Induced mutagenic frequency and spectrum of chlorophyll mutants in black gram, *Vigna mungo* (L) Hepper

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ABSTRACT

The present investigation was conducted during the Kharif season of 2013. Seeds of black gram, *Vigna mungo* (L) Hepper variety VBN 4 and ACM 07002 were exposed to gamma rays doses at 200, 250, 300 and 350 Gy and EMS doses at 20, 30 and 40 mM. Wide range of chlorophyll mutations affected almost all the parts of plant and were isolated in M_2 generation. The chlorophyll mutant frequency was higher (2.3) in EMS treatment as compared to gamma treatment in VBN 4 whereas in ACM culture the frequency was high (2.1) in gamma ray treatment compared to EMS treatment. The spectrum of chlorophyll mutants with albino, xantha, chlorina and viridis was obtained. Among the chlorophyll mutants occurrence of viridis was rare and albino was predominant.

Keywords: ACM 07002; chlorophyll mutants; EMS; gamma ray; VBN 4

INTRODUCTION

Black gram, *Vigna mungo* (L) Hepper is popularly known as Urd bean, Urd or Mash. It is an important food legume crop of the Indian subcontinent and is widely cultivated grain legume in the Indian sub-continent comprising India, Burma, Bangladesh and Sri Lanka (Bhosale and Hallale 2011). Creation of variability through pollination and artificial hybridization is very difficult as the flowers are cleistogamous and delicate to handle. Even if hybridization is carried out the seed set is less than 5 per cent. Also this crop lacks proper male sterility system commercially to be utilized for hybridization. Hence the investigation was aimed at creation of variation through physical (gamma ray) and chemical (EMS) mutagens. Chlorophyll mutations are used as markers in genetic, physiological and biochemical investigations and can be used as an index in evaluating the mutagenic action of different mutagens in several crops (Reddy and Annadurai 1991).

MATERIAL and METHODS

The present investigation was conducted at Agricultural College and Research Institute, Killikulam during Kharif 2013 with the promising cultures of black gram viz VBN 4 and ACM 07002. The seeds for M_2 generation of VBN 4 and ACM 07002 culture were procured from Agricultural College and Research Institute, Madurai. The variety VBN 4 and culture ACM 07002 were treated with gamma rays at four different doses ranging from 200 to 350 Gy with an interval of 50 Gy and ethyl methane sulphonate (EMS) at three different doses ranging from 20 to 40 mM with an interval of 10 mM. The M_2 generation was raised from M_1 plant basis following plant to progeny method in a randomized block design with three replications in Kharif season. The seeds were sown with adequate spacing of 30 × 10 cm. The recommended agronomic practices and plant protection measures were followed uniformly for all the treatments. The M_2 population of each treatment was subjected to screening for different chlorophyll mutations. The pertinent values were calculated and the number of mutations per 100 plants of M_2 generation was estimated according to the method of Gaul (1957). The different types of chlorophyll mutants scored were albino, xantha, chlorina and viridis in different treatments. The spectrum of chlorophyll mutants was classified according to the terminology of Gustafsson (1940).

RESULTS and DISCUSSION

The chlorophyll mutants (Plate 1) were recorded within 10 days after sowing. The chlorophyll mutant frequencies were estimated in M_2 seedling basis for gamma ray and EMS treatment and the results are furnished in Table 1. The gamma ray-induced chlorophyll mutant frequency ranged from 0.7 (200 Gy) to 2.1 (300 Gy) on M_2 seedling basis. The EMS-induced chlorophyll mutant frequency ranged from 0.6 (20 mM) to 2.3 (40 mM) on M_2 seedling basis in VBN 4 variety. Similarly in ACM 07002 culture-induced chlorophyll mutant frequency ranged from 0.6 (200 Gy) to 2.1 (300 Gy) on M_2 seedling basis in gamma ray treatment and under EMS treatment-induced chlorophyll mutant frequency ranged from 0.6 (20 mM) to 1.6 (20 mM). The chlorophyll mutant frequency was high in higher doses in M_2 generation. These results are in line with those of Gautam and Mittal (1998). The chlorophyll mutant frequency was higher (2.3) in EMS treatment compared to gamma treatment in VBN 4 whereas in ACM culture frequency was high (2.1) in gamma ray treatment compared to EMS treatment (Fig 1).

The spectrum of chlorophyll mutation observed in segregation M_2 generations was analysed and their relative percentages are furnished in Table 2. In the present investigation the four different spectrums of chlorophyll mutants were obtained viz albino, chlorina, xantha and viridis which is in accordance with Lal et al (2009), Selvam et al (2010), Bhosale and Hallale (2011) and Kousar et al (2013) in blackgram.

Table 1. Frequency of chlorophyll mutants in M_2 generation for VBN 4 and ACM 07002

Treatment	Number of M_2 seedlings				Mutant frequency/ 100 M_2 plants scored	
	Scored		Segregated			
	VBN 4	ACM 07002	VBN 4	ACM 07002	VBN 4	ACM 07002
γ - rays (Gy)						
Control	290	286	-		-	-
200	545	540	4	3	0.7	0.6
250	444	491	6	6	1.4	1.2
300	421	433	9	9	2.1	2.1
350	407	396	7	7	1.7	1.8
EMS (mM)						
Control	290	286	-	-	-	-
20	527	514	3	3	0.6	0.6
30	449	468	5	5	1.1	1.1
40	395	378	9	6	2.3	1.6

Table 2. Spectrum of chlorophyll mutants in M_2 generation for VBN 4 and ACM 07002

Treatment	Total number of mutants in M_2		Spectrum (relative %) of chlorophyll mutants							
			Albino		Xantha		Chlorina		Viridis	
	VBN 4	ACM 07002	VBN 4	ACM 07002	VBN 4	ACM 07002	VBN 4	ACM 07002	VBN 4	ACM 07002
γ-ray (Gy)										
Control	-	-	-	-	-	-	-	-	-	-
200	4	3	25.00	33.33	50.00	33.33	25.00	33.33	-	-
250	6	6	16.67	50.00	33.33	33.33	16.67	16.70	33.33	-
300	9	9	55.6	44.4	22.2	22.2	22.2	33.33	-	-
350	7	7	57.14	28.6	28.57	28.6	14.28	42.9	-	-
EMS (mM)										
20	3	3	-	33.33	33.33	33.33	66.67	33.33	-	-
30	5	5	40.0	40.0	-	40.0	60.0	20.0	-	-
40	9	6	44.4	33.33	11.1	33.33	44.44	33.33	-	-

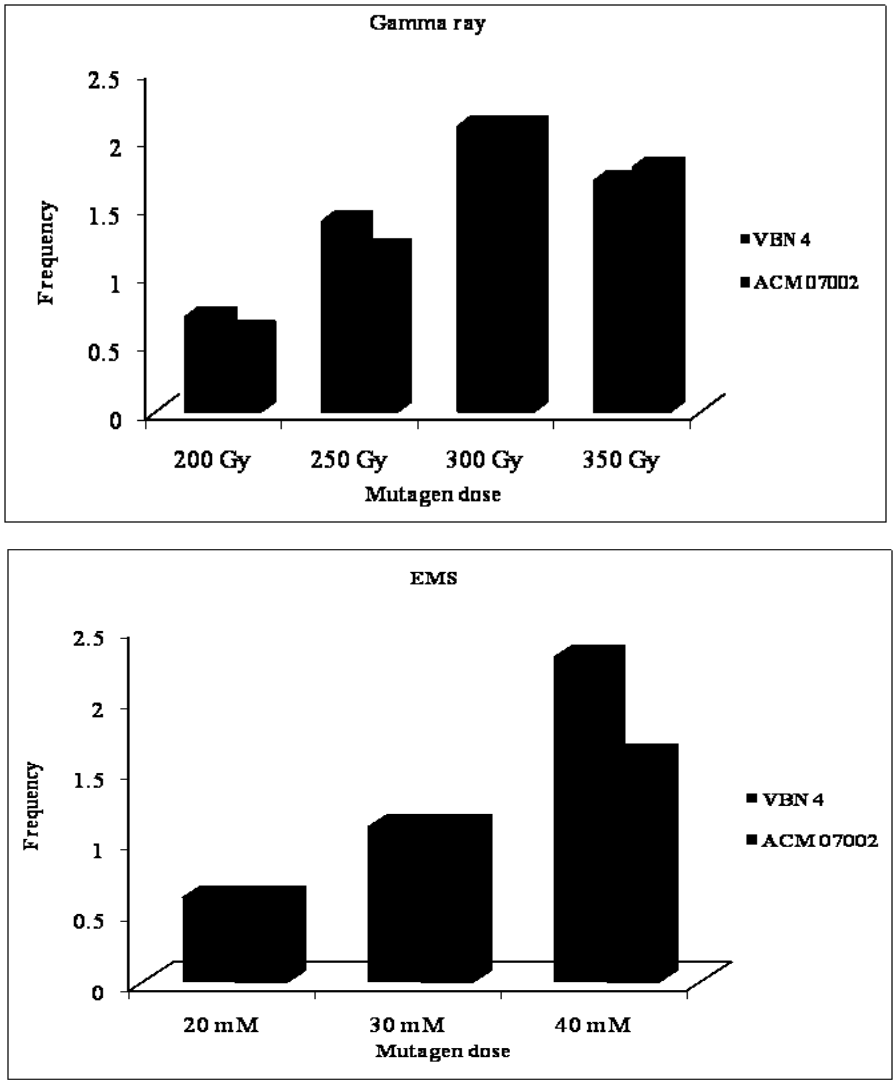
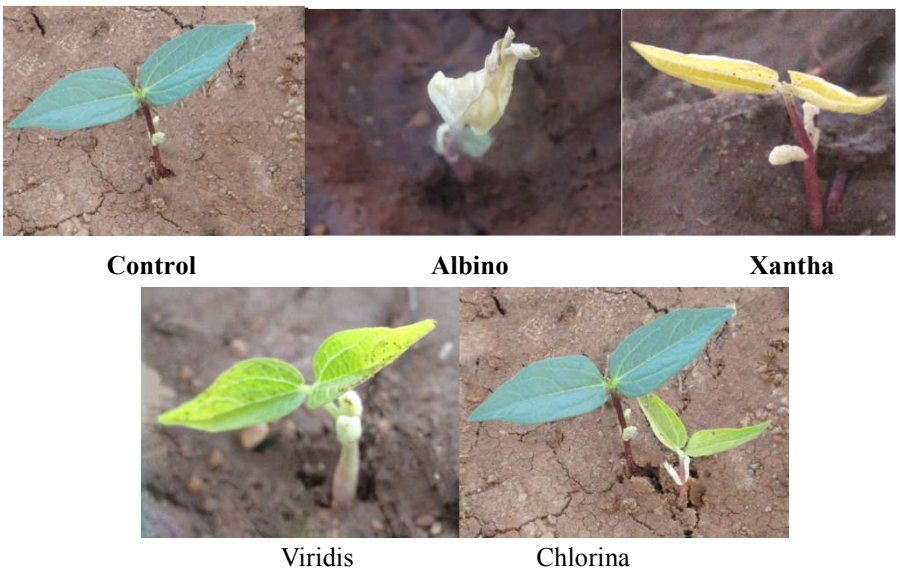


Fig 1. Frequency of chlorophyll mutants in M_2 generation for VBN 4 and ACM 07002



Control **Albino** **Xantha**

Viridis Chlorina

Plate 1. Field view of different types of chlorophyll mutants

In VBN 4 variety the spectrum comprised albino, xantha, chlorina and viridis. Among them the occurrence of viridis type of chlorophyll mutants was less and albina types was more in gamma ray treatment. The maximum frequency of albina was recorded in 350 Gy. These results are in agreement with those of Arulbalachandran and Mullainathan (2009). The spectrum comprised albino, xantha and chorina. Among them the occurrence of xantha type of chlorophyll mutants was less and chlorina types was more in EMS treatment. The maximum frequency of chlorina was recorded in 20 mM.

These results are in conformity with the work of Senapati et al (2008). The spectrum comprised can be represented by albino >xantha >chlorine >viridis in gamma rays treatment. In the EMS treatment the spectrum comprised can be represented by chlorine >albino >xantha. Similarly in ACM 07002 the spectrum comprised albina, chlorina and xantha. Among them the occurrence of xantha type of chlorophyll mutants was less and albina types was more in gamma ray treatment. The maximum frequency of albina was recorded in 250 Gy. The spectrum comprised albino, xantha and chlorina. Among them the occurrence of chlorina type of chlorophyll mutants was less and albino and xantha types was equally distributed in EMS treatment.

The spectrum comprised can be represented by albino >chlorine >xantha in gamma rays treatment. In the EMS treatment the spectrum comprised can be represented by albino= xanthan >chlorina.

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