

## GAMMA IRRADIATION I: EFFECT ON GERMINATION AND GENERAL GROWTH CHARACTERISTICS OF PLANTS—A REVIEW

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### Abstract

Qualitative and quantitative improvement of economically important plant species has remained a prime priority of mankind for decades. These improvements are generally correlated with successes in germination and the growth attributes of plants which are controlled by genes. To attain the desirable growth characters in plants of interest, selective screening is carried out in their predecessors followed by successive evaluation, which imposes the cost of time and labor. In order to manage time and labor and to obtain high-quality progenies, gamma irradiation may prove a suitable alternative method to selective screening for bringing mutational changes in plants in short period of time. Exposure of plants' propagating organs to gamma irradiation can either result in genomic damages corresponding to growth abnormalities or gene reshuffling after DNA repair with consequent healthy results. This paper reviews up to dated literature on the effect of gamma irradiation on germination and growth attributes of economically important plants.

**Key words:** Mutation, Selective breeding, Genes, Hormesis, Genome modification, Free radicals, ROS.

### Introduction

Plants—both wild and cultivated play a significant role in food production for human and animals and ecosystem consistency. They serve as a potent source of diverse medicines and ornamentation. To obtain maximum benefits from plant resources, random and systemic efforts have been directed for many decades to improve the qualitative and quantitative attributes of economically important plant species. One of the most ancient and target oriented methods in the improvement of plants is plant breeding which employs successive assortment and obsession of predecessor plants for creating progenies with desired traits (Moose & Mumm, 2008). During last few decades several improvements have been made in breeding programs for development of improved varieties of economically important plants and such quests continues for more accomplishments. Nevertheless, classical breeding methods—which only rely on the selection of superior traits and their fixation in descendants—offer labor and time hurdles besides several other challenges associated with agriculture and consumers (Collard & Mackill, 2008). Thus, to direct the prevailing issues with classical breeding methods, alternative methods based on reasonable cost, time and labor management principles for improvement of plant resources are critically necessary.

Mutation and molecular breeding, which make use of different mutagenic agents and molecular tools in traditional breeding, offer sound and attractive choice for plant breeders to raise desired traits in plants (Majeed *et al.*, 2017). Although mutation and molecular breeding of plants can be attained by several available techniques such as marker-assisted selection and quantitative trait loci mapping, the use of mutagenic agents like gamma irradiation seems reasonably economical and easy to practice as compared to other molecular techniques (Thomson *et al.*, 2009; Çelik & Atak, 2017). Moreover, a handful of research during the last few years highlighting the successes of gamma irradiation in the improved genetic

characteristic of plants make the use of radiation technique more attractive. Data suggests that the number of mutant varieties of different plants obtained mostly through gamma-induced mutation exceeds 2200 (Maluszynski, 2001; Ahloowalia *et al.*, 2004). Thus there is an attractive and potential space for radiation-induced mutation research which can boost breeding for the quality of plant species. The aim of this paper is to highlight the effect of gamma irradiation on germination and growth parameters of economically important crops by reviewing relevant literature survey.

### Gamma irradiation: Sources and mechanism of action:

Gamma irradiation along with other high energy rays is a type of electromagnetic waves which owe high penetration capacity into molecules and can bring about ionization of the subject material by removing their electrons (UNSC 2000). Unlike X-rays which are also part of electromagnetic spectrum, gamma rays retain more energy and thus have a greater capacity for penetration into the matter and ionization potentials (Burchfield, 2009). Such radiation generally originates naturally from galaxies and solar radiation as cosmic rays, from the decaying nuclei of radioactive materials which are external and internal earthy in nature and widely include isotopes of radium, potassium, uranium, cobalt, cesium, lead and carbon among many others (Underhill, 1995). Radioactive isotopes of certain atoms possess unsteady nuclei which can undergo decaying process over time resulting in the production of gamma rays (Fairand, 2001). Artificial sources of radiation are manmade gamma rays emitters such as Co-60 or Cs-137 cells which are devised generally for industrial and medical purposes. Besides industrial and medical installation of gamma sources, testing of nuclear weapons and subsequent fallout of their radiation and nuclear power generations in reactors are also some of the important artificial sources contributing to the dissemination of gamma rays (Charles, 2001). Although generally considered as extremely hazardous for the health of the

living organism, the use of gamma rays still has some beneficial application in agriculture, medicine and power generation. Mutation breeding of plants with gamma rays is one of the potentially useful applications in agriculture for desired outcomes.

Exposure of plant materials specifically seeds to gamma irradiation results in mutagenic changes in living cells by several means. Since gamma radiation owes high energy and greater penetrability into exposed cells and tissues, DNA of the subject material may undergo severe alterations. These alterations can either result from direct physical strikes of gamma irradiation on DNA (Fig. 1) or due to production of reactive O<sub>2</sub> species such as hydrogen peroxides, hydroxyl ions and other active atomic oxygen which can further interact with DNA and other cellular components and biomolecules resulting in the ionization, functional changes of proteins and enzymes and overall metabolic activities (Zaka *et al.*, 2004; Majeed *et al.*, 2017).

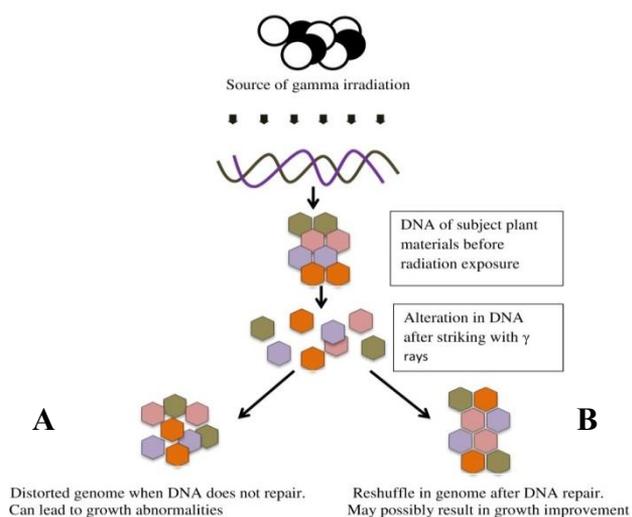


Fig. 1. Changes in DNA of subject plant materials after exposure to gamma irradiation. A- DNA damage is permanent (lethal effects); B- DNA repairs (genes reshuffling)

In particular, ionization in cell environment and direct physical strikes of gamma irradiation can lead to disruption of host DNA which can bring about significant changes in the inherited traits. The disruption of DNA may be permanent which could cause lethal effects or it may be temporary because structural molecules of DNA allow it to repair after damage in most instances following the nucleotides excision restoration mechanism (Ali *et al.*, 2015). Changes in DNA of plant seeds after exposure to radiation can serve a potential source of variation in descendants. Mutant offspring resulted from irradiated seeds can exhibit either abnormality in germination, morphology, and growth or these traits may be positively influenced which generally depend on repair of DNA and rearrangement of genes. Besides genomic modifications, radiation exposure can also cause changes in cell cycle patterns, hormonal balance, metabolic pathways and enzymatic alterations (Melki & Marouani, 2010). Radiation intensity and exposure period of subject seeds are important determinants in creating abnormal or desirable mutants of plants (Majeed *et al.*, 2017).

**Effect on germination of plants:** Germination of seeds is an important preliminary phase during the development, growth, and production of plants which is regulated by several factors ranging from internal cellular contents including genes to external environmental conditions (Koornneef *et al.*, 2002; Miransari & Smith, 2014). Successful germination of seeds overcoming internal and external stresses can result in stable morphological and growth performance of plants. These barriers in germinability of seeds can be influenced in both positive and negative directions by  $\gamma$ - radiation exposure as a result of mutation inductions depending on cellular abnormalities or stimulatory modifications triggered by radiation doses. In several studies, lethal and stimulatory effects of gamma irradiation on germination percentage, emergence, and survival of seedlings of different plant species have been reported (Table 1). Borzouei *et al.*, (2010) treated two wheat genotypes with different doses of gamma irradiation (100-400 Gy) which resulted in delayed germination time but no effects were observed on germination percentage. *Terminalia arjuna*, an important medicinal plant, showed positive responses in terms of germination speed and vigor index at lower doses (25 Gy) of gamma irradiation (Akshatha *et al.*, 2013). Marcu *et al.*, (2013b) documented enhanced germination (%) and germination index of *Lactuca sativa* at 30 Gy. Beyaz *et al.*, (2016) concluded that radiation doses 100 and 150 Gy were effective in stimulating germination of *Lathyrus chrysanthus*. Aref *et al.*, (2016) revealed that 5 Gy had a stimulatory effect on germination of *Datura innoxia* but doses exceeding 5 Gy negatively influenced germinability. Verma *et al.*, (2017) have also reported that germination and seedling survival of *Cuminum cyminum* improved at a lower dose (100 Gy) but declined at higher doses particularly at 500 Gy. In a recent study, Hussain *et al.*, (2017) showed dose range 0.5-5 kR as an effective treatment for enhancing germination of sunflower.

**Growth response of plants to radiation stress:** Plants are living organisms and they respond to growth challenges in a systematic manner. Growth challenges may be indigenous or exogenous in nature. They are supposed to show better growth and vigor if they possess genetic traits capable of coping with environmental stresses and other growth challenges. Several mechanisms govern the growth and developmental phases of plants and their adaptation to imposed challenges which depend on proper mobilization of growth hormones, regulation of cell cycle, activation of concerned enzymes, appropriate functioning of metabolic machinery and timely expression of genes which regulate growth phenomena (Gray, 2004; Santiago *et al.*, 2013; Salehin *et al.*, 2015). Boosting of growth controlling factors and associated mechanisms in plants can lead to desirable improvements in their growth characteristics. Gamma irradiation, particularly at lower doses, has been widely known to promote growth attributes of several economically important plants (Kim *et al.*, 2004). Data presented in Table 2 show effect of different radiation doses on growth characters of plants. Enhanced growth measured as shoot length and diameter and leaf area of *Capsicum annuum* L. was attained at radiation doses 2-4 Gy with respect to control (Kim *et al.*, 2005). Stimulatory responses

of hypocotyl and root growth of *Lactuca sativa* at gamma irradiation doses (2-30 Gy) while reduced shoot and root lengths in *Zea mays* were recorded at 0.1-1 kGy (Marcu *et al.*, 2013a,b). Ilyas & Naz (2014) observed enhanced number of leaves, root and shoot growth of *Curcuma longa* at 60 Gy treatments. Lower radiation dose (5 Gy) significantly increased growth rate of roots and shoots of *Datura innoxia L* (Aref *et al.*, 2016). El-Gazzar *et al.*, (2016) recorded significantly improved growth performance of *Vicia faba* at doses  $\leq 100$  Gy; however, higher doses had negative effects on the plant. Plant height and leaf area were increased when pre-germination radiation treatment (100 Gy) was applied to seeds of *Vigna unguiculata* (Olasupo *et al.*, 2016). On the other hand, Majeed *et al.*, (2016) revealed significant decline in overall growth and yield attributes of pea exposed to gamma irradiation doses 0.8-3.6 kGy. Shoot length, stem diameter, leaf length and width of *Abelmoschus esculentus* were significantly enhanced at radiation doses 400 Gy when compared to non-radiated plants (Asare *et al.*, 2017). Hong *et al.*, (2017) demonstrated that gamma radiation doses in the range between 10 and 15 Gy positively influenced growth traits of common wheat (*Triticum aestivum L.*). Wang *et al.*, (2017) demonstrated that 80 kR radiation doses increased the number of branches and stem diameter of medicinal plant *Sophora davidii*.

Germination and growth response of different plants to gamma irradiation treatment generally depends on the quantity of radiation dose, exposure time and plant species (Marcu *et al.*, 2013b; Majeed *et al.*, 2017). In many studies it has been established that lower doses of radiation often cause germination and growth improvements of plants while higher doses result in growth abnormalities, germination retardation or even death of exposed plants (Kovalchuk *et al.*, 2003; Majeed *et al.*, 2016; Hong *et al.*, 2017). Improvement in germination and growth traits of plants exposed to low doses of gamma irradiation may be attributed to positive mutational effects on genes controlling these traits, rapid DNA repair mechanism and stimulation of hormones and enzymes which are actively involved in germination and growth processes. Lower doses might possibly accelerate cell division in meristematic tissues which could contribute to improved germination and plant growth (Dhakshanamoorthy *et al.*, 2011). Moreover, lower doses may cause an increase in antioxidant potentials and better communications among growth hormones in radiated cells which may lead to enhanced growth (Wi *et al.*, 2007). Growth abnormalities and low germination of plants in response to higher doses of gamma irradiation is widely attributed to negative mutation, ionization of water present in cells and subsequent formation of reactive oxygen species and free radicals which can interact with other cellular molecules potentially imposing negative structural and functional changes in them (Wang *et al.*, 2017). Marcu *et al.*, (2013a) argued that creation of free radicals in cells in response to gamma irradiation can affect proteins, lipids, enzymes and cellular molecules which may cause increased antioxidant responses and alteration cell membrane permeability potentials. These changes can trigger growth abnormalities.

Table 1. Effect of different doses of  $\gamma$ -radiation on germination parameters of different plants

$\gamma$ - radiation doses	Plant species	Germination parameters	Effects	Reference
100-400 Gy	<i>Triticum aestivum L.</i>	Mean germination time; germination (%)	Decrease in mean germination time at higher doses but no effect on germination percentage	Borzouci <i>et al.</i> , (2010)
10-30 Gy	<i>Triticum durum Desf.</i>	Germination speed and germination capacity	No effect	Melki & Marouani (2010)
100-250 Gy	<i>Amaranthus cruentus L</i>	Percent germination	Drastically decreased at 250 Gy	Ayneband & Afsharinafar (2012)
0-200 Gy	<i>Terminalia arjuna</i>	Germination (%), speed, vigor index	Significantly increased at 25 Gy	Akshatha <i>et al.</i> , (2013)
0.1-1 kGy	<i>Zea mays L.</i>	Percent germination, germination index	Decrease with increasing dose	Marcu <i>et al.</i> , (2013a)
2-70 Gy	<i>Lactuca sativa</i>	Percent germination, germination index	Enhanced at 30 Gy	Marcu <i>et al.</i> , (2013b)
200-350 Gy	<i>Zea mays L.</i>	Germination rate and percentage	Germination percentage decreased at all doses	Nepal <i>et al.</i> , (2014)
10-100 Gy	<i>Curcuma longa</i>	Germination rate and percentage	No effect on germination rate	
50-250 Gy	<i>Lathyrus chrysanthus</i>	Germination	No effect on germination (%) but delay in germination rate	Ilyas & Naz (2014)
5-80 Gy	<i>Datura innoxia L</i>	Germination	100-150 Gy; improved germination	Beyaz <i>et al.</i> , (2016)
10-100 Gy	<i>Solanum tuberosum L</i>	Germination	Stimulation at 5 Gy	Aref <i>et al.</i> , (2016)
100-500 Gy	<i>Cuminum cyminum L</i>	Germination, seedling survival	20 Gy stimulated germination	Salomón <i>et al.</i> , (2017)
0.5-5 kR	<i>Helianthus annuus. L</i>	Germination time and percentage	Enhanced germination at 100 Gy but reduced at higher doses	Verma <i>et al.</i> , (2017)
			Improved germination percentage but delayed germination time	Hussain <i>et al.</i> , (2017)

Table 2. Growth response of different plants to exposure of gamma irradiation

$\gamma$ -radiation doses	Plant species	Growth attributes	Effects	Reference
100-400 Gy	<i>Triticum aestivum</i> L.	Root and shoot growth	Improved at 100Gy but decline at higher doses	Borzouei <i>et al.</i> , (2010)
300-500 Gy	<i>Abelmoschus esculentus</i> L.	Shoot height, number of branches and leaves plant <sup>-1</sup>	Significant improvement over control	Hegazi & Hamideldin (2010)
20-80 kR	<i>Lepidium sativum</i> L.	Root, shoot length, leaves and branches plant <sup>-1</sup>	Increase in doses decrease growth parameters	Majeed <i>et al.</i> , (2010)
10-30 Gy	<i>Triticum durum</i> Desf.	Epicotyl and shoot length	18-33% increase at 20 Gy	Melki & Marouani (2010)
50-800 Gy	<i>Oryza sativa</i>	Seedling length, number of tillers	Negative effect on seedling length but no effects on tillers	Harding <i>et al.</i> , (2012)
0.1-1 kGy	<i>Zea mays</i> L.	Shoot and root height	Significantly decrease	Marcu <i>et al.</i> , (2013a)
2-70 Gy	<i>Lactuca sativa</i>	Hypocotyl and root growth	Increased at doses 2-30 but decreased at 70 Gy	Marcu <i>et al.</i> , (2013b)
10-100 Gy	<i>Curcuma longa</i>	Root and shoot length, leaf number and width	Enhanced at 60 Gy	Ilyas & Naz (2014)
5-80 Gy	<i>Datura innoxia</i> L.	Root and shoot growth rate	Enhanced at 5 Gy	Aref <i>et al.</i> , (2016)
20-200 Gy	<i>Vicia faba</i> L.	Morphology and growth	Improved at $\leq$ 100 Gy	El-Gazzar <i>et al.</i> , (2016)
100-500 Gy	<i>Vigna unguiculata</i> L.	Plant height, leaf area	Improved at 100 but declined at higher radiation doses	Olasupo <i>et al.</i> , (2016)
0.5-5 kR	<i>Helianthus annuus</i> . L.	Radicle, plumule growth, root number	Up to 83% improvement in growth compared to control	Hussain <i>et al.</i> , (2017)
400-1000 Gy	<i>Abelmoschus esculentus</i> L.	Shoot length, stem diameter, leaf length and width	Lowest dose (400 Gy) enhanced the growth but higher doses significantly decreased the growth	Asare <i>et al.</i> , (2017)
10-150 Gy	<i>Triticum aestivum</i> L.	Plant growth	Enhanced growth at 10-15 Gy but doses greater than 20 Gy reduced growth attributes	Hong <i>et al.</i> , (2017)

## Conclusion

Gamma irradiation provides a feasible choice to plant breeders for bringing desired traits concerned with better germination and plants' growth thus avoiding high throughputs of time, labor and cost generally related with selective breeding methods. Since these traits are controlled by genes, positive fluctuation in genomic structure of plants of interest by exposure to gamma irradiation can enhance their germination and general growth characters. It is evident from the surveyed literature that in many instances, low doses of gamma irradiation promote germination and growth characters of different plants while higher doses have detrimental influences. Lower doses stimulate the growth of subject plant by either direct genome modifications or regulation of cellular process which include hormonal signaling, enhancing enzymatic efficiency, increasing anti-oxidative potentials, cell membranes modification etc. which could lead efficient cell division, high photosynthesis rate and improved capacity of plants to cope with environmental stresses. Conversely, higher doses cause genomic damage, production of free radicals and reactive oxygen species which influence germination and growth factors in a negative manner resulting in arrested germination, survival and growth abnormalities. Although the optimum dose of gamma irradiation for growth stimulation is difficult to suggest for mutation breeding since different plants respond differentially to applied doses; however, it seems that in most studies radiation doses lower than 100 Gy correspond to enhanced germination and growth performance of many plant species.

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