REPORT OF THE TWENTY-EIGHTH ALFALFA IMPROVEMENT CONFERENCE

July 13-16, 1982
University of California
Davis, California

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TWENTY-EIGHTH ALFALFA IMPROVEMENT CONFERENCE

Program Chairman - B. Melton

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Introduction

The 28th National Alfalfa Improvement Conference (NAIC) began with a pre-conference tour that originated in Bakersfield, California. Two days were spent traveling through the California Central Valley. Stops were made to observe alfalfa forage and seed production and both industry and public research programs. Other types of agriculture unique to the area were also observed. The formal part of the program was held on the University of California-Davis campus. Dr. Charles E. Hess, Dean, College of Agricultural and Environmental Sciences welcomed the participants. The formal conference included two and one half days of paper presentations and poster sessions.

This report includes the opening address by Dean, Hess, two special addresses, "History of the improved alfalfas" by Ward C. Waterman and "The story of certified alfalfa seed production in California" by Frank Parsons, abstracts of research studies reported at the conference; volunteer abstracts and brief articles submitted by scientists not attending the conference, committee reports, business transacted, and information on distribution of conference reports and membership.

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Members of the executive committee of the 28th NAIC included: B. Melton, Chairman, Las Cruces, NM; B.P. Goplen, Saskatoon, Sask.; W.J. Knipe, Woodland, CA, I.I. Kawaguchi, Bakersfield, CA; B. Hartman, Kerman, CA: J.E. Winch, Guelph, Ont.; E.L. Sorensen, Manhattan, KS, L.R. Teuber, local arrangements, Davis, CA, and D.K. Barnes, permanent secretary, St. Paul, MN. Copies of this report are available from D.K. Barnes, USDA-ARS, Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108.

Welcome to the National Alfalfa Improvement Conference

Charles E. Hess
Dean, College of Agricultural and Environmental Sciences
University of California, Davis, CA

It is a pleasure to welcome the National Alfalfa Improvement Conference back to Davis after twenty years. Since your last meeting in Davis, in 1962, many things have changed on the Campus as well as the issues confronting us in the state and nation. The enrollment in the College of Agricultural and Environmental Sciences has grown to approximately 4,700 undergraduate students and about 1,100 graduate students. This was about the size of the total Campus when the conference was held here in 1962. Now the total Campus has an enrollment of about 19,000 undergraduate and graduate students.

Among the major concerns confronting us today are things such as productivity, water, energy, integrated pest management, and genetic engineering. In the area of productivity, which is an important today as it was back in 1962, we are still attempting to get as much output per unit of input that is possible. There have been many important gains, such as short stature rice developed for the Sacramento Valley, a new variety which as replaced all the varieties in a period of about three years. An intensive program in strawberry breeding and selection has led to the average yield of about 19 tons per acre compared with three to six tons per acre in other areas. An exciting development in terms of increasing productivity is the development of genetically dwarf fruit trees, such as the peach, which enables us to get much higher plant populations per acre and into productivity in a much quicker time.

A second major area of concern is water. We are looking for ways of managing water better, of recycling drainage water, and looking to the development of plants which have greater tolerance to high salt levels. For example, Professor Emanuel Epstein has selected a barley which can actually be irrigated with sea water during its growth and development and it will actually produce a reasonable yield. We certainly are not advocating the use of sea water as an irrigation source for the Central Valley, but it does indicate the potential that does exist in plants for salt tolerance. Professor Bill Rains, Chair of the Department of Agronomy and Range Science, has been using tissue culture techniques to develop salt tolerant alfalfa. The work in this area ranges from breeding to molecular biology and one of our faculty has identified a gene in bacteria which provides salt tolerance for that organism and has been able to transfer the gene from one bacteria to another. In fact, he has been able to transfer the gene into rhizobia and produce rhizobia strains which can fix nitrogen under high salt levels.

Another are of major concern is energy. Presently, we have gone through a period of some deemphasis with the approach from the federal level being one of more exploration and the oversupply of oil has reduced the price somewhat in the past year. Still, energy uses in farming constitute a major cost of operations. Again, we are looking for conservation of energy in food and fiber production, harvesting, and processing as well as looking at agriculture as a potential source of energy from biomass.

Integrated pest management has been and continues to be a major research emphasis in California to try to develop plants which are resistant to insects and disease, to develop biological control strategies as an alternative to chemical control, and to make the chemicals that we do use more effective and as safe as possible. Mathematical modeling is a component of the more efficient use of pesticides and, to facilitate the timing of crop planning and the timing of the use of either biological or chemical control methods.

We have a very active program in genetic engineering and have been the focus of considerable interest as we attempt to develop policy guidelines for faculty who choose to be involved in private research ventures as well as conducting their University research activities.

So, as you see, it is a very interesting and challenging time to be involved in agricultural and environmental research, teaching, and extension. Although a number of changes have taken place in the twenty years since you have been here, Davis still remains a friendly, bike-oriented, energy-conscientious college town. I hope that you enjoy your stay at UC Davis and that you return home with good memories and lots of new ideas and information that can be applied in your own research, teaching, and extension programs.

History of the Improved Alfalfas

Ward C. Waterman
W-L Research, Inc.
Bakersfield, CA

My personal history of the improved alfalfas in the market place begins with the year of 1949. In the late 1940's, there were a number of new improved alfalfa varieties all waiting for the one answer -- where could they be produced in volume for the market place. There was Ranger, Buffalo, Atlantic and Narragansett, all ready to go with Vernal just two years away.

Going into the 1950's, all commercial alfalfa seed produced in the United States, the northern hardys, the intermediates, and the non-hardys were harvested from the hay fields left for seed. Except for a few such as Grimm and Ladak, they were all designated for their degree of hardiness by their state of origin -- Montana common, Nebraska common, Kansas common, California common, etc. There was also some alfalfa seed imported from Argentina, Chile, and Canada.

Beginning with the year of 1950, following two or three years of trial production with three or four small growers over the state, a substantial acreage was planted with these new improved alfalfas. They were grown out of their areas of adaptation, in the irrigated farming sections of the San Joaquin Valley in California. They were planted in rows 40 inches apart to cultivate out all second generation volunteer growth following each harvest. It proved successful with a larger volume of alfalfa seed for market than anyone had anticipated.

New grower oriented seed companies were organized to process and to market this new volume of alfalfa seed. By 1953, about 90% of this production in California was being processed and marketed by four companies: the Arnold Thomas Seed Service, Calapproved (a Farm Bureau growers' cooperative), J.C. Loomis Company, and Maricopa Seed Farms. The managements of these four new companies were convinced that these new varieties were sufficiently superior to completely replace the common alfalfas in short order and that there should be a favorable market for about all the seed we would produce.

How little did we know about, or understand, the market place. For the next ten years the new improved alfalfas replaced very little of the common seed harvested from the Great Plains hay fields. Every pound of alfalfa seed harvested from those hay fields moved into the market place. There was never a time that the mid-western wholesale seed trade refused to buy this common seed, if the price was below that of the certified new varieties. By 1953, it became obvious that the wholesale seed trade east of the Rocky Mountains was truly overwhelmed with the new, high volume production. These wholesalers had no program to promote the sales of these new alfalfas. All they knew to do was try to be competitive with each other with a favorable price to the retail store. Their ideas of values for these new varieties was below our costs of production.

The Certified Alfalfa Seed Council was organized in the fall of 1952 by the four new companies with the desperate feeling that the answer was to conduct an all-out sales and promotion program throughout the major consuming states (the northeast one-fourth of the United States). There was a need to tell our story of production to the Extension Service, to the Ag-economists, and to the farm publications.

In 1955, about the third year of the Council's activities in the midwest, an invitation was extended to all of the farm publication editors, far and wide, to meet with us in St. Paul, Minnesota. Following this meeting the prestigious publication, Successful Farming, Des Moines, Iowa, lauched an all-out farmer survey in all of Iowa and a good portion of those states adjoining Iowa. The survey asked the farmer to indicate, in their order of importance, the sources of information that influenced him the most in his decision-making process. The list presented for his answers included the farm publications, Extension Service Bulletins, County agent meetings and conferences. Number one, head and shoulders over all other sources for information listed, was his neighbor. The farm publications had a slight edge over the Extension Service and the County Agents. These all averaged about 15% in Iowa. In Nebraska, the Extension Service scored eight percent. One Nebraska legislator was so incensed that he proposed that the state eliminate the Extension Service as a gross waste of money. It came to a vote in their unicameral legislature. The Extension Service survived by only a few votes.

A short time after this, a team of Rural Sociologists at Iowa State University published the conclusions of their study of the Iowa farmers decision-making process. The title of their publication is "The Diffusion Process." It confirmed and elaborated upon the findings of the Successful Farming survey. It also exonerated both the Extension Service and the farm publications for their low scores.

The bottom line from this study determined that only one farmer in twenty-five could read a bulletin, a news article, or attend County Agent meetings, and trust his own judgement. The Rural Sociologists named this one farmer in twenty-five the Prime Innovator. The other twenty-four looked to him for their decision-making. If it worked for him, it would work for them. This is the so-called neighbor diffusion process.

By the fall of 1956, we new-comers in the seed industry from the West, having expected the improved alfalfas to take over in short order, knew we were now facing the reality of a long, slow transition period in the consuming areas. We were facing the probability of major economic problems for this seed industry, both in the production areas and in the marketing areas. This was becoming a basic reality in the market place that seemed to mandate the proprietary approach as essential for survival.

By 1958, it became completely obvious to these four seed companies that the proprietary approach was essential -- to follow the pattern of the hybrid corn industry. The four companies had to develop their own private varieties in order to establish an orderly program from the seed grower through to the effective marketing program that can be done only through the franchised proprietary programs.

The four original seed companies who organized the Certified Alfalfa Seed Council new became three. The J.C. Loomis Company and Maricopa Seed Farms became the Waterman-Loomis Company (now W-L Research, Inc.) with Dr. David F. Beard from USDA as their plant breeder. The Arnold Thomas Seed Service employed Dr. Howard Carnahan from USDA, and Calapproved merged with Caladino to become Cal/West. They employed Dr. Iver Johnson from Iowa State as their breeder.

About this time, Northrup King and Company launched their program to blend (scientifically) the public varieties (varieties not stated on the bag) which they called their 919 Brand. This was a successful program to help move the huge production into the market place on a semi-proprietary approach. They were followed by a number of others in the wholesale industry. It was a major contribution to help postpone the eventual collapse of the public variety commodity markets. Beginning of the end of a number of huge wholesalers, i.e. Rudy Patrick, Peppard, Albert Dickinson, Teweles, Mangelsdorf. The year 1957 was the peak year for California production -- 63,000,000 pounds of eleven public varieties, of Blue Tag certified alfalfa seed was produced.

The proprietary alfalfa seed industry now consists of probably a dozen companies, all with their own breeding programs. The proprietary industry is now well endowed with a competent staff of plant breeders. It took the hybrid corn industry about thirty years to convert their market to all commercial hybrids. The alfalfas may see another twenty-five years before the commercial proprietary varieties can claim a high percentage of the market.

There is not short cut from seed grower to the consumer to date. There are four primary components, all equally essential, to have an orderly program. You have the grower, the seed company with the plant breeder, the wholesale-retailer in the consuming area, and the consumer. Unless you have a favorable economic position for each of these four, the program will fail.

The large volume of the public varieties marketed as a commodity, became increasingly unfavorable for three of these four primary components. Only the consumer had a favorable break in the low price. And even the low price did little to accelerate the Diffusion Process.

It is generally conceded that the consumption of the new improved public varieties of alfalfa never exceeded 40% of the total market for the hay planting.

The California Crop Improvement Association has consistently played an important role in the development and growth of this new alfalfa seed industry. Following the conception of a pregnant idea in the minds of Dr. Tysdal, USDA, the breeder of Ranger alfalfa, and some of his colleagues, to produce the seed in the west, out of the area of adaptation, the California Crop Improvement Association took full charge of this new agronomic pregnancy. Under the very able direction and care of Frank Parsons, this new child was born and nursed into a very precocious new industry.

It was the learned foresight of Frank Parsons who recognized that the Certified Alfalfa Seed Council would falter, depending upon voluntary grower contributions. He took the Council under the umbrella of the Crop

Improvement Association in the 1950's, and it has maintained a viable program to this date.

As one of the leaders in the affairs of AOSCA, Frank Parsons played a dominant role in the development and format of the National Certified Alfalfa Review Board. This program has proven to be a truly essential adjunct to the alfalfa seed industry. To my knowledge, no responsible alfalfa plant breeder, either public or in private industry, would think of releasing a new variety until after it has been approved by the Review Board. In these last 19 years (1962), one hundred eighty five alfalfa varieties have been approved as worthy of certification by the Review Board. Nineteen of these, about 10 percent, have been discontinued, withdrawn by their breeders as being obsolete. Of the total number of one hundred eightly five varieties, 45 were produced by breeders as public varieties, and one hundred forty by breeders in industry to be released as a proprietary. In these last four years, 1978-81, there have been six public varieties and 53 proprietary varieties.

The sales of alfalfa seed, as a one variety proprietary, now approach 40% of the total market. An additional 25% of the total market are improved alfalfas, variety not stated, usually packaged as a blend. The remaining 35% of the market consists of alfalfa seed harvested from the hay fields. This seed is usually labeled with its state of origin and, supposedly, the variety of the hay field. The hay fields producing this seed are primarily in the northwestern states, the Rocky Mountain areas, and the Great Plains states. The varieties now being grown for hay in these areas range from the older public varieties to occasional fields planted with the more recent varieties.

For a conclusion to this brief history of the improved alfalfas in the market place, let me say this: "In this nation of free enterprise, with the freedom to make our own mistakes, about 30 years ago, there were four fools who tread where the angels feared to go, in this new alfalfa seed industry."

The Story of Certified Alfalfa Seed Production in California

Frank Parsons
California Crop Improvement Association
University of California, Davis, CA

This is too long of a story to be told adequately in fifteen minutes, and too fascinating for me to tell in the manner it deserves. California was not famous for its alfalfa seed, prior to the late 1940's. Infamous might be a better word, as far as the north central-northeast U.S. was concerned. Seed of California origin was presumed to be no good, would not survive the winters, nor be adapted in any way. Actually, for many years seed of winter hardy varieties growing in a small area of northern California was being shipped into the north central states, but I doubt if the words "California Origin" appeared in very large type on the labels. California growers had for years grown rather large quantities of seed of strictly non-hardy types, for use in the state or for export to areas where these types were adapted. Certification of alfalfa seed in the state was confined to very small amounts of such varieties as California Common, African, and Indian.

Then came Ranger! Many of you are too young to have been around then, and certainly not aware of the splash it made. Bacterial wilt was a serious problem, and getting steadily more serious. Ranger alfalfa was obviously a variety which gave promise of being desirable in a large area. Most of this area, however, had long since found that producing seed in dependable and adequate amounts was usually not possible. What to do? If the advantages of using Ranger were to be realized fully where was the seed to come from?

There wasn't much question it had to be from the western states. Montana, Idaho, and to some extent other states had traditionally grown such seed. Constant and adequate supplies, though, were doubtful. California was the place where seed yields were good, quality excellent, and where amounts necessary for supplying the rest of the country might be counted on. But, there was a stigma surrounding alfalfa seed grown in a warm climate. There were questions, such as would its winter hardiness, and maybe some of its other good features be lost.

A few alfalfa workers had been quietly postulating that seed could be grown outside its region of adaptation with no deleterious effects if it were kept to one generation of production. I am not sure as to the sequence of events, but conferences were held and rules for regulating such production were drawn up. Of course certification people were brought in to the discussion very early. If we were going to have assurance that seed was in fact only one generation out of its region of adaptation, certification was the answer. Certified seed standards provided for such things as freedom from varietal mixtures, isolation, etc. With alfalfa, however, what constitutes one generation of seed production? Limiting it to just one year's production was not feasible. Out of the numerous conferences came the decision that seed could be produced from stands up to six years old, when cultivars were grown outside the region of adaptation. Volunteer plants had to be controlled and either foundation or registered class seed had to be planted to produce certified seed.

California came onto the scene, with certified Ranger alfalfa being grown on a small acreage in 1946, with dramatic increases for many years thereafter. Other varieties followed, like Buffalo and Atlantic. As soon as California grown certified seed became available experiment station workers began to put out "Trueness to Type" plots to try to determine if this warm country grown stuff really was as good as if grown in a climate similar to that where it was to be planted for hay production. There were still skeptics, who were afraid this new source of seed would not be satisfactory. Gradually, however, all this concern faded into the background and California certified alfalfa seed began hitting the market in large amounts. There no longer was any reason for planting anything other than the best variety for one's area. New seed companies appeared, dealing exclusively in alfalfa seed -- for the most part certified.

From nothing in the mid-40's, California production of certified alfalfa seed, for export largely, rose to 63,000,000 pounds in 1957! This was grown on 130,000 acres, 60,000 of which was in Kern County (surrounding Bakersfield). Sooner than this, however, it became obvious that seed production was surpassing consumption. Seed at times was piling up without a market. In 1953 four seedsmen, Lloyd Arnold, Harold Loomis, Evart Vandermuelen, and Ward Waterman, got together to try doing something to stimulate alfalfa usage. The question as Ward expressed it was "Are we overproduced, or under consumed?" With the help of Richard Crabb, who was an advertising agency person in Chicago, they got the Certified Alfalfa Seed Council underway. The Council, supported by grower money, is alive and well today -- still doing a great job extolling the virtues of alfalfa and working closely with land grant institutions in the consuming states. Other western states are now participating with California, in support of the Council.

I must say something about production problems and methods. We planted in rows from the very beginning, for various reasons. First, the planting seed (foundation) was scarce and we found we could get good stands with a pound per acre, occasionally going to considerably less if a small increase of a new variety was necessary and only a few pounds or ounces of seed available. Then, too, by putting it in rows we could control weeds and volunteer alfalfa plants better, and get good seed production the first year. Also, our growers understood they were growing a crop considerably different than hay. They were growing alfalfa for its seed, like corn or wheat, and they'd better manage it as such. Naturally there were problems. Fortunately there was a man who worked out the solutions. His name is Luther Jones.

Luther was as good a down-to-earth agronomist as the University of California ever had. Assigned to seed production problems, he probably came closer to earning his salary than many of the rest of us in Agronomy. At least the farmers with whom he worked came to believe they were getting full benefit out of their tax money going into the University. Luther was the right man for his time. Our growers had to learn fast, how to best grow alfalfa seed and he helped teach them.

Row spacing, how to establish a stand, how much to irrigate, how best to control weeds -- all had to be done properly just to get the crop to the stage where even more critical problems faced the grower. How to control harmful insects, usually the lygus bug, how to manage the beneficial insects needed for pollination -- these matters became important as the season progressed. Incidentally, the honey bee is our most reliable pollinator despite many efforts to utilize leaf cutters and alkali bees. The bee people deserve much credit for our success in producing alfalfa seed. Most of our growers rely on the commercial bee keeper for pollination.

Once a crop of seed is made the grower is still not out of the woods. We used to cut and windrow alfalfa, harvesting from the windrow. A brisk wind could roll those windrows all over the place and create an enormous seed loss. Defoliation and harvesting the standing plant is now preferred. Seed losses from shatter are less, though still a threat. And, a lot of seed can be lost out the back end of the harvester, if not properly adjusted. Also, damage to the seed coat can be disastrous if the harvester is not set properly.

Maintaining an interested in alfalfa seed production has not always been easy. Yields are a bit too unpredictable, with the best management. A 500 pound state average may sound pretty good, but it won't keep the better grower in business. Then, too, yields often just begin to taper off in an area, for no apparent reason. I mentioned Kern County earlier. Not much alfalfa seed is grown there anymore. Production has shifted, now heavy in Fresno County.

With the rapid growth in certified alfalfa seed production in California, certification had to grow -- not only in volume, but in efficiency. Shipments of seed to other states often involved interagency certification, which provided for such things as rebagging -- to get the seed into customer packages -- or recleaning, perhaps to remove something which hadn't been gotten the first time; or blending, to bring several small lots into one large uniform one. We had to streamline office procedure to stay on top of the increased work load. To keep ahead of the enormous number of samples coming in we had to adopt faster methods for examining and testing the seed in the laboratory. Jones was active here too. He worked with Rodney Cobb machines to devise ways to expedite the work. Their belt fed machines for conveying seed under the eyes of a technologist looking through a low power microscope are still in use. Their discovery that germination can be predicted on the basis of seed coat injury, as observed under 10X magnification, speeded up the certification process enormously. This isn't orthodox seed germination technique but it serves for certification.

We began to accept the seedman's test, "Seed Handler's Test" it is called, for the test information needed on the label. We took such label information off the certified seed tag, and began to issue serially numbered tags and labels with only variety and crop name on them. We developed a system for "accredited" handlers, where more responsibility is placed on the handler for supervising seed conditioning, for drawing samples and for attaching labels. We began allowing what was called "Pre-tagging", where handlers were permitted to attach tags at time of cleaning, on the assumption that the seed would meet certification standards. A skilled operator can usually tell if a lot is going to make it. "Hold" tags were used on pretagged lots, to assure that the seed was not moved until it was in fact certified.

I think we can state that during this period, of the 1950's and 60's, seed certification came of age. It recognized industry needs and met them. It became capable of dealing with large quantities of seed, and of providing services adequate to supply any need for agriculture's planting seeds across North America. I am not saying that California certified alfalfa was responsible for all this progress. Other crops were involved. However, our great leap into alfalfa seed production certainly sharpened the need to really "get with it."

We were not far into this alfalfa revolution when private varieties began to show up. I won't attempt to say who was first, but first one company and then another established alfalfa breeding programs. You are well aware of what has happened since. A high percentage of the varieties and acreages is now proprietary. I like to think that certification played an extremely important role in this development. It provided the means wherein companies could establish identity, and a certain degree of acceptability, for their varieties. Plant Variety Protection Act was then something for the future. It is rather significant that when the act finally was passed it contained Title V, which provides for sales of a variety only as certified.

Over 100 varieties, mostly proprietary, are being grown for certification at the present time in California. According to my calculations, and I haven't taken the time to check the records, we have produced and certified 1 billion pounds of alfalfa seed. Where we go in the future remains to be seen, but I maintain California has made quite a splash in supplying alfalfa seed of improved varieties for the United States.

Morphology and Mechanical Strength of Alfalfa Leaves: Application to Breeding a Bloat-safe Alfalfa

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Compared to bloat-safe legume species, the bloat-causing legumes have a fast initial rate of digestion in the rumen (1). Our strategy in breeding a bloat-safe alfalfa cultivar has been to reduce bloat potential by selecting for low initial rates of digestion. The object of the work in this report was to examine the relationships among leaf morphology, mechanical strength and initial rate of digestion. Comparisons of bloat-safe and bloat-causing legumes have indicated that leaf morphology influences digestion and that the following characteristics might reduce the bloat-causing potential of alfalfa:

a) thicker and/or stronger layer (cuticle and outer epidermal cell wall),
b) thicker and/or stronger mesophyll cell walls,
c) a more prominent secondary and tertiary vein structure.

Such changes should reduce the initial rate of digestion by:

a) slower leaching of nutrients from the cell,
b) providing barriers to microbial penetration of the leaf,
c) allowing less tissue damage during chewing,
d) slowing the rate of cell rupture.

Since these changes should likely increase the mechanical strength of the leaf, we have used a number of mechanical treatments to disrupt leaves (2), and have subsequently measured the extent of tissue damage and cell rupture.

Although alfalfa leaves have a relatively low resistance to mechanical damage, it is possible to select plants which incur less tissue cell rupture. Of the mechanical treatments used, sonication has allowed the greatest number of plants to be evaluated per day with minimal equipment requirements. With field-grown alfalfa, values obtained by this method correlate with 3 hr 'in situ' nylon bag digestion results, indicating a relationship between mechanical strength and initial rate of digestion.

We have completed the first cycle of selection of alfalfa plants using the sonication method. Future plans call for several more cycles of selection followed by an evaluation of the bloat-causing potential of the selected plants.

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Nylon Bag Selection for Reduced Bloat Potential in Alfalfa

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Our main screening method to breed a bloat-safe alfalfa is a modified nylon bag technique using fistulated cattle. Using this technique we are selecting alfalfa plants with cells more resistant to microbial degradation. Selection is based on a minimum of two cuts and duplicate samples for each cut. We are able to analyze 40 plants/day, in duplicate, on a routine basis. Because of a relatively high daily variation (due in part to the cattle variation), selections are made from plants analyzed within days. Time of digestion (3-4 hours) of the plant samples in the fistulated cattle is adjusted to obtain approximately 50% DM loss, at which time there is about 8% of cell wall (NDF).

Because we are selecting for a character which is influenced considerably by the environment, we are gradually finding the biological parameters for optimum selection. Reproducibility between cuts (within genotypes) is best when alfalfa is in the lush, pre-bud stage of growth. Reproducibility is low with drought-stressed plants, old and diseased plants, and from winter growth of alfalfa plants in a greenhouse (low light intensity, cooler temperature). Under such conditions variation is considerably reduced, and all plants tend to resemble the LIRD (low initial rate of digestion) selections. Progress was impeded and a heritability study lost by a severe drought at Saskatoon in 1981.

Differences between the LIRD and HIRD (high initial rate of digestion) selections made at Saskatoon have been independently confirmed by rumen microbiologists (Cheng and Fay) using an in vitro digestion technique. Similarly Cheng and Fay (personal communication) have demonstrated a greater DW loss by leaching in the HIRD selections. Preliminary data indicate differences in mineral composition of cell wall fractions between LIRD and HIRD plants (Lees, unpublished data).

Spaced plants were established at Lethbridge in 1981 to provide for a preliminary evaluation in 1982 of LIRD and HIRD cycle 1 selections in sheep feeding trials. Release of mesophyll cell contents into ruminal contents will be measured. Cycle 2 selections will be completed at Saskatoon in 1982 and a third cycle in 1984 before final synthesis of LIRD and HIRD populations and subsequent animal pasture trials to verify reduced bloat potential of the LIRD synthetic.

Testing a Heterozygous Block Concept in Advanced Generations of Medicago sativa - M. falcata Hybrids

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Medicago falcata germplasm has played a significant role in the breeding of cultivated alfalfa. Variegated flowers represent the co-expression of M. falcata genes for yellow pigment and M. sativa genes for blue-purple. Plants with variegated flowers usually segregate indicating genomic heterozygosity at least for chromosome segments controlling flower color.

In crosses of plants carrying M. sativa or M. falcata germplasm, Srivatanaponge and Wilsie (3) found that highest yields were obtained from interspecific intervariety crosses, followed by intraspecific intervariety crosses, and finally by intravariety crosses. Durnev (2) reported an increase in yield in plants with variegated flowers over plants with pure parental flower colors. Burton (1), however, could show no significant correlation between yield and flower color in two different segregating F_2 populations. "Honeoye," bred in part by selecting plants with variegated flowers out of Saranac (R. P. Murphy, personal communication), has a high percentage of variegated flowers and outyields Saranac at several locations in the North Central Region. The experimental hypothesis that we tested was that the genomic heterozygosity of loci linked to flower color loci could contribute a measurable increase in vigor.

Seventy-two segregating S_1 families from plants with variegated flowers from four sources were studied along with plants segregating in five other unrelated hybrid populations. Dry weight of spaced plants was measured twice and open pollinated seed production was scored once; data were analyzed using a nested design. Among the flower colors segregating within entries (purple, variegated, yellow and cream), there was no trend for the plants with variegated flower color to be superior for either forage yield or open pollinated seed production. Heterozygosity which was lost by fixing the flower colors of the parental species, when compared with the total heterozygosity between the two genomes, was not large enough to show a diminished heterosis in this study.

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Yield in a Diallel Cross of Alfalfa with Half-Sib Seed Lines from Four Cultivars as Parents

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A partial diallel cross with 32 parental lines of alfalfa was evaluated to determine genetic variability and heterosis expressed for forage yield when pest resistant, inbred parents (MSA and MSB) were crossed with noninbred, vigorous germplasm ('Appalachee' and 'Arc'). The parental lines consisted of eight half-sib family seed lines from each of the four cultivars and were obtained from remnant seed from the experiment reported by Rowe and Hill (1981). Progeny from intercrossing each of the parental lines (parental syn-2's) were also included in the experiment.

Variation among parental syn-2's was significant, but the mean square for populations was more than 20 times larger than the mean square for lines within populations. General combining ability variation was significant, and the component due to cultivars from which the parents were derived was more than 20 times larger than that for lines within cultivars. Except for the component due to cultivars from which the parents were derived, specific combining ability effects were not significant. The relative importance of general combining ability to specific combining ability and of cultivars to lines within cultivars would probably have been different if clones instead of seed lines had been used as parents.

The crosses had significantly greater average yield than the mean of the parental syn-2's for each of the parental line combinations. The greater yield in the crosses was probably the combined result of heterosis in the crosses and inbreeding in the parental syn-2's.

Crosses with lines from MSA or MSB as one of the parents yielded significantly less than crosses between lines from Appalachee and Arc, and crosses between lines from MSA and MSB yielded significantly less than crosses from any other cultivar combination. The inbreeding that resulted from prolonged, intense selection in MSA and MSB appears to be great enough to prevent their use as commercial cultivars individually or in crosses with each other. The inbreeding depression in crosses is not completely eliminated when parental lines from MSA or MSB are used in combination with unrelated, noninbred parents.

The parental syn-2's from MSA had greater average yield than those from MSB, but parental lines from MSA had lower general combining ability than those from MSB. The results suggest that performance of advanced generations of parental lines may not be a good indicator of their performance in crosses.

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Effectiveness of Selection in Tandem Generations for the Same or Correlated Trait

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Theoretical studies on the relative effectiveness of various breeding schemes (Hill and Haag, 1974; Rowe and Hill, 1981), though somewhat interesting and informative, are sometimes faulted for their simplistic and naive assumptions about the genetics of a breeding situation. One deviation from reality is the assumption of random mating equilibrium (RME) in the population subjected to selection (i.e. gamete and genotype frequencies are entirely determined by allele frequencies in the population). Deviations from RME are expected for the population which has been previously selected for the same or correlated trait or which has been intercrossed with genetically foreign germplasm. Since RME is approximated only after 4 or 5 generations of random mating, most cycles of selection are on populations not in RME.

This study investigated the effect that deviation from the assumption of RME in the selected population has on the covariance of selection. The covariance relates the genotypic value of individuals or progenies screened to the frequency of desirable allele in the selected individual or progeny. The genetics at a single locus with two alleles, (B and b where B is the desired allele) with additive genetic action and four types of dominance genetic action was investigated for mass selection and for half-sib progeny test selection (HSPT).

The response surface of the covariance as a function of the frequencies of gametes BB and Bb indicated that when the frequency of BB was greater than p^2, where p is frequency of B in the population, the covariance was increased up to 200% of the covariance with the selected population in RME. Similarily when the frequency of gamete BB was below p^2, the covariance was reduced below that with selection in a population in RME and approached zero in some cases. The difference in covariance associated with mass selection and half-sib progeny test selection is 2:1 (Hill and Haag, 1974), but for the population not at RME this ratio holds only for additive genetic action. The HSPT ranged from 80 to 120% of one-half of the covariance with mass selection for different gamete frequencies with each type of dominance.

Thus it is beneficial to increase the frequency of BB gametes, but a practical problem arises in the identification of the genotypes which produce the BB gamete (genotypes BBBB, BBBb, and BBbb) when there is dominance. A practical caution from this study is that the gain per cycle of selection for a relatively rare allele could be improved by using a second generation of random mating between selection cycles (i.e. select for an uncorrelated trait for 1 generation) to improve the covariance of selection.

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Hexaploid Alfalfa Derived From 3x X 6x Crosses: A Status Report

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Artificially produced hexaploid alfalfa (2n=6x=48) has had limited value to the breeder because of its low vigor and fertility, due, in part, to colchicine-induced chromosome doubling during hexaploid synthesis. Triploid by hexaploid (3x X 6x) crosses are a relatively simple method of producing hexaploid plants in which the detrimental effects of colchicine treatment may be reduced. Furthermore, 3x X 6x crosses are a potentially powerful mechanism for combining exotic diploid and elite tetraploid germplasm, albeit at the hexaploid level. The objectives of this research were to study vigor, fertility and chromosomal stability in hexaploid alfalfa derived from 3x X 6x crosses, and to compare these characteristics in the hexaploids to those in naturally tetraploid alfalfas.

A higher percentage of hexaploid plants were produced from 3x X 6x crosses in which the 6x parent originated form Wisconsin germplasm WI6XGP-1[1], compared to those crosses in which the 6x parent was produced by colchicine treatment of triploids. Furthermore, fewer hexaploid plants were produced in 3x X 6x crosses in which the parent was derived from a *Medicago sativa* ssp. *coerulea* (2x) X *M. sativa* ssp. *sativa* (4x) cross, compared to crosses in which the 3x parent was a *M. falcata* (2x) X *M. sativa* ssp. *sativa* (4x) hybrid.

Hexaploids having *M. sativa* ssp. *coerulea* in their pedigrees were generally not as fertile nor as vigorous as those derived from *M. falcata* sources. All hexaploids derived from 3x X 6x crosses were less fertile, and most were less vigorous than tetraploid checks. Improved fertility and vigor were observed in Syn. 1 populations produced by intercrossing the original (3x X 6x)derived hexaploids. Two cycles of recurrent selection for hexaploidy in (3x X 6x)-derived materials produced a population having 25.7 % hexaploid plants (21.1% after one cycle). The percentage of "near hexaploid" plants (2n=47, 48 or 49) was 51.4% in this population after two cycles of selection (43.7% after one cycle).

These observations suggest that it is possible to make gains in fertility and chromosomal stability by selection in large, broad-based hexaploid populations derived from 3x X 6x crosses. Further research is needed to determine if adequate levels of vigor, fertility and chromosomal stability can be achieved for development of hexaploid cultivars.

[1] Derived primarily from spontaneous hexaploids from the cultivar 'Saranac'.

Relationship Between Seed Weight and Yield Heterosis in a Diallel Among Nine Germplasm Sources

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Nine primary alfalfa germplasm (GP) sources were introduced into the United States between 1850 and 1947 (Barnes et al., 1977). These nine GP sources represent most of the genetic diversity now available in U.S. alfalfa cultivars. In order of decreasing winter hardiness, they are Medicago falcata, Ladak, M. varia, Turkistan, Flemish, Chilean, Peruvian, Indian, and African. Most new alfalfa cultivars utilize a broad germplasm base, often containing genes from most of the nine original GP sources.

Several studies have reported positive correlations between alfalfa seed size and subsequent seedling vigor. We believed that the relationship between combining ability for seed weight and seedling vigor deserved further investigation. Our objectives were to examine general and specific combining ability for seed weight, seedling vigor, root yield, and forage yield in a diallel among the nine primary alfalfa GP sources, and to evaluate the use of hybrid seed weight as a rapid assay to identify alfalfa germplasms with superior combining ability for these characters.

The nine alfalfa GP sources were crossed in diallel fashion (including reciprocals). Seven genotypes were used to represent each GP source in seven different nine parent diallels. Seeds/pod and seed weight were determined for seed from parents, single crosses. Seed was planted into fertilized greenhouse sand benches. Seven weeks after emergence, seedling vigor was measured and entries were transplanted to space plant nurseries at Becker and Rosemount, MN. All entries were subsequently evaluated for secondary and fibrous root score, crown and root weight, forage yield, and fall dormancy reaction.

Greenhouse results indicated that in crosses among the nine alfalfa GP sources, number seeds/pod and seed weight were primarily controlled by additive gene action, while seedling vigor was primarily controlled by non-additive gene action. Hybrid seed weight was positive correlated with seedling vigor (r=0.25*, d.f.=72). However, significant maternal effects and seed parent X pollen parent interaction effects on seed weight led to inconsistent relationships between hybrid seed weight and seedling vigor.

In the field, additive gene action was important in the expression of all characters. Non-additive gene action was important in the expression of secondary root score, root weight, and second harvest forage yield. Generally, the more dormant sources produced larger root and greater first harvest forage yields, while non-dormant sources produced larger crowns and greater second harvest yields. Fall growth scores were useful in explaining patterns of root and forage production over the season. No correlation was observed between seed weight and total forage yield. Heterosis for seed weight appeared confounded with seed parent and pollen parent effects on seed weight when sources with divergent fall dormancy reactions were hybridized. However, controlled crossing between GP sources possessing similar dormancy reactions might improve the usefulness of seed weight as an assay to rapidly evaluate diverse alfalfa GP sources for combining ability.

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Seed Yields from Breeder and Foundation Seed of Eight Alfalfa Cultivars 1/

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The purpose of this study was to compare seed yields from breeder and foundation seed classes of eight alfalfa (Medicago sativa L.) cultivars, 'Agate', 'Arc', 'Cody', 'Dawson', 'Kanza', 'Ramsey', 'Ranger', and 'Vernal'. Breeding methods used in developing the cultivars and generations of synthesis of breeder and foundation seed classes varied among cultivars.

Breeder and foundation seed of the eight alfalfa cultivars were seeded at 2.2 kg/ha in a randomized block split-plot design of cultivars as main plots and seed classes subplots with 3 replications in a field near Caldwell, Idaho, in the spring of 1976. The experiment was managed to maximize seed yield. Seed yields were determined in 1977 and 1978.

Seed yields obtained from breeder seed averaged over eight alfalfa cultivars were lower than those obtained from foundation seed in 1977 and for the 1977-1978 average. Seed yields obtained from breeder seed were lower than those obtained from foundation seed for 2 of 8 cultivars, Ramsey and Vernal, in 1977 and for the 1977-1978 averages. Alfalfa breeders need to conduct seed yield tests that include at least one commercial seed generation of their experimental cultivars compared with a check cultivar.

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Introduction to Problems in Continuous Alfalfa

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Alfalfa, Medicago sativa L., is a potentially long-lived perennial that is well known for soil improvement and for livestock feed. While alfalfa is most commonly grown in rotation with other crops, land can be continuously cropped with alfalfa. As stands decline, production declines, and reseeding and continued cropping may be considered. The cause of stand decline should be identified and will give some clues as to the possible future success with continued alfalfa cropping. Success of future establishment and production may depend on diseases, nematodes, drainage, fertilizer, soil moisture, soil type, variety, cutting management, and their interactions or other problems. Reseeding to thicken a stand, retarding or killing remaining vegetation by chemicals followed by no-till interseeding, spring or fall plowing before reseeding, and rotation with another crop for one or more years before reseeding are options to consider.

The purpose of this presentation is to briefly review published work on problems associated with continuous alfalfa.

In the 1909 text, "Alfalfa Farming in America", J. E. Wing said "It is notable that some of the very oldest books on agriculture in referring to alfalfa say land should not be sown again to alfalfa until it has rested some seasons" (27). Furthermore, "There are hidden influences that we do not understand that make soils unfriendly to plants that have grown in them too long".

From 1907 to 1911 Schreiner and his associates published evidence of "soil-sickness" due to organic chemicals directly or indirectly associated with low productivity of soils, including soils continually cropped (22). From research in Kansas and Nebraska that was initiated about ten years later it was apparent that moisture, including subsoil moisture, becomes a limiting factor in alfalfa that is cropped continuously under nonirrigated or limited rainfall conditions (5, 11, 12, 13). That research in Kansas and Nebraska led to the expression that alfalfa should be grown only once per generation on a given field because of its ability to deplete soil moisture.

According to Rice (21) beneficial and harmful interactions among plants were documented more than 2000 years ago. Theophrastus, in his "Enquiry into plants" written before 285 B.C., pointed out that the legume "chickpea" does not invigorate the ground as other related plants but "exhausts" it instead. Moreover, "it destroys weeds and above all and soonest caltrop". Pliny in his "Natural History" written in the first century A.D. has many references to apparent effects we now term "allelopathic", the term most often used according to the definition of Rice in 1974 (19) as "any direct or indirect harmful effect of one plant on another through production of chemical compounds that escape into the environment". Rice explained (20) that many factors, both genetic and environmental affect the amounts of potential phytotoxins produced. Once the phytotoxins are produced and escape into the environment, they begin to be decomposed either by microorganisms or by chemical action not involving microorganisms. Thus, potential allelopathic effects depend basically on the relative rates of addition of the allelochemics to the environment and decomposition, or inactivation. Rice (20) concluded "There is little doubt that all plants, including microorganisms, produce compounds which, if present in appropriate concentrations, can inhibit or stimulate the growth of other plants". Rice (21) also reviewed "Some roles of allelopathy in pasture and forage crops" in 1980. No references were made to allelopathy in alfalfa. However, allelopathic effects of legumes other than alfalfa were presented and discussed. Autotoxicity is the term being used in those situations where a plant produces compounds that are toxic to itself. Both terms, allelopathy and autotoxicity, have been used in recent reports on alfalfa seedling growth, establishment, and production.

Thickening an established alfalfa stand by interseeding has not been successful, in general. Growth from old plants may shade new seedlings and prevent their establishment. Ten or more old alfalfa plants per m^2 in notillage seeding of alfalfa into old stands of alfalfa seriously hindered establishment of new seedings (16). Alfalfa contains water soluble substances toxic to itself and to other plants, and water extracts of immature alfalfa forage had the highest phytotoxic effects on corn seedlings in a laboratory study (6). Water-soluble substances toxic to new alfalfa seedlings could be accidentally incorporated into the soil while interseeding alfalfa to thicken old stands. Common mulching materials including alfalfa contain phytotoxic substances (6).

Autotoxicity for alfalfa seedling growth was reported in laboratory and greenhouse studies (8, 10, 14, 15, 17). The autoxicity was reported in fresh alfalfa leaves, stems and crowns, and in dry hay, old roots, soil, and soil residues. Damping-off of alfalfa seedlings due to Phythium spp. was effectively controlled by fungicide seed treatment in greenhouse tests with no apparent detrimental effect on Rhizobial inoculation (1, 24). A systemic fungicide used as a soil drench controlled Phythium spp. under continuous alfalfa field conditions in Nevada (25). In a prepared seedbed in Illinois, preplant incorporation of a fungicide did not improve alfalfa stands (3). Poor alfalfa establishment and persistence on a coarse loamy sand in Minnesota was probably due to a fungi-nematode interaction (23). Phytotoxic substances in the soil were associated with continuous alfalfa cropping in Alberta, Canada (4, 15, 26).

In Nebraska, relative five-year total alfalfa yields for land not previously cropped to alfalfa, land continued in an old alfalfa stand, and for land reseeded to alfalfa were 100, 69, and 68%, respectively (12). It was no more difficult to secure a satisfactory stand of alfalfa on upland soil previously in alfalfa than on land never having grown alfalfa in Nebraska (13). Old alfalfa stands were plowed and the land was cropped to annual grain crops before a new alfalfa seeding was made. In Illinois, relative stands after six years of corn-alfalfa, corn-soybean-alfalfa, and continuous alfalfa cropping sequences were 100, 83, and 43%, respectively, while relative yields were 100, 92, and 50%, respectively (14). Alfalfa was treated as an annual, fall-plowed and spring-seeded each year for six years in the continuous alfalfa cropping sequence. There were two years of alfalfa in the corn-soybean-alfalfa sequence and three years of alfalfa in the corn-alfalfa sequence where alfalfa was also treated as an annual. In Virginia, the six-year average yield of eight alfalfa cultivars grown on land not previously in alfalfa was 27% higher than the yield of the same cultivars grown on land previously in alfalfa (2).

Some allelopathic compounds have inhibited the uptake of various minerals (20). Periodic soil testing and the application of fertilizer can correct nutrient deficiencies that result from cropping to alfalfa (9).

Evidence for genetic selection in alfalfa against toxic soil conditions in Alberta was negative (4, 15) but data in Illinois indicated genetic variability was present in alfalfa cultivars for allelopathic effects (14). The toxicity of wheat straw extracts to germination and growth differed among cultivars (7). Selection and breeding for beneficial allelopathic effects has been suggested (18, 21).

The purpose of this symposium is to hear presentations of current research on problems with establishing and producing alfalfa in continuous cropping systems. There is a need to define those field conditions and cultural practices that prevent establishment and production under continuous cropping, or make continuous alfalfa noneconomic compared with other competitive crops. Our communications with the popular press must carefully state whether our results were obtained under laboratory, greenhouse, or field conditions. Blanket statements that you can't grow alfalfa continuously are not scientifically correct and are detrimental to all aspects of alfalfa production.

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Evidence of Auto-Toxicity in Alfalfa

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The practice of planting alfalfa into alfalfa sod or depleted stands usually has been unsuccessful. This may be due to a high level of pathogenic organisms in the soil, auto-toxicity or both. If these obstacles can be overcome, then the farmer could plant alfalfa after alfalfa or seed into depleted stands to increase production. The reasons why one cannot successfully pursue this practice is not fully known and is the topic of this report.

In a greenhouse study 'Lahontan' and 'Moapa 69' alfalfa were grown in soil in which alfalfa had been grown the two previous years and in a fallow soil of the same origin. Dried alfalfa shoots or roots at 0.5% of the weight of the soil were added either before or after steam pasteurization or fumigation. Seedlings of both 'Lahontan' and 'Moapa 69' were smaller when grown in soil which alfalfa had been the previous crop when compared to those grown in fallow soil even though both soils were either steam pasteurized or fumigated. Addition of ground foliage or roots to both soils after steam pasteurization or fumigation also reduced the size of the plants when compared to soils where plant tissue was not added. Addition of foliage and roots prior to soil treatment had a lesser effect on growth. The smaller seedling size is attributed mainly to auto-toxicity of alfalfa even though some pathogens may have been present after steam pasteurization or fumigation or were introduced by the addition of plant material to the treated soil.

Field studies are being initiated at four locations in Nevada to further investigate the role of both pathogens and auto-toxicity when establishing alfalfa in depleted stands or where alfalfa has been the previous crop. Greenhouse studies will also be conducted to further study auto-toxicity of alfalfa.

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The Effect of Allelopathy or Autotoxicity on Alfalfa Seedling Establishment

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Greenhouse tests were conducted to locate the influence of previously grown alfalfa upon seedling establishment. Treatments used were: (1) soil that had supported growth of alfalfa plants, from which all plant material had been removed, (2) alfalfa tops mixed into greenhouse potting soil, (3) alfalfa roots mixed into potting soil and (4) potting soil alone was used as a control. The stand obtained in soil where alfalfa had been grown was not substantially reduced, however, where tops, or roots to a lesser extent, had been incorporated in the soil the stand was reduced in most cases.

In another study, alfalfa seeds were germinated in plastic boxes on moistened blotters. The controls were moistened with water only, whereas half of the others had water containing an alfalfa extract from leaves and the other half had water containing an alfalfa extract from stems. In both cases, seed germination where alfalfa extracts had been added was severely reduced.

A four-month-old stand of alfalfa and an adjoining strip of weedy ground were plowed at the same time. The ground was worked and planted to alfalfa in the fall. The seedlings from several paired blocks were counted. The area where alfalfa was plowed under had 43% fewer plants than where weeds had been turned under.

In another field test, soil that had alfalfa incorporated with a roller-harrow was compared with soil that had been in corn the previous season. The stand of new alfalfa was reduced 85% by the incorporation of alfalfa residue.

Allelopathy or autotoxicity is not only a recent concern. In 1909 (1), it was stated:

> It is notable that some of the very oldest books on agriculture say in referring to alfalfa: 'It endures for many years and afterward may be plowed up and the land sown to corn. Land should not be sown again to lucerne (alfalfa) till it has rested for some seasons.' It is safe to assume that the ancients had seen signs that alfalfa best liked fresh land.

The inability to get consistent and discerning results has probably prevented an explanation of this problem many years ago.

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Allelopathic Effects of Alfalfa

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Several experiments have been conducted at University of Illinois, Urbana, IL to determine the role of allelopathic effects of alfalfa on the establishment of alfalfa. By means of correcting any soil fertility differences and protecting the alfalfa seedlings from fungal attack, results suggest that there is a release of phytotoxic factors from the previous alfalfa crop on re-establishing alfalfa. Field results indicate that the best preceding crop for alfalfa establishment is corn, followed by the various small grains, soybeans, and the worst preceding crop is alfalfa (1). Considering other forage legumes to follow alfalfa in rotation, red clover is the best choice, followed by birdsfoot trefoil. Forage grasses would be good choices to follow alfalfa, either smooth brome or orchardgrass. Early results indicate there are no major genetic differences among cultivars. There was no evidence supporting saponins as being phytotoxic to alfalfa. It is known that saponins are released by alfalfa roots, but whether they are released in proportion to their tissue content is unknown. Exudates of different saponin level cultivars indicated that saponin content is not the suspected phytotoxic factor. There was a strong relationship between radicle elongation and the fall growth characteristics of the strain releasing the exudate suggesting that phytotoxic substances may play a role in fall dormancy.

Laboratory techniques indicate that three-day old alfalfa seedlings release a toxic compound that inhibits germination and retards seedling elongation. Fingerprinting these compounds will enable the plant breeder to select genetic resistance or tolerance to these water soluble compounds.

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Effects of Methyl Bromide and Systemic Fungicide Metalaxyl on Alfalfa Stand Establishment, Persistence, Yield and Disease Incidence

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Damping off seriously reduced stand density of newly seeded alfalfa, in a field which was occupied by alfalfa for seven preceding years, as well as in a field which had not been seeded to alfalfa for 11 years. Soil fumigation with methyl bromide (970 kg/ha) prior to seeding reduced soil populations of pythiacious fungi, reduced damping-off and increased alfalfa stand establishment by about 80%. The systemic fungicide metalaxyl (2) with specific activity limited to phycomcetous fungi, reduced soil populations of those fungi only slightly, if at all when applied as a drench at seeding (2.25 kg a.i./ha), but increased alfalfa stand establishment by more than 200% over the control, and by nearly 80% over methyl bromide. Respective stand counts taken in 90 cm of row for the control, methyl bromide, and metalaxyl treatments were 33, 66 and 90 (at establishment) 16, 31 and 41 (April, 1979), 21, 42 and 40 (May, 1980) and 31, 53 and 66 (December, 1980). Alfalfa dry matter yields (MT/ha) for the first cutting in 1979 were 1.41, 2.64 and 3.88 for the control, metalaxyl and methyl bromide respectively [HSD (.05)= 0.40]. Only SW-32, a nondormant germplasm yielded significantly less than the other alfalfas, and its stand count was also significantly less than the others. Average yields for the second and third cuttings during the 1980 season (MT/ha) were 1.97, 2.64 and 3.25 for the control, metalaxyl and methyl bromide respectively [LSD (.05)= 0.40]. Yields for Washington SNI, 'Washoe' and Syn-XX were 3.56, 3.31 and 2.87 MT/ha respectively. These were significantly higher than 'Agate', SW-32 and 'Narragansett' which yielded 2.08, 2.02 and 1.90 MT/ha respectively [LSD (.05)= 0.34]. The former three all have resistance to stem nematode *Ditylenchus dipsaci* whereas the latter three do not. Although the field was heavily infested with *D. dipsaci*, similar yield differences failed to occur in 1979, possibly due to delayed activity by the nematode. Destructive samples taken in December, 1980 were examined for stem nematode, clover root curculio (*Sitona hispidula*), crown rot complex and Phytophthora root rot (*Phythophthora megasperma* f. *medicaginis*). Washington SNI, 'Washoe' and Nevada Syn-XX had significantly fewer plants with stem nematode symptoms than the others. Treatments appeared to have no major effect on stem nematode except that metalaxyl appeared to increase the resistance in Nevada Syn-XX. No differences among treatments were detected in the other disease or insect evaluations. The early advantage metalaxyl gave in stand establishment is attributed to control of *Pythium* sp. since 'Agate' with resistance to *P. megasperma* f. *medicaginis*, failed to establish any better than those cultivars and germplasm without resistance (1).

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Legume Production Following Fallow, Legumes, or Grasses

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Allelopathy occurs when a plant reduces germination, growth or development of another species. If the inhibition is of the same species, it is autotoxicity. In either case, reduced yield of the desired species is generally the end result.

The objective of this experiment was to determine if allelopathic or autotoxic effects exist by determining the yields of alfalfa (A), red clover (RC), or birdsfoot trefoil (BFT) seeded on a prepared seedbed following plowed-under A, RC, BFT, rye, sorghum-sudangrass (SS) or fallow soil. Six treatments of 6 x 30-ft. plots in four replications on a Conover loam, pH 6.8, were used to provide different sources of potential allelopathic or autotoxic material:

Species	Seeded	Method	LB/A	Fertilizer
Saranac A	8/23/79	Broadcast Seeder	20	0+100+100
Arlington RC	8/23/79	Broadcast Seeder	20	0+100+100
Mackinaw BFT	8/23/79	Broadcast Seeder	20	0+100+100
Wheeler Rye	9/9/79	11-Hole Drill	120	25+100+100
Haygrazer SS	5/23/80	11-Hole Drill	40	54+100+100
None, Fallow	--	--	--	0+100+100

Legumes established in 1979 were cut and removed on June 6 and July 22, 1980. Legume regrowth and the first growth of the SS and rye were sprayed with glyphosate at 2 lb/a.i./acre on August 1. The area was plowed on August 14, disked, and dragged for an excellent seedbed. The three legumes were seeded with a fertilizer-grain drill with band seeder and press wheels on August 26, 1980. Three harvests were made in 1981. One of alfalfa was on June 17, 1982.

Yield of 3 legumes, T/A, 12% H_2O year after seeding, and of A in the 2nd year after seeding on a plowed, prepared seedbed after 3 legumes, 2 grasses and fallow were plowed under.

Crop Seeded on Crop Plowed Under	Crop Plowed Under: Check Fallow	A	RC	BFT	SS	Rye	LSD 5%	CV %
				1981				
Alfalfa	5.85	5.73	5.90	5.76	4.77	5.30	.60	7.7
Red Clover	5.13	5.06	5.10	4.93	5.13	5.02	.77	10.8
Trefoil	5.08	5.09	4.97	5.06	4.98	4.96	.70	9.7
				1982				
Alfalfa	3.28	3.58	3.46	3.50	3.44	3.06		

Data show there was no significant difference between the first and second-year yields of alfalfa following plowed-under A, RC, BFT, or rye compared to A seeded on a fallowed seedbed. Only A following SS had a significantly lower yield than when seeded on fallow ground. There was no significant difference between the first-year yield of R or BFT following any plowed-under crop or fallow.

Results of this experiment, based on yield data, indicate there was no autotoxic or allelopathic effect of A, RC, or BFT seeded on plowed and tilled soil after that legume or either of the other two legumes, rye, or sorghum sudangrass (exception is A following SS) was plowed-under before seeding.

Alfalfa Establishment and Production With Continuous Alfalfa and Following Soybeans [1/]

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Alfalfa (Medicago sativa (L.)) is commonly grown in rotation with other crops but land can be continuously cropped with alfalfa. The purpose of this study was to determine the alfalfa stand establishment and production of cultivars that differed in disease resistance in a typical continuous alfalfa cropping situation, with limited supplemental irrigation.

Two alfalfa cultivar experiments were spring-seeded in 1970 at the Nebraska Agric. Exp. Stn. Mead Field Laboratory on a Typic Argiudoll soil with no known prior alfalfa cropping. Stand and forage yield were determined in 1970-72. The area was plowed in late May of 1975 after the first cutting and fallowed. Soybeans were seeded in 1976 on subareas planned for seeding alfalfa in 1977 and 1978, and in 1977 on the subarea planned for seeding alfalfa in 1978.

Two alfalfa experiments were spring-seeded per year in 1976, 1977, and 1978. In foliar fungicide experiments, stands in the year of seeding were excellent in 1976-1978. The decline in stand from 1977 to 1978 was greater for 'Ranger' than for 'Arc'. Anthracnose and phytophthora contributed to stand losses. Forage yields did not differ among fungicide treatments or between cultivars in the year of seeding or in subsequent years. Differences in forage quality among fungicide treatments and between cultivars were few and inconsistent. Anthracnose and foliar diseases were at subeconomic levels. In fungicide seed treatment experiments, stands in the year of seeding were excellent in 1976-1978 and did not differ among treatments or between cultivars. Stand decline in 1978 was greater for 'Dawson' than for 'Agate'. Forage yields did not differ among treatments or between cultivars in the year of seeding with one exception. Forage yields of Dawson were less than those of Agate subsequent to the year of seeding.

Initial stands were excellent on the same land area where alfalfa was seeded in 1970 with no known previous alfalfa cropping, after plowing the five-year old stand of alfalfa in 1975 and seeding alfalfa in 1976, and after one or two years of soybeans and seeding alfalfa in 1977 and 1978, respectively. Average forage yields of the experiments seeded in 1970 were 26% higher in the year of seeding, and 40 and 35% higher in the first and second years after seeding, respectively, than in the experiments seeded in 1976 to 1978.

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Interference Between Companion Crops and Alfalfa

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Approximately 90% of the alfalfa in Minnesota is established with a small grain companion crop, usually oats. The use of companion crops is generally considered to be detrimental to alfalfa establishment and subsequent yield. However, we previously reported that an alfalfa cultivar x oat companion crop interaction occurred for both alfalfa forage and oat grain yield (1). Additional research has been conducted to determine the extent of the interference between oat and alfalfa cultivars (2).

Five oat (Multiline E77; Froker, Lang, Noble and Stout) and six alfalfa (Agate, Cody, DuPuits, Iroquois, Washoe, and MN Allelopathy) cultivars were established together at Waseca, Rosemount and St. Paul, MN. Oat grain and alfalfa forage yield were measured. All oat cultivars reduced alfalfa yields (averaged over cultivars and locations) with Stout causing the largest alfalfa yield reduction and Froker the smallest. At Rosemount, Stout reduced the yield of all alfalfa cultivars except Cody, whereas Froker caused no yield changes. At Waseca, Stout only reduced yield of Iroquois; Froker reduced yield of Washoe and DuPuits.

Only the MN Allelopathy selection reduced oat grain yield over all oat cultivars and environments. Three oat-alfalfa cultivar combinations resulted in grain yield changes over all environments: Noble/Cody had a 3.5 quintals/ha reduction in oat grain yield; Stout/MN Allelopathy had a 3.0 quintals/ha reduction, and Lang/Iroquois had a 2.2 quintals/ha reduction.

In the past, the recommendation for selection of an oat cultivar for use as a companion crop has been to select a short, early maturing cultivar. Our research has indicated that interactions of alfalfa and small grain cultivars may preclude use of this general recommendation. However, it may be possible to select oat cultivars for broad based compatibility with alfalfa cultivars.

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Breeding for Increased N_2-fixation Potential in Alfalfa

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We have been conducting research on alfalfa N_2-fixation for nearly eight years. Our program has three long-term goals: 1) to develop an in-depth understanding of N_2-fixation in alfalfa, 2) to breed alfalfa that is more effective in N_2-fixation, and 3) to develop strategies to apply N_2-fixation information and improved germplasm to agricultural production systems. This report is intended to provide an update on the breeding program.

Early studies established that alfalfa genotypes differed in N_2-fixation and that an association existed between high acetylene reduction rates (AR) and high top weight, high root weight, large nodule mass, and many fibrous roots (3). Nodule mass was shown to be the primary factor affecting AR. The other traits were associated with AR only because they were associated with nodule mass (4). Nodule mass accounted for about 40% of the AR variation among genotypes. We hypothesized that host-Rhizobium interactions were important sources of differences in N_2-fixation. Hardarson et al. (2) demonstrated that alfalfa genotypes selected for high N_2-fixation preferentially nodulated with more effective Rhizobium strains than did alfalfa genotypes selected for low N_2-fixation. We also learned that our previous base populations were susceptible to several important pests.

The last several years have been spent developing six base populations of alfalfa that have adequate levels of resistance to important pests and that when used in strain crosses will provide a heterotic yield response. These populations are now being used in a multiple-step selection program for increased N_2-fixation. The steps include selection for plant vigor at 4-weeks and 13 weeks (1st regrowth) followed by selection for high nodule mass and then high AR. Plants are grown in the greenhouse in nil-nitrate sand benches. The sand is fertilized with P and K and micronutrients and inoculated with mixed cultures of high and low effectiveness strains of Rhizobium. About 5% of the seedling plants are finally selected. These are interpollinated within each base population. The progenies of the selected populations and crosses between the selected populations are evaluated in the field for N_2-fixation by the N^{15} dilution method.

Research by Groat (1) demonstrated an association between AR and the activities of two nodule enzymes, glutamate synthase (GOGAT) and phosphoenolpyruvate carboxylase (PEPC). Efforts are underway to incorporate selection for increased nodule enzyme activity into the breeding program.

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Effect of Seeding Year Harvest Management on Legume Nitrogen Availability for Succeeding Crops

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Forage legumes were once extensively used as green manure crops to improve soil fertility and to increase the yields of subsequent crops in rotations. Interest in the use of legumes as N sources declined when inexpensive forms of inorganic N fertilizer became available. However, recent increases in energy costs have made inorganic N fertilizer more expensive and organic N sources more attractive. The use of symbiotically fixed N produced by properly managed forage legumes in crop rotations may be a means to reduce agricultural energy costs. Our objectives were to determine the effects of harvest management on alfalfa (Medicago sativa L.), red clover (Trifolium pratense L.), and sweet clover (Melilotus officinalis L.) herbage and root dry matter and N yields, and the dry matter and N yields of a subsequent sorghum-sudangrass (Sorghum bicolor x sudanese) crop.

Non-dormant, 'CUF-101' and 'Ardiente,' and dormant, 'Saranac' and 'Agate' alfalfa; 'Arlington' red clover; and 'Madrid' sweet clover were seeded and in the seeding year the legumes were subjected to four harvest management treatments: (H1) harvest with herbage removal except fall regrowth; fall herbage regrowth, crowns, and roots incorporated, (H2) harvest with complete herbage removal including fall regrowth; only crowns and roots incorporated, (H3) cut as H1 with no herbage removal; cut-herbage, fall regrowth, crowns, and roots incorporated, (H4) no harvests; accumulated herbage, crowns, and roots incorporated. The legumes were incorporated into the soil in the fall of the seeding year with a moldboard plow. 'Trudan 8' sorghum-sudangrass was seeded in the spring of the following year. The 'non-harvest' treatment (H4) resulted in a lower total legume N yield than the other harvest treatments for each legume except sweet clover, but a greater N incorporation into the soil than for the treatments in which forage was removed from the plots (H1 and H2). The greatest average N incorporation into the soil (316 kg N/ha) occurred when the legumes were cut and the forage returned to the plots (H3), while treatments H1 and H2 in which all or a portion of the herbage was removed added an average of 86 kg N/ha. Within treatments H1 and H2, non-dormant alfalfa cultivars had lower N yields and N incorporation than dormant alfalfa cultivars and red clover. Sweet clover N yield and incorporation was less than for other legumes within managements H1 and H2, but similar within H3.

Total dry matter and N yields of the subsequent sorghum-sudangrass crop were not affected by previous legume species or cultivars within harvest management treatments. Harvest managements H3 and H4 which incorporated more N to the soil resulted in greater sorghum-sudangrass dry matter and N yields than H1 and H2.

A N budget for the legume/sorghum-sudangrass cropping system indicated that the 'no harvest' treatment (H4) resulted in an average addition to the soil of 40 kg/ha while treatments H1, H2 and H3 resulted in N deficits of 41, 67 and 16 kg/ha.

Stand and $N_2[C_2H_2]$ Activity of Four Groups of Alfalfa Cultivars Grown Under a Line-Source Sprinkler

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The alfalfa cultivars used in the United States originated in various ways and in a wide range of environments. We wanted to test whether selection of alfalfa for high forage yield in favorable environments had been accompanied by a reduction in drought resistance of young plants when these cultivars were grown in more stressful environments. Thirty eight populations were placed into four groups differing in origin, breeding, and adaptation. Group 1 included 14 newly released cultivars, most of which had been intensively bred for maximum forage production in humid or irrigated environments. Group 2 was composed of 10 less highly selected and "older" cultivars, some of which are widely used for dryland hay production in subhumid regions. A third group consisted of 10 cultivars intended primarily for dryland pasture and range plantings. The fourth group had 4 populations which were wild or which had not been intensively selected for agronomic traits.

Field experiments were seeded on 24 June 1980 and 14 July 1981. Planting rates were adjusted to provide one viable seed per 5 cm of row with the 18.3 m long rows at right angles to a line source sprinkler. Four replications were planted each year. The irrigation system produced a water application pattern that was uniform along the length of the line but which decreased linearly from an optimal amount adjacent to the line to zero approximately 12 m distant from the line. Irrigation began the day after each seeding and continued on alternate days thereafter until the completion of the experiments. Sufficient water was applied each time to initiate surface runoff along the source line. Data were obtained on stand, plant weight, leaf water potential, and acetylene reduction activity.

No significant differences in leaf water potential or acetylene reduction activity, $N_2[C_2H_2]$, were found among the groups of populations. Recently released cultivars were superior to all other groups in average stand and plant weight irrespective of drought stress. The commonly encountered contention of farmers and ranchers that "older" cultivars give better stands in drought stressed environments than more recently released cultivars was not supported by the data from these experiments. Although mature plants of cultivars bred specifically for use on dryland pastures and rangelands may survive better under drought and grazing stress than other cultivars, we found no evidence of superior stands, growth, or acetylene reduction during the seedling year.

A Survey of Alfalfa Diseases in Wyoming

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A survey was conducted in Wyoming during 1980-81 to identify diseases in irrigated alfalfa. Randomly selected fields were checked in all major alfalfa producing areas. In addition, "Disease Detection Plots" were established at 6 locations in the state to measure relative severity of foliar diseases. The test consisted of 11 alfalfa cultivars and lines recommended for characterizing disease resistance in alfalfa (1), and were planted in a randomized complete block design with 4 replications. Plots were given a subjective rating for diseases that occurred during 1981. Five test sites located at Afton, Powell, Laramie, Torrington and Riverton, WY were irrigated and one site located at Sheridan was dryland.

General Survey

Diseases found included: alfalfa stem nematode, root-knot nematode, Phytophthora root rot, spring black stem, downy mildew, common leaf spot, winter crown rot and Verticillium wilt. Diseases were generally widespread with the exception of Verticillium wilt, which was localized in north central Wyoming; winter crown rot, found in one field in northeastern Wyoming; and root-knot nematode, which occurred only in sandy soils of west central Wyoming.

Many fields where alfalfa stem nematode and/or Phytophthora root rot were found had severe stand decline. Spring black stem was frequently associated with discolored crowns and upper roots and may also be involved in stand reduction. Losses due to "winter kill" in Wyoming have recently been associated with the stem nematode (2) and may be associated with other crown and root diseases.

Disease Detection Plots

Foliar diseases detected at the five irrigated sites included; spring black stem, downy mildew and common leaf spot. Spring black stem was found at all sites except Torrington and was most severe at Laramie and Afton. Downy mildew was found at all sites and was most severe at Laramie and Afton. Common leaf spot occurred at Riverton, Powell and Torrington, but was not detected in Laramie or Afton. High elevation and cool summers may explain the increased severity of spring black stem and downey mildew and the absence of common leaf spot at Laramie and Afton.

Yellow leaf blotch was the only disease of importance found at the dryland site (Sheridan) but was not detected at any of the irrigated sites.

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Verticillium Wilt of Alfalfa in Pennsylvania

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Verticillium wilt of alfalfa was detected in Pennsylvania for the first time in August 1981. It occurred in a 1978 seeding in Centre County. The fungus Verticillium albo-atrum Reinke and Berth was isolated and identified and its pathogenicity verified in controlled inoculations.

During August and September 1981, plants suspected of having Verticillium wilt were collected in Bedford, Bradford, Carbon, Centre, Crawford, Dauphin, Fayette, Jefferson, Lancaster, Luzerne, Mifflin, and Northampton Counties. Verticillium wilt was identified in three additional sites in Centre and one site in Mifflin Counties. In 1982, suspected plants were collected in Bedford, Bradford, Centre, Dauphin, Huntington, Jefferson, Juniata and Lancaster Counties. Verticillium wilt was detected at new sites only in Centre and Bradford Counties. Verticillium wilt has not been detected south of the central valleys in Pennsylvania. Additional surveys will be made throughout the 1982 growing season.

Controlled inoculations of red clover indicate that this species is probably a primary survival host for the pathogen. Individual leaves are killed, but heavily infected plants persist indefinitely in the greenhouse. Other research on Verticillium wilt at the U.S. Regional Pasture Research Laboratory include host range, insect vector, and mode of pathogenicity investigations.

Crown and Root Fungal Diseases in The Major Alfalfa Production Areas of Egypt

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A survey was made of crown rots, root rots, and wilts of alfalfa in several regions of Egypt. The incidence of these diseases, the fungi associated with diseased roots, and the pathogenicity of the fungi to alfalfa seedlings were determined. Incidence of disease was highest in southern Egypt in the main production areas of New Valley and Esna. Rhizoctonia solani and Fusarium spp. were the most common fungi isolated. No pathogenic fungi were isolated from plants growing in the high soil salinity area of Siwa Oasis in the western desert. Root diseases increased with age of stand, and rots were more common than wilts. The known alfalfa pathogens R. solani, Sclerotium bataticola, F. oxysporum, and F. semitectum were identified. Fusarium fusarioides, F. equiseti, and Mucor tax. spp. III were recorded for the first time on alfalfa anywhere, and F. acuminatum for the first time in Egypt.

Evaluation of Verticillium Wilt Resistance in Field Trials and Growth Chamber Tests

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Verticillium wilt of alfalfa, caused by Verticillium albo-atrum Rke. et Berth. (Vaa) was first identified in the Midwest in 1980. As part of a program undertaken to investigate epidemiology and control of the disease, alfalfa breeding lines and cultivars were evaluated for Verticillium resistance in field trials and in growth chamber tests. Material in the field was scored on the basis of foliage and root symptoms 4.5 mo after plants had been root-dipped in a spore suspension of Wisconsin Vaa isolates and transplanted into the field. Material remaining in the field 5 mo after transplanting was evaluated to determine the percentage of plants showing Verticillium wilt symptoms (% wilt). In May 1982, one year after transplanting, plants were counted to determine percent winterkill for each of the 52 experimental lines and cultivars. Inoculation in growth chamber tests was carried out according to the procedure described by Christen and Peaden (1), also using Wisconsin Vaa isolates. Material in the growth chamber test was evaluated twice, at 18 and 41 days after inoculation, on the basis of foliage symptoms. Linear regression was used to compare disease severity ratings from growth chamber and field studies. Correlation coefficients between growth chamber disease severity ratings (at both 18 and 41 days) and the four parameters measured in the field to assess cultivar performance are presented in Table 1, where the range of values given represents that observed for the two field locations. Correlation coefficients between the four parameters measured in the field are presented in Table 2.

Table 1. Ranges of correlation coefficients (r) obtained by correlating disease severity values from growth chamber tests with four different parameters of field performance at two locations

Field response	Growth chamber evaluations (18 days)	(41 days)
Foliage symptoms	.338* - .362*	.554** - .594**
Root symptoms	.477** - .511**	.601** - .713**
% wilt (October 1981)	.305* - .365*	.597** - .644**
% winterkill (May 1982)	.254* - .496**	.417** - .513**

* = significant r value at P = 0.05; ** = significant at P = 0.01

Table 2. Correlation coefficients (r) between several measurements of field performance. Range given for each combination represents that observed at the two locations.

	Foliage symptoms	Root symptoms	% wilt
Root symptoms	.668** - .694**		
% wilt	.675** - .847**	.609** - .726**	
% winterkill	.621** - .790**	.453** - .633**	.667** - .749**

Reference

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Yield Reduction and Stand Losses of Alfalfa Associated With Phytophthora Root Rot in Wyoming

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Phytophthora root rot (PRR), caused by *Phytophthora megasperma*, has been found in the major alfalfa producing areas of Wyoming. Until recently, loss of alfalfa in stands less than 4 years old had been attributed to winterkill or unknown factors. It has now been determined that such loss of stands is, in part, attributable to PRR. Crop rotation has been suggested as a means of control of PRR, but the most effective method has been the use of PRR resistant cultivars (1). The objective of this study was to evaluate stand loss and reduction in forage yield of 20 alfalfa cultivars seeded in an area naturally infested with *P. megasperma*.

Twenty alfalfa cultivars were seeded on April 29, 1981, in a field which had been planted in barley for the previous 3 years. By July, several plants displayed symptoms of PRR, which was later verified in the laboratory. Stand counts were made in the fall of 1981 and in the spring of 1982. Forage yields were also taken in the fall of 1982.

The eight cultivars with the greatest number of plants/m^2 in the fall were all known to be PRR resistant, except 'Ranger' which ranked seventh. In spring stand counts, two of eight cultivars with the greatest number of plants/m^2 were known to be susceptible to PRR. Of the cultivars that ranked in the bottom 12, only one was classified as PRR resistant. All the remaining cultivars have been classified as either PRR susceptible or had an unknown reaction to PRR. Stand counts in the fall ranged from 54 to 134 plants/m^2. By the following spring, counts ranged from 12 to 102 plants/m^2.

Yields of cultivars susceptible to PRR were greatly reduced, although there was no statistically significant difference between the top 16 cultivars according to Duncan's multiple range test. All cultivars susceptible to PRR had forage yields less than those known to be resistant to PRR.

Spring seeded alfalfa stand and forage yields can be severely reduced by PRR in the first year. The study will be continued for several years to evaluate the persistance of each cultivar in a *P. megasperma* infested area.

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Effect of Metalaxyl on Phytophthora Root Rot and Yield of Alfalfa

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Phytophthora root rot induced by Phytophthora megasperma f. sp. medicaginis is a serious, destructive disease of alfalfa (Medicago sativa L.) that limits production in most alfalfa-growing regions of the world. In the Piedmont region of North Carolina, seeding is usually done in the fall (August 15-September 25) to take advantage of late summer rains and to allow plants to become well-established before cold weather. Most alfalfa is grown in heavy, clay soils (Cecil Series) that have a high water-holding capacity during the fall, winter, and spring. During the growing season, they are considered well drained when evapotranspiration is high. Wet-soil favors growth of P. megasperma f. sp. medicaginis and Phytophthora root rot is most severe on saturated or poorly drained soils. Estimates of yield losses caused by soil-borne, root-rotting fungi might be biased when assessments are made on above-ground responses (such as yield or stunting) that do not coincide with root damage. The purpose of this study was to evaluate a method for sampling fields of alfalfa several times to determine when Phytophthora root rot develops and to relate Phytophthora root rot to yield. Cultivars susceptible to Phytophthora root rot were planted in 3 experiments at 2 locations (Wake and Rowan counties, NC). Plants in one experiment in Rowan Co. were inoculated with mycelium of Phytophthora megasperma f. sp. medicaginis; natural inoculum was also present in all plots. Metalaxyl (Ridomil) at 0, 0.58, 1.13, and 2.28 kg (a.i.)/ha was applied once in the fall at seeding, and once in the spring when regrowth began. Metalaxyl was selected because it controls diseases caused by species of Phytophthora in other crops. Five destructive samples were taken from each plot between 11/80 and 7/81 and evaluated for plant number and weight per 0.3 m of row and for root rot. Dry matter was harvested three times during the spring and summer of 1981.

In Rowan Co., metalaxyl treatment had no significant effect on seedling number and plant weight. Root rot in metalaxyl-treated plots was significantly lower in noninoculated plots in May and June and in inoculated plots in May. Yields were significantly greater in inoculated and noninoculated metalaxyl-treatment plots than in controls in June, but were similar in May and July. Root rot scores (averaged across metalaxyl treatments) on 4/1, 5/11, 6/17, and 7/29 were 4, 6, 56, and 66%, respectively, in the noninoculated plots and 3, 7, 44, and 53%, respectively, in the inoculated plots. Rainfall (24 cm) between 5/11 and 6/17 was about twice normal. In Wake Co., metalaxyl treatment had no significant effect on seedling number, plant weight, root rot, or dry matter yields. Root rot scores (averaged across metalaxyl treatments) on 3/26, 5/18, 6/24, and 8/24 were 3, 2, 3, and 3%, respectively. Rainfall (14 cm) between 5/18 and 6/24 was about 1/2 normal. In the absence of conditions (e.g. wet soils) favorable to development of Phytophthora root rot, metalaxyl treatment had no significant effect on seedling number, plant weight, root rot, or dry matter yields.

Inheritance of Fusarium Wilt Resistance in Alfalfa

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The inheritance of resistance to Fusarium wilt, caused by *Fusarium oxysporum* f. sp. *medicaginis*, was studied in two genetically unrelated alfalfa (*Medicago sativa* L.) germplasm sources.

Alfalfa clones were selected as being representatives of the different disease classes described by Frosheiser and Barnes (1). One set of six clones was selected from winter-hardy germplasm and another set was selected from non-dormant germplasm. Source of the winterhardy germplasm was 'MnFW-H' (2), produced by intercrossing resistant plants from 27 cultivars and germplasm pools. Parents for the non-hardy germplasm were selected from 'UC Cargo','CUF-101' and 'Moapa 69'. Three susceptible clones were selected from a two-clone synthetic (MnGN-1) derived from the cultivar 'Appalachee'.

Two sets of six-parent complete diallels with reciprocals and selfs were produced from each germplasm source. In addition, parents of each diallel were testcrossed onto susceptible clones and intercrossed with parents of the other diallel.

The progenies were grown in the greenhouse for 10 weeks, then immersed for 20 minutes in an inoculum containing 1.5×10^6 conidia/ml. The following day all plants were transplanted into the field. Each plot was split into two subplots, so that a cross and its reciprocal were always paired in two replications.

MnGN-1, 'Narragansett', 'Agate' and Moapa 69 were included as checks. The number of established plants was recorded 2 weeks after transplanting and used as the base population for each plot. Dead plants were recorded throughout the season and surviving plants were rated for disease severity 3 months after transplanting.

A qualitative genetic analysis was performed. It demonstrated that resistance to Fusarium wilt in both the winterhardy and the non-hardy germplasm sources was controlled by one dominant gene (FW_1) and by one incompletely dominant gene (FW_2). The FW_2 gene in the absence of FW_1 produced resistant plants when quadruplex, intermediately resistant plants when triplex and susceptible plants when duplex, simplex or nulliplex.

The inheritance patterns for Fusarium wilt resistance indicated in this study suggest that response to selection should be rapid, especially in germplasm sources carrying the dominant gene. Phenotypic recurrent selection should be effective at increasing Fusarium wilt resistance in most alfalfa populations.

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Factors Influencing Evaluation of Fusarium Wilt Resistance in Selected Alfalfa Populations in the Southeast USA

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Fusarium wilt of alfalfa caused by Fusarium oxysporum f. sp. medicaginis is implicated in reducing yield and persistence of alfalfa stands in the southeast USA. The present study was initiated to investigate isolate variability, effectiveness of inoculation techniques under field and greenhouse conditions, disease reactions of cultivars in geographically separated wilt nurseries, and the effect of soil water matric potential on disease severity.

Isolates of Fusarium oxysporum f. sp. medicaginis from Minnesota, Pennsylvania, Maryland, and North Carolina did not differ in growth in response to temperature or in virulence. Conidia of four isolates from North Carolina were combined to inoculate resistant (Moapa, Liberty, and NCMP-2) and susceptible (Apalachee and Narragansett) alfalfa in the field and in greenhouse tests. Field and greenhouse results were highly correlated ($P=0.01$). Wilt resistance in 10 cultivars and 4 breeding lines adapted to the southeast USA was evaluated at two locations. Entries with the highest level of wilt resistance were germplasms NCMP 9, NCMP 11, and NCMP 13 and cultivars Cimarron and Saranac AR; Apalachee was highly wilt susceptible and the remaining entries were moderately wilt resistant. The disease reaction of entries common to wilt nurseries in North Carolina and Minnesota were similar, although disease scores were higher in North Carolina than Minnesota. This is probably because plants remained in the field in North Carolina about 2 months longer than in Minnesota and because soil temperatures were generally warmer in North Carolina.

Following inoculation with F. oxysporum f. sp. medicaginis, 2- to 3-month-old seedlings of alfalfa cultivars resistant (Moapa) and susceptible (Narragansett) to Fusarium wilt were replanted in the greenhouse or outdoors and grown at different soil moisture regimes. Each soil moisture treatment was defined as a range of soil matric potential. Tensiometers or gravimetric techniques were used to monitor moisture extraction and regulate soil irrigations. Yields of noninoculated Moapa and Narragansett plants, as well as inoculated Moapa plants, increased with increased soil moisture. Yield of inoculated Narragansett plants was reduced in comparison with noninoculated plants and there was no increase in yield in response to increased soil moisture. Severity of Fusarium wilt for both cultivars did not increase by increased soil moisture in greenhouse experiments. A significant increase in mortality was observed for Narragansett plants in the first 4 weeks following inoculation in an outdoor experiment. Afterwards, this difference was no longer observed. In all experiments, the basic characterization of Moapa and Narragansett as populations resistant and susceptible, respectively to Fusarium wilt was never altered. Although increased soil moisture may have an effect on the initial rate of disease development, there was no evidence that it altered the basic characterization of Moapa and Narragansett as cultivars resistant and susceptible, respectively, to Fusarium wilt.

Effects of Inoculum Concentration and Temperature on Anthracnose Severity in Alfalfa

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Soon after Flemish alfalfa (*Medicago sativa* L.) germplasm was introduced into the United States, it was found to be highly susceptible to anthracnose caused by *Colletotrichum trifolii* (5). Control of the disease was achieved by developing resistant cultivars (1, 2), but resistance was not lasting and within 10 yrs, pathogenic races had developed (3, 6). The new race capable of inducing disease in previously anthracnose-resistant cultivar Arc was designated race 2 (4). The growth rate of race 1 (isolate PA) and race 2 (isolate NC 4) between 4 and 36 C in culture is similar (7). However, the interaction among temperatures, races, and cultivars on anthracnose severity has not been studied. Since four germplasms are available that differ only by their selection for resistance to race 1 (1), this study was done to evaluate how inoculum concentrations and temperature influence anthracnose severity.

Conidia of race 1 or 2 of *C. trifolii* at concentrations of 1×10^3, 1×10^4, 1×10^5, or 1×10^6 conidia/ml were used to inoculate 3-week-old seedlings of 12 cultivars of alfalfa susceptible or resistant to anthracnose. Disease developed in susceptible cultivars inoculated with all concentrations of conidia and the percentage of plants scored 3, 4, or 5 (1-5 scale) increased sigmoidally as inoculum increased; the asymptote was generally reached at 1×10^5. The concentration where 50% of the seedlings were scored 3, 4, or 5 was between 1×10^3 and 1×10^4 conidia/ml. The disease curves for race 1 and 2 were similar for the four concentrations of conidia.

Saranac, Saranac AN 4, Vernal, Vernal AN 4, Glacier, Glacier AN 4, Team, and Arc were inoculated with 1×10^6 conidia/ml of race 1 or 2 and incubated at 12, 16, 20, or 24 C during infection. Disease was generally more severe in seedlings i) inoculated with race 2 than race 1, ii) incubated at the highest temperatures; and iii) in cultivars not previously selected for resistance to *C. trifolii*. Susceptibility to race 1 was temperature dependent. Among cultivars inoculated with race 2, susceptibility was temperature independent and resistance was temperature dependent. There was an interaction between temperatures and cultivars for race 2. Vernal AN 4 was resistant to race 2 at 12 C, moderately resistant at 16 C and susceptible at 20 or 24 C. Saranac AN 4 was more resistant to race 2 than Glacier AN 4 at 16 C. At 12, 20, and 24 C, the level of resistance to race 2 in Saranac AN 4 and Glacier AN 4 was similar.

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Breeding for Resistance to the Lesion Nematode (*Pratylenchus*) in alfalfa

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During the last 20 years it has become increasingly difficult to establish and maintain alfalfa at the North Central Experiment Station in Grand Rapids, MN. Seedling roots developed necrotic lesions followed by destruction of the secondary roots. Root injury was followed by stunting of the vegetative growth and subsequent plant death. Sheaffer et al. demonstrated that the unthrifty appearance of the plants was associated with high populations of lesion nematodes. Nematicide application significantly increased yield and stand persistence of alfalfa. Griffin reported that the most important root-lesion nematode associated with alfalfa was *Pratylenchus penetrans*, but at least six other *Pratylenchus* species can cause injury. Above-ground symptoms of root-lesion nematode injury resemble those caused by other plant pathogens causing plant stunting and characterized by small leaf size and shortened internodes.

A breeding program was initiated to determine if resistant germplasm could be developed. Plants (clones) varying in plant vigor and appearance were selected from a two-year-old cultivar trial having a high nematode population (700 to 800 nematodes/58 cm^3 of soil). These selections, their S_1 and half-sib progeny were evaluated for resistance both in field and laboratory tests. Significant variability was observed among clones for top weight, root weight, fibrous root score, and numbers of nematodes in the root. Evaluation methods in the growth chamber and the field rated clones similarly ($r=.72$, 18 d.f.) for root-lesion nematode resistance. Root fresh weight appeared to be the best selection criterion for laboratory evaluations and top dry weight was the best selection characteristics for field evaluations. We believe it should be possible to develop cultivars with resistance to the root-lesion nematode.

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Resistance to Race 1: A Prerequisite for Induced Resistance to Race 2, by Race 1 of *Colletotrichum trifolii*, on Alfalfa

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Approximately 80% of Arc alfalfa seedlings are resistant to race 1 of *Colletotrichum trifolii*. However, Arc is very susceptible to race 2 of this fungus. Less than 1% resistant plants can be found in any Arc population. As reported earlier a majority of Arc seedlings can be temporarily "immunized" from a lethal infection of race 2, by a "protective" inoculation with race 1. In one of the "protection" experiments, we transplanted surviving race 2 "protected" Arc seedlings, singly, to 10 cm plastic pots. After recovery, 162 plants were challenged with race 1 via needle inoculation. One hundred and sixty-one (99.3%) were found to be resistant to race 1. Plants were cut back, allowed to recover, and similarly inoculated with race 2. One hundred fifty-one of 156 of the remaining race 2 "protected" plants were found to be susceptible to race 2. In an experiment in which protection to race 2 was measured in Arc, 'Saranac AR', 'Team', 'Riley', and 'Baker', disease reactions of the "protected" survivors were as follows: In Arc, 100% of the survivors were resistant to race 1, 95% were susceptible to race 2; in Saranac AR, 100% of the survivors were resistant to race 1, 58% were susceptible to race 2; in Team 95% of the survivors were resistant to race 1, 67% were susceptible to race 2, however, in Riley, only 42% of the survivors were resistant to race 1, and of these 43% were susceptible to race 2; and in Baker, 53% of the seedlings were resistant to race 1, of these 40% were susceptible to race 2. From these data, we suggest that resistance to race 1 is a prerequisite to protection from race 2 in Arc, Saranac AR and Team, but not necessarily in Riley and Baker.

Utah Alfalfa Variety Testing Results -- Quality and Quantity

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Alfalfa variety trials have been conducted in Utah for several years. Several varieties have performed very well in the high mountain valleys of Utah as well as the desert regins. The top variety as per yield in Utah is Deseret, a USDA release from the Crops Laboratory located in Logan, Utah. Other varieties which have done very well are: Anchor, WL-309, WL-312, Vangard, and Thor. In relation to the yield trials, some quality work was initiated in which quality was evaluated for the to five alfalfa varieties. The quality analysis was run for these varieties two weeks prior to harvest, at harvest, and two weeks after harvest using infrared analysis. Interest in Utah is very keen for quality results. Dairy farmers as well as alfalfa hay brokers are interested in these trial results.

Response of Alfalfa to Soil Water Deficits

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Although many studies have reported yield increases due to alfalfa irrigation when soil water has been limited, little information is available on alfalfa plant and soil water relationships which might be used to improve water management. Our objective was to determine effects of soil-induced plant water deficits on seasonal growth patterns, forage quality, forage yield, water use, and water-use efficiency of alfalfa.

During 1981, established alfalfa grown on a sandy soil was subjected to four water supply levels designated: H (high irrigation), MH (medium high irrigation), ML (medium low irrigation), and U (unirrigated, rainfall only). Soil water depletions occurred for all treatments to at least 1.90-m depths. Alfalfa in H and MH treatments maintained midday plant water potentials (ψ_{mp}) at -7 to -13 bars throughout the season. For alfalfa subjected to ML and U treatments, ψ_{mp} reached -27.5 and -40 bars, respectively.

Dry weight (DW) accumulations for alfalfa in H and MH treatments increased with successive harvests during the summer while harvest DW's for alfalfa in ML and U treatments declined. Relative growth rate (RGR) declined sharply as ψ_{mp} decreased. At moderate plant water stress (ψ_{mp} of -15 to -20 bars), little growth occurred and under severe water stress RGR's were negative due to leaf loss. Reduced soil water availablilty resulted in increased leaf:stem weight ratios and *in vitro* dry matter disappearance (IVDMD), but only under severe and long-term plant water stress. Crude protein (CP) concentration was unaffected by plant water status.

Water use rates, after canopy closure at a leaf area index (LAI) of 1.5 for well-watered alfalfa ranged from 5.3 to 10.0 mm/day during the summer. Despite moderate (MH) and severe (U) plant water stress, alfalfa used water at an average daily rate of 6.9 and 2.6 mm, respectively.

It appeared that irrigation water could be used effeciently on coarse-textured soils by moderate water application to alfalfa at 50% depletion of extractable soil water, an apparent threshold for maintenance of favorable plant water status. A close association between forage yield of individual harvests and cumulative ψ_{mp} indicates potential for initiation of irrigations when plant measurements indicate soil water depletions to a threshold level.

Effects of Rhizo-Kote Seed Coating on Alfalfa Growth and Yield in Minnesota and Central California

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The long term success of an alfalfa field is dependent on many limiting factors. Among these are seedbed preparation, management practices, weed control, nodulation and soil fertility, and weather. Competition among plants develops if any one of the factors affecting growth becomes limiting. These limiting factors can affect stand establishment, plant growth and ultimately yield through a series of complex growth and development process interactions. The yield of alfalfa in a field community is affected by the level of interspecific competition with weeds. Intraspecific competition between alfalfa plants will also affect yield through effects on the yield components of the alfalfa population, namely plant numbers, plant size and the number and size of stems and leaves.

Three different field studies were established during 1980 to examine some of the plant population parameters affecting yield and the response of yield and yield components to Rhizo-Kote, a commercial lime based coating containing rhizobia bacteria, compared to moist-inoculated seed. One study in the spring of 1980 was established in South-eastern Minnesota using typical grower practices. Two small-plot studies at several planting rates were established on the CelPril Industries research farm in Manteca, California, one in the spring of 1980, another in the fall of 1980. Parameters measured included planting rate, plant counts at 3 weeks and 18 months, plant size, stem counts, alfalfa yield, and in Minnesota, weed yield.

In Minnesota, under grower field conditions, second year alfalfa yields averaged approximately 1.5 tons per acre alfalfa and 1.6 tons weeds, for two cuttings. In California under field plot conditions second year alfalfa yields averaged 11.2 tons per acre total for 6 cuttings with no weeds. Plant counts at 3 weeks and 18 months for comparible planting rates were lower in Minnesota than in California, indicating lower emergence and survival rates.

Alfalfa yields were significantly higher for the Rhizo-Kote treatments in Minnesota but in the California research plots no yield differences were seen between either the treatments or the planting rates.

Field seed emergence percentages in California were seen to be related to the number of seeds planted. As planting rate increased the percent seed emergence decreased. No difference was seen between treatments at high planting rates, but an increase in percent emergence was seen for the Rhizo-Kote treatment at low planting rates. Overall emergence rates were lower in Minnesota than in California. Seedling survival at 18 months was not seen to be related to planting rate.

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Effects of Three Management Regimes on Seed Yield of Three Types of Alfalfa Cultivars

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Time of clipping has been observed to influence subsequent blooming and seed set patterns for certain alfalfa (*Medicago sativa* L.) lines. The purpose of this study was to determine the effects of three management regimes on seed yield of three types of alfalfa.

Management regimes (whole plots) of no clip, early clip (10 May 1977 and 20 May 1978), and late clip (20 May 1977 and 2 June 1978) were imposed upon the following alfalfa cultivar types (sub-plots) in 1977 and 1978; Flemish -- moderately winterhardy ('Saranac' and 'DuPuits'), winterhardy ('Agate' and 'Vernal'), and pasture -- very winterhardy ('Drylander' and 'Travois'). Plots were seeded in 1976 at a rate of 2.2 kg/ha of seed per acre and harvested in 1977 and 1978. Plots consisted of two 3m rows 46 cm apart on beds 91 cm wide for rill irrigation as needed. Normal cultural practices were employed with leafcutter bees used as pollinators. At harvest, seed plots were mowed and air dried prior to threshing and seed cleaning in the laboratory. Data were analyzed for the split-split plot design.

Highest seed yields of all cultivars were obtained in both years when the plots were not clipped. The 1977-78 average yields for no clip, early clip, and late clip were 768, 465, and 552 hg/ha, respectively. Interactions of cultivar types and cultivars within types with managements were not significant ($P=0.05$). Flemish cultivars were highest yielding and pasture types were the lowest yielding each year. Cultivars within types did not differ significantly in seed yield. The 1977-78 average yields for Saranac, DuPuits, Agate, Vernal, Drylander, and Travois were respectively 682, 695, 616, 641, 450, and 487 kg/ha. The results indicated that clipping decreases seed yields and that cultivar types can be managed uniformly.

Efficient Hybridization of *Medicago dzhawakhetica* and *M. sativa* Using a *M. sativa* Mutant Lacking Post-Meiotic Cytokinesis

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Plant materials used in this study included one accession of *M. dzhawakhetica* (UAG 98) from Lesin's collection at the University of Alberta in Edmonton, three clones of *M. falcata*, and several ploidy levels and mutants of *M. sativa*. Diploid, tetraploid, hexaploid and octoploid *M. sativa* clones were used. At the diploid level, normal clones of CADL, clones homozygous for *rp*, and clones homozygous for *jp* were used. The *rp* gene conditions 2n pollen formation in alfalfa (McCoy, 1982), and the *jp* gene results in 4n pollen due to the lack of a post-meiotic cytokinesis. Plants homozygous for *jp* are effectively male-sterile, and were only used as female parents. All combinations of crosses between the various *M. sativa* clones and the *M. dzhawakhetica* accession were made.

The results indicate 2x *M. sativa* clones homozygous for *jp* are very effective in producing interspecific hybrids with 4x *M. dzhawakhetica*. 2x *jpjp* clones crossed with 4x *M. dzhawakhetica* resulted in 2.41 seeds/pollination. In comparison, crosses between 4x, 6x or 8x *M. sativa*, and 4x *M. dzhawakhetica* resulted in zero seed production. The only hybrids were produced from 2x *M. sativa rprp* clones x 4x *M. dzhawakhetica* (0.02 seeds/pollination). 2x *M. falcata* x 4x *M. dzhawakhetica* (0.02 seeds/pollination), and 4x *dzhawakhetica* x 2x *M. falcata* (0.01 seeds/pollination). The very efficient hybridization with *jpjp* clones may be due to some abnormality in female gametophyte development that results in an acceptable embryo: endosperm balance not produced in normal clones. Lesins (1961) also found that uneven ploidy levels were necessary for successful hybridization, however his crosses resulted in only 0.35 seeds/pollination.

Surprisingly, almost all hybrids were triploid (2n = 3x = 24), with one *M. sativa* genome and two *M. dzhawakhetica* genomes. Root-tip chromosome counts were made on 216 F_1 hybrids, and 211 were triploid, three had 23 chromosomes, and two had 25 chromosomes. Chromosome pairing was studied in nine F_1 hybrids. Trivalent frequency ranges from 0 trivalents per cell in one plant to an average of 1.9 trivalents per cell in another plant. This indicates that some plants have the potential for genetic exchange between genomes.

Morphologically, the F_1 hybrids are prostrate similar to the *M. dzhawakhetica* parent. Flower size is intermediate, although 4x *M. dzhawakhetica* flowers are smaller than 2x *M. sativa* flowers. Most F_1 hybrids are extremely weak, although one *M. sativa jpjp* clone produced hybrids more vigorous than the *M. dzhawakhetica* parent. The F_1 hybrids are completely male-sterile. Female fertility in crosses with normal CADL clones and *rprp* clones is very low (0.05 seeds/pollination).

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Selection of Alfalfa Cell Lines Resistant to Culture Filtrates of *Fusarium oxysporum*

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Four germplasms were utilized to select alfalfa cell lines resistant to a toxic component(s) of *Fusarium oxysporum* culture filtrates. The 4 germplasms were Moapa 69 (80% resistant plants), Narragansett (less than 20% resistant plants), Regen S (developed for regeneration potential from tissue culture, less than 20% resistant plants), and I 13 (an experimental *Fusarium* resistant line developed in Hungary).

Callus cultures established from cotyledon sections of Moapa 69 (10 cultures), Narragansett (10 cultures), and I 13 (7 cultures) were selected for their ability to proliferate as callus on Shenk and Hildebrandt (SHDN) medium (with kinetin and 2,4-D). Four cultures of Regen S were selected for their capacity to regenerate plants from tissue culture. Plants corresponding to each of the tissue cultures were propagated as stem cuttings to test each clone's response to the pathogen and to fungal culture filtrates.

The callus culture selection experiments involved the replacement of 20% of the water in SHDN media with *Fusarium* culture filtrate. Susceptible cell lines are killed when transferred to this media.

By selecting resistant colonies and increasing the percentage of filtrate in the medium, we have been successful in selecting a resistant cell line that is capable of growth on 100% culture filtrate medium. Plants have not yet been regenerated from this cell line. In addition to the selection of resistant lines starting from susceptible material, screening of cell lines from plants which are resistant to the pathogen has produced interesting results.

All 10 clones of Narragansett are susceptible *in vitro* to the filtrate and *in vivo* to the pathogen. Eight of the 10 Moapa 69 clones were susceptible to the filtrate. The 2 *in vitro* resistant clones were the only *in vivo* resistant clones in the Moapa 69 selections. The I 13 clones were all resistant to the pathogen, but only 4 were resistant to the *Fusarium* culture filtrate.

The toxic component(s) in the *Fusarium* culture filtrates are heat stable, nonproteinaceous, and soluble only in polar solvents. Thin-layer chromatography and callus culture assay has shown the toxic components to be free of fusaric and fusidic acids. The toxin(s) are biologically active in very small concentrations.

An *in vitro* alfalfa cell line selection approach coupled with toxin elucidation and classical breeding procedures may allow a more precise identification of the genotypes and the resistance mechanisms in *Fusarium* resistant plants.

Variation in Protoclones Regenerated from Nonmutagenized Calli of Two Regen S Alfalfa Selections

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Reports of alfalfa plant regeneration from protoplasts (2,3,4,5) make little mention of phenotypic or genotypic variation in the regenerates. Yet variation following regeneration from nonmutagenized cell cultures is common in many species. We find that variation may be frequent in nonmutagenized alfalfa protoclones.

Protoclones were regenerated from two selections (RS-K1 and RS-K2) of Regen S alfalfa (1) as previously described (3). Four single ramet replications of each regenerate were field-planted in a randomized complete block design in the spring of 1981. Ramets of both parents also were included. Twenty-one and 61 protoclones of RS-K1 and RS-K2, respectively, had either 3 or 4 surviving ramets in late fall. Plants were rated in mid-April of 1982 for winter damage (1 = no damage, 5 = plant dead). RS-K1 and RS-K2 had scores of 1.7 and 1.2, respectively, while 43 and 20% of their protoclones were rated at 3.0 or higher. Plants were harvested in mid-May for forage dry weight determination. Two of 12 RS-K1 and none of 51 RS-K2 protoclones had greater ($P = 0.05$) mean dry weights than their parents when clones with 3 or more replicates were compared. None of the RS-K1 and 24 of the RS-K2 protoclones were significantly inferior to their parents. Numbers of inferior protoclones were actually underestimated because of winterkill. Numerous variations, including changes in plant height and leaf shape, have been observed. Cytological studies of protoclones revealed changes in karyotype, including aneuploids, translocations, and increased ploidy.

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Collecting Alfalfa Germplasm in Turkey

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Scientists are in general agreement that alfalfa Medicago sativa L., most likely originated in the area of Asia-Minor, Iran, Turkey, and Northern Afghanistan. It is also recognized that the alfalfa weevil has existed in Asia-Minor as long as alfalfa has existed. Therefore, to provide the extremes of germplasm variability for plant breeding and entomological research (in particular, alfalfa weevil resistance research) collection of alfalfa in that part of the world is of great importance.

In 1981, we participated in a collection trip to Turkey which was organized through the Aegean Regional Agricultural Research Institute at Izmir. We were accompanied on the expedition by Dr. Ernest Small, taxonomist, Agriculture Canada, Biosystematics Research Institute, Ottawa and a Turkish scientist, a senior technician, and a driver for the vehicle from Izmir. The collection took place between July 15 and August 15. The party was transported by a 4-wheel drive Chevrolet carryall approximately 4,300 miles during the exploration and traveled throughout most of eastern Turkey.

In total 189 Medicago and other seed samples were collected; 14 Rhizobium collections; approximately 300 hebarium samples (collected by Dr. Small); and numerous Medicago insect samples (collected by Dr. Ratcliffe). The 189 seed samples consisted of 131 perennial Medicago single plant and population (cultivated and wild) collections, twelve annual Medicagos, 34 Lotus species and 12 Onobrychis, Trigonella, and other species. The insect collections included 33 live adult alfalfa weevils which are being held in quarantine at BARC by Dr. Ratcliffe.

Great diversity in the types of Medicago sativa subsp. sativa was seen, particularly in flower color, pod coiling, flower and pod size, and growth habit characteristics. A number of diploid types were collected. Collections of Medicago sativa subsp. falcata, M. marina and M. papillosa were also made. Twenty-eight "native populations" (primitive cultivars) of cultivated M. sativa subsp. sativa were collected from village farmers. Apparently, alfalfa first became popular as a cultivated crop about the time of the Ottoman Empire (1300-1500 AD) when the soldiers traveling throughout Turkey began to rely on the alfalfa hay grown in the central province of Kayseri for their horses. All of the cultivated alfalfas in Turkey are purple flowered, coiled pod types.

Many of the irrigated alfalfa fields which we saw in Kayseri province had severe infestations of stem nematodes. Previous U.S. introductions from this part of Turkey have demonstrated low to moderate levels of stem nematode resistance. This resistance has perhaps resulted from many years of natural selection in the Kayseri area. There was little evidence of other foliar, root or crown diseases. The climate in eastern Turkey is generally warm and dry during the growing season. Most of the cultivated alfalfa is grown under irrigation. Dodder was a severe weed problem in many of the alfalfa fields throughout eastern Turkey. Ninety-five percent of the alfalfa is harvested by hand and stored as loose hay or hand-rolled bundles in stacks for winter feeding.

The alfalfa weevil was recognized as a damaging insect by most farmers in eastern Turkey. Damage was occasionally severe enough on the first crop to require some chemical control, sporadically available through the government extension office. The farmers did not know of any type of alfalfa that was resistant to the weevil. No other insect problem was recognized by farmers, and their alfalfa fields were generally free of damage. However, pea aphid (both green and red forms) and several species of snout beetles (probably Sitona) were frequently found in fields of Eastern Turkey.

The soils throughout eastern Turkey are believed to be near pH 7. Farmers had essentially no knowledge of the presence of the rhizobium nodule on the alfalfa roots. No one used inoculum when planting seeds and were generally unaware that nitrogen was fixed from the air by the nodules on the roots. Some farmers applied fertilizer or manure to their alfalfa fields but most saw little response from its application.

Because of the high elevations in Eastern Turkey we had orignally expected to find an alpine climate with scattered trees and evergreens covering the land. In fact, there were essentially no trees on the hillsides and mountainsides, even at elevations in excess of 2,000 meters. However, perennial Medicagos were found easily in most locations in the east and northeast of Turkey.

Most notable on the collection trip was the decimation of the countryside. Literally all vegetation was grazed to the ground by sheep, goats and cattle (primarily sheep), making it very difficult to find flowering Medicagos. We found the best collection sites to be on steep road banks and in and around villages where there are rock walls and other obstructions keeping animals from reaching them.

All of the people that we met were quite courteous, cooperative, and most willing to be helpful to us.

Description of Several Variants Observed in Plant Introductions Being Increased at Reno, Nevada

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Visual observations and cytological studies of pollen of a number of plant introductions being increased at Reno, Nevada have identified several variants. A virescent mutant was identified in P.I. 247789, a tetraploid germplasm from Peru. Unlike the virescent mutant previously described by Stanford (1969) this mutant is not temperature sensitive. Plants were grown in the field, greenhouse and three growth chamber environments (constant 10°C, constant 20°C, and constant 30°C). Expression of the trait was consistent in all environments. Inheritance data indicated the virescent trait is controlled by a single recessive gene, designated *vr*.

In P.I. 258752, a diploid *M. falcata* germplasm from the USSR, we identified a new male-sterile mutant. Cytological studies of male-sterile plants indicated breakdown occurs immediately after pachytene. Preliminary inheritance data indicates male sterility is controlled by a single recessive gene.

In P.I. 325381, a diploid *M. sativa* (syn. *M. coerulea*) germplasm from the USSR, we identified one plant that produced jumbo pollen (54-64μm) instead of normal pollen (30-39μm). Cytological examination indicated that meiosis was normal through telophase II. However, after meiosis there is no post-meiotic cytokinesis. Inheritance studies indicated that jumbo pollen formation is controlled by a single recessive gene that is allelic to a previously identified mutant in CADL.

A minimum of 50 plants of each of the following diploid P.I.'s was screened for 2n pollen frequency: 179370, 251690, 258752, 314093, 315460, 315465, 315471, 315479, 315481, 325381, 325382, 325407. Interestingly most P.I.'s had a few (range 0 to 8) plants with a descernible level of 2n pollen (1 to 5%). In addition, P.I.'s 315460 and 325382 had one plant each with a high level of 2n pollen (greater than 15%). Allelism tests with the *rp* gene that conditions 2n pollen formation in CADL (McCoy, 1982) are being conducted. In addition, analysis of F_1 hybrids between *M. coerulea* P.I.'s and *M. falcata* P.I.'s demonstrated an increased frequency of plants with a discernible level of 2n pollen. A minimum of 50 plants of each of nine hybrid families was examined. All F_1 families produced several (range 3 to 16) plants with 1% or greater 2n pollen.

Two P.I.'s (P.I. 244317, a tetraploid germplasm from Spain, and P.I. 304525, a tetraploid germplasm from Turkey) are sensitive to the pesticide DIBROM. Application of DIBROM at normal rates resulted in die-back of the above-ground portion of the plants. Four to six weeks was required for regrowth to commence. An inheritance study of DIBROM sensitivity is being conducted.

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The Importance of Leaf Frost Resistance to the Winter Survival of Seedling Stands of Alfalfa in Northwestern Canada

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Alfalfa's ability to survive winter depends in part upon its capacity to synthesize, translocate and store food reserves in the roots and crowns during the fall. To survive in northern latitudes the plant must synchronize its development sufficiently early in the fall to accumulate food reserves prior to the first "killing frost". Although survival does not depend upon overwintering leaves, the cold resistance of the leaves may govern the length of time that food reserves are translocated to the crown and root in the fall. The objective of this study was to employ controlled freezing facilities in the field to define the influence of frost on the leaves of four alfalfa selections in the fall and on plant survival the following spring.

Plants propagated from single plant selections of the cultivars Saranac and Luna (*Medicago sativa* L.), Beaver (*M. media. Pers*) and Anik (*M. falcata*. L), were transplanted into the field in May and subjected to a range of freezing temperatures at three week intervals during August and September.

Leaves on all plants had the capacity to harden during the fall but the selection from Anik was consistently the most frost hardy. In mid-August, a -6°C temperature caused 50% leaf injury in the Anik selection but in late September, -12.5°C caused less than 35% injury. Temperatures ranging from -4 to -5°C caused 50% leaf injury to the selections of Beaver, Saranac and Luna in mid-August while in late September temperatures ranging from -9.5 to -10.5°C were required to produce similar amounts of injury. Thus, the Anik selection appeared to start hardening about 3 weeks earlier than all other selections.

The potential for winter injury was very high in the spring as a result of leaf-damage in mid-August. This effect diminished as the date of the first fall frost was delayed from early to late September.

Long term frost records at Beaverlodge, Alberta (55.12°N lat., 11.16°W long) indicate that because of the ability of alfalfa leaves to harden, early fall frosts are not of sufficient intensity to have a major effect on leaf injury and subsequently the winter survival of seedling stands of alfalfa at this location.

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History of Germplasm Involvement by the National Alfalfa Improvement Conference

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While the first conference met in 1934, it was in 1948 that the objectives of the conference were reviewed and stated by H. M. Tysdal as follows: "Through a cooperative integrated program to exchange information and materials with the objectives first to develop, test, and establish, and second to preserve and to make available on a national basis superior alfalfas for the maintenance of a permanent agriculture and for the betterment of the seed, forage, and livestock industries of this and other countries". In a broad sense, nearly all efforts by the National Alfalfa Improvement Conference (NAIC) have involved germplasm.

The need for multiple pest resistance, forage and seed yield, and persistence in alfalfa cultivars was discussed at the first conference in 1934. Alfalfa germplasm exchange among investigators, seed production of experimental strains in the west for return to the originator, the need for fundamental work, and voluntary contributions to alfalfa conferences and reports were discussed in relation to breeding "ideal" alfalfa cultivars. Proposals were made at the second conference to develop a system of listing strains and seed available with certain characteristics; to standardize certain types of tests and note taking procedures; and to appoint a secretary to keep people informed, list available seed, and to issue an annual report. Much of the germplasm involvement since then originated and centered around committee activities.

Committees on the Seed Increase of Experimental Strains were continually faced with poor and unstable seed yields in the midwest and east and searched for cooperative seed production in the irrigated west. The NAIC appealed for help directly to the Secretary of Agriculture and received funds in 1936. Presentations and discussions at several alfalfa conferences and joint committee meetings with the International Crop Improvement Association (later called AOSCA, Association of Official Seed Certifying Agencies) led to the development of regulations for producing certified seed of 'Ranger' outside of its area of adaptation about 1946. Standards developed for Ranger seed production became the basis for current methods. Many discussions on the need for reliable stock seed production of small-seeded legumes and grasses resulted in the National Foundation Seed Project that was organized in 1949. The NC-83 project, "Seed production of breeding lines of insect-pollinated legumes", initiated in 1965, with an objective on seed increase of experimentals, resulted from alfalfa conference activities.

Committees on Variety Testing initially discussed methods of testing strains and later discussed testing cultivars from public and private programs. Numbers of stations, years, and cuttings, generation of synthesis, harvest management, data obtained in the year of seeding, stand estimates, and other matters were considered without setting standards. The use of standard forage yield check varieties was initiated by the Central Alfalfa Improvement Conference (CAIC) in 1963 and the Eastern and Western Conferences adopted the practice. "Uniform Nurseries" to test experimental strains and cultivars were organized through USDA leadership at Beltsville, MD, and conducted from 1937 to 1954. Similar tests were regionalized in 1955 and reported by regional conferences. Experimental strains of various origin were assigned "A" numbers and clones exchanged among workers were assigned "C" (conference) numbers by conference secretaries through the mid-1960's.

The Committee on Release of New Cultivars functioned from 1952 to 1960 in an advisory capacity to evaluate data and to recommend release of cultivars developed in public programs. However, some public cultivars had been released without consideration by the committee. With the advent of a number of private alfalfa breeding programs about 1960, the role of the committee was questioned, and the committee was terminated.

The Committee on Cultivar Descriptions listed cultivars with published descriptions in conference reports from 1964 to 1976. The conference decided in 1976 that descriptions of cultivars that received favorable review by the National Certified Alfalfa Variety Review Board (NCAVRB) would be sent to the NAIC members annually.

Committees on Genetics, Breeding, Gene, Chromosome and other Nomenclature have functioned for 30 years. Breeding and genetic terms were defined. Note-taking procedures were developed for plant characteristics, disease, and insect resistance. Standard descriptions were made for growth stages. The principles of gene nomenclature elaborated by the International Committee on Genetic Symbols and Nomenclature at the 10th International Congress of Genetics were considered adequate for alfalfa. A chromosome numbering system and a key to the identification of the chromosomes in diploid Medicago sativa L. were made. Geneticists were requested to send genetic stocks to the National Seed Storage Laboratory. Nomenclature was considered for insect biotypes and physiological races of pathogens in 1972. The assignment of three-letter symbols for traits was suggested to assure long-term computer use. An Illustrated Summary of Genetic Traits in Tetraploid and Diploid Alfalfa was published in 1967 (D. K. Barnes and C. H. Hanson, USDA Tech. Bull. 1370) and 26 added references were listed in the 1976 Conference Report.

The Committee on Listing Available Breeding Lines has been active for about 20 years. Officially released breeding materials have been listed in conference reports. "Improved Breeding Lines of Alfalfa" 1978, USDA, ARM-W-5 by O. J. Hunt et al. listed lines released from 1965-1978. Indices listed the lines by origin (state, federal, private industry, and Canada) and use (botanical, genetic, pest resistance, etc.).

Committees on Screening Breeding Materials for Pest Resistance have been active since 1952 when initial screening procedures were described. The need for genetic diversity in pest resistance has been recognized for at least 25 years. The use of standard checks for disease and insect resistance was proposed by the CAIC in 1971 and endorsed by the NAIC in 1972.

The Committee on Standard Tests for Characterizing Disease and Insect Resistance of Alfalfa Cultivars has been active since 1972 when a report on screening procedures, standard evaluation methods, and standard checks was issued. The report became the basis for "Standard Tests to Characterize Pest

Resistance in Alfalfa Varieties", 1974, ARC-NC-19 by D. K. Barnes et al. The use of this publication resulted in unifying descriptions of pest resistance in cultivars and germplasm releases. ARC-NC-19 (revised) with 18 coauthors is scheduled for publication in the fall of 1982. The publication lists persons and locations that can provide information about cultivar evaluation tests for specific types of pest resistance. Seed of the 17 check cultivars recommended in standard tests is available from J. H. Elgin Jr., USDA/ARS at Beltsville, MD.

The National Certified Alfalfa Variety Board which met for the first time in January 1962 was the result of meetings by the alfalfa improvement conference, the American Seed Trade Association and the International Crop Improvement Association (AOSCA). The board consists of five members, one from AOSCA, one plant breeder from the ASTA, one member at large from the ASTA, one public plant breeder from the alfalfa improvement conference, and one plant breeder from the USDA/ARS. The board reviews and evaluates information on new alfalfa cultivars presented by public and private breeders and expresses an opinion on whether a new variety merits certification. The board reports to AOSCA those varieties that are distinctive and merit certification. Varieties that do not receive a favorable review are reported only to the secretary of AOSCA and the originator of the variety. The NAIC secretary sends a copy of the AOSCA report to members.

The Committee on Alfalfa Variety Certification has functioned continuously since 1966. Committee recommendations stimulated research that resulted in reducing the isolation distance for producing the certified seed class in fields larger than 2 ha (5 acres). Another study supported the adequacy of the 0.4 km (1320 feet) isolation for foundation seed production. In 1978 the NAIC adopted the "isolation zone" concept, that is, eliminating the isolation requirement for alfalfa where the isolation zone is less than 10% of the entire field to be certified (area of field usually within the isolation strip or border) provided there is a clear line of demarcation between adjacent cultivars. This change in alfalfa isolation requirements for the certified seed class was subsequently adopted by AOSCA, OECD (Organization for Economic Cooperation and Development), and Canadian seed organizations where seed is to be used for forage purposes only. Age of stand, cropping history and control of volunteer seedlings were recognized as added problems in maintaining varietal integrity. AOSCA standards state the originators of cultivars or their designee are responsible for specifying age of stand for all seed classes of certified seed. Federal standards for producing hybrid seed were developed.

Various Germplasm Committees functioned from 1960 to 1972. When the National Seed Storage Laboratory at Ft. Collins, CO, was constructed, the conference resolved to assemble seed of cultivars, breeding, genetic, and otherwise unused stocks for deposition. Since 1962, originators of cultivars were encouraged to send seed to the laboratory. Much committee work has been concerned with foreign plant introductions. Storage of seed of all plant introductions in the laboratory was recommended as a germplasm reservoir. It was also recommended that if seed of an introduction is very limited in quantity when received, that it be increased before storage and evaluation. The following terminology for seed of plant introductions was recommended: a) that open-pollinated seed be listed with the original PI number plus the letters O.P.; b) to maintain the original PI number if seed is increased in a cage or isolation; c) if a PI is increased by hand pollination, that it be labeled with the original PI number plus Sib or S_1 as appropriate. Alfalfa breeders voluntarily increased seed of plant introductions where the seed supply was most critical. Committees worked during 1968 to 1972 on the development of a system for obtaining, recording, and retrieving data on plant introductions that could be computerized. Two broad-based germplasm pools NC-83-1 and -2, that included selected plant introductions with pest resistance, were produced in the NC-83 project and distributed through the Nebraska Agric. Exp. Stn. and the Regional Plant Introduction Station at Ames, IA.

The Committee on Preservation of Germplasm was established in 1972 and continues to function. In its initial years, the committee continued to stimulate interest in the seed increase of plant introductions, the incorporation of Canadian germplasm into the U.S. collection, germplasm exchange with foreign countries, germplasm pool development, simultaneous seed and *Rhizobia* collection, and domestic seed collection from old stands particularly in range areas of the western states and adjacent areas of Canada.

Alfalfa is one of eight crop species that was included originally in a Germplasm Resources Information Project sponsored by the USDA and initiated in 1978 to develop an information system to service the National Plant Germplasm System (NPGS). The system will allow data to be collected, stored, and updated and will permit multiple location access for data input and queries. The Regional Plant Introduction Station, Iowa State University, Ames, IA, is the alfalfa center. An Alfalfa Crop Advisory Committee (ACAC) was appointed. The committee members are from federal and state government, universities, private industry, and Canada. Membership of the NAIC Committee on Preservation of Germplasm is identical to the Alfalfa Crop Advisory Committee. The NAIC/ACAC committee developed descriptor lists, defined the descriptors and set priorities for research on plant introductions. The committee is developing evaluation strategies for the most important descriptors, with emphasis on pest resistance, using standard tests. Scientists in eleven states showed interest in evaluations. The joint committee also reviews and sets priorities on alfalfa seed collection proposals from all regions before consideration by regional and national germplasm committees. Several USDA/ARS committee members have worked closely with the Ames Station to identify plant introduction accessions for seed increase, assist in coordinating seed increases, and in planning explorations for seed and *Rhizobium* nodules. Plans were made for USDA personnel at Reno, NV, to increase seed of about 150 accessions per year starting in 1979. Seed increases in 1979 were variable but the 1980 and 1981 seed crops were successful. Recent plant explorations include the Andes of northern Chile, 1980; domestic collection in old stands of western U.S. and adjacent areas of Canada, 1980; Peru, Bolivia, and Ecuador, 1981; and Turkey, 1981.

SUMMARY

Because of the NAIC and its members, and through the cooperation of breeders, entomologists, pathologists, agronomists, and seedsmen, North America has a large number of multiple pest resistant alfalfa cultivars and a germplasm improvement program.

Response of Seedling Alfalfa Plants to High Levels of Chloride-Salts

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Salinity is a problem of many soils of Arizona and other southwestern states. Alfalfa is only moderately tolerant of saline conditions. Yet, alfalfa is a very important economic crop in Arizona.

Plant damage from a high salt concentration in the external medium most commonly is due to 1) reduced water potential that makes it difficult for the plant to obtain water, and 2) toxic effects due to certain ions. One of the most prevalent ions in southwestern soils and water is chloride (Cl^-) and alfalfa is readily damaged by this ion. Under desert conditions of Arizona, Cl^- may be in the irrigation water or accumulate in the soil. Chloride is highly mobile in the soil, except under low rainfall conditions. Alfalfa has a deep root that readily absorbs Cl^-, which accumulates in the plant tissue without becoming incorporated into organic molecules.

Most studies of salt tolerance in alfalfa at the University of Arizona have been conducted using seed germinated in various concentrations of NaCl solution. Therefore, we initiated a series of greenhouse experiments with young, established alfalfa plants to determine their response to several common salts, their survival to increasing levels and adaptation to Cl^-, and the influence of plant age, temperature, soil moisture, and light on survival. This information is needed to establish a screening procedure for the alfalfa breeder to identify salt-tolerant alfalfa genotypes and to develop salt-tolerant strains or cultivars.

All studies were conducted in the greenhouse with plants of 'Lew' alfalfa grown from seed in one-liter, plastic pots filled with Mohave sandy loam soil (2.7 pounds of soil/pot). Twenty or 25 plants/pot were grown to 4 to 5 weeks of age (5 to 7 inches tall) and treated with a salt solution, usually KCl. Plant survival was evaluated 4 weeks after salt treatment. Except at time of salt treatment, plants were watered only with distilled water.

Seedling alfalfa plants survived moderate rate of Cl^- (405 to 810 lbs Cl/A), but survival decreased markedly as rate of Cl^- was increased. All rates of salt were applied on a surface-area basis. Few, if any, plants survived 2430 lbs Cl/A applied in a single dose. Survival was not significantly different whether the chloride-salt was NaCl or KCl, when equal amounts of Cl^- were applied. Survival was much higher with the application of high rates of K_2SO_4, Na_2SO_4, and NA_2HPO_4 than with KCl or NaCl. The chloride salt chosen for use in subsequent trials was KCl. The Na^+ ion from NaCl caused the potted soil to deflocculate, especially at the soil surface and hindered water movement through the soil.

Young alfalfa plants increased in Cl^- tolerance as they matured. At all ages tested (2- to 7-week-old plants), a higher percentage of the plants was killed when the herbage was removed just before the Cl^- was applied. Plants survived application of Cl^- better at moderate (80°F) than at high (100°F) temperatures and when the soil was dry rather than when it was wet. However, plant survival was not significantly affected by light intensity.

At high rates of Cl^-, foliage of treated plants showed wilting within 2 to 8 hours and the top growth has collapsed by 24 to 48 hours after treatment with KCl. Most damage and killing of plants occurred within 7 days, even though data on survival were not taken until 4 weeks after exposure to salt. Chloride accumulated in higher concentrations in leaflets than in stems. Lowest concentrations of Cl^- were found in roots. All roots appeared to be alive at the end of the 4-week recovery period, even plants where all shoots had been killed shortly after salt treatment.

Plants survived high total application rates of Cl^- when they were allowed time to adjust following exposure to sub-lethal levels of Cl^-. This adaptive response makes it difficult to select Cl^- tolerant plants under field conditions. Exposure to variable and unknown amounts of Cl^- in the soil and/or irrigation water during seedling establishment makes it difficult to decide on the amount of chloride-salt to apply to reach the selection pressure needed to identify tolerant plants.

The above results were used to develop a procedure to screen alfalfa seedling plants in the greenhouse for Cl^- tolerance (salt tolerance). Full text of the above results with data and details of the screening procedure developed can be found in the Journal of Plant Nutrition 4(2):143, 1981.

A Systematic Program of Seed Increase and Phase-out Needed for Public Alfalfa Cultivars

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Alfalfa (*Medicago sativa* L.) was introduced into North America from 1850 to 1947 as nine distinct sources of germplasms from different regions of the world (Barnes et al., 1977). All of the present U.S. cultivars were developed from those germplasm sources. Prior to about 1925 most alfalfa breeding research in the United States was concerned with selecting germplasms with increased winterhardiness. 'Grimm' (1900), 'Cossack' (1907), and 'Ladak' (1910) were very winterhardy cultivars that were released during that period. During the next 25 to 30 years the primary objective was to develop cultivars that were winterhardy and resistant to bacterial wilt caused by *Corynebacterium insidiosum* (McCull) H.L. Jens. 'Ranger' (1940) and 'Vernal' (1953) are examples of cultivars that results from this effort.

During the last 25 years many diseases, insects, and nematodes have been recognized as important problems on alfalfa (Barnes et al., 1974). The public breeding programs were responsible for developing new selection and breeding methods and releasing cultivars with resistance to numerous pests.

INCREASE IN CULITVAR NUMBERS

New cultivars were released at a rate of about 0.3 cultivar per year from 1900 to 1940 (Fig. 1). The number increased to about one new cultivar per year from 1941 to 1960 and to about 12 per year since 1960. This dramatic increase in cultivar release was due to the expansion of private plant breeding programs and to the development of cultivars with multiple pest resistance. The expansion of private breeding programs began about 1958.

The increased numbers of alfalfa cultivars has created at least three problems. These include: 1) isolation of cultivars during seed production, 2) testing of all adapted cultivars in each state, and 3) discontinuing seed production of obsolete cultivars. The first two problems have affected both public and private cultivars. Therefore cooperative research was conducted to determine if isolation requirements could be decreased for the certified seed class in seed production fields larger than 2 ha without affecting the phenotypic integrity of the cultivar (Brown et al., 1980). These and other unpublished data were responsible for changes in seed isolation approved in 1978 by the Canadian Seed Growers Association and the Association of Official Seed Certifying AGencies in the United States. Evaluation of the large numbers of cultivars is managed by different methods in different states and Canada. The practice of

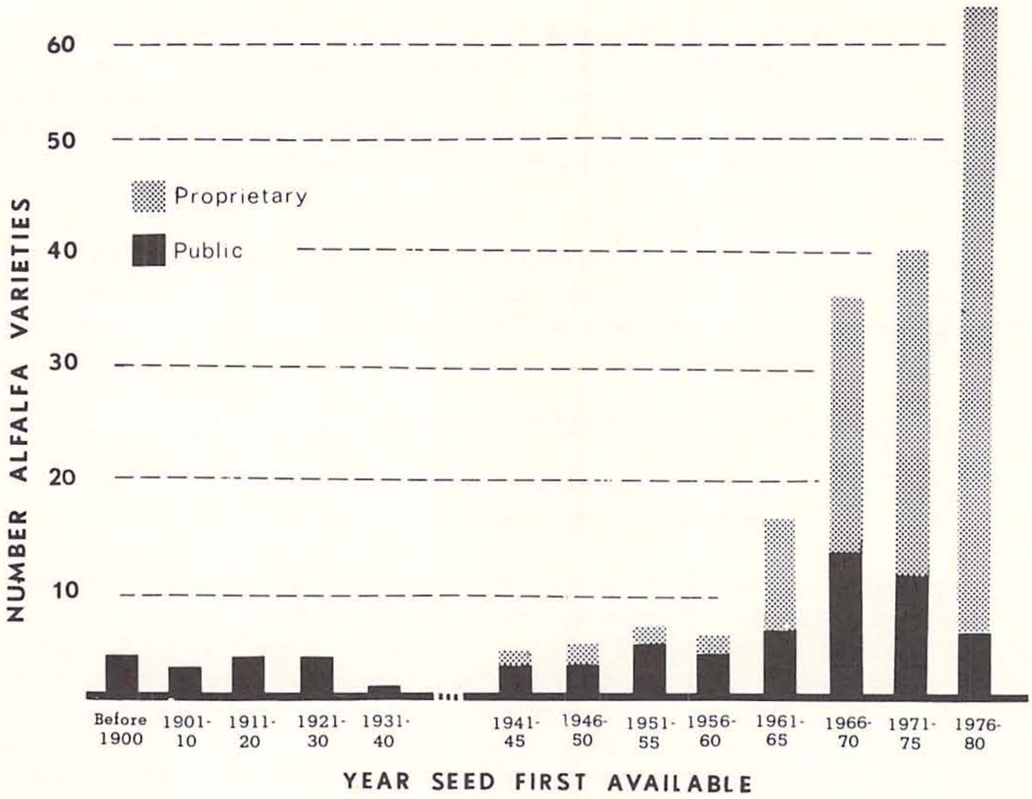

Figure 1. Numbers of alfalfa varieties released by public and private plant breeding programs in the United States.

industry paying a fee for the inclusion of their cultivars in public tests has increased the numbers of privately-owned cultivars being evaluated in many states. Uniform descriptions of new U.S. cultivars are also developed each year by the National Certified Alfalfa Variety Review Board, thus assuring impartial descriptions of both public and private cultivars.

Discontinuing seed production of obsolete cultivars is a problem unique with public cultivars. Cultivars developed by private plant breeding programs usually follow a systematic program of seed increase and phase-out. This is possible because stock seed, seed production, and marketing are exclusively controlled. Most public varieties released prior to 1980 have no provision for discontinuing seed production. This is further complicated since the area of seed production is usually different from the area of forage production. During 1981 certified seed of alfalfa cultivars, which are obsolete, was being produced. We have chosen the cultivars, Grimm and Ranger, to describe the problems that can be encountered when attempting to discontinue seed production of an obsolete cultivar.

GRIMM

Grimm was the first winterhardy alfalfa cultivar developed in the United States. A farmer, Wendelin Grimm, planted seed which he carried from Germany to his Minnesota farm in 1858 (Brand, 1911). Brand reported that in the beginning Grimm's alfalfa had a low percentage of hardy plants and that seed produced on the surviving plants provided for continual reseeding. Natural selection for increased winterhardiness and recombination of the surviving plants continued until about 1975, at which time the strain appeared quite uniform. About 1905 scientists at the University of Minnesota recognized and helped publicize the unique winterhardy characteristics of Grimm alfalfa. There was no systematic method of seed increased for Grimm. According to Walster (1924) the buyers of Grimm alfalfa were encouraged to ask for a pedigree to insure that it traced back to the original source of Grimm. Certified seed fields of Grimm were established in many locations using pedigreed seed. The lack of isolation requirements, length of stand limitations, and introduction of other germplasm undoubtedly were conducive to changes in the cultivar during the last 75 years.

Samples of authentic "1900 Grimm" are not available. However, a sample of 1925 certified Grimm was retained by the Wisconsin Agricultural Experiment Station. Wheeler (1951) described the original Grimm as being very winterhardy and susceptible to bacterial wilt. The 1925 certified seed lot fit that description, therefore we have considered it representative of the original Grimm. In 1977 we evaluated the bacterial wilt resistance and winterhardiness of 12 seed lots produced and sold as Grimm in 1973 through 1976.

All 12 Grimm seed lots tested were significantly different from the 1925 check lot for at least one characteristic (Table 1). We concluded that most, if not all, seed being currently produced as Grimm differs from the original cultivar. Since basic seed stocks are not available we recommend that certification and sale of Grimm alfalfa be discontinued. Similar arguments can be made for discontinuing the certification and sale of other obsolete cultivars such as Cossack and Ladak.

Table 1. Performance of Grimm seed lots marketed in 1973-76 as compared to a check seed lot.

Seed Lot	Certified	% Bacterial wilt resistant plants	Winterhardiness index†
3596	yes	47**	7.0
3595	no	40**	6.9
3593	no	30**	5.9**
3586	no	29**	6.0**
3594	no	24**	6.0**
3589	no	23**	6.8
3592	yes	22**	6.6
3597	no	21**	6.5
3588	no	18**	6.1**
3591	no	13	5.8**
3590	yes	9	6.3*
3587	yes	9	6.0**
Grimm (1925 check lot)	yes	3	6.8
Vernal	yes	47	6.8
Saranac	yes	--	6.0

*,** Statistically different than Grimm check lot at the 0.05 and 0.01 levels respectively.
†Based on fall growth after cutting spaced plants during 1st week of September: 1=tallest (least winterhardy), 9=shortest.

RANGER

Ranger was the first alfalfa cultivar developed in the United States with both winterhardiness and resistance to bacterial wilt (Kehr, 1959). Ranger was released by the USDA and the Nebraska Agric. Exp. Stn. in 1940. However, it was not until 1950, the second year after initiation of the National Foundation Seed Project, that certified Ranger seed became available in substantial quantities. Between 1950 and 1959 more than 200 million pounds of certified Ranger seed was produced (Kehr, 1959)

As new cultivars became available it became apparent that forage yields of Ranger were often lower than those of newer cultivars. Ranger is susceptible to most foliar diseases and insect pests. The level of bacterial wilt resistance in Ranger is also lower than most of the newer cultivars. The USDA and the Nebraska Agric. Exp. Stn. released the multiple pest resistant cultivars: 'Dawson' (1967), 'Baker' (1976), and 'Perry' (1979). These cultivars plus other public and private cultivars should have replaced Ranger. Nevertheless, Ranger seed production has continued because seed producers like its high seed yielding capacity, there is a market for the seed, and there is no age of stand limitation on seed production in the area of adaptation.

In 1972 the Nebraska Agric. Exp. Stn. initiated a program to discontinue Ranger seed production by terminating release of breeder seed. In 1975 the regional forage breeding committee (NCR 36) requested the National Foundation Seed Project to: 1) reclassify its inventory of Ranger foundation seed to certified seed and sell it and, 2) initiate no future contracts for Ranger foundation seed production. Regional action was necessary because regional action had been required to enter the cultivar in the National Foundation Seed Project. Unfortunately, registered seed was produced in 1975 because neither the Nebraska Station nor the National Foundation Seed Project could regulate the production of this class of seed. In addition, 3,072 hectares of certified Ranger were produced in 1975. It was apparent that action to discontinue Ranger seed production had to be cooperative between the Nebraska Agric. Exp. Stn., the Association of Official Seed Certifying Agencies (AOSCA), and the seed growers.

In 1976, AOSCA adopted a procedure for discontinuing alfalfa cultivars produced under seed certification. The Nebraska Agric. Exp. Stn. informed AOSCA of its 1972 and 1975 official actions to discontinue Ranger seed production. Nevertheless applications to AOSCA were made by seed growers for certifying significant acreages of registered and certified Ranger seed production in 1978.

In March 1979, the Director of the Nebraska Agric. Exp. Stn. wrote a letter to the Directors of Agricultural Experiment Station Directors inthose states that produced certified Ranger seed in 1978 requesting their cooperation in phasing-out Ranger seed production.

It was expected that no new fields would be established for certification and that current fields would be phased-out. AOSCA was informed in June 1979 of the Director's letter and replies, and asked to transmit the letter to its members.

A comparison of AOSCA Production Publications 32, 33, and 34 for 1978, 1979, and 1980, respectively, cast doubt on how rapidly Ranger seed production would decline. The number of hectares applied for registered production was 121, 96, and 52 in 1978, 1979, and 1980, respectively. Applications for certified production were 2,169, 2,154, and 1,255 hectares in 1978, 1979, and 1980, respectively, and new fields were seeded for certified production in two states in 1979 and five states in 1980.

Based on experiences with Ranger and Grimm we concluded that it is difficult to discontinue seed production of previously released public cultivars. Provisions must be made at the time of release to systematically increased and phase-out the seed production of a cultivar. Several methods that a breeder can use to regulate seed production of released cultivars are:

1. Recognize only three generations (breeder, foundation, and certified) of increase with the releasing agency maintaining control of foundation production.
2. Specify age of stand limitations for each generation and for maximum age of any specified field.
3. Protect a cultivar name through registration under the Plant Variety Protection Act certification provision, thereby preventing the selling of "uncertified" seed with the cultivar name.

Public plant breeders have the primary responsibility for insuring that programs are developed for the systematic increase and phase-out for their cultivars.

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Genetic Variability of the Root System Size and its Relation to Yield

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Relatively little progress has been reached by breeding for forage yield, not only in the U.S.A. (7) but also in Czechoslovakia (2). Greater success was reached indirectly by breeding for disease and insects resistance in areas of their regular occurrence. Alfalfa is perennial, harvested in more cuts in contrast to most crops where the progress in breeding has been compared. It could be one of the reasons for the problem described above. Alfalfa root systems and crowns must survive not only each winter, but also after each cut. Regrowth in spring and after cuts is therefore correlated with size of the root systems in field conditions, and also with forage production (5). Complex designs for evaluation and selection for forage yield are often used, but the root system is mostly evaluation only for disease resistance.

A significant correlation between root system size and yield or fertility was found in experiments with sufficient room for root growth (3, 4, 5). It therefore could be expected that selection for increased yield would also mean selection for increased root system size. But the expectation is not reliable because the correlation mentioned is not absolute, since stresses change during crop growth. In particular, the correlation between forage production and root system size in controlled environments of glasshouses and growth chamber was not significant.

This is because no great stress probably occurred there and there was only limited room for root growth (6). Strains with greater root systems responded to the change of environment by greater yield increment as compared with strains with smaller root systems (r=0.520) (6). The winter survival of these strains was also better (r=0.370) (4).

Limited information is available concerning the amount of genetic variability and mode of inheritance for alfalfa root traits (1). Examination of combining ability for root system size showed that general combining ability amounted to about 60%, which creates real prospects for selection. It was calculated that root system size was most closely correlated with forage yield of all selection criteria evaluated (4). Therefore, it may be that some progress in breeding for forage productivity in alfalfa may be reached by selection for root system size.

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The Seed Mass per Plant of Some American Lucerne Varieties in Czechoslovakia

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Fourteen U.S. varieties and hybrids and two Czechoslovak varieties were evaluated for seed yield at a plant spacing of 0.5 x 0.5 m with 4 replications, consisting of 144 plants per variety.

The seed mass per plant of American lucerne varieties and hybrids was low, except for the variety Apex that equalled the control variety Palava (Table 1). The seed yield was closely connected with the original germplasm of the variety and the intensity of seed breeding. Apex and A24 come from French varieties of 'Flamande' type which have a good yielding ability under our conditions. Iroquois and Mark II are related cultivars. The remaining varieties were low seed yielding under our conditions.

The influence of individual years on the seed mass per plant of the whole variety set is best demonstrated by relative comparison of its values in individual experimental years: 1971 = 100%, 1972 = 5%, 1973 = 44%. The genotype and environment shares in the seed yield formation were: genotype = 5.3%, year = 89.9%, other factors = 4.8%. In the unproductive year 1972 low temperatures (average: 19.7°C and 17.3°C) prevailed at the time of blossom and seed formation (July-August), and extreme precipitation represented nearly one half of the whole year's average (July-August: 298.3 mm).

In three experimental years, the most frequent plants in the whole variety set were those of the lowest seed mass - to 5 g (64.7%), and most productive plants represented (40.01 - 45.00 g of seed per plant) only 0.1 g.

The variability of seed mass per plant according to years, varieties, and in the whole set had abnormal Poisson's distribution.

For further selection the variety Apex appeared to be a suitable one.

Table 1. The seed mass per plant of American lucerne varieties and hybrids (RIPP, Piestany, 1971-73).

Variety (hybrid)	Seed yield per plant		
	g	relative %	in points 1-9
Hodoninka	4.83	69.40	3
Palava	6.96	100.00	5
Cayuga	3.97	57.04	2
Iroquois	5.42	77.87	4
Mark II	5.40	77.59	4
Saranac	4.82	69.25	3
Norseman	3.27	46.98	2
Ladak 65	4.13	59.34	2
A-59	3.42	49.14	2
A-24	5.40	77.59	4
Dawson	3.11	44.68	1
Titan	4.37	62.79	3
Apex	6.46	92.82	5
Endura	4.61	66.24	3
MX-82 (CMS hybrid)	4.25	62.50	3
TX-202 (CMS hybrid)	5.12	73.56	3

$\bar{x}$ = 4.73 g, s = 0.86, cv = 18.18%
Hd $P_{0.05}$ = 1.43 g, 20.55%; Hd $P_{0.01}$ = 1.93 g, 27.73%

The Combining Ability of Lucerne Clones with Fertility Characters

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In 1978-80 we evaluated 8 lucerne clones in two diallelic sets according to the following fertility characters: seed yield, pollen fertility, number of ovules in the ovary, number of seeds per flower and per pod, and the percentage of pod setting. Clones with combined yield capacity for mass and for seed were selected from French varieties Orca, Omega and F 34, and from the German variety Langensteiner.

Clones with higher level of self-fertility increased their seed yield in progeny. In the most fertile cross combinations the higher seed yield was combined with higher pollen fertility and higher number of ovules in the ovary.

When evaluating fertility characters, both GCA and SCA, were of primary relevance (with seed yield and pollen fertility). The best GCA in fertility characters was found in parental clone 1 (selection from Orca variety) and clone 8 (selection from F 34 variety) which were utilized in further improvement of yield capacity of lucerne for seed growing.

The results showed the importance of self-fertility, pollen fertility, and made necessary the thorough analysis of fertility characters with components of synthetic and hybrid lucerne populations under less favorable conditions for the purpose of seed growing.

Somatic Embryogenesis in Callus and Suspension Cultures of Lucerne

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This paper describes an induction system of somatic embryoids in callus and cell cultures of lucerne and their regeneration into intact plants. Phenotypic characteristics were evaluated in regenerants. The possibility of using in vitro techniques for propagation and breeding lucerne are discussed.

Callus cultures were derived from hypocotyl, coteledons, shoot, petiole and ternate leaves on modified Blaydes medium to which 100 μM 2,4,-D and 5 μM KIN were added. Primary callus was induced during 10 days of cultivation in vitro of the above-mentioned medium. Callus tissue was transferred to identical basic medium, to which 1 μM NAA and 10 μM KIN were added. During a 20-day cultivation light green centres containing embryoids began to form on the callus surface.

Process of somatic embryogeneis continued during passaging on solid or liquid medium (cultivation on horizontal shaker) of identical composition. Secondary embryoids differentiated on the surface of primary embryoids after long-term cultivation. Histological analysis confirmed the rise of secondary embryoids from epidermis cells. After transferring individual embryoids or their clusters on a basic medium without growth regulators, the embryoids germinated into whole plants.

Frequency of the establishment of embryoids and regenerants was observed in callus and cell cultures derived from different organs. Sequence of embryogenetic ability was as follows: Petioles, shoots, hypocotyls, cotyledons, leaves. Regenerated plants were transplanted directly into soil. In shoots where good roots were not established rhizogenesis was supported by transfer ring regenerants on the basic medium with 1 μM NAA. The plants from which tissue cultures of petiole and leaf were taken distinctly differed in their ability of somatic embryogenesis, which was quantitatively evaluated according to the number of regenerants. Clones with high embryogenous ability were selected by replicated explanation of petiole and leaves from regenerants of four generations. Clones without the ability to form embryoids under given conditions were obtained within the same cultivar. Contrast genotypes will be used for genetic analysis.

In order to evaluate phenotypes of established regenerants they were divided into groups in accordance with the kind of tissue culture they were derived from petioles, shoots, hypocotyls, cotyledons, leaves. Plants within each group were tested for stainability of pollen, number of eggs in ovary, leaf area, width and length of leaves.

Average stainability of pollen within a group was between 58-68%, width of leaves approximately 9-11.5 mm, length of leaves between 21.4 mm and 25.2 mm and leaf area from 4.2 cm^2 to 5.8 cm^2. Leaf area indices (LAI) were 2.14-2.78.

Based on double hierarchical grouping, significant differences were found in given characteristics both among groups and among plants within each group. The only exception was leaf area in regenerants derived from cotyledons and shoots. As far as this character is concerned no significant difference was observed among the given groups of plants.

The described procedure of somatic embryogenesis in lucerne seems to be very perspective. Possibility of obtaining high numbers of generations within a short time interval and their considerable phenotype diversity draws attention to their possible use in breeding.

Pollination of Lucerne in Enclosures

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The problem of proper pollination and mutual crossing of chosen genotypes often arises in breeding lucerne. In view of solving similar problems pollination of lucerne in enclosure by insect pollinators has been tested at our Institute.

For this purpose cages made of polyester nets (mesh size 2 x 3 mm) were used. The cages were of different size, the covered area was mostly 4 and 6 m^2, height 185 cm.

Plants were planted in beds covered by nets before flowering. In one instance flowering plants grown in small pots (10 cm) made of burnt clay were transferred to cages. Pots were rooted in soil.

Leafcutter bees, Megachile rotundata F. and four species of bumble bees (Bombus agrorum, B. lapidarius, B. lucorum and B. terrestris) were used for pollination of lucerne in cages.

Similar experiments have also been taking place in growth chambers under the following conditions: illumination 25.000 lux, a 16-hour day with temperatures 24 and 26°C, an 8-hour night with temperature 19°C. Temperature and light intensity graduately decreased one hour before switching off and increased after switching the light on. Apart from leafcutter bees and bumble bees (B. lucorum) also honey bees were tested in growth chambers.

Leafcutter bees, M. rotunda, adapted themselves well to limited room conditions. In one experiment they pollinated lucerne satisfactorily even in a narrow tunnel made by covering the plant row by net. Since additional feeding of leafcutter bees has not been successfully solved by now, it was necessary to avoid overstocking of the cages with resulting disproportion between demand and supply of food. With the rate of approximately one female for 1.5 m^2 all matured flowers were visited and tripped in succession. However, leafcutter bees were active only under temperatures higher than 21°C.

Adaptation to growth chamber conditions lasted 7-10 days. Females showed a higher degree of adaptability than males who died soon after emerging from cocoons. Of 15 incubated cocoons only 3 females succeeded in overcoming artificial environmental conditions. However, they were able to pollinate the given lucerne area of 4 m^2 very well.

Bumble bees seemed most suitable for pollination of lucerne in enclosure since they responded favourably to such conditions though smaller colonies reacted better than stronger ones. Of four species used the best pollinators were B. lucorum and B. terrestris since they visited lucerne in the open as well. Worker bees of B. agrorum and B. lapidarius learned to trip flowers and collect pollen after 3-5 days as soon as pollen storages in nests were consumed.

Proper pollination by bumble bees required the presence of younger brood in colonies. Presence of the queen was not inevitable since brood of laying workers sufficiently stimulated the colony towards pollen collection and tripping of flowers.

Easy application of additional feeding was another advantage of using bumble bees since there was not any danger of starvation of the colonies in case of occasional lack of flowers in enclosures. Bumble bees were easy to handle; mother colony could be divided into smaller pollination units. Moreover, bumble bees pollinated flowers under comparatively lower temperatures than leafcutter bees.

In an experiment performed in growth chamber in March the female of B. lucorum started brood cells and reared workers in a nesting box. Both, the queen and her workers, pollinated lucerne flowers very efficiently under artificial conditions thus confirming excellent adaptability of bumble bees.

Honey bees proved to be good pollinators of lucerne in growth chamber, too. Workers of the used colony were F_2 crosses of /Apis mellifica carnica x ligustica/ x carnica. They were used for pollination from December 3 to March 15 and collected both nectar and pollen. They learned to trip the flowers in order to obtain access to pollen. The nucleus used had about 4,000 individuals in the first stage. The colony gradually dwindled to about half that number and this strength was further maintained on the same level by brood rearing. Additional feeding by honey and pollen was necessary.

Seed yields of plants pollinated in enclosure differed in accordance with the material used. The average was 1-4 g of seeds per plant. Highest yield was obtained in p-cross of plants precultivated in pots. Pollination under these conditions was ensured by B. lucorum and B. terrestris bumble bees and the average for flower was 1.57 seeds.

The use of bees for lucerne pollination in enclosure brings multiple effect in breeding work. However, some questions of managing pollinators remain to be solved so that they would be available not only in growing season but during winter months as well.

Evaluation of Yield Dynamics in Alfalfa

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The performance differences among varieties and other kinds of breeding material the results are dependent on a number of factors which can decisively influence the interpretation of the results. There are common influences, i.e. the effect of the trial proper specified by the year of layout and implemented by the influence of the year of vegetation including the harvest year proper, then the effects of sites, harvest years and single cuts. Harvest techniques can also play a role, especially in evaluating green and dry matters. Routine evaluation is based on single trial results, mostly with regard to total annual production regardless of the above-mentioned classification. One of the possibilities of global evaluation is the construction of multidimensional variate including the criteria mentioned. However, multidimensional variate can be defined by various means.

First possibility is the construction of a complex trait consisting of a group of traits, where each trait is dependent on another one at least. Such a complex trait can be reduced to few artificial traits by multidimensional methods. These traits can in some cases be interpreted in basic form. When such interpretation is not possible, rotation of artificial traits is performed, which should enable their interpretation. Considering basic traits means to exploit gradually maximum of the total variance in case of their mutual independence. It usually happens that most information is included in the first two or three artificial traits. Artificial traits can be identified with the axes of rectangular coordinates in two-dimensional or three-dimensional systems. In these coordinates the selection goal chosen can be fixed. It can be given as the best performing variety or characteristics determined by the breeder (3).

A further possible approach consists of homogenizing the experimental material, i.e. excluding the influence of trials (layout years), which leads to decreasing of standard deviations and enables to define multidimensional variate with highly correlated components. These correlations facilitate the reduction of material into some artificial variables which could be taken as a system of coordinates. In it, varieties and original traits are presented. In this way the whole complex can be simplified graphically. Configuration of vectors and points can be practically checked by means of known facts, e.g. by the correlation of green and dry matter. In the absence of discrepancies with well-known facts, practical conclusions on performance and representation of varieties can be derived from graphical representation (1).

Another procedure is based on the fact that both individual cuts within a year and yields of successive years are not statistically independent. Should the performance trials respect the effects of the environment as well, then the effect of years of vegetation should be recorded by means of further trials (years of layout). These trials can obviously be laid out on new field plots only, so that under these circumstances the effect of years of layout is confounded with the effect of the site (soil). When apart from green matter yields, also dry matter yields (hay) and proteins are evaluated, we speak of performance trials with three (3) different categories of characters (viewpoints of classification or factors), the variants of which are not statistically independent. Such a complex of questions can be worked out mainly by means of partial analyses in the form of MANOVA when variants of a certain category of characters (e.g. cuts in a given harvest year in green matter or harvest years in a given cut and for a certain yield of character) form a multidimensional variable.

A procedure was worked out for summarizing the results of these partial analyses into a single common information (2). Simultaneously, it was shown how additional one-dimensional analyses of variance help the interpretation of partial multidimensional analyses as well as the possibility of using objective decomposition of the interactions occurred, in particular their linear components (4).

In addition the possibility of graph interpretations of results was shown and the solution of some methodology questions on these types of trials was given (5).

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Maintenance of Male Sterility in Back-Crosses

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After crossing of sterile forms A_2, A_3 with sterility maintainers, only sterile plants were found in the F_1 progeny. After back-crossing of sterile hybrid plants with sterility maintainers selected from inbred progenies of I_1 and I_2 generations, some semi-sterile and fertile segregates were observed in the BC_1 generations. The numbers of semi-fertile and fertile plants further increased in the BC_2 generation (Table 1).

Table 1. % pollen fertility in back-crossing BC_1, BC_2 generations.

Cross combination	Number of plants with % pollen fertility				Average % pollen fertility
	ms 0-10%	sms 11-50%	sf 51-75%	f 76-100%	
BC_1 generation					
A_2x0/1/x0I_1	83	--	--	1	1.7
A_2x0/3/x0I_1	43	--	--	--	0.5
A_2x0/3/x0I_{1-4}	10	2	--	1	12.9
A_2x0/5/x0I_1	74	5	--	--	2.0
A_3x0/1/x0I_1	74	2	--	--	1.9
A_3x0/2//x0I_1	9	1	1	--	8.2
A_3x16/1/x16I_1	92	2	--	--	2.2
A_3x16/2/x16I_1	31	4	1	1	8.9
A_3x16/x16I_1	43	5	--	--	4.3
A_3x16/x16I_1	88	24	3	1	9.8
A_3x29/x29I_1	56	4	--	--	4.1
A_3x29/x29I_1	87	40	1	2	12.8
A_3x46/2/x46I_1	38	1	--	--	1.1
A_3x46/x46I_1	80	2	--	--	1.9
A_3x46/x46I_1	18	5	1	1	13.6
A_3xMiro/xMiro I_1	19	--	--	--	2.8
A_3xMiro/x Miro I_1	43	15	--	--	8.6
BC_2 generation					
A_2x0x0I_1/x0I_2	105	2	--	--	1.6
A_2x0x0I_1/x0I_2	21	8	9	--	22.0
A_2x0/1x0I_1/x0I_2	--	1	3	10	83.0
A_3x0/1x0I_1/x 0I_2	12	--	--	--	2.6
A_3x0/1x0I_1/x0I_2	25	5	1	--	9.3
A_3x0I_1x0I_2/x0I_2	66	1	--	--	1.4
A_3x0I_1x0I_2/x0I_2	12	10	5	--	27.8
A_3x16x16I_1/x16I_1	24	--	--	--	1.4

$A_3x16/2x16I_1/x16I_1$	22	5	--	--	7.5
$A_3x16x16I_1/x16I_2$	27	--	--	--	3.3
$A_3x16/2x16I_1/x16I_2$	3	6	1	1	29.8
$A_2x0/3x0I_{1-5}/17/x16I_{1-7}$	13	--	--	--	1.9
$A_2x0/3x0I_{1-8}/7/x16I_{1-6}$	14	--	--	--	0.7
$A_2x0/5x0I_{1-1}/4/x16I_{1-7}$	15	--	--	--	0.9
$A_2x0/5x0I_{1-6}/7/x18I_{1-5}$	13	--	--	--	0.8

Maintainer plants that transmit a high pollen fertility can decrease the frequency of sterile plants in further generations when used in propagation of sterile plants in further generations when used in propagation of CMS. After self-pollination of sterility maintainers segregation was obtained with plants having a low percentage of pollen fertility, and self-sterility began to appear. Genotypes with a full ability to maintain pollen sterility in generative propagation were not obtained. These findings can affect the breeding models of lucerne heterotic hybrids.

Research on the Expression of Male Sterility in F_1 Lucerne Hybrids

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The beginning of lucerne heterosis research in Czechoslovakia is connected with obtaining sterile plants and with the study of their response to pollen sterility and fertility expressions in F_1 generation. Male-sterile plants marked as A_1, A_2, A_3, A_4 were obtained from the world collection of lucerne in 1970 after determination of pollen fertility by Lugol solution. Sterile plants A_1, A_3 come from the Bulgarian local population Bojnica, A_2 from Hungarian "Bekeszentandrasi", A_4 from Polish "Miechowska".

After open-pollination of male-sterile plants A_1, A_2, A_3, A_4 under field conditions progeny of F_1 generation was obtained. There were different frequencies of sterile (ms), semi-sterile (sms), semi-fertile (sf), and fertile (f) phenotypes (Table 1).

Table 1. Number and % of phenotypes per fertility class after open-pollination of sterile plants.

Sterile plants	Number of phenotypes with % pollen fertility								
	0-10		11-50		51-75		76-100		
	ms		sms		sf		f		S
A_1	25	(36.05)	41	(41.88)	10	(9.83)	34	(22.21)	110
A_2	22	(18.68)	28	(21.70)	4	(5.09)	3	(11.51)	57
A_3	16	(6.88)	5	(7.99)	0	(1.87)	0	(4.24)	21
A_4	36	(37.37)	41	(43.41)	13	(10.19)	24	(23.02)	114
S	99		115		27		61		302

$X^2 = 30.80$; P 0.5 = 8.34; P 0.001 = 27.87

Segregation of sterile, semi-sterile, semi-fertile, and fertile phenotypes in F_1 generation was not random in the investigated sterile plants, but it was associated with their sterility type. If phenotypically sterile plants were similar genotypically, the same results would be obtained in pollen fertility in F_1 progeny after crossing with the same fertile pollen parents. The existence of such dependence would suggest that all the sterile plants would be maintained by the same sterility maintainers.

From the results presented in Table 2, it follows that the average percentage of pollen fertility in F_1 generation of sterile plants A_1, A_2, A_3 crossed with the same fertile plants differed considerably (the sterile plant A_4 died out).

Table 2. The average pollen fertility percentage of sterile plants F_1 hybrids with the same fertile plants.

Father-fertile plants	Mother-sterile plants		
	A_1	A_2	A_3
0	58.80	2.08	0.50
1	63.12	15.00	14.67
3	85.50	48.33	23.33
7	40.83	7.63	5.82
13	48.75	56.67	8.50
16	39.45	--	3.38
34	50.71	29.64	3.93
59	87.50	73.00	22.69
72	47.50	27.69	23.83
80	56.00	41.75	41.82
88	46.71	41.44	29.27
112	35.90	24.60	34.85
113	83.60	31.50	23.25
Bronka	41.25	31.33	15.13
D	56.62	30.67	4.40
Miro	35.35	10.00	1.83
xy	42.44	26.78	9.50
Average	54.11	31.10	16.86

Paired testing hybridization of sterile plants A_1, A_2, A_3 with equal fertile plants show that phenotypically equal plants differ genotypically as well as in the type of cytoplasmic nucleus-male sterility (CMS). The initial breeding material for sterility maintainers formation was obtained in higher number with the sterile plant A_3 than with A_2. No fertile analogue was obtained for maintaining of the male sterility of A_1.

From the above it follows that with lucerne on the tetraploid level, more than one type of CMS will occur. Because of that, it will be necessary to work out the details, of breeding of lucerne heterotic hybrids using sterile forms for which sterility maintainers may be obtained most easily.

Performance of Polycross Progenies of Alfalfa Derived from Partly Inbred Parents
I. Forage Yield

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An alfalfa crop is high yielding when it is vigorous and persistent at the same time.

Concerning persistence, research on the development of the alfalfa canopy (Rotili, 1979) detected a positive correlation between the persistence of the alfalfa crop and the homogeneity of its constituents (the individual plants) for such characters as regrowth, velocity of growth and flowering. Selfing allows the best homogenization of the said characters.

Concerning vigor, previous experimental results (Rotili, 1976) showed that selfing, combined with selection in competitive conditions, effectively improved forage yield. The present program carried out in Lodi tends to verify those results in frequent cutting conditions.

Two parental populations (cultivars Leonicena and Cantoni) were chosen for this research. Within each cultivar, selection was practised for plants with dry matter weight exceeding the mean by two standard deviations. These plants were self-fertilized. Intense positive selection for vigor was practised between as well as within families after each generation of selfing.

Parental populations and selfed families were evaluated under competition and frequent cutting conditions. At every level of selfing, plants chosen were polycrossed by hand in the greenhouse. The progenies $S_0 \times S_0$, $S_4 \times S_4$, and $S_2 \times S_2$ were grown in concrete boxes (80 cm long, 25 cm wide and 60 cm high); 40 seedlings of each entry were planted in a double row in the box in a randomized block design with 7 replications. Data were collected on the 25 central plants of each row. Dry matter weight, earliness and mortality were determined. Cuts were made every 23-26 days.

From the results concerning the parental progenies common to each level of inbreeding, the following observations can be made: 1) Theoretically, vigor of polycross progenies derived from autotetraploid parents will decrease as their degree of inbreeding increases (Busbice, 1969; Demarly, 1968; Gallais, 1968). However, in this study, where selection in competitive conditions was practised, there is a favourable effect of selfing on the mean. Many progenies increased significantly with inbreeding. This indicates that selection of vigorous progenies during the selfing phase was successful. These results confirm those obtained in the previous experiment (Rotili, 1976); 2) The effect of selection is greater within S_2 families than within S_4 families.

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Performance of Polycross Progenies of Alfalfa Derived From Partly Inbred Parents. II. Saponin and Leaf Protein Content.

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Our breeding work is designed to develop alfalfa (Medicago sativa L.) cultivars for intensive management systems and industrial transformations. The method is based on the use of two generations of selfing combined with selection in competitive conditions. Breeding for quality concerns protein and saponin content of the leaves.

In the present experiment, saponin and leaf protein content of polycross progenies (45 $S_0 \times S_0$, 43 $S_1 \times S_1$ and 103 $S_2 \times S_2$) were analysed. Leaves for analysis were collected at the second and fourth cut. The saponin content was evaluated by means of a biological assay which utilizes the growth inhibition of the fungus Trichoderma viride (Zimmer et al., 1967). The protein content was detected by the colorimetric Autoanalyser method. It is important to underline that no selection was practised for either saponin or for protein content.

Concerning the protein content of leaves, the present results confirm the previous ones (Rotili et al., 1976, 1978, 1981) on the low variability of this character. The strategy of improving this character through the increase of the resistance to frequent cutting therefore seems to be the best way to obtain a higher amount of protein without a much lower amount of forage yield per hectare.

Concerning the saponin content, the following observations were made from Table 1:

1. The mean performance of the two populations is similar at every level of inbreeding.
2. The variability between families increases with inbreeding: the families with the lowest leaf saponin content are at the S_1 and S_2 level.

The correlation between S_1 and S_2 families was high for both populations. No correlation was observed between dry matter yield and saponin content.

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Table 1. Percent growth inhibition of *Trichoderma viride* colonies. Mean values and range of variation. Average of two cuts.

		Cultivar	
		Leonicena	Cantoni
$S_0 \times S_0$	Mean	43.7	45.0
	Range	37.5 to 51.8	39.5 to 50.5
$S_1 \times S_1$	Mean	35.8	35.6
	Range	28.4 to 42.9	22.1 to 48.4
$S_2 \times S_2$	Mean	41.7	39.3
	Range	29.5 to 49.4	22.3 to 50.1

Productivity and Survival of Alfalfas S_1 and F_1 Plants Grown Under Competitive Conditions

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In order to evaluate the importance of intra-specific competition in alfalfa breeding and to study the consequences of different percentages of self-pollinated seeds on productivity of alfalfa meadows a trial was carried out at Perugia (Italy) during 1981, using a randomized block design with four replicates. Three seed mixtures having rations of self-pollinated seed (S_1)/cross pollinated seed (F_1) of 1/9, 3/7. 1/1 were sown, seed by seed, in micro-plots (60 x 40 cm) with a seeding rate of 1600 seeds/m².

Data were collected on: establishment, green matter yield per plant of selfed and crossed origin and dry matter yield per plot at 1st, 2nd, and 3rd cut, number of plants alive in the fall season.

The results can be summarized as follows:

1 The evaluation and the selection of single alfalfa plants grown in competitive conditions are possible and could be useful for breeding purposes;

2) The productivity of S_1 plants was much lower than that of F_1 in each of the three mixtures; such differences were greater than those normally shown in spaced plant trials;

3) No differences in dry matter yield per plot were shown among the mixtures. On the whole, an increase or a decrease in S_1 seed percentage would not seem very important with respect to the productivity of alfalfa meadows in the first year;

4) Within each mixture it was possible, on the basis of data collected on the 3rd cut, to identify some F_1 phenotypes (about 10% of the seeds sown) characterized by a high green matter yield per plant;

5) No differences in establishment were shown between S_1 and F_1 seedlings; on the contrary, in each mixture fall survival percentages of S_1 plants were significantly lower than those of F_1; that points out the presence of a selective S_1 elimination which seems to start, in the Italian environmental conditions, with the summer period of water and thermal stresses. The intensity of selection tends to increase with increasing age of the plants, and in an

alfalfa meadow which is one year old, only a few S_1 plants would be present, irrespective of their percentage in the initial seed lot.

Hybrid Alfalfa for a Frequent Cutting Schedule

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After more than ten years of research work on hybrid alfalfa, about 500 three-way hybrids were produced (1). The best hybrid made with a cytoplasmic male-sterility system received favourable review in 1981, and approved for certification by name of "KM-Hybridalfa." This hybrid has outyielded the Europe variety by 8, 12, and 33% in the 2nd, 3rd, and 4th years, respectively, when harvested on a cutting cycle of 28 days at 5 locations (2). On a standard system of cutting, however, hay yield of this hybrid has not differed significantly from Europe. Consequently "KM-Hybridalfa" had a high persistence enabling it to be utilized in a more intensive cutting cycle.

These data also show that winterhardiness, shorter dormancy, and rapid recovery after cutting are not necessarily incompatible characteristics of alfalfa.

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Crude Protein Content of Alfalfa as Affected by Inbreeding

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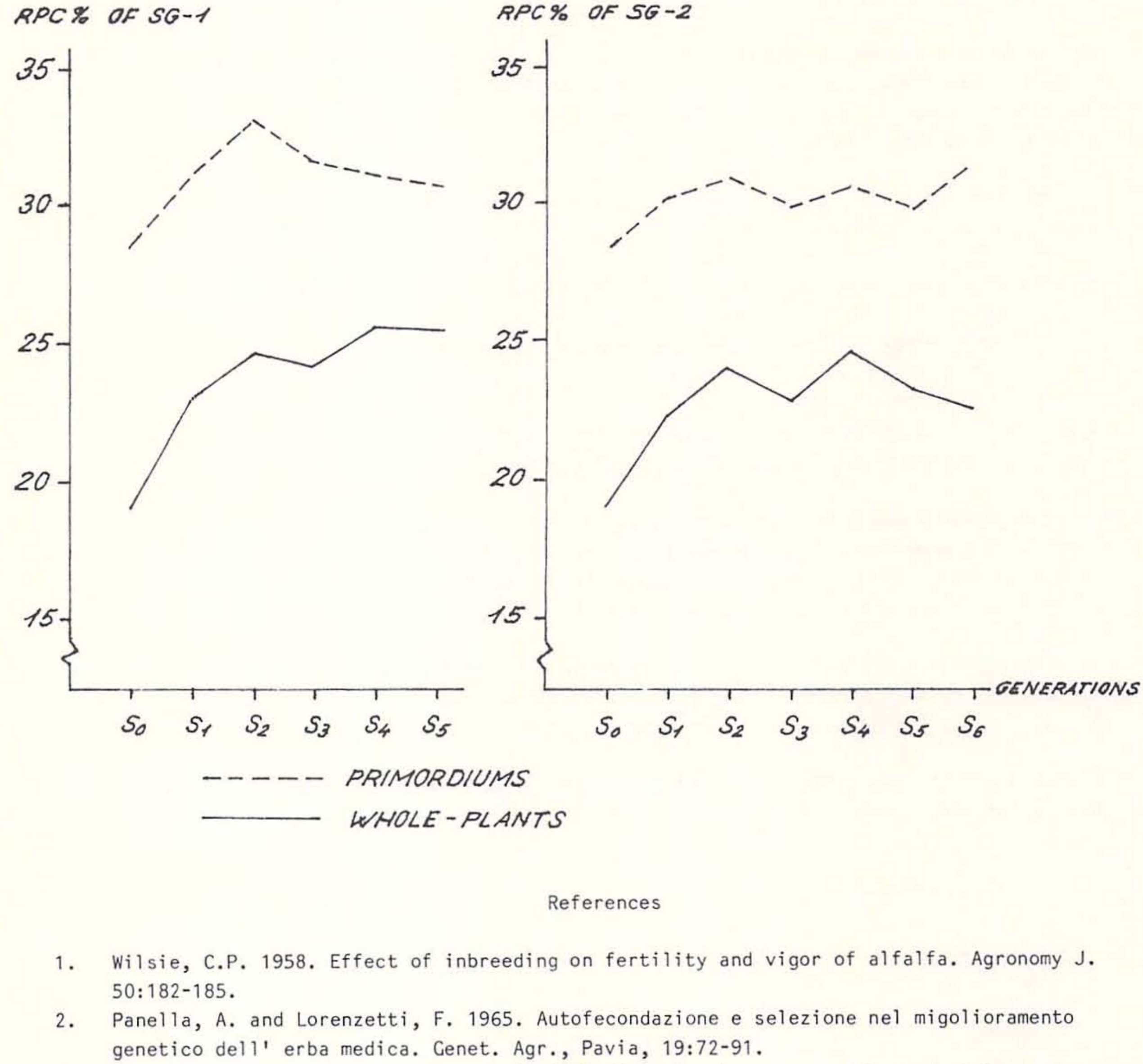
Spaced plants of two lots of selfed generations (SG-1, SG-2) and a mixed open pollinated population (OPP) have been tested for raw (crude) protein content (RPC) on an individual plant basis. Primordia* and whole plants** were taken for samples, then dried at 60°C and Kjeldahl analysis followed. Results are briefly summarized in figures 1-2. Some of our conclusions are as follows:

1. Mean value of RPC (crude protein) is higher and its range narrower in SFs than in OPP.
2. Within generations variability shows no discernible tendency when sequenced according to intensity of inbreeding.
3. Mean RPC values of generations follow a set pattern: they are always highest in S_2 compared to S_1 and S_3; their cumulative tendency over S_0-S_2 invariably breaks in S_3, never consistently returns in further generations.
4. Pattern of RPC mean values over S_1-S_3 was consistent with and agreed with former findings on selfing [dry matter and quality/quantity relationships (1, 2, 3)].

MEAN ROW PROTEIN VALUES IN FUNCTION OF GENERATIONS
FOR SG-1 AND SG-2.

FIGURE 1. FIGURE 2.



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*Taking as many primoriums as possible from each plant with 4-5 cm stem.
**Taking the whole above-ground plant at the time of appearance of 2 open florets.

Research on Combining Ability of Back-crossed Alfalfa CMS Lines

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Using a controlled pollination method, a new hybrid alfalfa named "KM-Hybridalfa" was produced in Hungary, and received state registration in 1981. The cytoplasmic male sterility makes it possible to cross parents with good combining ability in a controlled way. A further objective is to improve the combining ability of the parents.

This paper is to report a part of a long-term task where the objective is to produce hybrids between inbred lines using cytoplasmic male sterility. The selection of parents is essentially based on the progeny test, because the "provocation" of the initial material -- in this field of breeding -- is the crossing alone.

We began the inbreeding of male sterile (cms) parents with the BC-method. Our fundamental objectives were:

1. How does the inbreeding (that is the selection of lines) affect the productivity of progenies?
2. Is there a negative effect caused by the male sterility of female parent in the productivity of its progenies?

In our earlier research on testing sterile clones and cms F_1 we experienced that productivity of progeny depended on the general combining ability (GCA_F) of the female parent.

In our research there were 63 families obtained from crossing of 9 male sterile and 7 fertile strains. The experimental design was a split-plot, with plot size: 0.54 m² in 4 replications, started in August 1980, so in 1981 we had a stand comparable to a 2-year-old planting. Results were computed by variance analysis (VA, Table 1).

The progeny test confirmed our earlier statement referring to the genetical importance of the heterozygote of male parents: i.e. crosses between non-inbred fertile parents there were no statistically differences based on progeny average.

By increasing the intensity of inbreeding of the female parent the frequency of 3 and 4 allelic interactions have decreased in the progenies, a symptom well demonstrated by the decrease of SCA values.

	max.	min.
SCA_{F1} 109	42.73	-55.85
SCA_{BC1} 109	23.51	-61.77
SCA_{BC2} 109	15.84	-24.82
SCA_{BC3} 109	16.04	-19.49

The increase of homogeneity of the female parent resulted in a greater "hybrid homogeneity" as evidenced by reduced CV values of the progenies shows: F_1-109 = 19.8%; BC_3-109 = 12.3%. From the table of variance analysis, it can be seen by the productivity of progenies that the additive material effects were significant for the 9 male sterile parents. The BC-progenies outyielded the cms F_1-109 progenies by 7-26 per cent on the average of 7 testers (Table 2).

On the basis of another similar experiment, we studied the relation between the sterility of female parents and the mean average of green forage yield of progenies grouped according to females. A low r-value (0.52; n=9) was obtained, which does not suggest a close relationship.

Correlations were also determined between seed setting capability of the cms-lines (female fertility) and the 1st year green forage yield of their progenies.

The r-values of the correlation between pod/100 flowers and seed/100 flowers on the one hand, and 1st year green forage yield of the progenies on the other, were r=0.013 (n=63) and r=0.044 (n=63), respectively. This indicated that seed setting ability of female parents did not effect the fodder yielding potential of combinations based on cytoplasmic male sterility. However, seed setting data of female parents cannot be fully ignored, because this character plays an important role in the propagation of the new combinations.

Table 1. Variance analysis of the 9 x 7 NC-2 experiment.

Source of variance	SQ	DF	MQ	F-test	Statistical variance components
Replication	28.20	3			
Fertiles (Male)	2.15	6	0.36	1.55^{NS}	σ^2_M = 26.51
Error (rm)	3.41	18	0.19		σ^2 = 0
Steriles (Female)	24.32	8	3.04	13.14***	σ^2_F = 991.93
Genetical Interaction	12.60	48	0.26	1.14^{NS}	σ^2_{MF} = 78.11
Error (rmf)	38.86	168	0.23		σ^2_{e1} = 2.31

*** = Significant at 0.01 level, NS = non significant.

Table 2. Average of progeny of the male sterile parents for one year and five cuttings.

Code of the male sterile parent	Green yield kg/plot	Green yield relative
1 FM-2	4.3	125.8***
2 F_1 137	4.3	124.0***
3 BC_3 109	4.3	123.5***
4 FM-1	4.2	122.0***
5 KM-Hybridalfa+	3.9	114.0***
6 BC_1 109	3.9	113.2***
7 BC_2 109	3.7	107.0
8 F_1 140	3.6	104.2
9 F_1 109	3.4	100.0

+TC-hybrid as maternal parent.

***P=significant as .007 per cent level.

The Relationship between Fusarium Wilt and Agronomic Characteristics of Some Hungarian Lucerne Varieties

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We have analyzed the relationship between Fusarium wilt tolerance and some agronomic characters as reflected by a number of Hungarian cultivars which are mainly of sativa types: Kisvardai-1, Kisvardai-2, KM-Hybridalfa, Mv-Syn-alfa, Nagyszenasi, Szarvasi-1, Szarvasi-2, Tapioszelei-1, Verko, Vertibenda.

Fusarium wilt reduced green forage yield in the 4th year of the stand.

There was a positive relationship between rapidity of regrowth and Fusarium tolerance (r=0.86). Those cultivars proving themselves to be persistent showed less susceptibility to Fusarium wilt (Szarvasi-2, 67.7%, Hybridalfa, 60.2%), and had rapid recovery after cutting.

Seed yields of the varieties were significantly different. The differences were much greater than the differences of green forage yield. Erectness and seed yield showed a close correlation (r = 0.70). There was no relationship between seed yield and Fusarium susceptibility (r=0.18). Differences in winterhardiness were very small, therefore no relationship was found between this character and green forage or seed yield. The relationship between recovery and winterhardiness was negative (r=-0.42) while erectness and winterhardiness showed a positive (1) correlation (r=+0.48). Cultivars showing agronomic factors which beneficially influenced green forage yield had a relatively low seed production.

Severity of Wilt in Different Varieties of Alfalfa as Affected by Harvesting Schedule

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In Hungary, Fusarium and Verticillium are the major causal agents of wilt, depending on the given ecological conditions. Cultural practices play an important role in the development of these diseases. Observations carried out under conditions of provocation (stress) suggest that disease is more severe in dense stands used for green fodder than in spaced plants intended for seed production.

We established a trial with different cultivars (Europe, KM-Hybridalfa, Leonicena, Szarvasi-2, Verko, Vertibend) to determine their disease reaction under different cutting schedules (28, 35, 42 days between cuts).

Cultivar	Wilt index at the end of the third year according to intervals of cutting: 28 days	35 days	42 days
Europe	82.3	59.8	55.0
KM-Hybridalfa	46.7	16.6	20.5
Leonicena	43.0	26.1	16.6
Szarvasi-2	67.2	27.3	10.4
Verko	77.7	62.4	59.8
Vertibenda	74.7	57.3	54.9

The wilt index of southern cultivars with 6 cuts per year was 52.2%, while the Flamande types could only be cut 4 times per year to give a similar (56.6%) wilt index. The relationship between green forage yield and wilt index was very high (r=0.95) with the most intensive cutting schedule and less (r=0.33) when only 4 cuts per year were taken. When a comparison was done between entries with the same number of cuts, yield performance within a life span varied strikingly. Yield performance, however, appeared to be similar when a comparison was made between 6-cut southern types and 4-cut Flamande types. By choosing an optimal number and time of cuts, severity of Fusarium wilt can be reduced and life span of the stand increased.

Female Sterility in Alfalfa (Medicago sativa L.)

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Female-sterile plants fs-1 and fs-2 were selected from progeny of a mutation derived from the alfalfa cultivar Warminska. Female-sterile mutants were crossed to 20 various plants of male-steriles and normal bisexuals.

Morphological shape of plants and flowers in fertile and female-sterile forms were similar, but the fs-2 plant was characterized with self-tripping.

Female-sterile plants and normal-fertile components used for crossings had 74-95 per cent pollen stainability and shed much pollen. The fs-2 plant had pollen sacks which did not dehisce in tripped flowers.

The fs-1 represented a type of semi-female sterility because 4.5-8.6 per cent of 1170 pollinated florets set pods. Female sterility expression in the fs-2 was stronger. This mutation has proved entire female-sterile because in 1510 pollinated florets, only 0.0-1.8 per cent set pods. Seed setting for fs-2 was also low in various crossing combinations.

Male functions of fs-1 and fs-2 mutants in reciprocal crosses were normal. Male-sterile plants pollinated with fs-pollen set 100 per cent of pods in hand-tripped florets. Sexual functions of female organs in the fs-1 and fs2 were very limited inspite of its floret structure being normal.

Observed female sterility appeared to be inheritable because of the phenotypical trait expression in progenies from various crossings. The female sterility character was transmitted to F_1 progeny only in the combinations where fs-plants were used as the maternal parent. The female sterility traits was not observed in the reciprocal crosses. Not one of 417 F_1 plants obtained from female-fertile x female-sterile combinations segregated fs-phenotype. These results indicate a cytoplasmic-genic type of inheritance. The cytoplasmic factor and one recessive gene system is expected on the basis of our preliminary experiments.

If entire female-sterile populations were bred, they would be exploited as pollen-producing components in hybrid breeding. On the other hand, a high frequency of female sterility genes in alfalfa populations perhaps could be a reason for seed set reduction.

Segregation of F_1 progeny of crosses of female-sterile and normal plants.

Crossing variants	Number of pollinated florets	Number of set seeds	Pod setting per percent	Phenotypic segregation in F_1 progeny: Seed number per pod	No. of F_1 plants	Female fertile plants	Female sterile plants
fs 1 self-pollinated	200	8	4.5	1.1	6	1	5
fs 1 x fs 2	70	5	8.6	0.8	5	1	4
fs 1 x Mf	900	66	7.3	1.2	57	52	5
ms x fs 1	270	497	100.0	3.3	257	257	0
fs 2 self-pollinated	290	0	0	0	0	0	0
fs 2 x fs 1	110	2	1.8	1.0	1	1	0
fs 2 x Mf	1110	6	0.5	1.0	2	1	1
ms x fs 2	170	316	100.0	3.4	160	160	0

fs - female-sterile selections
Mf - normal hermaphrodite plants
ms - male-sterile plants (ES rf_2 rf_2 rf_2 rf_2 genotype)

Evaluation of Alfalfa under Dryland Conditions

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Alfalfa varieties and ecotypes of different origin have been evaluated in northern Syria under dryland conditions (350-400 mm rainfall). Dry matter yield in the first year after establishment ranged from 1.5 m T/ha to 2.5 m T/ha over four cuts.

The seasonal distribution of dry matter yield suggest that the materials can be grouped into two distinct types. One group includes winter-dormant types (Iran, Turkey, Yugoslavia, Lebanon) which gave poor dry matter yield in the first cut, but considerably higher yield in the second and third cuts. The second group, which is winter-growing, gave a very similar dry matter yield in the first two cuts. Some varieties belonging to the second group (Mesa Sirsa, Arizona, Hayden and Sonora) were also the most productive in terms of total dry matter yield.

Breeding Alfalfa for Tolerance to Multiple Cutting

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Use of alfalfa for preparation of fodder rich in protein is promising not only in the main zone where alfalfa is being grown (Middle Asia, Northern Caucasis, Ukraine) but also in the new regions of its growing (Non-chernozem zone, Baltic Republics, and Byelorussia).

It was assumed that with early cutting of sward and increasing number of cuttings, the harvest of grass with high content of protein can be raised. However, investigations have shown that when cutting alfalfa hay types in the budding stage, the sward badly weakened and killing of plants was often observed. In consequence of severe weather conditions in this zone, the plants have no time to store a sufficient supply of food reserves for developing subsequent shoots. After a two-year cutting of sward at the early stage of growth, the killing of plants reaches up to 75-85%, and when harvesting at the beginning of flowering, 10-15%. Usually alfalfa is cut 1-2 times at the beginning of flowering when green mass contains maximum quantity of protein (15-16%).

The task is to create a variety, which after 3-4 cuttings at the budding stages, gives quick regrowth and accumulates food reserves, mainly from buds of the tillering zone for development of shoots.

Interspecific and intraspecific hybridization with subsequent selection is used in the breeding work. Interspecific hybrids obtained from the artificial crossing of species belonging to *Medicago sativa*, *M. falcata*, and *M. varia* serve as the initial material. Hybrids for multiple cutting were created on the basis of biotypical selection from simple hybrids (F_2-F_3) and subsequent pollination of them in the polycross nurseries.

As the result, the promising simple and complex hybrids have been obtained. Evaluation of these hybrids was done by multiple-cut usage (3-4 cuts at the budding stage) when protein content in herbage was 22-24%. Great attention had been paid to the rate of sward recovery following cutting, resistance to *Fusarium oxysporum* and unfavourable weather conditions. Hybrids MH 2, MH 8, MH 9 possessing high resistance to multiple cutting at the budding stage have been developed. Total yield of protein at the cost of additional cuttings and increasing content in the plants during harvest at the early stage of vegetation increased by 20-30%.

Hybrids provided an increase in dry matter production in total for 3 years vs. the standard by 22.1-23.1%. Habit of shoot formation and form of root system greatly influences the sward resistance to frequent cutting. It is very important for the non-chernozem zone that alfalfa should form more shoots from the root-crown buds and the root system should have a tap-coronal or tap-branching form.

Alfalfa Research in Pakistan

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in cooperation with USDA under PL-480 Project No. FG-Pa-253/PK-ARS-48(N)

1) Polycross test with 16 varieties of alfalfa

This study was carried out to test the general combining ability of 16 varieties of alfalfa so that best combining lines having greater potential for green fodder yield may be selected for use in synthetic production of alfalfa varieties. Seeds of these varieties were sown in field rows randomly and allowed to cross freely using honeybees. The polycrossed seed of each variety was collected separately and was sown in a polycross test during 1979. The data was recorded on green fodder yield and its components for 1979 and 1980. The six sources having the highest fodder yield potential were the U.S. varieties AS-13 (17.1 m tons/acre), C/W/5 (17.90 m tons per acre), AS-13R (17.2 m tons/acre) and 185 alfalfa (17.1) and two Pakistan varieties, type 18 (18.3 m tons per acre) and Punjab selection (17.4 m tons/acre).

2) Fertilizer trials with alfalfa

Studies with the following treatments were carried out from 1976-77 to 1977-78.

Treatment No.	Fertilizer treatments N	P	K lbs/acre
T1	0	0	0
T2	30	30	0 at sowing
	30	30	0 after first cutting
	30	60	0 after second cutting
	30	90	0 after third cutting
	30	120	0 after fourth cutting
	30	150	0 after fifth cutting
T3	30	100	0 at sowing
T4	30	180	0 at sowing
T5	60	180	0 at sowing
	Variety - Type 8/9		

The maximum yield of green fodder was 29.02 m tons per acre, obtained from fertilizer treatment T5, where 60 lbs nitrogen and 180 lbs $P_2 05$ acre was applied at sowing, followed by T3 where 30 lbs nitrogen and 100 lbs $P_2 05$ acre was applied at sowing (27.3 m tons/acre), and T4 (30 lbs nitrogen and 180 lbs $P_2 05$/acre) applied at sowing (27.3 m tons per acre). T2 where nitrogen and phosphorus were applied in split applications at different cuttings yielded 27.0 m tons/acre as compared to 22.3 m tons per acre for the control.

3) Effect of different intervals of cutting frequencies on the green fodder yield of alfalfa

The results of three years studies (1978-80), revealed that different cutting frequency treatments significantly affected the different morphological characters like plant height, stem thickness, number of tillers/plant and fodder yield. An interval of 29 days gave the highest green fodder yield of 14.2 m tons per acre followed by 36 days intervals (12.8 m tons), 22 days cutting frequency (7.8 m tons per acre) and 15 days cutting intervals (6.6 m tons) per acre.

4) Height of cutting experiment with alfalfa

The results of three years studies (1978-80), showed that maximum green fodder yield from alfalfa was obtained when the crop was cut at two inches level above the ground (29.2 m tons per acre), followed by cutting alfalfa close to the ground (24.5 m tons per acre). The lowest green fodder yield (23.5 m tons) resulted from a cutting level of four inches above the ground.

Committee on Preservation of Germplasm

This is a continuing committee. The former alfalfa GRIP Committee [Germplasm Resources Information Project, an information system being developed for the National Plant Germplasm System (NPGS)] was renamed the Alfalfa Crop Advisory Committee (ACAC). Membership on this NAIC Committee and on the ACAC Committee is identical. Several significant contributions have been made to alfalfa germplasm since 1980, when a report was given at the 27th NAIC and included in the proceedings.

The alfalfa advisory committee has been working on the initial phases of a seven-phase plan for the conservation and use of plant genetic resources. The plan originated in 1979 with the National Plant Genetics Resources Board. The alfalfa committee is one of eleven advisory committees.

A Proposal for the Evaluation of Alfalfa Germplasm was presented by D. K. Barnes at the August 1981 meeting of the ACAC. In the 1978 and 1979 committee meetings, lists were made of high, medium and low priority descriptors (characteristics) on which information was desired. Descriptors were defined and evaluation strategies were discussed. All descriptors will be included in the Germplasm Resources Information Network (GRIN), the information system designed to serve the NPGS. Descriptors regarding resistance to 12 diseases, 9 insects, and 3 nematodes were given high and medium priority and will be addressed first. Standard evaluation procedures are available for 9 of the 12 diseases, 5 of the 9 insects, and the 3 nematodes. State and federal alfalfa scientists with expertise in pest resistance evaluation were surveyed with regard to their interests in evaluating accessions. An estimated annual budget of about $60,000 would enable the evaluation of about 150 accessions for 18 descriptors. Continuous seed increase and evaluation was considered necessary to evaluate the present and future collections. A semitechnical report on the status of Alfalfa Germplasm in the United States is being prepared by committee members.

The NPGS Registry Committee met at Peoria, Illinois, in 1981. A registry-specific descriptor list was developed, and the use of a universal identifier number was discussed. At a subsequent GRIP Coordinating Committee meeting it was decided that the Plant Inventory (PI) Number would function as the primary identifier, but supplementary identifiers could be used.

Alfalfa exploration and seed collection trips during 1980-81 included the following: The Andes of Chile, 1980, W. R. Kehr; Domestic collection in the U.S. and Canada with emphasis on old stands and range areas, 1980, M. D. Rumbaugh, R. J. Lorenz, A. C. Wilton, and W. R. Kehr; Bolivia, Peru, and Ecuador, 1981, M. D. Rumbaugh and W. F. Lehman; Turkey, 1981, J. H. Elgin Jr. and R. H. Ratcliffe. No alfalfa seed collection trips were scheduled for 1982. Collection proposals are planned for Morocco; Bulgaria, Hungary, and Romania; North Africa; Peru; Israel and southern Mediterranean areas; Yugoslavia and Greece.

About 170 foreign accessions were increased under cages at Reno, Nevada, in 1979 and 1980. About 200 cage increases were made in 1981. Seed production per accession has varied greatly, from none to a pound or more. Initial funds for this program were for a five-year program managed through the cooperation of the Regional Plant Introduction Station at Ames, Iowa, and the University of Nevada at Reno. Continued annual support is anticipated so that accessions can be increased soon after collection.

A National Seed Repository of U.S. alfalfa cultivars was established at the Plant Introduction Office (PIO) at Beltsville, Maryland, in 1981 to meet foreign requests. The Repository is supervised by G. A. White and J. H. Elgin Jr. All public and private breeders were invited to contribute seed. The PIO will fill requests by sending 25 grams of seed.

The *Medicago* collection of Karlis Lesins has been given to the Devonian Botanic Garden, Edmonton, Canada. The collection will be catalogued and assessed for species of the most interest, seed increase and maintenance status.

National concern on the adequacy of the National Plant Germplasm System has been expressed in several committee reports and in a report to the Congress by the comptroller general. The current status, and its strengths and weaknesses were evaluated, and a long-range plan (1983-1997) was made by the USDA/ARS in the interest of strengthening the total germplasm system. Douglas R. Dewey was recently appointed by the USDA/ARS national technical advisor for "Crop Germplasm Enhancement".

Publications

Kehr, W. R. 1981. Collecting alfalfa seed in the Andes of Chile. Proc. 17th Central Alfalfa Improvement Conference, E. Lansing, Michigan. p. 11.

Rumbaugh, M. D., W. R. Kehr, R. J. Lorenz, W. F. Lehman, and A. C. Wilton. 1981. Recent alfalfa germplasm acquisitions from North and South America. Agronomy Abstracts. Annual meetings American Soc. of Agron., Atlanta, Georgia. p. 71-72.

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Report of the Committee on Available Breeding Lines of Alfalfa

Responses to questionnaires yielded the following list (Table 1) of germplasm releases not previously recorded in conference reports. Table 2 contains a list of cultivars that were available before the National Alfalfa Variety Review Board was organized and the status of basic seed stocks and acreages in seed production as of April 1982. The table also contains a listing of cultivars that were reported as discontinued. Examination of records from the National Seed Storage facility at Ft. Collins reveals that 52 of the 243 previous germplasm releases are stored. Forty eight of the germplasm releases were clonal material, two of which are represented by self seed stored at Ft. Collins. One hundred sixteen of the releases are registered in Crop Science.

One hundred eighty five varieties have been favorably reviewed by NAVRB (through Dec. 1981). Ninety nine have been registered with Crop Science (includes 19 pre-NAVRB and Canadian). Forty nine of those reviewed before 1981 are not stored at the Ft. Collins facility. The varieties Canadian Variegated, Ferax, Kaw, Macsel and Liberty are also not stored at Ft. Collins. A list of germplasm releases and varieties that are not stored is being forwarded to the seed storage laboratory with a recommendation that originators be contacted with a request for seed. Retirement and discontinuation of alfalfa research programs at a number of locations provide a distinct possibility that germplasm may be lost. Up to 5 pounds of Buffalo Foundation seed may be requested from Pioneer Seed Co. at Kerman, California as long as the excess supply lasts. Contact Boyd Hartman before January 1, 1983.

The committee recommends that:

1. An updated list of discontinued varieties should be prepared.
2. Guidelines on discontinued varieties should be made available to certification agencies.
3. An updated list of released germplasm for which seed or clones are no longer available should be prepared.
4. An updated supplement or a revision of ARM-W-5 (197) "Improved Breeding Lines of Alfalfa" should be prepared within three to four years.
5. The committee should continue its assignment to compile a list of available breeding lines for the 1984 conference. This should include lines that are not officially released and a listing of germplasm stored in archives at various locations that might be available for limited distribution.
6. Strong emphasis should be placed on registration of germplasm and variety releases in Crop Science to provide readily available documentation of this material as it is used in research programs.
7. A list of "endangered" varieties should be mailed to appropriate conference members with a request for information on seed stored in locations other than the National Seed Storage facility.

Submitted by,

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Real Michaud
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Table 1. Alfalfa germplasm releases since 1980 N.A.I.C.

State/Agency /Company	Contact	Name of Germplasm	Stock (seed or clone)	Description
Kansas AES	E.L. Sorensen	KS145	seed	Derived from Cody and DuPuits. Resistant to anthracnose, downy mildew, bacterial wilt, pea aphid, spotted alfalfa aphid.
Kansas AES/ USDA	E.L. Sorensen	K78-10	seed	Derived from cvs Anchor and Elga. Resistant to downy mildew, anthracnose, pea aphid and spotted alfalfa aphid. (CROP SCI. 21:476, 1981)
Nevada/USDA	B.D. Thyr	NMP-8 CLS_5	seed	Common leaf spot resistant. Non-dormant germplasm.
Nevada/USDA	B.D. Thyr	NMP-46	seed	Blue alfalfa aphid-resistant winter-hardy germplasm developed from Wash. SNI.
Nevada/USDA	B.D. Thyr	NMP-47	seed	Blue alfalfa aphid-resistant non-hardy germplasm developed from Southwest Intercross (SNI).
Nevada/USDA	B.D. Thyr	NMP-50	seed	Blue alfalfa aphid-resistant semi-hardy germplasm developed from 'Washoe'.
Nevada/USDA	B.D. Thyr	NMP-11	seed	Common leafspot-resistant alfalfa germplasm developed from P.I. numbers 196233, 196247, 209091, 262550, 277425, 292773 and 302929.
Nevada/USDA	B.D. Thyr	NMP-12	seed	Common leafspot-resistant alfalfa germplasm developed from P.I. numbers 167263, 196228, 204460,

State/Agency /Company	Contact	Name of Germplasm	Stock (seed or clone)	Description
Nevada/USDA	B.D. Thyr	NMP-12 (continued)		251205, 251689, 251830, 287884, 287886, and 341814.
Nevada/USDA	B.D. Thyr	NMP-13	seed	Common leafspot-resistant alfalfa germplasm developed from P.I. numbers 182239, 196225, 253445, 255962, 260246, 300578 and 384507.
Nevada/Oregon /USDA	B.D. Thyr	GXB GXC GXE	seed	Dormant alfalfa germplasm well adapted to irrigated areas of the Intermountain West and Pacific Northwest regions.
New Mexico AES	Bill Melton	EUPH-5	seed	Non-dormant, of El Unico parentage with high levels of resistance to <u>Phytophthora</u>.
New Mexico AES	Bill Melton	EUAN-5	seed	Non-dormant of El Unico parentage with high levels of resistance to races 1 and 2 of anthracnose.
New Mexico AES	Bill Melton	MAN-5	seed	Semi-dormant of Mesilla parentage with high levels of resistance to races 1 and 2 of anthracnose.
North Carolina/ USDA	Will A. Cope	NCMP 1	seed	Developed by selection for pest resistance, plant habit, and genetic diversity. Moderate resistance to bacterial wilt, pea aphid and high resistance to phytophthora root rot, anthracnose (race 1) and spotted alfalfa aphid.
North Carolina/ USDA	Will A. Cope	NCMP 2	seed	Originated from a cross between a weevil-resistant population and

State/Agency /Company	Contact	Name of Germplasm	Stock (seed or clone)	Description
North Carolina /USDA	Will A. Cope	NCMP 2 (continued)		Apalachee. Moderate levels of resistance to bacterial wilt and anthracnose (race 2) and high levels of resistance to phytophthora root rot and anthracnose (race 1). Moderately resistant to the pea aphid and yield is above the average of the common commercial varieties.
North Carolina /USDA	Will A. Cope	NCMP 10	seed	Originated from a cross between Saranac An 4 and a weevil-resistant population. Moderate resistance to anthracnose (race 2) and high resistance to bacterial wilt and anthracnose (race 1) and pea aphid. Yield over two years averaged one ton per acre greater than "Arc".
North Carolina /USDA	Will A. Cope	NCWMP 22	seed	Developed by a three way cross between three weevil-resistant populations. Highly resistant to spotted alfalfa aphid, phytophthora root rot, and anthracnose (race 1); moderately resistant to bacterial wilt and pea aphid.
Univ. of Alberta	K. Lesins		seed	Ten to 12 plants self fertile and self tripping in greenhouse. Progeny show resistance to inbreeding depression. 200 seeds from each plant available on request.
Utah/USDA	M.D. Rumbaugh	GP52-III	seed	Derived from 3 cycles of selection from C3. Adapted to grazing in

State/Agency /Company	Contact	Name of Germplasm	Stock (seed or clone)	Description
Utah/USDA	M.D. Rumbaugh	GP52-III (continued)		dryland pastures and rangelands. Dormant with resistance to Fusarium.
Utah/USDA	M.D. Rumbaugh	BC-79		Traces to introductions by N.E. Hansen. Sixty to 80 percent root spreading with resistance to bacterial wilt and Fusarium root rot. Winter dormant for range and pasture in semi-arid areas.
Utah State Univ./USDA	M.D. Rumbaugh	U-5560	seed	A low saponin, bacterial wilt resistant strain.
Washington AES /USDA	J.H. Elgin, Jr. (now at BARC)	W10	seed	Intercrosses among Arc, Agate, Apalachee, Beltsville 2-An_4, Beltsville 3-An_4 and Nev. Syn WW. Moderate to low levels of resistance to anthracnose, bacterial wilt, Phytophthora, root-knot nematode and stem nematode, pea aphid and spotted alfalfa aphid. (CROP SCI. 22:163, 1982)
Wisconsin	E.T. Bingham	'Regen-S'	seed	A cultivated tetraploid developed from 'Saranac' by three cycles of recurrent selection for regeneration of plants from callus. Selected genotypes will also regenerate from protoplasts. Exp. Sta. Release, Dec. 7, 1978.

Table 2. Present seed stored and acreage of alfalfa cultivars which were available prior to NAVRB organization (Includes some cultivars available since NAVRB that are discontinued).

Name	Location of seed	Class of seed	Amount stored	Acreage in production
A-59*	Pratt County, KS, processed by Farm Research, Inc.	Registered		50 A(1980)
	Idaho	Certified		21 A
	Wright City, MO Embro Seed Co.			
African*	California	Foundation	550# (1954-55)	
Alfa*		None reported		
Apalachee*	Oxford, NC	Breeder	700 g	Discont.
Arc*	Oklahoma	Foundation		
Atlantic*	New Jersey	Breeder(1954)	125# 5-8% germ)	
	New Jersey	Breeder(1952)	5#	
Beaver*	Canada	Foundation		35-73 A
		Certified		5,015 A
	Idaho	Certified		96 A
Buffalo*	Frederick, OK	Certified		100 A
	Kerman, CA	Foundation	480#	(Contact Boyd Hartman if interested in this seed. It will be destroyed after 8/1/82)
California Common*	El Centro, CA	First generation after certified	450 g	

*See sample stored in National Seed Storage Laboratory, Fort Collins, CO.

Name	Location of seed	Class of seed	Amount stored	Acreage in production
Caliverde*	El Centro, CA	Foundation	340 g (1954)	
		Certified	1675 g (1955)	
Canadian (ON) Variegated*		None reported		
Cherokee*	Oxford, NC	Breeder	25#	Discont.
Chilean 21-5*	California	Certified	100 g (1958)	
Cody*	Oklahoma	Registered		37 A(1981)
		Certified		337 A(1981)
Cossack*		None reported		
Culver*	Indiana		50# (Reported to be in cold storage but unable to find it)	
	Indiana	Basic (?)	Small seed lots available from Dr. John Axtel	
DuPuits*	Northern Nevada	Non-certified		?
Ferax*	Univ. of Alberta	Breeder	10#	
Fremont*	Laramie, WY	Breeder	2#	
		Foundation	10#	
Grimm*	Saskatchewan, Canada	Breeder (?)	5#	
	Canada	Foundation		3 A
Hairy Peruvian*	California	Common	3500 g (1962-63)	
Hardigan *		None reported		
Hardistan*		None reported		

Name	Location of seed	Class of seed	Amount stored	Acreage in production
Indian	California	Breeder	250 g	
		Parent stock (before 1955)	200 g	
Kaw		None available per E. Sorensen, KS		
Ladak*	Idaho	Certified		306 A
	Northern Nevada	Non-certified		?
	Montana	Certified		940 A
Lahontan*	Idaho	Certified		14 A
	Northern Nevada	Non-certified		?
		Breeder	Available	
Liberty	Oxford, NC	Registered	900 g	
	Oklahoma	Foundation	Available	
Macsel		None reported		
Meeker Baltic*	South Dakota	None available		
Moapa*	California	Certified	1005#	
	Nevada	Breeder	Available	
Narragansett*	Idaho	Certified		14 A
	Northern Nevada	Certified		40 A
	New York	Breeder	None available	
Nemastan*		None reported		
New Mexico* 11-1	Hatch, NM	Registered, certified	1700#	13 A
	Galem, NM	Certified	1750#	13 A
		Breeder	5#	
		Registered	Some	
Nomad	Oregon		None in 1981	

Name	Location of seed	Class of seed	Amount stored	Acreage in production
Orestan*	Ontario, OR	Registered	458#	
Rambler*	Oregon	Certified		354 A
	Canada	Foundation		20 A
		Certified		1602 A
	Idaho	Certified		65 A
	*Swift Current, Canada	Breeder	75#	
Ranger*	Idaho	Certified		910 A
		Registered		16 A
	Colorado	Certified		50 A
	Madras, OR	Registered	4718#	
	Northern Nevada	Certified		620 A
	*Univ. of Nebraska Bill Kehr	Breeder	5#	
	Nebraska	Certified	50#	
	Nebraska	Certified		2132 A
	Washington	Certified		112 A
Socheville*		None reported		
Talent*	Oregon State Univ.	Breeder	3#	
	Oregon	Certified		5 A
Team*	Beltsville, MD	Breeder	150#	Discont.
Teton*	South Dakota	Breeder	83# (1960)	
Turkistan*		None reported		
Vernal*	Idaho	Certified		3681 A
	Colorado	Registered		15 A
	Northern Nevada	Certified	2#	400 A

Name	Location of seed	Class of seed	Amount stored	Acreage in production
Vernal (continued)	Montana	Certified		324 A
	Washington	Certified		5422 A
		Foundation	340#	
	Canada	Certified		351 A
Williamsburg*		None reported		
Zia*	Pecos Valley area, Carlsbad and Hagerman, NM	Non-certified	Estimated 35-40 T (Mostly carryover seed)	Approx. 300-400 A
	New Mexico	Non-certified	50,000#	300 A

Committee on Potential Use of Tissue Culture in Alfalfa Improvement

This report summarizes the scope of current tissue culture research on alfalfa, clover species, and birdsfoot trefoil in North America, and cites related research from other parts of the world. To conserve space, certain chemical symbols and terms are not defined; it is recommended that project leaders be contacted for full details.

In California, Bill Rains (UC-Davis) is selecting for salt tolerance in cultivated alfalfa and has regenerated plants from salt tolerant callus. Currently, comparative studies on ion uptake of the salt-selected and non-selected cells are being carried out to determine possible mechanisms and regulations involved in the ability of salt-selected cells to grow on high NaCl levels. One alfalfa line being used is 'Regen-S.' Regarding salt tolerance, Michael Smith and J. A. McComb in Australia, reported that W75RS (Regen-S) was more salt tolerant than cvv., 'Hunter River,' 'Cuf 101,' and 'Hasawi' and that tolerance was manifested at both the callus and whole plant level.

T. J. McCoy (USDA-ARS, Reno) and T. R. Knous (UN-Reno) are using culture filtrates of the Fusarium wilt pathogen for selection of alfalfa cell lines resistant to a toxic component(s) produced by Fusarium disease. Two different cellular responses have been observed in two different alfalfa lines. Resistant plants of 'Moapa 69' are susceptible to the culture filtrate as cells in culture; whereas resistant plants of a Hungarian experimental line are resistant to the filtrate. Different responses may reflect different mechanismsof resistance.

At Kansas State, Lowell Johnson (Manhattan) is using mesophyll-derived protoplasts of alfalfa to (1) study variability of protoclone regenerates, (2) develop protoplast fusion methods to eventually transfer pest resistance and other desirable traits from sexually isolated species, and (3) use pathogen produced toxins for _in vitro_ selection for disease resistance. Regen-S selections have been used thus far but regenerable selections from Kansas cultivars are being sought. Published reports of alfalfa regeneration from protoplasts include those from Johnson's group, Kao's laboratory (Sask., Canada) and Cocking's group (Nottingham, GB).

Glenn Collins (UK, Lexington, KY) is using alfalfa and red clover as regenerating legume species in research aimed at regenerating soybean and other recalcitrant legume species. Both biochemical and developmental processes will be monitored while manipulating media and culture conditions. Other work with clover species includes virus-elimination via meristem-tip culture, interspecific hybrid embryo rescue, and regeneration of plants from protoplasts.

In New Mexico, Greg Phillips (Las Cruces) is screening 1000 regionally adapted genotypes for regeneration to underpin somatic cell selection research for stress tolerance; namely, tolerance of limited phosphorus. In a pilot study using red clover selected under conditions of limited phosphorus, regenerated plants exhibited improved phosphorus efficiency and increased forage concentrations of phosphorus and total nitrogen.

At MONSANTO, Donna Mitten (St. Louis) indicates that the definition of the reduced nitrogen requirements for regeneration has been instrumental in improving regeneration responses. Somatic embryo formation requires a minimum of 12.5 mM NH_4^+ in the regeneration medium for optimal expression. A survey of nine germplasm sources of modern alfalfa has shown 80 percent of the genotypes from the cv. 'Ladak' to regenerate from tissue culture. Some regeneration was found among all the germplasm sources except Falcata and Indian. In vitro selections for salt tolerance have yielded many presumptive variants which are being used in future studies.

In South Carolina, Earlene Rupert (Clemson) has found regenerable genotypes in three cultivars of T. repens ('Tillman,' 'Regal' and 'Sacramento'). Explants from hypocotyl sections are induced to callus on media containing either Picloram or 2,4-D, and then regenerated on media containing BA, kinetin or 2iP. Callus material of the most productive cultures will be used to duplicate the alfalfa and red clover cell-plant systems. Embryo culture and embryo callus culture have been used extensively in the Clemson program to obtain wide hybrids involving eight species of Trifolium, section Lotoidea. Several years ago, meristem culture was used to eliminate viruses from the parental clones of Tillman white clover.

Ike Kawaguchi and Keith Walker are developing commercial quantity breeding stocks of alfalfa with high capacities for regeneration at Plant Genetics, Inc. (Davis, California). This material will be a cornerstone for exploiting cell and molecular biology in varietal improvement. In addition, David Stuart is initiating fundamental investigations in the control of somatic embryogenesis by ammonium ion. This research is at an early stage in our laboratories.

At Wisconsin, Ted Bingham (Madison) is focusing on analyzing the genetic control of many variants of HG2 alfalfa recovered in an experiment designed to yield amino acid over-producers. Amino acid variants have tended to be sterile, hindering gene transfer, but some other interesting variants have been genetically transmitted, such as a dominant semi-dwarf. Cells were mutagenized; hence, most variants are probably due to induced mutations. Some spontaneous variants have also been recovered and one of them with improved clonal yield and fertility appears to be due to chromosome substitution. This spontaneous improvement in culture was not as great, however, as that obtained in four generations of conventional selection using selfed progeny of HG2.

Red clover and birdsfoot trefoil are being used in several applications of tissue culture by Dwight Tomes and his associates at the University of Guelph (Guelph, Ont.). These include germplasm storage, somatic cell selections and analysis of variation arising in tissue culture. They are able to store genotypes of birdsfoot trefoil and red clover up to one year at 4°C without subculture while progeny tests are completed. This is particularly useful in birdsfoot trefoil because repeated propagation by cuttings has been unsuccessful. Physiological differences are evident between in vitro and crown bud cuttings, but the in vitro propagation does not appear to induce genetic changes in either species. In vitro selection of birdsfoot trefoil for 2,4-D tolerance and altered nitrogen metabolism has been successful and altered genotypes are being analyzed genetically.

At Agrigenetics, Dale Hanson (Madison) is optimizing alfalfa regeneration from mesophyll and suspension culture protoplasts in preparation for experiments on DNA transformation, cell physiology and genome expression. About 70 percent of isolated protoplasts survive and undergo cell division using optimized culture conditions in V47 salts with an osmoticum of 10 percent manitol, simple organic components, 0.5 mg/l zeatin, 1 mg/l NAA, and 0.2 mg/l 2,4-D. Callus differentiation into embryos was obtained by acute 2,4-D treatment and was genotype dependent.

Dick Smith (USDA-ARS, Madison) has regenerated both red and crimson clover (T. pratense and T. incarnatum, resp.) from hypocotyl, petiole, and ovary explant tissues. Current activities include recurrent selection for improved regeneration, cold storage of tissue, and meristem culture for virus elimination. After one cycle of selection, regeneration capacity of red clover was increased from 4 percent to 42 percent.

Conclusions: Tissue and meristem culture are being used by several groups for virus elimination in the small seeded forage legumes and this use is expected to continue. The basic research applications of disease toxins in tissue cultures are being exploited in the characterization of resistance mechanisms, and eventually should find applied use in screening for some forms of disease resistance. Screening for salt and herbicide tolerance using tissue culture is under way in several laboratories. Regeneration of whole plants often has been genotype specific in alfalfa and clover species, whereas it is less dependent on genotype in birdsfoot trefoil. The tendency for most genotypes of Ladak to regenerate may be related to its ability to develop adventitious shoots from root segments as reported in 1950 by Dale Smith. Since alfalfa, birdsfoot trefoil and several clover species can be regenerated from callus, suspension cultured cells and sometimes protoplasts, somatic cell fusion within and among these legumes is a logical next step and is underway in a few laboratories.

Some variants have already been recovered from somatic cell selection and are being analyzed. Spontaneous variation arising in culture is also being evaluated, and protoplasts are being prepared for various aspects of genetic engineering. At this time, most applications of tissue culture in alfalfa improvement involve production or transfer of simply inherited traits which are not obtainable using conventional methods. The major effort now devoted to conventional breeding of quantitative traits is expected to remain unchanged.

Respectfully submitted by:

E. T. Bingham (Chair)
G. B. Collins
D. J. Hanson
L. B. Johnson
T. J. McCoy
D. H. Mitten
G. C. Phillips
D. W. Rains
E. A. Rupert
R. R. Smith
D. T. Tomes
K. A. Walker

Committee on Standard Tests for Characterizing Disease and Insect Resistance of Alfalfa Cultivars

The committee was charged by the 27th Alfalfa Improvement Conference to complete its task begun in 1977 of revising the standard tests bulletin (ARS-NC-19) originally published in 1974 with 2,700 copies (supply now exhausted). The committee has completed its assignment. The final version of the new bulletin has been submitted to the USDA, North Central Region, Office of Information for publication. It is expected to be available about October 1982. The new bulletin will include several new maps and testing procedures as well as revisions to maps and procedures published previously. Copies will be distributed to all scientists on the National Alfalfa Improvement Conference mailing list. Sufficient additional copies should be available for classroom use.

Note: A collection of standard seed lots of the 21 check cultivars recommended in the standard tests bulletin is being maintained at the Beltsville Agricultural Research Center. In most cases, foundation seed are available; however, registered or certified seed have been substituted where necessary. These seed lots serve as a uniform source for the check cultiars and will be supplied in amounts up to 20g to those conducting evaluations in which the standard checks are needed.

Requests for seed of the standard checks should be addressed to J. H. Elgin, Jr., USDA, ARS, Field Crops Laboratory, Bldg. 001, BARC-West, Beltsville, MD 20705.

Respectfully submitted by:

F. I. Frosheiser
K. T. Leath
W. F. Lehman
M. W. Nielson
R. H. Ratcliffe
E. L. Sorensen
D. K. Barnes
J. H. Elgin, Jr., Chrm.

Committee on Industry Relations

Recently several of us discussed the need for more interaction between the National Alfalfa Improvement Conference (NAIC) and the various segments of the alfalfa industry. There is a need for more communication among researchers, producers, and consumers to convey industry needs to research agencies and provide an opportunity for industry to facilitate NAIC activities.

I propose that the incoming chairman name a committee to include the Conference secretary, two other members of the NAIC, and one member each from the American Dehydrators Association, the American Seed Trade Association, and the Certified Alfalfa Seed Council.

The committee would have the charge to promote a close relationship between industry and NAIC and enhance the overall development of the alfalfa industry.

Respectfully submitted,

E. L. Sorensen

E. L. SORENSEN, Chairman

Resolution Committee Report

Be it resolved that the 28th National Alfalfa Improvement Conference in session at the University of California, Davis, on July 13-15, 1982, adopts the following resolutions:

1. Our sincere appreciation is extended to the administration of the University of California, Davis; to the Agricultural Experiment Station, and to the Agricultural Extension Service, for providing facilities and staff for making this conference possible. Special appreciation is extended to Dean Hess for his words of welcome.

2. A special thank you is extended to Dr. Larry Teuber for his tremendous effort in the planning and coordination of the conference.

3. An additional thank you goes to Ike Kawaguchi and Vern Marble for their successful leadership in organizing the pre- and post-conference tours.

4. Furthermore, we express our appreciation to those many people from industry who contributed to the highly successful pre- and post-conference tours. Contributors to the tours include:

   Hosts

   W-L Research, Inc., J. B. Boswell Company, Pioneer Hi-Bred International, Northrup-King Co., Jackson-Perkins Rose Farms, Cal/West Seeds, Dairyland Seed Co., Inc., Lovelock Seed Co. Inc., University of California, Davis, University of Nevada, and USDA.

   Refreshments

   California Seed Assoc., Nevada Seed Council, Kern County Hay Growers Assoc., Cel Pril Industries Inc., San Janquin Hay Growers Assoc., Kamprath Seeds, and Plant Genetics, Inc.

   Food

   W-L Research, Inc. - Bosque dinner
   J. B. Boswell Company - Luncheon
   Pioneer Hi-Bred International - Mexican dinner
   Cal/West Seeds - Luncheon and refreshments
   Northrup King Co. - Luncheon and refreshments
   Lovelock Seed Co., Inc. - Luncheon and refreshments

5. Many thanks are also extended to the alfalfa seed industry for sponsoring the special luncheon during the conference. Contributors include:

Arnold Thomas Seed Service, Inc., Cal/West Seeds, Dairyland Seed Co., Inc., DeKalb Ag Research, Inc., Farm Seed Research Corp., FFR Cooperative, Great Plains Research Co., Inc., Land O'Lakes, Inc., Lovelock Seed Co., Inc., North American Plant Breeders, Northrup King Co., Pioneer Hi-Bred International, W-L Research, Inc., Plant Gentics, Inc.

6. Additional people to whom we want to give special recognition for the success of the conference include: Lloyd Stockton, Boyd Hartman, Bill Knipe, Don Smith, Bob Shesley, Shirley Bell, and the many others who assisted in carrying out the details to conduct the conference.

7. Finally, we express our appreciation to the officers of the NAIC for their efforts in organizing an excellent program that we have enjoyed at Davis, California.

Respectfully submitted,

Don W. Graffis
James H. Elgin, Jr., Chairman

Secretary's Report

The NAIC is in its forty-eighth year. The national conferences have evolved from a small group of 27 scientists from eight states, to this meeting of nearly 250 people for nearly all states and providences of the U.S. and Canada plus representatives from many foreign countries. The numbers of disciplines represented has increased dramatically and the number of researchers from the private sector is nearly equal to those in the public sector. Many of the most dramatic changes in the NAIC have occurred in the last six to eight years. This is illustrated by the increases in numbers of scientists receiving NAIC mailings. Accurate numbers of members are not available prior to 1976, except that when I became secretary in 1974 the mailing list included about 100 people.

	Number of Scientists Receiving NAIC Mailings			
Source	1976	1978	1980	1982
United States	168	225	294	334
Canada and Mexico	28	51	59	65
Non-North American	30	53	102	138
Total	226	329	455	537

Continued membership growth and increased numbers of activities is good news. There is also some bad news. The position of permanent secretary was established by the early conferences as part of the duties of the USDA leader of alfalfa research . The NAIC secretarial duties have been considered part of the person's USDA assignment. This allowed use of governmental mail privileges and the USDA published the NAIC Proceedings. The current level of NAIC activities, the potential USDA budget restraints, and the increasing NAIC time requirement by the secretary necessitates that some changes in NAIC operations be considered.

Presently there are about two or three NAIC mailings made each year. In the last two years printing costs for those mailings plus an address directory cost $1273.52 (see Treasurer's Report for NAIC 6-23-82). All monies for printing costs have usually come from registration surpluses of the previous national conference, interest on the NAIC bank account, and sale of address directories. The 27th NAIC income did not keep pace with the 28th NAIC activities. The deficit of the NAIC operation has come from non-USDA monies in the Minnesota Alfalfa Breeding Project.

<u>Secretary's Financial Report for NAIC</u>

Balance in account 6-26-80	521.12	
owed to MN Alfalfa Project	(214.55)	
Balance on hand 6-26-80		306.57
<u>Income</u>		
Interest 6-26-80 to 6-23-82	44.37	
Sale of directories	14.00	
Registration surplus 27th NAIC	200.00	
		258.37
<u>Disbursements</u> (Printing Costs)		
Announcements of reginal meetings, 1980 National Certified Alfalfa Variety Review Board Report and related materials (2-19-81)	(124.28)	
Mailing 27th NAIC Report and related materials, including printing of 1981 address directory (6-22-81)	(632.38)	
Verticillium wilt report (10-31-81)	(136.23)	
Notice 28th NAIC and request for papers (12-17-81)	(77.05)	
Information for 28th NAIC Registration and 1981 National Certified Alfalfa Variety Review Board Report and related materials (3-17-82)	(218.58)	
Program 28th NAIC	85.00	(1273.52)

NAIC payment on printing bill	622.37	
MN Alfalfa Project Debit (27th and 28th NAIC)	(865.70)	
Balance in account 6-23-82	157.12	
Balance on hand 6-23-82		(708.58)

Based on the current and projected situation I recommend the NAIC consider the following recommendations:

1. Establish a committee to study the NAIC financing and report recommendations for future funding to the 29th NAIC.

2. Restructure the position of permanent secretary to provide assistance in editing and publishing the Conference Report to make it more timely.

3. Change the title of Permanent Secretary to Secretary or corresponding secretary.

4. Appoint a committee to determine the interest and feasibility of an international alfalfa newsletter.

It has been a pleasure to work with Bill Melton, those responsible for hosting the 28th NAIC, and all of the membership that make the NAIC such an active organization. It is my observation that, "The alfalfa plant and the NAIC both share the necessary ingredients to make a highly effective symbiosis."

Respectfully submitted,

Don Barnes

DONALD K. BARNES, Secretary NAIC

NOTE: During the subsequent business meeting the first three recommendations were discussed and voted on. 1) It was agreed that a finance committee would be appointed by the 29th NAIC. 2) It was voted that a new office of vice-chairman be elected and that it would be the responsibility of the person in that position to edit the NAIC report. The vice-chairman will become chairman of the following conference. 3) The title of Permanent Secretary will be changed to Secretary.

Nominations Committee Report

The Nomination Committee presented the name of Robert "Bob" R. Kalton, Director of Research, Land O'Lakes, Webster City, IA for Chairman and the name of Bernie P. Goplen, Agriculture Canada, Saskatoon, Saskatchewan for Vice-Chairman. Drs. Kalton and Goplen were unanimously elected as Chairman and Vice-Chairman, respectively, of the 29th NAIC. The Executive Committee for the 28th NAIC served as the Nominating Committee.

The Executive Committee of the 28th NAIC recommended to the Association of Official Seed Certifying Agencies (AOSCA) that L.R. Teuber, assistant professor, University of California-Davis serve as alternate delegate from the NAIC on the National Certified Alfalfa Variety Review Board beginning July, 1982. He will become delegate on July 1, 1984.

Jim Moutray, Director of Research, North American Plant Breeders, Ames, IA was elected by the alfalfa industry members to serve as their representative on the Executive Committee of the 29th NAIC.

Committee on Location of 1984 National Alfalfa Improvement Conference

An invitation was received to hold the 1984 NAIC at Lethbridge, Alberta, Canada. The proposed dates are July 17, 18 and 19. M.R. (Mike) Hanna will chair the Local Arrangements Committee. The invitation was accepted.

Lethbridge is in an irrigation district where alfalfa is grown for hay, cubes, and seed production. The Agriculture Canada Research Station at Lethbridge conducts a number of research projects on alfalfa, covering seed production, breeding, utilization, diseases, and insect pests.

An invitation was received to hold the 1986 NAIC at the University of Georgia, with J.H. Bouton as Chairman of Local Arrangements.

The locations committee consisted of J.H. Bouton, R.S. Fulerson, M.S. Offutt, and R.E. Howarth, Chairman.

History of the National Alfalfa Improvement Conference

No.	Year	Location	Chairman	Secretary
1	1934	Lincoln, NE	T.A. Kiesselbach	H.M. Tysdal
2	1934	Washington, D.C.	A.J. Pieters	H.M. Tysdal
3	1935	St. Paul, MN	H.L. Westover	H.L. Westover
4	1936	Madison, WI	R.A. Brink	H.M. Tysdal
5	1937	Chicago, IL	R.A. Brink	H.L. Westover
6	1938	Manhattan, KS	H.M. Tysdal	H.L. Westover
7	1939	New Orleans, LA	H.M. Tysdal	H.L. Westover
8	1940	Fort Collins, CO	L.F. Graber	H.L. Westover
9	1942	St. Louis, MO	L.F. Graber	H.L. Westover
10	1946	Logan, UT	J.W. Carlson	H.M. Tysdal
11	1948	Lincoln, NE	C.O. Grandfield	H.M. Tysdal
12	1950	Lethbridge, Canada	T.M. Stevenson	O.S. Aamodt
13	1952	Raleigh, NC	R.P. Murphy	O.S. Aamodt
14	1954	Davis, CA	O.F. Smith	H.O. Graumann
15	1956	St. Paul, MN	C.P. Wilsie	H.O. Graumann
16	1958	Ithaca, NY	C.H. Hanson	H.O. Graumann
17	1960	Saskatoon, Canada	J.L. Bolton	C.H. Hanson
18	1962	Davis, CA	E.H. Stanford	C.H. Hanson
19	1964	Lafayette, IN	R.L. Davis	C.H. Hanson
20	1966	Univ. Park, PA	H.L. Carnahan	C.H. Hanson
21	1968	Reno, NV	W.R. Kehr	C.H. Hanson
22	1970	Urbana, IL	R.R. Hill, Jr.	C.H. Hanson
23	1972	Ottawa, Canada	D.H. Heirichs	C.H. Hanson
24	1974	Tucson, AZ	Dale Smith	C.H. Hanson and D.K. Barnes
25	1976	Ithaca, NY	M.W. Pedersen	D.K. Barnes
26	1978	Brookings, SD	M.D. Rumbaugh	D.K. Barnes
27	1980	Madison, WI	E.L. Sorensen	D.K. Barnes
28	1982	Davis, CA	B.A. Melton	D.K. Barnes

National Alfalfa Improvement Conference (NAIC)
Mailing List Questionnaire

Returning this questionnaire indicates that you would either like to be added to the NAIC mailing list or that you have an address or activity change.

1. Name: ____________________ 2. Date: ____________________

3. Mailing Address: __________ 4. Office Telephone No. __________

____

____

5. Present activities with alfalfa: Check appropriate blank(s):

Research Activities	Non-Research Activities
A ___ Breeding	J ___ Administration
B ___ Entomology	K ___ Extension
C ___ Nematology	L ___ Forage Producer
D ___ Pathology	M ___ Marketing
E ___ Physiology and Microbiology	N ___ Seed Producer
F ___ Forage Production	O ___ Student
G ___ Seed Production	P ___ Teacher
H ___ Utilization	Q ___ Certification and Variety Protection
I ___ Chemical and Quality Analysis	R ___ Writer or Publisher
	S ___ Other ____________________

6. Would you like new variety and germplasm release information?
Yes ____ No ____

For Canadian and USA Scientists Only:

7. Which Regional Alfalfa Improvement Conference(s) would you like to receive information about? _____ Eastern, _____ Central, _____ Western

8. What best describes your employment situation: ____ USDA, ____ SAES, ____ U.S. Private Industry, ____ Canadian Public, ____ Canadian Private

NOTE: Please call this questionnaire to the attention of your colleagues and employees who you think should be on the NAIC mailing list.

Return to: D.K. Barnes
Department of Agronomy and Plant Genetics
University of Minnesota
St. Paul, MN 55108