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A Peer Review on The Development of Agronomic Traits Through Physical and Chemical Mutagens

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Abstract

The initial step in mutational breeding is to recognize or identify the appropriate varieties containing the suitable genes which donot exist in nature. In nature, the existing varieties which are found to undergo variation are as a result of mutation. This review paper is mainly focussed on the several types of mutagens and improvement of common tropical and temperate region crops through induced mutationespeciallyphysical and chemical mutagens. The crops include the commonly existing plants which provide higher yield with abundant nutritional value, or applied as a remedy for ailments or diseases. An assessment has been conducted on several types of physical as well as chemical mutagens on tissue culturedexplants to have a peer insight into the screened genotypes providing better agronomic behavior.Different detection techniques like RAPD, cell viability assays, flame photometryfor identifying mutation in the genotypes have been integrated in this study with an objective to aid a researcher in identifyingthe correct approach of mutational breeding for further crop improvement studies.

Introduction

Over several decades, agricultural biotechnology has owned the platform of economic development worldwide.The different agronomic traits prevalent in nature has been subsequently modified through plant breeding techniques by researchers to achieve the best varieties. Plant breeding techniques is the other name for spontaneous mutation. With the advent of Mendelian genetics followed by hybridization technology, cross breeding techniques were adapted in order to obtain the progeny superior to the parental genotypes. Later in 1920s and 1930s, the work of Lewis john Toddler on application of X-Rays laid the footprints of mutational breeding. Hence, it was deciphered that genetics is the basis of both spontaneous and induced mutations leading to mutational breeding for gaining better traits in plants whereas mutation is the basis of variation [3].Spontaneous mutation occurs by chance and thus it becomes a tedious task to procure developed beneficial traits in plants [4]. However, through induced mutations, desired improved

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characteristics can be introduced and new cultivars can be obtained. The most common examples are edible banana or seedless grapes, many roots and tuber crops,development of new colours in ornamental plants[5,6,7,8].Our study is focussed on the most commonly available plants in nature which provide a high nutritional value or are applied for therapeutic applications and the effect of study of physical mutatgens (radiation) and chemical mutagens (Ethyl methyl sulphonate, Colchicine, sodium azide). Different techniques for detection of chromosomal rearrangements are discussed to have a clear analysis of the mutagenesis study to be carried out in future research investigations.

Mutational Breeding through mutation

Mutagenesis is the phenomenon through which sudden heritable changes occur in plants which is not a result of recombination or genetic assortment but induced through physical and chemical mutagens [9]. A mutagen is likely to be chemical, physical or biological that alters the hereditary material such as DNA, of an individual. There are different types of mutagens such as physical mutagen, chemical mutagen and biological mutagens. In most of the cases mutagen causes cancer where it is believed that mutagens behave as carcinogens. All of them have unique characteristics mutational signature. Mutagens cause changes in the genetic material such as DNA and therefore also called genotoxic. They usually affect the transcription and replication of the DNA, which lead to the cell death. The consequence ofmutations may also be loss of function for a specific type of gene. Different mutagen act on the DNA in different way. It may also result in causing chromosomal breakages and also chromosomal rearrangement and also chromosomal instability. In some cases, mutagens also modify the DNA sequence. The first mutagens identified is carcinogens, and its shown to be linked to cancer. The exposure to radiation and cancer is observed in 1902, just in six years after the discovery of X-rays by Wilhelm Rontgen and radioactivity by Henri Becquerel. The first fungi mutants were created by Georgii Nadson and German Filippov under the ionizing radiation in 1925 [10].Mutagens also alter the DNA sequence of an organism which include substitution, insertions and deletion of one or more nucleotide bases in DNA sequences. In most of the cases, many are silent mutations or non-functional sequencescausing no visible effects at all, but in some cases they occur in non-coding portions

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Types of Mutagens

There are mainlythree different type of mutagens mainly a physical mutagen, chemical mutagenic agent or biological mutagen. They may cause the direct damage to the DNA, and then result in replication error. Mutagenesis is described as a treatment of biological material to a mutagen. The physical or chemical type of mutagens is usually used in the plant breeding programs to artificially generate genetic variation in order to development of new varieties as a result of which to increased yield, earliness and also resistance to the disease and also reduced

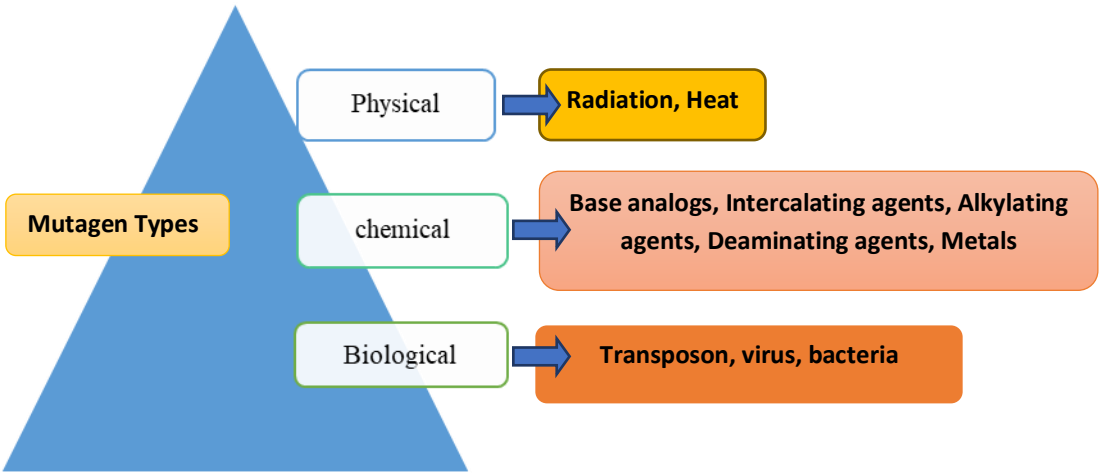

Figure 1 Types of mutagens affecting agronomic traits

plant height. The different types of mutagens are shown pictorially in Figure 1.

Physical mutagens

X-rays and UV lights are physical mutagens. UV light is considered as a non-ionizing radiation and the most commonly used ionizing radiational agents such as x-rays, gamma rays and cosmic rays ionizes the cellular water content and release hydroxyl free radicals (OH). One of the most important aspects of X-rays is that the higher dose of x-rays is fatal for an organism. In work reported previously[11], the genetic control of ionizing-induced mutagenesis in *Saccharomyces cerevisiae* was investigated by measuring the reversion frequency of cycl-9, it is a ochre allele of the structural gene for iso-1-cytochrome c [12] in a variety of radiation strains.Effect of physical mutagen on plant is shown pictorially in Figure 2.

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Apart from the above-mentioned physical mutagens, the common examples of physical mutagens are beta particles and alpha particles, protons, ion beam, neutrons [as reviewed from Table 2 of references 13,14].

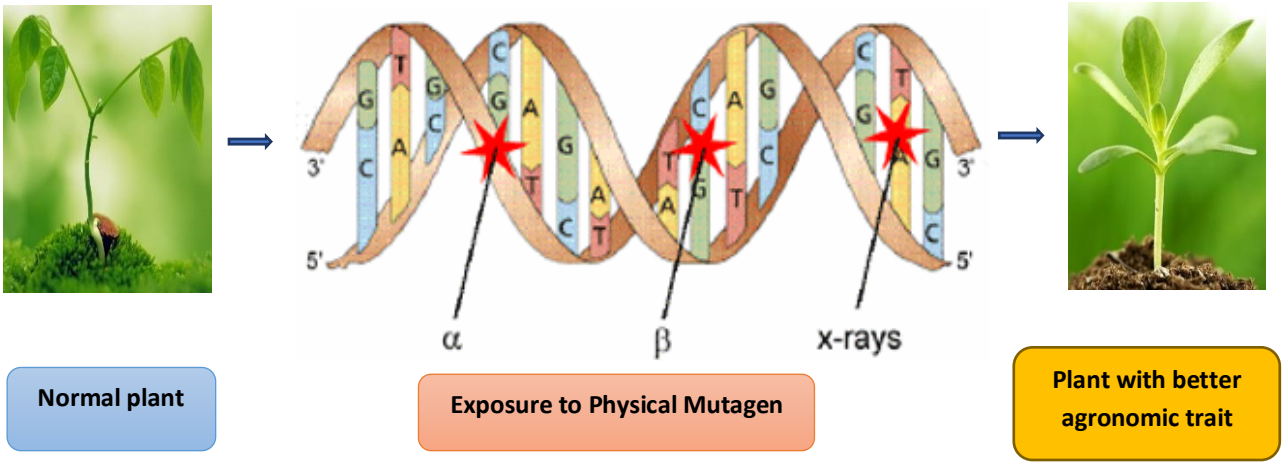

Figure 2Effect of physical mutagen on plant

Chemical mutagens

Most chemical mutagens are azides and alkylating agents. The most popular chemical mutagensis ethyl methanesulfonate. Alkylating agents are very reactive, even with the water. Alkylation of DNA leads to unstable trimesters, which release the alkyl group and interface with DNA replication.The three most common forms available are intercalating agent, base analogue and reacting chemicals.

Intercalating agent has thepotential to interpolate between two base pairs in double stranded DNA helix altering the DNA structure at that definite location.Examples are acridine orange, ethidium bromide, etc.the effect of chemical mutagen is pictorially shown in Figure 2.

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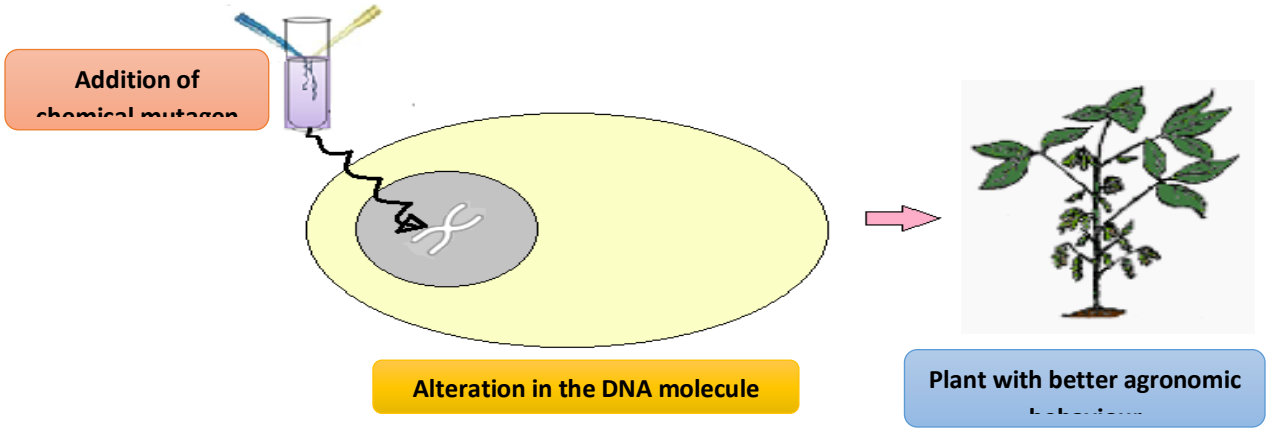

Figure 3 Effect of chemical mutagen on plants

In base analogues, the chemicals with morphological similarity to normal nitrogen bases are substituted at those specific locations instead of the latter. The most common example is 2-aminopurine, an analogue to adenine,5-bromourcail, an equivalent to the nitrogen base thymine. Reacting chemicals modify or change the DNA causing mutation by reacting with the nitrogenous base that is bound to the nucleic acid sequence. For instance, nitrous acids bind to bases for modifying purine with the removal of amino group.

Common examples of chemical mutagens are alkylating agents, azides, hydroxylamine, antibiotics, nitrous acid, acridine orange, and base analogues [as reviewed from Table 2 of references 14,15].

Biological mutagens

Insertion sequence (IS) and transposons elements are biological mutagens. The common examples are bacteriophage MU, mutator gene. These are referred to as the jumping genes which move from one location of the gene to the other and result in mutation. Although similar in behavior to insertional elements, transposons are larger in size compared to IS. Transposons possess unique properties executed by transposase gene and also have inverted repeat 9IR) sequences.

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Recent advancements in study of mutational behavior by physical mutagen (Radiation)

Bracken fern *Pteridium aquilinum v. arachnoideum* is consumed as a diet in the temperate and subtropical region specially, Japan and Brazil. Mann-Whitney U-test was conducted to study the breakage or chromosomal aberrations or re-arrangements in the consumers and non-consumers group [16] as a result of exposure to physical mutagen, radiation. No correlation was found in human alcohol consumption, smoking or gender, variation was speculated only in brecken fern.

Gamma radiation study was explored through the RAPD analysis in sunflower seeds where the magnetic field (MF) was altered by exposure to radiation. MF energy has the capability to change the reactivity of the radicals generated by radiation. Previous studies have shown the effect of carcinogenesis on magnetic field but this research revealed a new horizon to the study of MF with the radiation of selected mutant [17].

Seed of black gram commonly known as urdbean was investigated by researchers for the study of physical mutagen like gamma radiations, EMS and DES. The germination percentage was observed in M1 generation with 60Kr gamma radiation and a linear relationship was observed in higher doses of radiation. It was inferred that the gamma rays have a massive effect on chromosomal morphology, anatomy and physiology of the plant [18].

Physical mutagen such as gamma radiation and DES have also been studied on two varieties of khesari mainly DL-250 and the PLK-750.Dried seeds were exposed to 15Kr, 35Kr, 50Kr of gamma radiations along-with different doses of DES (0.5% for var. DL-250 and 0.25% for var. PLK-750).Pollen sterility showed a dose dependent growth on gamma radiations while survival percentage showed a subsequent decrease. It was observed that PLK-750 trait showed more sensitivity compared to DL-25 [19]. Studies have also been done on tomato (*Lycopersicon esculentum*Mill. Var. Co-2) to study the effect gamma rays and NMU on seed germination and seedling survival after 10,20,30,40Kr. The survival percentage decreased with an increase of the effect of gamma rays.

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Recent achievements in development of agronomic traits by chemical mutagen(Ethyl methyl sulphonate, Sodium azide, Colchicine)

The following chemical mutagens (ethyl methane sulphonate (EMS), diepoxybutane-derived (DEB), sodium azide) and physical mutagens (Gamma rays, X-rays and fast neutrons) are most profoundly found to modify the different functional traits in rice and other agronomic crops. In *Oryza sativa*(430mutant varieties) the effective use of these chemical and physical mutagenshave been investigated. The experiment was carried on *Oryza sativa* L. sp. indica cv. MR219 with exposure to the chemical mutagen EMS at increasing concentrations ranging from 0.25%, 0.50%, 0.75%, 1.25%, 1.5% to 2% and determined by MI generation. With subsequent increase in dose of EMS, a gradual decrease in root germination and length, height of the seedling was was observed as compared to non-treatment control.

Research studies have revealed the effect of ethyl methane sulfonate (EMS) and N-nitrose-N'- ethyl urea (ENU) mutagenic on Plovdiv (normal bean). Seven days leaf petiole explant was cultivated on MS mediumcontaining1 mM BAP with increasing concentrations of EMS and ENU treatment at different time intervals. The study concluded that ENU showed a stronger effect than EMS on the callus growth of Plovdiv[20]In transgenic potato tubers, Or genes originally obtained from orange cauliflower mutant facilitated the carotenoid pathway. Moreover, it increased the levels of three additional metabolite intermediates of phytoene, phytofluene, and z-carotene[21].

Leaf explants treated with Ethyl methyl sulfonate (EMS) was grown in vitro on MS medium fortified with Benzyl-adenine (BA) and 1-Naphtalene acetic acid (NAA).A total of 24 mutants comprising of different traits such as dwarfism, leaf and flower morphology were studied at the molecular level. Significant variation was observed in different parts of the plant. RAPD analysis using 33 primers was conducted where approximately 50% polymorphism was exhibited [22].

Chlorophyll mutations is a progressive study to investigate the different physiological and biochemical pathways occurring in plants and to investigate the effect of mutagens on the genetic elements at specific loci [23]. The results are obtained in the M2 generation and provides important conclusive inference [24]. This an informative study which provides a broadened view of the efficacy and effectiveness of different mutagens on the variability study of the genetic

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constituent of plants. Investigations have been done on the horse-gram variety by Shirshat and coworkers to study the efficacy of several physical mutagens such as EMS, NEU and SA [25].

Salim Khan and co-workers investigated the consequence of chemical mutagens on plants [26]. Sodium Azide, a chemical mutagen is considered for inducing mutation leading to developed agronomic traits in plants. The chemical was found to decrease the cellular calmodulin level, and its effect was visible in mitotic index acting as a calcium ion provider in signal transduction and cell division. It assists in blocking of secretion and accumulation of Camp. The effect of Sodium azide has been observed in tomato andin root length, seedling height and yield per plant. Moreover, sodium azide is found to affect the physiology and decrease cyanide resistant respiration in tobacco callus[26].

Production variations in tomato (Var. roman VF) was investigated through chemical mutagens such as colchicine treatment and para dichlorobenzene. Increase in leaf length and height with subsequent decrease in the height of the seedling, branch number and percentage of germination was observed by colchicine treatment while in para-dichlorobenzene, reduction in size of the abovementioned parameters was not revealed when treated at a higher concentration. However, a lower value ranging from 1mM to 4mM of the chemical mutagens were studied to be effective in inducing mutation to obtain variability [27].

Polyploidation is generally applied for growing pineapple (*Ananas comosus* (L. Merr) callus using varying concentrations of colchicine ranging from 0%, .01%, 0.05%, 0.1%. Group randomized Design (GRD) was used to measure the response and thickness of grown callus at different concentrations. 0.1% was seen to be the most effective in inducing pineapple explants callus.

Spinacia oleracea, an important green leafy vegetable has immense nutritional and therapeutic use. It is considered as a perishable vegetable since the leaves after detachment from the plant are dehydrated due to excessive respiration and transpiration loss. Several attempts were taken by crushing the leaves to a powdered form in a cabinet dryer with a gradual increase of isothermal temperatures from 50 to 80°C at intervals of 10°C to increase the shelf-stability of the plant. Physical and optical parameters were evaluated depending on the factors such as dehydration temperature, types of treatment and particle size. 60°C for unbalanced and 70°C for balanced were found to be appropriate for green leafy powder [28].

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The genetic variability of hexaploidy oat crop was manifested by the application of physical or chemical mutagen. It was observed to be dose dependent and the decrease was according to the increase of the subsequent increase of mutagen [29].

Enhanced metabolic traits are achieved through induced mutagenesis which is listed in the following Table 1.

Enhanced characteristics	**Crop**	**Research publications**
Quality of oil	Soyabean canola, sunflower	[30-32],[33], [34]
Nutritional enrichment of protein	Maize, soyabean	[35], [36]
Enriched amylose content consumed by diabetes individuals	Cassava	[37]
Obovate leaf pod with modified traits	groundnut	[38]
Oilseed meals for poultry feed which is low in phytic acid	soybean	[39]
High resistant starch consumed by diabetes patients	rice	[40]

Techniques used for analysis of mutagenesis

Presence or absence of specific region of DNA is determined through Polymerase Chain Reaction (PCR). Apart from this, various techniques are available to determine the mutagenic sequence in DNA such as protein truncation test, southern hybridization, amplified fragment length polymorphism (AFLP), nucleotide sequencing etc [26].

DNA polymorphism study was carried out by RAPD analysis on Oil palm, a high yielding potential plant with high storage of lipids. Based on similar morphological features, the common ones dura(D), pisifera(P) and tenera(T) were investigated. 185 bands (approximately 300 to 1500 base pairs) were studied with 28 primers and a similarity index was observed among the three varieties with 750 basepairs whereas dura and tenera showed more matches [41].

Wheat grass, a commonly used plant is widely used since ancient times for the cure of different ailments and also for its high nutritional value. Various forms of the plant is available but the proper composition of minerals present required for formulation is still in vague. Flame

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photometry technique was applied for determination of minerals required for the formulation procedure. It is a very simple, robust and inexpensive technique which is also less time consuming[42].

Mitragyna speciosa korth (Ketum), a traditional medicinal plant is widely utilized as it provides energy for increased work efficiency, cure fever, cough and cold, diarrhea. The mutagenic and anti-mutagenic testing was studied through phytochemical screening as well as genotoxic and cytotoxic testing was done in order to validate its efficacy as a traditional medicine. Two strains of *Salmonella typhimurium*, TA 98 and TA100, histidine requiring mutants were studied for studying mutagenesis against a liver metabolizing enzyme through plate incorporation method. Antimutagenicity experiment was conducted by a modified plate incorporated procedure to explore the effects of *M. speciosa* extracts on 2-NF, NaN3 and 2-AA induced mutagenicity. The incubation rate of mutagenicity was calculated by the equation

Inhibiton rate (%) = 1- (T / M) X 100%

where M is the number of revertants per plate in positive control and T is the number of revertants per plate in the presence of mutagens. ANOVA test was used to analyze the result. The absence of mutagenic activity was visible in frame shift (TA 98) and base pair (TA 100) in presence of S9 mix, it was observed that the highest number of mean plate was TA 98 strain at the dose of 12.5mg/ml. But in the presence of S9 mix, all concentration of *M. speciosa,* induced strong antimutagenic activity[43].

Protoapigenone application proved that DNA damage was responsible for killing of lung cancer cells [44]. Protoapigenone is obtained from lady fern plant *Thelypteris torresiana.* In H1299 (human lung adenocarcinoma) and NB4 (human acute promyelocytic leukemia), G1 population was monitored through flow cytometry and the cell viability was checked. Comet assay did not show tailing whereas tailing was observed in proptoapigenone treated cells.

Conclusions:

Different physical mutagens like radiation effect on plants and chemical mutagens like EMS, sodium azide and colchicine have been studied on various tissue cultured medicinal explants as well as the tropical crops. Genetic traits have been induced through the mutagenesis studies which have produced improved varieties with high nutritional as well as medicinal value

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according to their potential use. Different methods for studying DNA polymorphisms such as RAPD, simple PCR techniques, cell viability assays and analysis of cells through flow cytometry, flame photometry studies have been identified. This paved the way for further scope in research and development of plants with better agronomic importance.

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Disclosure statement

There is no conflict of interest by the authors.

References

[1] F.J.Novak, H. Brunner. Plant breeding: induced mutation technology for crop improvement. IAEA Bull.vol,4,pp.25-33, 1992.

[2] Q.Y.Shu, B.P.Forster,H. Nakagawa. Principles and applications of plant mutation breeding. In: QY Shu, Forster BP, Nakagawa H, editors. Plant mutation breeding and biotechnology. Wallingford: CABI; pp. 301-325, 2012.

[3] M.C.Kharkwal. A brief history of plant mutagenesis. In: Shu QY, Forster BP, Nakagawa H, editors. Plant mutation breeding and biotechnology. Wallingford: CABI;. pp. 21-30, 2012.

[4] W.E. Lonnig. Mutation breeding, evolution, and the law of € recurrent variation. Recent Res Dev Genetics Breeding. vol,2, pp.45-70,2005.

[5] B.S.Ahloowalia,M. Maluszynski. Induced mutations: a new paradigm in plant breeding. Euphytica. vol,118 (2) pp.167-73,2001.

[6] C. Broertjes,A.M. Van Harten. Applied mutation breeding for vegetatively propagated crops. Vol.12, Developments in crop science. Amsterdam: Elsevier Publ, pp. 197-204, 1988.

ISSN: 0971-1260
Vol-22-Issue-17-September-2019

[7] M.Chai,Y.W. Ho,K.W. Liew, et al. Biotechnology and in vitro mutagenesis for banana improvement. In: Jain SM, Swennen R, editors. Banana improvement: cellular, molecular biology, and induced mutations. Enfield (NH): Science Publishers, Inc. pp. 59-77,2004.

[8] S. Predieri. Mutation induction and tissue culture in improving fruits. Plant Cell Tissue Organ Culture.vol,64 (23),pp.185-210,2000.

[9] R. Roychowdhury,J Tah. Mutagenesis: a potential approach for crop improvement. In:Hakeem KR, Ahmad P, Ozturk M, editors. Crop improvement: new approaches and modern techniques. New York (NY): Springer; pp. 149-187, 2013.

[10] Russian Trail in the Discovery of DNA structure. Science, August,2018

[11] R.H. McKee, C.W. Lawrence. Genetic analysis of gamma-ray mutagenesis in yeast.I. Reversion in radiation-sensitive strains. Genetics, vol, 93(2), pp. 361-373, 1979.

[12] M. Stewart, C. Murphy, J.W. Fristrom. The recovery and preliminary characterization of X chromosome mutants affecting imaginal discs of Drosophila melanogaster. Dev. Biol.27, pp.71-83,1972.

[13] Wani MR, Kozgar MI, Tomlekova N, et al. Mutation breeding: a novel technique for genetic improvement of pulse crops particularly Chickpea (Cicer arietinum L.). In: Parvaiz A, Wani MR,Azooz MM, Lam-son PT, editors. Improvement of crops in the era of climatic changes. New York (NY): Springer; 2014. p. 217-248.

[14] Mba C, Afza R, Shu QY. Mutagenic radiations: X-rays, ionizing particles and ultraviolet. In: Shu QY, Forster BP, Nakagawa H, editors. Plant mutation breeding and biotechnology. Wallingford: CABI; 2012. p. 8390.

[15] Wani MR, Kozgar MI, Tomlekova N, et al. Mutation breeding: a novel technique for genetic improvement of pulse crops particularly Chickpea (Cicer arietinum L.). In: Parvaiz A, Wani MR,

[16] R.C. Recouso, R.C. Stocco dos Santos, R. Freitas, A. Freitas. Clastogenic effect of bracken fern (Pteridium aquilinum v. arachnoideum) diet in peripheral lymphocytes of human consumers: Preliminary data veterinary and comparative oncology, vol, 1(1), pp. 22-9, April 2003.

[17]Gunhan Erdem, employment of RAPD technique to assess the genetic stability of *Helianthus annuus* treated with different mutagenic agents, Sevil Oldacay, vol 5, pp. 277-281, 2004.

ISSN: 0971-1260
Vol-22-Issue-17-September-2019

[18] C. Thilagavathi. Influence of physical and chemical mutagen on quantitative characters of vigna mungo (L. Hepper), L. Mullainathan, vol 1(1), pp. 06-08, 2011.

[19] Sushil kumar. Effect of physical and chemical mutagen and their interactions on germination, growth, fertility and yield in two cultivars of khesari, vol, 11(2), pp. 147-151, 1998.

[20] M.R. Veltcheva, D.L. Svetleva.In vitro regeneration of Phaseolus vulgaris L. via organogenesis from petiole explants, Journal of Central European agriculture, vol, 6, pp. 53-58, 2005.

[21] A. B. Lopez, J.V. Eck, B.J. Conlin, D. J. Paolillo, J. O. Neill. Effect of the cauliflower Or transgene on carotenoid accumulation and chromoplast formation in transgenic potato tubers, *Journal of Experimental Botany*, vol, 59, pp. 213–223, February 2008.

[22] M. Behera, J. Panigrahi, R. R. Mishra, S. P. Rath. Analysis of EMS induced invitro mutants of Asteracantha longifolia(L.) Nees using using RAPD markers, vol.11, pp. 39-47 , 2012.

[23] Sato, M., Gaul, H. Effect of ethyl methane sulphonate on the fertility of barley. Radiation botany, vol, 7, pp. 7-15, 1967.

[24] D.L. Svetleva Induction of chlorophyll mutants in common bean under the action of chemical mutagens ENU and EMS. J. Central Euro. Agri.vol,5(2), pp. 85-90, 2004.

[25] R. K. Shirshat, M. N. Mohrir. M. A. Kare, A.S. Bhuktar. Induced mutations in Horsegram: mutagenic efficiency and effectiveness. Recent Research in Science and Technology, vol 2(7), pp. 20-23, 2010.

[26] S. Khan, Sodium azide: a chemical mutagen for enhancement of agronomic traits of crop plants, Fahad Al-Qurainy, vol, 21, 1-21, 2009

[27] M.A. Adelanwa, M.L.Habeeb, E.B. Adelanwa. Morphological studies of the effect of colchicine and paradichlorobenzene on tomato (*Lycopersicon esculentum*).Journal ofEnvironmental Issuesand Agriculture in Developing Countries, vol. 3, pp. 122-127, 2 August 2011.

[28] A. Anu, K. Prasad. Studies on Spinach Powder as Affected by Dehydration Temperature and Process of Blanching, International Journal of Agriculture and Food Science Technology, vol, 4, pp. 309-316, 2013.

ISSN: 0971-1260
Vol-22-Issue-17-September-2019

[29] Jefferson Luis Meirelles Coimbra, genetic variability induced by chemical and physical mutagenic agents in oat genotype, Fernando Iraja Felix de Carvalho, vol 4, 48-56, 2004

[30]W.R. Fehr, E.G. Hammond. U.S. Patent No. 6,133,509. Washington (DC): U.S. Patent and Trademark Office; 2000.

[31] E.G. Hammond, W.R. Fehr. Registration of A5 germplasm line of soybean. Crop Sci. vol, 23, pp. 192-193, 1983.

[32] A. Patil , S.P. Taware, V.S. Rao . Development of a high oleic soybean mutant and its stability across the environments. In: Shu QY, editor. Induced plant mutations in the genomics era. Proceedings of an International Joint FAO/IAEA Symposium. Rome: Food and Agriculture Organization of the United Nations; pp. 200-202, 2009.

[33] S.P. Landge ,Y.Y. Barve , R.K. Gupta, et al. Development of B. napus canola quality varieties suitable for Indian agro-climatic conditions by induced mutations. In: Shu QY, editor. Induced plant mutations in the genomics era. Proceedings of an International Joint FAO/IAEA Symposium. Rome: Food and Agriculture Organization of the United Nations;pp. 322-326, 2009.

[34] J.M. Fern-andez-Mart-ınez ,B. P-erez-Vich , L. Velasc. Mutation breeding for oil quality improvement in sunflower. In: Shu QY, editor. Induced plant mutations in the genomics era. Proceedings of an International Joint FAO/IAEA Symposium. Rome: Food and Agriculture Organization of the United Nations; pp. 177-181, 2009.

[35] N.B. Tomlekova. Induced mutagenesis for crop improvement in Bulgaria. Plant Mutation Rep. vol, 2(2): pp. 4-27, 2010.

[36] C. Yathaputanon, B. Kumsueb, A. Malipan. et al. Protein content in high-protein soybean mutants in Thailand. In: Shu QY, editor. Induced plant mutations in the genomics era. Proceedings of an International Joint FAO/IAEA Symposium. Rome: Food and Agriculture Organization of the United Nations; 2009. pp. 195-196.

ISSN: 0971-1260
Vol-22-Issue-17-September-2019

[37] Ceballos H, Sanchez T, Denyer K, et al. Induction and identification of a small-granule, high-amylose mutant in cassava (Manihot esculenta Crantz). J Agr Food Chem. 2008;56(16):7215-7222.

[38] Hamid MA, Azad MAK, Howlidar MAR. Development of three varieties of groundnut with improved quantitative and qualitative traits through induced mutation. Plant Mutation Rep. 2006;1(2):1417.

[39] Landau-Ellis D, Pantalone VR. Marker-assisted backcrossing to incorporate two low phytate alleles into the Tennessee soybean cultivar 5601T. In: Shu QY, editor. Induced plant mutations in the genomics era. Proceedings of an International Joint FAO/IAEA Symposium. Rome: Food and Agriculture Organization of the United Nations; 2009. p. 316318

[40] Amano E. Use of induced mutants in rice breeding in Japan. Plant Mutation Rep. 2006;1(1):2124.

[41] D K Sathish, RAPD markers for identifying oil palm (Elaeis guineensis jacq.) parental varities (dura & pisifera) and the hybrid tenera, C Mohankumar, vol 6, pp. 354-358, july 2007.

[42]K. Shah, T. Desai. Formulation and evaluation of wheat grass topical gel, Pharma Science Monitor, vol,3, pp. 3010-3017, 2012.

[43] R. Ghazali,R. Abdullah, N. Amli, N. Fadilah Rajab.Mutagenic and antimutagenic activities of *Mitragyna speciosa* Korth extract using Ames test, Journal of Medicinal plant Research, vol 5(8), pp.1345-1348, 18 April, 2011.

[44] Chien-Chih Chiu, fern plant derived protoapigenone leads to DNA damage, apoptosis and G2/M arrest in lung cancer cell line H1299, Hsueh-Wei Chang, vol 28, pp. 501-506, 2009