The Pharma Innovation Journal 2021; 10(12): 2520-2522

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2021; 10(12): 2520-2522

www.thepharmajournal.com
Received: 02-09-2021
Accepted: 13-10-2021

Anisa M Nimbal
Assistant Seed Production Officer, AICRP-NSP (Crops), Seed Unit, UAS, Dharwad, Karnataka, India

V Rudra Naik
Professor and Deputy Director of Research, Directorate of Research, UAS, Dharwad, Karnataka, India

Corresponding Author:
Anisa M Nimbal
Assistant Seed Production Officer, AICRP-NSP (Crops), Seed Unit, UAS, Dharwad, Karnataka, India

Effect of gamma rays irradiation on seed germination, seedling growth and plant survival of Horsegram genotypes

Anisa M Nimbal and V Rudra Naik

Abstract
In the present study, the pure, healthy and dry seeds of four horsegram varieties of *viz.*, VLG 50, White seeded horsegram, SHG-07 and Tegur local were irradiated with four different doses of gamma rays *ie.*, 100, 200, 300 and 400 Gy for study of the effect on seed germination and seedling vigour such as germination per cent, shoot length (cm), root length (cm) and dry weight (g). For LD50 recorded near to the 400 Gy gamma rays as per survival per cent in the all four horsegram varieties. The M_1 generation was raised in the laboratory condition. The observations were recorded on germination per cent, shoot length, root length, and seed vigour index (length) and analyzed. The results showed that in M_1 generation (laboratory). Among four genotypes, VLG 50 was observed more sensitive, whereas, the cultivar Tegur local exhibited more resistance for most of all traits. The results of the study clearly indicated that different doses of gamma rays could be effectively utilized to create genetic variability for different quantitative traits in all the four genotypes.

Keywords: Horsegram, germination, seeding morphology, gamma radiation

Introduction
Horsegram (*Macrotyloma uniflorum* Lam. Verdc.) is an arid legume crop extensively cultivated in India, Shri Lanka, Maynmar and Australia. In India the crop is grow both in kharif and rabi season Horsegram is a promising nutritious crop; seeds contain high lysine content than chickpea and Tur (Yadav, 2004) [11]. The average productivity of horsegram crop is low. The causes of low productivity are many, but non-availability of high yielding varieties is the most important one. Success of conventional breeding is primarily dependent on the existing genetic variability for target traits (Ara *et al*, 2009) [1]. Natural variability is limited in horsegram and hybridization is difficult because of its cleistogamous, and small flower structure. Also, altering the seed coat colour in some of the genotypes is needed as the consumer preference is not black seeded one. Induced mutagenesis is one of the effective tool to create genetic variability so that improvement in yield, disease resistance, early maturity and grain quality can be achieved (Kharkwal *et al*, 2004) [6]. At present, gamma rays are one of the most used mutagens due to the lower cost (easy availability) and increased efficiency (higher penetration into matter) compared to other mutagens (Moussa 2006 Spencer-Lopéz *et al*., 2018) [7, 9]. However, induced mutation studies are scanty in horsegram. Keeping this in view, the present investigation was under taken in order to generate preliminary information for determining suitable doses of gamma radiation in inducing mutations in four different varieties of horsegram.

Materials and Methods
Genetically pure, healthy and matured seeds of four horsegram varieties *viz.*, VLG 50, White seeded horsegram, SHG-07 and Tegur local were subjected to Induced mutagenesis by exposing to different doses of gamma rays (100 Gy, 200 Gy, 300Gy and 400 Gy) at Bhabha Atomic Research Centre (BARC), Trombay. Non- irradiated seeds were treated as control. Under laboratory condition, 100 seeds per treatment along with control of all four varieties were sown for germination test and seedling characteristics study during *kharif* -2020 at seed Unit, UAS, Dharwad. Germination of seeds were carefully examined everyday and the emergence of cotyledon leaf was taken as the indication of germination. Count on germinated seeds for each treatment was taken on eighth day after sowing and germination percentage was calculated.

After fifteen days of sowing, shoot and length were measured in centimeter and seedlings were dried in hot air oven at constant 50 ºC temperature for 48 hours dry weight (g) and total number of seedlings survived were counted after fifteen days of sowing and plant survival percentage were calculated in the M_1 generation.

Results and Discussion

The effect of different doses of gamma rays in all four genotypes on germination percentage, shoot length, root length dry weight and percent decrease over the control is presented in Table1 and Table 2. All the varieties shown sensitive reaction to different doses. Based on survival percentage the LD_{50} for germination was found to be near 400 Gy in all four varieties. The physiological damage in terms of reduction germination percentage, survival percentage and seedling characters revealed that gamma radiation is more deleterious to the horsegram genotypes. To detect response of any crops to mutagenic treatment seed germination is used as the most important criteria. Both physical and chemical mutagens cause physiological damages in different biological materials during M_1 generation (Gaul, 1970) [4]. In the present study, control treatment of all the four genotypes recoded highest mean germination percentage (83,90,90,86) and survival percent (99,97,99,100) compared to different treatments whereas, lowest mean germination percentage (41,61,60,57) and survival percent (37,58,57,55) was exhibited by the treatment 400 Gy compared to different treatments (Table 1). Thus, results indicate that increased mutagens doses caused a progressive increase in biological damages as a measure of reduction in germination, shoot and root length in all four genotypes. Similar trend was observed in many leguminous crops by several workers (Bholbat *et al.* 2012, Datir *et al.*, 2007 and Hemavathy (2015) [2, 3, 5]. Among the genotypes, VLG -50 and Tegur local were most sensitive and least sensitive genotype with respect to different doses of gamma radiation treatment respectively. Differential genotypic radio-sensitivity to different mutagen doses within a species have also been reported by several workers *viz.*, Datir *et al*, 2007 [3], Sundesha, *et al.*, 2019 [10] and Priyanka *et al.*, 2020 [8] in horsegram.

The effect of induced mutation treatment on the initiation of the germination has been attributed to chromosomal organization Datir *et al.,* 2007 [3] and Chromosomal aberrations impacted by irradiation caused reduction in fertility and increase in physiological disorders leading to seedling injury, slow growth and mortality Sundesha *et al.*, 2019 [10]. Mutagen treated seed with lowered respiratory quotient may act in inhibition of germination. Thus, reduction in the germination could be due to the enhanced development of free radicles, causing lethality. Cumulative effect of all these factor may be attributed as the reason for reduction of plant survival.

Similar to the germination percentage, control treatment of all the four genotypes recoded highest mean shoot length (17.51,13.77,12.08,14.72 cm), root length (19.0,16.52,13.67,16.65 cm) and dry weight (0.232,0.201,0.187,0.234 g) compared to different treatments whereas, lowest mean shoot length (11.09,8.28,8.6,9.75cm), root length (10.96,9.3,8.3,8.87cm) and dry weight (0.188,0.187.0.143,0.147g) was exhibited by the treatment 400 Gy compared to different treatments (Table 2). Increased doses of gamma radiation resulted decline in seedling morphological traits *viz.*, shoot length, root length and dry weight revealing differential response of genotypes to different doses. These results are in accordance with the result observed by Sundesha *et al.,* 2019 [10] and Priyanka *et al.*, 2020 [8].

Table 1: Effect of different doses of gamma rays on seed germination and seed survival per cent in mungbean cultivars under laboratory conditions

Varieties	Treatments	Number of seeds sown	Seed germination (%)			Seed survival (%)		
			Number of seeds germinated	Mean germination in per cent	Reduction over control per cent Number of seeds survival	Number of seeds germinated	Mean germination in per cent	Reduction over control per cent Number of seeds survival
VLG-50	Control	100	83	83	-	99	99	-
	100 Gy	100	76	76	8.43	71	71	13.41
	200 Gy	100	70	70	15.66	67	67	18.29
	300 Gy	100	66	66	20.48	61	61	25.61
	400 Gy	100	41	41	50.60	37	37	54.88
White seeded Horsegram	Control	100	90	90	-	97	97	0.00
	100 Gy	100	85	85	5.56	83	83	4.60
	200 Gy	100	84	84	6.67	81	81	6.90
	300 Gy	100	70	70	22.22	67	67	22.99
	400 Gy	100	61	61	32.22	58	58	33.33
SHG-07	Control	100	90	90	-	99	99	0.00
	100 Gy	100	86	86	4.44	86	86	3.37
	200 Gy	100	82	82	8.89	80	80	10.11
	300 Gy	100	73	73	18.89	70	70	21.35
	400 Gy	100	60	60	33.33	57	57	35.96
Tegur local	Control	100	86	86	-	100	100	0.00
	100 Gy	100	84	84	2.33	83	83	3.49
	200 Gy	100	76	76	11.63	75	75	12.79
	300 Gy	100	62	62	27.91	62	62	27.91
	400 Gy	100	57	57	33.72	55	55	36.05

Table 2: Per cent reduction in shoot length (cm) and Root length (cm) and in horsegram cultivars under different treatments in laboratory conditions

Varieties	Treatments	Shoot length (cm)		Root length (cm)		Dry weight (g)	
		Mean (cm)	Per cent reduction over control	Mean (cm)	Per cent reduction over control	Mean (cm)	Per cent reduction over control
VLG-50	Control	17.51	-	19	-	0.232	-
	100 Gy	15.26	12.85	13.53	28.79	0.168	27.59
	200 Gy	14.92	14.79	12.36	34.95	0.175	24.57
	300 Gy	13.21	24.56	13.71	27.84	0.183	21.12
	400 Gy	11.09	36.66	10.96	42.32	0.188	18.97
White seeded Horsegram	Control	13.77	-	16.52	-	0.201	-
	100 Gy	12.2	11.40	15.67	5.15	0.174	13.43
	200 Gy	9.42	31.59	11.72	29.06	0.2	0.50
	300 Gy	8.6	37.55	9.77	40.86	0.163	18.91
	400 Gy	8.28	39.87	8.3	49.76	0.183	8.96
SHG-07	Control	12.08	-	13.67	-	0.187	-
	100 Gy	11.02	8.77	12.74	6.80	0.162	13.37
	200 Gy	9.42	22.02	12.48	8.71	0.152	18.72
	300 Gy	8.6	28.81	9.77	28.53	0.163	12.83
	400 Gy	8.12	32.78	8.3	39.28	0.143	23.53
Tegur local	Control	14.72	-	16.65	-	0.234	-
	100 Gy	13.65	7.27	16.44	1.26	0.204	12.82
	200 Gy	13.55	7.95	16.25	2.40	0.191	18.38
	300 Gy	12.85	12.70	16.12	3.18	0.166	29.06
	400 Gy	9.75	33.76	8.87	46.73	0.171	26.92

Conclusion

In the present investigation, among four horsegram genotypes VLG-50 was found more sensitive whereas, Tegur local was observed more resistant for different doses of gamma radiation exposure. The gamma radiation doses LD_{50} *ie.*, 400Gy or below 400 Gy could be utilized in irradiation of all four horsegram genotypes. Overall studies confirmed that gamma radiation could be used for inducing genetic variability or beneficial mutations in horsegram.

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