

## Effect and Sensitivity of Gamma Irradiation to Various Biometrical Traits of Cowpea (*Vigna unguiculata* (L.) Walp.)

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**ABSTRACT:** The present study was conducted to assess the effect and sensitivity of gamma rays to different biometrical traits in two popular high yielding, genetically diverse grain type cowpea (*Vigna unguiculata* (L.) Walp.) varieties viz. Paiyur 1 and Goa cowpea 3. Cowpea is a highly self pollinated crop with limited variability. Inducing mutations can thus play an important role in creating variations within the crop and offer a better opportunity for developing new desirable traits. The two cowpea varieties selected for the study have not undergone mutation studies so far. Hence, an attempt was made to identify desirable mutants by gamma irradiating them. The two cowpea varieties were exposed to varying doses (9 doses) of gamma rays to study their effect on germination, plant survival, seedling length, vigour index, pollen fertility and various quantitative traits like days to 50 per cent flowering, days to maturity, plant height, number of primary branches per plant, number of pod clusters per plant, number of pods per cluster, number of seeds per pod, pod length, hundred seed weight and seed yield per plant. LD<sub>50</sub> was calculated for each of the two varieties on the basis probit analysis and the optimum dose were obtained as 200 Gy for Paiyur 1 and 400 Gy for Goa cowpea 3. A dose dependant decrease was noticed for most of the characters in M<sub>1</sub> generation. The results indicated that the reduction in germination per cent, shoot length and root length over control was noticed in all mutagenic treatments in both the cultivars, while increased pollen sterility was associated with corresponding increases in dose of mutagens.

**Keywords:** Cowpea, Gamma rays, *in vitro* studies and biometrical traits.

## INTRODUCTION

Cowpea (*Vigna unguiculata* (L.) Walp.), often known as ‘black-eyed pea’, is a significant food legume crop that is widely grown throughout the world in Southern Europe, Africa, Central and South America, Asia, Oceania, and the United States (Quin, 1997). Cowpea is considered to be one of the most important food sources and offers nutritional security in the semi-arid regions of Sub-Saharan Africa due to its innate drought tolerance and capacity to flourish even in marginal lands. It is used as a vegetable, food, forage crop in the tropics (Steele and Mehra 1972).

The cowpea is also popularly known to be a ‘*poor man’s meat*’ owing to its presence of higher level of carbohydrate (57%), higher quantity of quality protein (21-33%) and rich in lysine and tryptophan content compared to other cereals. Also, it is a rich source of minerals (calcium, zinc and iron) and amino acids (β-carotene, thiamine, riboflavin and folic acid). Cowpea leaves, serves as a green nutritious fodder for the milch animals due to the presence of higher protein content (27-34%). After the harvest of pods, the dried plant

could be used as fodder for ruminant animals during the lean season and the *in situ* decomposition of cowpea roots in the soil enhances the nitrogen level to the tune of 40-80 kg N ha<sup>-1</sup> by the symbiotic nitrogen fixation bacteria, *Bradyrhizobium* spp. (Quin, 1997).

In India, cowpea is cultivated in an area of 654 lakh hectares with a production of 599 lakh tonnes. The productivity of cowpea is 916 kg ha<sup>-1</sup> (Joshi *et al.*, 2018). The major cowpea growing states are Maharashtra, Karnataka, Tamil Nadu, Gujarat, Madhya Pradesh and Andhra Pradesh. In Tamil Nadu, cowpea is cultivated in an area of 65,836 hectares with a production of 50,145 tonnes (2019-20) ([https://aps.dac.gov.in/APY/Public\\_Report1.aspx](https://aps.dac.gov.in/APY/Public_Report1.aspx)).

The productivity of cowpea is 760 kg/ha which is much below the national average. The poor productivity of cowpea is due to its cultivation in the infertile soils under rainfed conditions, propensity towards natural vagaries of monsoon and pathogenic organisms, asynchronous pod maturity, shedding of newly formed pod causes poor sink realisation, inherent nature of indeterminate growth habit and long duration of the crop.

The strategies for improving the cowpea productions are (i) to enhance the productivity level and bridging the yield gap and (ii) development of high yielding short duration varieties having multiple resistance to accommodate 2-3 crops in a year. Such early maturity varieties can be accommodated well under intercropping situations with sugarcane.

Quantum of genetic variability present in a population is a prime requisite for any crop improvement programme, which ultimately results in the development of high yielding varieties. The availability of natural variability in the gene pool of *Vigna unguiculata* (L.) Walp. is not sufficient to evolve high yielding cultivars. Hence, it is essential to create genetic variability through specialised techniques like induced mutation by employing physical or chemical mutagens. Many high yielding varieties in castor (Ankinudu *et al.*, 1968), wheat (Swaminathan, 1969), sesame (Sharma, 1993), cowpea (Dhanavel *et al.*, 2008), black gram (Thilagavathi and Mullainathan 2009) and soybean (Pavadai *et al.*, 2010) were developed by mutation breeding approach.

Both physical and chemical mutagens are extensively used for the creation of genetic variability. However, in the present study, physical mutagen like gamma rays is employed to create genetic variability in cowpea. Gamma rays can induce multiple types of DNA damage, ranging from nucleotide modifications to DNA strand breaks (e.g., oxidized base, a basic sites, single-strand breaks (SSBs), double-strand breaks (DSBs)). If this DNA damage fails to be repaired or is repaired imprecisely, mutations such as single-base substitutions (SBSs), deletions, insertions, inversions, or translocations may occur at the genome scale and finally lead to changes in the phenotypic traits.

The aim of the present study is to assess the effect and sensitivity of gamma rays on germination, shoot length, root length, seedling height, vigour index, plant survival, pollen fertility and other quantitative traits in  $M_1$  generation.

## MATERIALS AND METHODS

Material of this consists of two popular high yielding grain type cowpea varieties *viz.* Paiyur 1 and Goa cowpea 3. Pure seeds were obtained from Regional Research Station, Paiyur, Tamil Nadu, India and ICAR-Central Coastal Agricultural Research Institute Goa, India respectively.

Two hundred healthy seeds of Paiyur 1 and Goa cowpea 3 were selected and exposed to nine doses of gamma rays *viz.*, 100 Gy, 150 Gy, 200 Gy, 250 Gy, 300 Gy, 350 Gy, 400 Gy, 450 Gy, 500 Gy of gamma rays at Indira Gandhi Centre for Atomic Research, Kalpakkam, Tamil Nadu. Non-irradiated seeds of these two varieties were considered as control. Hundred seeds in each variety and dosage have been reserved for conducting *in vitro* studies and the remaining seeds were utilised for raising the  $M_1$  generation.

The *in vitro* study was conducted with two replications in the Laboratory of Department of Plant Breeding and Genetics, Anbil Dharmalingam Agricultural College and Research Institute, Tiruchirappalli. The gamma ray treated seeds were transferred to moist filter paper for germination studies at the rate of 50 seeds per replication. For comparison, untreated seeds were soaked in normal water for 6 hours and directly placed in the moist filter paper to serve as a control. The treated and untreated seeds were observed on 7<sup>th</sup> day after sowing for assessing the germination percentage. Shoot length, root length, seedling height were recorded 14 days after sowing. Vigour index is calculated based on the formula suggested by Abdul-Baki and Anderson (1973). The dose of mutagen at which 50 per cent mortality is observed were considered as LD<sub>50</sub> value for the variety / genotype. The LD<sub>50</sub> value was calculated based on probit analysis.

The  $M_1$  generation of selective doses was raised during late *Rabi* 2021 season in the experimental field of Department of Plant Breeding and Genetics, Anbil Dharmalingam Agricultural College and Research Institute, Tiruchirappalli. Both treated and untreated seeds were sown in a RBD with two replications. The experimental plots size was 3.3 × 2.5 m<sup>2</sup>. The rows were spaced 30 cm apart with an inter plant distance of 15 cm.

***In vitro* and field observations recorded.** For each dose of mutagen, the mean values for germination percentage, survival percentage, shoot length, root length, seedling height and vigour index were recorded under *in vitro* conditions in the laboratory; whereas, observations *viz.*, plant height at 30 days after sowing, plant height at maturity, days to 50% flowering, days to maturity, number of primary branches per plant, number of pod clusters per plant, number of pods per cluster, pod length, number of seeds per pod, hundred seed weight and seed yield per plant were recorded from all the  $M_1$  plants in selected doses and control varieties from each replication.

Pollen fertility was also determined by staining the pollens with 1 per cent Iodine-Potassium Iodide stain. The fully stained pollen grains having proper shape were considered as fertile, while the unstained, abnormal shaped and improperly filled pollen grains were categorised as sterile. Pollen fertility per cent (%) was measured as the ratio of fertile to the total number of pollens observed in the microscopic field (10x). This observation was based on 10 randomly sampled flower buds. For this study, one bud was randomly sampled from each of the 10 randomly sampled plants of each plot of each dose.

Pollen fertility percentage = Total number of well stained pollen grains/Total number of well stained and unstained pollen grains × 100.

**Statistical analysis.** The biometrical observations that were made at the relevant phases of the crop were subjected to first order statistical analysis.

## RESULTS AND DISCUSSION

### A. Effect of gamma irradiation on germination and survival percentage

The effect of mutagen on various traits observed viz. germination per cent and survival per cent are presented in Table 1 and 2. Both traits showed a linear, dose-dependent negative relationship as the mutagen's dose was increased. The values of the traits showed decreasing trend with the increase in the dose of

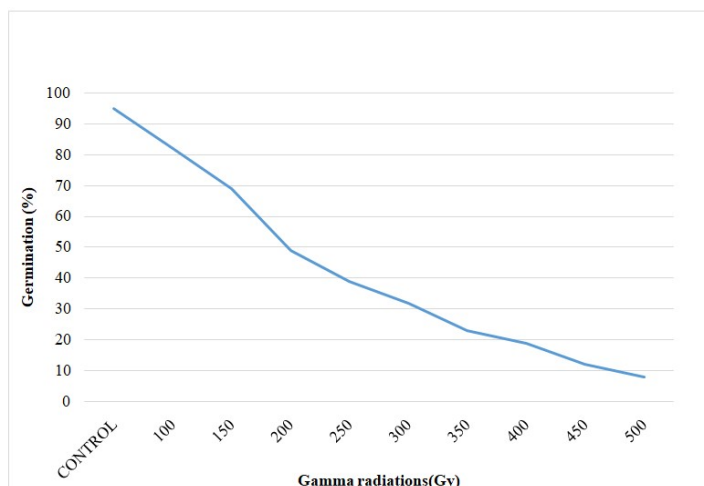
mutagen. Mohammad *et al.* (2018) reported that disturbances caused by the mutagen at cellular and physiological levels interfere with the biological processes of the tissues and exhibited significant change in the phenotype. The seed germination percentage ranged from 8 (500 Gy) to 82 (100 Gy) per cent in Paiyur 1 and 25 (500 Gy) to 88(100 Gy) per cent in Goa cowpea 3. The Paiyur 1 control exhibited maximum germination of 95.0 per cent; whereas Goa cowpea 3 registered 93 per cent (Fig. 1 and 2).

**Table 1: Effect of gamma irradiation on the germination and survival percentage of Paiyur 1 cowpea.**

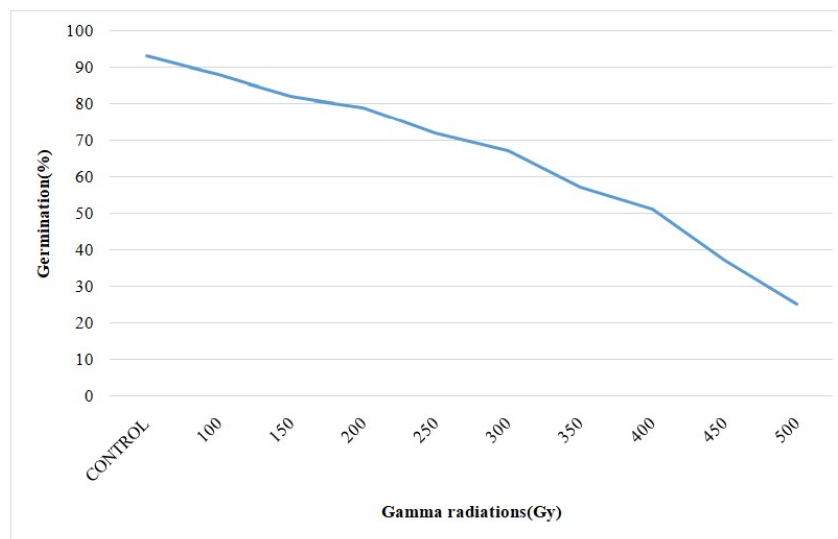
| Treatment | Germination per cent | Per cent over control | % reduction over control | Survival per cent | Per cent over control | % reduction over control |
|-----------|----------------------|-----------------------|--------------------------|-------------------|-----------------------|--------------------------|
| 100 Gy    | 82                   | 86.32                 | 13.68                    | 76                | 86.36                 | 13.63                    |
| 150 Gy    | 69                   | 72.63                 | 27.37                    | 60                | 68.18                 | 31.82                    |
| 200 Gy    | 49                   | 51.58                 | 48.42                    | 47                | 53.40                 | 46.59                    |
| 250 Gy    | 39                   | 41.05                 | 58.95                    | 34                | 38.63                 | 61.36                    |
| 300 Gy    | 32                   | 33.68                 | 66.32                    | 28                | 31.82                 | 68.18                    |
| 350 Gy    | 23                   | 24.21                 | 75.79                    | 19                | 21.59                 | 78.40                    |
| 400 Gy    | 19                   | 20.00                 | 80                       | 15                | 17.04                 | 82.95                    |
| 450 Gy    | 12                   | 12.63                 | 87.37                    | 8                 | 9.09                  | 90.90                    |
| 500 Gy    | 8                    | 8.42                  | 91.57                    | 5                 | 5.68                  | 94.31                    |
| Control   | 95                   | 100                   | 0                        | 88                | 100                   | 0                        |
| Mean      | 37                   | -                     | 61.05                    | 32.44             | -                     | 63.13                    |
| SE        | 8.50                 | -                     | -                        | 9.34              | -                     | -                        |

**Table 2: Effect of gamma irradiation on the germination and survival percentage of Goa cowpea 3 cowpea.**

| Treatment | Germination per cent | Per cent over control | % reduction over control | Survival per cent | Per cent over control | % reduction over control |
|-----------|----------------------|-----------------------|--------------------------|-------------------|-----------------------|--------------------------|
| 100 Gy    | 88                   | 94.62                 | 5.37                     | 85                | 95.50                 | 4.49                     |
| 150 Gy    | 82                   | 88.17                 | 11.83                    | 78                | 87.64                 | 12.36                    |
| 200 Gy    | 79                   | 84.95                 | 15.05                    | 74                | 83.15                 | 16.85                    |
| 250 Gy    | 72                   | 77.41                 | 22.58                    | 69                | 77.53                 | 22.47                    |
| 300 Gy    | 67                   | 72.04                 | 27.96                    | 64                | 71.91                 | 28.08                    |
| 350 Gy    | 57                   | 61.29                 | 38.71                    | 53                | 59.56                 | 40.44                    |
| 400 Gy    | 51                   | 54.84                 | 45.16                    | 49                | 55.06                 | 44.94                    |
| 450 Gy    | 37                   | 39.78                 | 60.22                    | 34                | 38.20                 | 61.80                    |
| 500 Gy    | 25                   | 26.88                 | 73.11                    | 21                | 23.59                 | 76.40                    |
| Control   | 93                   | 100                   | 0                        | 89                | 100                   | 0                        |
| Mean      | 62                   | -                     | 33.33                    | 58.56             | -                     | 34.20                    |
| SE        | 7.10                 | -                     | -                        | 7.06              | -                     | -                        |



**Fig. 1.** Dose response relationship for germination percentage in cowpea variety Paiyur 1 after treatment with different doses of gamma irradiation.



**Fig. 2.** Dose response relationship for germination percentage in cowpea variety Goa cowpea 3 after treatment with different doses of gamma irradiation.

In terms of percentage reduction over the control, the range was from 13.68(100 Gy) to 91.57 (500 Gy) per cent in Paiyur 1 and from 5.37 (100 Gy) to 73.11(500 Gy) per cent in Goa cowpea 3. Similar results showing dose dependent decrease in germination were earlier reported by Ramya *et al.* (2014), Uma and Salimath (2001) and Akilan *et al.* (2020). Fifty per cent reduction in germination was observed around 200 Gy-250Gy in Paiyur 1 and 350 Gy - 400 Gy in Goa cowpea 3.

The survival rate also showed a similar decreasing trend in all mutagenic treatments compared to its respective control (Paiyur 1: 88% and Goa cowpea 3: 89 %) on 30<sup>th</sup> day. The survival percentage ranged from 5.68 (500 Gy) to 86.36 (100 Gy) per cent in Paiyur 1 and 23.59 (500 Gy) to 95.50 (100 Gy) per cent in Goa cowpea 3.

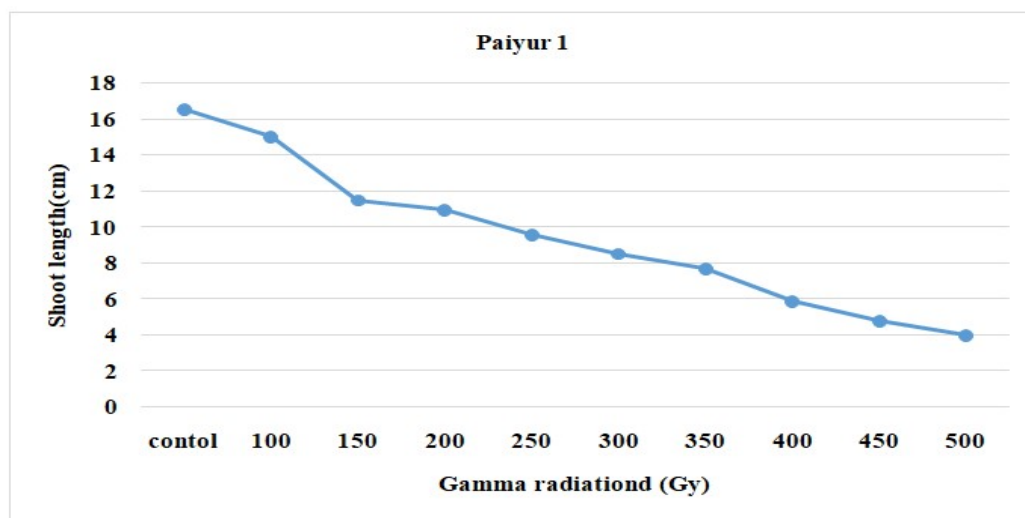
The percent reduction of survived seedlings over the control also exhibited a reduction ranging from 13.63 (100 Gy) to 94.31 (500 Gy) per cent in Paiyur 1 and from 4.49 (100 Gy) to 76.40 (500 Gy) per cent in Goa cowpea 3. The inability of the mutagen treated cells to repair the damage done to them could be the cause of the reduction in survival percentage. These findings are similar with the previous reports of Ugorji *et al.* (2012), Dhanavel and Girija (2009) and Kumar V Ashok (2010).

#### *B. Effect of gamma irradiation on root length, shoot length, seedling height and vigour index*

The effects of mutagen on shoot length, root length, seedling height and vigour index are presented in Table 3 and 4.

**Table 3: Effect of gamma irradiation on the shoot length, root length, seedling length (at 14 DAS) and vigour index in Paiyur 1 cowpea.**

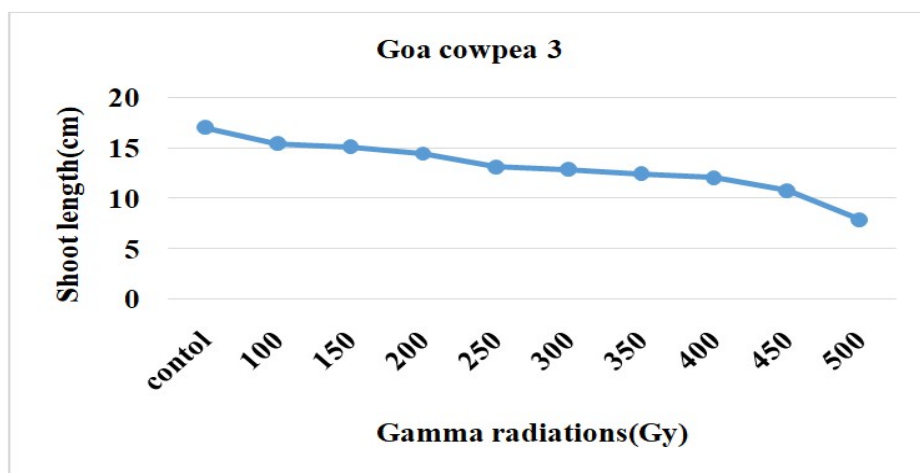
| Treatment | Shoot length (cm) | Per cent over control | % reduction over control | Root length (cm) | Per cent over control | % reduction over control | Seedling length (cm) | Per cent over control | % reduction over control | Vigour index | Percent over control |
|-----------|-------------------|-----------------------|--------------------------|------------------|-----------------------|--------------------------|----------------------|-----------------------|--------------------------|--------------|----------------------|
| 100 Gy    | 15.04             | 90.88                 | 9.12                     | 16.15            | 94.62                 | 5.39                     | 31.19                | 92.77                 | 7.23                     | 25.57        | 80.07                |
| 150 Gy    | 11.50             | 69.49                 | 30.51                    | 12.36            | 72.41                 | 27.59                    | 23.86                | 70.97                 | 29.03                    | 16.46        | 51.54                |
| 200 Gy    | 10.96             | 66.23                 | 33.77                    | 11.72            | 68.66                 | 31.34                    | 22.68                | 67.46                 | 32.54                    | 11.11        | 34.79                |
| 250 Gy    | 9.56              | 57.76                 | 42.24                    | 10.68            | 62.56                 | 37.43                    | 20.24                | 60.20                 | 39.79                    | 7.89         | 24.71                |
| 300 Gy    | 8.52              | 51.49                 | 48.52                    | 9.15             | 53.60                 | 46.40                    | 17.67                | 52.56                 | 47.44                    | 5.65         | 17.70                |
| 350 Gy    | 7.69              | 46.46                 | 53.53                    | 8.24             | 48.27                 | 51.73                    | 15.93                | 47.38                 | 52.62                    | 3.66         | 11.47                |
| 400 Gy    | 5.91              | 35.71                 | 64.29                    | 6.82             | 39.95                 | 60.05                    | 12.37                | 37.86                 | 62.14                    | 2.41         | 7.57                 |
| 450 Gy    | 4.79              | 28.94                 | 71.06                    | 5.31             | 31.11                 | 68.89                    | 10.1                 | 30.04                 | 69.95                    | 1.21         | 3.79                 |
| 500 Gy    | 3.99              | 24.11                 | 75.89                    | 4.66             | 27.30                 | 72.70                    | 8.65                 | 25.72                 | 74.27                    | 0.69         | 2.16                 |
| Control   | 16.55             | 100                   | 0                        | 17.07            | 100                   | 0                        | 33.62                | 100                   | 0                        | 31.93        | 100                  |
| Mean      | 8.66              |                       | 47.66                    | 9.45             |                       | 44.61                    | 19.667               |                       | 46.11                    | 10.66        |                      |
| SE        | 1.18              |                       |                          | 1.23             |                       |                          | 2.41                 |                       |                          | 2.74         |                      |



**Fig. 3.** Dose response relationship for shoot length in paiyur 1 cowpea variety after treatment with different dose of gamma radiations.

**Table 4:** Effect of gamma irradiation on the shoot length, root length, seedling length (at 14DAS) and vigour index in Goa cowpea 3.

| Treatment | Shoot length (cm) | Per cent over control | % reduction over control | Root length (cm) | Per cent over control | % reduction over control | Seedling length (cm) | Percent over control | % reduction over control | Vigour index | Per cent over control |
|-----------|-------------------|-----------------------|--------------------------|------------------|-----------------------|--------------------------|----------------------|----------------------|--------------------------|--------------|-----------------------|
| 100 Gy    | 15.45             | 90.62                 | 9.38                     | 18.09            | 94.91                 | 5.09                     | 33.54                | 92.88                | 7.117                    | 29.51        | 87.88                 |
| 150 Gy    | 15.09             | 88.50                 | 11.50                    | 16.29            | 85.46                 | 14.53                    | 31.38                | 86.90                | 13.09                    | 25.73        | 76.62                 |
| 200 Gy    | 14.42             | 84.58                 | 15.43                    | 15.47            | 81.16                 | 18.84                    | 29.89                | 82.77                | 17.23                    | 23.61        | 70.31                 |
| 250 Gy    | 13.12             | 76.95                 | 23.05                    | 14.12            | 74.08                 | 25.92                    | 27.24                | 75.44                | 24.56                    | 19.61        | 58.40                 |
| 300 Gy    | 12.88             | 75.54                 | 24.46                    | 14               | 73.45                 | 26.55                    | 26.88                | 74.44                | 25.56                    | 18.00        | 53.62                 |
| 350 Gy    | 12.44             | 72.96                 | 27.04                    | 13.47            | 70.67                 | 29.33                    | 25.91                | 71.75                | 28.25                    | 14.77        | 43.98                 |
| 400 Gy    | 12.07             | 70.79                 | 29.21                    | 12.8             | 67.16                 | 32.84                    | 24.87                | 68.87                | 31.13                    | 12.68        | 37.76                 |
| 450 Gy    | 10.78             | 63.22                 | 36.77                    | 12.1             | 63.48                 | 36.52                    | 22.88                | 63.36                | 36.64                    | 8.46         | 25.20                 |
| 500 Gy    | 7.87              | 46.16                 | 53.84                    | 9.09             | 47.69                 | 52.31                    | 16.96                | 46.96                | 53.03                    | 4.24         | 12.63                 |
| Control   | 17.05             | 100                   | 0                        | 19.06            | 100                   | 0                        | 36.11                | 100                  | 0                        | 33.58        | 100                   |
| Mean      | 12.68             |                       | 25.63                    | 13.94            |                       | 26.88                    |                      |                      | 26.29                    | 17.40        |                       |
| SE        | 0.781             |                       |                          | 0.86             |                       |                          |                      |                      |                          | 27.44        |                       |



**Fig. 4.** Dose response relationship for shoot length in Goa cowpea 3 variety after treatment with different dose of gamma radiations.



The length of the seedlings was recorded after 14 days. Mean seedlings length ranging from 8.65 (500 Gy) to 31.19 cm (100 Gy) in Paiyur 1 and from 16.96 (500 Gy) to 33.54 cm (100 Gy) in Goa cowpea 3 (Fig. 7, 8). In terms of percentage reduction in the seedling length, it ranged from 7.23 (100 Gy) to 74.27 per cent (500 Gy) in Paiyur 1 and from 7.12 (100 Gy) to 53.03 per cent (500 Gy) in Goa cowpea 3. The reduction in length was also observed in soybean (Pepol, 1989) due to irradiation.

The vigour index showed a maximum value of 25.57 (100 Gy) and minimum of 0.69 (500 Gy) in Paiyur 1. Percentage reduction of vigour index over the control revealed minimum reduction in 100 Gy and maximum reduction in 500 Gy. Highest and lowest vigour index was reported in 100 Gy (29.51) and 500 Gy (4.24) respectively in Goa cowpea 3. Percentage reduction of vigour index followed the similar trend as in Paiyur 1.

#### C. Effect of gamma irradiation on pollen fertility

The mean pollen fertility in both mutant populations and the percentage drop with its respective control are presented in Table 5. The control varieties Paiyur 1 and Goa cowpea 3 exhibited maximum pollen fertility of 92.95 and 91.53 per cent respectively. A gradual reduction in pollen fertility with increased dose of the mutagen has been observed. Pollen fertility was found to be maximum with lesser dose of 100 Gy (88.09 per cent in Paiyur 1 and 88.38 per cent in Goa cowpea 3) and minimum with higher dose 500 Gy (29.65 per cent in Paiyur 1 and 37.80 per cent in Goa cowpea 3) in both the varieties. The reduction in pollen fertility in terms of percentage ranged from 5.23 (100 Gy) to 68.10 per cent (500 Gy) in Paiyur 1 and from 3.45 (100 Gy) to 58.70 per cent (500 Gy) in Goa cowpea 3. The increased pollen survival in lesser dose has been mainly attributed to chromosomal interchange, least chromosomal aberration and gene mutation. Ramya *et al.* (2014), Kumar *et al.* (2009) recorded reduction in pollen fertility in comparison to the control.

#### D. Effect of gamma irradiation on other biometrical traits

The other biometrical traits *viz.* days to 50% flowering, days to maturity, plant height at maturity, number of primary branches per plant, number of pod clusters per plant, number of pods per cluster, pod length, hundred seed weight, seed yield per plant were recorded and the mean values were presented in Table 6 and 7. All the traits showed decrease in the mean values with increase in the dose of the mutagen in the M<sub>1</sub> generation.

The variability of quantitative characters influencing yield was much greater in mutagenic progenies than in control (Prasad, 1976). These mutagens' capacity to infiltrate living organisms' cells and interact with DNA results in the general harmful effects linked to their mutagenic capabilities. Thus their effects are mainly due to the direct interactions between the mutagen and the DNA molecules (Mensah *et al.*, 2007).

Mutagens have the potential to induce physiological functions, which often manifests growth retardation and

unrestricted cell death in M<sub>1</sub> generation (Mak *et al.*, 1986). This is consistent with recent research, which showed that gamma rays had an inhibitory influence on yield performance.

The height was measured at 30 days and at maturity. There was a reduction in the height observed with increasing mutagenic dose. The control heights were found to be 20.4 cm (30 days old seedling) and 72.5 cm (at maturity) for Paiyur 1. While for Goa cowpea 3 it was found to be 16.5cm (30 days old seedling) and 32.5 (at Maturity). Thereafter, heights of the plants reduced in both the cultivars.

The number of days to 50% flowering showed a declining trend in the selective doses over the control in both the varieties. Fifty per cent flowering was attained in 55 days in Paiyur 1 and 68 days in Goa cowpea 3 variety. All the investigated mutagenic treatments noticed lower mean plant heights than the control. The control Paiyur 1 and Goa cowpea 3 recorded mean plant height of 72.5 cm and 32.5 cm respectively. Also, number of primary branches per plant showed gradual reduction as compared to the control (Paiyur 1: 7.68 and Goa cowpea 3: 5.4). Similar results of reduction in the number of primary branches per plant in sesame in M<sub>1</sub> generation were reported by Prabhakar (1985).

Number of pod clusters per plant showed decreasing values with increase in the mutagenic dose as compared to control (Paiyur 1: 19.34 and Goa cowpea 3: 4.89). Number of pods per cluster showed a declining trend and ranging from 3.5 (150Gy) to 2 (250Gy) as compared to the two control varieties *viz.*, Paiyur 1 (4) and Goa cowpea 3 (3). Reduced pod count could be caused likely by the toxicity of the mutagen, inhibiting action of enzymes, and changes in enzyme activity. Pod length also showed a slight decline in the mean values varied from 14.18 cm (150Gy) to 12.11 cm (250Gy) in comparison with control (15.83 cm) in Paiyur 1 and from 18.5cm (350Gy) to 16 cm (450 Gy) compared to control (23.77cm) in Goa cowpea 3.

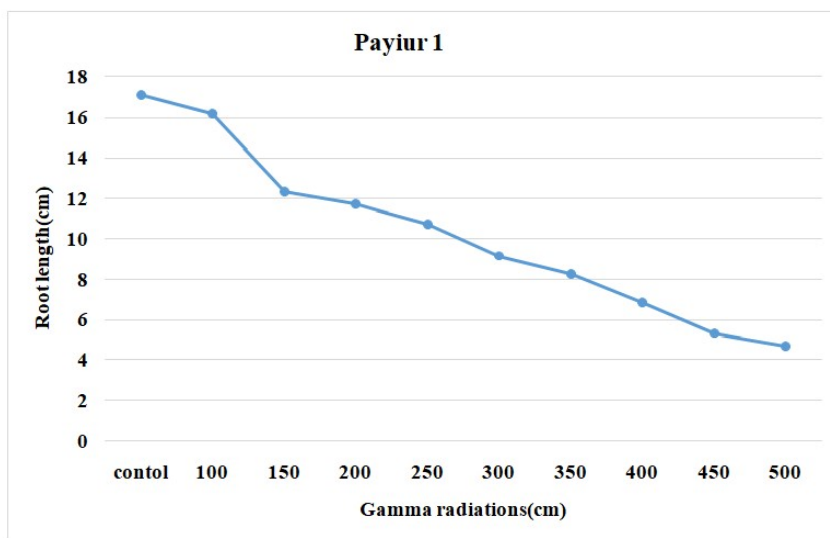
Number of seeds per pod did not show much variation in the three doses of Paiyur 1 variety as compared to control. The mean number of seeds per pod was 13 in all the studied doses. In Goa cowpea 3 variety, control had registered an average of 19 seeds per pod whereas, 350 Gy showed a mean of 10 seeds per pod and 400 Gy showed an average of 9 seeds per pod. Hundred seed weight also showed a similar kind of reduction over the control in both the varieties.

In all mutagenic treatments, the seed yield per plant showed a dose-dependent, negative, and linear relationship with the increased dose of mutagen. The reduction in this trait may also be attributed to the increase in seed sterility at higher doses of the treatment.

Similar results were observed in different crops by several mutagens. Banu *et al.* (2005) observed reduced seed yield per plant in combined treatments with gamma rays and EMS in *Solanum melongena* L.

**Table 5: Effect of gamma irradiation on pollen fertility in Paiyur 1 and Goa cowpea 3 varieties.**

| Treatment | Paiyur 1         |                       |                          | Goa cowpea 3     |                       |                          |
|-----------|------------------|-----------------------|--------------------------|------------------|-----------------------|--------------------------|
|           | Pollen fertility | Per cent over control | % reduction over control | Pollen fertility | Per cent over control | % reduction over control |
| 100 Gy    | 88.09            | 94.77                 | 5.23                     | 88.38            | 96.55                 | 3.45                     |
| 150 Gy    | 86.33            | 92.87                 | 7.12                     | 79.23            | 86.56                 | 13.44                    |
| 200 Gy    | 78.81            | 84.79                 | 15.21                    | 73.07            | 79.83                 | 20.17                    |
| 250 Gy    | 59.77            | 64.30                 | 35.70                    | 70.3             | 76.80                 | 23.19                    |
| 300 Gy    | 55.29            | 59.48                 | 40.52                    | 68.16            | 74.46                 | 25.54                    |
| 350 Gy    | 50.84            | 54.70                 | 45.30                    | 61.48            | 67.16                 | 32.83                    |
| 400 Gy    | 44.55            | 47.92                 | 52.07                    | 49.88            | 54.49                 | 45.50                    |
| 450 Gy    | 37.79            | 40.66                 | 59.34                    | 46.08            | 50.34                 | 49.66                    |
| 500 Gy    | 29.65            | 31.90                 | 68.10                    | 37.80            | 41.30                 | 58.70                    |
| Control   | 92.95            | 100                   | 0                        | 91.53            | 100                   | 0                        |
| Mean      | 59.01            |                       | 36.51                    | 63.82            |                       | 30.28                    |
| SE        | 7.06             |                       |                          | 5.50             |                       |                          |



**Fig. 5.** Dose response relationship for root length in Paiyur 1 cowpea varieties after treatment with different dose of gamma radiations.

**Table 6: Effect of gamma irradiation on various quantitative traits for Paiyur 1.**

| Treatment | Days to 50% flowering | Days to maturity | Plant Height (cm) |          | No. Of Primary Branches / Plant | % over control | No. Pod Clusters / Plant | % over control | No. Of Pods / Cluster | % over control | Pod Length (cm) | % over control | No. Of seeds / pod | % over control | 100 Seed Weight (g) | % over control | Seed Yield / Plant (g) | % over control |
|-----------|-----------------------|------------------|-------------------|----------|---------------------------------|----------------|--------------------------|----------------|-----------------------|----------------|-----------------|----------------|--------------------|----------------|---------------------|----------------|------------------------|----------------|
|           |                       |                  | 30 DAS            | Maturity |                                 |                |                          |                |                       |                |                 |                |                    |                |                     |                |                        |                |
| 150 Gy    | 48                    | 83               | 14.8              | 69       | 5.83                            | 75.91          | 15.1                     | 78.07          | 3.5                   | 87.5           | 14.18           | 89.57          | 13                 | 100            | 8.68                | 87.67          | 25.6                   | 78.77          |
| 200 Gy    | 52                    | 88               | 12.58             | 68.47    | 4.46                            | 58.07          | 14.65                    | 75.75          | 2                     | 50             | 13.72           | 86.67          | 13                 | 100            | 8.5                 | 85.85          | 21.47                  | 66.06          |
| 250Gy     | 53                    | 90               | 11.6              | 66.34    | 3.17                            | 41.27          | 12.88                    | 66.60          | 2                     | 50             | 12.11           | 76.50          | 13                 | 100            | 8.4                 | 84.84          | 19.53                  | 66.09          |
| Control   | 55                    | 90               | 20.4              | 72.5     | 7.68                            | 100            | 19.34                    | 100            | 4                     | 100            | 15.83           | 100            | 14                 | 100            | 9.9                 | 100            | 32.5                   | 100            |
| Mean      | 51                    | 87.75            | 12.99             | 6.94     | 4.48                            |                | 14.21                    |                | 2.5                   |                |                 |                | 13                 |                | 8.53                |                | 22.2                   |                |
| SE(m)     | 1.53                  |                  | 0.9               | 0.81     | 0.77                            |                | 0.67                     |                | 0.5                   |                |                 |                | 0                  |                | 0.081               |                | 1.79                   |                |

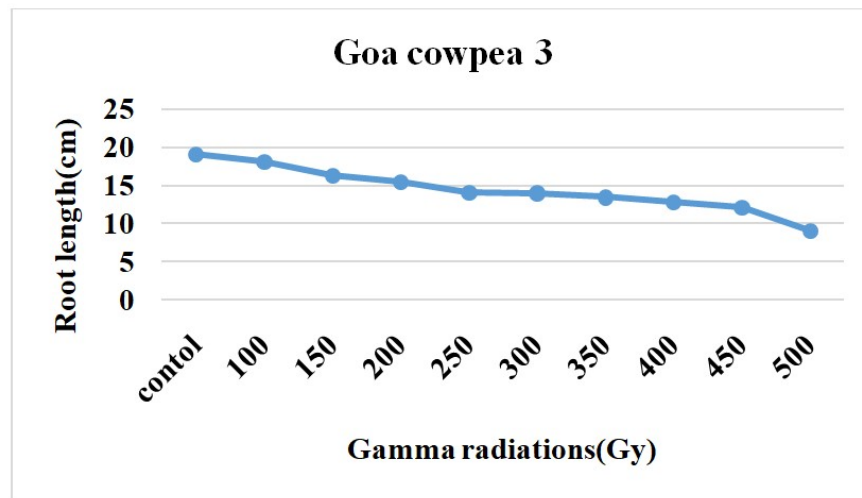


Fig. 6. Dose response relationship for root length in Goa cowpea variety after treatment with different dose of gamma radiations.

Table 7: Effect of mutagenic treatment on various quantitative traits for Goa cowpea 3.

| +       | Days to 50% flowering | Days to maturity | Plant Height (cm) |          | No. Of Primary Branches / Plant | % over control | No. Pod Clusters / Plant | % over control | No. Of Pods / Cluster | % over control | Pod Length (cm) | % over control | No. Of seeds / pod | % over control | 100 Seed Weight (g) | % over control | Seed Yield / Plant (g) | % over control |
|---------|-----------------------|------------------|-------------------|----------|---------------------------------|----------------|--------------------------|----------------|-----------------------|----------------|-----------------|----------------|--------------------|----------------|---------------------|----------------|------------------------|----------------|
|         |                       |                  | 30 DAS            | Maturity |                                 |                |                          |                |                       |                |                 |                |                    |                |                     |                |                        |                |
| 350 Gy  | 60                    | 96               | 8.77              | 22       | 4.89                            | 90.56          | 3.65                     | 74.64          | 2.6                   | 86.67          | 18.5            | 77.83          | 10                 | 52.63          | 22.58               | 98.60          | 29.8                   | 97.07          |
| 400 Gy  | 64                    | 100              | 7.5               | 21.5     | 4.5                             | 83.33          | 3                        | 61.35          | 2                     | 66.67          | 17.5            | 73.62          | 9                  | 47.37          | 20.5                | 89.51          | 25.4                   | 82.74          |
| 450Gy   | 65                    | 102              | 5.9               | 20.2     | 3.2                             | 59.26          | 2.4                      | 49.08          | 1                     | 33.33          | 16              | 67.31          | 10                 | 52.63          | 18.9                | 82.53          | 22.9                   | 74.6           |
| Control | 68                    | 106              | 16.5              | 32.5     | 5.4                             | 100            | 4.89                     | 100            | 3                     | 100            | 23.77           | 100            | 19                 | 100            | 22.90               | 100            | 30.7                   | 100            |
| Mean    | 63                    | 101              | 7.39              | 21.23    | 4.19                            |                | 3.02                     |                | 1.86                  |                | 17.33           |                | 9.67               |                | 20.66               |                | 26.03                  |                |
| SE(m)   | 1.53                  |                  | 0.83              | 0.54     | 0.51                            |                | 0.36                     |                | 0.47                  |                | 0.73            |                | 0.33               |                | 1.06                |                | 2.01                   |                |

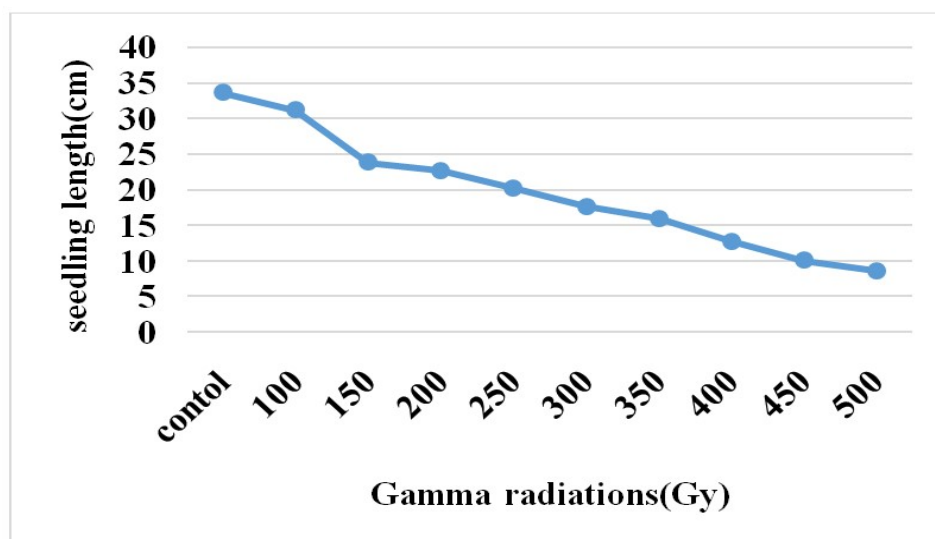
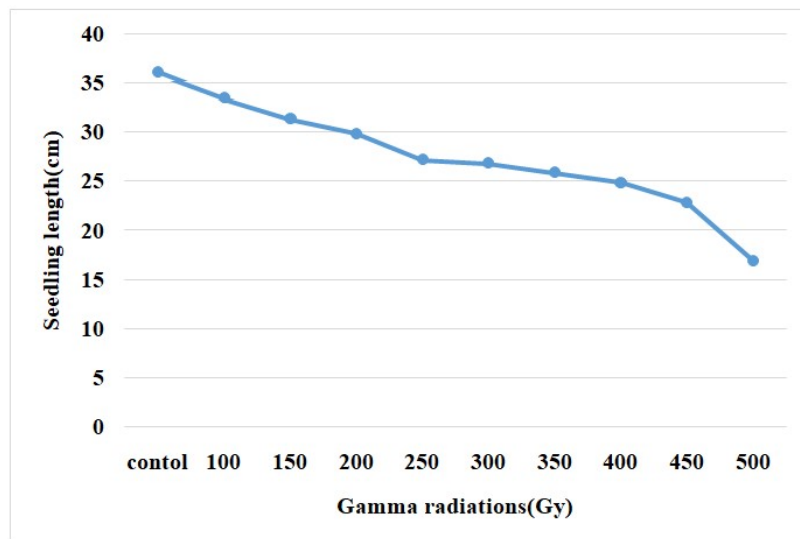
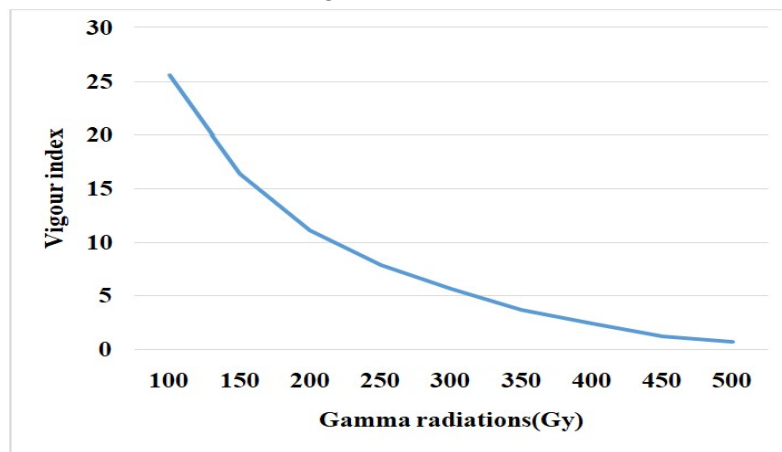


Fig. 7. Dose response relationship for seedling length in Paiyur 1 cowpea variety after treatment with different dose of gamma radiations.

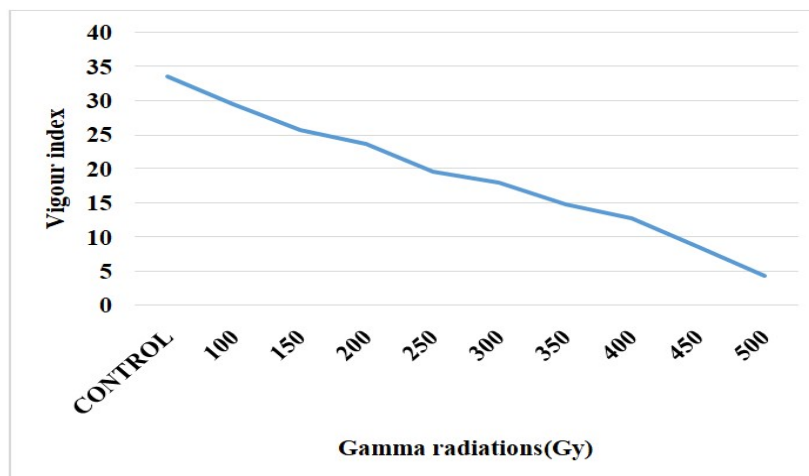




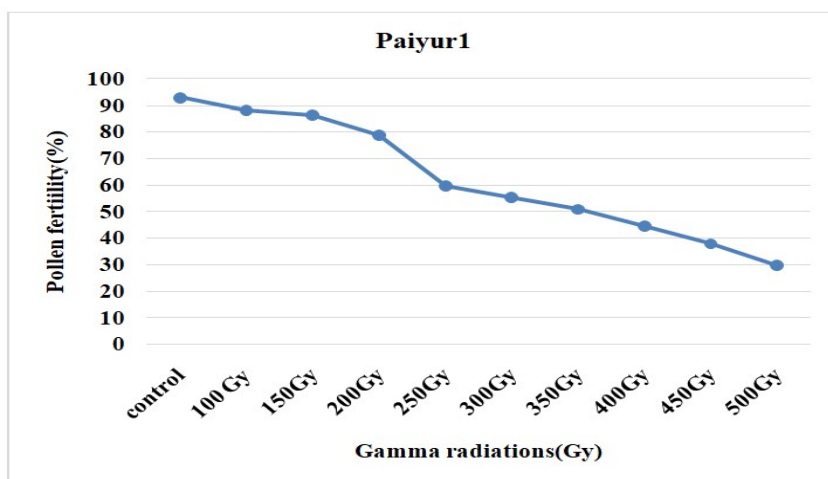
**Fig. 8.** Dose response relationship for seedling length in Goa cowpea 3 cowpea variety after treatment with different dose of gamma radiations.



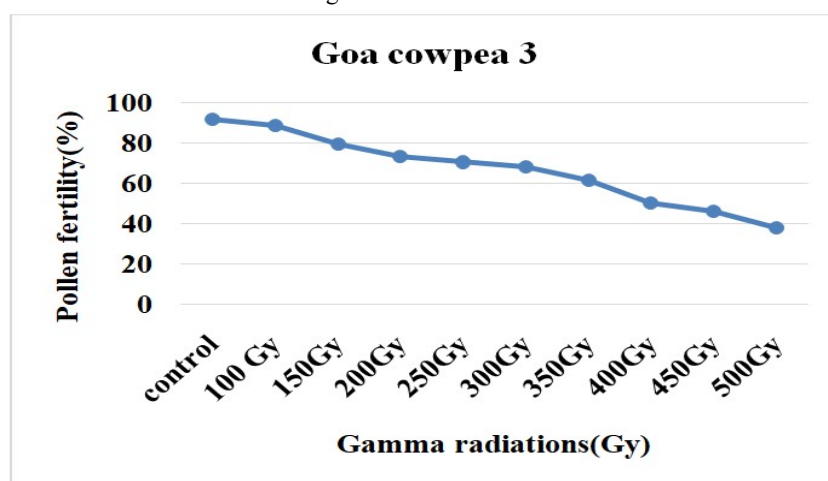
**Fig. 9.** Dose response relationship for vigour index in Paiyur 1 cowpea variety after treatment with different dose of gamma radiations



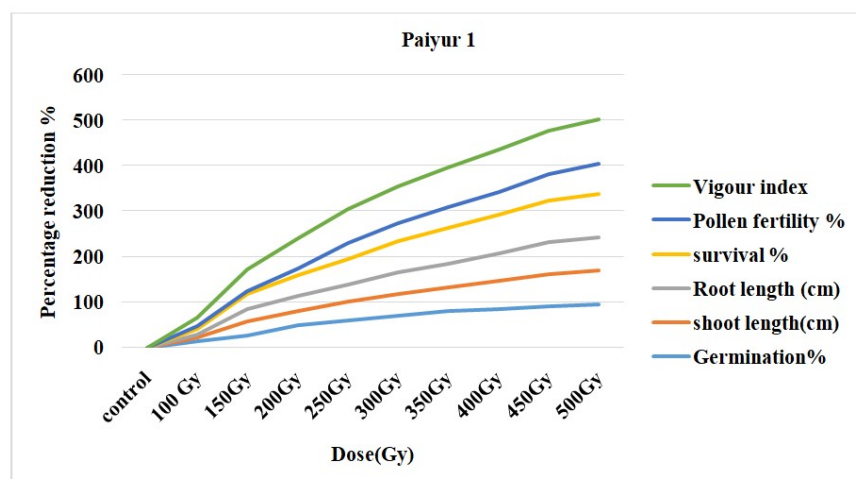
**Fig. 10.** Dose response relationship for vigour index in Goa cowpea 3 variety after treatment with different dose of gamma radiations.



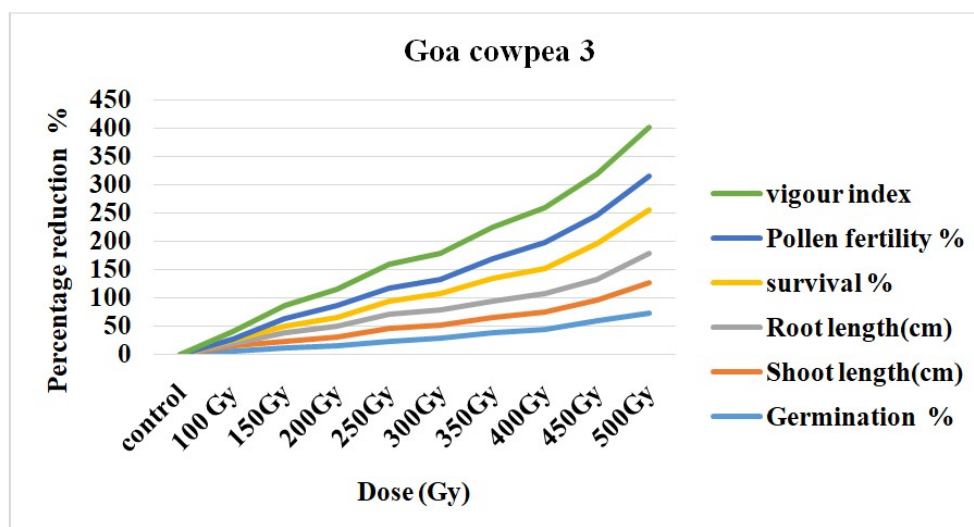
**Fig. 11.** Dose response relationship for pollen fertility in cowpea variety Paiyur 1 after treatment with different doses of gamma irradiation.



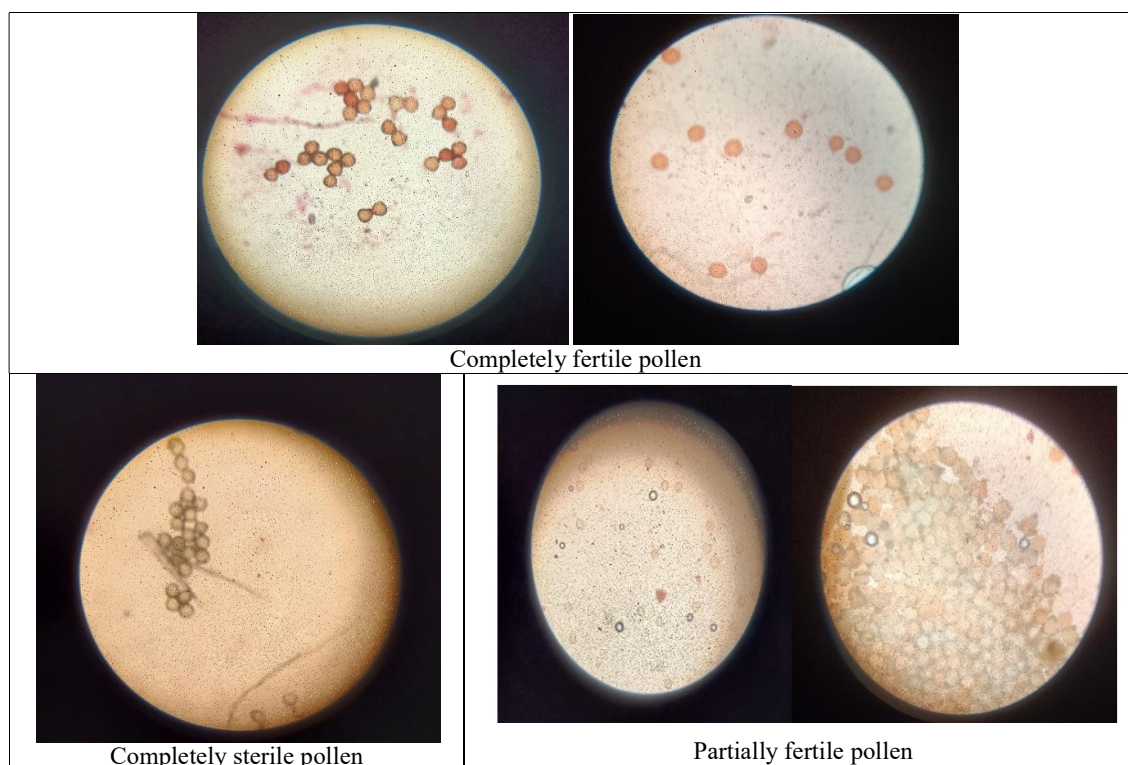
**Fig. 12.** Dose response relationship for pollen fertility in cowpea variety Goa cowpea 3 after treatment with different doses of gamma irradiation.



**Fig. 13.** Effect of gamma irradiation on six quantitative traits in  $M_1$  generation of Paiyur 1 cowpea.



**Fig. 14.** Effect of gamma irradiation on six quantitative traits in  $M_1$  generation of Goa cowpea 3.



**Fig. 15.** Pollen grains complete fertile, partial fertile and sterile nature due to the effect of gamma irradiation.

All the morphological characters of  $M_1$  generation showed a decline with increasing dose of the mutagenic treatment compared to control. The quantitative characters gradually increased with increasing dose of the mutagen. The maximum reduction of quantitative characters was observed at 250 Gy in Paiyur1 and at 450 Gy in Goa cowpea 3.

The limited morphological differences may result from physiological and other genetic disruptions such as chromosomal damage, altered coiling, failure, or restricted pairing of chromosomes. Such results were earlier reported in linseed (Rai, 1978), green gram (Koteswara Rao *et al.*, 1983), cowpea (Odeigah *et al.*, 1998) and niger (Naik and Murthy, 2009) crops.

## CONCLUSIONS

Effect and susceptibility of the two cowpea varieties viz., Paiyur 1 and Goa cowpea 3 for gamma irradiation showed altered / modified phenotypes observed in the M<sub>1</sub> generation of mutagen-treated plants. The genetic materials of the two cowpea varieties were severely harmed by the gamma irradiation. From the foregoing discussion, it was concluded that, all the quantitative characters showed a decreasing trend with increasing dose of the mutagen. They exhibited a negative and linear relationship with increasing mutagenic dose.

## FUTURE SCOPE

There is limitation in cowpea production due to indeterminate growth habit, asynchronous pod maturity, shedding of newly formed pods, presence of anti-nutritional factors, low yielding and long duration of the crop. Due to its indeterminate growth continuous flower production is there, which leads to continuous pod formation at different times. As a result, multiple harvesting is required (2-3 pickings). This demands for more labour and increases the cost of cultivation. Thus, mutation can be focussed on identifying and developing cowpea lines with determinate growth habit which will be amenable for mechanical harvesting and help in increasing the income of the farmers.

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**Conflict of interest.** None.

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