two decigrams of free acid were suspended in water and one equivalent of potassium hydroxid added. Unfortunately, this quantity was not sufficient to dissolve the substances completely, so that a slightly greater quantity of the alkali had to be used. This solution was then treated with one equivalent of barium chlorid. On standing in the desiccator a crystalline barium salt formed. This salt was evidently different from the normal barium salt, which is so insoluble that it precipitates at once. It was also of different appearance under the microscope, consisting of a few small needles in clusters, which apparently were the normal salt, and more abundant larger single needles, apparently the acid salt. The presence of the normal salt in small quantities under the conditions of the experiment was probably due to the fact that an excess of alkali had to be used in dissolving the substances. The barium content of this preparation was determined, 0.207 gram yielding 0.0692 gram of $BaSO_4$, equivalent to a barium content of 20.2 per cent.

Calculated for Ba $(C_{17}H_{18}O_6)$	28. 1 per cent.
Calculated for Ba $(C_{17}H_{19}O_6)_2$	17. 7 per cent.
Found	20. 2 per cent.

Apparently, as shown by the microscope, the preparation consisted of a mixture of two salts.

It has not been found possible to identify mycophenolic acid with any known compound. The substance of Gosio referred to above very greatly resembles it, though these substances are probably not identical. However, Gosio's characterization of this substance was based on a very small quantity of material, so that his formula $C_9H_{10}O_3$, based on a single combustion, can not be regarded as final. The chief points of difference between the substance described by Gosio and mycophenolic acid are the percentage composition and the behavior with ferric chlorid. Gosio's substance gives an intense blue color with ferric chlorid in alcoholic solution. Mycophenolic acid gives a violet color in aqueous solution, while in alcoholic solution with a trace of ferric chlorid it gives a violet color which becomes bright green on addition of an excess of the reagent.

In one particular mycophenolic acid resembles Gosio's substance but differs from penicillic acid. It is not toxic. Ten milligrams were dissolved in water with the aid of a little sodium carbonate and injected subcutaneously into a mouse. No untoward effects whatever were noted. It differs furthermore from penicillic acid in being present chiefly in the mycelium in the early stages of growth. In old cultures it is found both in the culture fluid and in the mycelium, perhaps because with the gradual production of basic substances it is dissolved. The question whether toxic phenolic substances are found in the culture fluid or only in the mycelium is one that has been much discussed by students of pellagra. When the substances are insoluble acids with soluble salts like mycophenolic acid, their distribution is probably only a question of the reaction of the medium. When the reaction is acid they will be found in the mycelium, as lichen acids incrust the lichen thallus. When the medium contains available bases they will become more or less dissolved in the medium.

With the advancing age of the culture, mycophenolic acid gradually increases in quantity until under the conditions employed in these experiments at the end of two weeks the maximum yield is obtained.

After that time the quantity present is apparently constant. When grown in the "Long Blake" bottles charged with 250 cubic centimeters of culture fluid, the yield at the end of about two weeks averages about 0.07 gram of the crude acid per bottle.

Since *Penicillium stoloniferum* is found so commonly in the United States and has been isolated in this laboratory from spoiled maize, it is not easy to understand why it so rarely, if ever, causes spoiled maize in the United States to give the ferric-chlorid reaction. The first explanation to present itself was that the American organism might be a different strain or perhaps a "physiological variety."

To solve this question, Dr. Thom very kindly furnished a specimen of his type culture. This specimen was grown side by side with the Italian organism. It grew rather more slowly than the latter and there were slight differences in appearance. The cultures gave a good ferric-chlorid reaction, very similar in shade to that given by the Italian organism, but when the attempt was made to separate mycophenolic acid from the cultures of the American organism none could be found. In its place was found a quite different substance or mixture of substances. As this material has not yet been obtained in satisfactory crystalline form, not much can at present be said of its properties.

The different biochemical behavior of the two strains from the two continents is certainly suggestive. Whether these two strains are really physiologically different can not as yet be decided. The American organism used is an old one, having been propagated by Dr. Thom in the laboratory for a number of years. Possibly this long artificial propagation has altered its behavior. It is proposed to continue the investigation of this problem by comparing the two cultures on hand with a number of other recently isolated strains.

No extended physiological studies were undertaken on *Penicillium stoloniferum*. A few observations were made incidentally. The organism always produced alcohol, as shown by applying the iodoform test to the distillate. No quantitative determinations were made, but the amount of alcohol formed, as judged by the iodoform test, seemed to be decidedly less than that produced by *P. puberulum*. *P. stoloniferum* produces a small amount of oxalic acid, as shown by the method used for *P. puberulum*. It seems to be present in somewhat larger amounts and at an earlier stage of growth than in cultures of *P. puberulum*. Finally the mycelium of *P. stoloniferum* seems to be very rich in mannitol.

SUMMARY.

Of six species of Penicillium from maize examined, only two elaborated substances toxic to mice. Two of these species, one toxic, the other nontoxic, were studied in detail.

The first, identified as *Penicillium puberulum* Bainier, elaborates a toxic product which was isolated and for which the name "penicillic acid" and the formula $C_8H_{10}O_4$ are suggested. This substance behaves like a monobasic acid. It is toxic to animals when injected subcutaneously, causing death in a dosage of about 0.2 to 0.3 gram per kilo of body weight. The formation of penicillic acid is more abundant when the air supply is limited and the reaction of the medium is acid. The form in which nitrogen was offered the fungus seems also to have some influence on its formation.

Penicillium puberulum Bainier was always found to produce alcohol when grown in the presence of sugar. Old cultures contain minute amounts of oxalic acid. In the presence of sugar and leucin no amyl alcohol is produced, although leucin is consumed. A small quantity of volatile acid is, however, formed. In the presence of sugar and tyrosin neither tyrol nor tyrosol is produced, though tyrosin is consumed. A small quantity of volatile acid is formed.

The second organism, *Penicillium stoloniferum* Thom, was nontoxic. Unlike the other five studied, it was isolated from Italian maize. It elaborates a new phenolic acid, for which the name mycophenolic acid and the formula $C_{17}H_{20}O_6$ are suggested. This substance behaves like a weak dibasic acid and, like penicillic acid, resembles the lichen acids in many ways. Among the other metabolic products of the organism, alcohol, oxalic acid, and mannitol were found.

In the present paper it has been shown that species of Penicillium so closely related that until recently they were not distinguished by morphologists differ quite markedly in their metabolism. It is greatly to be desired that the whole genus be studied biochemically. The chemical findings will no doubt supplement the morphological in many important ways. Indeed, as indicated by the constant presence of alcohol and the formation of penicillic acid by *Penicillium puberulum* and the formation of mycophenolic acid by *P. stoloniferum*, it is not impossible that characteristic chemical properties may help to distinguish between species or strains not now sharply separated by morphologists.

Therefore the present investigation furnishes additional data for explaining the discrepancies in different biochemical investigations on molds. Previous investigators may have failed to realize that the products elaborated vary with the species, with the reaction of the medium, with the aeration, and perhaps with the nature of the nitrogenous food supply, and that it is exceedingly difficult to distinguish between quite distinct species. They may also have underestimated the difficulties of distinguishing between the individual species.

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