

Improvement of major ornamental crops through mutation breeding

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The improvement in ornamental crops can be achieved by mutation induction through physical and chemical mutagens at different concentration by using various plant materials, even through the most of the mutations are deleterious and lethal. The gamma irradiations at 1 K rads to 7.5 K rads proved best according to crop response. As the doses of mutagens increase, the quality parameters of plant decrease. So, it has been suggested to use mutagens at lower concentrations (Rose- 5 to 7.5 K rads, Chrysanthemum- 1.5 to 2 K rads, Gladiolus- 1 to 5 K rads, Tuberoses- 0.5 to 1 K rads, Orchid and Carnation- 5 to 10 G rays under *in vitro*) to obtain useful mutants. Further, different cultivars also differed in their response to resist mutations. The changes in flower colour and forms due to mutation can be easily detected from its phenotypic expression as well as from cytological studies. A overall review of research work indicated that mutation breeding is one of the most powerful methods for developing new varieties which is very successful in ornamental crops.

Improvement in flower crops is very important to generate the interest by evolving new forms, new colours and their large scale production. New varieties can be developed through sexual breeding which provides thrilling results but is time consuming while mutation breeding takes about three times less time and even produce an unusual type.

Induced mutation breeding holds promise for effective improvement and it has led to a great burst of flower colour, form, pattern and other variations in different ornamentals.

In general mutation refers to sudden heritable change in the phenotype of an individual. In molecular term, mutation is the permanent and relatively rare change in the number or sequence of nucleotides.

Mutation breeding refers to the genetic improvement of crop plants for various economic characters through the use of induced mutations. Mutations are induced by mutagens. Generally mutagens are classified in two ways *i.e.* physical mutagens and chemical mutagens. Different types of radiations like gamma rays, X-rays, alpha

particles, beta particles, ultra violet rays and fast and thermal neutrons are considered into physical mutagens while chemical mutagens include ethyl methane sulphonate, methyl methane Sulphonate, ethyl ethane sulphonate, ethylene amines, 5-Bromo Uracile, 2-Amino purine, acriflavin, proflavin, nitrous acid, hydroxylamine, sodium azide, etc. Mostly physical mutagens are used for development of new varieties in ornamental crops and among all physical mutagens, gamma radiations are more preferable due to its properties like sparsely ionizing, deeply penetrating and it is non-particulate Kaicker and Vishnuswarup (1992), Shobha *et al.* (2001), Arnold (1998), Diltla (2003), Banerji and Datta (2002), Dhaduk (1992), Misra and Mahesh (1993), Misra and Bajpai (1983), Abraham and Desai (1976), and Gupta *et al.* (1984). The treatment of mutations is applied on seed, seedlings, cuttings, any plant part and also on *in vitro* plantlets. Under *in vitro* condition Shobhana and Rajeevan (2003), Sooch *et al.* (1999) and Singh *et al.* (1999) were obtained good results with gamma rays at low concentrations.

Mutations occur in cells in two ways. Firstly, by alteration in nuclear DNA which is also known as point mutation and it cause addition, deletion, transition and transversion in nucleolus of cell. Thus it changes the structure of nuclear DNA and mutations occurred. In second way, the mutagen makes the change in cytoplasmic DNA that is also called as cytoplasmic mutation. The best example of cytoplasmic mutations is 'Male sterility' induced in some ornamentals crops.

Mutation is cheap and rapid method of developing new varieties as compared to hybridization methods. It is more effective for the improvement of oligogenic characters than polygenic traits. It is the simple, quick and best way when a new character is to be induced in vegetatively propagated crops. An induced mutation takes lesser time for release of new variety. Mutations provide new variety is identical to parent variety except for the character improved.

While under limitations of mutations, most of the mutations are deleterious and undesirable. Also, identification of micro-mutations is usually very difficult

which are more useful to breeder like increase in yield. Since useful mutations are produced at a very low frequency (0.1%), a very large plant population has to be screened to identify and isolate desirable mutants. Mutation breeding has limited scope for the genetic improvement of quantitative or polygenic characters.

Some important characteristics of mutations are as below:

- Mutations may be more or less permanent depend on genotypic characters of plant and its stability to sustain sudden changes occurred in cell.
- Mutations occurred at very low frequency which is only 0.1%.
- Rate of mutation varies from gene to gene and species to species.
- Direction of changes developed by mutations is may be from dominant to recessive or vice-versa.
- Effects of mutations are mostly harmful.
- Mutation occurred on gene and the gene where the mutation affects is known as 'Hot spot'.
- Mutations occurred randomly that means anywhere and on any cell which is not predicted before its occurrence.
- Mutations may be occurred once or repeatedly.
- Most of mutations are lethal which are recognized by its mortal or lethal effect in which maximum plants are died due to the drastic changes in physiological process and chemical reactions. So the rate of survival is very less in almost all plants when the doses exceed their LD₅₀.

Induced mutations are useful in crop improvement in five principal ways:

- Development of improved varieties
- Creation of genetic variability
- Production of haploids
- Induction of cytoplasmic male sterility
- Overcoming self incompatibility

Brief review of research work:

Rose:

The cultivars of rose were treated with different doses of gamma irradiations observed by Kaicker and Vishnuswarup (1992) that when control shown maximum number of sprouts in Christian Dior, Queen Elizabeth and Kiss of Fire which was followed by treatment of 5 Kr in all cultivars. The decreasing trend was observed in number of sprouting as the doses increased. In case of chemical mutagens, the maximum number of sprouts (62) was observed 2% DES in Christian Dior and 32 sprouts in Kiss of Fire at 0.25% EMS treatment while maximum mutants *i.e.* 3 were obtained in 0.1% E. I.

Shobha *et al.* (2001) reported that a mutant of

'Wouburn Gold' in rose which had maximum flavanol (31.20 mg%), anthocyanin (4.12), luecoanthocyanin (0.32) and total carotenoid (0.49) contents that provided tangerine orange petal colour. In general, all mutants shown more flavanol, anthocyanin, luecoanthocyanin and total carotenoid contents then their original cultivars and colour change was also recorded in all mutants.

The colour changed in miniature rose cvs. Putlock, Blue Blood, Mountie and Dark Red Mountie recorded by Arnold *et al.* (1998) from light to dark as gamma rays doses increases. Number of flowers / plant, flower diameter was found to be maximum in 50 G rays treatment which were consequently decreased further. The maximum numbers of point mutations were observed at 200 G rays in Putlock and Blue Blood while at 100 G rays in cvs. Mountie and Dark Red Mountie.

Chrysanthemum:

The significant reduction in plant survival (%), vegetative and floral characters of chrysanthemum in all cultivars at 2.0 K rads of gamma rays treatment over control were observed by Dilt (2003). The somatic colour mutations were observed in Ellen Van Langen, Gulmohar, Shyama and Snow Ball. They also indicated significant reduction in number of flower heads/plant (66.53) and flower head size (6.27 cm) in treated plant material as compared to untreated plants. Among all the cultivars 'Gulmohar' showed better performance.

The phenotypic characters of cv. SUREKHA and its four gamma rays induced mutants were studied by Banerji and Datta (2002) and found that the performance of mutant 'Creamish White' was good for maximum number of characters. The maximum petiole size and number of chloroplast were observed in original cultivar of chrysanthemum 'Surekha' over its four mutants. In 'Yellow' mutant maximum number of branches, leaves, maximum head height, stomata/unit area with significantly maximum width of guard cells and width of pore at 0.1% were recorded than cv. SUREKHA and its other mutants. Mutant of 'yellow Background' shown maximum leaf size, head weight, per cent moisture, length of pore then all others. Significantly maximum height of the plant, number of ray florets per head, head diameter, fertility and size of pollen grains were reported in 'Creamish White' mutant of cv. SUREKHA. Only maximum number of heads/plant, size of florets and size of chloroplast was found to be more in 'Lighter mutant' while chromosomal aberrations percentage recorded significantly higher at 0.1% in 'Yellow mutant', 'Yellow Background' and 'Creamish white mutant' then original. Significantly maximum per cent somatic mutation (45.45) was recorded at 250 G rays.

The all vegetative parameters, per cent survival and flower head number were decreased with increase in gamma irradiations treatment in chrysanthemum cv. SUREKHA. Also showed increase in per cent abnormal plants, per cent abnormal leaves and floral characters for the same cultivar with increase the dose of gamma irradiations.

Gladiolus:

The plant height, spike length, number of florets per spike, number of corms per plant and weight of one corm were significantly maximum at 1.0 K rads gamma rays treatment in gladiolus in all four cultivars viz. *Gladiolus callianthus* var. *murielae*, Christian Jane, Psittacinus Hybrid and Oscar which was indicated by Misra and Mahesh (1993). Significant decrease in number of days required to sprout in 1.0 K rads treatment for cultivar Oscar and at 2.5 K rads for *Gladiolus callianthus* var. *murielae* was also observed. In general, the decreasing trend was observed in all parameters with the increase in doses of gamma irradiations.

Under Ph. D. studies by Dhaduk (1992) suggested that 3 and 5 K rads gamma irradiations showed maximum per cent survival and 1 K rads were found to be better for longevity of spikes for all four cultivars of gladiolus.

The significantly maximum sprouting per cent (99.55) and per cent survival (100 %) were observed by Misra and Bajpai (1983) in gamma rays at 3 K rads and 4 K rads for all varieties of gladiolus viz., Blue Lilac, Jo Wagenaar, Sanc Souci, Snow Princess, Himprabha, Ratna's Butterfly, Picardy, Sylvia and Murielea. Among all varieties 'Sylvia' had shown maximum per cent sprouting (78.5%) and 'Blue Lilac' recorded significantly maximum per cent survival (77.35%) in physical and chemical mutagens. The interaction effect of varieties and treatments was also studied by the scientist and revealed that the minimum days required for sprouting was observed in cv. HIMPRABHA at 3 K rads of gamma irradiations (14 days) and 28 days at 1.00% Ethyle Methane Sulphonate. The mutagenic treatment delayed the sprouting than control. The 100% sprouting was recorded with Blue Lilac and Sylvia at 7 K rads of gamma rays while all the chemical mutagens proved to be sub-lethal to lethal for all varieties. In case of per cent survival (100%), all varieties at 5 K rads proved best but in chemical mutagens only MNH at 0.05% showed 100 % sprouting.

The corms of gladiolus var. 'Spic and Span' treated with 4 hours of continuous radiation from a Cobalt-60 irradiator (treatment was given by placing the plant material at certain distance from the centre of gamma source) by Sheehen and Sagawa (1960) and obtained the

earlier sprouting from 2,784 to 13,040 rads treatments. Abnormalities were noted in the treatments of 7,520 to 33,840 rads as expressed in the forms of leathery, wrinkled and narrow leaves which increased with increased doses of severe leaf deformities. Twin buds and semi-double florets on a few spikes were also observed. Dhara and Bhattacharya (1972) also stated the affectivity of gamma-irradiation on the gladiolus corms. They noted double flowers, stunted spikes and leaf variegation.

Banerji *et al.* (1981) and Banerji and Datta (1987) irradiated dormant corms of *Gladiolus psittacinus* var. Hookeri cultivars Orange and Red with 2.5, 5, 7.5, 10 and 12.5 K rads of gamma rays. Reduction in sprouting, plant height, sprout number, spike length, number of leaves per corm, number of florets per spike, corm size and number of corms and cormels were recorded after irradiation. Morphological abnormalities and chromosomal aberrations like early separation, bridges, fragments and laggards increased with increase in exposure to gamma rays. Days to sprout, emergence of spike and first floret opening delayed in higher doses. Flowering completely ceased at the highest dose. LD₅₀, on survival basis, was found to fall in between 10 to 12.5 Krads. From later experiment, they isolated one mutant at 2.5 Krads treatments.

Irradiation treatment given to the seeds of cv. VISTA BONITA with X-rays by Hubbard (1966) and observed that the normal and faster germination in the same year, whereas in the following season 40-44 per cent of 136 seedlings evenly bloomed (four time more than the control) from salmon to deep blotched pink via various shades with seedlings having florets ranging from 200 to 500 size. Different abnormalities appeared as ruffling of petals, unopening of florets, increased floret parts, bicolouration, short spike, poor texture and attachment of the spike and small cormels. Durst (1973) and Yamato *et al.* (1958) treated seeds to a dose of 6000 R X-rays and observed some mutants which are lighter in colour.

The cormels of eight varieties of gladiolus were exposed to thermal neutrons and X-rays by Jenkins (1961). He reported that even low rate of both types of irradiations reduced the germination of the cormels and height of the plants. The heavier doses (15,000 to 20,000 R) were lethal and none of the cormels germinated in this experiment. Electric shock treatment of an ultra high frequency electric field of 7.7 nm, 40.7 MHz and 30 V has been given on the growing plants and during the formation of vegetative organs (corms) by Sozonova and Syrovatka (1974). They reported growth stimulation of the corms from irradiated plants in the year of treatment. The optimum results were obtained by treating the plant

by 15 minutes. Some of the variants detected in the above experiment have been isolated, multiplied and released as new varieties.

Tuberose:

The maximum survival, leaf number, plant height and number of plant flowered at 0.5 K rads gamma irradiations for both single and double tuberose for large and small size bulbs were observed by Abraham and Desai (1976). They also reported treatment of fast neutrons at 142 rads was best for survival, leaf number, plant height and number of plants flowered in single and double tuberose for both bulb size. The number of leaves, height of plant, total number of flowering plants and total survival percentage of plants were decreasing with increase in doses of both mutagens *viz.* gamma rays and fast neutrons. While comparing the two cultivars on the basis of all the biological end points selected, the double type was more sensitive than the single type, both for gamma rays and fast neutrons treatments.

Shukla and Datta (1993) revealed that leaf length and leaf width was significantly higher at 500 rads and decreased at 0.01% in tuberose cv. SINGLE at 1500 rads of gamma irradiations. In case of cv. DOUBLE, leaf length and leaf width were increased till 1000 rads of gamma rays treatment which gave maximum length and width, afterward it was started to be decreasing. LD₅₀ for sprouting percentage of both types of tuberose was 500 rads and it was decreased with increase in the doses and it may prove lethal at 1500 rads.

Gupta *et al.* (1984) reported that the 'Rajat Rekha', mutants of single tuberose gave maximum leaf length, leaf width, number of spikes/plant and number of flowers/spike as compared to its control while 'Svarna Rekha' which is mutant of double tuberose showed maximum plant height, number of flowers / spike, flower length as well as length of spikes. Thus, it is proved that the mutants of single and double types of tuberose had maximum good vegetative and floral characters over their respective controls.

Orchid:

In vitro studies of *Dendrobium* cv. Sonia-28 x Emma done by Shobhana and Rajeevan (2003) found that ⁶⁰CO irradiations at 10 Gy was good for days to shoot and root formation (94.0) and days taken for planting (293.0). Field evaluation of same seedling revealed that non-irradiation seedling performed better, but mutation observed in irradiated one.

Carnation:

Sooch *et al.* (1999) revealed that 1 K rads gamma irradiations increases the number of shoots (7.17), shoot length (1.38 cm), number of roots (18.30) and root length (1.18 cm) while decreases the days for root initiation (6.90).

Singh *et al.* (1999) observed that 5 Gy ⁶⁰CO dose was found to be significantly higher for different vegetative characters of carnation cv. Espana under *in vitro* conditions. Also recommended 5 Gy gamma radiations were significantly better for all floral characters except colour variation.

Cytological studies for realization of mutation:

Banerji and Datta (2002) concluded that 'Creamish White' mutant of chrysanthemum showed higher values for all cytological characters expect for stomata per unit area and per cent pollen grain fertility. Further, they studied root tip mitosis in chrysanthemum cv. SUREKHA and found that 250 G rays gave better chromosomal aberration for different characters.

Shukla and Datta (1993) revealed that 1500 gamma radiations was found to be effective on chromosomal aberrations of root tip mitosis in single and double tuberose.

Dhaduk (1992) reported that the early separation, Bridges and laggards type of chromosomal aberrations were maximum during root tips mitosis in vM₁ gladiolus in all four cultivars studied.

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