

Research Application Summary

Crop improvement through use of mutation breeding-a Review

Afutu, E.¹, Asare, P.A.¹, Adu, M.¹, Amenorpe, G.², Addy, S.N.T.T.³, Lumor, P.¹, Asare-Bediako, E.¹, Danquah, O-A.¹ & Rubaihayo, P.⁴

¹Department of Crop Science, School of Agriculture, College of Agriculture and Natural Sciences, University of Cape Coast, P. O. Box 5007, Cape Coast, Ghana

²Nuclear Agriculture Research Centre (NARC), Biotechnology and Nuclear Agriculture Research Institute (BNARI), Ghana Atomic Energy Commission (GAEC), P. O. Box LG80, Legon, Ghana

³Council for Scientific and Industrial Research, Crops Research Institute, Fumesua, Kumasi, Ghana

⁴Department of Agricultural Production, School of Agriculture, College of Agricultural and Environmental Sciences, Makerere University, P.O Box 7062, Kampala, Uganda

Corresponding author: emmanuel.afutu@ucc.edu.gh

Abstract

The sustainability of agriculture in sub-Saharan Africa is threatened by rapid population explosion and climatic change, resulting in challenges with food sustainability and malnutrition. Conventional breeding approaches alone cannot produce sufficient varieties to match the ever increasing demand for nutritious food because of associated limitations. On the other hand, transgenic breeding is still plagued with controversies. Plant mutation breeding is thus an alternative approach for developing crop varieties. There is therefore need to invigorate mutagenesis of crops in Africa to facilitate crop improvement. Mutagenesis can develop exceptional useful plants in a relatively short time. It can broaden genetic base and increase genetic diversity as a prerequisites for successful conventional plant breeding. The fast debilitating effect of climate change can also be mitigated by developing drought resistant cultivars for future generations with mutagenesis. Mutation breeding can therefore contribute towards attainment of some of the United Nation's Sustainable Development Goals, such as ending hunger, achieving food security and zero poverty. In this paper we highlight prospect of mutation breeding for crop improvement in Africa

Key words: Crop improvement, food security, improved seeds, mutation breeding, sub-Saharan Africa, *Vigna unguiculata*

Résumé

La durabilité de l'agriculture en Afrique sub-saharienne est menacée par l'explosion démographique rapide et le changement climatique, ce qui entraîne des problèmes de durabilité alimentaire et de malnutrition. Les méthodes conventionnelles de sélection ne peuvent à elles seules produire suffisamment de variétés pour répondre à la demande toujours croissante d'aliments nutritifs en raison des limitations associées. D'autre part, la sélection transgénique est toujours en proie à des controverses. La sélection végétale par mutation est donc une approche alternative pour développer des variétés végétales. Il est donc nécessaire de revigorer la mutagenèse des cultures en Afrique pour faciliter l'amélioration des

cultures. La mutagenèse peut développer des plantes utiles exceptionnelles en un temps relativement court. Elle peut élargir la base génétique et augmenter la diversité génétique comme condition préalable à une sélection végétale conventionnelle réussie. L'effet négatif du changement climatique peut également être atténué en développant des variétés résistantes à la sécheresse pour les générations futures avec mutagenèse. L'élevage par mutation peut donc contribuer à la réalisation de certains des objectifs de développement durable des Nations Unies, tels que l'élimination de la faim, la sécurité alimentaire et la pauvreté zéro. Dans cet article, nous mettons en évidence la perspective de la sélection par mutation pour l'amélioration des cultures en Afrique

Mots clés : Amélioration des cultures, sécurité alimentaire, semences améliorées, sélection par mutation, Afrique subsaharienne, *Vigna unguiculata*

Introduction

The Millennium Development Goals (MDGs) ended in 2015 after having aspired to halve extreme poverty and hunger, promote gender equality and reduce child mortality. Sub-Saharan Africa is said to have made considerable progress regarding the MDGs (Madzivhandila *et al.*, 2016). The number of people suffering from hunger in SSA was for example, believed to have been halved (FAO, 2015a, 2015b) and the pervasiveness of hunger in the region was reduced by around 31% between 1990/1992 and 2015 (FAO, 2015c; Madzivhandila *et al.*, 2016). The sequel of the MDGs is the Sustainable Development Goals (SDGs), whose development agenda is more people-centered and has strong focus on food and nutrition security. Despite the recorded decline in the rate of poverty and pervasiveness of undernourishment in SSA, the total number of malnourished people is continually rising on the continent. Approximately, there has been 24% increase in the number of undernourished between the base year of 1990s and 2016 (from 175.7 million in 1990-1992 to an estimated 217.8 million in 2014-2016) (Madzivhandila *et al.*, 2016). This translates into approximately 1% annual percentage growth rate in the number of undernourished people in SSA and suggests that the MDGs nutrition targets were not fully achieved in the region. Many factors have been cited for the partial achievement of the nutrition targets of the MDGs but recent escalated world food prices and increased frequency and intensity of droughts in SSA have been implicated (Madzivhandila *et al.*, 2016).

In recent years, many agricultural technologies, policies and farming systems have been promoted to increase crop yields, ensuring continuous cultivation, and more so, avoiding total crop failure due to biotic and abiotic stresses in sub-Saharan Africa. Some of these farming systems and or policies have evolved through many forms; from diversification and more recently, to what is known as sustainable intensification (SI). Sustainable intensification have been touted as a quicker route to closing yield gaps between actual and potential yields and therefore increasing agricultural productivity rapidly, which altogether, are essential for poverty reduction and improved livelihoods (Delve *et al.*, 2016). Despite its widely recognized contribution to food and nutrition security, SI is not a panacea to all the food challenges in SSA (Katerere *et al.*, 2016). There is currently more room for SI to be holistic and advance beyond just top-down technologies but rather also utilize indigenous knowledge, practices and solutions (Katerere *et al.*, 2016). Available data on agricultural productivity and intensification indicate that yields of cereal crops, agricultural value added per worker and total factor productivity in SSA are lower than in Asia and Latin America (Delve *et al.*, 2016). To bridge this gap, fundamentals of SI in SSA should include the use of improved seeds and fertilizers, within the framework of Integrated Soil Fertility Management (ISFM) (Katerere *et al.*, 2016).

To circumvent the numerous challenges facing the African farmer, there should be more efficient, and a quick way of achieving multiple breeding objectives while utilizing minimum amount of resources. Precise crop improvement techniques are needed to develop climate smart varieties, adaptable to abiotic and biotic stresses (Raina *et al.*, 2016). Opportunely, adoption of improved varieties and hybrids, and agro-chemicals is increasing over time in many countries (Delve *et al.*, 2016). Integrating these technologies promise generating higher benefits for smallholder farmers in SSA. Different plant breeding tools and methods have been employed by plant breeders in SSA to develop new and improved seeds of various food and nutrition security crops. The different plant breeding tools and methods applied have their attendant challenges and advantages. This paper therefore, looks at mutagenesis and makes a case for its adoption and application in the plant breeding programmes in Africa as a quick and efficient way of achieving both food and nutritional security in Africa.

Mutagenesis. Mutagenesis is a process of developing mutation. Mutation is a stable and reproducible change in the genetic information of an organism, under natural or artificial conditions. Mutations occur when faults in DNA or chromosome repair occur (Forster and Shu, 2011). Mutations are heritable changes in the DNA of a living cell, not controlled by segregation or recombination of genes. Mutation occurs by either altering nuclear DNA (point mutations) or changing the cytoplasmic DNA (cytoplasmic mutation). The best example of useful cytoplasmic mutation is cytoplasmic male sterility. When mutation is purposefully directed to produce useful plants, it is called mutation breeding (Pathirana, 2011; Acquaaah, 2012). Mutation was originally defined by de Vries (1901, 1903; 1905) to be a “sudden” heritable change in a genetic material that lead to expression of clear, immediately observable changes in phenotype. Forster and Shu (2011) emphasized that mutation can be a subtle heritable change in the genetic material that lead to expression of small and subtle changes which are not immediately seen as changes in phenotype and are mostly biochemical in nature and are detected using appropriate technologies. They thus effectively nullified the premise and the use of the word “sudden” in defining mutation (Forster and Shu, 2011).

Freisleben and Lein (1944) lay claim to the first use of the term mutation breeding; they used it to refer to the purposeful production of mutant lines for crop improvement. A mutant is an individual carrying a mutation that may be revealed using molecular means or identified by phenotyping tools. Crop improvement by means of induced mutation is however reported to have begun over eight decades ago following the discovery of mutagenic effects of radium ray on *Datura stramonium* (Gagar and Blakeslee, 1927) and in some cereals and *Drosophila* with X-ray (Kharkwal *et al.*, 2004). Mutation breeding is currently widely used to refer to the use of mutagens to induce new traits in the development of crop varieties (Forster and Shu, 2011). Mutation breeding has become very popular for many breeders increasing elite genotypes and harnessing desired genetic diversity for socio-economic development. Accordingly, there are currently several crop varieties possessing important traits developed through induce mutation breeding techniques (Forster and Shu, 2011).

Methods for generating induced mutant varieties could be divided into two, viz., forward and reverse genetic mutagenesis. The former employs both chemical and radiation in induction while the reverse genetic mutagenesis involves insertional mutagenesis, Target Induced Local Lesions in Genome (*TILLING*) and next generation sequencing. Insertional mutagenesis involves agrobacterium mediated transformation, virus induced gene silencing, RNA mediated interference and transposon tagging. However, Forster and Shu (2011) opined that these could be grouped into three types of mutagenesis: the induced, the insertion and the side-directed. Induced mutagenesis involves the use of radiation

or chemical as mutagens. Insertion mutagenesis on the other hand involves inserting at least one nucleotide into a sequence of DNA with the aid of T-DNA (T-DNA insertion mutagenesis) during genetic engineering or the activation of transposon elements using “jumping gun” during “jumping gene” transformation (transposon or transposition mutagenesis). Side directed mutagenesis occurs when mutation is created at a definite site within a molecule of DNA. Mutations are generally random events. A modern biotechnology technique uses site-directed mutagenesis to induce target mutations. However, conventional mutagenesis remains unpredictable and cannot be directed to specific genes.

Mutagenesis and plant breeding. A number of factors in nature act to alter the genetic constitution of plants permanently and thus produce spontaneous mutation, which is a mutation without the aid of man. These factors may include the interaction of seeds with background radioactivity, cosmic rays, and presence of soil chemicals. The frequency of spontaneous mutation is highly influenced by elements of weather (Medina *et al.*, 2005). Unlike spontaneous mutation, induced mutation breeding involves three important steps. The first is selection of parental material for starting mutation breeding. The selected material should be the best variety available and seed should be pure. The second is the part of the plant to be treated since the mutagen dose differs for different plant tissues. Lastly, the dose for mass treatment must be determined before actual treatments of propagules. After mutation induction, it is useful to conduct mutant screening and confirmation (Forster and Shu, 2011). Useful mutant screening involves searching for exceptional useful mutants from a myriad of subtle and sudden mutant plants, of which most are useless mutations. For example, an M2 plant (first mutated generation is termed M1 for seed propagated crops, next mutated generation is M2, followed by M3, etc) reaching anthesis three days earlier than their parents may be screened as useful mutant for early flowering and thus early maturing mutants. Similarly, mutant plants without disease symptom might be screened as useful host plant mutant for disease resistance. It has been argued that a plant with mutated flowering characteristic is more of a ‘putative mutant’ than a ‘true mutant’ given that flowering is controlled by genes which are influenced by environmental factors (Forster and Shu, 2011). Likewise, plants having mutations in many other traits including disease resistance may not be true mutants because absence of disease may merely be due to the absence of the pathogen. However, when mutants and parental controlled lines are cultivated under the same soil and environmental condition and the performance of the mutant line is significantly better and useful than a parental line, a mutant is established (Amenorpe *et al.*, 2010). Mutant breeding involves screening large number of induced genotypes before getting few progenies as selected putative individuals. Mutagenesis in crops has been applied to different breeding objectives. Some of these include crop domestication and adaptation to new environments, development of resistance to biotic and abiotic stresses, and, improvement of crop quality and nutritional traits (Pathirana, 2011).

Application of mutagenesis in the production of new crop varieties. Cereal mutagenesis (Fig. 1), has dominated the world’s genetic enhancement of crops. A total of 1,549 cereal mutant varieties have been released, accounting for 47.3% of the total number of mutant varieties released worldwide (FAO/IAEA, 2018). Other edible crop categories include grain legumes (463 mutant varieties) representing 14.1%, 42 tree fruits representing 1.3%, and, 19 root and tuber crops representing 0.6% (FAO/IAEA, 2018). The portion of the pie chart in Figure 1, which is labelled “others” (1,205 mutant varieties) represent 36.8% of the total mutant varieties registered and this comprises of crop groups such as ornamentals, grasses, fibre crops, medicinal plants, industrial oil crops, etc. Among the cereals, maize (*Zea mays*) constitute the highest proportion of mutants released, followed by rice (*Oryza sativa*), barley (*Hordium vulgare*), and others (FAO/IAEA, 2018).

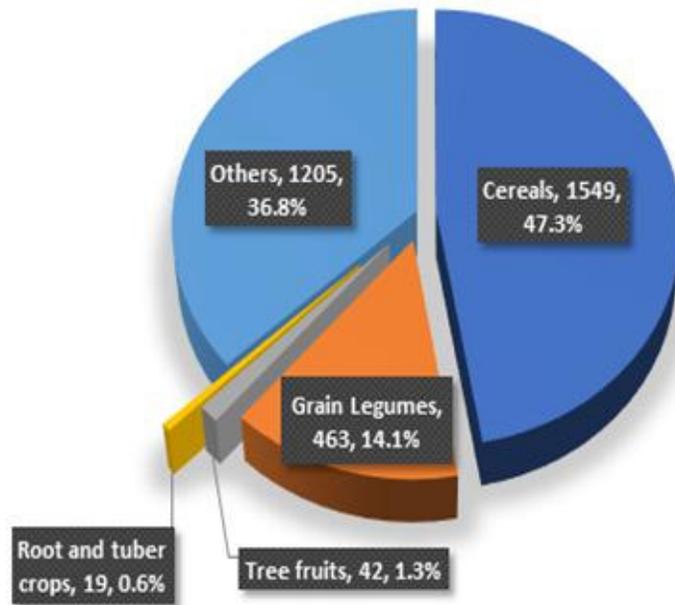


Figure 1. Number of mutant varieties released as at April, 2018 analysed based on different end-uses or type of crop. Data source: (FAO/IAEA, 2018).

Out of the 463 mutant varieties of grain legumes and pulses registered, 167 (36.1%) are soybeans followed by groundnut, common bean, cowpea and pigeon pea with 73 (15.8%), 57 (12.3%), 10 (2.2%) and 6 (1.3%) mutant varieties respectively, and these five (5) crops constitute 68% of the total grain legumes and pulses registered (FAO/IAEA, 2018). A breakdown of mutant varieties developed and registered in different continents are presented in Figure 2. Out of the total 3,278 mutant varieties registered, Asia has the highest number (1,993), representing 60.8% followed by Europe with 955 mutant varieties representing 29.1%, North America with 51 (6.1%) and the last three continents are Africa, Latin America and, Australia and the Pacific with 69 (2.1%), 51 (1.6%) and 10 (0.3%) mutant Varieties, respectively (FAO/IAEA, 2018).

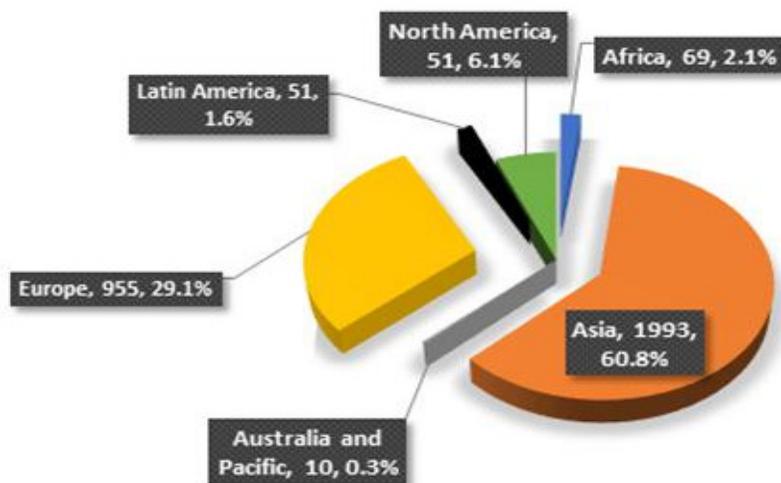


Figure 2. Number of mutant varieties released among continents as at April, 2018.

Figure 3 shows a breakdown of the 69 mutant varieties developed from Africa as at April 2018 (FAO/IAEA, 2018). The crop with the highest number of mutants developed within the continent is rice (39 mutant varieties) representing 56.5% of the total mutant varieties, followed by sorghum and sesame with 8 (11.6%) and 5 (7.2%) mutant varieties, respectively (FAO/IAEA, 2018). There are 3 (4.3%) mutant varieties each for cowpea and soybean, 2 (2.9%) mutants each recorded for common bean, finger millet and safflower while there is 1 (1.4%) mutant variety each registered for the following crops; Banana, Cassava, Chickpea, Maize and wheat. These figures suggest that Sub-Saharan Africa is not yet taking advantage of the benefits that mutation breeding offers, unlike Asia, Europe and North America which produce enough food to feed their people and also, export to other continents.

Mutagenesis – The way forward to achieving food and nutritional security in Sub-Saharan Africa (The case for mutagenesis in breeding programmes in Africa). “We may search for mutable plants in nature, or we may hope for species to become mutable by artificial methods. The first promises to yield results most quickly, but the scope of the second is much greater and it may yield results of far more importance. Indeed, if it once should become possible to bring plants to mutate at our will and perhaps even in arbitrarily chosen directions, there is no limit to the power we may finally hope to gain over nature”(a quote from Hugo de Vries adapted from Forster and Shu, 2011).

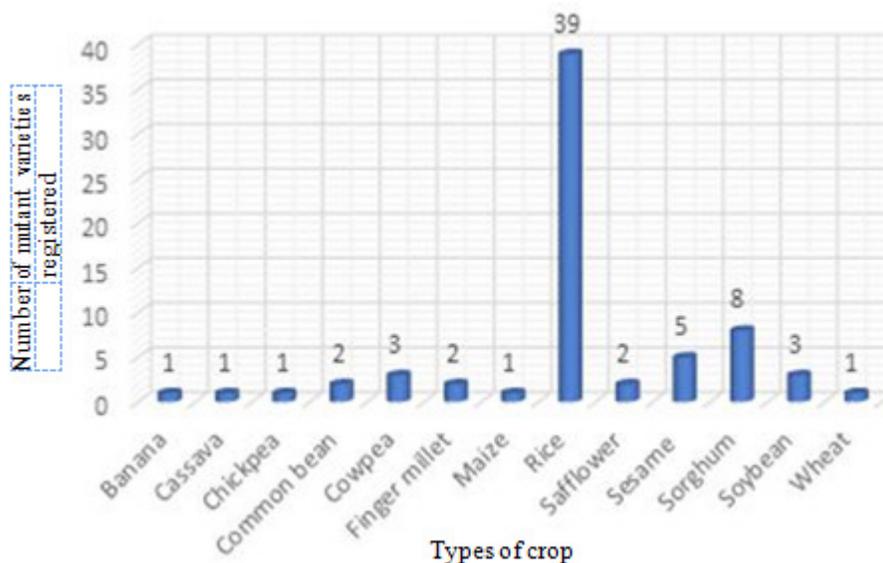


Figure 3. Breakdown of the 69 mutant varieties released in Africa as at April, 2018.

An evidential repercussion of rapid population growth in Africa than other continents is that there is an increase in the absolute number of undernourished people in Sab Saharan Africa compared to other continents (IBRD/World Bank, 2015; FAO, 2015, Delve *et al.*, 2016; Madzivhandila *et al.*, 2016). Crop Improvement must follow similar trends for sustainable production and supply of farm commodities to support the survival and livelihoods of the populations in SSA. Mutation breeding offers the opportunity for cheap, simple, efficient, rapid, and yet environmentally friendly, development of superior crop cultivars. Moreover, physically or chemically induced mutations are very crucial for induced mutagenesis, allowing for the generation of mutations for any gene of interest and the

predictable development of multiple mutations for genes of interest. That means single mutagenic treatment may suffice for improvement of several traits. This phenomenon is common in mutation of major genes, because when a gene that has influence on expression of multiple traits undergo mutation it might be accompanied by changes in several traits simultaneously. According to Forster and Shu (2011), induced mutations occur randomly across the whole genome and within any locus or gene. This does not only provide the probability of generating mutations for any gene of interest but also, enables the development of multiple mutations for any target gene in a predictive manner. This phenomenon known as pleiotropy is common in mutation breeding. It allows multiple breeding objectives to be achieved within a relatively short period compared to conventional breeding approaches which require several generations of backcrossing or two-way and three-way crosses before stacking relevant genes.

In contrast to conventional breeding, mutation breeding is also advantageous in fixing a defect in an otherwise elite cultivar, and yet retaining all its agronomic and quality characteristics. It is the only straightforward technique for improving seedless crops (Pathirana, 2011). Therefore, the adoption and application of mutagenesis in plant breeding programmes in sub-Saharan Africa would result in rapid development of entirely new cultivars using minimum resources. This is one sure way of matching rapid varietal development with rapid population growth in Africa. It is therefore, not surprising, to see that in terms of distribution of mutant varieties, Asia is leading in the release of mutant varieties (FAO/IAEA, 2018).

According to Pathirana (2011), a desired mutant gene may be incorporated in a commercial cultivar through conventional breeding. When one or more of the mutant cultivars possessing the desired characters is or are poorly adapted, back-crossing with elite commercial cultivar becomes indispensable. This increases the turnaround time of the breeding programme and results in a decrease in the resources spent on developing an improved variety to meet a breeding objective targeted at addressing a challenge or source of stress which requires urgent attention. Also some crop species and varieties are characterized by poor combining ability of their parental genotypes when exploited in conventional crossbreeding programmes. For example, in aromatic rice cultivars, crossbreeding with non-aromatic cultivars may result in reduced aroma and grain quality (Bourgis *et al.*, 2008; Pathirana *et al.*, 2009; Pathirana, 2011). Induced mutation may be employed in such situations to circumvent this bottleneck and generate cultivars with desired traits. Thus, in cowpeas for example, it is possible to develop in one genotype, an insect pest resistant and high yielding mutant variety within a relatively shorter period. This would help avert an otherwise devastation due to the insect pest which would have resulted in a total crop failure, thus, affecting food and nutritional security. Induced mutation techniques are also ideal in circumstances where genes conferring detrimental characters are strongly linked to genes controlling traits of interests, where induced mutations may be used to generate crossing-over event or isolation of an independent mutation for the traits of interest (Pathirana, 2011).

Conclusion

If sub-Saharan Africa is to produce enough food to match its rapid population growth, to ensure food and nutritional security and export, we must add induced mutagenesis to our plant breeding mix. The development of useful mutant cultivars has been revolutionizing agriculture not only in densely populated developing countries but also in agriculturally advanced countries.

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