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# DETERMINATION OF DOSES OF PHYSICAL, CHEMICAL AND THEIR COMBINATION OF MUTAGENS ON SEED GERMINATION AND RADICLE LENGTH IN OKRA (*ABELMOSCHUS ESCULENTUS* (L.) Moench.)

Sam Ruban J., Ebenezer Babu Rajan R. and Kader Mohideen M.

Department of Horticulture, Faculty of Agriculture, Annamalai University, Annamalainagar – 608 002, Tamil Nadu, India.

# Abstract

The Study on determination of doses of mutagens in bhendi cv. Arka Anamika reveals that the  $LD_{50}$  value was 40 kR for gamma rays, 0.4 per cent for EMS and 0.8 per cent for DES. Chemical mutagens when applied singly exerted greated inhibitory effect on seed germination and radical length as compared to gamma irradiation in bhendi. Among the mutagens, it was noted that the combination doses caused significant reduction in germination and radical length than single mutagenic treatments. Based on the germination level, the doses of 40 kR gamma in combination with 0.3, 0.4 and 0.5 per cent of EMS were considered as the  $LD_{50}$  value. At higher concentration, the growth of radical was completely inhibited in combination doses.

Keywords : Mutagens, LD50 value, germination

## Introduction

Mutagens has been an important tool in the hands of plant breeders to create variability and widen the genetic base in crop plants. Radiations were found to be the most efficient mutagen for higher rate of mutation. However, the chemicals were also found to be useful since they cause less damage but are more specific in their mutagenic effect. Though most of the workers have employed physical and chemical mutagens singly, the investigation involving combination of mutagens are meager in bhendi combination of mutagenic treatments is one of the methods employed for the enhancement of mutagenic efficiency (Nallathambi, 1983). Thus, radiation combined with chemical mutagens is expected to increase the mutation frequency and widen the spectrum. Yashvir (1975) studied the combined effect of radiations and chemicals and reported height reduction in okra. The main objective of the breeders is to obtain maximum frequencies of positive mutations. But, it is not clear which levels of mutagens have to be applied to attain this objectives. Hence, the investigation was undertaken in bhendi cv. Arka Anamika to fix the optimum dose (LD<sub>50</sub>) for physical, chemical mutagens and their combinations.

# Materials and Methods

The study was carried out at the vegetable field unit of Department of Horticulture during 2001. One physical mutagens i.e., gamma ray and two chemical mutagens viz., ethyl methane sulphonate (EMS) and diethyl sulphate (DES) were employed. The seeds were exposed to gamma irradiation treatment of 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 kR. Ten concentrations of EMS and DES viz., 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1.0 per cent were tried along with a control. The seeds subjected to mutagenic treatments were allowed to germinate in petridishes and radical length were observed in lab conditions. The germinated seeds placed in petridishes were counted from  $2^{nd}$  to  $7^{th}$  day. The number of seeds germinated from each doses of treatment was counted and the percentage was worked out. Also the root length from the cotyledonary node to the tip of the primary root was measured and expressed in centimetres.

#### **Results and Discussion**

The germination percentage of mutated seeds showed a range of 26.56 per cent for gamma 100 kR to 96.28 per cent for 10 kR gamma radiations as against 100 per cent germination in control (Table 1). As the dose levels increased, the germination showed a decrease. Similar results have been reported by Singh *et al.* (1998). The radical length was also affected due to different irradiation treatments. It was lowest (2.80 cm) in 100 kR gamma as against the highest (4.20 cm) in control followed by 10 kR with 4.0 cm. The dose of gamma 40 kR in which 50 per cent of reduction in germination was recorded. Accordingly, the doses of 30, 40 and 50 kR of gamma ray doses were fixed as the LD<sub>50</sub> value.

The chemical mutagen viz., EMS showed a germination range of 21.50 per cent for 1.0 per cent 95.17 per cent for 0.1 per cent as compared to control which registered 100 percent germination. Similarly, the



readicle length also showed market reduction with increase in concentration. More than 50 per cent reduction in germination was recorded at 0.4 per cent EMS concentration. Hence, the doses of 0.3, 0.4 and 0.5 per cent of EMS were fixed as the LD<sub>50</sub> value. Similarly, based on the germination percentage, the LD<sub>50</sub> value has been fixed for DES also. The results showed a range in germination of 37.50 per cent for 1.0 per cent to 98.52 per cent for 0.1 per cent as compared to 100 per cent in control. Similarly, the radical length showed a range from 0.10 cm in 1.0 percent DES to 3.95 cm in 0.1 per cent as against the control with 4.10 cm. With DES treatments, 50 percent reduction in germination was observed at 0.8 per cent. Hence, LD<sub>50</sub> value was found to be 0.7, 0.8 and 0.9 per cent for DES treatments. This is in line with the results obtained by Jeevanandam et al. (1985) and Suryakumari (2002).

In the combination treatment of gamma with EMS showed a marked reduction as compared to single treatment (Table 2). The combined treatment of 100 kR gamma with 0.5 EMS recorded lowest germination of 8.90 per cent as compared to highest (62.58 in 20 kR gamma with 0.3 per cent EMS treatment, whereas in control it was 100 per cent. Higher concentration the growth of radical was completely inhibited as reported by Ramalingam (1980). Based on the results, the doses of 40 kR gamma combined with 0.3, 0.4 and 0.5 percent of DES were fixed as the LD<sub>50</sub> value. The combination study of gamma with DES indicated minimum germination of 19.80 per cent in 100 kR gamma with 0.9 per cent DES as against a maximum of 65.50

percent with 20 kR gamma with 0.7 per cent DES treatment. The length of the radical also showed an inverse relationship with the dose levels (Yashvir, 1975). Based on the results, the doses viz., 0.7, 0.8 and 0.9 percent DES combined with 50 kR gamma was fixed as the  $LD_{50}$  value.

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Table 1 :  $LD_{50}$  value for Gamma rays, EMS and DES for germination (Per cent) and radicle length (cm) under laboratory condition in okra

	Gamma rays				EMS		DES		
Tr. No.	Dose (kR)	Germination (%)	Radicle length (cm)	Dose (kR)	Germination (%)	Radicle length (cm)	Dose (kR)	Germination (%)	Radicle length (cm)
$T_1$	Control	100.0 (90.00)	4.2	Control	100.00 (90.00)	3.9	Control	100.0 (90.00)	4.1
$T_2$	10	96.2 (78.88)	4.0	0.1	95.0 (77.30)	3.8	0.1	98.5 (81.93)	3.9
T <sub>3</sub>	20	81.7 (64.70)	3.8	0.2	80.3 (63.72)	3.6	0.2	92.7 (74.36)	3.9
$T_4$	30	72.4 (58.34)	3.7	0.3	56.2 (48.60)	2.5	0.3	87.6 (69.46)	2.9
T <sub>5</sub>	40	52.1 (46.24)	3.5	0.4	52.5 (46.43)	2.4	0.4	81.5 (64.53)	2.8
T <sub>6</sub>	50	54.3 (47.49)	3.6	0.5	50.6 (45.36)	2.1	0.5	73.1 (59.34)	2.4
T <sub>7</sub>	60	44.6 (41.95)	3.4	0.6	47.9 (43.81)	1.9	0.6	65.2 (53.88)	2.0
T <sub>8</sub>	70	43.7 (41.40)	3.2	0.7	41.9 (40.34)	1.0	0.7	53.6 (47.09)	1.2
T <sub>9</sub>	80	41.5 (40.11)	3.1	0.8	34.8 (36.18)	0.2	0.8	50.5 (45.30)	0.5
T <sub>10</sub>	90	28.7 (32.42)	3.0	0.9	24.7 (29.86)	0.1	0.9	48.5 (44.14)	0.3
T <sub>11</sub>	100	26.5 (31.02)	2.8	1.0	21.5 (27.62)	0.0	1.0	37.5 (37.76)	0.1
SED		0.43	0.13		0.73	0.09		0.59	0.16
CD (p = 0.05)		0.88	0.26		1.47	0.19		1.18	0.27

Figures in parentheses indicate transformed values

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Tr.	Gamma + EMS			Gamma + DES				
No.	Dose	Germination	Radicle	Dose	Germination	Radicle		
		(%)	length		(%)	length (cm)		
			( <b>cm</b> )					
$T_1$	Control	100 (90.00)	4.0	Control	100.0 (90.00)	4.0		
T <sub>2</sub>	20 kR + 0.3 % EMS	62.5 (52.29)	3.9	20 kR + 0.7 % DES	65.5 (54.03)	3.9		
T <sub>3</sub>	20 kR + 0.4 % EMS	60.5 (51.06)	3.9	20 kR + 0.8 % DES	60.6 (51.17)	3.8		
$T_4$	20 kR + 0.5 % EMS	57.2 (49.18)	3.4	20 kR + 0.9 % DES	53.7 (48.87)	3.8		
T <sub>5</sub>	40 kR + 0.3 % EMS	52.6 (46.53)	3.4	40 kR + 0.7 % DES	55.6 (48.22)	3.9		
T <sub>6</sub>	40 kR + 0.4 % EMS	50.5 (45.29)	3.0	40 kR + 0.8 % DES	56.5 (48.73)	3.7		
T <sub>7</sub>	40 kR + 0.5 % EMS	48.8 (44.33)	2.8	40 kR + 0.9 % DES	52.1 (46.23)	3.5		
T <sub>8</sub>	60 kR + 0.3 % EMS	46.2 (42.87)	2.2	60 kR + 0.7 % DES	50.9 (45.53)	3.2		
T <sub>9</sub>	60 kR + 0.4 % EMS	41.3 (40.00)	2.0	60 kR + 0.8 % DES	50.6 (45.37)	3.0		
T <sub>10</sub>	60 kR + 0.5 % EMS	40.5 (39.52)	1.8	60 kR + 0.9 % DES	49.5 (44.71)	2.5		
T <sub>11</sub>	80 kR + 0.3 % EMS	37.5 (37.80)	1.9	80 kR + 0.7 % DES	42.5 (40.69)	2.8		
T <sub>12</sub>	80 kR + 0.4 % EMS	33.1 (35.18)	1.5	80 kR + 0.8 % DES	40.0 (39.23)	2.6		
T <sub>13</sub>	80 kR + 0.5 % EMS	30.5 (33.57)	1.4	80 kR + 0.9 % DES	37.0 (37.46)	2.0		
T <sub>14</sub>	100 kR + 0.3 % EMS	27.0 (31.13)	0.9	100 kR + 0.7 % DES	26.1 (30.76)	1.9		
T <sub>15</sub>	100 kR + 0.4 % EMS	21.0 (27.27)	0.3	100 kR + 0.8 % DES	25.5 (30.28)	1.3		
T <sub>16</sub>	100 kR + 0.5 % EMS	18.9 (25.77)	0.0	100 kR + 0.9 % DES	19.8 (26.42)	0.5		
	SED	1.25	0.33		0.35	0.12		
	CD ( $p = 0.05$ )	2.58	0.69		0.71	0.25		

**Table 2 :**  $LD_{50}$  value for combination of mutagens (Gamma + EMS and Gamma + DES) for germination (per cent) and radicle length (cm) under laboratory condition in okra.

Figures in parentheses indicate transformed values