

## **Chapter 5.**

### **Cowpea (*Vigna unguiculata*)**

*This chapter deals with the biology of cowpea (Vigna unguiculata). It contains information for use during the risk/safety regulatory assessment of genetically engineered varieties intended to be grown in the environment (biosafety). It includes elements of taxonomy, centres of origin and distribution, crop production and cultivation practices, morphological characters, reproductive biology, genetics and genome mapping, species/subspecies hybridisation and introgression, interactions with other organisms, human health considerations, common pests and pathogens, and biotechnological developments.*

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## Introduction

Cowpea (*Vigna unguiculata* (L.) Walp.) is grown in tropical Africa, Asia, North and South America mostly as a grain, but also as a vegetable and fodder crop. It is favoured because of its wide adaptation and tolerance to several stresses. It is an important food source and is estimated to be the major protein source for more than 200 million people in sub-Saharan Africa and is in the top ten fresh vegetables in the People's Republic of China (hereafter "China").

In the English-speaking parts of Africa it is known as cowpea whereas in the Francophone regions of Africa, the name "niébé" is most often used. Local names for cowpea also include "seub" and "niao" in Senegal, "wake" or "bean" in Nigeria, and "luba hilu" in the Sudan. In the United States, it is typically referred to as blackeye beans, blackeye peas, crowder peas and southern peas. On the Indian subcontinent it is called "lobia" and in Brazil it is "caupi." In China it is called "long bean" or "asparagus bean".

## Species or taxonomic group

### *Classification and nomenclature*

Cowpea (*Vigna unguiculata* (L.) Walp.) belongs to the family *Fabaceae* (*Leguminosae* is also used as the family name with *Papilionoideae* as the subfamily), genus *Vigna*, and section *Catjang* (Verdcourt, 1970; Maréchal, Mascherpa and Stainier, 1978) (Table 5.1).

Table 5.1. Classification of cowpea (*Vigna unguiculata* (L.) Walp.)

Taxonomic placement	Scientific name
Kingdom	<i>Plantae</i>
Division	<i>Magnoliophyta</i>
Class	<i>Magnoliopsida</i>
Order	<i>Fabales</i>
Family	<i>Fabaceae</i>
Sub-family	<i>Faboideae</i>
Tribe	<i>Phaseoleae</i>
Sub-tribe	<i>Phaseolinae</i>
Genus	<i>Vigna</i>
Section	<i>Catjang</i>
Species	<i>unguiculata</i>
Botanical varieties	1. <i>Vigna unguiculata unguiculata</i> var. <i>unguiculata</i> 2. <i>Vigna unguiculata unguiculata</i> var. <i>spontanea</i>

Annual cowpea has two botanical varieties (Table 5.1), the cultivated *Vigna unguiculata unguiculata* var. *unguiculata* and the wild form *V. u. u.* var. *spontanea*, both of which are inbreeding. *V. u. u.* var. *spontanea* is typically found mostly near the borders of cultivated cowpea fields and within them.

Cultivated cowpeas have been divided into five cultivar groups based mainly on pod, seed and ovule characteristics (Pasquet, 1999; 1998) (Table 5.2).

*Unguiculata* is the largest cultivar group. The cultivar group *Sesquipedalis* (variously known as "asparagus bean", "yardlong bean", "long bean" or "snake bean") has more than 16 ovules and seeds spaced within the pod. Recent molecular evidence suggested that it is a subspecies (Xu et al., 2012; 2010).

Table 5.2. The five cultivar groups of cultivated cowpea

Cultivar group	Selected feature
unguiculata	Includes most African grain and forage types. More than 16 ovules/pod.
melanophthalmus	Blackeye pea types. Less than 17 ovules/pod. Grown mostly in the Americas.
biflora (Catiang)	Smooth seed in short erect pods. Common in India. Less than 17 ovules/pod.
sesquipedalis	Asparagus or yard-long beans. Very long pods consumed fresh, especially in the People's Republic of China.
textilis	Rare form with very long peduncles once used for fibre in Africa.

The wild cowpeas in the subspecies *unguiculata* currently are described as being the variety *spontanea* (previously included in the subspecies *dekindtiana*, i.e. in Padulosi [1993]). Var. *spontanea* are similar to domesticated cowpea landraces except that the pods are small and dehiscent, and the seeds are ten times smaller than cultivated cowpea. The seed coat of *spontanea* is hard, thick and impermeable to water. There are no obvious barriers to hybridisation or recombination between members of these five different cultivar groups or with the wild cowpeas (var. *spontanea*) in the subspecies *unguiculata*.

The *Vigna unguiculata* species complex is currently divided into 11 subspecies (Padulosi, 1993; Padulosi and Ng, 1997; Pasquet, 1997, 1993a, 1993b). Ten of the subspecies are perennial and one, cowpea, is annual (Table 5.3). Plants from these subspecies have exhibited varying degrees of crossability with cultivated cowpea. Note that another taxon, *Vigna monantha* Thulin from coastal Somalia, may warrant reclassification as a new *Vigna unguiculata* subspecies.

Table 5.3. The *Vigna unguiculata* (L.) Walp. subspecies complex

Subspecies	Perennial	Annual	Habitat
<i>aduensis</i> <sup>2</sup>	Yes		Montane forest areas in Ethiopia north of the Blue Nile (altitude 1 400-2 600 m).
<i>alba</i> <sup>1</sup>	Yes		In the coastal plains from SãoTomé and Gabon to north-western Angola.
<i>baoulensis</i> <sup>2</sup>	Yes		West African rain forest area, from Sierra Leone to eastern Cameroon.
<i>burundiensis</i> <sup>2</sup>	Yes		Mainly found in forest margins, gallery forest margins or cleared grasslands in the subhumid and humid zones in Burundi, Uganda and the Kakamega forest in western Kenya.
<i>dekindtiana</i> <sup>1</sup>	Yes		In semi-arid zones with a disjunct distribution in the mountains from southern Angola and Zimbabwe, and a few specimens observed in northwest Zambia (altitude 1 400-1 900 m) and possibly in West Africa.
<i>letouzeyi</i> <sup>2</sup>	Yes		The Congolese basin rainforest from Cameroon and Gabon to the border of the Democratic Republic of the Congo with Uganda.
<i>pawekiae</i> <sup>2</sup>	Yes		Montane forest of eastern Zimbabwe to south-western Ethiopia through Malawi, eastern Tanzania, Ngorongoro and the major Kenyan mountains. Also observed in the mountains east of Lake Tanganyika (altitude 1 400-2 600 m).
<i>pubescens</i> <sup>1</sup>	Yes		In the coastal Indian Ocean plain from Maputo to Kenya. (A few specimens have also been collected in swamps in Burundi, southern Sudan, south-western Tanzania and Uganda).
<i>stenophylla</i> <sup>1</sup>	Yes		Complex distribution where pubescent forms (var. <i>protracta</i> (E. Mey.) Mithen) are in the back of the coastal sand dunes in eastern Cape Province, at higher elevation from Transkei northward, on the eastern slopes of the Drakensberg at 500-1 500 m elevation, in Swaziland and east of Mpumalanga and Northern Province. Narrow leaflet forms (var. <i>stenophylla</i> (Harv.) Mithen) occur at low elevations in north-eastern Natal, Swaziland and Kruger Park plain, and at 1 200-1 500 m elevation in the high veld of West Mpumalanga, Gauteng and the northern part of Free State. Scabrous lobed-leaflet forms (var. <i>kgalagadiensis</i> Mithen) found in north-eastern Namibia, Botswana, Zambian Barotseland and north-western Zimbabwe.
<i>tenuis</i> <sup>1</sup>	Yes		In two different areas: Zambia-Zimbabwe-Malawi at 1 200-1 800 m and in a coastal area from southern Natal to mid-Mozambique.
<i>unguiculata</i> <sup>1</sup>		Yes	Widely cultivated especially in West Africa (see Figure 5.3).

Notes: 1. Most cultivated cowpeas and the subspecies *alba*, *dekindtiana*, *pubescens*, *stenophylla* and *tenuis* (and var. *spontanea*) are highly self-pollinated. Previously, these subspecies were pooled into the subspecies *dekindtiana* and it is convenient here to call these wild cowpea subspecies the “*dekindtiana* group”. 2. The subspecies *aduensis*, *baoulensis*, *burundiensis*, *letouzeyi* and *pawekiae* are all out-crossing. Previously, these subspecies were pooled into the subspecies *mensensis* and they are described here as the “*mensensis* group”.

### ***Description of the plant***

The cowpea *Vigna unguiculata* (L.) Walp. is an annual herbaceous legume cultivated for its edible seeds or for fodder. Cultivated cowpeas are herbaceous annuals that are either erect, prostrate or climbing annuals with a tap root and virtually all are glabrous. They are mostly grown for grain but a small proportion (about 10%) are grown as green leafy vegetables and fodder in Africa or as fresh pods in eastern Asia (Boukar et al., 2015).

Cowpea *V. unguiculata* can grow up to 80 cm and up to 2 m for climbing cultivars. It has a well-developed root system. Germination is epigeal with the first pair of true leaves being simple and opposite and subsequent leaves being trifoliate with oval leaflets (6-15 cm long and 4-11 cm broad) and alternate. The papilionaceous flowers are born on racemose inflorescences at the ends of peduncles that arise from leaf axils and can be white, yellowish, pale blue or violet. Peduncles are stout and grooved and usually much longer than the leaves (2-20 cm long). For each inflorescence, flowers are sequentially produced in alternating pairs on thickened nodes at the tip with cushion-like extra-floral nectaries between each pair of flowers. The flower is large (standard is 2-3 cm in diameter), with a straight keel, diadelphous stamens (one free and nine fused), a sessile ovary with many ovules, and a style that is bearded along the inside and ends in an oblique stigma. Pods occur in pairs forming a V, mostly pending and vertical, but they can be erect. They are cylindrical, 2-6 cm long and 3-12 mm broad and contain 8-20 seeds. Seeds can be white, pink brown or black (Heuzé et al, 2013) (Figure 5.1).

Figure 5.1. **Aerial parts of cowpea (*Vigna unguiculata* (L.) Walp.)**

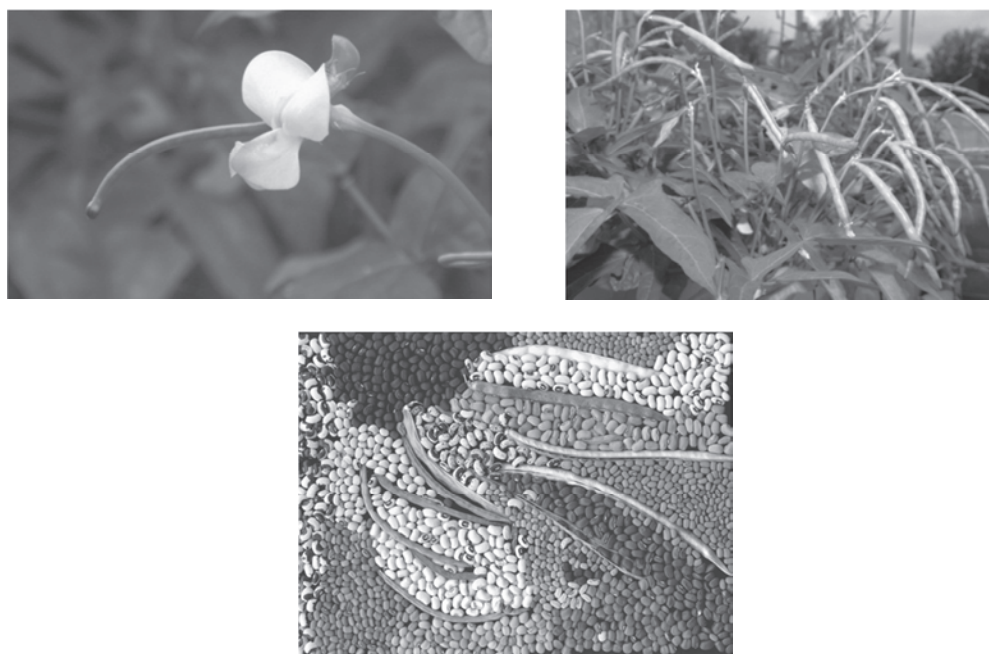


*Note:* This line drawing shows leaves, stems, petioles, flowers and pods (main image), together with the reproductive organs consisting of stamens (nine fused and one free) and pistil with its curved style with brush below the stigma (bottom left) and parts of the corolla (bottom right); the standard (top), two wings (middle) and keel (bottom).

*Source:* Steward (1958), digitized by BHL wiki and licensed under CC BY-NY-SA 4.0.

The corolla is yellowish-white to violetish-white with violet wings and mature seed colours vary from white through brown to black (Figure 5.2).

Figure 5.2. Cultivated cowpea flower, pods and seeds



*Note:* Picture of cowpea flower (top left), immature green pod (top right), maturing pods with an illustration of the great variety of seed colours (bottom).

*Source:* Courtesy Carl Davies, CSIRO.

Cultivated cowpeas are mostly indeterminate and some have the potential to produce multiple flushes of flowers (Gwathmey, Hall and Madore, 1992) that live for less than one year. The wild relatives of cowpeas, which are perennial (Table 5.3), have fleshy roots and the capacity to resprout after a dry or cool season.

## Geographic distribution, habitats, crop production, centres of origin and diversity

### *Geographic distribution*

Cultivated cowpeas are grown as warm-season-adapted annuals in tropical and subtropical zones (as defined by Hall [2001]) in all countries in sub-Saharan Africa and in Asia, South America, Central America, the Caribbean, the United States and around the Mediterranean Sea. In subtropical zones temperatures are only suitable for cowpea in the summer, whereas temperatures are suitable year-round in tropical zones. The vast majority of the world's cowpea production (over 95%) takes place in sub-Saharan Africa (Figure 5.3), with about 12.5 million hectares under cultivation worldwide in 2014 (Singh et al., 2002; FAOSTAT, 2014) (Table 5.4). Asia is the second largest producing region, representing less than 3% of the global production in average over the 1993-2014 period (Figure 5.3), most of it being cropped in Myanmar (FAOSTAT, 2014).

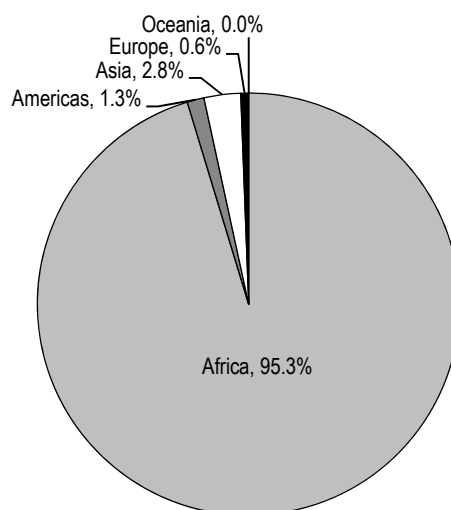
In Africa, cowpea can be cultivated up to 1 800 m altitude but is mainly grown in the lowlands. The centre of maximum diversity of cultivated cowpeas and land races is found in West Africa in a region comprising the Sudan savannah zone of Nigeria (at 4 million ha, Nigeria has the largest area of cowpea cultivation according to FAOSTAT),

Table 5.4. Global production of cowpeas (dry) in million metric tonnes (MMT)

Cowpea production	Average 1993-2014	2010	2011	2012	2013	2014
<b>World</b>	<b>4.59</b>	<b>6.91</b>	<b>4.78</b>	<b>8.25</b>	<b>8.03</b>	<b>5.59</b>
Africa	4.37	6.57	4.50	7.95	7.78	5.35
<i>including – Nigeria</i>	2.53	3.37	1.64	5.15	4.63	2.14
– Niger	0.79	1.77	1.52	1.33	1.63	1.59
– Burkina Faso	0.37	0.63	0.44	0.60	0.58	0.57
– Tanzania	0.13	0.15	0.17	0.18	0.19	0.19
– Cameroon	0.10	0.15	0.16	0.15	0.17	0.17
– Mali	0.10	0.13	0.13	0.14	0.17	0.15
– Kenya	0.07	0.07	0.08	0.11	0.13	0.14
Asia	0.13	0.23	0.19	0.19	0.15	0.15
<i>including – Myanmar</i>	0.11	0.21	0.17	0.16	0.12	0.12
Americas	0.06	0.09	0.07	0.09	0.08	0.07
Europe	0.03	0.03	0.03	0.02	0.02	0.02
Oceania	0.00	0.00	0.00	0.00	0.00	0.00

Source: FAOSTAT (2014).

Figure 5.3. Cowpea production share by region, average, 1993-2014



Source: FAOSTAT (2014).

central Burkina Faso, Ghana, Togo, northern Benin and the north-western part of Cameroon (Padulosi and Ng, 1997). Substantial cowpea cultivation also occurs in the semi-arid Sahelian zone, which is a transition zone between the Sahara desert in the north and the Sudan savannah zone in the south. The Sahel encompasses northern and central Senegal and southern Mauritania in the west to central Sudan in the east, passing through central Mali, northern Burkina Faso, southern Niger (at 5 million ha, Niger has the largest area of cowpea cultivation) and central Chad. Significant cowpea production also occurs in the northern Guinea savannah zone and the forest and southern Guinea savannah zones of West Africa, the United Republic of Tanzania and Uganda, and some cowpeas are cultivated in central, southern and north-eastern Africa. Many areas where cultivated

cowpeas are grown and the locations where the wild cowpea *V. unguiculata* var. *spontanea* has been found are shown in Figure 5.4.

Figure 5.4. **Distribution of cultivated and wild cowpeas in Africa**



*Note:* Areas with cultivated cowpea are shown in grey, while the black dots indicate the locations where wild cowpea *V. unguiculata* var. *spontanea* occurs.

*Source:* Adapted from Remy Pasquet.

The wild relatives of cowpea are widely distributed across sub-Saharan Africa (Figure 5.5). They occupy a range of habitats (described in Table 5.3) to an elevation of 2 600 m. *Vigna monantha* has been found in Somalia in the coastal plain from Hobyo to Bender Bayla.

Figure 5.5. **Distribution of the wild relatives of cowpea in Africa**



*Source:* Adapted from Pasquet (1996).

In Asia, cowpea (“asparagus bean”) ranks as one of the top ten fresh vegetables. It is cultivated across a broad geographic range, except for some permanently cold regions. According to the FAO statistics, Myanmar is the main cowpea producer in Asia (FAOSTAT, 2014). China, India, Japan, Korea and Thailand are among the major asparagus bean-producing countries. The estimated annual cultivation area in Asia in total



is 1 million ha, China alone making up roughly one-fifth of the world's fresh pods production with over 1.5 million tonnes (equivalent to an additional 0.2 MMT of dry matter). Compared with the African cowpea, "asparagus bean" is more adapted to cool climates and is less tolerant to very high temperatures.

### *Ecosystems and habitats of native and naturalised cowpea*

Cowpeas and their wild relatives have persisted for thousands of years in sub-Saharan Africa with many occurring in West Africa and southern Africa. While some wild relatives are persistent from year to year due to their fleshy roots and ability to resprout after a dry or cool season, most wild relatives persist through the production of hard seed that can remain viable for several years in the soil.

The wild cowpeas *V. unguiculata* var. *spontanea* clearly benefit from human disturbance as shown in the following examples from the Africa region. In the Milalani wild population in coastal Kenya, the population has increased after each mechanical clearing of the roadsides. In a long-term seed-supplementation trial in Muhaka field station in Kenya, the plots that were ploughed every year had more wild cowpea plants than the undisturbed plots (R.S. Pasquet, personal communication). While *Vigna unguiculata* var. *spontanea* can be found in natural ecosystems from Cameroon eastward with clear examples in eastern Cameroon, Uganda and the western Ethiopian lowlands, it seems only to be found in disturbed places (fields, field margins, roadsides and fallows) in Burkina Faso, western Niger and northern Ghana. In the West African Sahel, cowpea is also widely cultivated for fodder. For farmers mainly focusing on fodder, fodder from wild cowpea (as well as domesticated-wild F<sub>1</sub> hybrids and their progenies) may be considered as being equivalent to fodder from domesticated cowpea. Often wild cowpea plants are not uprooted from the field, and appear to be tolerated in the agro ecosystem. The hybrid progenies may even end up being used by farmers for sowing and may be considered as fodder landraces. Wild cowpeas and wild relatives of cowpea do not appear to represent a significant weed problem in sub-Saharan Africa (Huesing et al., 2011).

Those few cowpea landraces that produce some hard seeds that can survive for several years in the soil may have a tendency to persist in and around cultivated fields. Domesticated cowpea can theoretically survive as feral plants, as was shown for example in Japan (Berville et al., 2005). However, this rarely has been observed in Africa; for example, a few small feral populations observed in coastal Kenya were not seen in consecutive years.

### *Centres of origin and diversity*

Several hypotheses have been proposed for the domestication of cowpea in different parts of sub-Saharan Africa (summarised in Ba, Pasquet and Gepts, 2004). It is likely that cowpea was domesticated only once, probably in West Africa about 2000 B.C. (Padulosi and Ng, 1997), and that the progenitor of cultivated cowpea was the wild cowpea *V. unguiculata* var. *spontanea* (Pasquet, 1999). In West Africa, where most of the world's cowpea is cultivated, there are many weedy forms that are intermediates between truly wild forms and very small-seeded cultivated cowpeas (Rawal, 1975). Recent molecular evidence shows that the "asparagus bean" has undergone a severe genetic bottleneck during domestication in Asia from its African progenitors (Fang et al., 2007; Xu et al., 2010).

The greatest genetic diversity in wild relatives of cowpea has been found in southern Africa in a region encompassing Namibia from the west, across Botswana, Zambia,



Zimbabwe and Mozambique to the east, and South Africa and Swaziland to the south (Padulosi and Ng, 1977). This genetic diversity includes many primitive traits that were lost in domestication such as perenniality, hairiness, small size of seeds and pods, hard seeds, pod shattering and outbreeding. Cultivated cowpeas also are present in this region. The South African Transvaal may have been the centre of speciation of *Vigna unguiculata* due to the presence there of the most primitive subspecies (Padulosi and Ng, 1977).

### *Crop production and management practices*

#### Africa

Most cowpea grown in the African region is intercropped with sorghum (*Sorghum bicolor*) or pearl millet (*Pennisetum glaucum*), and sometimes with other crops such as maize (*Zea mays*), cassava (*Manihot esculenta*) or cotton (*Gossypium* spp.) (Blade et al., 1996). The crop is typically planted at wide spacing (1 m) irregularly through young stands of the component cereal or other crop. Because the cowpea is planted after cereal crop establishment, at low density and without inputs, dry grain cowpea yields in the range of 300 kg/ha only are typically achieved in such systems. In Senegal, most of the cowpea production is sole-cropped (Thiaw, Hall and Parker, 1993), in part due to the light sandy soils and availability of horse-drawn peanut seed drill which can easily be modified to plant cowpea in rows, making possible animal-draft cultivation to control weeds. In the last decade, an increasing portion of the cowpea crop in other parts of Africa has been planted in pure stand, at relatively higher density, using improved varieties and with agricultural inputs, especially insecticides, resulting in average yields of between 1-2 tonnes/ha. Strong demand for cowpea-based foods in urban areas and good prices are driving this transition to more intensified production practices.

Figure 5.6. Cowpea field, Shawula district, Swaziland



Source: Courtesy EcoPort ([www.ecoport.org](http://www.ecoport.org)). Author Roger P. Ellis.

Cowpea is a legume species usually considered as being resistant to droughts. Droughts often occur in the Sahelian zone and Sudan savannah zones (Dancette and Hall, 1979). Cowpea has a greater ability to withstand these droughts and to produce significant grain than any other crop grown, including the drought-resistant crops pearl

millet, sorghum and peanut. In addition, cowpea hay is an important source of forage for livestock, which plays a particularly critical role in feeding animals during the dry season in many parts of West Africa (Singh and Tarawali, 1997; Tarawali et al., 2002, 1997).

Figure 5.7. Cowpea (*Vigna unguiculata* (L.) Walp.) straw as feed for cattle



Source: IITA Image Library, licenced under CC BY 3.0.

### Other regions of the world

In Asia and Brazil, both sole-cropping and intercropping are practiced (Pandey and Ngarm, 1985; Watt, Kueneman, and de Araújo, 1985), while in the United States generally only sole-crops are grown. In Brazil and India, some intercropping of cowpea is still practiced, but the majority of the crop is produced under sole-cropping with inputs. Cowpea production in the United States is entirely mechanised with machinery and agronomic practices adapted from other crops such as common beans or soybeans. Large growers in Brazil have adopted similar modern farming practices to produce high yields (Freire Filho et al., 2011).

In China, “asparagus bean”, as a vegetable, is usually intercropped with common bean or cucumber. Smallholder farming and hand-harvest of the immature fresh pods of asparagus bean still remains the dominant production system in China, as pod quality/appearance, rather than yield, is usually more important.

## Reproductive biology

### *Generation time and cropping season duration*

#### *Domestic cowpeas*

Domesticated cowpeas are annuals with duration from sowing to harvest varying from two to six months. Cowpeas are grown as a rainfed crop and the dates of sowing and maturity must fit the timing of the rainfall and the hydrologic budget (Dancette and Hall, 1979). Cultivars vary in their responses to photoperiod and temperature as they influence the time of budding and flowering. A classification of these responses by Ehlers and Hall (1996) includes three photoperiod classes (day-neutral, quantitative short-day and obligate short-day), three juvenility classes (short, intermediate and long), three classes of

heat-induced floral bud suppression (no bud suppression, partial and complete bud suppression) and two classes of pod-setting ability under hot long days (low and high). Semi-arid, subhumid and humid zones are considered as they were defined by Hall (2001).

In the semi-arid Sahelian zone of Africa, where the growing season usually is very short due to a short rainy season, adapted cowpea cultivars include:

- erect day-neutral ones with a short juvenile period that have a cycle length of 60 days
- spreading day-neutral ones with a slightly longer juvenile period that have a cycle length of 70 days (Hall, 2004)
- dual-purpose, spreading, short-day ones with a longer cycle of about 90 days for producing hay and grain.

Note that day-neutral cultivars have a fairly constant cycle length because time of flowering is not influenced by photoperiod, but is rather influenced by temperature which is relatively constant in tropical zones.

In the wetter semi-arid Sahelian and subhumid Sudan savannah zones to the south, adapted cowpea cultivars include ones with different types of short-day requirements for flowering. The beginning of the rainy season, which determines the time of sowing, can be much more variable than the end of the rainy season, which determines the optimum time for harvest. Adapted cowpea cultivars with an appropriate short-day requirement reach maturity at the optimum time for harvest even with substantial variation in sowing date. Thus, these cultivars have a variable cycle length depending on the date of sowing.

Further south in the wetter subhumid Sudan and humid Guinea savannah zones, cowpea cultivars may be found that are day-neutral but have a long cycle length due to a long juvenile period (Lush, Evans and Wien, 1980).

Most Chinese “asparagus bean” cultivars are day-neutral or weakly short-day.

### *Wild relatives of cowpea*

With respect to the wild relatives of cowpea, members of the *dekindtiana* group that are adapted to the Sudan savannah zone were observed to be obligate short-day plants (Lush, Evans and Wien, 1980). Members of *V. unguiculata* var. *spontanae* also are short-day plants. In contrast, members of the *mensensis* group, which are adapted to the more humid forest and southern Guinea savannah zones, were observed to be day-neutral with a long juvenile period (Lush, Evans and Wien, 1980). In areas of East Africa where there is a bimodal rainy season, wild relatives of cowpea have been observed to have a cycle length of one to two years. They germinate during the beginning of one rainy season and produce fruits during this rainy season, and then survive the dry season using carbohydrate reserves in the fleshy roots and grow again at the commencement of the next rainy season producing more fruits and then survive the dry season. These wild relatives of cowpea are presumed to be day-neutral in their flowering behaviour.

### **Reproduction characteristics**

#### *Pollen dispersion*

There is no mechanical dispersion of pollen from the flowers of cultivated cowpeas because the anthers release pollen during the first half of the night when the flowers are

still closed (Ladeinde and Bliss, 1977), and the pollen is sticky and heavy. The cuticle which protects the stigmatic surface breaks and releases a stigmatic exudate during the second half of the night at which time self-fertilisation can begin. Subsequently, the flower opens during the early morning and then closes in the late morning.

### *Pollination characteristics*

In general, cultivated cowpeas have a high level of self-pollination. Based on their work in Texas, Blackhurst and Miller (1980) noted that the pollination process in cultivated cowpeas is complete before the flower opens. However, once they have begun flowering, cultivated cowpeas, wild cowpeas and wild relatives have the ability to produce flowers every day for several weeks (Gwathmey, Hall and Madore, 1992). Consequently, some opportunities for cross-pollination occur providing pollinators are present. Outcrossing in limited amount has been observed and quantified in literature. Fatokun and Ng (2007) report it at two locations in Nigeria and one location in Benin, and in one case pollen travelled up to 31 m between parental plants. The authors concluded that outcrossing occurred at a frequency of less than 1%. In Senegal, outcrossing rates at 2% have been observed. In the south-eastern United States, outcrossing of 0-1.4% was observed with six cultivars (Williams and Chambliss, 1980). Some non-quantitative observations have also been made. Significant outcrossing has been observed in cowpea fields that are next to wild lands in Botswana. In California, some cowpea cultivars have exhibited a few percent outcrossing in some locations.

Cross-pollination is usually less than 1%, but will vary somewhat with the cultivar and, more particularly, with the population of some insects. In several cases, the pollinators are not known, but honeybees (*Apis mellifera*) have been observed around cowpea flowers and thus have been implicated in pollination (Ige, Olotuah and Akerele, 2011). Purselove (1968) reported that the extra-floral nectaries at the base of the corolla attract ants, flies and bees, but noted that a heavy insect would be required to depress the wings of the flower and expose the stamens and stigma (tripping). In coastal Kenya and Burkina Faso, several large carpenter bee species (*Xylocopa* spp.) and leafcutter bee species (*Megachilidae* spp.) were considered potential cross-pollinators of cowpea (R.S. Pasquet, personal communication), and it was shown that these same leafcutter and carpenter bees were the likely pollinators of the wild progenitor of cowpea (Kouam et al., 2012). Casual observations made in California and Texas (United States) and Nigeria indicate that large bumblebees (*Bombus* spp.) may be responsible for the cross-pollination that occurs in cowpeas in these regions.

Inter-specific crossing between wild and cultivated cowpeas are rare (see the description under the section “Species/subspecies hybridisation and introgression” on the next page).

### *Seed viability*

Cultivars of domesticated cowpeas usually do not create long-lived seed banks in the soil because their seed coats typically are permeable to water and the seeds have little dormancy (Lush, Evans and Wien, 1980). Some land races and cultivars with smooth seed coats can have some hard seeds.

Wild cowpeas and relatives of cowpea have dormant seeds due to the impermeable nature of their seed coats (Lush and Evans, 1980). These hard seeds can survive for several years in the soil, especially if the soil is dry.

## Genetics and genome mapping

Cowpea is a diploid with  $2n = 2x = 22$  chromosomes, one of which is short (19  $\mu\text{m}$ ), 7 are medium length (26-36  $\mu\text{m}$ ) and 3 are long (41-45  $\mu\text{m}$ ) (Frahm-Leliveld 1965; Mukherjee 1968). The genome size is about 613 Mb (Arumuganathan and Earle, 1991). Chloroplasts are maternally inherited (Corriveau and Coleman, 1988). The wild subspecies also are diploid with  $2n = 22$  (Vikal and Satija, 1992; Venora and Padulosi, 1997; Adetula, 2006).

Much progress has been made recently in developing genetic maps of cowpea using a range of methods: restriction fragment length polymorphism (RFLP), random amplified polymorphic DNA (RAPD), amplified fragment length polymorphism (AFLP), genomic scar markers (SCAR), simple sequence repeat (SSR), single-nucleotide polymorphism (SNP) and phenotypic markers (Timko, Ehlers and Roberts, 2007; Andargie et al., 2011; Lucas et al., 2011) together with information on genome organisation (see the Cowpea Genomic Initiative developed by the Department of Biology of the University of Virginia at: <http://cowpeagenomics.med.virginia.edu>).

Of note is the recent construction of a high-density cowpea consensus genetic map based on SNP markers together with information on genome organisation (Muchero et al., 2009; Lucas et al., 2011). An SNP-based genetic map has also been constructed for asparagus bean (Xu et al., 2011).

Based on these platforms, quantitative trait loci (QTLs) governing many agricultural and adaptive traits such as leaf morphology, foliar thrips resistance and drought tolerance, have been mapped (Muchero, Ehlers and Roberts, 2010a, 2010b; Muchero et al., 2009; Pottorff et al., 2012). A high quality bacterial artificial chromosome- (BAC-) based physical map is also available for cowpea (790 contigs and 2 535 singletons), and the genome assembly of cowpea is underway (Close et al., 2011).

## Species/subspecies hybridisation and introgression

### *Natural interspecific crossing (extent, sterility/fertility)*

Floral morphology favours either autogamy (self-pollination) or allogamy (outcrossing) in different groups of the *V. unguiculata* species complex. Most cultivated cowpeas and members of the *dekindtiana* group are highly self-pollinating in that their anthers usually are in contact with their stigmatic surface. The *mensis* group of subspecies exhibits high levels of outcrossing and has anthers that are a few millimetres below the stigmatic surface, with the stigmatic surface oriented upwards and its lower part protected by a beard of long hairs (Lush, 1979).

To date, no successful natural or artificial crosses have been reported and subsequently confirmed between any member of the *Vigna unguiculata* species complex and any other species. Although *Vigna schlechteri* and *Vigna vexillata* are the closest species to *Vigna unguiculata*, numerous attempts to cross either of these species with *V. unguiculata* have failed (Mithen, 1989; Barone, Del Giudice and Ng, 1992; Fatokun, 2002; Fatokun, Perrino and Ng, 1997).

Wild cowpeas in the *mensis* group with floral morphologies that favour outcrossing function differently than the cultivated cowpea. If their flowers are not tripped by a heavy bee, they may remain open until late into the afternoon (Lush, 1979) and can eventually reopen the following morning. This wild cowpea group has much higher levels of cross-pollination than cultivated cowpeas, but does not readily cross with

cultivated cowpea. Studies have been conducted in coastal Kenya with cultivated cowpea and a wild cowpea *V. unguiculata* var. *spontanea* that had an outcrossing floral morphology. The level of outcrossing was less than 2%. Cultivated cowpeas readily cross with wild cowpeas in the same subspecies (i.e. var. *spontanea*) and can be crossed with members of the other subspecies of *Vigna unguiculata* but with varying degrees of difficulty.

### ***Experimental crosses***

The subspecies from the *mensis* group are not readily crossed with cultivated cowpea although it is possible, while some subspecies from the *dekindtiana* group are more easily crossed with cultivated cowpea (Sakupwanya, Mithen and Matangandura-Mhlanga, 1989; Kouadio et al., 2007, 2006). Breeders working with the subspecies *dekindtiana* have obtained many viable progeny after a simple hybridisation with cultivated cowpeas. In contrast, with plants from the subspecies *pubescens*, they have found it useful to backcross the F<sub>1</sub> with a parent because most of the F<sub>1</sub> seed were shrivelled and had low levels of germination and emergence. Crossability of plants from the subspecies *temuis* with cultivated cowpeas has been found to be intermediate in ease between *dekindtiana* and *pubescens*.

The overall message is that crosses appear possible among all members of the *Vigna unguiculata* complex but they vary from being easy to being difficult.

### ***Information and data on introgression***

A very high frequency of progeny from naturally formed interspecific hybrids between wild and cultivated cowpeas would have one or more domestication traits that significantly reduce their persistence in wild ecosystems. However, as feral wild x cultivated plants are sometimes used for forage by farmers, it is likely that hybridisation between such plants and wild cowpeas will occur and that the progeny would have an essentially wild phenotype with high survival potential in natural ecosystems.

## **General interactions with other organisms (ecology)**

### ***Potential positive effect of cowpea on cereal production***

Cultivated cowpeas play a critical role in the cereal-based intercropped and rotational cropping systems where they are often grown in sub-Saharan Africa, in terms of nutrient improvement and resistance to certain pests.

Cultivated cowpeas have symbiotic relations with rhizobia (Elowad and Hall, 1987) and mycorrhizae (Kwapata and Hall, 1985) that enhance the flow of reduced nitrogen and phosphate into the cropping system. These nutrients frequently limit the productivity of cereals in sub-Saharan Africa, and associated legumes can bring a beneficial effect.

Certain cowpea genotypes can cause suicidal germination of the seeds of the weed parasite *Striga hermonthica*, which is a major pest of pearl millet, sorghum and maize that has been difficult to solve by other means (Singh and Matsui, 2002). Some cowpea genotypes can reduce the reproduction of certain plant parasitic nematodes (including *Scutellonema cavenssi*) that can damage pearl millet, sorghum and peanut (Germani, Baujard and Luc, 1984; Hall et al., 2003).

Consequently, cowpea can enhance the edaphic conditions and thus the productivity of the cereals and other crops that are grown in rotation or as intercrops with it. An increase in the area of cowpea cultivation over present levels in sub-Saharan Africa

would not only benefit cereal productivity but also livestock production, whole farming systems and human nutrition and welfare.

### ***Pests and diseases***

Cowpeas are host to a range of pests and diseases such as insects and mites, viruses, fungal and bacterial diseases, nematodes and parasitic weeds. These may affect the whole plant, the flower or the pod and are detailed in Annex 5.A1, together with information on plant resistance and methods for pest control and management. The pests of major economic importance are *Maruca vitrata*, *Aphis craccivora*, *Clavigralla tomentosicollis*, *Megalurothrips sjostedti* and *Callosobruchus maculatus*.

### **Human health and biosafety**

Like other grain legumes, cowpeas contain a range of anti-nutritional factors such as hemagglutinin, tannin, trypsin inhibitors, oxalate, phytate, polyphenols and oligosaccharides (Sreerama et al., 2012; Afiukwa et al., 2012). The levels of anti-nutritional factors in cowpea are similar to those in the widely consumed food legume, chickpea (see Table 5.5).

Table 5.5. Anti-nutritional factors in the grain of chickpea and cowpea

Anti-nutritional factor	Chickpea	Cowpea
Phytic acid (mg/g)	12.1	14.0
Polyphenols (mg GA/g)	10.8	12.1
Oligosaccharides (mg/g)	34.9	31.7
Raffinose	8.6	10.3
Stachyose	19.1	17.8
Verbascose	7.2	3.6
Trypsin inhibitor activity (Units/g)	6 452	6 981
Trypsin inhibitor activity [IC <sub>50</sub> (µg/ml)]	44.8	38.2

Source: Adapted from Sreerama et al. (2012).

Cowpea grains complement the grains of cereals as foods for people by enhancing the quantities and qualities of proteins and vitamins. For example, cowpea grains have substantial levels of folic acid, which is a critical vitamin for all people and especially pregnant women since it prevents the occurrence of neural tube defects such as spina bifida in infants. Fresh and dry grains of early season cowpea cultivars and fresh pods and leaves are often an important source of food during the “hungry period” occurring two months prior to the main cereal harvest in the Sahelian and savannah zones (Dancette and Hall, 1979). Cowpea is a staple crop having a greater ability to withstand these droughts and to produce significant grain than any other agricultural plant grown in these zones, including the drought-resistant grain crops pearl millet, sorghum and peanut (Turk, Hall and Asbell, 1980; Ziska and Hall, 1983; Petrie and Hall, 1992; Singh and Matsui, 2002; Hall, 2004).

The grain is the most important part of the cowpea plant for human consumption. The seeds are most often harvested and dried for storage and consumption at a later time, either after cooking whole or after being milled like a flour product and used in various recipes (Nielsen, Ohler and C. Mitchell, 1997; Ahenkora, Adu Dapaah and Agyemang, 1998). As such, cowpea plays a critical role in the lives of millions of people in the developing world, providing them a major source of dietary protein that nutritionally



complements low-protein cereal and tuber crop staples. The nutritional profile of cowpea grain is similar to that of other pulses, with a relatively low fat content and a total protein content that is two- to fourfold higher than cereal and tuber crops. Similar to other pulses, the storage proteins in cowpea seeds are rich in the amino acids lysine and tryptophan when compared to cereal grains, but low in methionine and cysteine when compared to animal proteins. Total seed protein content ranges from 23% to 32% of seed weight (Nielsen, Brandt and Singh, 1993; Hall et al., 2003; Boukar et al., 2011).

In the south-eastern parts of the United States, portions of West Africa, Asia, and in the Caribbean, consuming fresh seeds and green pods is preferred to the cooked dry seeds (Nielsen, Ohler and C. Mitchell, 1997; Ahenkora, Adu Dapaah and Agyemang, 1998). In many parts of Africa and Asia, in addition to the seeds, the fresh or dried leaves are also consumed as a side dish or as part of a stew and provide significant nutritional value. In addition to human consumption, cowpea leaves and stems (stover) are also an important source of high-quality hay for livestock feed (Tarawali et al., 2002; 1997). Fresh pods of asparagus bean provide people in Asia with a source of energy protein, multiple vitamins and minerals.

## *Annex 5.A1.*

### **Common pests and pathogens**

#### **Cowpea pests and economic consequences**

There are many pests and diseases of cowpea (Table 5.A1.1) although insects tend to be the most economically important. There are good levels of host plant resistance for many of these pests in the cowpea germplasm, and it is being successfully deployed by the cowpea breeders.

However, there are several important pests for which strong cultivar resistance is not available in the primary gene pool. These are flower thrips (*Megalurothrips sjostedti*), pod-sucking bugs (*Clavigralla tomentosicollis*) and the podborer (*Maruca vitrata*) (Jackai and Daoust, 1986; Jackai and Adalla, 1997; Dreyer, Baumgärtner and Tamò, 1994). About two to three sprays of insecticide are needed to prevent significant economic losses by: 1) flower thrips reducing flower production; 2) pod-sucking bugs reducing pod and seed development; and 3) podborers damaging peduncles, floral buds, flowers, green pods and developing grain. Most African farmers do not apply insecticides to cowpea and as a consequence grain yields are 10-20% of what might be obtained with a complete spraying regimen (Jackai and Adalla, 1997).

Cultivated cowpea flowers are also visited by forage bees. In Africa, several bees have been observed on cowpea flowers (Table 5.A1.3) (Pasquet et al., 2008; Asiwe, 2009; Ige, Olotuah and Akerele, 2011).

#### **Podborer**

Many scientists consider the podborer to be the most damaging and economically important insect pest of cowpea in sub-Saharan Africa except for in the Sahelian zone, where it rarely occurs. In reviewing the biology of the podborer, Singh and Jackai (1985) noted that the female moth lays up to 200 eggs on flower buds, flowers and tender leaves of cowpea. Eggs hatch in two to three days, and there are five larval instars. Larval development takes about 8-14 days. The late larval instars can be identified by the black dots on their body. A two-day prepupal period follows the larval period, during which feeding ceases. The pupal stage takes six to nine days, and the pupae are initially green or pale yellow but later darken to greyish brown. Pupation occurs in the soil in a double-walled pupal cell, and adults emerge after about 5-10 days and have a life span of 5-15 days. The early larvae, in the absence of flower buds and flowers, feed on young tender shoots and peduncles. Later, when the flower buds and flowers are formed, they move to and feed on floral parts and subsequently on green pods. Pod damage consists of tunnelling by foraging larvae and is particularly dramatic, hence the common name of this insect. Infested pods are often webbed together with leaves, flowers and other pods.

The International Institute of Tropical Agriculture (IITA), headquartered in Nigeria, has devoted much effort over three decades to developing methods for controlling podborer in cowpea (Oghiakhe, Jackai and Makuanjuola, 1995; Jackai, Padulosi and Ng, 1996). At this time there is no domesticated cowpea with adequately strong resistance to podborer (Adekola and Oluleye 2008), and conventional breeding may have little chance of producing cowpea cultivars with adequate resistance to podborer (Machuka, 2002).

Resistance to stem damage is available in many cultivars, but high levels of resistance to feeding damage in flowers and pods is not available in cultivated cowpeas (Jackai, Padulosi and Ng, 1996). There is some evidence that pods held together at a wide angle above the crop canopy suffer less damage than pods produced within the canopy and separated by a narrow angle (Oghiakhe, Jackai and Makuanjuola, 1995; Singh, 1980). Cultivars with pods held above the canopy are useful but have a disadvantage. Pods are not very active in photosynthesis and when above the canopy, they reduce the amount of solar radiation reaching the leaves. Studies with cowpea genotypes having different canopy architecture indicated the pods-above-the-canopy trait can reduce photosynthetic efficiency and crop growth rates by as much as 54% (Kwapata, Hall and Madore, 1990). Variations in crop management practices such as cowpea spacing (Asiwe et al., 2005) or sole cropping versus various types of intercropping (Jackai and Adalla, 1997) were shown to have little influence on the populations of podborer or the damage they cause to cowpea.

The use of plant-derived insecticides to control podborer has been studied with emphasis on the neem tree (*Azadirachta indica* A. Juss). Extracts from the kernel, seed and leaves of neem have been shown to cause growth disruption, feeding inhibition, deterrence and mortality in podborer but they are not as effective as synthetic insecticides (Jackai and Adalla, 1997). Applying pesticidal forms of *Bacillus thuringiensis* to control podborer has had limited success (Taylor, 1968). This pesticide is broken down by the ultraviolet rays of the sun and usually is only effective for a few hours.

Attempts to develop biological control methods for podborer have failed in the past (Waterhouse and Norris, 1987). More recent research suggests that the podborer is native to southeastern Asia and its parasitoids are being sought in south-east Asia and tested for their efficacy and specificity (Tamò et al., 1997). Currently, biological control methods are being actively studied and several promising candidates (Table 5.A1.2) are emerging (Tamò et al., 2012).

Use of synthetic insecticides is considered the most effective and dependable means for controlling podborer in cowpea (Asiwe et al., 2005). Insecticides are often not locally available or are too expensive for smallholder farmers. Health problems related to misuse of insecticides (Coulibaly and Lowenberg-DeBoer, 2002; Maumbe and Swinton, 2003) are another reason for considering alternative solutions to the podborer problem.

## Hairy caterpillar

In the Sahelian zone, which is the second most important area where cowpeas are grown, insect pest pressure is low but on occasions hairy caterpillar (*Amsacta moorie* Butler syn. *Amsacta moloneyi* Druce) can totally destroy large areas of the crop and cultivar resistance is not available. At the beginning of the rainy season in the Sahelian zone of Senegal, waves of female *Amsacta* moths emerge and lay eggs on a large range of plant species (Ndoye, 1978). They will feed on a range of grasses, pearl millet, sorghum and peanut but they show preference for cowpea. If the cowpea plants are young when they are infested, they are defoliated and killed. If the cowpea plants are large, they can outgrow the attack and are only partially defoliated. Usually, however, the waves of hairy caterpillars arrive when the cowpea plants are young.

Hairy caterpillar can be controlled by synthetic insecticides; however, farmers usually do not have the spraying equipment or supplies of insecticide to enable them to control the sporadic large waves of hairy caterpillar that occasionally occur in the Sahelian zone.

In cases where hairy caterpillar is not present, useful yields of cowpea often can be obtained in the Sahelian zone without using insecticides, which is one reason why many farmers in this zone do not have either sprayers or insecticides.

Table 5.A1.1. Pests and diseases of cowpea

Insects and mites	Podborers – <i>Maruca vitrata</i> * – <i>Cydia ptychora</i> Hairy caterpillar ( <i>Amsacta moorie</i> )* Storage pests – <i>Callosobruchus maculatus</i> * – <i>Bruchidius atrolineatus</i> Thrips – <i>Megalurothrips sjostedi</i> * – <i>Sericothrips occipitalis</i> – <i>Frankliniella schultzei</i> Pod-sucking bugs – <i>Clavigralla tomentosicollis</i> * – <i>Riptortus dentipes</i> – <i>Anoplocnemis curvipes</i> Lygus bugs ( <i>Lygus hesperus</i> ) Cowpea curculio ( <i>Chalcodermus aeneas</i> ) Stink bugs ( <i>Nezara viridula</i> ) Aphids – <i>Aphis craccivora</i> – <i>Myzus persica</i> – <i>Aphis gossipii</i> Green leafhopper ( <i>Empoasca kraemeri</i> ) Foliage beetles – <i>Ootheca mutabilis</i> – <i>Medythia quaterna</i> Flower beetle ( <i>Mylabris pustulata</i> ) Greasy cutworm ( <i>Agrotis psilon</i> ) Bean shoot fly ( <i>Ophiomyia phaseoli</i> ) Bean pod fly ( <i>Melanogromyza sojae</i> ) Red spider mite ( <i>Tetranychus urticae</i> )	Fungal and bacterial diseases	Septoria leaf spots – <i>Septoria vignae</i> – <i>S. vignicola</i> Scab ( <i>Elsinoë phaseoli</i> ) Brown blotch ( <i>Colletotrichum capsici</i> and <i>C. truncatum</i> ) Cercospora leaf spot ( <i>Cercospora canescens</i> ) Fusarium wilt ( <i>Fusarium sp</i> ) Rusts – <i>Uromyces appendiculatus</i> – <i>Phakopsora pachyrhizi</i> Anthracnose ( <i>Colletotrichum destructivum</i> ) Powdery mildew ( <i>Erysiphe polygoni</i> ) Ashy stem blight ( <i>Macrophomina phaseolina</i> ) Ascochyta blight ( <i>Ascochyta phaseolorum</i> ) Pythium stem rot ( <i>Pythium aphanidermatum</i> ) Sclerotium stem rot ( <i>Sclerotium rolfsii</i> ) Bacterial blight ( <i>Xanthomonas campestris</i> ) Bacterial pustule ( <i>Xanthomonas axonopodis</i> )
Viruses	Cowpea aphid-borne mosaic virus (CABMV)** Blackeye cowpea mosaic virus (BICMV)** Cucumber mosaic virus (CMV)** Cowpea mosaic virus (CPMV)** Cowpea severe mosaic virus (CSMV)** Southern bean mosaic virus (SBMV)** Cowpea mottle virus (CPMoV)** Cowpea golden mosaic virus (CGMV) Cowpea chlorotic mottle virus (CCMV)	Nematodes	Root knot nematode – <i>Meloidogyne incognita</i> – <i>M. javanica</i> ) Cyst nematode ( <i>Heterodera</i> spp)
Parasitic weeds	Striga ( <i>Striga gesnerioides</i> ) Alectra ( <i>Alectra vogelii</i> )		

Notes: \* No strong host resistance. \*\* Seed-borne viruses.

## Pest predators

As in all cropping systems there are a variety of natural enemies feeding/developing on cowpea insect pests. These natural enemies include more than 25 parasitoid species belonging to the families listed in Table 5.A1.2 (Jackai and Daoust, 1986; Bottenberg, Tamò and Singh, 1998; Adati et al., 2008). In addition to parasitoids, generalist predators

also feed on cowpea insect pests (Table 5.A1.3). These include mites, beetles, ants, bugs and spiders (Bottenberg, Tamò and Singh, 1998; Adati et al., 2008).

Table 5.A1.2. Parasitoids and entomoviruses attacking the podborer *Maruca vitrata* in West Africa

Parasitoids	Status	Stage attacked*	Reference
<b>Hymenoptera, Trichogrammatidae</b>			
<i>Trichogrammatoidea eldanae</i>	Indigenous	Egg	Arodokoun et al. (2006)
<b>Hymenoptera, Eulophidae</b>			
<i>Tretrastichus</i> sp.	Indigenous	Pupa	Usua and Singh (1978)
<b>Hymenoptera, Braconidae</b>			
<i>Apanteles taragamae</i>	Introduced	Larva	Srinivasan et al. (2007)
<i>Bassus bruesi</i>	Indigenous	Larva	Arodokoun et al. (2006)
<i>Bracon</i> sp.	Indigenous	Larva	Arodokoun et al. (2006)
<i>Braunsia</i> sp.	Indigenous	Larva	Usua and Singh (1978)
<i>Braunsia kriegeri</i>	Indigenous	Larva	Arodokoun et al. (2006)
<i>Dolichogenidea</i>	Indigenous	Larva	Arodokoun et al. (2006)
<i>Phanerotoma</i> sp.	Indigenous	Egg-larva	Usua and Singh (1978)
<i>Phanerotoma leucobasis</i>	Indigenous	Egg-larva	Arodokoun et al. (2006)
<i>Pristomerus</i> sp.	Indigenous	Larva	Arodokoun et al. (2006)
<i>Testudobracon</i> sp.	Indigenous	Larva	Arodokoun et al. (2006)
<b>Diptera: Tachinidae</b>			
<i>Aplomya metallica</i>	Indigenous	Larva	Agyen-Sampong (1978)
<i>Cadurcia</i> sp.	Indigenous	Larva	Arodokoun et al. (2006)
<i>Nemorilla maculosa</i>	Indigenous	Larva	Srinivasan et al. (2007)
<i>Pseudopetichaeta laevis</i>	Indigenous	Larva	Usua and Singh (1978)
<i>Thecocarcelia incedens</i>	Indigenous	Larva	Agyen-Sampong (1978)
<i>Thelairosoma palposum</i>	Indigenous	Larva	Usua and Singh (1978)
<b>Entomoviruses</b>			
<b>Baculoviridae</b> MaviMNPV	Introduced	Larva	Lee et al. (2007)
<b>Cypoviridae</b> MaviCPV	Indigenous	Larva	Tamò et al. (2003)

Source: Adapted from Tamò et al. (2012).

Table 5.A1.3. Non-pest arthropods associated with cowpeas

Families containing natural enemies of cowpea pests	Generalist predators	Bees that forage on cowpea flowers
Braconidae	Phytoseiid mites	Honey bees ( <i>Apis mellifera andonsonii</i> )
Chalcididae	Coccinellid beetles	Carpenter bees ( <i>Xylocopa</i> sp)
Encyrtidae	Staphilinid beetles	Digger bees ( <i>Anthophora</i> sp)
Eulophidae	Mantodea	Bumble bees ( <i>Bombus</i> spp)
Ichneumonidae	Formicid ants	Leaf-cutting bees ( <i>Megachile</i> spp)
Pteromalidae	Anthocoridae bugs	
Scelionidae	Spiders	
Tachinidae		
Trichogrammatidae		

## *Annex 5.A2.*

### **Biotechnological developments**

#### **Biotechnological approaches in cowpea improvement**

The goal of cowpea breeding programmes is to develop consumer-preferred varieties with high yield and resistance to biotic and abiotic constraints to production. Traditional plant-breeding approaches to cowpea improvement have had many successes over the last 30 years. Recent figures from the Food and Agriculture Organization's statistics (FAOSTAT) show an impressive increase in the productivity of cowpea globally. Three principal methods are used in breeding the self-pollinating cowpea: pedigree, mass selection and single seed descent. The pedigree method, often with slight modifications, is the one most frequently used. Selections are based largely on the main character of interest, for example, resistance to the parasitic weed *Striga*. Detailed data on maturity, time to flower, growth habit, and grain and fodder yields are collected and the most promising single plants selected for advancement. Other traits of interest are selected for as well, including seed colour, seed texture, seed size and leaf yield. The relative importance of these traits varies with the particular breeding programme. For example, leaf yield is more important in eastern and southern Africa while west and central African breeding projects lay more emphasis on grain and fodder yields.

Varieties are available that can yield more than 1 tonne/ha. Over the years, improvements have resulted in more than a doubling of the average yield of the crop, from about 200 kg/ha to about 500 kg/ha. However, even this still-modest level of productivity can only be guaranteed if one or two insecticide sprays are applied.

Unfortunately, there are no utilisable resistance genes for post-flowering insect pests in the cowpea genome. There is little prospect for genetic improvement of cowpea by wide-crossing. Cowpea is extremely well-isolated from other *Vigna* species that might provide sources of resistance genes. Many efforts have sought to create viable wide crosses between cowpea and its nearest relatives, but the gulf has proven too wide. For example, it is known that resistance to some insects such as the legume podborer, *M. vitrata*, exists in a distant relative of cowpea, *V. vexillata*, but interspecific genetic barriers prevent hybridisation. What is true for *M. vitrata* is also true for the cowpea bruchid, and for pod-sucking bugs and thrips. Lack of resistance genes is a major bottleneck that limits the success of conventional cowpea breeding. Biotechnological approaches to finding these genes outside the cowpea genome and transferring them into cowpea may progress cowpea improvement. Given the successes with other crops such as maize, tomatoes, sweet potato and cotton, biotechnological approaches to introduce insect resistance and other traits are being explored for cowpea.

#### **Improved cowpeas developed by using biotechnologies**

The first reported use of genetic transformation in cowpeas was conducted by Garcia and colleagues (García, 1986; García et al., 1987) using *Agrobacterium tumefaciens* as the gene vector and although antibiotic-resistant callus was obtained, no whole plants were regenerated. Later, mature de-embryonated cotyledons were used as target tissues for gene transfer (Muthukumar et al., 1996). The authors obtained transgenic plants after selection on the antibiotic, hygromycin. However, transmission of the transgenes to the

next generation could not be demonstrated. When the particle gun was used to deliver genes to cowpea, it was found that they were transmitted to only a small proportion of the progeny and that there was no evidence for stable integration of the transgenes (Ikea et al., 2003). A very promising regeneration and transformation system was described by Kononowicz et al. (1997) and although not pursued at the time, it formed the basis of a system that turned out to be reproducible and that obeys Mendelian rules of inheritance (Popelka et al., 2006). Critical features of this system include suitable explants from cotyledonary nodes or embryonic axes and a tissue culture regime without auxins in the early stages, but which includes a cytokinin at low levels during shoot initiation.

There are now several reports showing experimental evidence for reproducible gene transfer to cowpea, including genes for podborer (Higgins et al., 2012), cowpea weevil (Solleti et al., 2008) and for weed control (Citadin, Cruz and Aragão, 2013) as well as a range of model genes to evaluate the technology (Citadin, Cruz and Aragão, 2013; Behura et al., 2014).

The first insect resistance trait being tested using biotechnology is against the legume podborer, *Maruca vitrata*. The cowpea podborer belongs to the Pyralidae, the family to which the European corn borer (ECB) belongs. ECB, a major pest of maize in the eastern United States, can be controlled by means of maize hybrids genetically engineered to express the *cryIAb* gene from *Bacillus thuringiensis* (often referred to as *Bt*). In the US Corn Belt, about one-quarter of maize now carries the *cryIAb* gene. The protein product of this gene has been shown to be toxic to *M. vitrata* when fed in the diet ( $LC_{50}=0.03 \mu\text{g/g}$  diet) (Srinivasan, 2008). Accordingly, genetic transformation of cowpea to express the *cryIAb* protein has the prospect of imparting *M. vitrata* resistance. The African Agricultural Technology Foundation (AATF) based in Kenya is implementing a programme to develop genetically engineered maruca-resistant cowpeas. The bred lines contain the *cryIAb* gene, with the *nptII* gene used as selectable marker. Being under testing phase, some varieties are expected to reach the African market around 2017 (AATF, n.d.).

Another constraint that cannot be adequately addressed through conventional breeding is resistance to cowpea weevil. While it is true that there are cowpea cultivars derived from the landrace TVu2027 with moderate resistance to cowpea weevil, this resistance has already been incorporated into many cowpea varieties and has been widely disseminated, both in Africa and beyond. It now appears that there are populations of cowpea weevil that can overcome this resistance. Numerous genes have the potential to confer resistance to the cowpea weevil if transferred into cowpea and expressed in the seed. The most advanced of these involves transferring an  $\alpha$ -amylase inhibitor ( $\alpha$ AI) gene from common bean into cowpea. The  $\alpha$ AI protein protects common bean seeds against cowpea weevil and certain other bruchids, though not against the common bean weevil. When  $\alpha$ AI was linked to a strong seed-specific promoter and transferred into garden pea using gene technology, the garden pea seeds, which are normally susceptible to cowpea weevil, proved to be highly resistant (Shade et al., 1994). By transferring the common bean  $\alpha$ AI gene into cowpea and expressing it in the seeds, it should be possible to introduce a new source of weevil resistance into cowpea. However, some uncertainty hangs over this undertaking as the  $\alpha$ AI protein may not be produced in the recipient plant exactly as it is in the donor parent. This has been observed with  $\alpha$ AI expressed in garden peas. The  $\alpha$ AI protein from garden peas had small mass difference from that of the protein from common bean, a difference probably due to a variation in the degree of post translational modification in the recipient species. The possibility that this variant



protein – which still inhibits insect  $\alpha$ -amylase and blocks weevil growth and development – might cause toxicity or allergenicity in consumers of the transformed seed has to be addressed (Prescott et al., 2005), although in a recent comprehensive study this was considered to be unlikely (Lee et al., 2013).

In those cropping areas where cowpea is grown as a sole crop, it could be desirable and feasible to control weeds using a herbicide. It was recently shown that a biotechnological approach could be used to introduce tolerance to a Group B herbicide into cowpea (Citadin, Cruz and Aragão, 2013). This could open the way to a no-tillage farming system for cowpea.

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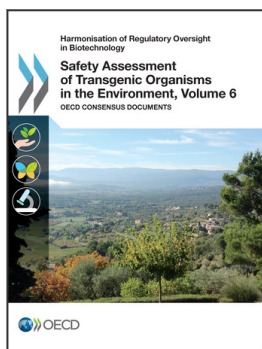
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