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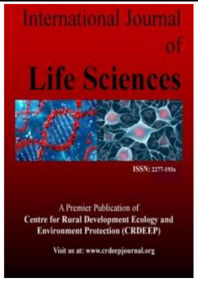

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Review Paper

The Role of Mutation breeding for Innovative Dry Land Crop Production in Ethiopia in the Face of Global Climate Change: A Review

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ABSTRACT

Currently, increasing crop yields to ensure food security is a major challenge in the dry lands of Ethiopia due to the synergistic effects of the agro ecological factors of the dry lands, anthropogenic factors, shrinking resource, lack of scientific agriculture and climate change that poses major bottlenecks for attempts for food security packages. Ethiopia has planned Climate Smart agriculture (CSA) through its climate resilient green economy strategy which creates a win-win situation between climate change and agricultural development. Climate-smart dry land agriculture can maintain crop productivity and lessen the impact of climate change in one way or another. To solve the aforementioned bottlenecks of dry land agriculture, it is scientifically proved that mutation breeding has great contribution. Crop varieties generated through the exploitations of mutation breeding are significantly contributing to global food and nutritional security and improved livelihoods. Mutation breeding is an important tool in crop improvement and is free of the regulatory restrictions imposed on genetically modified organisms. In the International Atomic Energy Agency (IAEA) mutant database, over 3000 officially released mutant varieties have been released worldwide in cereals, ornamental plants, fruits, vegetables, and oil crops. As a result, sustainable food production has been maintained. The genetic fidelity of the regenerated plants is highly desirable for developing new improved plant varieties and a useful as a reliable tool for feeding the ever-growing human population, especially under climate change and limited arable land. But Ethiopia, with its vast area dry land agriculture sector coupled with the ever increasing climate change impact is not taking advantage of the fortunes of the cost effective technology of Mutation breeding. Hence this paper identified the gaps and tried to synthesize the available scientific works on the significance of mutation breeding for proactive dry land agriculture and upon molding the information provided its own analysis-synthesis on the ways forward.

Introduction

Climate change is predicted to have major adverse consequences for the world‘s ecosystems and societies. Although a global phenomenon, the severity of the adverse effects of climate change will differ significantly across regions, countries and socioeconomic groups. Poor countries will suffer more, with the poorest in the poor countries likely to suffer most. Africa is highly vulnerable to the potential impacts of climate change and Ethiopia is often cited as one of the most vulnerable and with the least capacity to respond and adapt. Agriculture is the backbone of Ethiopia‘s economy, contributing 42% of the GDP and supporting 85% of employment (FDRE 2011). Agricultural production in Ethiopia is dominated by small-scale subsistence farmers, and is mainly rain-fed, thus highly exposed to climate variability and extremes. According to the World Bank (2006), current rainfall variability already costs the Ethiopian economy 38% of its growth potential. Climate change is likely to worsen this already distressing situation. [6].

The major predicted impacts of climate change on Ethiopia‘s agriculture include frequent droughts and dry spells, shortened growing season, and increased incidence of pests and diseases (NMA 2007). Without effective adaptation, there is likely to be a decrease in the total area suitable for crop production in the country. A study based on the Ricardian method predicts that a unit increase in temperature could result in reduction of the net revenue per hectare by US$177.62 in summer and US$464.71 in winter seasons [17].

Increasing crop yields to ensure food security is a major challenge. Amongst the obstacles against this are the changing climate (increasing temperatures and more erratic rainfall) which most often compromise crop productivity (Parry et al. , 2005) and the need to produce addition al food and crops for bioenergy whilst minimizing the carbon costs of production There is therefore an urgent requirement for new higher yielding varieties with improved nutrient and water use efficiency [83].

Review of Scientific Literatures

Causes and challenges of climate change

The emission of greenhouse gases from anthropogenic activities such as industrial process, land use change and agriculture are the main drivers of climate change. In Ethiopia, agriculture contributes 80% of total country's green house gases emission and CH, N4O and CO, respectively, contributed 71.5%, 14.58% and 13.92% to aggregated emission (UNFCCC 1995). 22 To calm down the impacts of climate change, countries should act now, act together and act differently on the stabilization of greenhouse gases concentrations in the atmosphere at a level that would prevent dangerous anthropogenic interference with the climate system within a time frame sufficient to allow ecosystems to adapt naturally to climate change, to ensure that food production is not threatened and to enable economic development to proceed in a sustainable manner (UNFCCC 1992). How can agricultural greenhouse gases emissions be reduced or sequestration enhanced while maintaining and even increasing food supply, particularly in dry land agriculture? This can be answered by adopting climate-compatible agricultural development strategies which encompasses development, mitigation and adaptation strategies [11].

Climate- smart agriculture in the dry lands

Climate-smart agriculture can be defined as agriculture that sustainably increases productivity, resilience (adaptation), reduces/removes greenhouse gases (mitigation), and enhances achievement of national food security and development goals [2,15,21].Agricultural development, particularly in dry lands, is a victim of climate change because it is estimated that higher temperatures could reduce crop yield by 10-20% in Sub-Saharan Africa by 2050. In return agricultural development is one of the causes of climate change because it is responsible for 10-12% of human-generated greenhouse gases emissions each [12].Agricultural development must be effective in terms of food production, reducing GHGs emission and helping farmers adapt to climate change [14,4]. Ethiopia has planned CSA through its climate resilient green economy strategy which creates a win-win situation between climate change and agricultural development [1, 3, 25].CSA in dry lands need the management of grazing lands, agricultural lands, and water and vegetation resources. For example, climate-smart livestock production is less 16,929,022t CO2/yr than the conventional livestock production. Conservation agriculture characterized by three principles, namely: continuous minimum mechanical soil disturbance; permanent organic soil cover; and diversified crop rotations/ plant associations could maximize production and reduces GHG emissions (FAO 2008, 2001, Hobbs et al. 2008). There is a need to enhance physical water productivity defined as the ratio of the amount of agricultural output to the amount of water used and economic productivity defined as the value derived per unit of water used [7].The ways in which vegetation resources are used and managed determine the future direction of climate change in drylands [1].

Climate Change and Its Impact on Plant Genetic Resources

The most profound and direct impacts of climate change over previous decade and the next few decades will surely be on agriculture and food security. The effects of climate change will also depend on current production conditions .The area where already being obstructed by other stresses, such as pollution and will likely to have more adverse impact by changing climate. Food production systems rely on highly selected cultivars under better endowed environments but it might be increasingly vulnerable to climate change impacts such as pest and disease spread.

If food production levels decreases over the year, there will be huge pressure to cultivate the crops under marginal lands or implement unsustainable practices that, over the long-term, degrade lands and resources and adversely impact biodiversity on and near agricultural areas. In fact, such situations have already been experienced by most of the developing countries. These changes have been seen to cause a decrease in the variability of those genetic loci(alleles of a gene)controlling physical and phenotypic responses to changing climate [26].Therefore, genetic variation holds the key to the ability of populations and species to persist over evolutionary period of time through changing environments[27]. If this persists, neither any organism can predict its future (and evolutionary theory does not require them to) nor can any of those organisms be optimally adapted for all environmental conditions. Nonetheless, the current genetic composition of a crops species influences how well its members will adapt to future physical and biotic environments. The population can also migrate across the landscape over generations. By contrast, populations that have a narrow range of genotypes and are more phenotypic ally uniform may merely fail to survive and reproduce at all as the conditions become less locally favorable. Such populations are more likely to become extirpated (locally extinct), and in extreme cases the entire plant species may end up at risk of extinction. For example, the Florida Yew (*Torreya taxifolia*) is currently one of the rarest conifer species in North America. But in the early Holocene (10,000yearsago), when conditions in southeastern North America were cooler and wetter than today, the species was probably wide spread. The reasons for that are not completely understood, but *T.taxifolia* failed to migrate towards the north ward as climate changed during the Holocene. Today, it is restricted to a few locations in the Apalachicola River Basin in southern Georgia and the Florida panhandle. As the *T. taxifolia* story illustrates, once plant species are pushed into marginal habitat at the limitations of their physiological tolerance, they may enter an extinction vortex, a downward cycle of small populations, and soon [28, 29].Reduced genetic variability is a key step in the extinction vortex .Gene banks must be better to respond to novel and increased demands on germplasm for adapting agriculture to climate change. Gene banks need to include different characteristics in their screening processes and their collections need to be comprehensive, including what are now considered minor crops, and that may come with huge impact on food baskets.

Crop Induced mutations

In situations where it is either impossible or impractical to source heritable variations from existing germplasm, the induction of allelic variations becomes an appealing option. Mutation, the heritable alteration to the genetic blueprint, has been the main driver for evolution and hence speciation and domestication of both crops and animals. Following the sublime discovery of X-rays and other forms of radiation in the early 20th century and the subsequent demonstration that these could alter the genetic material permanently, scientists have induced mutations in plants using both physical and chemical agents [30-32]. Induced mutation is hence an established crop improvement strategy and is credited with the development of over 3,200 officially released elite crop varieties and ornamental plants being cultivated all over the world [33]. The induction of mutation is a chance event so scientists traditionally enhance their chances of success at inducing useful mutation events by generating massive numbers of putative mutants that are then subsequently screened. This is expensive and time-consuming with the associated sheer drudgery cited as main reason for seeking other means for exploiting heritable variations in crops. Biotechnology applications are now being used to enhance the efficiency levels for producing and evaluating large populations. For instance, the high throughput reverse genetics technique, TILLING, short for Targeted Induced Local Lesions IN Genomes [12, 33, 35, and 36] permits the efficient screening of large populations of plants for specific mutation events [37-39,40-47]. The specificity, and hence efficiency, of TILLING - it identifies mutation events in predetermined genome regions - holds great promise for the use of induced mutations to broaden the genetic base of crops. Cell and tissue biology techniques are also used to enhance the efficiency of mutation induction. For instance, with doubled haploidy [48,49], homozygosity of the mutated segments of the genome is achieved rapidly while in vitro propagation techniques are used to dissociate chimeras quickly (to generate solid homohistonts) and to produce and manage large mutant populations in cost-, time-, and space efficient manners [50]. The critical importance of other uses of cell biology techniques, for instance, in germplasm conservation, in overcoming hybridization barriers and in therapid multiplication of disease-free planting materials makes it an indispensable tool in crop improvement in general.

Concepts of Mutation breeding

Mutagenesis is the process whereby sudden heritable changes occur in the genetic information of an organism not caused by genetic segregation or genetic recombination, but induced by chemical, physical or biological agents.[28].Mutation breeding employs three types of mutagenesis. These are induce d mutagenesis, in which mutations occur as a result of irradiation (gamma rays, X- rays, ion beam, etc.) or treatment with chemical mutagens; site-directed mutagenesis, which is the process of creating a mutation at a defined site in a DNA molecule; and insertion mutagenesis, which is due to DNA insertions, either through genetic transformation and insertion of T-DNA or activation of transposable elements. [29, 56] Plant breeding requires genetic variation of useful traits for crop improvements.[34] However, multiple mutant alleles are the sources of genetic diversity for crop breeding as well as functional analysis of the targeted gene in many cases . The key point in mutation breeding is the process of identifying individuals with a target mutation, which involves two major steps: mutant screening and mutant confirmation.[56] Mutant screening is a process involving selection of individuals from a large mutated population that meet specific selection criteria, e.g. early flowering, disease resistance as compared to the parent. However, these selections are often regarded as putative mutants or false mutants. Mutant confirmation, on the other hand, is the process of reevaluating the putative mutants under a controlled and replicated environment using large samples. Through this process, many putative mutants are revealed to be false mutants. In general, the mutations that are important in crop improvement usually involve single bases and may or may not affect protein synthesis.[52]

Mutation breeding strategy for obtaining mutants

Any mutation breeding strategy requires several sequential steps. The effectiveness of mutation breeding over other breeding methods depends up on the efficacy of selection of useful variant mutants in the second (M2) or third(M3) generation. The first step in mutation breeding is to reduce the number of potential variants among the mutagenized seeds or other propagules of the first (M1) plant generation to a significant level to allow close evaluation and analysis.[51] Determination of the target population size in the first generation of mutants is a prerequisite for potential success in any mutation breeding programme.

The targeted population should be fixed so as to allow a high number of mutation measurements. Thus, the population size should be managed effectively by the breeder. It should be noted that the population size depends on the inheritance pattern of the target gene.

Therefore, it is advisable to select mutagens that give a high mutation frequency so as t o reduce the population size of the M generation. [51] Genetically, Ml1mutant plants are heterozygous. This is because only one allele is affected by one mutation during treatment. However, the probability of having a mutation on both the alleles concurrently is a product of individual probability of mutation. Therefore, its occurrence is extremely low. Moreover, in M, only dominant mutations can be identified, while it is impossible to identify a recessive mutation expression at this stage. In this case, a plant breeder should attempt screening mutations in subsequent generations where segregation will occur [51]. Consequently, the plant breeder generates homozygotes for dominant or recessive alleles. Caution should be taken to pr event cross pollination among the Ml population as this would lead to generation of new variation which will be difficult to differentiate from the effect of mutation.[51,53] Screening and selection start in the M12 generation. Roy Chowdhury*etal*.[54] discuss three main types of screening/selection techniques. These are physical/mechanical, visual/phenotypic and other methods. Physical or mechanical selection can be used efficiently to determine the shape, size, weight, density of seeds, etc., using appropriate sieving machinery. Visual screening is the most effective and efficient method for identifying mutant phenotypes. Visual/phenotypic selection is often used in selection for plant height, adaptation to soil, growing period, disease resistance, colour changes, earliness in maturity, ion-shattering, climate adaptation, etc. In the category o f ‘others’, physiological, biochemical, Chemical, physio-chemical procedures for screening may be used for selection of certain types of mutants. When a mutant line appears to possess a promising character, the next stage is seed multiplication for extensive field trials. In this case, the mutant line, the mother cultivar and other varieties will be tested.

The methods for comparative trials of mutants are the same as t hose for any other newly developed varieties. The purpose of field trials is to find whether the mutant promises to become a commercial variety that is s superior to the mother cultivar.

Prior to release as a commercial variety, the promising mutant should be studied for combinations of different characters like growth habit, structure and yield components in a wide range o f environments under varying water availability, plant density, sowing dates, etc. [51]

Applications of Mutation breeding in basic research

Global food security deteriorated drastically in 1960's when developing countries like Pakistan and India were desperately short of the food supply. Fortunately, agriculture research responded with a new production technology which has popularly been called as "Green Revolution Technology". This aided to avoid large scale starvation for around four decades however, food security problem has again seen a major deterioration in the last few years; sky high food prices and once again poor people of the world are challenged with severe malnutrition the underlining causes that drove to food security deterioration; increasing fertilizer and fuel prices, erratic rain falls, severe drought conditions, excessive floods, divert of food grains into biofuel production will remain for the decades to come. Food security will even get worse since the population is still expanding while no significant increase in arable lands is foreseen. Therefore a newer green revolution is required to solve the problem of food insecurity in the decades to come. The gigantic advent of induced mutation breeding is anticipated to promise a sound solution to further increase food production by both increasing grain production and stability. In this regard, induced mutagenesis is gaining importance in plant molecular biology as a tool to identify and clone genes and to study their structure and function [41]. The application of mutation techniques has generated a vast amount of genetic variability and is playing a significant role in plant breeding and genetics and advanced genomics studies.

Recently mutation breeding techniques have also been integrated with other molecular technologies such as molecular marker techniques or high throughput mutation screening techniques are becoming more powerful and effective in breeding crop varieties.

Mutation breeding is entering into a new era; molecular mutation breeding. Therefore induced mutation breeding will continue to play a significant role in improving world food security in the coming years and decades.

The widespread use of mutation techniques in plant breeding programmes throughout the world has generated thousands of novel crop varieties in hundreds of crop species, and billions of dollars in additional revenue [34]. The wide spread use of induced mutations in plant breeding programs has led to the release of elite mutant plant varieties. Such mutants play a significant role in designing crops with improved yield and yield contributing traits, quality and longer shelf life, enhanced stress tolerance and reduced agronomic inputs. The knowledge of biochemistry, physiology and development of plants has rapidly advanced with the introduction of T-DNA insertional mutagenesis. The auxin mutants such as aux1, pid, mp and lop1 have suggested implications in auxin transport, inhibition, uptake and signal transduction [42]. The understanding mechanism of cytokinin action was elucidated with the identification of mutants with elevated cytokinin level (amp1), photomorphogenic mutant (det1, cop) cytokinin resistant mutant and cell division mutants [43]. Schmulling et al. in 1997 identified Cytokinin mutants such as ckr1, ein2, cry1, stp1 and zea3 in Arabidopsis thaliana [44]. These mutants have elucidated the role of cytokinin- regulated genes in diverse biological processes, ranging from cell division, photosynthesis, chloroplast development, disease resistance and nutrient metabolism.

Chandler and Robertson, 1999 elucidated the mechanism of action of growth hormone gibberellin with the screening of dwarf le mutant of pea and dwarf mutants of maize [45]. Several dwarf mutants such as d8 in maize and Rht3 in wheat are GA deficient and do not respond to applied GA3 [63]. These dwarf mutants have contributed significantly in developing resistant and high fertilizer responsive varieties.

Several ABA deficient mutants such as aba1 in Arabidopsis and aba2 in N. plumbaginifolia [47.49] and ethylene response mutants have been isolated [50]. These mutants are highly valuable and have a major role in increasing the shelf life of fruits and extended flower-life and delayed senescence as shown by its transfer to tomato and petunia [67].

A series of homeotic mutants with defective flowers have been identified in Petunia, Antirrhinum and Arabidopsis. The isolation of these mutants has contributed significantly to understand patterns of flower development [68]. Homeotic mutants for leafy cotyledons lec are defective in the maturation of embryos which remain green have been developed through insertional mutagenesis [69]. The mutants which determine the development of seed e.g. fis mutant have a crucial role in understanding the apomixes [70]. The developmental patterns in crop plants play a vital role in yield and yield attributed traits. The manipulation of these patterns will assume a new dimension in plant breeding in near future.

Participatory plant breeding

Factoring in the perspectives of the growers and other stakeholders such as consumers, extensionists, vendors, industry, and rural cooperatives in the crop improvement endeavor of developing new varieties is known as Participatory Plant Breeding (PPB) ; [67]). The need for this paradigm in plant breeding is probably greatest in developing countries relative to the industrialized countries where market forces determine agricultural research and development (R&D) themes including plant- breeding objectives. By having farmers and other end-users involved in the development of varieties, feedback mechanisms are enhanced hence improving the relevance of the breeding activities to the needs of the growers.

Farmers' participation in plant breeding can be categorized under the three stages of design, testing, and diffusion[69].During the design stage, breeding goals are set and variability to be used created while at the testing stage, the breeding materials are evaluated and narrowed down to the few promising ones. The diffusion stage encompasses activities spanning varietal release, on-farm trials under farmer management and the identification of the mechanisms for the dissemination of the seeds and planting materials of the improved varieties.

Farmers, as the custodians of PGRFA, have over the several millennia o f selecting from, improving, and exchanging local genetic diversity contributed immensely to the diversity of plants we grow. With the upsurge in the ready availability of modern crop varieties bred in research institutes, the roles of farmers in ensuring diversity and adding value to PGRFA have waned significantly. One effect of this shift is the precariously narrow genetic base of the modern crop varieties. The obvious threat that this poses to food security calls for the systematic re-integration of farmer s' knowledge and perspectives in the developing of modern crop varieties. PPB is avertable and validated means for ensuring this.

Future prospects of Mutation breeding

In recent years interest has rekindled in mutation research since induced mutagenesis is gaining importance in plant molecular biology as a tool to identify and isolate genes and to study their structure and function. These studies will definitely have a major impact on the future crop improvement programmes [71]. Mutation in association with the new technology of genetic engineering will constitute tools of plant breeders in near future. Although most of the varieties released so far has been developed from a mutation in combination with the direct selection. In the present era in vitro culture and molecular methods have resulted in the creation of new and wide paradigm in the utilization of mutation breeding for crop improvement. Recently, heavy ion beam irradiation has emerged as an effective and efficient way of inducing mutation in many plant varieties because of its broad spectrum and high frequency [72]. In recent years in vitro mutagenesis technique has enhanced the crop yield and germplasm innovation by the development of quality and improved resistance traits [73]. In in vitro culture techniques, a small amount of tissues and calli can be subjected to mutagenesis for the betterment of crop species [74].

Currently, the use of in vitro mutagenesis is low, very little number of plants such as banana and sugarcane have been regenerated through this technique. On the other hand, many seed propagated plants such as wheat, rice, maize and barley can now be regenerated from cell suspension cultures [74]. In future development of in vitro cell selection techniques for disease resistance would be equally important. A coordination of the recent techniques of anther and microspore culture, cell suspension, irradiation of haploid cells and chromosome doubling and regeneration of doubled haploid plants could be utilized to obtain genotypes with desired traits [75].

The induced mutation has also proved useful in the preparation of genetic maps that will facilitate molecular marker assisted plant breeding in future [76]. Mutation breeding has become increasingly popular in recent times as an effective tool for crop improvement [77].

The direct use of mutation in the development of molecular maps in structural and functional genomics could lead to rapid improvement of plant yield and quality. The molecular techniques of DNA fingerprinting and molecular mappings such as RAPD (Random Amplified Polymorphic DNA,) AFLP (Amplified Fragment Length Polymorphisms) and STMS (Sequence-Tagged Microsatellite Sites) have contributed significantly in the screening and analysis of mutants. Site directed insertion of transgenes based on chimeric RNA/DNA oligonucleotides as done in tomato [80] and maize and mutant tagging will be widely used in gene technology [79]

Conclusion and Recommendation

In the current world scenario of crop production there are prevailing dangers of Genetic erosion, shrinking natural resources, loss of wild type genetic resources, climate change, anthropogenic factors, biotic and a biotic stresses, alarming population growth, etc are posing an alarm for future global food security. Ethiopia is even more vulnerable to the aforementioned scenarios unless coping up measures are aggressively taken. Hence this paper concluded and recommends innovative and proactive responses for the aforementioned factors. With this connection and taking a piece of the solution, mutation breeding will help in broadening the gene pool of crop species and developing varieties tolerant to the prevailing biotic and a biotic stresses, biofortified varieties and climate proof varieties in which a segment of this solution will be an input for climate smart agricultural packages thereby contributing its own for food security. Hence I recommend if our country Ethiopia will take advantage of the unleashed potential of mutation breeding thereby reinovating the exhausted potential of conventional breeding approaches hitherto, specifically for the dry land agricultural discourses.

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