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Mutation Breeding and Its Importance in Modern Plant Breeding: A Review

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This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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Review Article

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ABSTRACT

A mutation is an abrupt, heritable alteration in a living cell's DNA that is not brought about by genetic recombination or segregation. The deliberate use of mutations in plant breeding is known as "mutation breeding." Mutation breeding provides the advantage of improving a fault in an otherwise excellent cultivar without sacrificing its agronomic and qualitative features, in contrast to hybridization and selection. There is no simpler solution than mutation breeding to enhance seedless crops. These benefits have led to the development of a market for mutation breeding in plant breeding since the initial release of mutant cultivars derived from fundamental mutation research in Europe. Both physical and chemical mutagens have improved methods for inducing mutations in major crops, and strategies for selecting mutant populations have been detailed. A broad range of mutations that have not been previously documented have been detected, and new mutagenic factors like cosmic rays and ion beam radiation are being studied. However, ionising radiation and alkylating chemicals continue to be widely used. The efficiency of mutant breeding has increased as a result of the advent of reliable in vitro methods for numerous crop species. In vitro methods are particularly effective because they can manage sizable mutagenized populations in a small area, have a quicker progeny turnover rate in vegetatively propagated species, and can

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screen for a variety of biotic and abiotic stress factors in the culture environment. Over the last ten years, there have been significant advancements in mutant screening, with reverse genetic methods now being prioritised. Thus, the combination of molecular methods and mutation techniques is opening up new and intriguing possibilities for contemporary plant breeding.

Keywords: Mutation breeding; agriculture; modern breeding; mutagenesis; crop improvement.

1. INTRODUCTION

Forster and Shu (2012) condensed the definition of "mutation" from a series of articles by de Vries (1901, 1903, 1905) to mean abrupt heritable changes in an organism's genetic material that are unrelated to recombination or segregation. A mutation is an abrupt, heritable alteration in a living cell's DNA that is not brought about by genetic recombination or segregation. The deliberate use of mutations in plant breeding is known as "mutation breeding." Genetic variety in living organisms can be increased through spontaneous (natural) or purposefully produced mutations. Numerous plant species have been domesticated in crops thanks to spontaneous mutant features, which has had a huge impact on civilization [1-3]. The genetic variety that enabled crop adaptation to new conditions was made possible by mutations; many of our key crops are farmed in locations that are distant from the progenitors' centres of origin. Consequently, main crop species are suited to a broad variety of longitudes, latitudes, and altitudes; these also differ significantly in terms of physical circumstances (such as light, temperature, soil, and water) and biological factors (such as pests and diseases).

1. Breeding plants is essentially the human-guided evolution of crop plants, with genetic variety serving as the basis. Crossbreeding can be used to accomplish a crop's breeding goals when desired variety exists across its cultivars. But when one or more of the parent cultivars with the desirable traits for crossbreeding are not well suited, a back-crossing technique must be utilised to regain the elite type. A further issue with traditional crossbreeding is that certain parental genotypes have inadequate combining capacity. For instance, cross-breeding aromatic rice varieties with non-aromatic rice varieties will result in a loss of scent and quality. Aromatic rice cultivars also have low combining ability. In these situations, it may be advantageous to use mutation induction to create cultivars within certain germplasm pools that have the necessary features. Genes influencing desirable features can occasionally be closely connected to genes delivering undesirable characteristics. In this case, the induction of mutations may lead to the isolation of an independent mutation for the desired phenotype or a crossing over event. For crops lacking seeds, like edible bananas or seedless grapes, the only feasible method to enhance variety and create new cultivars would be through mutation induction. This also holds true for many root and tuber crops, as well as the emergence of unique hues and variations in species of ornamental plants that are vegetative propagated. The first articles on induced mutations date back eighty-three years, to (Muller and Stadler) for more than 50 years, plant mutant breeding has been actively used, and developing nations are helped to swiftly embrace new technologies by the Joint FAO/IAEA Division, which is headquartered in Vienna. With the use of this strategy, mutation technology has been used widely and 3211 registered mutant varieties have been released in over 170 different plant species to date. A few of these mutant cultivars have completely changed agriculture in both agriculturally developed and heavily populated emerging nations. “Mutation breeding was also able to embrace and utilize the most recent results and technological breakthroughs in plant genomes and molecular biology research thanks to the use of molecular and genomic tools for mutant screening and characterization” [4].

1.1 Types of Mutation

Since whole genome analysis methods are still relatively new, most research on induced mutations in plants has concentrated on visible features. Thus, the initial classification of mutant plants was based on the phenotype that they displayed. One excellent example of a database where mutant genetic stocks are categorised by

phenotype and supported by textual descriptions and photographic exhibits is the barley genetic stocks database (*http://ace.untamo.net*). Lundqvist *et al.* (2012) provides “information on the genetics of the designated mutant (dominant/recessive, number of alleles, and map position), the source of mutation (natural or induced, and the genotype in which it occurred), the deployment of the mutant, and references”.

1.1.1 Genome mutations

These mutations cause aneuploidy, or the addition or deletion of chromosomes within a genome, as well as ploidy, or variations in the number of genomes. By creating haploid embryos, pollen irradiation has been utilised to decrease the number of genomes (Gustafson and Ekberg 1995; Murovec and Bohanec 2012). Plant breeders can benefit from the usage of haploids (Forster and Thomas, 2005). “Fascinatingly, a chromosomal spindle attachment failure during mitosis resulted in an induced mutant in Arabidopsis that was detected by TILLING-facilitated haploid production” (Ravi and Chan 2010). “Both autoploidy and alloploidy can be attained spontaneously or through induction as genome additions. Numerous crop species, such as strawberry (8x), bread wheat (6x), plum (6x), durum wheat (4x), triticale (4x), potato (4x), tobacco (4x), chrysanthemum (4x), tart cherry (4x), cotton (4x), and banana, are either autoploid or allopolyploid. Because these mutations occur at random, the cause of them is difficult to pinpoint. Even in a healthy, uncontaminated cell, spontaneous mutations have a non-zero chance of occurring. In humans, oxidative DNA damage occurs 10,000 times per cell per day, while in rats, it occurs 100,000 times per cell per day. The specific change can be used to identify spontaneous mutations” [5]. Crop output, enrichment of gene variety (new genes introduced), genetic buffering and heterosis promotion, and morphological enlargement of the nucleus, which results in cell and plant tissues, are among the benefits of polyploidy. Old Rubber, soybeans, rice, and maize are examples of allopolyploid crops that essentially act like diploids during meiosis by forming bivalents. Interactions, exchanges, and fusions both within and between genomes can result in genomic rearrangements. Chromosome translocation, duplication, and deletion mutations can occur spontaneously and are frequently linked to the evolution of species, but they can also be produced. The Ph1 gene tightly regulates chromosomal pairing in allopolyploid species like wheat (which has three genomes: AABBDD), ensuring that recombination is limited to homologous chromosomes. Nevertheless, a mutation in the Ph1 gene known as the ph1b mutation permits homoeologous pairing of chromosomes from different genomes, thereby facilitating gene transfer between the DD, BB, and AA genomes of bread wheat during meiosis. This mutation has been utilised in wheat breeding and genetics (Law and Worland 1987). The first documentation of mutant selection in plant breeding: maturity and other traits in cereals in China is found in an ancient Chinese book. The first verifiable (spontaneous) plant mutant, the larger celandine 'incisa' mutant, was described in 1590.

1.1.2 Chromosome mutations

Deficits in one or more chromosomes, extra chromosomes, chromosome replacements, and chromosome rearrangements are examples of aneuploids. They may arise organically or be created by crossing, particularly in situations where parents are making unequal contributions quantity of genomes or chromosomes. Radiation can cause aneuploidy by causing chromosomes to randomly delete. A wheat euploid is renowned and widely applied in plant breeding and genetic research (Law and Worland 1987; Shimelis and Spies 2011). Professor Ernest Robert Sears was a pioneer in the discovery and investigation of wheat aneuploidy [6]. Chromosomal breakage and subsequent rearrangements lead to chromosomal rearrangements. Although chemical and physical mutagens can also cause these changes, ionising radiation is the main cause of these alterations. Errors in repair processes may result in translocations, inversions, duplications, and deletions. About 90% of deletion mutations caused by ionisation radiation are fatal.

1.1.3 Gene mutations

Smaller modifications, including single nucleotide changes or tiny indels, are frequently seen in mutations inside genes. These mutations could produce a new allele and be functional. They therefore offer valuable novel variation, which makes them interesting for plant breeding. Ashikari et al. [7] examined the semidwarf mutant in rice, which is significant for agriculture. They found that the GA20 oxidase gene (GA20ox-2) had nucleotide deletions or changes that caused either an internal stop codon or a single amino

acid mutation. Resistance to more nucleotide changes has emerged in crop plants, including rice, wheat, barley, and soybeans. diseases and pests, improved abiotic stress tolerance, plant height, and biochemical quality [8]. The majority of reverse genetics In order to demonstrate the usefulness of gene mutations for functional genomics and agricultural enhancement, TILLING projects have concentrated on the recovery of point mutations targeted to gene sequences. Gene function is unaffected by mutations that happen in non-genic areas. These are known as quiet mutations and are frequently neutral. Typically, they entail single-base alterations, or point mutations, which have no effect on transcription [9]. Chemical mutagens like alkylating chemicals often cause silent mutations. Silent deletions can also occur from deletions of intergenic regions, where regulatory elements remain unaffected. However, a frame shift in a gene's coding sequence might result from base loss or insertion. A premature stop codon and a malfunctioning peptide may result from this. One instance of biological mutagenesis is transposon insertion, in which transposable elements (TEs) relocate and reintroduce themselves into the genome. Numerous environmental stressors have the ability to cause transposon migration, which frequently leads to the loss of gene function (null alleles). Examples of significant crop traits resulting from transposon insertion are provided by Lisch [10] in a recent review. These include the colour shift of grapes from black to green (chardonnay), red fruit in oranges and grapefruits, seedless apples, tomato fruit shape, and common morning glory flower colour. Rapid advancements in molecular genetics and genomics have made a variety of DNA technologies available for crop improvement, as well as transforming breeding and bringing molecular plant breeding methodologies to fruition [11].

1.1.4 Non-nuclear mutations

In higher plants, mutations involving plasmons or extranuclear DNA impact the genetic material of mitochondria, chloroplasts, and organelles. Principal instances include chloroplast changes that provide antibiotic resistance in rapeseed [12] and Nicotiana tabacum [13], as well as cytoplasmic male sterility—a trait that is very advantageous in F1 hybrid breeding. Furthermore, significant commercial characteristics like the colour of citrus fruits and the leaves of tobacco and decorative plants are impacted by changes in chlorophyll [14]; (Pathirana 2011).

1.1.5 Mutation mimics

Physiological abnormalities, such as soma clonal variations created in certain in vitro culture procedures, are the main component of mutation mimics. A phenotype, such as stunted growth or sterility, that resembles a known genetic effect may be caused by the physiological illnesses. Many times, the physiological disorder's underlying cause is unclear, but DNA Methylation is one of these reasons. After clonal tissue culture multiplication, oil palm offers a well-known and disturbing example of aberrant (mantle) fruit development that wasn't noticed until years after planting when the clones reached maturity [15,16]. Regarding mutant breeding, one of the main reasons mutations cannot be chosen or phenotyped in M1 populations is because these populations frequently exhibit high levels of physiological abnormalities as a result of the mutagenic therapy.

1.1.6 Plasmon mutations

The extranuclear genetic systems of the mitochondria and chloroplast are distinct in higher plants. It has been demonstrated that these genomes' mutations offer benefits for agriculture. Numerous writers have demonstrated that nitroso chemicals cause plasmon mutations to occur often. Chloroplast mutations for antibiotic resistance have been produced in Nicotiana through the use of nitroso chemicals. These mutations also cause the chlorophyll mutations that were seen in the first-generation following mutagen therapy. Since these mutations frequently arise as chimaeras, they can be advantageous in the development of ornamental crops with different leaf or fruit traits. Plasmid mutations are not Mendelian in nature; instead, they are maternally inherited and are transported through the cytoplasm. Van Harten *et al* goes into great detail on two more significant traits inherited by plasmonic genes: resistance to various fungal illnesses and cytoplasmic male sterility.

2. MUTAGENS AND ITS TYPES

Plant genome sequences have been modified by a wide range of distinct mutagens. These fall into the general categories of chemical, biological, and physical agents. “Physical mutagens can be

divided into two main categories: ionising atomic particle irradiation, which includes X-rays, gamma rays, and cosmic rays; these subatomic particles include electrons, protons, neutrons, deuterons, alpha, and beta particles" [17]; (Mba et al. 2012). "Fast neutron and ion beams are the most often used for inducing plant mutations" (Burtscher and Casta 1967; Li et al. 2002); [18]; (Tanaka et al. 2010). "Mutations can be induced using mutagens or mutagenic agents, which are chemical or physical agents that enhance the frequency of mutations. Mutations induced this manner are referred to as induced mutations. Induced mutations occur when a gene comes into contact with mutagens or other environmental factors" [19]. "The first person to report induced mutations in plants by X-ray genetic experiments on barley and maize was Stadler [20].

Higher plants are exposed to ionising radiation through two major interactions with genetic material: light can produce purine or pyrimidine and can also induce photochemical damage, dimers, which cause point mutations in the DNA sequence" (Pathirana 2011); [21,22]. High doses will cause more biological damage because, generally speaking, the effects of ionising radiation are proportionate to the energy absorbed in the exposed tissue and the applied dose. Plant breeding for crop improvement has made extensive use of ionising radiation; according to http://mvd.iaea.org, physical mutagens were responsible for over 80% of released mutant cultivars, while gamma radiation was responsible for over 60% of known mutant cultivars. Plant mutant breeding programmes of the FAO and IAEA have made available gamma irradiators and protocols. Physical mutagens can be applied to or have been applied to many kinds of plant materials; however, compared to seed irradiation, lesser dosages are employed for soft tissues (those with a high-water content). While some non-ionizing radiation, such UV light, can be employed, its capacity to penetrate tissue is known to be limited because it has a lower energy than gamma and X-rays (Mba et al. 2012).

Only low-density subjects, including spores or pollen grains, as well as thin single-cell layer samples, are suitable for ultraviolet treatments [23,24]. Because it cannot penetrate tissue, its use in plant breeding has been restricted.

The earliest application of chemical mutations, particularly mustard gas, was documented by Auerbach and Robson (1944), who also examined the results with ionising radiation upon the production of mutations. Many chemical mutagens have been identified to far; however, the majority are members of the alkylating agent class, which includes methyl nitrosourea (MNU), ethyleneimine (EI), EMS, diethyl sulphate (dES), ethylene nitroso urethane (ENU), and ethyl nitroso urethane (ENU) [25]. These substances react with genetic elements to produce N3 adenine, N3 cytosine, and alkylated O6 guanine. O6 guanine tends to convert G: C to A: T base pairs during alkylated base repair, N3 adenine leads in A: T to T: A transversions, and N3 cytosine frequently produces C: G to T: A transitions and C: G to G: C transversions. [26,27].

These single nucleotide mutations result in missense mutations, truncations, and an expansion of alleles; nonsense mutations can lead to premature stop codons and deletions of splicing sites [28]. Because of its efficiency and convenience of use—particularly its detoxification through hydrolysis for disposal—EMS is the most often used chemical mutagen in plant genetics (Pathirana 2011). More than 60% of the mutant cultivars created more than one-third of chemical mutagenesis treatments come from EMS, MNU, and ENU treatments (http://mvd.iaea.org). Additional non alkylating mutagens consist of (a) nitric oxide and nitrous acid; (b) base analogues and their derivatives; (c) some antibiotics, such as azaserine, mitomycin C, or streptozotocin; and (d) topoisomerase and intercalating agents.

Leitão [27] has described their mutagenic properties and application in plant mutation induction.

The act of producing a mutation at a specific location in a DNA molecule is known as this sort of mutagenesis [29]. Sodium azide, a common pesticide, bactericide, and industrial nitrogen gas generator, has been the most often employed non-alkylating mutagen in plant mutagenesis [30]; (Gruszka et al. 2012). In barley and broad beans, for instance, mixtures of chemical and physical agents have also been employed [31]. Girija as well as in a 2009 study, Dhanavel evaluated the effects of chemical and physical mutagenesis in plants, finding that gamma Ray affects the cytological, genetic, and developmental processes in plants. morphogenetic, physiological, and biochemical characteristics. Therefore, it was suggested that a combination of treatments

would be beneficial in creating a greater variety of mutations.

Transposable elements or the T-DNA of Agrobacterium tumefaciens are used in biological mutagenesis. The reader is directed to more review studies on these subjects, including Gierl et al. (1989) and Bennetzen (2000) on transposable elements and Azpiroz-Leehan and Feldmann [32] on T-DNA mutagenesis. We merely provide a cursory overview of transposable elements in relation to mutation induction here. Retrotransposons and DNA transposons make up 15–80% of TEs, which make up the majority of eukaryotic genomes. The rice genome, for instance, has 30% of TEs. Specifically, the majority of repetitive DNA in plant genomes is made up of long terminal repeat (LTR) retrotransposons, which also significantly increase the size of genomes in species with bigger genomes, like maize, which has a genome size of 2.3 Gb and more than 75% of LTR retrotransposons (Sanmiguel and Bennetzen 1998). As demonstrated by the mutator class of transposons in maize and the TOS17 mutant population in rice (Meeley and Briggs 1995; Miyao et al. 2003); [33], crop plants may be susceptible to transposon mutagenesis provided transposons can be activated. McClintock's (1950) first discovery of transposons in maize kernel variegation opened the door for their subsequent application as crucial instruments for examining gene function, gene isolation, and gene cloning (Marks and Feldmann 1989; Zhu et al. 2012); [10]. Transposon insertion causes a disturbance in gene function or expression, which is the main way that transposable elements cause mutations. Marker-assisted selection is another application for transposons (Kalendar et al. 1999). Additionally, they are used in research on the composition and evolution of plant genomes (Fedoroff 2000). Although chemical and ionising radiation have proven effective in causing a variety of mutations in plants, it can be challenging to determine the precise relationship between the altered gene and the mutant phenotype. However, this can be done in Mutagenesis caused by transposon (Miyao et al. 2003; Zhu et al. 2012). However, physical mutagens like gamma rays have been employed as a stress management to promote transposon mutagenesis and transposon activity (Walbot 1988).

2.1 Physical Mutagens

Before the effects of specific compounds on DNA were described in the mid-1940s, ionising radiation was the only tool utilised in mutation research. X-rays were employed initially, but as the IAEA made g-rays from radioactive sources like 60Co and 137Cs available to many developing nations, they gained popularity. It is also possible to use fast neutrons from nuclear reactors, especially since the Joint FAO/IAEA Division in Vienna offers this irradiation service. DNA strands can cross-link when chromosomal

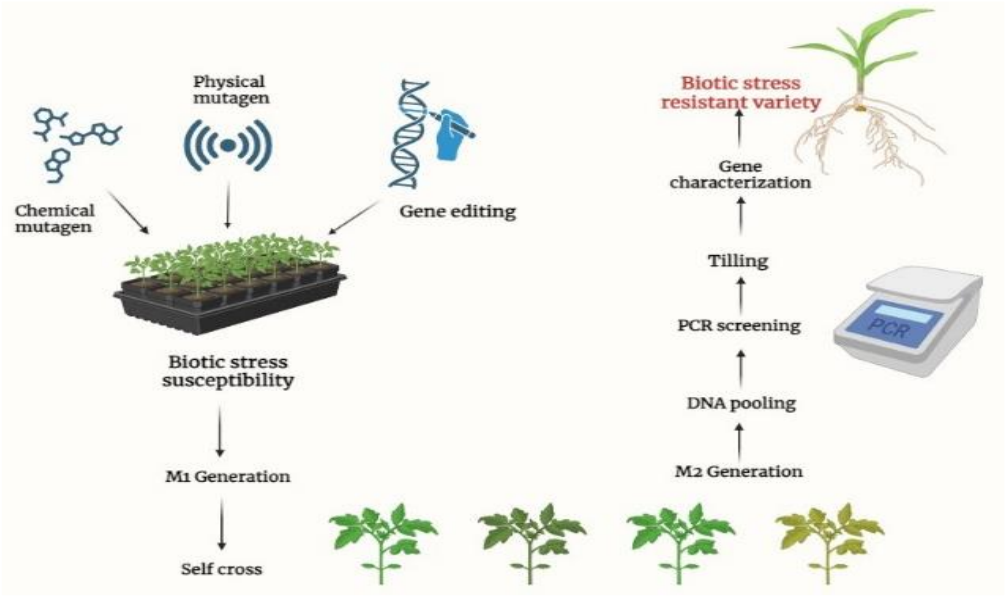

Fig. 1. Types and mechanism of mutation

breaks are caused by ionising radiation. Therefore, more harmful consequences than those of chemical mutagens that cause small DNA modifications like base-pair substitution should be anticipated. UV radiation is non-ionizing among physical mutagens. As a result, they are effective in creating purine or pyrimidine dimers, which lead to point mutations, and have low penetration. Pollen in the late or early uninucleate phases can be efficiently exposed to UV radiation. For the principal crop species, radiation dosages and conditions for irradiating seeds and buds have been optimised and published. As information may not be easily accessible, treatment conditions and dosages for novel or minor crop species must be determined through experiments. Temperature, oxygen and moisture contents, and storage conditions after radiation are the primary external elements that impact radiation.

2.2 Chemical Mutagens

Due to the harmful consequences of ionising radiation and the high rates of chromosome abnormalities it causes, scientists have been searching for other ways to cause mutations. Consequently, a wide range of chemical mutagens have been identified. However, a large number of chemical mutagens make it challenging to develop standard guidelines and treatment requirements. The literature has addressed the classification of chemical mutants, treatment strategies, post-treatment management, and post-treatment selection for the major crop species. Since sodium azide's mutagenic effects were first reported. no new chemical mutagens that are often used in plant breeding have been found.

Alkylating agents are the most commonly employed chemical mutagens; EMS is the most well-liked due to its efficiency and handleability, particularly its hydrolysis-based detoxification process for disposal. The other common alkylating agent is nitroso compound, but because to its increased volatility and sensitivity to light, further measures must be taken.

3. THE PAST, PRESENT, AND FUTURE OF MUTATION BREEDING IN CROP IMPROVEMENT

Plant breeding can be carried out using a variety of methods, from as simple as choosing plants with desired traits for propagation to more intricate molecular approaches. In recombinant DNA research, the use of radioactively labelled probes has become standard practice for cloning, mapping plant genes, and transgenesis, especially for RFLP, or microsatellite-based DNA fingerprinting. The utilisation of high throughput platforms like TILLING (Targeting Induced Local Lesions in Genomes) for the assessment of mutant crop varieties for particular sequence genomic modification has been made possible by recent advancements in publicly available genomics resources. With the advancement of TILLING technology over the past ten years, chemically induced mutagenesis has seen a resurgence in application. Surprisingly, better agronomic and botanic properties characterize the majority (48 percent) of the mutant varieties recorded in the Mutant Variety Database. This could be owing to the fact that botanic and agronomic features are easily observable, and for the most part, screening does not require specialized equipment [34]. Mutagenesis in TILLING is linked to the extraction of chromosomal DNA from each mutant and the use of sophisticated molecular tools to the population's molecular level screening. In actuality, TILLING employs the reverse genetic approach, which is high throughput, inexpensive, and compatible with a wide range of organisms, by using conventional mutagenesis and nucleotide polymorphism finding methods. The worldwide research community has received hundreds of induced mutations thanks to large-scale TILLING techniques. The use of nuclear techniques in plant breeding has been primarily directed towards inducing mutations. Since the discovery of X-rays, the use of ionising radiation, such as X-rays and gamma rays for creating variation, has become an established technology. Advances in mutation breeding techniques, such as in vitro mutagenesis, promise to increase further the improvement of crop varieties. Plant breeders have applied in vitro culture for rapid multiplication, molecular methods to select desired genotypes, mutagenesis to increase variation, and varied environmental conditions to manipulate traits [35-38].

3.1 Past Achievements

Over the course of the roughly 80-year history of induced mutations, numerous instances of the creation of novel and useful modifications in plant traits that considerably boost the potential yield of certain crops have been documented. The main goals of mutant breeding are to enhance the incidence of viable mutations. as well as the

frequency and spectrum of mutations. The primary goal has been to improve the well-adapted cultivars by the modification of characteristics like as disease resistance, maturity, and seed size, all of which are essential for raising yield and yield-attributed features. Through the process of mutant breeding, a variety of characteristics have been enhanced, including tolerance to biotic and abiotic stressors, length of maturity and flowering, and other traits that contribute to yield. Legumes and cereals are significant food crops, and plant breeders have focused much of their attention on improving these crops over time. These crops have been enhanced in the past via introduction, selection, and hybridization employing genetic variability that has been released through recombination or genetic variability that is already present. These days, induced mutagenesis offers the chance to produce previously unidentified alleles, resulting in a great deal of genetic variety. It is clear from the list of mutant cultivars created in legumes that this potential has been utilised in both cereals and legumes [39-42].

3.2 Basic Ongoing Research

In the 1960s, when developing nations like Pakistan and India were severely short on food, there was a sharp decline in global food security. Thankfully, agricultural research came out with a new production technique known as "Green Revolution Technology" in response. This helped to prevent widespread starvation for about 40 years, but in recent years, the problem of food security has gotten worse once more. The world's poor are once again facing extreme malnutrition due to skyrocketing food prices. The underlying causes of this decline include rising fuel and fertiliser prices, unpredictable rainfall, extreme drought conditions, frequent floods, and the diversion of food grains into the production of biofuel. The application of mutation techniques has generated a vast amount of genetic variability and is playing a significant role in plant breeding and genetics. 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. Food security will worsen even further because a newer green revolution is required to solve the problem of food insecurity in the decades to come. Induced mutation breeding will therefore continue to play a significant role in improving global 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 additional revenues. 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. Elite mutant plant types have been released as a result of the widespread usage of induced mutations in plant breeding programmes. These mutants are important for creating crops that have higher yields and features that contribute to yield, higher quality and longer shelf lives, increased stress tolerance, and lower agronomic input requirements. Since the discovery of T-DNA insertional mutagenesis, our understanding of plant physiology, biochemistry, and development has rapidly expanded. Auxin transport, inhibition, uptake, and signal transduction have all been linked to auxin mutants like aux1, pid, mp, and lop1. The finding of mutants with high cytokinin levels (amp1), photomorphogenic mutants (det1, cop), cytokinin-resistant mutants, and cell division mutants clarified the mechanism of cytokinin activity. Cytokinin mutants in Arabidopsis thaliana, including ckr1, ein2, cry1, stp1, and zea3, were discovered by Schmulling et al. in 1997. These mutants have clarified the function of genes regulated by cytokinin in a variety of biological processes, including cell division, photosynthesis, chloroplast formation, resistance to illness, and nutrition metabolism.

By screening dwarf le mutants of pea and dwarf mutants of maize, Chandler and Robertson (1999) clarified the mechanism of action of the growth hormone gibberellin. A number of dwarf mutants, such Rht3 in wheat and d8 in maize, are GA deficient and do not react to administered GA3. These dwarf mutants have made a substantial contribution to the development of cultivars that are highly fertiliser-responsive and lodging-resistant. Numerous ABA-deficient mutants have been identified, including ethylene response mutants and aba1 and aba2 in Arabidopsis and N. plumbagin folia, respectively. These mutants are extremely useful and have a significant role in prolonging fruit shelf life, prolonging floral life, and delaying senescence, as evidenced by their transfer to tomato and petunia.

There are a number of homeotic mutants in Petunia, Antirrhinum, and Arabidopsis that have malformed blooms. Understanding patterns of

floral development has greatly benefited from the isolation of these mutants. Through insertional mutagenesis, homeotic mutants for leafy cotyledons lec have been created that are deficient in the development of embryos that remain green. Understanding the apomixes depends critically on the mutations that control seed development, such as the Fis mutant. Crop plants' developmental patterns have a significant impact on yield and attributes linked to yield. In the near future, plant breeding will take on a new dimension with the manipulation of these patterns.

3.3 Future Prospectus

Since induced mutagenesis is becoming more and more important in plant molecular biology as a method to find and isolate genes and explore their structure and function, interest in mutation research has revived recently. Future crop development initiatives will undoubtedly be greatly impacted by this research. In the near future, genetic engineers will use mutation in conjunction with new technologies as tools for plant breeders. Despite this, the majority of the cultivars that have been made public to date have been created by a combination of direct selection and mutation. The use of mutant breeding for crop improvement has taken on a new and broad paradigm in the modern period because to advances in molecular technologies and in vitro culture.

Because of its high frequency and wide range, heavy ion beam irradiation has become a popular and efficient method of inducing mutation in a wide variety of plant species. Through the production of increased resistance traits and higher quality, in vitro mutagenesis has improved crop yield and germplasm innovation in recent years. A limited number of tissues and Calli may be treated to mutagenesis in in vitro culture techniques in order to improve crop species. Nowadays, in vitro mutagenesis is not widely used; very few plants, including sugarcane and bananas, have been successfully regenerated using this method. However, many plants that are propagated from seeds, including barley, wheat, rice, and maize, may now be grown again from cell suspension cultures. It would also be crucial to create in vitro cell selection methods for disease resistance in the future. To create genotypes with desired features, it may be possible to coordinate the latest techniques of anther and microspore cultivation, cell suspension, irradiation of haploid cells, chromosome doubling, and regeneration of doubled haploid plants. The created mutation has additionally shown use in the creation of genetic maps that will support marker-assisted molecular plant breeding in the future. Recently,

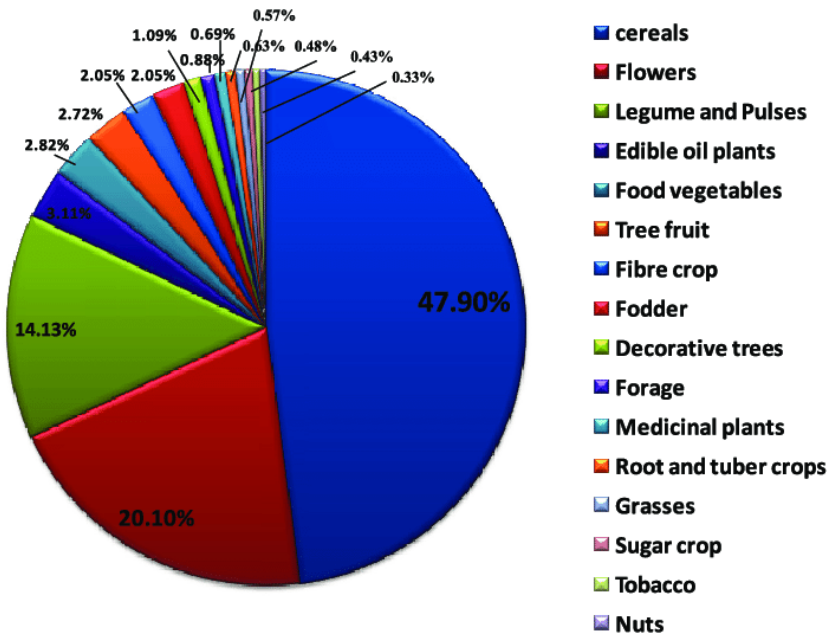

Fig. 2. The Pie chart representing distribution of officially released mutants from different plant categories from all over world. Data collected from MVD 2021
(https://mvd.iaea.org/#!Search) accessed on Jan 20, 2021

there has been a growing interest in mutation breeding as a useful technique for crop development. Plant production and quality could be rapidly improved by using mutation directly in the creation of molecular maps in structural and functional genomics.

4. CONCLUSIONS

Induced mutations have been a significant factor in plant breeding for fifty years, helping both rich and emerging nations produce more food. In addition to using cutting-edge labs and contemporary genomic technologies for mutation induction and discovery, traditional mutation breeding is still employed for the good of communities. Mutation breeding has shown adaptable, practical, and ready for application on any crop provided that the aims and selection criteria are clearly defined. There are currently 3211 registered mutant varieties across more than 170 different plant species.

Since conventional breeding methods have been used for a long time to narrow down genetic variability, induced mutagenesis is one of the most important strategies for increasing genetic variation and diversity in crops in order to get around the bottleneck conditions. Although induced mutagenesis is almost seven decades old, it has been shown to be effective in unlocking plant genetic resources' potential and providing plant breeders with the raw materials needed to create the desired smart crop varieties. Crop varieties produced by utilising mutation breeding are greatly improving livelihoods and contributing to global food and nutritional security.

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Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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