**Gamma rays induced chromosomal aberrations in tomato (Solanum lycopersicum L.)**

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**ABSTRACT:** Gamma irradiation is one of the physical mutagens that are widely used for induced mutations, food sterilization and medicinal healing. In the present study, irradiation techniques were applied to investigate the effect of gamma irradiation on seed germination, meiotic behavior and pollen fertility of tomato (*Solanum lycopersicum* L.). The seeds of *Solanum lycopersicum* (var. Bharat ratan -221) were irradiated with different doses of gamma rays (10, 15, 20, 25, 30, 35 and 40 kR). Results showed that seed germination and pollen fertility in M1 generation reduced steadily with the increasing doses of gamma rays. During the study, increases in meiotic abnormalities with increasing doses were noticed in mutagenic population of gamma rays in M1 generation. The most common chromosome abnormalities observed include univalents, multivalents, bridges, laggards, fragments, micronuclei and disturbed polarity, etc.

**KEYWORDS:** Tomato, *Solanum lycopersicum* L., Seed germination, Meiotic abnormalities, Gamma rays.

The cultivated tomato, *Solanum lycopersicum* L. (2n=24) belongs to the varied family Solanaceae, which includes more than 3,000 species, occupying a wide variety of habitats (Knapp 2002). *Solanum lycopersicum* L., was previously recognized as *Lycopersicon esculentum* Mill., but data from both morphology and molecular sequences support its inclusion in the large genus *Solanum* L., and a revised new nomenclature has resulted (Peralta and Spooner 2001).

After the discoveries of (Muller and Stadler), a large amount of genetic variability has been induced by various mutagens and contributed to modern plant breeding. For the past five decades the induced mutation had played a major role in the development of superior plant varieties are food crops especially cereals and pulses. The genetic variation may be created by induced mutation through the physical and chemical mutagens. These mutagens induced various abnormalities resulting in new varieties with desired characters. One of the chief advantages of mutation breeding applied to this crop that it can give rise to many different alleles with different degree of trait modifications (Chopra 2005).

Gamma rays are the most energetic form of electromagnetic radiation; their energy level is from ten to several hundred kilo electron volts and they are considered as the most penetrating compared to other radiations (Kovacs and Keresztes 2002). These radicals can damage or change important components of plant cells. They have been reported to affect differentially the morphology, anatomy, biochemistry and physiology of plants depending on the radiation dose (Asghar et al. 2003).

It is known to be the most popular mutagens for their simple application, good penetration, reproducibility, high mutation frequency and less disposal problems (Chahal and Gosal 2002).

**MATERIALS AND METHODS**

**Plant material** Experimental plant material selected for the present investigation was *Solanum lycopersicum* L. The seeds of this variety were procured from Daulatganj Market Ujjain, Madhya Pradesh.

**Gamma rays treatment** Approximately 500 dry and healthy seeds of the selected cultivars were subjected to various doses of gamma rays at the Bhabha Atomic Research Center, Mumbai with the treatment doses 10, 15, 20, 25, 30, 35 and 40 kR. About 150 irradiated and control seeds were sown in separate pots (five seeds in each pot).

**Cytological preparation and meiotic analysis** To determine the meiotic behavior, buds of appropriate size were collected from the field between 9 - 12 a.m. and fixed in Carnoy’s solution (glacial acetic acid : alcohol in the ratio of 1:3) for at least 24 hrs. Squash technique and 2% aceticarmine (as a stain) were used for cytological preparation. Microscopic photographs were taken from the temporary slides with the aid of an Olympus microscope.

**RESULTS**

The germination percentages of treated seeds and plant survival of M1 generation were studied, and were found to be inversely proportional to the dose given (Table 1). The germination percentage was reduced from 86.25% to 43.75% when the dose was increased from 10 kR to 40kR respectively. The plant survival percentage was reduced from 86.95% to 55.17% when the dose was increased from 10kR to 40kR respectively.
Meiosis was completely regular in control plants with 12 bivalents (2n = 24) at diakinesis and metaphase I stage (Figs. 1) and chromosomes were seen separating usually at anaphase stages (Figs. 2) without any abnormality. At metaphase I, the treated material showed univalents and multivalents (Figs. 3, 4, 5). The percentage of cells, showing univalents with 20kR and 40kR treatment was 0.47% to 2.60% respectively. The percentage of cells showing multivalents also increased from 0.93% to 2.17% when the dose was increased from 15kR to 40kR, respectively.

At anaphase, laggards, bridges formation and fragment of chromosomes were observed. Laggards were the most frequent abnormality observed in the treated sets (Fig. 6), the highest score for laggards, expressed in terms of percentage of anaphase I and telophase I cells having laggards, was observed at the radiation dose of 40 kR.

Table 1. The percentage of seed germination and plant survival in M1 generation of *Solanum lycopersicum* L. after different doses of gamma rays.

| Treatment dose | No. of Seeds sowed | Seed germination | Plant survival |
|----------------|--------------------|------------------|----------------|
|                |                    | No.  | % age  | No.  | % age  |
| Control        | 80                 | 78   | 97.5   | 77   | 98.71  |
| 10kR           | 80                 | 69   | 86.25  | 60   | 86.95  |
| 15kR           | 80                 | 61   | 76.25  | 52   | 85.24  |
| 20kR           | 80                 | 53   | 66.25  | 40   | 75.47  |
| 25kR           | 80                 | 49   | 61.25  | 34   | 69.38  |
| 30kR           | 80                 | 43   | 53.75  | 26   | 63.41  |
| 35kR           | 80                 | 39   | 48.75  | 22   | 57.89  |
| 40kR           | 80                 | 35   | 43.75  | 16   | 55.17  |

Table 2. The percentage of meiotic abnormalities in M1 generation of *Solanum lycopersicum* L. after different doses of gamma rays.

| Treatment doses | Frequency of abnormal PMCs at Metaphase-I/II (%) | Frequency of abnormal PMCs at Anaphase/telophase- I/II (%) |
|----------------|-----------------------------------------------|---------------------------------------------------------|
|                | Total PMCs scored   | Mult. | Uni. | Total | Fr. | Lag. | Br. | Mic. | Total |
| Control        | 250                | -     | -    | -     | 260 | -    | -   | -    | -     |
| 10kR           | 217                | 0.93  | -    | 0.93  | 246 | -    | -   | 0.40 | 0.40  |
| 15kR           | 213                | 0.95  | 0.47 | 1.42  | 239 | -    | -   | 0.83 | 0.41  |
| 20kR           | 210                | 1.37  | 0.91 | 2.28  | 236 | 0.42 | 0.84| 1.27 | 1.69  |
| 25kR           | 218                | 1.74  | 1.31 | 3.05  | 219 | 0.92 | 0.91| 1.82 | 2.28  |
| 30kR           | 229                | 2.07  | 1.65 | 3.72  | 215 | 0.93 | 1.39| 1.86 | 2.32  |
| 35kR           | 241                | 2.17  | 2.60 | 4.77  | 221 | 1.35 | 1.80| 2.26 | 2.71  |
| 40kR           | 230                | 2.17  | 2.60 | 4.77  | 209 | 1.43 | 1.91| 2.39 | 2.87  |

Abbr: Mult. - Multivalents, Uni. - Univalents, Fr. – Fragments, Lag. – Laggard, Br. – Bridge, Mic. - Micronuclei
Figures 1-10. 1. Diakinesis showing 12 bivalents; 2. PMCs showing normal anaphase I with 12=12 segregation; 3. PMCs showing univalent; 4, 5. PMCs showing multivalent; 6. Laggard at anaphase I; 7. Bridge at anaphase I; 8. Fragment at telophase I; 9, 10. Micronuclei at telophase II.
(1.91%). No laggards were observed in pollen mother cells of control progenies. Occurrence of cells having laggards had an increasing trend with increasing dose of radiation intensity (Table 2). Bridges and fragments at anaphase I and telophase I were observed in nearly all the doses (Figs. 7, 8), while the frequency of both abnormalities was higher in higher doses (Table 2).

Bridges were observed with or without fragments. The higher doses showed increasing percentage of bridges at anaphase I with the increased frequency from 0.83-2.39% in 15 kR and 40 kR, respectively (Table 2). The frequency of micronuclei in telophase II was very irregular not only within the treatment as a whole, but also within the PMCs of the same plant (Figs. 9, 10). The highest percentage of micronuclei was observed in 40kR of radiation dose (2.87%). Thus, in the present study, the total abnormality percentage showed an increasing trend with the increasing irradiation dose.

**DISCUSSION**

Seed germination in treated population was found to decrease with increasing doses of the gamma rays. Similar results were also observed (Borzouei et al. 10). The conflicting response in the extent of induced meiotic chromosomal aberrations may be accredited to the differences in the genotypic constitution of the plant and the mechanism of action of gamma rays. The percentage of plant survival decreased with the increasing intensity of gamma radiation. The results are in accordance with the studies of Chaudhary (1983). The increase in the frequency of chromosomal damage with increasing radiation dose may be responsible for reduction of plant survival in the present investigation.

The formation of univalents and multivalents at metaphase I has been reported in different plants such as barley (Kumar and Singh 2003). Occurrence of univalents may be due to the failure of pairing among homologous chromosomes indicating non-homology between chromosome. Multivalents may be recognized as pairing due to translocation and inversion (Dixit and Dubey 1986).

Laggards may also be attributed to the failure of the multivalents to separate accurately (Ganai et al. 2005). Chromosome breakage and reunion of its broken ends may result in formation of bridges (Ignacimuthu and Babu 1989). Bridges occur due to sister chromatids chromosomes exchange followed by delayed or failure of their separation at later stages. Bridge formation observed by Ahmad and Yasmin (1992). The fragments at metaphase may be due to the failure of broken chromosome to recombine. The fragments might have arisen due to the stickiness of chromosome and the consequent failure of the arrival of chromatids at the poles. Fragments may also be acentric chromosomes formed as a result of inversion (Agarwal and Ansari 2001).

Occurrence of micronuclei may be associated with the fragments and lagging chromosomes that could not reach the pole get involved with the daughter nuclei as recognized by Kumar and Dubey (1980). Disturbed polarity at telophase may cause due to disturbed spindle formation, also investigated by several workers as (Verma and Raina 1991) in Crotalaria juncea, (Verma and Sharma 2000) in Phlox drummondii., and (Verma et al. 2017) in Capsicum annum.

**CONCLUSION**

Cytological analysis has been considered as one of the reliable approaches to evaluate the potential of a mutagen by assessing the frequency of the cellular damage it causes in plants and provides the better prospects of selecting the appropriate mutagenic dose and by using which the positive mutations with desired characters could be induced in order to improve the plant. The present investigation revealed that lower doses of the gamma rays caused less cellular damage hence, can be optimized for inducing desired agronomic traits in tomato.

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