

Chapter 6

Cowpea [*Vigna unguiculata* (L.) Walp.] Breeding



**Ousmane Boukar, Abou Togola, Siva Chamarthi, Nouhoun Belko,
Haruki Ishikawa, Kanako Suzuki, and Christian Fatokun**

Abstract Cowpea, *Vigna unguiculata* (L.) Walp., is an important grain legume grown and consumed not only in the dry savannah areas of Sub-Saharan Africa but also in many other tropical and subtropical regions. It provides income, food and nutrition security to millions of people. Several studies have led to a better understanding of the taxonomy of cowpea and its wild relatives. The species diversity, distribution and evolution of cowpea have been intensively explored. The crop is mainly cultivated in intercropping system where its low plant population does not allow the full expression of the yield potential of the cultivars being grown. Considerable challenges affect the production of this crop despite its comparatively better adaptation to harsh environments. The available genetic resources maintained in the different gene banks are being used for the improvement of cowpea. Germplasm diversity and cultivars characterization were conducted in different studies. Sources of resistance/tolerance to key biotic and abiotic stresses are being identified and introgressed genes involved in new breeding lines are being developed. Improvement strategies were developed to address the major constraints to production while also taking consumer preferences into consideration. Breeding approaches of self-pollinated crops were used in the breeding programs. Application of biotechnology has been suggested to address intractable problems. Considerable effort has been made to genetically transform cowpea. Recent development of genomic resources should support the implementation of molecular breeding to complement conventional breeding and to enhance genetic gain. Key elements needed for successful application of molecular breeding tools include the availability of a high-throughput genotyping platform, high-quality consensus genetic maps,

O. Boukar (✉) · A. Togola · S. Chamarthi · N. Belko
Cowpea Breeding Unit, International Institute of Tropical Agriculture, Kano, Nigeria
e-mail: o.boukar@cgiar.org; a.togola@cgiar.org; s.chamarthi@cgiar.org; n.belko@cgiar.org

H. Ishikawa · C. Fatokun
Cowpea Breeding Unit, International Institute of Tropical Agriculture, Ibadan, Nigeria
e-mail: h.ishikawa@cgiar.org; c.fatokun@cgiar.org

K. Suzuki
Cowpea Breeding Unit, International Institute of Tropical Agriculture, Lusaka, Zambia
e-mail: kanasuzu@shinshu-u.ac.jp

improved phenotyping capability and identification of markers closely linked to target traits. Progress is being recorded in many of these areas which should allow the development of modern breeding programs that will result in effective and efficient development of improved resilient cowpea cultivars.

Keywords Conventional breeding · Cowpea · Genetic resources · Genomics · Modern Breeding · *Vigna unguiculata*

6.1 Introduction

Cultivated cowpea, also called black-eyed pea (*Vigna unguiculata* (L.) Walp.) is a commonly grown and consumed grain legume, which is very well adapted to the dry savannah areas of Sub-Saharan Africa (SSA). It belongs to the subfamily Faboideae, tribe Phaseoleae, subtribe Phaseolineae, genus *Vigna* section *Catiang*. The genus *Vigna* is divided into numerous species with numbers ranging from about 150 (Verdcourt 1970) to 184 (Phillips 1951) and 7 sections (Verdcourt 1970). The classification of *Vigna* species has remained generally inconclusive for quite some time. Several researchers have made efforts to resolve the *Vigna* species classification issue (Maréchal et al. 1978; Padulosi 1993; Pasquet 1993; Pienaar 1992). In recent reports, Pasquet and Padulosi (2012) synthesized the work of Vaillancourt et al. (1993) and Delgado-Salinas et al. (2011) on the taxonomic boundaries of the genus *Vigna* thus suggesting the existence of five subgenera as follows:

- (a) *Lasiospron*;
- (b) *Vigna* (now including yellow and blue flowered species such as *V. subterranean*);
- (c) *Haydonia*;
- (d) *Ceratotropis* (Asian subgenus);
- (e) *Plectotropis* (including section *Catiang* with two species *V. unguiculata* {cowpea} and its sister species *V. schlechteri* Harms previously referred to as *V. nervosa* Markotter).

De Leonardis et al. (1993) reported that members of section *Catiang* have canoe-shaped keel pointed like a beak at the top and pollen grains have reticulate surfaces. However, the cultivated cowpea along with its cross compatible wild relatives are grouped in *Vigna unguiculata* subspecies *unguiculata*.

Pasquet and Padulosi (2012) concluded that the position of *Vigna* within Phaseolineae is established. Cultivated cowpea, *Vigna unguiculata* subspecies *unguiculata* is divided into four cultivar groups as follows: *Unguiculata*, *Sesquipedalis*, *Biflora* and *Textilis*. The cultivar group *Unguiculata* is made up of the cowpea generally grown for the protein-rich grains and fodder for livestock while *Sesquipedalis* comprises the yard-long-bean which is most commonly grown in Asian countries, especially India and China. It is suggested that this cultigroup evolved from cowpea in Asia following selection for long podded types that are

consumed as vegetable – both grains and fleshy pods. The cultigroup *Textilis* has long fibrous peduncles, which are used in northern parts of Nigeria to make rope from the fiber.

The primitive wild relatives of cowpea such as *Vigna unguiculata* ssp. *dekindtiana*, ssp. *stenophylla*, ssp. *tenuis*, ssp. *pubescens* and ssp. *protracta* as well as several cultivars like ssp. *tenuis* var. *tenuis*, var. *oblonga*, var. *parviflora*, var. *ovata* and ssp. *protracta* var. *protracta*, var. *rhomboidea* among others, are most commonly found in southern parts of Africa. These are distributed across from Namibia through Zambia, Botswana, Zimbabwe, Mozambique, Eswatini (Swaziland) and South Africa (Padulosi et al. 1991).

6.1.1 Domestication

The West and Central Africa subregions have been suggested to be the center of origin of cultivated cowpea since it is there that the greatest amount of germplasm is present. The immediate progenitor of cultivated cowpea has been reported to be *Vigna unguiculata* ssp. *dekindtiana* var. *dekindtiana*. It is the most cross compatible with cowpea and seeds of some *dekindtiana* lines though small in size are bigger than those from the wild types found in southern parts of Africa. In a review of previous work that was based on plant morphology, Faris (1965) suggested there was enough evidence to show that cultivated cowpea was domesticated in West or Central Africa. The movement of cowpea to other parts of the world such as Europe, Asia and the Americas is said to have taken different routes. While cowpea movement to Asia especially India was from West Africa through northeastern Africa along with sorghum, which is adapted to similar agro-ecologies as cowpea (Faris 1965; Pant et al. 1982) movement to other parts especially to the USA was through African slaves who took along some seeds (Whit 2007). Movement of cowpea to Europe was through Egypt in North Africa (Ng and Singh 1997). The yard-long-bean, *V. unguiculata* ssp. *unguiculata* cultivar group *Sesquipedalis*, evolved in Asia where it is most commonly found and cultivated for consumption of the fresh green pod with seeds as a vegetable.

6.1.2 Importance

Cowpea is a food and nutrition security crop in different parts of SSA. Farmers and food vendors derive income from cowpea, which also provides fodder for ruminants. Being a legume, cowpea fixes atmospheric nitrogen some of which it uses for its growth and development and leaves some in the soil for the benefit of companion and following crops. The rapid growth rate of cowpea in the field enables the canopy to cover the soil thereby helping to reduce soil erosion. Apart from being a source of food, feed and income cowpea also contributes to the sustainability of the

cropping system and the environment. The grains are processed into several types of dishes for human consumption. The most common dish made of cowpea across several parts of West Africa is *akara* or *kosai*. To make *akara* the cowpea grain is ground into a paste and deep fried in oil in small balls. Cowpea is a known source of dietary protein in many communities where meat is very expensive to purchase. Besides the protein content which can be up to 32% on a dry weight basis (Nielsen et al. 1993) the grains also contain carbohydrates (62% soluble carbohydrates) and according to Boukar et al. (2011) minerals such as iron (33.6–79.5 mg/kg), zinc (22.1–58 mg/kg), phosphorus (3450–6750 mg/kg), calcium (310–1395 mg/kg) magnesium (1515–2500 mg/kg) and potassium (11,400–18,450 mg/kg). Compared with some other food legumes cowpea is rich in amino acids such as lysine, methionine and tryptophan but deficient in sulfurous amino acids. The grains also contain trypsin inhibitor, but the level is about one-half that contained in soybean (Lambot 2002). Cooking, however, inactivates the inhibitor. Cowpea leaves are also consumed as a vegetable in several East African countries especially Kenya and Tanzania. The green leaves contain 29–43% protein on a dry weight basis with younger ones having higher amounts (Nielsen et al. 1997). Because cowpea, when compared to several other crops, is drought tolerant, it promises to be better suited to the dry savannah regions that are already characterized by increased frequency of short raining seasons due to climate change.

Most farmers in the dry savannah areas of SSA keep livestock, which they feed with cowpea haulm that is well appreciated by the farmers because of the appreciable level of protein present. Many farmers derive almost the same amount of income from sales of grain as from fodder harvested from their fields. Breeders have in recent times devoted some attention to increasing fodder quality and yield in cowpea because of the economic value of the haulm (Samireddypalle et al. 2017). A number of dual purpose (grain and fodder) cultivars have been selected by breeders to meet the farmers' needs for livestock fodder. The genetic variation present among different cowpea lines in fodder quality and quantity gives breeders opportunities to make progress in selecting for lines with higher levels of these attributes.

6.1.3 Selection and Early Improvements

The history of cowpea cultivar development is recent. Spillman (1911, 1913) in the USA carried out genetic studies in cowpea and reported the inheritance of a number of traits that relate to seed. In India, Roy and Richaria (1948) were the first to report making crosses in cowpea with the aim of generating segregating populations from which selections were made for lines with early maturity and other desirable attributes. Not much activity was reported until the 1960s towards the development of improved cowpea cultivars in Africa where the greatest quantity is produced and consumed. The earliest report of cowpea improvement in Africa was at the Potchefstroom College of Agriculture, South Africa (1948). The cowpea breeding

work initiated at the institute was aimed at developing cultivars that are erect with long peduncles that carry pods above the canopy to enable harvesting with a mower and possessing resistance to leaf spot and nematodes. In SSA, the first report of cowpea breeding work in 1956 was from the Northern Nigeria Regional Department of Agriculture. Concerted breeding efforts were initiated in the early 1970s at the International Institute of Tropical Agriculture with collection and ex-situ conservation of several germplasm lines. The collection efforts resulted in the acquisition of several germplasm lines from different parts of SSA and from outside the region.

Germplasm collection was followed with evaluations for identification of superior genotypes, recombining desirable traits from two or more parents, advancement of segregating populations and selecting those showing desired attributes. Selected lines were later tested across many agro-ecologies (IITA 1972). Those showing superior performance in comparison with existing farmers' cultivars were distributed to interested farmers in various communities. Initial selection efforts were focused on breeding lines with early maturity, disease resistance, high grain yield and white or brown seed coat color. With time however, emphasis was placed on selection of lines with resistance to insect pests, diseases (fungal, bacterial, viral), early to medium maturity, day neutral characteristic, rough seed coat texture and high grain yield. In more recent times, selection has focused on developing breeding lines with resistance to *Striga*, high grain yield, large seed size and dual purpose. It has been difficult to find germplasm lines that show sufficiently high levels of resistance to insect pests such as flower bud thrips, maruca pod borer and a complex of pod-sucking bugs. Hence, cowpea cultivars available to farmers can perform optimally when adequate protection against insect pests is provided. The development and release of cultivars with insect resistance will enable farmers to grow cowpea more profitably, enhance their health as they will no longer need to handle potentially toxic synthetic insecticides and to protect the environment because no chemicals will be released while protecting the crop in the field.

This chapter is devoted to cowpea as an important but *orphan* grain legume crop of SSA. The topics covered include cultivation practices, germplasm biodiversity and conservation, crop improvement using both conventional and new methods such as transgenics and molecular marker-assisted selection.

6.2 Cultivation and Traditional Breeding

Cowpea is grown in different agro-ecological zones of the world. Some key cowpea producing countries are shown in Fig. 6.1. Figures shown for Brazil, Benin, Botswana, Chad, Ghana, Guinea-Conakry, Namibia, Sierra Leone, South Sudan and Togo were obtained through cowpea scientists working in the countries. Its production and utilization depend mostly on the agro-climatic conditions of the production environment and the socioeconomic conditions, ethnic culture and traditions of the people. Different biotic and abiotic stresses adversely affect cowpea production. Cowpea research activities (Appendix I) are being implemented in many cowpea

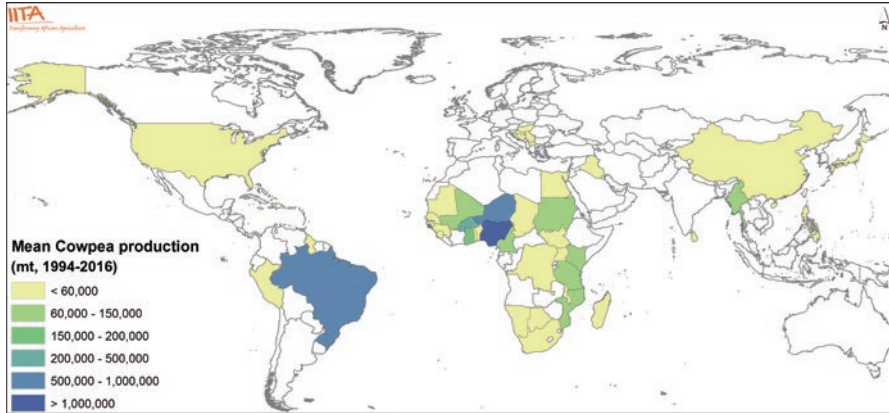


Fig. 6.1 Cowpea production in the world. (Source data: FAOSTAT (2018) and personal communications with scientists (2018))

producing countries to alleviate these constraints. Conventional breeding methods have been used to develop a number of cultivars (Appendix II) and modern breeding tools of molecular markers are now being applied following recent developments of genomic resources.

6.2.1 Current Cultivation Practices

Agronomic practices and the level of inputs (quality seeds, fertilizers, agrochemicals) used for cowpea production differ greatly among farmers. In SSA some farmers still grow local landraces (mostly spreading types) in association mainly with cereals (sorghum, millet, maize). These landraces are characterized by photosensitivity. Cowpea is grown in different types of soils preferably well-drained sandy loams. Generally heavy clay soils are avoided, as the crop is sensitive to waterlogging conditions. Under traditional cultivation, a majority of farmers plant cowpea on flat soil with no ploughing although a few use animal drawn ridgers and still fewer use tractors for land preparation. Weeding of the field is carried out using hand-held hoes. Cowpea yield increases with good land preparation.

Farmers in SSA do not apply fertilizer on cowpea fields. However, the soils are poor with deficiencies in nitrogen, phosphorous and organic matter content. For the crop to produce optimal yield it is recommended to have a starter dose of nitrogen of up to 20 kg/ha. Increase in yield is associated with phosphorus application as single superphosphate at 40 kg P₂O₅/ha (Suzuki et al. 2018). Fertilizer application is done at planting or 7–10 days afterwards.

Typically, cowpea is planted randomly at up to 1.0 m distance between plants and along with cereals. Usually the companion cereal crop is planted first and cowpea later. Where planting is done following straight rows, 0.8050×0.40 m is used for local prostrate cultivars while 0.50×0.20 m is used for the erect and semi-erect types. Using row planting allows maximizing the crop density and facilitates the application of inputs.

When planted as sole crop or within an organized system, planting is done such that the maturity period of the crop coincides with dry weather. Pod rots cause considerable grain yield loss if harvesting occurs during humid cloudy weather. In addition to the maturity of the cultivars, photosensitivity is also taken into consideration when planning to plant cowpea. Early photo-insensitive cultivars are planted earlier while prostrate photosensitive types may be delayed.

Under intercropping, less than 5 kg seeds are used for planting 1 ha. Generally, these are spreading/prostrate types. In monocropping, 20–25 kg of seeds are required for 1 ha. Most of these are erect and semi-erect types. Until recently seeds were distributed mainly from farmer to farmer. In many cases, grains are purchased and used as seeds. With the efforts of different projects and the deployment of extension agents, farmers organize themselves in groups to produce and sell seeds in their communities. In some countries, seed companies are emerging and they acquire foundation seeds from research institutes and produce certified seeds that are sold in small packs.

In SSA, seeds are usually not treated prior to planting although it is recommended to use seeds free of diseases and insect pests. A good plant stand in the field should lead to high yield. Weed control is one activity that contributes to higher yield in the field. Cowpea may suffer when competing for light, water and nutrients with weeds at its early growth stages. Depending on the region, two to three hand weedings are carried out during the crop life cycle. In some areas, animal tractions are used for weeding and ridging the crop. When pre-emergence herbicides are used, the first weeding can be considerably delayed. Unfortunately, the requisite herbicides are not easily available and most farmers are not aware of their importance. What many farmers are aware of is the use of insecticides to protect their crop. Having one to two targeted insecticide applications is a prerequisite for good grain yield.

6.2.2 Current Agricultural Problems and Challenges

Despite its resilience to drought and low soil fertility, cowpea production is considerably affected by numerous constraints. In addition to biotic and abiotic stresses, there are agronomic practices and socioeconomic challenges that limit significantly the production of this crop in SSA.

6.2.2.1 Biotic Stresses

Insect pests are considered the most limiting factors for cowpea production in most parts of the tropics where appropriate insecticides are lacking or unaffordable by farmers. Insects attack cowpea from the seedling stage to seeds in storage. In addition, different groups of pests infest the cowpea plant at the same time (Jackai and Adalla 1997). At the seedling stage, aphids (*Aphis craccivora* C.L. Koch), beanflies (*Ophiomyia* spp.) leafhoppers (mainly *Empoasca* spp.), foliage beetles (*Ootheca* spp., *Medyrthia* spp. and others), the arctiid defoliator (*Amsacta moloneyi* Druce) and some foliage beetles infest cowpea. During flower bud initiation and flowering time, the most important insect pests are flower bud thrips (*Megalurothrips sjostedti* Trybom) and in some cases maruca (*Maruca vitrata* Fabr.). At podding stage there are *Maruca vitrata*, *Clavigralla* spp., *Acanthomia* spp. and *Riptortus* spp. causing damage to cowpea pods and seeds contained therein. During storage, bruchid (*Callosobruchus maculatus* Fabr.) is the most important insect pest (Boukar et al. 2013, 2015).

Several diseases afflict cowpea and may cause appreciable grain yield loss. The main categories of diseases are bacterial, fungal and viral. Among the bacterial diseases are bacterial blight and bacterial pustule. Major fungal diseases include anthracnose, *Macrophomina*, *Fusarium* wilt, web blight, brown blotch, *Cercospora* leaf spot, *Septoria* leaf spot and scab. Viral diseases recorded in cowpea production areas are cowpea yellow mosaic, cowpea aphid borne mosaic, black-eyed cowpea mosaic, cowpea severe mosaic and southern bean mosaic (Boukar et al. 2013, 2015).

Other biotic constraints are parasitic weeds and nematodes. *Striga gesnerioides* (Willd.) Vatke and *Alectra vogelii* Benth are main parasitic weeds that are found predominantly in West and Central Africa (WCA) and Eastern and Southern Africa (ESA) respectively. Nematodes such as root knot nematodes induce significant yield losses in susceptible cowpea cultivars.

6.2.2.2 Abiotic Stresses

Drought affects cowpea production adversely despite the ability of the crop to grow under hot weather conditions with little rainfall during the short (55 day) cropping season (Hall and Patel 1985). All the types of drought – seedling stage, reproductive stage and terminal can be experienced by cowpea.

Heat reduces significantly cowpea grain yield when high temperatures (>20 °C) occur late at night. This is because flower production and pollen viability are affected by high temperatures (Hall 2004). This author showed that each 1 °C above a threshold of 16 °C during the night leads to 4–14% reduction in grain yield.

Cowpea production is also reduced due to low soil fertility. In the savanna agroecology, low soil fertility is common due to low organic matter and phosphorous contents. It is reported that soil fertility is more limiting to cowpea grain and fodder production in the Sahelian zone than rainfall and the use of fertilizer can increase water-use efficiency (Penning de Vries and Djiteye 1991).

6.2.3 *Improvement Strategies*

To alleviate the various cowpea production constraints, improvement programs were established in several countries in Sub-Saharan Africa such as Nigeria, Niger, Senegal, Burkina Faso, Uganda, Kenya and Tanzania with major attention from 1960 onward (Singh and Ntare 1985). During the same period of time, there were substantial efforts in Asia to breed cowpea to suit local cropping systems and consumer tastes. Most of the literature on development of cowpea cultivars comes from India, as well as from other countries such as Bangladesh, Myanmar, China, Indonesia, Nepal, Pakistan, the Philippines, Sri Lanka and Thailand (Mishra et al. 1985). In Latin America, EMBRAPA in Brazil in collaboration with IITA, initiated early cowpea improvement programs. Other countries in this region with some research activities on cowpea include Colombia, Venezuela, Panama, Trinidad, Nicaragua, Jamaica and Guyana. Cowpea is grown mostly in all of the southern USA with extensive dry seed industries in both California and Texas. Cowpea breeding and evaluation programs have existed in the US since the latter part of the nineteenth century. Some of the main cowpea research institutions in this early period included the Arkansas Agricultural Research Station. Around 1980, cowpea research was being conducted at 28 different locations in the USA. Eight institutions, namely, Auburn University, Clemson University, Louisiana State University, Mississippi State University, Texas A&M University, University of Arkansas, University of California and the University of Georgia and the Agricultural Research Service of USDA had ongoing research programs with clearly identifiable cowpea breeding objectives (Fery 1985).

Early cowpea breeding efforts targeted a better understanding of the botany, morphology, physiology and production constraints. Later high yield potential and seed quality were considered as main objectives of the breeding programs. Responses to day length, crop maturity and crop position within the different cropping systems were also considered in the development of improved cowpea cultivars. Major steps taken in the development of better cowpea cultivars in the 1970s were germplasm collection, evaluation and maintenance and breeding for disease resistance. Subsequently, emphasis focused on breeding for insect resistance, early maturity and improved plant types with desired grain quality (Singh and Ntare 1985). Searches for sources of resistance to different diseases and insect pests were also initiated (Singh et al. 1983) followed by an intensive hybridization program to incorporate these key traits in improved breeding lines. In the 1980s, breeding efforts focused on seed-type preferences in the different regions and assessments of damages caused by insect pests along with multiple diseases. It was the aim of breeders at this earlier time to develop single lines with resistance to all the major cowpea diseases in the humid and subhumid tropics. Hence all segregating populations were evaluated for resistance to several diseases from F2 to F6 both under natural or supplemented infestation in the field and glasshouse. As for resistance to insect pests, efforts were devoted to aphids, thrips and bruchids. To this end germplasm lines were evaluated in the field and screenhouse for detection of resistant

lines. A germplasm line TVu3000 was found to be resistant to aphids and the dominant gene controlling the trait was transferred to several improved cowpea breeding lines. Similarly, line TVu 2027 and a land race from Ghana Sanzi were found to show tolerance to storage weevils and flower bud thrips, respectively. These tolerance genes have been incorporated into a number of improved breeding lines. As with insect resistance, germplasm lines were also evaluated for resistance to diseases and those that showed the desired levels of resistance were used as parents in crosses from which segregating generations were assessed for their reactions to the diseases. For example, a local land race, Dan Ila was detected as being resistant to bacterial blight. The resistance gene has been transferred to many improved breeding lines. During the same period, a systematic program was initiated to develop extra-early cowpea cultivars that fit into multiple-cropping systems (Singh and Ntare 1985).

In the 1990s, the IITA breeding program focused on the development of high yielding bush-type vegetative cowpea cultivars with different maturity periods (extra-early, early, medium, late), photoperiod sensitive and insensitive grain types and adaptation to different cropping systems (sole crop, intercrop) (Singh et al. 1997).

In the early 2000s, cowpea cultivars with preferred seed types resistant to biotic (diseases, insect pests, parasitic weeds) and abiotic (heat, drought) and adapted to both sole cropping and intercropping were developed (Singh et al. 2002). From 2010 and thereabouts these efforts were sustained with the aim of developing high yielding widely adapted and stable cowpea cultivars (Fig. 6.2) with resistance to major production constraints and with acceptable seed types (size, texture, color, protein and mineral content, improved cooking properties). The implementation of the Collaborative Research Program of CGIAR on Grain Legumes offered the opportunity to strengthen the development of cultivars with tolerance to drought and low-soil phosphorous and resistance to insect pests. Recent improvement strategies are building on the use of modern approaches in cowpea breeding (Ehlers et al. 2012).

6.2.4 Traditional Breeding Methodologies and Limitations

Being self-pollinated, cowpea cultivar development has benefited from the breeding methodologies applicable to this group of crops. Many cultivars were obtained at the beginning of the breeding programs using mass selection and pure line approaches. Landraces collected from farm fields were evaluated and single plants found to be of good performance were selected. In the case of mass selection, seeds from these plants were bulked and grown to produce improved populations where further selections could be repeated several times. For pure-line breeding, the seeds from each selected plant were sown as progeny rows. Seeds of the best rows were evaluated in replicated yield trials and superior lines selected to constitute new improved cultivars.

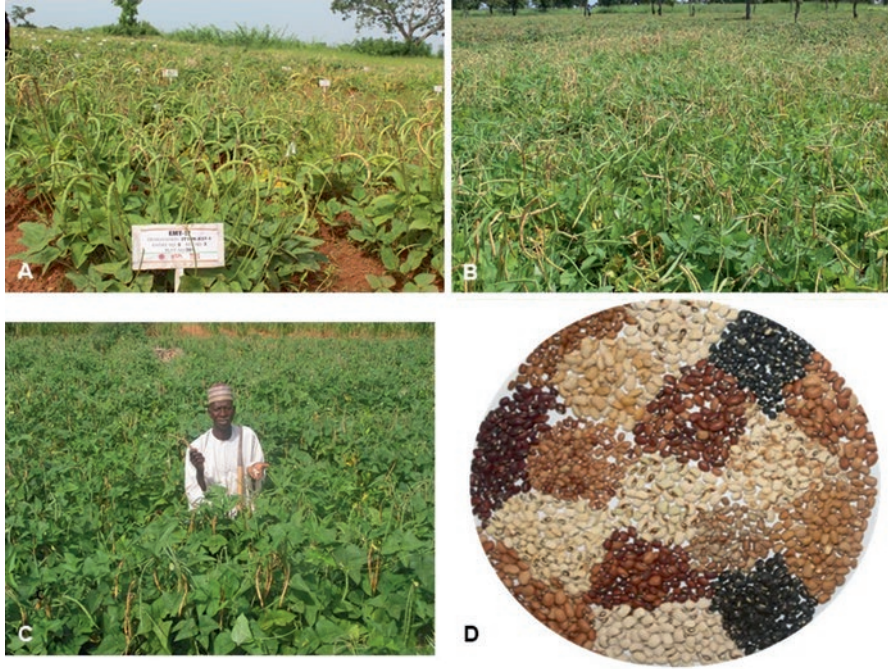


Fig. 6.2 Fields of improved cowpea cultivars. (a) evaluation of advanced lines in Tamale, Ghana, (b) Seed multiplication in Saria, Burkina Faso, (c) Farm field in Kano, Nigeria, (d) Seed diversity in cowpea breeding program, IITA, Kano, Nigeria

Because of the limited genetic variability associated with these two breeding approaches, segregating populations were produced from single or multiple crosses between two or more lines. Depending on the aim of the breeding program, the segregating populations were handled in a number of ways. The pedigree method of breeding is used largely in many cowpea breeding programs. This method has proved suitable for the short-term objective of developing cultivars with new combinations of horticultural characteristics and disease resistance (Fery 1985). Single seed descent has also been used especially for rapid generation of recombinant inbred lines for linkage mapping and QTL identification. Mehta and Zaveri (1997) have reported that single seed descent develops better progenies for yield and yield components than single plant selection. Another method that is commonly used in cowpea breeding programs is the backcross breeding method which has proved to be useful for transferring single resistance genes for specific production constraints into cowpea lines that have good yield performance or are preferred by farmers, but susceptible to or lacking this particular trait. To reduce the time and efforts for record keeping associated with pedigree method, breeders have also used bulk population method. In this method of breeding plants in the segregating populations are harvested in bulk through several generations under natural or artificial conditions.

The different combinations or modifications of the breeding methods mentioned above are being used across cowpea cultivar development programs. Depending on the specific characteristics of the parents that were crossed, the traits of focus in the program, and the targeted environments (off-season, screening facilities, presence or absence of inoculum), IITA often performs a combination of these conventional breeding methods. Fery (1985) reported that a combination of backcross-pedigree breeding method has been used in some programs to transfer desired traits from relatively unadapted genetic backgrounds into well-adapted commercial cultivars. Under this combination of methods only one, two or three backcrosses are conducted while the remainder of the breeding is handled through pedigree procedures.

Despite the progress achieved through conventional breeding methods, there are certain limitations associated with them. Sources of resistance to key production constraints such as insect pests, mainly the pod borer and pod sucking bugs, show low levels of expression in cowpea germplasm lines and cultivars. Unfortunately, a wild cowpea relative *Vigna vexillata* (L.) A. Rich which has good sources of resistance genes to these pests is not cross compatible with the cultivated lines. This has prevented the transfer of the resistance genes into cultivated cowpea. In addition, the traditional cowpea breeding approaches require up to a decade or more to develop improved cultivars largely due to the need to employ sequential and repeated phenotypic evaluations and performance trials (Ehlers et al. 2012). These authors pointed out that in many cases, complex, specialized conditions, techniques and skills are needed to assess phenotypes for selection.

6.2.5 Role of Biotechnology

With the advent of recent advances in biotechnology and genomics (see Sects. 6.4 and 6.5, respectively, of this chapter), cowpea genetic improvement can be made more efficient. With the successful genetic transformation of cowpea and stable transmission of the transgene to progeny according to the Mendelian law of inheritance (Popelka et al. 2006), it is now possible to develop transgenic Bt cowpea cultivars with resistance to the pod borer *Maruca vitrata*. Some modifications have been made to improve the genetic transformation systems, which have led to the development of new transgenic cowpea with resistance to bruchids and caterpillars (Higgins et al. 2012).

6.3 Diversity and Conservation of Germplasm

Major cowpea germplasm collections are conserved at the International Institute of Tropical Agriculture, Ibadan, Nigeria with 15,872 accessions from 90 countries, at Griffins, Georgia, USA with 7146 accessions from 50 countries and at Riverside,

California, USA with 4876 accessions from 45 countries. These gene banks represent the largest repertoires of cowpea biodiversity. In addition to cultivated accessions, IITA holds about 1818 accessions of wild relatives. After the Convention on Biological Diversity (1992), IITA's cowpea collections were placed under Article 15 of the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRF) thus making them available to the international community for research, food and agriculture.

6.3.1 *Germplasm Diversity*

Cowpea accessions maintained in the gene banks exhibit important phenotypic variations in qualitative traits such as plant type, seed coat color, flower color and quantitative agronomic traits such as yield, maturity or stress tolerance. Although most of the cultivated cowpea cultivars are erect to semi-erect, accessions with prostrate or climbing growth habits. Pods can be coiled, round, crescent or linear. Peduncles range from <5 cm to >50 cm long in the cultivated cowpea species. Porter et al. (1974) described 6 different patterns of pigmentation of flower and pod, 62 eye colors and 42 eye patterns on the seeds. The maturity cycle of the crop varies from 50 days to more than 120 days. Studies of diversity in cowpea germplasm using morphological and physiological traits have been reported in several publications (Egbadzor et al. 2014a; Ehlers and Hall 1996; Fery and Singh 1997; Perrino et al. 1993).

Recent studies describing cowpea germplasm diversity are based on the analysis of molecular markers. All types of molecular markers are used in the diversity analysis in cowpea. Allozymes (Panella and Gepts 1992; Pasquet 1999, 2000; Vaillancourt et al. 1993), seed storage proteins (Fotso et al. 1994; Opong-Konadu et al. 2005) and chloroplast DNA polymorphisms (Vaillancourt and Weeden 1992) were explored to describe the genetic relationships of some cowpea germplasm. Other markers employed in diversity studies include restriction fragment length polymorphisms (RFLP) (Fatokun et al. 1993), random amplified polymorphic DNA (RAPD) (Ba et al. 2004; Diouf and Hilu 2005; Fall et al. 2003; Mignouna et al. 1998; Nkongolo 2003; Xavier et al. 2005; Zannou et al. 2008), amplified fragment length polymorphisms (AFLP) (Fang et al. 2007), DNA amplification fingerprinting (DAF) (Simon et al. 2007), simple sequence repeats (SSRs) (Asare et al. 2010; Li et al. 2001; Ogunkanmi et al. 2008; Uma et al. 2009; Wang et al. 2008; Xu et al. 2010) and sequence tagged microsatellite sites (STS) (Vir et al. 2009). More recently single nucleotide polymorphism (SNP) markers described as more effective in diversity assessment compared with other markers such as AFLP and SSR (Acquaah 2007; Egbadzor et al. 2014b; Varshney et al. 2007) have been used in cowpea. Huynh et al. (2013) used SNP to study the structure of cowpea landrace and wild relative populations from African and non-African countries. SNPs have also been used in the estimation of genetic diversity and population structure of cowpea (Fatokun et al. 2018; Xiong et al. 2016).

Despite the existence of natural hybrids between wild and cultivated cowpea genotypes, and the wide variation in phenotypic traits among cowpea accessions, a narrow genetic variability is observed in the cultivated gene pool (Ehlers and Hall 1997). In several studies that evaluated genetic variability based on molecular markers, a single domestication event in the cowpea is considered the basis of the narrow genetic variability of the crop (Asare et al. 2010; Ba et al. 2004; Coulibaly et al. 2002; Padulosi and Ng (1993); Pasquet 2000) attributed the low genetic divergence in cowpea to the self-pollinating nature of the crop.

6.3.2 *Cultivar Characterization and Phylogeny*

Cowpea cultivars exhibit variable features including morphological, agronomic, physiological and molecular expressions. The collection and documentation of these specific characteristics are very valuable for breeding programs. Cultivar characterization adds value to the gene bank and to the breeding programs as the information generated guides the user to request specific cultivars.

The real center of origin for cultivated cowpea still remains hazy. Early reports have indicated Africa as area of domestication of cowpea, given the exclusive presence of wild cowpea relatives (Steele 1976). Faris (1965) using literature and extensive work involving morphological descriptors concluded that West or Central Africa is the center of domestication of cowpea while Coulibaly et al. (2002) using molecular markers presented northeastern Africa as area of early domestication of cowpea. The theory of West African center of origin is more widely accepted (Baudoin and Maréchal 1985; Maréchal 1978; Ng 1995). However, efforts are needed to clarify the dispersal of cultivated cowpea to other regions of the world, the region of first domestication and sub-domestication (Xiong et al. 2016).

Using a collection of cowpea landraces and wild annual cowpeas from both East and West Africa, Huynh et al. (2013) showed that there are two major gene pools for cultivated cowpea in Africa. Landraces from western Africa and eastern Africa form gene pools 1 and 2, respectively. These landraces are similar to the wild relatives available in the same regions. Therefore domestication processes responsible for the existence of the two gene pools occurred differently. These authors pointed out that landraces within Africa presented lower total genetic variation than landraces outside Africa. Xiong et al. (2016) found three well-differentiated genetic populations and admixtures associated with the regions and countries from where the cowpea cultivars were collected (Table 6.1). These authors also concluded that West and East of Africa are the first domestication regions of cowpea while India is a sub-domestication region.

Table 6.1 Regions and countries with same genetic populations and admixtures

Grouping	Regions	Countries
Cluster I	North America, Latin America, Oceania, Central and East Africa, India and South Africa	Asia (Afghanistan, Iran, Pakistan, Turkey, China), West Africa (Cameroon, Niger) and Europe (Hungary) plus the American cultivars
Cluster II	West Africa	South Africa, India and USA
Cluster III	The American, East Asian, Central West Asian, and European cultivars	Latin America (Brazil, Guatemala, Mexico, Paraguay), Southern Africa (Botswana, Mozambique, Zimbabwe), Central East Africa (Kenya) and Oceania (Australia).

6.3.3 Genetic Resources Conservation Approaches

Preventing the loss of agricultural biodiversity is of the highest global priority given the importance of genetic resources in world food security. A framework for the efficient and effective ex-situ conservation of globally important collections of cowpea was initiated in 2010 with key stakeholders through the support of Global Crop Diversity Trust and the leadership of IITA. An extensive survey on cowpea genetic resources conservation and use (collections, facilities, human resources, ongoing research, networks) was conducted in different institutes across various countries. From the data compiled, an international group of experts was constituted to discuss the state of cowpea conservation and use in Africa. A set of recommendations was made. Dumet et al. (2012) summarized these recommendations which are related to safe conservation of each unique *Vigna* accession, its importance and diversity, and the formation of global information portals.

Over 59,000 accessions of cowpea and other *Vigna* spp. (including wild cowpea relatives and other cultivated *Vigna* other than cowpea) are being maintained in ex situ conditions. International seed processing standards need to be followed to ensure safety of the collections. For cowpea and other *Vigna* germplasm, optimal processing includes artificial dehydration (down to 8% on average), germination tests prior to storage, adequate packaging for medium (5 °C) and/or long-term storages (−20 °C) and monitoring viability at regular intervals during storage. Given the difficulties of many countries to comply with these standards, the need for maintaining at least one copy of each unique accession under international standard storage conditions was recommended (Dumet et al. 2012). Germplasm regeneration procedures need to meet standards that insure genetic integrity and health; Dumet et al. (2008) reviewed these standard regeneration guidelines.

6.3.4 Cytogenetics

The genome size of cowpea is relatively small (620 Mb) consisting of $2n = 2x = 22$ chromosomes (Arumuganathan and Earle 1991). The chromosomes are described as extremely small. Detailed descriptions of the cowpea karyotype have been reported. Pachytene bivalents, cells in mitotic prometaphase and in metaphase, were used in the development of karyotypes. In addition, chromosomal banding patterns, karyotype comparisons among wild cowpea species, chromosomal distribution of ribosomal DNA (rDNA), a centromeric repetitive DNA family and Ty1-copia-like retrotransposable elements have been previously reported (Barone and Saccardo 1990; Pignone et al. 1990; Saccardo et al. 1992). While conducting a karyotypic analysis of mitotic chromosomes of 11 wild taxa of *Vigna unguiculata*, Venora and Padulosi (1997) reported a low degree of karyological variability despite the high morphological variability in cowpea.

As in some legumes, such as common bean, 18S–5.8S–25S and 5S rDNA distributions were observed in cowpea (Galasso et al. 1995) which have 18S–5.8S–25S rDNA at chromosomal termini and 5S rDNA proximally located on the same chromosome and have also centromere-specific satellite repeats. The identified Ty1-copia-type retrotransposons are dispersed relatively uniformly across all chromosomes except in centromeres and subtelomeres (Galasso et al. 1997). From molecular cytogenetic studies, Iwata-Otsubo et al. (2016) found very distinct chromosomal structures in cowpea that require further examination. Understanding chromosome structure and the distribution of a few of the major repeat families will help in the ongoing genome sequencing project. In addition, cowpea can benefit from progress made in the cytogenetics of common bean (*Phaseolus vulgaris*) a legume that has the same number of chromosomes and genome size as cowpea (Vasconcelos et al. 2014).

6.4 Molecular Breeding

The advent of genomic revolution, fast advances in genotyping capabilities and bioinformatics provide modern approaches which have the potential to accelerate the development of improved cowpea cultivars. Using conventional approaches requires more than a decade to complete the delivery of improved lines mostly due to several phenotypic evaluations and performance trials characterized by complex, specialized conditions, techniques and skills needed for selection of best lines. Molecular marker-assisted selection (MAS) uses markers that are linked to traits or to estimates of genotypic effects (QTL) instead of phenotypic measurement alone, and it provides a powerful and potentially cost- and time-saving avenues to increase the rates of genetic gain in plant breeding programs (Ehlers et al. 2012). Substantial development of genomic resources in cowpea has occurred recently and some applications of molecular breeding are being recorded in cowpea breeding programs (Fig. 6.3).

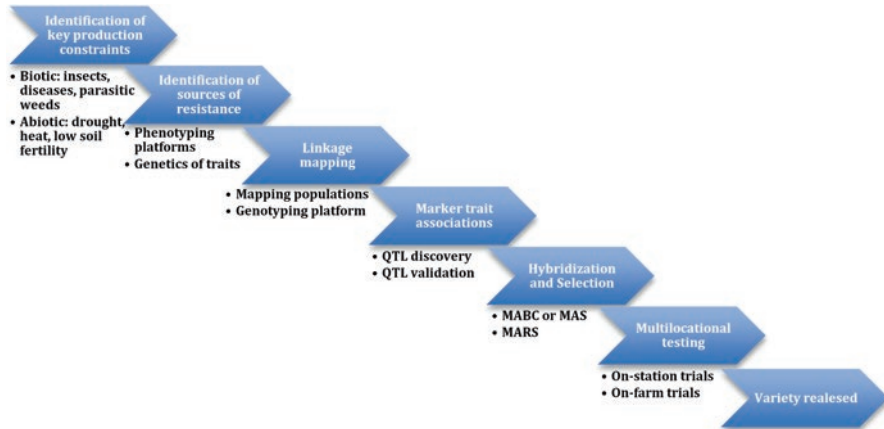


Fig. 6.3 Scheme of molecular breeding in cowpea. (Source: Boukar et al. 2016)

6.4.1 High-Throughput Genotyping Platform

Using methylation filtration (MF) technology, Timko et al. (2008) reported the sequencing and analysis of the gene-rich, hypomethylated portion of the cowpea genome characterized by more than 250,000 gene-space sequence reads (GSRs). The data generated provided an excellent starting point for both marker development and comparative genomics.

Efforts to develop microsatellite (SSR) markers needed by breeders in the implementation of modern breeding have been reported by several research groups. Gupta and Gopalakrishna (2010) have described the identification and development of unigene-based SSR markers in cowpea. These authors characterized and validated 102 SSRs out of 1071 SSRs found in 15,740 cowpea unigene sequences available from the National Center for Biotechnology Information (NCBI) database. Using de novo transcriptomic analysis of cowpea, Chen et al. (2017) identified valuable sets of SSR markers to be validated and used in different genetic and breeding studies.

Through the Generation Challenge Programme's (GCP) Tropical Legumes I project, the University of California Riverside (UCR) and partners developed a high-throughput genotyping platform based on the Illumina GoldenGate Assay for 1536 SNP loci. This resource represents 1536 expressed sequence tag (EST)-derived SNPs (Muchero et al. 2009a). KBiosciences in the UK converted about 1000 mapped SNPs from this platform for use with the single-plex KBiosciences KASPar genotyping platform. This made the platform more readily available, flexible and affordable to the cowpea breeding community (Ehlers et al. 2012).

Recently Muñoz-Amatriaín et al. (2016) reported resources developed from an IITA developed line, IT97K-499-35, and 36 diverse accessions leading to the development of an Illumina Cowpea iSelect Consortium Array, a genotyping assay for 51,128 SNPs.

6.4.2 High-Density Genetic Maps

For the implementation of modern molecular breeding program, consensus genetic linkage maps are key required genomic resources. Boukar et al. (2016) listed several linkage maps developed for cowpea. The first comprehensive cowpea genetic linkage map in terms of number and type of markers was developed by Ouédraogo et al. (2002). The total number of markers on this map is 441. Muchero et al. (2009a) reported the first cowpea consensus genetic linkage map, which consisted of 928 markers spanning 11 linkage groups over a total map size of 680 cM. Genetic maps from 6 RIL populations were merged to build this consensus map. Lucas et al. (2011) reported its improved version with 33% more bins, 19% more markers and had an improved order compared to the first consensus genetic map. Recently Muñoz-Amatriaín et al. (2016) used 5 biparental RIL populations to develop a consensus genetic map having 37,372 SNP loci mapped to 3280 bins with higher average density of 1 bin per 0.26 CM.

6.4.3 Phenotyping and Marker-Trait Association

The implementation of modern breeding requires high-throughput phenotyping platforms. Accurate phenotypic and genotypic data are also needed for the execution of an effective and efficient modern breeding. Screening protocols for both biotic and abiotic stresses require high levels of refinement to facilitate precise data measurements. Cowpea breeding programs are currently using the Breeding Management System (BMS) of the Integrated Breeding Platform (IBP) to design electronic field books that are uploaded into handheld devices (tablets, phones) to be used for data capture. In addition to the tablets, barcoding devices are being introduced in these breeding programs. These different tools will help in the reduction of errors and facilitate timely generation of accurate data. With the advent of advances in molecular marker technologies, precise phenotypic data are being used in combination with genotypic data to identify markers linked to target traits.

Recently, Boukar et al. (2016) reviewed extensively marker-trait association studies reported in the literature for cowpea. Several quantitative trait loci (QTL) were identified using different types of markers (Table 6.2).

6.4.4 Molecular Breeding Deployment

The genomic resources generated during implementation of the Tropical Legumes I project, have led to the initiation of molecular breeding of cowpea. Some of the strategies being adopted include marker-assisted backcross (MABC), marker-assisted selection (MAS) and marker-assisted recurrent selection (MARS).

Table 6.2 Mapping of some cowpea traits

Traits	Marker types	No. markers / QTL	Locations	References
Cowpea golden mosaic virus resistance	AFLP	3	Same linkage group	Rodrigues et al. (2012)
<i>Striga</i> resistance	AFLP	3–6	LG1	Ouédraogo et al. (2001)
	SCAR	2	LG 1	Ouédraogo et al. (2012)
	AFLP/ SCAR	4/1	Same linkage map	Boukar et al. (2004)
Bacterial blight resistance	SNP	3	LG3, LG5, LG9	Agbicodo et al. (2010)
Flower bud thrips resistance	AFLP			Omo-Ikerodah et al. (2008)
Seed size	SSR	6	LG1, LG10	Andargie et al. (2011)
Seed weight	RFLP	2	LG 2 LG6	Fatokun et al. (1992)
Seed weight	SSR	6	LG1, LG2, LG3, LG10	Andargie et al. (2011)
Seed size	SNP	10	LG5, LG7, LG2, LG6, LG8, LG10	Lucas et al. (2013b)
Charcoal rot resistance	SNP/ AFLP	9	LG2, LG3, LG5, LG6, LG11	Muchero et al. (2011)
Heat tolerance	SNP	5	LG2, LG7, LG6, LG10, LG3	Lucas et al. (2013a)
Drought-induced senescence	AFLP	10	LG1, LG2, LG3, LG5, LG6, LG7, LG9, LG10	Muchero et al. (2009b)
Maturity	AFLP	2	LG7, LG8	Muchero et al. (2009b)

Source: Adapted from Boukar et al. (2016)

MABC was conducted at IITA, Nigeria, at the Institut de l'Environnement et de Recherches Agricoles (INERA), Burkina Faso, the Institut Sénégalais de Recherches Agricoles (ISRA) Senegal and the Eduardo Mondlane University (EMU) Mozambique, for the introgression of *Striga* resistance (IITA, INERA, ISRA), seed size (INERA, EMU), drought tolerance and nematode resistance (EMU) into local cultivars or improved lines. Given that MABC is now routinely used in modern breeding programs, there is a high expectation that cowpea breeding programs will continue using similar strategies in their improvement activities.

Breeders at INERA and ISRA used MAS to develop improved lines with combined desirable traits from two crosses involving three elite parents, i.e. IT93K-503-1 x IT84S-2246 and Mouride x IT84S-2246, respectively. Using QTL information, an ideotype was constructed by aligning all favorable polymorphic marker alleles from among the two parents (Ehlers et al. 2012). The F_{2:4} breeding lines were genotyped and lines having the maximum number of favorable marker

alleles were selected. Further inbreeding of these lines and selection based on marker content led to phenotypic evaluations for target trait expression and replicated yield tests. Compilation of data needed for preparing the dossier to accompany application for their potential release as new cultivars is being carried out.

Under the TL I project, MARS scheme was also implemented in the participating countries which included Burkina Faso, Senegal, Mozambique and Nigeria. Elite by elite crosses were performed and selection indices based on grain yield and identified associated QTL were used to identify high yielding individuals with complementary favorable marker configurations. Intercross of lines with these complementary markers were performed to pyramid favorable alleles in individual background. After two to three cycles, lines with all favorable alleles were identified and are being tested for possible release.

6.5 Genetic Engineering

6.5.1 Cell and Tissue Culture Approaches

In in vitro plant regeneration two methods commonly used are somatic embryogenesis and organogenesis (Machuka et al. 2002). Plant hormones and other supplements added to the culture medium affect both methods. Several plant tissue culture techniques have been reported in cowpea to attempt to regenerate whole plants from various genotypes (Brar et al. 1999). These efforts are needed to implement gene transfer methodologies. Except for soybean, plant regeneration protocols in grain legumes have not been as reliable as for other crops. This is the reason why grain legumes are described as *recalcitrant crops* to in vitro manipulations (Monti et al. 1997). Concerted efforts were performed to discriminate new buds or induce their multiple proliferations by using different explant sources and several combinations of natural or synthetic plant growth factors.

Shoot Differentiation Using a modified B₅ medium containing coconut water from fresh local coconuts and a high cytokinin concentration, scientists at IITA reported that about 33% of explants (primary leaves and hypocotyl isolated from germinating seeds) differentiated some shoots (Monti et al. 1997). According to these authors, the histology of these explants showed that a strong proliferation occurred on the explant surface, at the epidermal level, where callus was formed. The fact that only the basal parts of young leaflets were able to produce shoots was due to the presence of formed meristems.

Somatic embryogenesis is used for most genetic transformation protocols with recalcitrant legumes given that embryogenic tissues are very prolific and usually originate from single cells (Hansen and Wright 1999). Ganapathi and Anand (1998) reported that induction of somatic embryos occurred in suspension cultures of calli derived from cowpea seedling leaf explants.

Kononowicz et al. (1997) developed a morphogenic system for cowpea using embryonic axis and cotyledonary base explants. Shoot meristem regeneration and morphogenesis were carried out on MS medium (Murashige and Skoog 1962) containing N⁶-benzyl adenine (BA) and NAA (naphthalene acetic acid) at different concentrations (10 μ M and 5 μ M for BA; 0.2 μ M and 0.05 μ M for NAA) with a pH of 5.8. In addition, media were supplemented with modified B5 vitamins (Gamborg et al. 1968). Through morphogenesis, up to 15 shoots can be regenerated from a single primary explant. Shoots obtained from cotyledon segments and embryonic axes cultures can be easily elongated and rooted. Rooting of cowpea plantlets is most successful in hormone-free MS medium with the addition of 1 mg/L of indole-3-acetic acid (IAA) or 0.05 mg/L of NAA.

Multiple Bud Regeneration Experiments were conducted in order to induce multiple bud proliferation from highly morphogenic cowpea tissues to provide a different approach to plant differentiation with the aim to obtain plants from transformed tissues (Monti et al. 1997). These authors also reported that cotyledon segments and embryonic axes from embryos of different ages of various cowpea genotypes were placed in media containing high concentrations of BAP (3–6 mg/L) and a low concentration of auxin by scientists at Purdue University. Cotyledon explants developed shoots at 50% frequency after 3 weeks of in vitro culture (Monti et al. 1997). They further reported that the herbicide thidiazuron was used as a plant growth regulator to induce multiple bud proliferation from cotyledonary and apical nodes. The best results in terms of frequency of multiple bud proliferation from apices were obtained with cvs. Cornetto and TVu9062 with an average of 87– 85%, respectively.

Multiple shoot formation was attempted through organogenesis from different explants including roots, stem pieces, intact immature cotyledons or protoplasts derived from immature cotyledons, leaves and stem apices (Machuka et al. 2002). At IITA, organogenesis was obtained in several genotypes including IT90K-277-2, IT89KD-288, IT83F442, IT86D-1010, IT93K-624, Vita3 and Ife Brown, when cultured in vitro (Machuka et al. 2002).

These attempts to regenerate cowpea were not successful enough to be used in the genetic transformation of the crop. The first successful genetic transformation of cowpea was reported by Popelka et al. (2006) using regeneration by organogenesis of a wide range of explants on culture media with moderate levels of cytokinin BAP. Longitudinally bisecting seeds through their embryonic axes and by removing both shoot and root apices produced the best explants for multiple shoot formation. These authors have conducted intensive cultivar and culture media comparisons to confirm that (1) there are special media preferences for particular cultivars, (2) the number of independent shoots was not affected by the polyamine growth regulator putrescine (but their development), (3) only deformed shoots could be obtained even at concentrations of thidiazuron as low as 0.1 μ M and (4) the effective way to overcome the difficulties in rooting of cowpea was to graft rootless shoots onto cowpea seedlings.

6.5.2 Transformation Systems

In crop improvement, genetic engineering becomes an attractive option when facing intractable problems which conventional breeding methods have not been able to resolve. The legume pod borer (*Maruca vitrata*) causes significant grain yield loss in cowpea if the crop is not protected with an appropriate synthetic insecticide. At IITA and partners breeding programs, concerted efforts were made to identify which among the more than 15,000 cultivated and wild relatives' germplasm lines show resistance to this troublesome insect pest of cowpea. According to Jackai and Daoust (1986), despite availability of a large number of accessions in the cowpea germplasm and cultivars only moderate levels of resistance to the pod borer and pod bugs have been detected. Wild cowpea relatives that show high levels of resistance to the insect pests (Singh et al. 1990) are not cross compatible with the cultivated cowpea (Fatokun 2002). Some *Baccillus thuringiensis* (*Bt*) protoxins were tested in artificial diets for their efficacies on maruca, the main culprit in damage to cowpea, and *CryIab*, *CryIC* and *CryIIA* were found to be most potent in curtailing the growth and development of the insect larvae (Machuka 2002). Successful in vitro culture is essential for genetic transformation in plants. Some efforts have been made towards developing a robust genetic transformation system for cowpea with little success. Cardi et al. (1990) isolated protoplasts from cowpea leaves and these were cultured in MS medium with 3% sugar and a pH of 5.8. Plating efficiencies of protoplasts varied with the cowpea lines used. Protoplasts proliferated and formed calli. Roots developed from the calli but no shoots were obtained. Kononowicz et al. (1997) reported successful transformation of cowpea using microprojectile bombardment and cocultivation with *Agrobacterium tumefaciens*. They first tested several cowpea plant parts as explants to establish regeneration, including shoot tips and axillary buds excised from 6–7 day old seedlings. The authors settled for embryonic axis and cotyledonary base as explants. Transient expression of a β -glucuronidase reporter gene was established in the transformed tissues. Organogenesis was observed when transformed tissue was placed in culture medium containing BAP. Popelka et al. (2006) were the first to report successful transformation of cowpea with the *Bt* gene that confers resistance against the larvae of the *Maruca vitrata* pod borer. Of the many cowpea lines tested by these authors for their ability to be genetically modified, only IT86D-1010 showed positive response. Genetically modified cowpea plants carrying the *Bt* gene have been evaluated in confined field trials in Nigeria and Burkina Faso (AATF 2016). Results showed good levels of expression of the gene in the cowpea lines as maruca larvae failed to cause damage to transgenic cowpea plants. The *Bt* gene has been transferred using the backcross method of breeding to some already released cowpea cultivars preferred by farmers in Nigeria. The protocol followed in producing transgenic cowpea, as described by Higgins et al. (2012), was able to transform cowpea with a gene from the common bean (*Phaseolus vulgaris*) that codes for α -amylase inhibitor. Three of four selected transgenic plants showed complete protection of the plants against the cowpea storage pest beetle *Callosobruchus maculatus*. With the

successes reported by Popelka et al. (2006) and Higgins et al. (2012) the genetic transformation of cowpea with desirable genes has become a reality.

6.6 Mutation Breeding

6.6.1 Conventional Mutagenesis

Mutation breeding is most often carried out on already existing elite crop cultivars. This derives from the fact that induced mutations usually affect simply inherited traits such as disease resistance, color, earliness to flower, etc. This method of breeding is therefore embarked upon especially when a desired simply inherited trait is lacking in an improved crop cultivar. The expectations are that induced mutation will result in plants expressing the desired traits. The International Atomic Energy Agency (IAEA) in collaboration with the Food and Agriculture Organization (FAO) supports the implementation of mutation breeding in its member states through the development and utilization of technologies that induce mutations of plants. Among these technologies, gamma irradiation and X-rays are the most commonly used. Adaptation to harsh conditions (drought, salinity, low soil fertility), enhancement of crop nutritional value and resistance to diseases and pests are the usual targets of the supported mutation breeding. Table 6.3 lists the released mutant cowpea cultivars created through the efforts of IAEA. Few reports are available in cowpea cultivar development using induced mutation. Physical and chemical mutagens have been applied for inducing mutagenesis in cowpea. Ojomo (1973) reported gamma radiation treatment caused numerous chromosome disorders as well as high genetic variation for number of pods per plant and grain yield in cowpea. Adekola and Oluleye (2007) irradiated an improved cowpea breeding line IT84S-2246 using two levels of gamma radiation – 196 and 245 Gy. Some plants in the M2 generation from seeds treated with 245 Gy dose were observed to have leaflets terminating in tendrils, broad leaflets, pigmented pods and plant parts and carried the pods above the canopy. Dwarf plants were recovered from the seeds treated with 196 Gy. Girija et al. (2013) irradiated cowpea seeds with gamma rays (20, 25, 30 KR) and as well treated with ethyl methyl sulfonate (EMS) (20, 25, 30 mM). In the M6 generation, several unique mutants were detected. The highest frequency of mutation was observed in flower color among progeny of seeds treated with 25 mM EMS. The most distinct mutants identified were associated with flower color, altered seed size, shape and seed coat color. Horn et al. (2017) irradiated four elite cowpea cultivars in Namibia. Four mutants were selected at M6 generation, which, at M7, showed broad adaptation in the country. They also had the highest grain yield that ranged from 1.95–2.83 mt ha⁻¹. Most of the high performing mutants were derived from seeds of one of the four cultivars (cv. Shindimba) that were irradiated. An observed interesting trait among the mutants was straight pod shape which farmers in the country prefer. Horn et al. (2016) irradiated three improved cowpea cultivars,

Table 6.3 List of released mutant cowpea in different countries

Cultivar name	Country	Registration year	Short description
Uneca-Gama	Costa Rica	1986	Developed by irradiation of seeds with gamma rays (100 Gy). Main attribute of mutant cultivar is high yield
V16 (Amba)	India	1981	Developed by treatment with chemical mutagen DMS. Main attributes of mutant cultivar are high yield, resistance to fungal and bacterial diseases
V37 (Shreshtha)	India	1981	Developed by treatment of seeds with chemical mutagen DMS. Main attributes of mutant cultivar are high yield, high vegetative growth, suitable also as fodder
V38 (Swarna)	India	1981	Developed by treatment of seeds with chemical mutagen DMS. Main attributes of mutant cultivar are high yield, early maturity, synchronous flowering, better quality pods and grains, resistance to diseases
V240	India	1984	Developed by treatment with chemical mutagen DMS. Main attributes of mutant cultivar are high yield, resistance to fungal, viral and bacterial diseases
Co 5	India	1984	Developed by irradiation of seeds with gamma rays (300 Gy). Main attributes of mutant cultivar are more nutritive forage cowpea, high yield (16%), comparable for intercropping with fodder cereals
Cowpea-88	India	1990	Developed by irradiation of F1 generation from cross Cowpea-74 X virus resistant strain H-2. Main attributes of mutant cultivar are high grain yield, high green fodder yield, resistance to yellow mosaic virus
ICV 11	Kenya	1985	Developed by irradiation of seeds with gamma rays. Main attributes of mutant cultivar are semi-erect, large leaves, green stem, green pods, maturity in 65 days, yield 1100 kg/ha, resistance to cowpea aphid
ICV 12	Kenya	1985	Developed by irradiation of seeds with gamma rays. Main attributes of mutant cultivar are higher yield than ICV 11 and resistance to cowpea aphids
TRC77-4 (Kalleshwari)	India	2007	Improved traits: yield
COCP 702 [=CoVu 702 & CO(CP) 7]	India	2002	Developed by irradiation of seeds with gamma rays (200 Gy). Main improved attributes of mutant cultivar are high yield and good quality
Gujarat cowpea-1	India	1984	Improved traits: yield, early maturity, root knot resistance

(continued)

Table 6.3 (continued)

Cultivar name	Country	Registration year	Short description
CBC5	Zimbabwe	2017	Developed through mutation induction using gamma irradiation of CBC1 seeds at 150 Gy. Main improved attributes are high grain and fodder yield (at least 18% yield advantage), good dynamic stability across environments (sites and seasons) and increased seed size (at least 8% seed size increment over parent)

Source: Adapted from Joint FAO/IAEA Mutant Varieties Database (2019)

namely IT81D-985, IT89KD-245-1 and IT87D-453-2, and detected substantial genetic differences among these cowpea genotypes following mutagenesis. Differences in flowering ability, days to maturity, flower and seed colors and grain yields were observed among the mutants. They identified and isolated ten phenotypically and agronomically stable novel mutants in M6 generation from each of the three cultivars. These promising mutant lines were recommended for tests across several agro-ecologies for their adaptability. Those with superior performance were to go for large-scale production or used as parents in the Namibian cowpea breeding program. In Nigeria, Odeigah et al. (1998) treated seeds of two cowpea breeding lines with three mutagens: gamma radiation, EMS and NaN_3 . The three lines responded differently to the mutagens. As expected, the mutagens had both desirable and deleterious effects on the plants resulting from treated seeds. An array of mutants was observed among plants in the M2. The observed mutants showed differences in plant morphology and physiology. They further reported that induced mutations resulted in plants with branched peduncles, pigmentation of pods and plant parts, male sterility, early maturity, as well as resistance to cowpea storage weevils and aphids. Some other observable traits among the mutant plants were stunting in growth, twining stem and spreading growth habit none of which is present in any of the untreated lines. The protein content in grains of some mutants were found to be higher by up to 13.3%, in a mutant from IT84E-124, and 13.64% from Vita 7, when treated with 1.0 mM NaN_3 . Olasupo et al. (2016) applied UV light on cowpea pollen with the aim of inducing mutations. The results showed that for the duration of exposure of pollen to the UV rays no visible morphological change was induced in the progeny derived from seeds using the pollen for pollination.

In India at least seven cowpea cultivars improved using mutation breeding procedures were released between 1981 and 2007 at the Bhabha Atomic Research Centre, Trombay, (Punniyamorthy et al. 2007). These mutant cultivars are characterized by high grain yield and early or medium maturity. Others have increased seed size, resistance to yellow mosaic virus or increased fodder yield. From the foregoing, it is evident that cowpea cultivar development has benefited from mutation breeding in different countries and the cultivars have in most instances met the expectations of farmers and consumers.

In cowpea, seeds and pollen grains have been treated with chemical and physical mutagens for the purpose of inducing mutations in the crop. There is however no report on the induction of mutagenesis in cowpea using in vitro cultured plant tissues. This is not surprising because totipotency is very difficult to achieve in cowpea. All of the reported transgenic events in cowpea have been carried out by coculturing existing plant tissues with *Agrobacterium* carrying the genes of interest (see Sect. 6.5 on genetic engineering above).

Ojomo (1973) reported that exposing cowpea seeds to gamma radiation resulted in numerous chromosomal disorders. However, most of the other above reports on mutation induction in cowpea did not mention whether the mutation induced intergenic (occurring within the DNA) or structural i.e. intragenic disorders that occur on the chromosome such as deletions, inversions, translocations or duplications, changes in chromosome numbers such as polyploidy, aneuploidy or haploidy. In a review of mutation breeding in different crops, Oladosu et al. (2016) concluded that base substitutions, a term implying nucleotide changes involving substitution of one base for another, are among the different types of mutations at the molecular level. According to these authors, base substitutions can happen through mis-pairing during replication of the base analogue in the treated DNA.

6.7 Hybridization and Heterosis

Cowpea is a highly self-pollinated crop. The cleistogamous flowers, which open for only 1 day, are large and showy which make hybridization relatively easy. The level of outcrossing in cowpea is generally low (Fatokun and Ng 2007) and could vary with the environment where the crop is grown. The anthers open to release pollen grains contained therein on the day the flower opens. The stigma is however receptive from a day before anthesis and remains so until day of flower opening. Insects, especially bumble bees, are the major cowpea pollinator. There is no clear evidence that color influences insect visits to flowers in cowpea. Leleji (1973) reported that bumble bees tended to visit purple colored flowers more whereas honey bees visited white colored flowers more frequently.

Strong cross-incompatibility exists between cowpea and many of its wild relatives, particularly those outside the section *Catiang*. Even among members of section *Catiang* difficulties are encountered when making crosses between some of them. For example, Fatokun and Singh (1987) had to use embryo rescue to enhance crossing between cultivated cowpea and *Vigna unguiculata* ssp. *pubescens*. However, the F1 hybrids showed partial fertility. All attempts so far made to cross *V. vexillata* to cowpea have not yielded positive results. *Vigna vexillata* has genes that confer resistance to many of the insect pests that cause serious yield losses in cowpea. Barone and Ng (1990) examined the causes of the incompatibility between cowpea and *V. vexillata*. They observed that pollen tube growth through the styles in interspecific crosses was arrested in stigmatic tissues and only 15–20% of ovules were fertilized. Embryos developed following interspecific hybridization did not go

beyond the globular stage before they started to collapse. Even when pods resulting from interspecific hybridizations were retained on the plants until maturity subsequent treatment with auxin did not result in any success (Fatokun 2002).

Varying levels of heterosis have been reported in cowpea. Agble (1972) reported seed size heterosis in crosses made between four local Ghanaian cowpea cultivars. Bhaskaraiah et al. (1980) in a 10×10 set of diallel crosses found relatively high heterosis for grain yield and pods per plant but lowest for 100-seed weight. Bhushana et al. (2000) generated 36 hybrids from line tester crossing design and found mid-parent heterosis of 112.4% for pods/plant, 105.32% for seed yield per plant and 30.31% for pod length. Mak and Yap (1977) evaluated heterosis for protein content in a diallel cross involving seven cultivars of yardlong bean. Heterosis ranged from 40.7% to 63.2% but only three F1 hybrids showed significantly higher parent values. Number of pods per plant showed the highest heterosis among yield components while seeds per pod, seed weight and pod length showed relatively low levels of heterosis. In spite of the reported levels of heterosis in cowpea no record is available of any hybrid cultivar released for cultivation. The drawback to developing hybrid cultivars in cowpea can be attributed to the high level of self-pollination in the crop. In addition, genetic but not cytoplasmic male sterility has been reported in cowpea (Ladeinde et al. 1980; Sen and Bhowal 1962). Cytoplasmic male sterility would be advantageous to a successful hybrid cultivar development program in cowpea.

6.8 Conclusions and Prospects

Cowpea possesses a high potential to play a strategic role in tackling the complex challenges of hunger, malnutrition, environmental sustainability, climate change and increasing food prices, confronting the global community in coming decades (Widders 2012). In the dry Sahel and Savannah areas where the crop is being largely produced, increasing human population is threatened by food and nutritional insecurity. Cowpea can contribute to alleviate hunger and malnutrition as it has demonstrated its ability to grow in these regions and to be the cheapest source of plant protein. Cowpea fodder also plays an important role as animal feed. Haulms constitute nutritious feed for animals, most especially for ruminants. Another characteristic of cowpea is its ability to fix atmospheric nitrogen through symbiosis with bacteria living in its root nodules. This contributes to the sustainability of the agricultural systems. The soils in the cowpea-growing areas are greatly degraded by the inability of farmers to provide adequate amounts of fertilizers to ameliorate the problem of poor soil fertility. Numerous factors including biotic and abiotic factors are limiting the production of this important crop. It is necessary that adequate attention be given to addressing the constraints that have continued to hamper the productivity of cowpea in the fields of SSA farmers, as this will enhance the sustenance of the contributions of the crop to food and nutrition security. Lack of sources of genes for some traits of interests (e.g. insect resistance) is the major limitation to

fully implementing conventional breeding in cowpea. In addition, conventional breeding is ineffective in realizing adequate genetic gain that could allow farmers in SSA to benefit optimally from cowpea production. The advent of new technologies and approaches such as molecular breeding and gene editing offer opportunities to modernize cowpea improvement programs. In recent times, several donor communities have advocated and supported the implementation of modern breeding approaches in the Consultative Group on International Agricultural Research (CGIAR) centers and some National Agricultural Research Services (NARS) programs. Cowpea programs at IITA and NARS are currently involved in the modernization of breeding approaches. With the continued support of these initiatives, there is hope that cowpea breeding programs will soon become more efficient and effective and sustainable production of cowpea will therefore be significantly increased in SSA.

Acknowledgements The authors would like to express sincere gratitude to all donors who have supported those of our activities reported in this review. We also wish to thank all colleagues at IITA and the collaborating National Agricultural Research Services who have provided the information summarized in [Appendix I](#).

Appendices

Appendix I: Main Research Institutions Relevant to Cowpea

Institution	Area of specialization	Research activities	Contact information including website
International Institute of Tropical Agriculture (IITA)	Genetic Improvement	Activities were carried out in order to develop improved lines with (1) high yield potential, (2) resistance /tolerance to both biotic and abiotic stresses, (3) good adaptation to monocropping and intercropping systems and (4) grain characteristics preferred by consumers and processors	IITA Kano o.boukar@cgiar.org IITA Ibadan c.fatokun@cgiar.org http://www.iita.org
	Natural resource management	Activities were carried out for development of useful methods to supply P nutrient on cowpea by using local materials, e.g. rock phosphate and organic matters. In addition, the utilization of arbuscular mycorrhizal fungi is also being tested as P uptake enhancer by cowpea	IITA Lusaka Zambia k.suzuki@cgiar.org http://www.iita.org

(continued)

Institution	Area of specialization	Research activities	Contact information including website
	System Research / Agronomy	Development of (1) village-based/ commune-based dissemination scheme for improved cowpea (AVEC-BF), (2) single-seed protein content evaluation technique, (3) high protein content management technique, and evaluation of yield-gap analysis in Burkina Faso	IITA Ibadan Nigeria h.ishikawa@cgiar.org http://www.iita.org
	System Research / Agronomy	Conduct research to identify best crop management (Planting dates, population, cropping sequence, fertilizer use) techniques that exploit the potential of improved cowpea cultivars and close the yield gap between what is obtained in the research stations and farm fields. Evaluate performance of cowpea cultivars in both sole and different intercropping systems since most cowpea is grown in intercropping in West and Central Africa. The use of cropping systems models to simulate cowpea growth and yield in diverse ecologies and cropping systems and management practices is evaluated in the Agronomy unit	IITA Kano a.kamara@cgiar.org http://www.iita.org
	IPM research	Pest management research focusing on West Africa is addressing critical insect pests such as the legume pod borer <i>Maruca vitrata</i> , for which a range of bio-pesticides and biological control agents have shown promising results. For aphids and thrips, novel sources of host plant resistance from the IITA-mini core are currently being evaluated, and can be integrated with biological control approaches	IITA Cotonou Benin m.tamo@cgiar.org IITA Kano a.togola@cgiar.org http://www.iita.org

(continued)

Institution	Area of specialization	Research activities	Contact information including website
	Genetic Resource	The IITA gene bank holds the world's largest and most diverse collection of cowpeas with 15,122 unique samples from 88 countries, representing 70% of African cultivars and nearly half of the global diversity and about 2000 accessions of cowpea wild relatives. Activities undertaken with IITA breeders include genotyping of the core collection (with University of California Riverside) and development of a trait based subset for drought and heat tolerance (with ICARDA). Wild relatives are being evaluated under a project supported by the Global Crop Diversity Trust	IITA Ibadan m.abberton@cgiar.org
	Socio-economics	Comprehensive household and plot level surveys are conducted in order to assess the adoption and ex-post impacts of improved cowpea cultivars on yield, farm income, food security and poverty. Gender differentials in adoption and impacts are also taken into consideration when conducting these assessments. Quantify ex ante poverty impact of improved cowpea technologies across drylands of Sub-Saharan Africa and South Asia	IITA Malawi a.alene@cgiar.org j.manda@cgiar.org s. gbegbelegbe@cgiar.org
Institut de l'Environnement et des Recherches Agricoles (INERA), Kamboinse and Saria, Burkina Faso	Genetic Improvement	Activities were carried out in order to develop improved lines with (1) high yield potential, (2) resistance /tolerance to both biotic and abiotic stresses, (3) good adaptation to monocropping and intercropping systems and (4) grain characteristics preferred by consumers and processors using classic and modern breeding tools	INERA/ CREAM-Kamboinse batiemo52@gmail.com INERA/DRREA-Sariahamasegua22@gmail.com

(continued)

Institution	Area of specialization	Research activities	Contact information including website
Institut de Recherche Agricole pour le developpement (IRAD), Maroua, Cameroon	Genetic Improvement	Activities were carried out in order to develop improved lines (1) well adapted to the Soudano Sahelian zone of Cameroon, (2) high yield potential and preferred by farmers, (3) resistance / tolerance to pest weeds (<i>Striga</i>), insects (aphids, thrips) and diseases (<i>Colletotrichum</i>)	IRAD Maroua (Cameroon) sobdagonne@gmail.com http://www.iradcameroun.org
Savanna Agricultural Research Institute (SARI), Tamale, Ghana	Genetic Improvement	Activities includes, developing improved lines with (1) high yield potential, (2) resistance /tolerance to both biotic and abiotic stresses, (3) good adaptation to monocropping and intercropping systems and (4) grain characteristics preferred by consumers and processors using both molecular and conventional techniques. Germplasm collection and evaluation. Yield evaluation of advance elite lines across multilocations and cultivar release and maintenance	Haruna_ mohammed67@yahoo.com Owusu owusuemmagh@yahoo.com Francis Kusi onkufra@yahoo.com
	On-farm Agronomy	Activities were carried out to test the performance of elite promising lines for release under the farmers' own environment and management	Julius Yirzagla yirzagla@yahoo.com Mahama Goarge Yakubu mgyakubu@yahoo.com
	IPM	Pest management research activities in Ghana include evaluating cowpea engineered with cry genes from <i>Bacillus thuringiencis</i> for control of the legume pod borer <i>Maruca vitrata</i> , survey of alternate host plants for <i>M. vitrata</i> as possible refugia for IRM in Bt-cowpea, evaluating cultivars for host plant resistance to thrips <i>Megalurothrips sjostedti</i> and also evaluating biorational botanical pesticides as a cheap source of pesticides for control of major insect pests of cowpea	Mumuni Abudulai mabudulai@yahoo.com S. K Asante skasante@yahoo.com

(continued)

Institution	Area of specialization	Research activities	Contact information including website
Institut d'Economie Rurale (IER), Cinzanna, Mali	Genetic Improvement	Breeding activities carried out in order to develop improved lines with:(1) high yield potential, (2) resistance /tolerance to both biotic and abiotic stresses, good adaptation to monocropping and intercropping systems and – 4 grain characteristics preferred by consumers and processors	SRA Cinzana diallo.sory@yahoo.fr
	Genetic Resource	The IER gene bank is not fully functioning. About 2000 accessions for the nine mandate crops (cowpea, sorghum, millet, groundnut). The cowpea mini gene bank is holding the largest and most diverse collection with more than 1000 accessions from different agro-ecologies in Mali	SRA Cinzana mousmandiaye@yahoo.fr
	IPM research	Identification of source of insects and disease resistance (parental lines) for cultivar development	SRA Cinzana zkouyate@yahoo.fr
	Agronomy	Conduct research to identify crop management (planting dates, population, fertilizer use). Evaluate performance of cowpea cultivars in sole and different intercropping systems since most cowpea is grown in intercrops in Mali	SRA Cinzana kmarcel59@yahoo.fr
	Seed production Unit	Production of breeder and foundation seeds to supply private seed companies; production of certified seeds to supply farmers	SRA Cinzana mousmandiaye@yahoo.fr
Institut National de la Recherche Agronomique du Niger (INRAN), Niamey, Niger	Genetic improvement	Creation of high yielding cultivars (grains, fodder) with tolerance to major biotic and abiotic stresses, adapted to mixed cropping	souleymanabdou@gmail.com masalif2000@yahoo.fr
	Socio-economics	Adoption studies, impact assessment	geribro@yahoo.fr
		Value chain	bokarmoussa@gmail.com
	Seed production	Production of foundation seeds to supply private seed companies; Production of certified seeds to supply farmers	salamiissoufou@yahoo.fr

(continued)

Institution	Area of specialization	Research activities	Contact information including website
	IPM	Laboratory and field screening of cultivars for tolerance to insects; Chemical and biological control of insects; Improved storage technologies	amadoulaouali@gmail.com
	Genetic resources	Management of the gene bank	issazakarym@yahoo.fr
Institute for Agricultural Research (IAR), Nigeria	Genetic Improvement	Activities were carried out in order to develop, release and register cowpea cultivars with (1) high yield potential, (2) resistance /tolerance to both biotic and abiotic stresses, (3) good adaptation to mono and intercropping systems	IAR Samaru Zaria mahammadlawan@yahoo.com binsaba@yahoo.co.uk mffaguji@hotmail.com
	Agronomy	Develop production guides in line with best recommended agronomic practices of the newly developed, and commercially released cowpea cultivars for recommendation to farmers	sojiolufajo@yahoo.com https://iar.abu.edu.ng/
	IPM research	Help in identifying source of insects and disease resistance (Parental lines) for cultivar development. Also, develop an Insects Resistance Management (IRM) plan(s) for an insect resistance cowpea cultivars developed for commercial use in Nigeria.	imutono@yahoo.com rsadamu@yahoo.com
	Genetic Resource	The IAR gene bank (currently not fully functioning) is designed to hold about 35,000 accessions for the nine mandate crops (Cowpea, Groundnut, Maize, Sorghum, Cotton, Castor, Jatropha, Sunflower and Artemisia). The cowpea mini gene bank is holding the country's largest and most diverse collection of cowpeas with more than 5000 accessions from 6 Nigerian agro-ecologies and 10 African countries, representing 60% of Nigerian cultivars, 40% of African cultivars and nearly 10% of the global diversity.	mahammadlawan@yahoo.com uwa6474@yahoo.com

(continued)

Institution	Area of specialization	Research activities	Contact information including website
	Seed production Unit	The IAR seed Production Unit is mandated for the production of foundation and certified seeds of the seven crops: cowpea, groundnut, maize, sorghum, cotton, castor, sunflower	almuh2013@yahoo.com
	Product Development Unit	The program is mandated for conducting researches: to determine the nutritional and biological value of released and candidate cultivars; assess the suitability of the cultivars for industrial processing and to improve the technology of local food processing. Also, monitor foods and feeds for toxic contaminants. The program participate generating new technologies and in training of both individuals and organization that are interested in using the technologies generated by the program	sanbugaje@gmail.com https://iar.abu.edu.ng/pages/prodctdeveloprech.html
	Socio-economics	Participatory Rural Appraisal for trait prioritization and inclusion in breeding objective, Adoption studies and Impact assessment	m.bellohassan@gmail.com
Federal University of Agriculture, Makurdi (FUAM), Nigeria	Genetic Improvement	Breeding activities carried out in order to develop improved lines with: (1) high yield potential, (2) resistance /tolerance to both biotic and abiotic stresses, (3) good adaptation to different cropping systems and (4) grain characteristics preferred by consumers and processors	FUAM uam.edu.ng lomoigui@cgiar.org lateeflekan@gmail.com
Institut Senegalais de Recherches Agricoles (ISRA), Bambey, Senegal	Genetic Improvement	Develop high yielding cowpea cultivars adapted to semiarid zones with resistance to relevant biotic constraints and good grain quality.	ndiaga.cisse@isra.sn ncisse@refer.sn www.isra.sn
	Pathology	Disease management, host plant resistance	sarrapenda@hotmail.com
	Entomology	Pest management, host-plant resistance	sardr@yahoo.com
	Genetics	Management of Genetic resources	miamybo@yahoo.fr
	Agronomy	Production management	aliouselbe11@yahoo.fr
	Seed production	Manage foundation seed unit	taffaguey@yahoo.fr

Appendix II: List of Main IITA Released Cultivars in Different Countries

Cultivars	List of countries	Main characteristics
TVx 3236	Togo, Uganda, Yemen, Angola, Botswana, Burkina Faso, Cameroon, Liberia, Mali, Mauritius, Nigeria, Senegal, Sierra Leone,	Scab resistance, <i>Cercospora</i> , brown blotch, thrips tolerance
IT82D-889	Philippines, Suriname, Somalia, Sri Lanka, Swaziland, Tanzania, Thailand, Zambia, Belize, Bolivia, Guinea Bissau, Liberia, Malawi	Extra early maturity, combined resistance to cowpea yellow mosaic and black eyed cowpea mosaic
IT82E-16	Egypt, Ethiopia, Ghana, Guinea Conakry, Lesotho, Malawi, Mozambique, South Africa, Eswatini (Swaziland), Zambia	Early maturity, Combined resistance to <i>Cercospora</i> leaf spot, brown blotch and anthracnose
VITA-3	Belize, Brazil, Fiji, Jamaica, Sierra Leone, Thailand, Venezuela,	Combined resistance to <i>Cercospora</i> leaf spot, brown blotch, anthracnose, bacterial pustule, bacterial blight, cowpea yellow mosaic virus and cowpea and black eyed cowpea mosaic
VITA-4	Central African Republic, Haiti, India, Liberia, Myanmar, Sri Lanka,	Resistant to bacterial blight, brown blotch, <i>Septoria</i> , scab and root knot
IT84S-2246-4	Benin, Guinea Conakry, Jamaica, Nigeria, USA	Early maturity, bruchid tolerant, scab resistance, root knot nematode resistance, aphid resistance
IT99K-573-1-1	Ghana, Niger, Nigeria, Sierra Leone, Tanzania	Medium maturity, resistance to <i>Striga</i> and <i>Alectra</i> , stem rot resistance
IT82E-32	Ethiopia, Lesotho, Sierra Leone, Somalia	Early maturity
IT99K-573-2-1	Ghana, Nigeria, Burkina Faso, Sierra Leone	<i>Striga</i> resistance, <i>Alectra</i> resistance
VITA-5	Central African Republic, Liberia, Togo, Yemen	Field tolerant to leafhoppers, resistant to anthracnose, bacterial pustule and <i>Cercospora</i>
IT82E-18	Australia, Belize, Central African Republic, Eswatini (Swaziland)	Early maturity
IT98K-205-8	Burkina Faso, India, Nepal, Niger	Early maturity, <i>Striga</i> resistance,
IT81D-994	Cameroon, Central African Republic, Nigeria	Medium maturity, bruchid tolerance, <i>Striga</i> resistance for race 4 and race 1, combined resistance to <i>Cercospora</i> leaf spot, brown blotch and anthracnose, <i>Septoria</i> leaf spot resistance, drought tolerance
IT82D-789	Sri Lanka, Suriname, Yemen	early maturity,

(continued)

Cultivars	List of countries	Main characteristics
IT83S-818	Central African Republic, Ghana, Mali	Medium maturity, combined resistance to cowpea yellow mosaic, blackeye cowpea mosaic and many strains of cowpea aphid borne mosaic
IT86D-1010	Paraguay, Sierra Leone, Sri Lanka	Early maturity, combined resistance to cowpea yellow mosaic, black eyed cowpea mosaic and cowpea aphid borne mosaic, black eyed cowpea mosaic and 5 strains of cowpea aphid borne mosaic.
IT87D-885	Equatorial Guinea, Haiti, Lesotho	Combined resistance to <i>Cercospora</i> leaf spot, brown blotch, anthracnose, bacterial pustule, bacterial blight
IT89KD-374	Mali, Niger, Nigeria	Early maturity, combined resistance to <i>Cercospora</i> leaf spot, brown blotch, anthracnose, bacterial pustule, bacterial blight, adapted to intercropping
IT90K-277-2	Cameroon, Nigeria, South Sudan	Combined resistance to cowpea yellow mosaic, cowpea aphid borne mosaic, black eyed cowpea mosaic, cowpea mottle, cowpea cucumber mosaic, combined resistance to <i>Cercospora</i> , brown blotch, adapted to intercropping
IT97K-499-35	Mali, Niger, Nigeria	Early maturity, <i>Striga</i> resistance, <i>Alectra</i> resistance, resistance to scab, brown blotch, <i>Cercospora</i>

References

- AATF (2016) African agricultural technology foundation 2016 annual report. AATF, Nairobi
- Acquaah G (2007) Principles of plant genetics and breeding. Blackwell Publishing, Malden
- Adekola OF, Oluleye F (2007) Induction of genetic variation in cowpea (*Vigna unguiculata* L. Walp.) by gamma irradiation. *Asian J Plant Sci* 6:869–873
- Agbicodo EM, Fatokun CA, Bandyopadhyay R et al (2010) Identification of markers associated with bacterial blight resistance loci in cowpea [*Vigna unguiculata* (L.) Walp.]. *Euphytica* 175:215–226
- Agble F (1972) Seed size heterosis in cowpeas (*Vigna unguiculata* (L.) Walp.). *Ghana J Sci* 12:30–33
- Andargie M, Pasquet RS, Gowda BS et al (2011) Construction of a SSR-based genetic map and identification of QTL for domestication traits using recombinant inbred lines from a cross between wild and cultivated cowpea [*V. unguiculata* (L.) Walp.]. *Mol Breed* 28:413–420
- Arumuganathan K, Earle ED (1991) Nuclear DNA content of some important plant species. *Plant Mol Biol Rep* 9:208–218
- Asare AT, Gowda BS, Galyuon IKA et al (2010) Assessment of the genetic diversity in cowpea (*Vigna unguiculata* L. Walp.) germplasm from Ghana using simple sequence repeat markers. *Plant Genet Resour* 8(2):142–150. <https://doi.org/10.1017/S1479262110000092>

- Ba FS, Pasquet RE, Gepts P (2004) Genetic diversity in cowpea [*Vigna unguiculata* (L.) Walp.] as revealed by RAPD markers. *Genet Resour Crop Evol* 51:539–550
- Barone A, Ng Q (1990) Embryological study of crosses between *Vigna unguiculata* and *V. vexillata*. In: Ng NQ, Monti LM (eds) Cowpea genetic resources. IITA, Ibadan, pp 151–160
- Barone A, Saccardo F (1990) Pachytene morphology of cowpea chromosomes. In: Ng NQ, Monti LM (eds) Cowpea genetic resources. IITA, Ibadan, pp 137–143
- Baudoin J, Maréchal R (1985) Cowpea taxonomy, origin and germplasm. In: Singh SR, Rachie KO (eds) Cowpea research, production and utilization. Wiley, New York, pp 3–9
- Bhaskaraiah KB, Shivashankar G, Virupakshappa K (1980) Hybrid vigour in cowpea. *Indian J Genet Plant Breed* 40:334–337
- Bhushana HO, Viswanatha KP, Runachala PA, Halesh GK (2000) Heterosis in cowpea for seed yield and its attributes. *Crop Res (Hisar)* 19:277–280
- Boukar O, Kong L, Singh BB et al (2004) AFLP and AFLP-derived SCAR markers associated with *Striga gesnerioides* resistance in cowpea. *Crop Sci* 44:1259–1264
- Boukar O, Massawe F, Muranaka S et al (2011) Evaluation of cowpea germplasm lines for protein and mineral concentrations in grains. *Plant Genet Resour* 9(4):515–522. <https://doi.org/10.1017/S1479262111000815>
- Boukar O, Bhattacharjee R, Fatokun C et al (2013) Cowpea. In: Singh M, Upadhyaya HD, Bisht IS (eds) Genetic and genomic resources of grain legume improvement. Elsevier, London, pp 137–156. <https://doi.org/10.1016/B978-0-12-397935-3.00006-2>
- Boukar O, Fatokun CA, Roberts PA et al (2015) Cowpea. In: De Ron AM (ed) Grain legumes, handbook of plant breeding. Springer, New York, pp 219–250. https://doi.org/10.1007/9781493927975_7
- Boukar O, Fatokun CA, Huynh B-L et al (2016) Genomic tools in cowpea breeding programs: status and perspectives. *Front Plant Sci* 7:757. <https://doi.org/10.3389/fpls.2016.00757>
- Brar MS, Al-Khayri JM, Morelock TE, Anderson JE (1999) Genotypic response of cowpea *Vigna unguiculata* (L.) to in vitro regeneration from cotyledon explants. *In Vitro Cell Dev Biol Plant* 35:8–12
- Cardi T, Valanzuolo S, Mazza P, Filippone E (1990) *In vitro* and *in vivo* response to Al³⁺ of *Vigna unguiculata* (L.) Walp. In: Ng Q, Monti L (eds) Cowpea genetic resources. IITA, Ibadan, pp 163–174
- Chen H, Wang L, Liu X, Hu L et al (2017) De novo transcriptomic analysis of cowpea (*Vigna unguiculata* L. Walp.) for genic SSR marker development. *BMC Genet* 18:65. <https://doi.org/10.1186/s12863-017-0531-5>
- Coulibaly S, Pasquet RS, Papa R, Gepts P (2002) AFLP analysis of the phenetic organization and genetic diversity of *Vigna unguiculata* L. Walp. reveals extensive gene flow between wild and domesticated types. *Theor Appl Genet* 104(2–3):358–366
- De Leonardi W, Fichera G, Padulosi S, Zizza A (1993) Preliminary studies on pollen and seed of wild germplasm accessions of *Vigna unguiculata* (L.) Walpers. In: Proceedings, 58th Congress of the Italian Botanic Society. University of Tor Vergata, Rome, 4–8 October 1993. 127:3
- Delgado-Salinas A, Thulin M, Pasquet R et al (2011) *Vigna* (Leguminosae) sensu lato: the names and identities of the American segregate genera. *Am J Bot* 98:1694–1715. <https://doi.org/10.3732/ajb.1100069>
- Diouf D, Hilu KW (2005) Microsatellite and RAPD markers to study genetic relationships among cowpea breeding lines and local cultivars in Senegal. *Genet Resour Crop Evol* 52:1957–1967
- Dumet D, Adeleke R, Faloye B (2008) Regeneration guidelines: cowpea. In: Dulloo ME, Thormann I, Jorge MA, Hanson J (eds) Crop specific regeneration guidelines [CDROM]. CGIAR System-Wide Genetic Resource Programme, Rome
- Dumet D, Fatokun C, Pasquet R et al (2012) Sharing of responsibilities of cowpea and wild relatives in long term conservation. In: Boukar O, Coulibaly O, Fatokun CA et al (eds) Innovative research along the cowpea value chain. Proceedings of the fifth world cowpea conference on improving livelihoods in the cowpea value chain through advancement in science, held in Saly, Senegal, September 27–October 1, 2010. IITA, Ibadan, Nigeria, pp 56–65

- Egbadzor KF, Danquah EY, Ofori K et al (2014a) Diversity in 118 cowpea [*Vigna unguiculata* (L.) Walp] accessions assessed with 16 morphological traits. *Int J Plant Breed Genet* 8:13–24. <https://doi.org/10.1186/2193-1801-3-541>
- Egbadzor KF, Ofori K, Yeboah M et al (2014b) Diversity in 113 cowpea [*Vigna unguiculata* (L.) Walp] accessions assessed with 458 SNP markers. *Springerplus* 3:541. <https://doi.org/10.1186/2193-1801-3-541>
- Ehlers JD, Hall AE (1996) Genotypic classification of cowpea based on responses to heat and photoperiod. *Crop Sci* 36:673–679
- Ehlers JD, Hall AE (1997) Cowpea (*Vigna unguiculata* L. Walp.). *Field Crops Res* 53:187–204
- Ehlers JD, Diop NN, Boukar O et al (2012) Modern approaches for cowpea breeding. In: Boukar O, Coulibaly O, Fatokun CA et al (eds) Innovative research along the cowpea value chain. Proceedings of the fifth world cowpea conference on improving livelihoods in the cowpea value chain through advancement in science, held in Saly, Senegal, September 27–October 1, 2010. IITA, Ibadan, Nigeria, pp 3–16
- Fall L, Diouf D, Fall-Ndiaye MA et al (2003) Genetic diversity in cowpea [*Vigna unguiculata* (L.) Walp.] varieties determined by ARA and RAPD techniques. *Afr J Biotech* 2:48–50
- Fang J, Chao C-CT, Roberts PA, Ehlers JD (2007) Genetic diversity of cowpea [*Vigna unguiculata* (L.) Walp.] in four West African and USA breeding programs as determined by AFLP analysis. *Genet Resour Crop Evol* 54:1197–1209
- FAOSTAT (2018) Database Accessed 23 Dec 2018. <http://www.fao.org/faostat/en/#data/QC>
- Faris DG (1965) The origin and evolution of the cultivated forms of *Vigna sinensis*. *Can J Genet Cytol* 7:433–452. <https://doi.org/10.1139/g65-058>
- Fatokun CA (2002) Breeding cowpea for resistance to insect pests: attempted crosses between cowpea and *Vigna vexillata*. In: Fatokun CA, Tarawali SA, Singh BB et al (eds) Challenges and opportunities for enhancing sustainable cowpea production. Proceedings of the world cowpea conference III held at IITA, Ibadan, Nigeria, 4–8 September 2000, pp 52–61
- Fatokun CA, Danesh D, Young ND (1993) Molecular taxonomic relationships in the genus *Vigna* based on the RFLP analysis. *Theor Appl Genet* 86:97–104
- Fatokun CA, Menancio-Hautea DI, Danesh D, Young ND (1992) Evidence for orthologous seed weight genes in cowpea and mung bean based on RFLP mapping. *Genet* 132:841–846
- Fatokun CA, Ng Q (2007) Outcrossing in cowpea. *J Food Agric Envir* 5:334–338
- Fatokun CA, Singh BB (1987) Interspecific hybridization between *Vigna pubescens* and *V. unguiculata* through embryo rescue. *Plant Cell Tissue Organ Cult* 9:229–233
- Fatokun C, Girma G, Abberton M et al (2018) Genetic diversity and population structure of a mini-core subset from the world cowpea (*Vigna unguiculata* (L.) Walp.) germplasm collection. *Sci Rep* 8:1–10. ISSN:2045-2322
- Fery RL (1985) Improved cowpea cultivars for the horticultural industry in the USA. In: Singh SR, Rachie KO (eds) Cowpea research, production and utilization. Wiley, New York, pp 129–135
- Fery RL, Singh BB (1997) Cowpea genetics: a review of the recent literature. In: Singh BB, Mohan Raj DR, LEN J (eds) Advances in cowpea research. Coproduction of IITA and Japan International Research Center for Agricultural Sciences (JIRCAS). IITA, Ibadan, pp 13–29
- Fotso M, Azanza JL, Pasquet R, Raymond J (1994) Molecular homogeneity of cowpea (*Vigna unguiculata*, Fabaceae) seed storage proteins. *Plant Syst Evol* 191:39–56
- Galasso I, Schmidt T, Pignone D, Heslop-Harrison JS (1995) The molecular cytogenetics of *Vigna unguiculata* (L.) Walp.: the physical organization and characterization of 18s-5.8s-25s ribosomal RNA genes, 5s ribosomal RNA genes, telomere-like sequences, and a family of centromeric repetitive DNA sequences. *Theor Appl Genet* 91:928–935
- Galasso I, Harrison GE, Pignone D et al (1997) The distribution and organization of Ty1-copia-like retrotransposable elements in the genome of *Vigna unguiculata* (L.) Walp (cowpea) and its relatives. *Ann Bot* 80:327–333
- Gamborg OL, Miller RA, Ojima K (1968) Nutrient requirements of suspension cultures of soybean root cells. *Exp Cell Res* 50:151–158

- Ganapathi A, Anand P (1998) Somatic embryogenesis from young leaves of cowpea (*Vigna unguiculata* (L.) Walp. (Abstract) in Plant biotechnology and in vitro biology for the 21st century. IX International Congress on Plant Tissue and Cell Culture, 14–19 June 1998, Jerusalem, Israel
- Girija M, Dhanavel D, Gnanamurthy S (2013) Gamma rays and EMS induced flower color and seed mutants in cowpea (*Vigna unguiculata* L. Walp.). *Adv Appl Sci Res* 4:134–139
- Gupta SK, Gopalakrishna T (2010) Development of unigene-derived SSR markers in cowpea (*Vigna unguiculata*) and their transferability to other *Vigna* species. *Genome* 53(7):508–523
- Hall AE (2004) Comparative ecophysiology of cowpea, common bean and peanut. In: Nguyen HT, Blum A (eds) *Physiology and biotechnology integration for plant breeding*. Marcel Dekker Inc, New York, pp 271–325
- Hall AE, Patel PN (1985) Breeding for resistance to drought and heat. In: Singh SR, Rachie KO (eds) *Cowpea research, production, and utilization*. John Wiley, New York, pp 137–151
- Hansen G, Wright SM (1999) Recent advances in transformation of plants. *Trends Plant Sci* 4(6):226–231
- Higgins TJV, Gollasch S, Movig L et al (2012) Genetic transformation of cowpea for protection against bruchids and caterpillars. In: Boukar O, Coulibaly O, Fatokun CA et al (eds) *Innovative research along the cowpea value chain. Proceedings of the fifth world cowpea conference on improving livelihoods in the cowpea value chain through advancement in science*, held in Saly, Senegal, September 27–October 1 2010, pp 131–137
- Horn LN, Habteab M, Ghebrehiwot HM, Shimelis HA (2016) Selection of novel cowpea genotypes derived through gamma irradiation. *Front Plant Sci* 7:262. <https://doi.org/10.3389/fpls.2016.00262>
- Horn L, Shimelis H, Sarsu F et al (2017) Genotype-by-environment interaction for grain yield among novel cowpea (*Vigna unguiculata* L.) selections derived by gamma irradiation. *The Crop J* 6(3):306–313. <https://doi.org/10.1016/j.cj.2017.10.002>
- Huynh B-L, Close TJ, Roberts PA et al (2013) Gene pools and the genetic architecture of domesticated cowpea. *Plant Genome* 6(3):1–8
- IITA. International Institute of Tropical Agriculture (1972) Grain legume program. IITA Annual Report, IITA, Ibadan, pp 13–19
- Iwata-Otsubo A, Lin J-Y, Gill N, Jackson SA (2016) Highly distinct chromosomal structures in cowpea (*Vigna unguiculata*), as revealed by molecular cytogenetic analysis. *Chromos Res* 24:197–216. <https://doi.org/10.1007/s10577-015-9515-3>
- Jackai LEN, Adalla CB (1997) Pest management practices in cowpea: a review. In: Singh BB, Mohan Raj DR, LEN J (eds) *Advances in cowpea research*. Coproduction of IITA and Japan International Research Center for Agricultural Sciences (JIRCAS). IITA, Ibadan, pp 240–258
- Jackai LEN, Daoust RA (1986) Insect pests of cowpea. *Annu Rev Entomol* 31:95–119
- Kononowicz AK, Cheah KT, Narasimhan ML et al (1997) Developing a transformation system for cowpea (*Vigna unguiculata* [L.] Walp.). In: Singh BB, Mohan-Raj DR, Dashiell KE, LEN J (eds) *Advances in cowpea research*. Co-publication of IITA and Japan International Research Center for Agricultural Sciences (JIRCAS). IITA, Ibadan, pp 361–371
- Joint FAO/IAEA Mutant variety database (2019) Accessed 28 Jan 2019. [https://mvd.iaea.org/#!Search?Criteria\[0\]\[val\]=cowpea](https://mvd.iaea.org/#!Search?Criteria[0][val]=cowpea)
- Ladeinde TAO, Watt E, Onajole AAO (1980) Segregating pattern of three different sources of male sterility genes in *Vigna unguiculata*. *J Hered* 71:431–432
- Lambot C (2002) Industrial potential of cowpea. In: Fatokun CA, Tarawali SA, Singh BB et al (eds) *Challenges and opportunities for enhancing sustainable cowpea production*. Proceedings of the world cowpea conference III held at the IITA, Ibadan, Nigeria, 4–8 September 2000, IITA, Ibadan, Nigeria, pp 367–375
- Leleji OI (1973) Apparent preference by bees for different flower colours in cowpea (*Vigna sinensis* (L.) Savi ex Hassk.). *Euphytica* 22:150–153
- Li CD, Fatokun CA, Ubi B et al (2001) Determining genetic similarities and relationships among cowpea breeding lines and cultivars by microsatellite primers. *Crop Sci* 41:189–197

- Lucas MR, Diop NN, Wanamaker S et al (2011) Cowpea-soybean synteny clarified through an improved genetic map. *Plant Genome* 4:218–224
- Lucas MR, Ehlers JD, Huynh BL et al (2013a) Markers for breeding heat-tolerant cowpea. *Mol Breed* 31:529–536
- Lucas MR, Huynh BL, da Silva Vinholes P et al (2013b) Association studies and legume synteny reveal haplotypes determining seed size in *Vigna unguiculata*. *Front Plant Sci* 4:95. <https://doi.org/10.3389/fpls.2013.00095>
- Machuka J (2002) Potential role of transgenic approaches in the control of cowpea insect pests. In: Fatokun CA, Tarawali SA, Singh BB et al (eds) Challenges and opportunities for enhancing sustainable cowpea production. Proceedings of the world cowpea conference III held at the IITA, Ibadan, Nigeria, 4–8 September 2000. IITA, Ibadan, Nigeria, pp 213–222
- Machuka J, Adesoye A, Obembe OO (2002) Regeneration and genetic transformation in cowpea. In: Fatokun CA, Tarawali SA, Singh BB et al (eds) Challenges and opportunities for enhancing sustainable cowpea production. Proceedings of the world cowpea conference III held at the IITA, Ibadan, Nigeria, 4–8 September 2000. IITA, Ibadan, Nigeria, pp 185–196
- Mak C, Yap TC (1977) Heterosis and combining ability of seed protein, yield and yield components in long bean. *Crop Sci* 17:339–341
- Maréchal R (1978) Etude taxonomique d'un groupe complexe d'espèces des genres *Phaseolus* et *Vigna* (Papilionaceae) sur la base de données morphologiques et polliniques, traitées par l'analyse informatique. Conservatoire et Jardin Botaniques, Geneve
- Maréchal R, Mascherpa JM, Stainier F (1978) Etude taxonomique d'un groupe complexe d'espèces des genres *Phaseolus* et *Vigna* (Papilionaceae) sur la base de données morphologiques et polliniques, traitées par l'analyse informatique. *Boiss* 28:1–273
- Mehta DR, Zaveri PP (1997) Single seed versus single plant selection in cowpea. *Legume Res* 20(2):130–132
- Mignouna HD, Ng NQ, Ikea J, Thottapilly G (1998) Genetic diversity in cowpea as revealed by random amplified polymorphic DNA. *J Genet Breed* 52:151–159
- Mishra SN, Verma JS, Jayasekara SJB (1985) Breeding cowpeas to suit Asian cropping systems and consumer tastes. In: Singh SR, Rachie KO (eds) Cowpea research, production and utilization. Wiley, New York, pp 117–123
- Monti LM, Murdock LL, Thottapilly G (1997) Opportunities for biotechnology in cowpea. In: Singh BB, Mohan Raj DR, Dashiell KE, LEN J (eds) Advances in cowpea research. Copublication of IITA and Japan International Research Center for Agricultural Sciences (JIRCAS). IITA, Ibadan, pp 341–351
- Muchero W, Diop N-N, Bhat PR et al (2009a) A consensus genetic map of cowpea [*Vigna unguiculata* (L.) Walp.] and synteny based on EST-derived SNPs. *Proc Natl Acad Sci USA* 106:118159–118164
- Muchero W, Ehlers JD, Close TJ, Roberts PA (2009b) Mapping QTL for drought stress-induced premature senescence and maturity in cowpea [*Vigna unguiculata* (L.) Walp.]. *Theor Appl Genet* 118:849–863
- Muchero W, Ehlers JD, Close TJ, Roberts PA (2011) Genic SNP markers and legume synteny reveal candidate genes underlying QTL for *Macrophomina phaseolina* resistance and maturity in cowpea [*Vigna unguiculata* (L.) Walp.]. *BMC Genomics* 12:8. <https://doi.org/10.1186/1471-2164-12-8>
- Muñoz-Amatriaín M, Mirebrahim H, Xu P et al (2016) Genome resources for climate-resilient cowpea, an essential crop for food security. *Plant J* 89(5):1042–1054
- Murashige T, Skoog F (1962) A revised medium for rapid growth and bioassays with tobacco tissue cultures. *Phys Plant* 15:473–497
- Ng N (1995) Cowpea *Vigna unguiculata* (Leguminosae-Papilionideae). In: Smartt J, Simmonds NW (eds) Evolution of crop plants, 2nd edn. Longman, Essex, pp 326–332
- Ng NQ, Singh BB (1997) Cowpea. In: Fuccillo D, Sears L, Stapleton P (eds) Biodiversity and trust. Cambridge University Press, Cambridge, pp 82–89

- Nielsen SS, Brandt WE, Singh BB (1993) Genetic variability for nutritional composition and cooking time of improved cowpea lines. *Crop Sci* 33:469–472
- Nielsen SS, Ohler TA, Mitchell CA (1997) Cowpea leaves for human consumption: production, utilization and nutrient composition. In: Singh BB, Mohan-Raj DR, Dashiell KE, LEN J (eds) *Advances in cowpea research*. Co-publication of IITA and Japan International Research Center for Agricultural Sciences (JIRCAS). IITA, Ibadan, pp 326–332
- Nkongolo KK (2003) Genetic characterization of Malawian cowpea (*Vigna unguiculata* (L.) Walp.) landraces: diversity and gene flow among accessions. *Euphytica* 129:219–228
- Odeigah PGC, Osanyinpeju AO, Myers GO (1998) Induced mutations in cowpea, *Vigna unguiculata* (Leguminosae). *Rev Biol Trop*. San José. 46(3) Online version ISSN 0034-7744
- Ogunkanmi LA, Ogundipe OT, Ng NQ, Fatokun CA (2008) Genetic diversity in wild relatives of cowpea (*Vigna unguiculata*) as revealed by simple sequence repeats (SSR) markers. *J Food Agric Envir* 6:253–268
- Ojomo AO (1973) Breeding and improvement of cowpea in the Western State of Nigeria. In: *Proceedings of the first IITA grain legume improvement workshop, 29th October – 2nd November*. IITA, Ibadan, Nigeria, pp 21–25
- Oladosu Y, Rafii MY, Abdullah N et al (2016) Principle and application of plant mutagenesis in crop improvement: a review. *Biotech Biotech Equip* 30:1–16. <https://doi.org/10.1080/13102818.2015.1087333>
- Olasupo FO, Ilori CO, Muiyiwa AA (2016) Radio-sensitivity of cowpea to ultra-violet radiation by pollen treatment. *Plant Breed Crop Sci* 8(11):228–239. <https://doi.org/10.5897/JPBCS2016.0602>
- Omo-Ikerodah EE, Fawole I, Fatokun C (2008) Genetic mapping of quantitative trait loci (QTLs) with effects on resistance to flower bud thrips (*Megalurothrips sjostedti*) in recombinant inbred lines of cowpea [*Vigna unguiculata* (L.) Walp.]. *Afr J Biotech* 7:263–270
- Oppong-Konadu EYR, Akromah IK, Adu-Dapaah OE (2005) Genetic diversity within Ghanaian cowpea germplasm based on SDS-PAGE of seed proteins. *Afr Crop Sci J* 13:117–123
- Ouédraogo JT, Maheshwari V, Berner DK et al (2001) Identification of AFLP markers linked to resistance of (*Vigna unguiculata* L.) to parasitism by *Striga gesnerioides*. *Theor Appl Genet* 102:1029–1036. <https://doi.org/10.1007/s001220000499>
- Ouédraogo JT, Gowda BS, Jean M et al (2002) An improved genetic linkage map for cowpea (*Vigna unguiculata* L.) combining AFLP, RFLP, RAPD, biochemical markers, and biological resistance traits. *Genome* 45:175–188. <https://doi.org/10.1139/g01-102>
- Ouédraogo JT, Ouédraogo M, Gowda BS, Timko MP (2012) Development of sequence characterized amplified region (SCAR) markers linked to race-specific resistance to *Striga gesnerioides* in cowpea (*Vigna unguiculata* L.). *Afr J Biotech* 11:12555–12562. <https://doi.org/10.5897/AJB12.805>
- Padulosi S (1993) Genetic diversity, taxonomy and ecogeographic survey of the wild relatives of cowpea (*Vigna unguiculata* (L.) Walpers). PhD thesis, Université Catholique, Louvain la Neuve, Belgium
- Padulosi S, Ng NQ (1993) A useful and unexploited herb, *Vigna marina* (Leguminosae-Papilionoideae) and the taxonomic revision of its genetic diversity. *Syst Geogr Plants* 62(1–4):119–126
- Padulosi S, Laghetti G, Pienaar B et al (1991) Survey of wild *Vigna* in southern Africa. *Plant Genet Resour Newsl* 83/84:5–8
- Panella L, Gepts P (1992) Genetic relationships with *Vigna unguiculata* (L.) Walp. based on isozyme analyses. *Genet Resour Crop Evol* 39:71–88
- Pant K, Chandel K, Joshi B (1982) Analysis of diversity in Indian cowpea genetic resources. *SABRAO J* 14:103–111
- Pasquet RS (1993) Classification infraspecificque des forms spontanees de *Vigna unguiculata* (L.) Walp. a partir de donnees morphologiques. *Syst Geogr Plants* 62:127–173
- Pasquet RS (1999) Genetic relationships among subspecies of *Vigna unguiculata* (L.) Walp. based on allozyme variation. *Theor Appl Genet* 98:1104–1119

- Pasquet RS (2000) Allozyme diversity of cultivated cowpea *Vigna unguiculata* (L.) Walp. *Theor Appl Genet* 101:211–219
- Pasquet RS, Padulosi S (2012) Genus *Vigna* and cowpea (*Vigna unguiculata* (L.) Walp.) taxonomy: current status and prospects. In: Boukar O, Coulibaly O, Fatokun CA et al (eds) Innovative research along the cowpea value chain. Proceedings of the fifth world cowpea conference on improving livelihoods in the cowpea value chain through advancement in science, held in Saly, Senegal, 27 September – 1 October 2010. IITA, Nigeria, pp 66–87
- Penning de Vries FWT, Djiteye MA (1991) La productivité des pâturages sahéliens: une étude des sols, de la végétation et de l'exploitation de cette ressource naturelle. Center for Agricultural Publishing and Documentation (Pudoc-DLO), Wageningen
- Perrino P, Laghetti G, Spagnoletti Zeuli PL, Monti LM (1993) Diversification of cowpea in the Mediterranean and other centers of cultivation. *Genet Resour Crop Evol* 40:121–132
- Phillips EP (1951) The genera of South African flowering plants. Government Printer, Pretoria
- Pignone D, Cecarelli S, Perrino P (1990) Chromosome identification in *Vigna unguiculata* (L.) Walp. In: Ng NQ, Monti LM (eds) Cowpea genetic resources. IITA, Ibadan, pp 144–150
- Pienaar S (1992) Genetic diversity, taxonomy and ecogeographic survey of the wild relatives of cowpea subspecies *protracta* var. *rhomboidea* Padulosi. *S Afr J Bot* 58(6):420
- Popelka JC, Gollasch S, Moore A et al (2006) Genetic transformation of cowpea (*Vigna unguiculata* L.) and stable transmission of transgenes to progeny. *Plant Cell Rep* 25:304–312
- Porter WM, Rachie KO, Rawal KM et al (1974) Cowpea germplasm catalogue no. 1. SAS Institute, Ibadan
- Potchefstroom College of Agriculture (1948) Annual report 1947–1948. Field Husbandry Research and Education College of Agriculture Potchefstroom, South Africa
- Punniyamorthy D, Reddy KS, Dhanasekar SP (2007) IANCAS bulletin, Nov 2007, pp 299–307
- Rodrigues MA, Santos CAF, Santana JRF (2012) Mapping of AFLP loci linked to tolerance to cowpea golden mosaic virus. *Genet Mol Res* 11:3789–3797. <https://doi.org/10.4238/2012.August.17.12>
- Roy RS, Richaria RH (1948) Breeding and inheritance studies on cowpea, *Vigna sinensis*. *Agron J* 40:479–489
- Saccardo, F, Del Guidice A, Galasso I (1992) Cytogenetics of cowpea. In: Thottappilly G, Monti LM, Mohan Raj, DR, Moore AW (eds) Biotechnology: enhancing research on tropical crops in Africa. CTA/IITA co-publication. IITA, Ibadan, pp 89–98
- Samireddypalle A, Boukar O, Grings E et al (2017) Cowpea and groundnut haulms fodder trading and its lessons for multidimensional cowpea improvement for mixed crop livestock systems in West Africa. *Front Plant Sci* 8:30. <https://doi.org/10.3389/fpls.2017.00030>
- Sen NK, Bhowal JG (1962) A male sterile mutant cowpea. *J Hered* 53:44–46
- Simon MV, Benko-Iseppon AM, Resende LV et al (2007) Genetic diversity and phylogenetic relationships in *Vigna savi* germplasm revealed by DNA amplification fingerprinting. *Genome* 50:538–547
- Singh BB, Ntare BR (1985) Development of improved cowpea varieties in Africa. In: Singh SR, Rachie KO (eds) Cowpea research, production and utilization. Wiley, New York, pp 105–115
- Singh BB, Chambliss OL, Sharma B (1997) Recent advances in cowpea breeding. In: Singh BB, Mohan Raj DR, Dashiell KE, LEN J (eds) Advances in cowpea research. Copublication of IITA and Japan International Research Center for Agricultural Sciences (JIRCAS). IITA, Ibadan, pp 30–49
- Singh BB, Ehlers JD, Sharma B, Freire Filho FR (2002) Recent progress in cowpea breeding. In: Fatokun CA, Tarawali SA, Singh BB et al (eds) Challenges and opportunities for enhancing sustainable cowpea production. Proceedings of the world cowpea conference III held at IITA, Ibadan, Nigeria, 4–8 September 2000. IITA, Ibadan, Nigeria, pp 22–40
- Singh SR, Singh BB, Jackai LEN, Ntare BR (1983) Cowpea research at IITA. Ibadan, Nigeria, IITA. Information series 14:1–20
- Singh SR, Jackai LEN, Dos Santos JHR, Adalla CB (1990) Insect pests of cowpea. In: Singh SR (ed) Insect pests of food legumes. Wiley, New York, pp 43–89

- Spillman WJ (1911) Inheritance of the 'eye' in *Vigna*. *Am Nat* 45:513–523
- Spillman WJ (1913) Color correlation in cowpea. *Science* 38:302
- Steele W (1976) Cowpea, *Vigna unguiculata* (Leguminosae-Papilionatae). In: Simmonds N (ed) *Evolution of crop plants*. Longman, London, pp 183–185
- Suzuki K, Fatokun C, Boukar O (2018) Responses of cowpea genotypes to indigenous rock phosphate application. *Agron J* 110:1–14
- Timko MP, Rushton PJ, Laudeman TW et al (2008) Sequencing and analysis of the gene-rich space of cowpea. *BMC Genomics* 9:103
- Uma MS, Hittalamani S, Murthy BCK, Viswanatha KP (2009) Microsatellite DNA marker aided diversity analysis in cowpea [*Vigna unguiculata* (L.) Walp.]. *Indian J Genet Plant Breed* 69:35–43
- Vaillancourt RE, Weeden NF (1992) Chloroplast DNA polymorphism suggests a Nigerian centre of domestication for the cowpea *Vigna unguiculata* (Leguminisae). *Am J Bot* 79:1194–1199
- Vaillancourt RE, Weeden NF, Barnard J (1993) Isozyme diversity in the cowpea species complex. *Crop Sci* 33:606–613
- Varshney RK, Chabane K, Hendre PS et al (2007) Comparative assessment of EST-SSR, EST-SNP and AFLP markers for evaluation of genetic diversity and conservation of genetic resources using wild, cultivated and elite barleys. *Plant Sci* 173:638–649
- Vasconcelos EV, Fonsêca AF, Pedrosa-Harand A et al (2014) Intra- and interchromosomal rearrangements between cowpea [*Vigna unguiculata* (L.) Walp.] and common bean (*Phaseolus vulgaris* L.) revealed by BAC-FISH. *Chromos Res* 23:253–266. <https://doi.org/10.1007/s10577-014-9464-2>
- Venora G, Padulosi S (1997) Karyotypic analysis of wild taxa of *Vigna unguiculata* (L.) Walpers. *Caryologia* 50:125–138
- Verdcourt B (1970) Studies in the leguminosae-papilionoideae for the flora of tropical East Africa. IV. *Kew Bull* 24:507–509
- Vir R, Bhat KV, Lakhanpaul (2009) Transferability of sequence tagged microsatellite sites (STMS) primers to pulse yielding taxa belonging to Phaseolae. *Int J Integr Biol* 5(1):62–66
- Wang ML, Barkley NA, Gillaspie GA, Pederson GA (2008) Phylogenetic relationships and genetic diversity of the USDA *Vigna* germplasm collection revealed by gene-derived markers and sequencing. *Genet Res* 90:467–480
- Widders IE (2012) Cowpea: a solution to global challenges. In: Boukar O, Coulibaly O, Fatokun CA et al (eds) *Innovative research along the cowpea value chain*. Proceedings of the fifth world cowpea conference on improving livelihoods in the cowpea value chain through advancement in science, held in Saly, Senegal, September 27–October 1, 2010. IITA, Ibadan, Nigeria, pp xi–xviii
- Whit WC (2007) Soul food as cultural creation. In: Bower A (ed) *African American foodways: explorations of history and culture*. University of Illinois Press, Urbana, pp 45–58
- Xavier GR, Martins LMV, Rumjanek NG, Filho FRF (2005) Variabilidade genética em acessos de caupi analisada por meio de marcadores RAPD. *Pesqui Agropecu Bras* 40:353–359
- Xu P, Wu X, Wang B et al (2010) Development and polymorphism of *Vigna unguiculata* ssp. *unguiculata* microsatellite markers used for phylogenetic analysis in asparagus bean (*Vigna unguiculata* ssp. *sesquipedialis* (L.) Verdc.). *Mol Breed* 25(4):675–684. <https://doi.org/10.1007/s11032-009-9364-x>
- Xiong H, Shi A, Mou B et al (2016) Genetic diversity and population structure of cowpea (*Vigna unguiculata* L. Walp). *PLoS One* 11(8):e0160941. <https://doi.org/10.1371/journal.pone.0160941>
- Zannou A, Kossou DK, Ahanchédé A et al (2008) Genetic variability of cultivated cowpea in Benin assessed by random amplified polymorphic DNA. *Afr J Biotech* 7:4407–4414