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Research Article

Mutagenic effectiveness, efficiency and dose optimization of gamma rays in papaya (*Carica papaya* L.) varieties

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Abstract
The present study was conducted to determine the mutagenic effectiveness, efficiency and lethal dose of gamma rays in papaya varieties CO7, Arka Prabhath and Arka Surya. Papaya seeds were exposed to various doses of gamma rays (0Gy, 100Gy, 150Gy, 200Gy, 250Gy, 300Gy, 350Gy, 400Gy, 450Gy and 500Gy) and the mutagenic effectiveness and efficiency of gamma rays were assessed based on the chlorophyll mutation frequency and biological damage in M_1 plant basis. There was a negative relationship between mutagenic effectiveness and efficiency with the doses of gamma irradiation. LD_{50} value was optimized in CO7, Arka Prabhath and Arka Surya as 354, 348 and 341Gy respectively. Generally, the germination, survival and growth rate of papaya decreased with an increase in gamma-irradiation doses. Lower doses of mutations created very few mutations and higher doses of gamma irradiation induced lethality and sterility of seedlings whereas, lethal dose 50 has proved to be more effective in inducing desirable mutations in papaya. Since identifying the mutagenic efficiency and effectiveness of the mutagen and optimization of lethal dose is the preliminary step in any mutagenic experiment, this study will offer a foundation for further gamma irradiation studies in papaya to develop desirable mutants.

Keywords: Papaya, Gamma rays, Mutagenic efficiency, Mutagenic effectiveness, LD_{50}

INTRODUCTION

Papaya (*Carica papaya* L.) is a semi-perennial herbaceous plant of the family Caricaceae believed to have originated from the Caribbean coast of Central America and introduced to India during the 16th century. This super fruit is also known as 'pawpaw' and is cultivated throughout the tropics and subtropics due to its wide ecological adaptability, easiness in cultivation, high palatability and year-round production. This crop is identified as relatively rich in vitamin A (2020 IU), vitamin C (46 mg/100g), vitamin B1(40 mg/100g), riboflavin, iron, calcium, dietary fibers and low in calories (32 kcal/100g) (Dinesh, 2010) with a plethora of uses in food, medicine, and industries.

Papaya is the fourth most important fruit crop worldwide and gained importance among farmers because of the early bearing capacity, higher productivity per unit area and fair economic returns. India leads in papaya production with 5988.8 thousand metric tonnes in an area of 138.4 thousand hectares with a productivity of 43.3 metric tonnes per hectare. The papaya export from India is 9.99 thousand metric tonnes which are worth of 4,150.44 Lakh Rupees (NHB, 2017-18). Despite its increase in demand and area, the production is not proportionally increasing because of yield loss due to various diseases. Among them, papaya ringspot is the most devastating disease which is caused by a potyvirus "*Papaya ringspot virus* (PRSV-P)" and is transmitted by aphids. Plants that are infected at early growth stages do not produce fruits and infection at later growth stages decreases the production and quality of fruits (Gonsalves *et al.*, 1998). No papaya varieties with resistance to this disease have been developed so far.

Traditional plant breeding programs require sufficient genetic variability in the gene pool for improvement of specific traits like disease resistance. Sources of disease resistance may be available in the wild relatives, however introgression of such traits into the cultivated varieties is difficult and time consuming. Trait improvement through mutation breeding is an innovative approach to generate new variations in the elite genotype without altering the genetic background of the crop. Induced mutagenesis is routinely used in crop improvement programs for generating novel variants. Among various radiation sources, gamma rays are very important in creating genetic variability through mutagenesis (Hong *et al*., 2022). Induced mutagenic studies have been conducted in different fruit crops to improve traits like disease resistance, dwarf stature and seedless fruits, nevertheless limited works have been reported in papaya (Pujar *et al.,* 2019; Sahu *et al*., 2019).

The selection of effective and efficient mutagen is of paramount importance in any mutagenic experiments to obtain promising mutations (Kumar *et al*., 2021). The usefulness of mutagen depends on the spectrum of chlorophyll mutations, lethality, sterility, mutagenic efficiency and effectiveness of the mutagen. Futhermore, reports on mutagenic efficiency in fruit crops are very few and no such studies have so far been done on papaya. Various studies have reported that more desirable mutations occur at the dose which causes the death of 50% of organisms or 50% of growth reduction (Alvarez-Holguín *et al*., 2019). Therefore, optimization of LD_{50} (lethal dose 50) is very important for any mutation breeding experiment. LD_{50} is the dosage of mutagen which causes 50 % lethality in the organism and changes with the species, the nature of the plant material and the stage of the crop. The present work was carried out with a view to study the biological responses of papaya to gamma irradiation and to determine the mutagenic efficiency, effectiveness and optimal dose (LD_{50}) of gamma rays in papaya varieties CO 7, Arka Prabhath and Arka Surya which are popular gynodioecious varieties cultivated in South India.

MATERIALS AND METHODS

Three popular gynodioecious papaya varieties namely CO 7, Arka Prabhath and Arka Surya were selected for the present study. The CO 7 seeds were collected from Horticultural College and Research Institute, Tamil Nadu Agricultural University, Coimbatore while seeds of Arka Prabhath and Arka Surya were collected from the Indian Institute of Horticultural Research, Bangalore. The dry papaya seeds of these varieties were exposed to nine different doses of gamma rays ranging from 100 to 500 Gy using ^{60}Co as a gamma source. Gamma-ray irradiation was performed at the gamma chamber facility of the Radiological Safety Division, Indira Gandhi Centre for Atomic Research, Department of Atomic Energy, Kalpakkam, Tamil Nadu, India. The irradiated seeds were sown in polybags filled with a mixture of red soil: FYM: sand in a ratio of 1:1:1 and maintained under controlled conditions for germination. The germination percentage was calculated after four weeks of sowing and growth parameters such as seedling height, seedling girth, number of leaves, petiole length and survival percentage were recorded after six weeks of sowing. Biological damages like lethality, injury and the occurrence of chlorophyll mutants were recorded in M_1 generation. The chlorophyll mutants were characterised and the mutagenic effectiveness and efficiency of gamma rays were determined using the formula given below.

$$\text{Mutation frequency } (M_1 \text{ plant basis}) = \frac{\text{Number of chlorophyll mutants} \times 100}{\text{Total number of plants}}$$

$$\text{Mutagenic effectiveness} = \frac{\text{Mutation frequency} \times 100}{\text{Dose of mutagen (Gy)}}$$

$$\text{Mutagenic efficiency} = \frac{\text{Mutation frequency} \times 100}{\text{Percent lethality (L) or Percent injury (I) or Percent sterility (S)}}$$

The experiment was conducted as a completely randomized design with three replications and 10 treatments *viz*., 0Gy, 100Gy, 150Gy, 200Gy, 250Gy, 300Gy, 350Gy, 400Gy, 450Gy and 500Gy. A total of 30 seeds per replication was irradiated with gamma rays. The lethal dose (LD_{50}) values of gamma irradiation in three papaya varieties were determined based on the germination per cent using Probit analysis. Data were subjected to the standard analysis of variance procedure using AGRES statistical computing software to test the significance of observed plant responses at different treatments.

RESULTS AND DISCUSSION

Chlorophyll mutants are not desirable in crop improvement programs but serve as an important parameter to determine the mutagenic efficiency (Eswaramoorthy *et al*., 2021). Several forms of chlorophyll mutations such as albino, xantha, viridis, maculata, striata, and chlorina have been reported in numerous crops using physical and chemical mutagens. In the present study, a wide spectrum of chlorophyll mutants such as xantha, chlorina,striata,virescent viridis and albino were observed in M_1 generation (**Fig.1**). Most of the chlorophyll deficient mutants died after a few days of germination whereas

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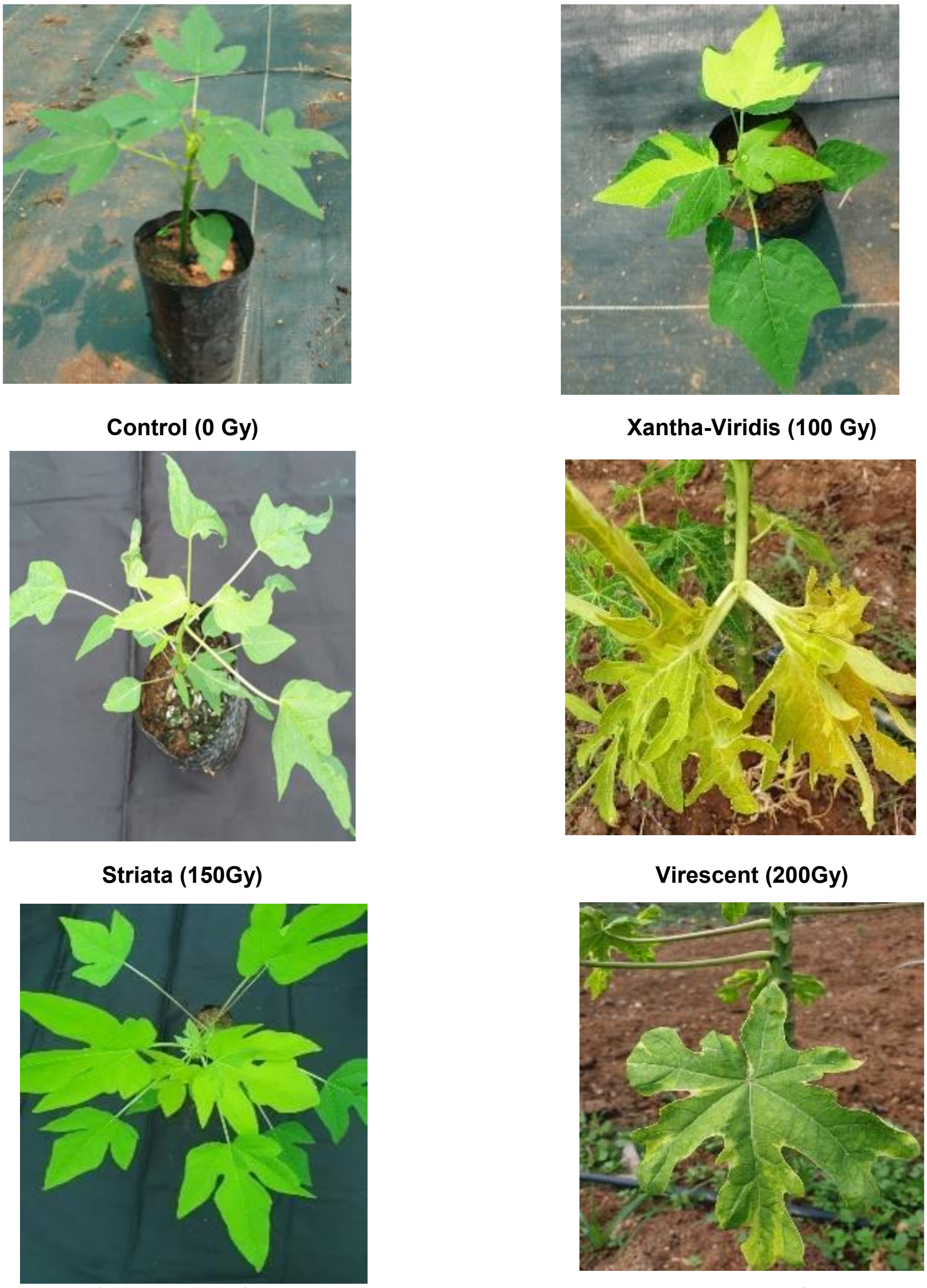

Fig.1. Spectrum of chlorophyll mutants in M_1 generation of papaya

some showed retarded growth because of the deficiency of chlorophyll. The average mutation frequencies of papaya seedlings when exposed to various gamma ray doses were 3.75, 4.05 and 4.11 in CO 7, Arka Prabhath and Arka Surya, respectively (**Table 1**). The total frequency of chlorophyll phenodeviants was found higher in Arka Surya followed by Arka Prabhath and lower in CO 7 variety. The occurrence of chlorophyll mutations in M_1 generation varied with genotypes and mutagen doses. Chlorina mutant was observed with maximum frequency whereas albino showed lower frequency which failed to survive after 10 days of germination. Similar findings were reported by Nura *et al*. (2021). In this study, no definite trend was observed in the occurrence of chlorophyll mutants in a dose based manner in any of the three varieties. The occurrence of chlorphyll mutants might be attributed to a variety of factors, including defective chlorophyll biosynthesis, chlorophyll degradation, and carotenoid deficiency (Goyal *et al*., 2019). Chlorophyll mutations are crucial for determining gene function as well as understanding chlorophyll metabolism and regulation in plants (Dwivedi *et al*., 2021). Hence chlorophyll mutations can be used as the most reliable marker for assessing the genetic impact of mutagenic treatments in different crops.

The results on the mutagenic efficiency and effectiveness of various gamma irradiation doses in three papaya varieties are presented in **Table 1**. Mutagenic efficiency is referred to as the actual ratio of the rate of mutations exhibited to the biological impairments that occurred in the plant population. Hence it is an important tool to determine the impact of a mutagen in biological system. Biological damage in response to gamma irradiation was determined in terms of lethality (reduction in germination), injury (reduction in shoot length) and sterility (reduction in pollen viability). Dutta *et al*. (2021) stated that the impact of a mutagen could be identified in the M1 generation in terms of lethality, injury and sterility. Therefore, the mutagenic efficiency and effectiveness were determined in M1 plant basis. High mutagenic efficiency in terms of lethality was found in 300 Gy (72.94), 250Gy (90.11) and 200Gy (90.76) in CO 7, Arka Prabhath and Arka Surya respectively. The least mutagenic efficiency was found at 500 Gy in all papaya varieties studied, since lethality was found to be maximum at this dose. The mutagenic efficiency consistently decreased with an increase in dosages of gamma irradiation. This is in close agreement with the reports of Dutta *et al*. (2021) in bitter gourd. Tamilzharasi *et al*. (2021) reported that there was a substantial positive correlation between mutation frequency and biological damage in the M1 generation when exposed to gamma radiation in blackgram. Mutagenic effectiveness refers to the rate at which a mutation is induced as a function of the mutagenic dose. The estimates of effectiveness of gamma rays ranged from 1.11 to 2.59, 1.0 to 2.96 and 1.0 to 2.08 in Arka Surya, Arka Prabhath and CO 7 respectively (**Table 1**). A critical examination of the data obtained in this study showed that the higher mutagenic efficiency and effectiveness coincide with the LD50 value. The assortment of effective mutagen and the dosages are crucial in the success of any breeding program. Hence it can be concluded that the lethal 50 doses of gamma rays may be preferable for effective mutation breeding in papaya in order to isolate beneficial mutants as it may cause less biological injuries.

The tolerance level of papaya seeds to gamma irradiation was studied in the M1 generation in terms of reduction in germination, survival and seedling growth characteristics. The seed germination percentage is an important variable in mutation study since it signifies the effectiveness of the mutagen. Results of gamma irradiation in papaya showed a significant difference in the germination rate among different treatments (**Table 2**). In general, maximum germination was observed in control and there is a gradual reduction in germination with the increase in irradiation doses in all three varieties. Germination percentage showed a broad range of variation from 70 % to 3.33% in Arka Surya. The highest germination (68.67%) was found in 100Gy followed by 150Gy among the irradiation treatments. Maximum germination percentage in Arka Prabhath (73.33%) and CO 7 (78.67 %) was recorded in 150 Gy. There was a regressing trend in germination above 150 Gy in CO 7 and Arka Prabhath. Arka Surya exhibited a steady declining pattern with a rise in doses. It can be concluded that the percentage of seed germination exhibited a gradual reduction with an increase in doses of gamma rays and higher doses above 400Gy were lethal in papaya varieties. Similar results were elicited in pummelo (Sankaran *et al*., 2021), rough lemon (Kaur and Rattanpal, 2010), barnyard millet (Ramesh *et al*., 2019) and blackgram (Veni *et al*., 2017). According to Mahadevamma *et al*. 2012, seed germination decreased with increased dosages up to 20 kR and the lethal dose in Sunrise Solo and Coorg Honey papaya was 9.5 kR and 7.5 kR respectively. Yadav *et al*. (2016) inferred that gamma irradiation of Papaya cv. Kesar King resulted in increased germination percentage, survival percentage, and plant growth in response to irradiation up to 10 kR and was reduced with further increase in doses. The adverse effect of gamma rays on seed germination might be due to the change in physiological and biological processes within the plants such as inhibition of DNA synthesis (Jahan *et al*., 2021) or suppression of enzyme synthesis (Kurobane and Yamaguchi, 1978). Yusuf and Nair (1974) explained that gamma irradiation impaired the synthesis of the enzyme involved in the formation of IAA from tryptophan as well as accelerated the degradation of the enzyme thus decreasing seed germination.

The plant survival percentage of gamma-ray exposed seeds along with the control was studied after 6 weeks of sowing. The highest survival percentage was observed in control in all the varieties followed by the treatment 150 Gy in CO 7 and Arka Prabhath and 100 Gy in Arka Surya **(Table 1**). The maximum survival percentage observed among irradiated treatments was 93.22% (150Gy), 93.64% (150Gy) and 90.3% (100Gy) in CO 7, Arka Prabhath and Arka Surya sequentially. Arka Surya exhibited the lowest survival per cent of irradiated plants with a range of 0 to 90.3%. There was an exorbitant lethality observed in higher doses at 400 Gy and no seedlings survived at 500 Gy in Arka Prabhath and Arka Surya. Only a few survived in CO 7. Even though the seeds exposed to higher doses of gamma irradiation germinated, most of them failed to survive after a few days. Significant differences in survival rate can be observed among different treatments in all three varieties. There was a declining trend in the survival of plants with an increase in doses of gamma rays. These results are in accordance with the reports in acid

Table 1. Mutation frequency, mutagenic effectiveness and efficiency in M_1 generation for three papaya varieties exposed to different doses of gamma rays

CO 7

Doses (Gy)	Total Number of plants	Number of chlorophyll mutants					MF	SP	SRP	ME	MEF
		Xantha	Viridis	Chlorina	Albino	Total					
100	66	1	-	-	-	1	1.52	96.00	5.09	29.76	1.52
150	67	-		1	-	1	1.49	95.45	2.78	53.69	1.00
200	63	1	1	-	-	2	3.17	95.65	5.52	57.47	1.59
250	60	1	-	1	-	2	3.33	90.48	7.73	43.10	1.33
300	48	-	1	2	-	3	6.25	90.00	8.57	72.94	2.08
350	42	-	-	1	1	2	4.76	89.38	11.00	43.29	1.36
400	30	-	2	-	-	2	6.67	86.67	18.22	36.59	1.67
450	20	-	-	1	-	1	5.00	80.00	40.00	12.50	1.11
500	19	-	-	-	1	1	5.26	77.78	40.44	13.01	1.05

ArkaPrabhath

Doses (Gy)	Total Number of plants	Number of chlorophyll mutants					MF	SP	SRP	ME	MEF
		Xantha	Viridis	Chlorina	striata	Total					
100	65	1	-	-	-	1	1.54	95.65	5.20	29.56	1.54
150	66	-	-	-	1	1	1.52	90.45	2.02	75.16	1.01
200	55	-	1	2	-	3	5.45	93.64	6.18	88.28	2.73
250	54	1	-	1	2	4	7.41	89.47	8.22	90.11	2.96
300	50	-	-	2	-	2	4.00	87.43	10.65	37.55	1.33
350	40	1	-	-	1	2	5.00	85.00	12.32	40.59	1.43
400	25	1	-	-	-	1	4.00	83.33	52.80	7.58	1.00
450	20	-	1	-	-	1	5.00	42.86	62.32	8.02	1.11
500	15	1	-	-	-	1	6.67	33.33	95.65	6.97	1.33

Arka Surya

Doses (Gy)	Total Number of plants	Number of chlorophyll mutants					MF	SP	SRP	ME	MEF
		Xantha viridis	Virescent	Striata	Chlorina	Total					
100	62	1	-	-	-	1	1.61	93.3	3.04	53.02	1.61
150	60	-	-	-	1	1	1.67	90.3	4.66	35.74	1.11
200	58	-	1	2	-	3	5.17	88.7	5.70	90.76	2.59
250	54	1	-	-	1	2	3.70	87.6	10.00	37.04	1.48
300	51	1	-	-	1	2	3.92	83.3	10.98	35.71	1.31
350	40	1	-	1	-	2	5.00	82.4	11.14	44.88	1.43
400	16	-	1	-	-	1	6.25	82.2	61.59	10.15	1.56
450	15	-	-	1	-	1	6.67	31.7	67.33	9.90	1.48
500	14	-	1	-	-	1	7.14	26.0	93.33	7.65	1.43

MF- Mutation frequency, SP-Survival percentage, SRP- Survival reduction percentage, ME-Mutagenic efficiency, MEF-Mutagenic effectivenes

lime (Devi *et al*., 2021) and sorghum (Wanga *et al*., 2020). Higher doses of gamma irradiation induce physiological changes in the cells which cause growth reduction and lethality of seedlings. Higher doses of gamma irradiation initiate a G2-phase cell cycle arrest that pauses cell division and hampers growth (Hong *et al.,* 2022).

Determination of the lethal dose (LD50) is of prime importance in framing a mutagenic experiment. The germination percentage is one of the imperative variables since it signifies the magnitude of damage caused as a result of gamma irradiation. The LD50 value of gamma rays for papaya varieties CO 7, Arka Prabhath and

Table 2. Probit analysis of three papaya varieties exposed to different gamma-irradiation doses

CO 7

Dose (Gy)	Log_{10} of doses	Germination percentage	Percent reduction over control	Observed mortality percentage	corrected mortality percentage	Empirical probit units	LD_{50} value
0(control)		83.33	-	16.67	0.00		
100.00	2.00	73.33	10.00	26.67	12.00	3.83	
150.00	2.18	78.67	4.66	21.33	5.60	3.41	354.67
200.00	2.30	70.00	13.33	30.00	16.00	4.01	
250.00	2.40	65.33	18.00	34.67	21.60	4.21	
300.00	2.48	61.00	22.33	39.00	26.80	4.38	
350.00	2.54	53.33	30.00	46.67	36.00	4.64	
400.00	2.60	30.00	53.33	70.00	64.00	5.36	
450.00	2.65	26.67	56.66	73.33	68.00	5.47	
500.00	2.70	12.00	71.33	88.00	85.60	6.06	

Arka Prabhath

Dose (Gy)	Log_{10} of doses	Germination percentage	percent reduction over control	Observed mortality percentage	corrected mortality percentage	Empirical probit units	LD_{50} value
0(control)		76.67	-	23.33	0.00		
100.00	2.00	72.23	4.44	27.77	5.78	3.43	
150.00	2.18	73.33	3.34	26.67	4.35	3.29	
200.00	2.30	63.33	13.34	36.67	17.39	4.06	
250.00	2.40	61.00	15.67	39.00	20.43	4.17	
300.00	2.48	60.00	16.67	40.00	21.74	4.22	348.49
350.00	2.54	52.00	24.67	48.00	32.17	4.54	
400.00	2.60	23.33	53.34	76.67	69.57	5.51	
450.00	2.65	20.00	56.67	80.00	73.91	5.64	
500.00	2.70	10.00	66.67	90.00	86.96	6.12	

Arka Surya

Dose (Gy)	Log_{10} of doses	Germination percentage	percent reduction over control	Observed mortality percentage	corrected mortality percentage	Empirical probit units	LD_{50} value
0(control)		70.00	-	30.00	0.00		
100.00	2.00	68.67	1.33	31.33	1.90	2.93	
150.00	2.18	67.67	2.33	32.33	3.33	3.17	
200.00	2.30	62.00	8.00	38.00	11.43	3.80	
250.00	2.40	60.00	10.00	40.00	14.29	3.93	
300.00	2.48	56.67	13.33	43.33	19.05	4.12	341.27
350.00	2.54	48.67	21.33	51.33	30.48	4.49	
400.00	2.60	21.00	49.00	79.00	70.00	5.52	
450.00	2.65	16.67	53.33	83.33	76.19	5.71	
500.00	2.70	3.33	66.67	96.67	95.24	6.67	

Table 3.Effect of gamma irradiation on seedling growth parameters in M1 generation of three papaya varieties.

Doses (Gy)	Arka Prabhath								Arka Surya								CO 7							
	SH	SH % OC	SG	SG % OC	NL	NL % OC	PL	PL % OC	SH	SH % OC	SG	SG % OC	NL	NL % OC	PL	PL % OC	SH	SH % OC	SG	SG % OC	NL	NL % OC	PL	PL % OC
0	13.10	100	1.70	100	11.00	100	5.70	100.	14.80	100.0	1.90	100.0	11.00	100.0	6.10	100	15.00	100.0	1.80	100.0	11.00	100	6.50	100.00
100	12.50	95.4	1.60	94.1	11.00	100.0	5.10	89.4	13.00	87.84	1.50	78.95	10.00	90.9	6.00	98.36	15.10	100.6	1.90	105.5	10.00	90.9	6.20	95.38
150	14.50	110.6	1.80	105.8	12.00	109.0	5.80	101.7	11.00	74.32	1.50	78.95	10.00	90.9	5.80	95.08	15.90	106.0	2.00	111.1	12.00	109.0	6.70	103.08
200	10.30	78.63	1.50	88.24	10.00	90.91	5.00	87.72	8.20	55.41	1.40	73.68	8.00	72.73	5.50	90.16	13.50	90.00	1.70	94.44	10.00	90.9	5.80	89.23
250	8.40	64.12	1.30	76.47	9.00	81.82	4.50	78.95	6.90	46.62	1.20	63.16	9.00	81.82	5.00	81.97	8.20	54.67	1.30	72.22	9.00	81.82	5.40	83.08
300	6.70	51.15	1.10	64.71	8.00	72.73	4.00	70.18	6.10	41.22	1.10	57.89	8.00	72.73	4.10	67.21	8.00	53.33	1.20	66.67	9.00	81.82	4.90	75.38
350	5.10	38.93	0.90	52.94	7.00	63.64	3.60	63.16	5.30	35.81	0.90	47.37	7.00	63.64	3.50	57.38	6.10	40.67	1.00	55.56	8.00	72.73	4.10	63.08
400	5.00	38.17	0.80	47.06	6.00	54.55	3.20	56.14	4.90	33.11	0.80	42.11	5.00	45.45	3.10	50.82	5.50	36.67	1.00	55.56	6.00	54.55	4.00	61.54
450	4.20	32.06	0.70	41.18	5.00	45.45	3.00	52.63	4.10	27.70	0.50	26.32	5.00	45.45	2.90	47.54	5.10	34.00	0.90	50.00	6.00	54.55	3.30	50.77
500	4.00	30.53	0.50	29.41	5.00	45.45	2.60	45.61	4.10	27.70	0.40	21.05	4.00	36.36	2.50	40.98	4.80	32.00	0.80	44.44	5.00	45.45	3.20	49.23
Mean	8.38		1.19		8.40		4.25		7.84		1.12		7.7		4.45		9.72		1.36		8.6		5.01	9.72
SEd	0.25		0.01		0.20		0.09		0.22		0.03		0.14		0.10		0.25		0.03		0.19		0.12	0.25
CD (0.5%)	0.52		0.03		0.44		0.20		0.46		0.06		0.30		0.23		0.52		0.06		0.40		0.25	0.52

SH-Seedling height (cm), SG-Seedling girth (cm), NL-Number of leaves, PL-Petiole length (cm), % OC – Per cent over control

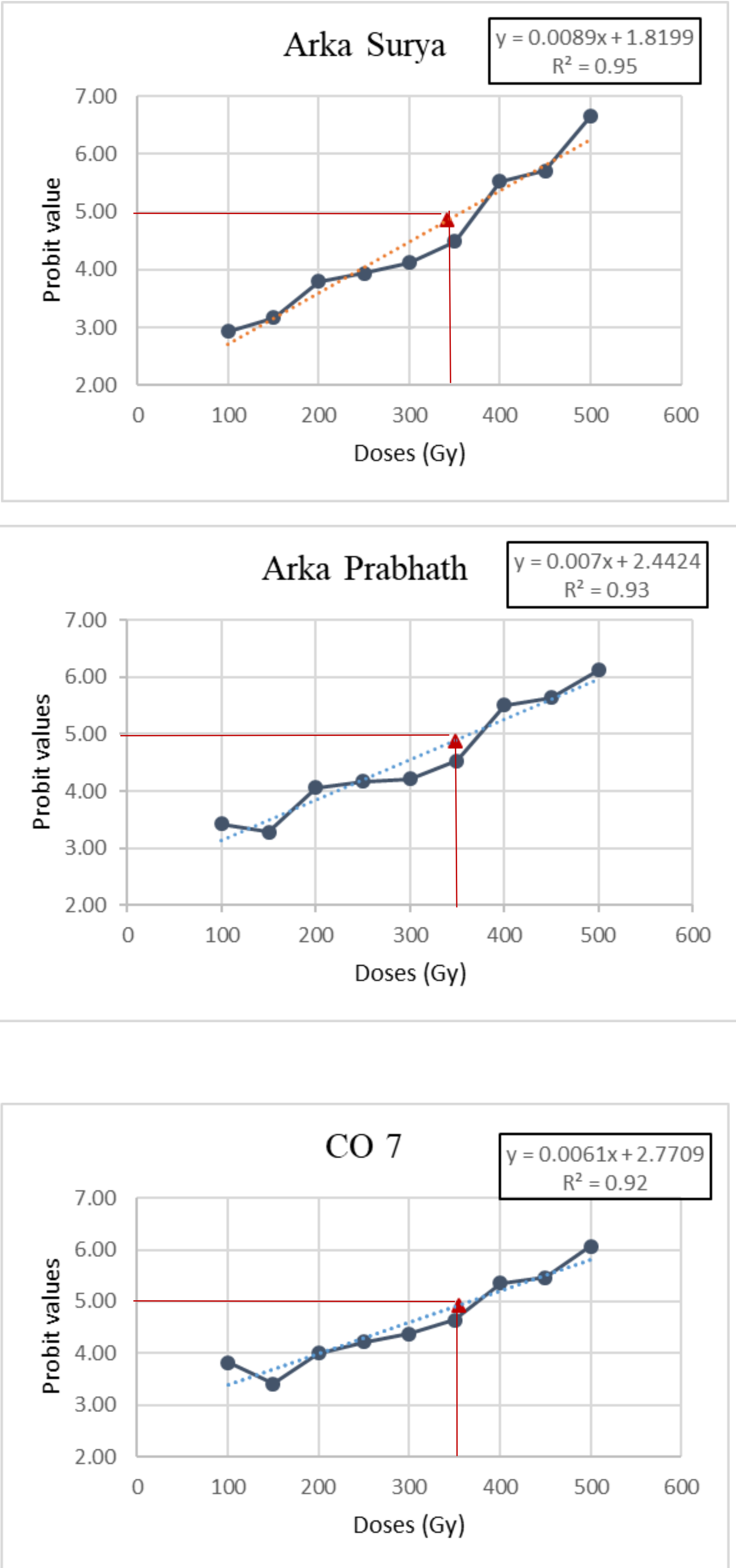

Fig. 2. Dose response curve of germination percentage (Probit analysis)

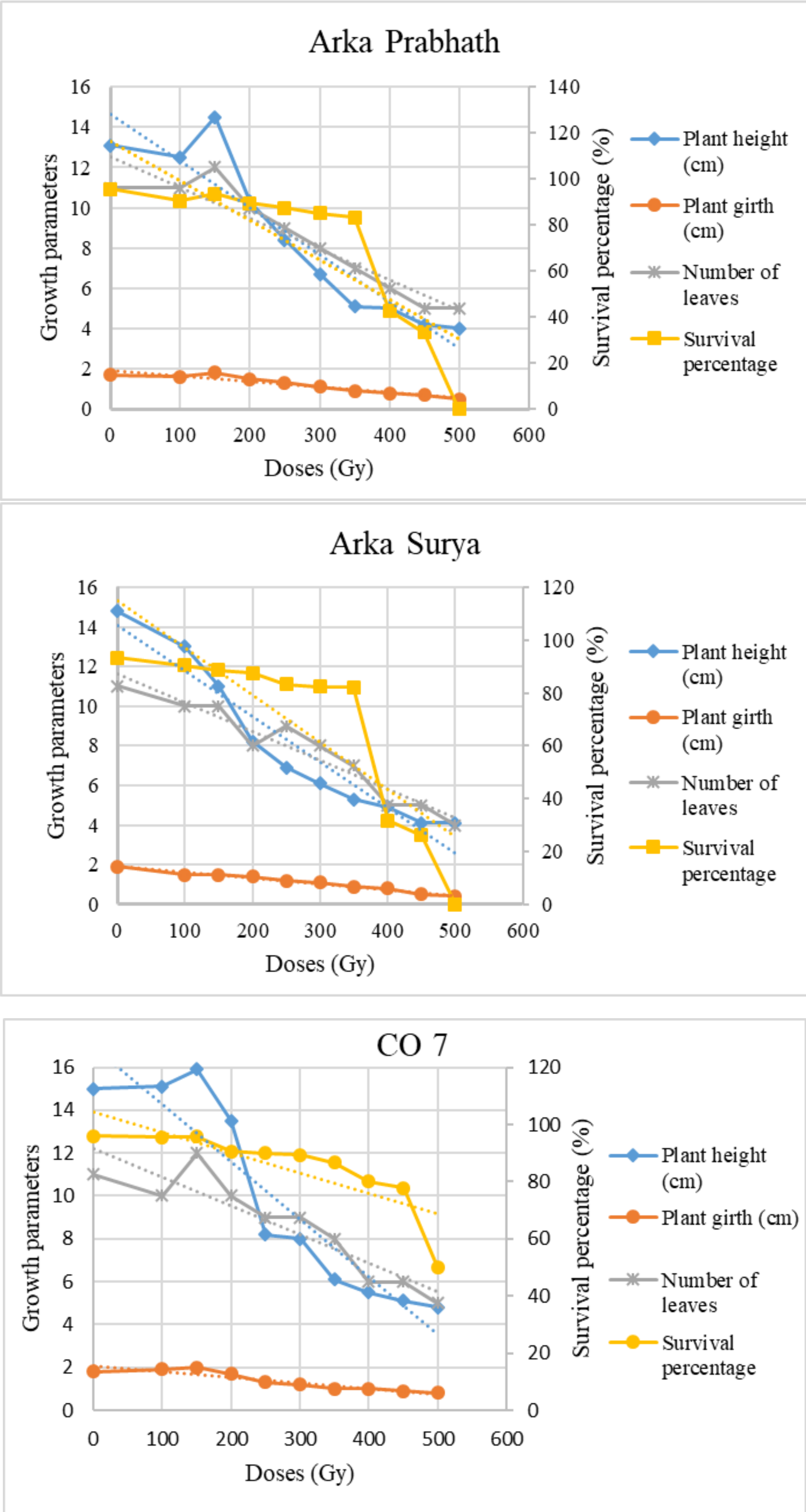

Fig. 3. Dose response curve of survival percentage and different seedling parameters

Arka Surya was determined based on the germination percentage using probit analysis (**Table 2**). The germination percentage was recorded up to four weeks after sowing. It is the minimum dose of gamma rays that permits fifty percentage germination of treated seeds. This is the optimum dose of gamma rays in papaya which produce mutants with minimum damage and maximum mutations. The findings revealed that the lethal dose of gamma rays in three papaya varieties ranged from 300 to 350 Gy and the established LD50 values were 354, 348 and 341 Gy for CO 7, Arka Prabhath and Arka Surya correspondingly (**Fig. 2**). Based on the data, it can be seen that the CO 7 variety had less sensitivity to gamma rays than Arka Prabhath followed by Arka Surya. Lethal dose differs with biological materials, nature of treatment and subsequent environmental conditions. Several studies of gamma-ray exposure in different papaya cultivars have been reported. Hang and Chau (2008) exposed the seeds of the papaya variety Dai Loan Tim to gamma rays ranging from 10 to 60 Gy. They indicated that LD50 of gamma rays in germinated papaya seeds was 30 Gy. Husselman *et al.* (2014) found that increased doses of gamma rays of 100 and 120 Gy were lethal to papaya when treated with dosages ranging from 0 to 120 Gy. The lethal dose of gamma rays for different fruit crops was reported by Surakshitha *et al*. (2017) in grapes and Murti *et al.* (2013) in strawberries.

The growth characteristics of mutant seedlings such as seedling height, seedling girth, number of leaves and petiole length were studied along with the control plants. Analysis of the variance of these traits showed that gamma irradiation had a significant effect on the growth of seedlings. Results presented a stimulative effect of gamma rays on the growth of seedlings at lower doses whereas became destructive at higher doses (**Table 3**). CO 7 and Arka Prabhath varieties displayed a revival in growth traits up to 150 Gy, above which a detrimental effect was noticed. Arka Surya exhibited a gradual reduction in growth with the increase in gamma-ray doses. The higher mean value of different seedling growth characteristics was observed in CO 7 followed by Arka Prabhath and Arka Surya. There was a clearcut reduction in the survival and growth of seedlings above the lethal dose of gamma rays in all varieties examined (**Fig. 3**). Seedling growth characteristics exhibited a regressing trend with enhance in gamma-ray doses in other crops such as Guava (Singh *et al*., 2018) and Acid lime (Devi *et al.,* 2021). Higher doses of gamma irradiation inhibit the endomitotic DNA synthesis during germination due to which, epicotyl elongation and cell elongation gets reduced in the seeds (Callebaut *et al.,* 1980). Increased dosages of gamma rays prompt the production of reactive oxygen species and lead to oxidative stress that damages the proteins, lipids and nucleic acids and alters the cellular metabolism in plants (Borzouei *et al*., 2010). From these results, it could be assumed that low doses of gamma rays caused a positive effect on the growth of seedlings. This can be substantiated by the hormesis phenomenon which is defined as the stimulating effect on living organisms for low levels of any factors that inhibit growth and development at higher levels (Amirikhah *et al*., 2021). Increased plant growth has been reported at lower doses of gamma rays in many crops such as grapes (Dev *et al*., 2016) and Lathyrus (Beyaz *et al.,* 2016). Even though lower doses of gamma rays induced different growth characteristics in seedlings, this is normally associated with low mutation frequencies (Mba *et al.,* 2010). Hence, the optimization of lethal dose is essential to obtain a higher rate of anticipated mutations with less injuries.

The outcome of the study validated that gamma irradiation can be used to generate significant variations in papaya. Chlorophyll deficient mutants were observed among the irradiated population. All biological parameters studied including lethality, sterility and seedling injury showed a steady increase with increasing gamma irradiation dosages. The mutagenic efficiency and effectiveness of gamma irradiation were found highest at LD50 and was decreasing with an increase in doses. Following the exposure of papaya seeds to various doses of gamma rays, their seedling growth pattern showed a regressing trend with an increase in doses. Lower doses of gamma rays exhibited a stimulation in certain growth traits whereas higher doses caused an inhibitory effect in papaya. Based on the current findings, the LD50 value of gamma rays was optimised as 354, 348 and 341Gy in papaya varieties CO 7, Arka Prabhath and Arka Surya respectively. The optimised LD50 dose can be utilized further for framing mutagenesis experiments for developing mutants with desirable characteristics in papaya.

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