Studies on Quantitative Characters for Gamma Rays Treatment in Soybean (Glycine max (L.) Merr.) Var. Co-1

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Abstract: Effect of gamma rays on Soybean (Glycine max (L.) Merr.) var. Co-1 plants of three generation viz., M1, M2 and M3. The mutagens like Gamma rays were treatments for various doses (10, 20, 30, 40, 50, 60, 70, 80, 90 and 100KR) separately. The morphology and yield parameters were carried out M1, M2 and M3 generations. The effect of mutagens on different growth parameters, yields and yield components and quality (protein and oil) were studied. The seeds were grown in the field and observation separate concentration. The following characters were recorded such as days to first followers, plant height, number of leaves per plant, number of branches per plant, number of cluster per plant, number of pods per plant, number of seeds per plant, seed yield per plant, protein content and oil content. The yield parameters like plant height, number of cluster per plant, number of seeds per plant and seed yield per plant were recorded the moderated and high mean value in the 50 KR of treated population of all generation with compared to control plants.

Keywords: Soybean, mutagens, Gamma rays.

1. Introduction

The present study was investigated through induced mutation by gamma rays and their effects on quantitative traits of soybean (Glycine max (L.) Merr.) Variety CO-1 by means of extended the genetic variability.
1.1. Importance of Soybean

Soybean, Glycine max (L.) Merr., combines in one crop both the dominant world supply of edible vegetable oil, and the dominant supply of high-protein feed supplements for livestock. Other fractions and derivatives of the seed have substantial economic importance in a wide range of industrial, food, pharmaceutical, and agricultural products (Smith and Huyser, 1987). The United States is the principal world supplier of soybeans (Jewell, 1988). Soybean protein is rich in valuable amino acid lysine (5%) in which most of the cereals are deficient. In addition, it contains a good amount of minerals, salts and vitamins (thiamine and riboflavin) and its sprouting grains contain a considerable amount of Vitamin C. Vitamin A is present in the form of precursor carotene, which is converted into vitamin A in the intestine. A large number of Indian and western dishes such as bread, `chapati’, milk, sweets, pastries etc., can be prepared with soybean. Wheat flour fortified with soybean flour makes good quality and more nutritious ‘chapati’. Soybean oil is used for manufacturing vanaspati ghee and several other industrial products. Soybean is used for making high protein food for children. It is widely used in the industrial production of different antibiotics. Soybean builds up the soil fertility by fixing large amounts of atmospheric nitrogen through the root nodules, and also through leaf fall on the ground at maturity. It can be used as fodder; forage can be made into hay, silage etc. Its forage and cake are excellent nutritive foods for livestock and poultry. Soybean being the richest, cheapest and easiest source of best quality proteins and fats and having a vast multiplicity of uses as food and industrial products is sometimes called a wonder crop. Soybean is one of the important crops of the world. Production of soybean in India at the present time is restricted mainly to Madhya Pradesh, Uttar Pradesh Maharashtra and Gujarat. It is also grown on a small acreage in Himachal Pradesh, Punjab and Delhi.

1.2. Taxonomy of Soybean Relatives

The soybean is a papilionoid legume (family Fabaceae, subfamily Faboideae), and a member of the tribe Phaseoleae, subtribe Glycininae. The subtribe to which soybean belongs consists of 16 genera, none of which, save for soybean (Glycine) and kudzu (Pueraria), are commonly known outside of botanical science. The genus Glycine is unique within the subtribe on several morphological and chromosomal characters, and does not seem to bear an especially intimate relationship with any other genus in the subtribe (Lackey, 1977). A single exception may be the genus Sinodolichos, a rarely-collected and poorly-known genus from Asia. Sinodolichos is unknown in the living state outside of Asia (Lackey, 1981a).

The genus Glycine is divided into two questionably distinct subgenera: Glycine and Soia. The first consists of six or seven perennial species primarily from Australia. The second consists of three annual species from Asia: Glycine max, Glycine soja Sieb. & Zucc., and Glycine gracilis Skvortz. The
first species is the cultivated soybean, the second species is the wild form of the soybean, and the third species is the weedy form of the soybean (Lackey, 1981a).

1.3. Mutation in Soybean

The mutagenesis was quickly treated in gene level so the portant of crops production. The FAO/IAEA was first news released the data base for most of mutant varieties of Soybean. Mutation breeding supplement conventional plant breeding as a source of increasing variability and could confer specific improvement without significantly altering its acceptable phenotype (Ojomo et al., 1979). Mutations, both spontaneous and induced have been eminently successful in changing the fatty acid composition of several oilseed crops. Three reasons have been quoted for attempts in changing seed oil quality by means of single gene mutations being exceptionally successful. Induced mutation was abnormal of the chromosome is the primary basis of genetic change; therefore, investigations on the mechanism of chromosome breakage, type of aberrations, and their genetic consequence form an integral part of most mutation studies (Zeerak, 1992). Induced mutagenesis has been recognized as the most efficient method for induction of morphological and genetical variabilities in plants especially in those with limited genetic variabilities, because in plants the gene replacement experiments through homologous recombination with introduced DNA sequences have met with limited success.

2. Materials and Methods

2.1. Collection of Seeds

Soybean seeds variety CO-1 were collected from Millet breeding station, Tami Nadu Agriculture University (TNAU), Coimbatore.

2.1.1 Mutagenic treatment - Gamma irradiation treatment

The seeds were treated with different dose of Gamma rays (10, 20, 30, 40, 50, 60, 70, 80, 90 and 100KR) treated from sugarcane breeding institute (ICAR) Coimbatore.

2.2. Laboratory studies (Mi Generation)

2.2.1. Germination studies

Germinated seeds will count from 3rd to 7th day emergence of cotyledonary leaf will be taken as the indication of germination. Germination percentage will be worked out for the treatment in each genotype separately and lethality will be found out based on the mean value of 10 replicates.

2.2.2. Shoot and root length
The shoot and root length (cm) will be measured ten randomly selected seedlings with ten replications on the 15th day with the effect of physical mutagen along with control.

2.2.3. Lethal dosage (LD50 Value)

The LD50 value for Soybean variety CO-1 was observed at 50KR of gamma rays.

2.3. Field studies (M1, M2 and M3 generations-Based on the LD50 value)

Treated and control seeds were sown in the field (3 replication) in a randomized Block design (RBD) in order to raise the M1, M2 and M3 generations. Each treatment of doses consists of hundred seeds including control. The seed to seed and row to row distance was maintained at 15×60cm respectively. Cultural operations were carried out viz., irrigation and weeding. The following characters such as Days to first flower, Plant height, Number of branches per plant, Number of leaves per plant, Number of cluster per plant, Number of pod per plant, Number of seeds per plant, Hundred seed weight, Seed yield per plant, Fresh weight per plant and Dry weight per plant were observed in all the treatments along with R1- R3 generations.

2.3.1. Days to first flower (days)

The number of days taken from sowing to first flower was recorded and expressed as number of days to first flower.

2.3.2. Plant height (cm)

The height of the plant from the base to the top of the plant of maturity was measured and expressed in cm.

2.3.3. Number of branches per plant

Number of branches arising from the main stem were counted and recorded at the maturity.

2.3.4. Number of leaves per plant

The number of leaves was counter and recorded as the number of leaves per plants

2.3.5. Number of clusters per plant

Total number of clusters at maturity time were counted and recorded as the number of clusters per plant.

2.3.6. Number of pods per plant

Total number of pods at maturity time were counted and recorded as the number of pods per plant.
2.3.7. **Number of seeds per plant**

Total number of seeds from individual plant were counted and recorded as the number of seeds per plant.

2.3.8. **Seed yield per plant**

Seed yield was worked out by using digital electronic balance and expressed in grams.

2.3.9. **Hundred Seed weight (g)**

Hundred Seed weight was worked out by using digital electronic balance and expressed in grams.

2.3.10. **Fresh weight per plant (g)**

Fresh weight was measured after harvest by using digital electronic balance and expressed in grams per plant.

2.3.11. **Dry weight per plant (g)**

Dry weight was measured after harvest by drying in hot oven at 60°C, using digital electronic balance and expressed in grams per plant.

2.4. **Biochemical Studies**

2.4.1. **Seed protein content (%)**

Two seeds from the same plant of each M3 plants were separately collected and ground in a mortar and the extracts were defatted by washing with three changes of cold acetone for 4 to 6 hrs. The acetone was removed by filtration and the extracts were air-dried at room temperature. The proteins from the defatted meal were precipitated with 10% trichloro-acetic acid and recovered by centrifugation at 5000 rpm for 30 minutes at 40°C. The protein content was then determined calorimetrically according to the method of Lowry et al., (1951) using bovine serum albumin as standard.

2.4.2. **Seed oil content (%)**

The oil content of the kernel was estimated with petroleum ether in Soxhlet extraction apparatus (Cox and Pearson, 1962).

3. **Results and Discussion**

Effect of Gamma rays on Soybean for R1, R2 and R3 generations was thoroughly investigated with effect of gamma rays on quantitative traits such as germination percentage (LD50 Value), Days to First flower, Plant height (cm), Number of leaves per plant, Number of branches per plant, Number of
cluster per plant, Number of pods per plant, Number of seed per plant, Seed yield per plant (g), 100 seed weight (g), Fresh weight per plant (g) and Dry weight per plant (g), Seed Protein content and Seed Oil content.

3.1. Determination of LD$_{50}$ Value

Important directions to be made in a mutagenesis experiment are the choice of doses of mutagen. It can be achieved by means of lethal dose (LD$_{50}$ value). It refers to 50 per cent reduction of germination seeds in given progeny. The determination of optimal mutagenic dose is not easy. Because, low and high mutagenic doses are having some merits and demerits generally, higher mutagenic doses provide a higher number of possible mutants.

The present investigation exhibited that increased doses of gamma rays treatment in the resulted in the decrease in germination percentage, 50 per cent reduction in seed germination at 50KR of gamma rays (LD$_{50}$ value 52.82) respectively.

3.2. Seed Germination, Seedling Survival

Different doses of gamma rays treatment showed gradual reduction of seed germination (10$^{th}$ day), seedling survival (30$^{th}$ day) and Plant height (30$^{th}$ day) than control. The effects of gamma rays were different doses on survival percentage, mutation frequency and mutagenic effectiveness. The survival percentage and mean value of M$_1$ generation were decreased with increase the dose of treatments.

3.3. M$_1$ Generation

Induced mutation procedure in the recent past has successfully been used for the improvement of various pulse crops. In the present investigation, all the parameters were gradually reduced in R$_1$ generation at maturity time except days to first flowering which increase the days indicated inhibitory effects. The present result confirms these earlier reports. Amarnath et al., (1991) reported that Genotypic and phenotypic variability and heritability of some quantitative characters in soybean (Glycine max (L.) Merr).The growth and other quantitative traits were proportionately decreased with increasing dose of gamma irradiation in the present study. Cheng et al., (1999) the structural, biochemical and genetic characterization of a new radiation-induced, variegated leaf mutant of soybean (Glycine max (L.) Merr.).

The decrease in survival percentage has been attributed to the physiological disturbance or chromosomal damage caused to the cells of the plant by the mutagen. Mensah (1977) recorded reduction in germination and survival percentage due to the effect of chemical mutagens in cowpea.
The M₁ generation was assessed at the field level to measure the intensity of injury caused by mutagenic treatments (Gaul, 1970).

Table 1. Effect of gamma rays on soybean in M₁ generation

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Days to first flower</th>
<th>Plant height (cm)</th>
<th>No. of leaves/plant</th>
<th>No. of branch/plant</th>
<th>No. of cluster/plant</th>
<th>No. of pod/plant</th>
<th>No. of seed/plant</th>
<th>Seed yield (g)</th>
<th>protein content (%)</th>
<th>oil content (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>35.58 ± 2.11</td>
<td>69.88 ± 1.94</td>
<td>68.32 ± 3.32</td>
<td>4.87 ± 0.61</td>
<td>23.21 ± 1.32</td>
<td>23.21 ± 1.32</td>
<td>80.21 ± 3.33</td>
<td>12.87 ± 0.98</td>
<td>36.14 ± 2.06</td>
<td>18.81 ± 0.78</td>
</tr>
<tr>
<td>Gamma rays 10 KR</td>
<td>36.32 ± 2.30</td>
<td>73.50 ± 2.30</td>
<td>65.11 ± 4.05</td>
<td>4.32 ± 0.35</td>
<td>21.47 ± 1.50</td>
<td>21.47 ± 1.50</td>
<td>76.32 ± 2.48</td>
<td>11.62 ± 0.35</td>
<td>36.37 ± 1.87</td>
<td>18.77 ± 0.41</td>
</tr>
<tr>
<td>20 KR</td>
<td>36.13 ± 1.58</td>
<td>74.41 ± 1.58</td>
<td>69.38 ± 3.98</td>
<td>4.55 ± 0.41</td>
<td>23.65 ± 1.44</td>
<td>23.65 ± 1.44</td>
<td>81.25 ± 1.65</td>
<td>12.35 ± 0.66</td>
<td>36.15 ± 2.04</td>
<td>19.24 ± 0.32</td>
</tr>
<tr>
<td>30 KR</td>
<td>35.11 ± 1.56</td>
<td>75.24 ± 1.56</td>
<td>71.45 ± 3.54</td>
<td>5.01 ± 0.20</td>
<td>24.55 ± 1.06</td>
<td>24.55 ± 1.06</td>
<td>80.69 ± 1.47</td>
<td>10.43 ± 0.84</td>
<td>37.02 ± 1.52</td>
<td>19.36 ± 0.41</td>
</tr>
<tr>
<td>40 KR</td>
<td>35.03 ± 1.30</td>
<td>75.66 ± 1.30</td>
<td>74.08 ± 1.47</td>
<td>5.36 ± 0.13</td>
<td>22.08 ± 1.11</td>
<td>22.08 ± 1.11</td>
<td>82.33 ± 1.08</td>
<td>13.24 ± 0.47</td>
<td>37.81 ± 1.22</td>
<td>19.04 ± 0.27</td>
</tr>
<tr>
<td>50 KR</td>
<td>33.44 ± 1.00</td>
<td>73.01 ± 1.00</td>
<td>78.26 ± 1.55</td>
<td>5.84 ± 0.11</td>
<td>26.78 ± 1.87</td>
<td>26.78 ± 1.87</td>
<td>85.46 ± 1.33</td>
<td>14.18 ± 0.98</td>
<td>38.54 ± 0.97</td>
<td>19.63 ± 0.21</td>
</tr>
<tr>
<td>60 KR</td>
<td>35.11 ± 1.41</td>
<td>74.22 ± 1.41</td>
<td>60.53 ± 1.07</td>
<td>3.62 ± 0.30</td>
<td>21.56 ± 0.59</td>
<td>21.56 ± 0.59</td>
<td>78.31 ± 1.52</td>
<td>12.56 ± 0.24</td>
<td>33.11 ± 0.56</td>
<td>18.42 ± 0.40</td>
</tr>
</tbody>
</table>

3.4. Effect of Gamma Irradiation on M₂ and M₃ Generations

In M₂ and M₃ generations all the parameters gradually increased in optimum doses. The present results confirm these earlier reports in black gram (Arulbalachandran 2006); Mung bean (Khan and Wani 2005): Zea mays (Gnanamurthy et al., 2011) and Cowpea (Dhanavel et al., 2012) The R₂ Plants raised in the field were examined to identify the mutants induced by gamma rays, as well as to find out their effects on various quantitative characters. The maximum mutants were observed at 50KR of gamma rays.

Table 2. Effect of Gamma rays on Soybean in M 2 generation (Mean value ± SE).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Germination %</th>
<th>Seedling survival %</th>
<th>Days to first flower</th>
<th>Root length (cm)</th>
<th>Shoot length (cm)</th>
<th>No. of fruit/plant</th>
<th>Seed yield (g)</th>
<th>Fresh weight (g)</th>
<th>Dry weight (g)</th>
<th>100 Seed weight (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>97.24</td>
<td>92.45</td>
<td>37.21</td>
<td>25.64</td>
<td>68.39</td>
<td>70.51</td>
<td>16.08</td>
<td>87.19</td>
<td>47.65</td>
<td>10.39</td>
</tr>
<tr>
<td>Gamma rays 10 KR</td>
<td>95.11</td>
<td>90.35</td>
<td>38.27</td>
<td>22.54</td>
<td>66.18</td>
<td>66.25</td>
<td>15.10</td>
<td>84.25</td>
<td>43.28</td>
<td>10.02</td>
</tr>
<tr>
<td>20 KR</td>
<td>84.27</td>
<td>83.06</td>
<td>38.08</td>
<td>20.16</td>
<td>62.05</td>
<td>61.48</td>
<td>14.35</td>
<td>80.04</td>
<td>39.49</td>
<td>9.87</td>
</tr>
<tr>
<td>30 KR</td>
<td>69.84</td>
<td>66.35</td>
<td>40.21</td>
<td>18.84</td>
<td>58.54</td>
<td>51.38</td>
<td>13.07</td>
<td>76.29</td>
<td>35.28</td>
<td>9.36</td>
</tr>
<tr>
<td>40 KR</td>
<td>58.87</td>
<td>54.08</td>
<td>40.65</td>
<td>18.23</td>
<td>51.20</td>
<td>47.59</td>
<td>12.54</td>
<td>71.24</td>
<td>34.07</td>
<td>9.04</td>
</tr>
<tr>
<td>50 KR</td>
<td>51.37</td>
<td>46.28</td>
<td>41.10</td>
<td>17.51</td>
<td>45.29</td>
<td>44.06</td>
<td>12.05</td>
<td>66.01</td>
<td>32.15</td>
<td>8.90</td>
</tr>
<tr>
<td>60 KR</td>
<td>37.39</td>
<td>35.24</td>
<td>43.08</td>
<td>15.29</td>
<td>40.07</td>
<td>41.17</td>
<td>11.04</td>
<td>59.81</td>
<td>27.40</td>
<td>8.81</td>
</tr>
</tbody>
</table>
Table 3. Effect of gamma rays on soybean in M3 generation (Mean value ± SE)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Days to first flower (Days)</th>
<th>Plant height (cm)</th>
<th>No. of leaves/plant</th>
<th>No. of branch/plant</th>
<th>No. of cluster/plant</th>
<th>No. of pod/plant</th>
<th>No. of seed/plant</th>
<th>Seed yield (g)</th>
<th>Protein content (%)</th>
<th>Oil content (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>36.81 ± 1.43</td>
<td>70.75 ± 4.32</td>
<td>64.27±1.99</td>
<td>64.32±0.11</td>
<td>21.62±1.54</td>
<td>49.62±1.82</td>
<td>79.81±3.81</td>
<td>10.21±0.97</td>
<td>37.56±1.27</td>
<td>18.05±1.71</td>
</tr>
<tr>
<td>Gamma rays 10</td>
<td>36.99 ± 1.25</td>
<td>75.66 ± 2.56</td>
<td>69.54±2.56</td>
<td>4.46±0.13</td>
<td>22.32±0.89</td>
<td>59.62±1.96</td>
<td>81.37±2.76</td>
<td>11.64±0.69</td>
<td>38.32±1.52</td>
<td>18.37±1.15</td>
</tr>
<tr>
<td>KR 20</td>
<td>35.47 ± 1.08</td>
<td>76.43 ± 5.04</td>
<td>73.21±4.17</td>
<td>4.51±0.15</td>
<td>20.81±0.94</td>
<td>60.39±3.71</td>
<td>80.26±5.52</td>
<td>12.07±0.77</td>
<td>38.50±1.90</td>
<td>19.22±1.36</td>
</tr>
<tr>
<td>KR 30</td>
<td>37.18 ± 0.69</td>
<td>74.41 ± 3.98</td>
<td>70.42±3.56</td>
<td>5.02±0.10</td>
<td>21.37±1.37</td>
<td>60.55±4.21</td>
<td>83.36±6.36</td>
<td>11.85±0.96</td>
<td>39.02±1.16</td>
<td>19.56±1.05</td>
</tr>
<tr>
<td>KR 40</td>
<td>35.24 ± 0.78</td>
<td>80.71 ± 2.54</td>
<td>75.37±1.82</td>
<td>5.07±0.09</td>
<td>24.39±0.99</td>
<td>61.27±2.96</td>
<td>82.97±5.91</td>
<td>12.39±0.82</td>
<td>39.13±2.37</td>
<td>18.85±0.58</td>
</tr>
<tr>
<td>KR 50</td>
<td>34.35 ± 1.14</td>
<td>81.02 ± 2.61</td>
<td>81.52±4.21</td>
<td>5.16±0.27</td>
<td>26.51±1.72</td>
<td>62.21±3.27</td>
<td>86.37±4.56</td>
<td>13.46±0.82</td>
<td>39.56±1.94</td>
<td>18.76±1.02</td>
</tr>
<tr>
<td>KR 60</td>
<td>37.65 ± 1.22</td>
<td>62.89 ± 2.78</td>
<td>61.32±2.37</td>
<td>5.02±0.16</td>
<td>20.12±1.01</td>
<td>50.29±3.11</td>
<td>84.80±3.82</td>
<td>12.55±1.27</td>
<td>38.06±2.07</td>
<td>18.22±0.89</td>
</tr>
</tbody>
</table>

4. Conclusion

The present investigation, different dose of gamma irradiation induced mutation in R1 generation inhibit seedling growth and other growth parameters in field condition. The inhibitory effects were due to physiological disturbances on soybean by gamma irradiation especially with imbalance of growth hormones. From the results LD50 value was fixed at 50KR gamma rays. In R2 and R3 generations 50KR of gamma rays showed more frequency of chlorophyll mutants, viable mutants, effectiveness and efficiency.

References


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