

Assessment of Mutagenicity Induced by Different Mutagens in Coriander (*Coriandrum sativum* L.)

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Authors' contributions

This work was carried out in collaboration among all authors. Authors PNK and MDV designed the study, performed the analysis of study, wrote the protocol and wrote the first draft of manuscript. Author SNH managed the analyses of study. Authors SND and SGW manage the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Coriander is one of the most widely used herb. Coriander generally grown for leaf and grain purpose throughout the world. Induction of mutation is considered to be effective in improving various characteristics in plants like seed germination, plant vigour, yield etc. In present study we used three different chemical mutagens Colchicine, Sodium azide (SA) and Methyl methane sulphonate (MMS) on four different varieties of coriander (Green wonder, Cross-91, Mrudul and Surabhi). The study was performed by exposing the seeds of four varieties of coriander (*Coriandrum sativum* L.) to Colchicine (0.01%,0.02%,0.03% and 0.04%), Sodium azide (SA) at (0.1%,0.2%,0.3% and 0.4%) and Methyl methane sulphonate (MMS) at (0.01%,0.02%,0.03% and 0.04%) respectively. The observations were made on seed germination in field and laboratory conditions and the observations on seedling height, number of primary branches and leaves were made in field conditions. All the mutagens significantly affect the germination and seedling growth. The study revealed that germination percentage, plant height, no. of primary branches and leaves decreased with increase in dose/concentration of mutagen. Among three different chemical mutagen we

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observed that colchicine is better mutagen for induction of mutagen after that sodium azide (SA) had good results for seed treatment. Some of the concentrations of methyl methane sulphonate (MMS) (0.01% & 0.02%) were better. Among all four varieties of coriander Cross-91 gave better result after that Mrudul and Surabhi respond well to some concentrations. We categories the effectiveness of mutagen to induce mutation in coriander as MMS>SA>Colchicine.

Keywords: Mutation; coriander; polyploidy; colchicine; methyl methane sulphonate; sodium azide.

1. INTRODUCTION

Coriander is an important seed and herb spice crop of Asia used in several culinary purposes and also for treatment of several diseases. India is the largest producer of coriander. It is prominently cultivated in Rajasthan, Andhra Pradesh, Gujarat and Madhya Pradesh. Approximately 80% of the world total coriander seed is produced in India of which more than 80% is produced in Southeast rajasthan [1]. Mutation breeding is powerful tool to enrich variation particularly for attributes of economical importance in crop like coriander where hybridization is difficult. Mutation is a physical or chemical agent that changes the genetic material usually DNA of an organism. Physical and chemical mutagen induces physiological changes (injury) gene mutation (point mutations) in the biological material in M1 generation [2]. Germination percentage, seedling height, percentage survival, shoot length decreased with increased in dose/concentration of the mutagen lower treatment of all the mutagens causes less biological damage and suitable for inducing desirable mutations in coriander [3]. Polyploidy play an important role in plant evolution and genetic variation creation. Polyploidy induction is carried out by using colchicine and ploidy level was determined by analysis of flow cytometry, after DNA quantification by spectrophotometric method it was observed that treated plant presented higher contents of cellular DNA than diploid plants. Polyploidy induction is effective method to increase plant performance [4]. Genetic variation is the most essential pre-requisite for successful crop improvement program as it provide spectrum of variants for the effective selection. This can be achieved through the process of mutation [5]. Lower concentration of chemical mutagens was more effective and efficient in inducing mutation with low biological damages and high mutation frequency in foxtail millet [6].

2. MATERIALS AND METHODS

The coriander varieties Green wonder, Cross-91, Mrudul, Surabhi were selected as base

population for experiment. The presoaked seeds of coriander varieties were treated with Colchicine (0.01, 0.02, 0.03 and 0.04%), Sodium azide (SA) (0.1, 0.2, 0.3 and 0.4%) and Methyl methane Sulphonate (MMS) (0.01, 0.02, 0.03 and 0.04%) respectively. Seeds are treated for 12 hrs after that removed air dried and sown in field directly and also in laboratory by using germination tray. The data on germination percentage recorded from 15 days after sowing in field and laboratory. And other biological parameters such as seedling height, number of primary branches and number of leaves were recorded on 40-50 days after sowing in field.

3. RESULTS AND DISCUSSION

3.1 Comparative Effects of Mutagens on Seed Germination

3.1.1 In laboratory

The data on seed germination in laboratory was influenced by different mutagenic treatments on coriander varieties compared with control (In percentage) and presented in Table 1 and Fig. 1 respectively. In this study we observe that all the concentration of colchicine, SA and MMS showed the effect on seed germination percentage when compared with control. Colchicine treated seeds showed variable result in germination percentage. Highest germination percentage was observed in Surabhi variety (93.33%) treated with 0.04% colchicine and lowest germination percentage was observed in green wonder (13%) treated with 0.03% of colchicine. Cross-91 and Mrudul also shows variable results for germination. In SA treatment as the concentration of sodium azide increases germination percentage tends to decrease similar results of decrease in germination was observed in coriander [3,7]. At the concentration of 0.01% MMS green wonder shows 73.33%, Cross-91 38.42%, Mrudul 36.28% and Surabhi 80% of germination respectively. But the germination percentage decreases above the 0.02% MMS concentration. Many workers have

reported the adverse effect of physical or chemical mutagen on plant survival, germination [8,9,10,11,12,13]. Sodium azide is the less effective and the most efficient mutagen and has been reported to be mutagenic in several crop [14,15]. Decrease in seed germination induced

by mutagenic treatment may be the result of damage of cell constituents at molecular level [16,17]. The inhibition of seedling growth due to mutagenic treatments might be due to auxin destruction or due to inhibition of auxin synthesis [18].

Table 1. Comparative effect of mutagens on seed germination percentage in laboratory

Treatment	Coriander varieties (Germination % laboratory)			
	Green wonder	Cross-91	Mrudul	Surabhi
Control	86	90	82.68	88.32
0.01% Col.	13.33	73.33	66.67	60
0.02%Col.	58	66.67	53.33	53.33
0.03%Col.	42	80	66.67	86.86
0.04%Col.	53.33	80	60	93.33
0.1% SA	50	80	46.67	87
0.2% SA	44	58.6	38.32	46
0.3% SA	38	60	32.44	33.33
0.4% SA	25	33.33	28.42	26.34
0.01% MMS	73.33	38.42	36.28	80
0.02%MMS	47	26	28.18	28.72
0.03%MMS	34	22	25.16	18.67
0.04%MMS	25	18.36	16.20	13.33

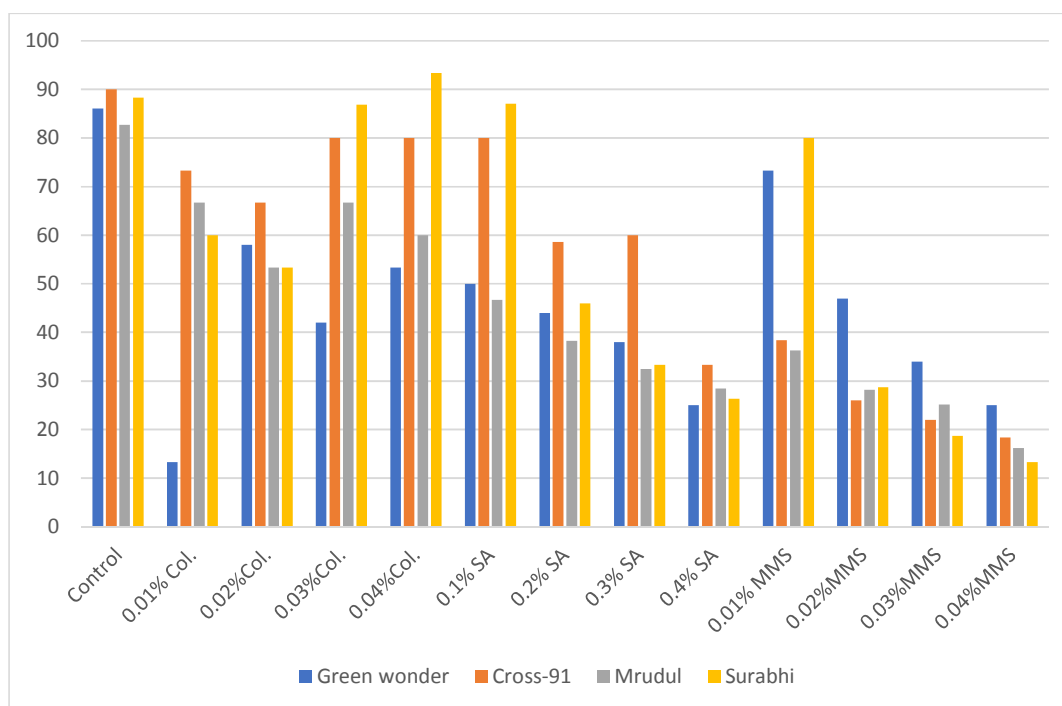


Fig. 1. Comparative effect of mutagen on seed germination in laborator

3.1.2 In field

In field condition all the concentration of colchicine showed 100% germination in all varieties (Table 2 and Fig. 2) there was no any effect of colchicine treatment on germination percentage. As compared to germination percentage in laboratory as the concentration of SA increases germination percentage decreases in field also. Similar results of decrease in seed germination percentage were observed in cluster

bean [19,20,21,22,23]. The effect of sodium azide on growth and variability induction was observed in *Helianthus annuus* [24]. Dose dependent reduction in germination was observed in coriander [25,26]. Among four varieties of coriander Cross-91 shows good result for all the mutagens treatments. It gives better result as compared to other varieties when treated with Sodium azide as well as Methyl methane sulphonate. (Table 2 and Fig. 2).

Table 2. Comparative effect of mutagen on seed germination in field

Treatment	Coriander varieties (Germination % field)			
	Green wonder	Cross-91	Mrudul	Surabhi
Control	100	100	100	100
0.01% Col.	100	100	100	100
0.02%Col.	100	100	100	100
0.03%Col.	100	100	100	100
0.04%Col.	100	100	100	100
0.1% SA	60	100	60	100
0.2% SA	40	60	40	60
0.3% SA	40	60	20	20
0.4% SA	20	40	20	20
0.01% MMS	60	80	60	80
0.02%MMS	40	60	60	40
0.03%MMS	20	40	40	40
0.04%MMS	20	40	20	20

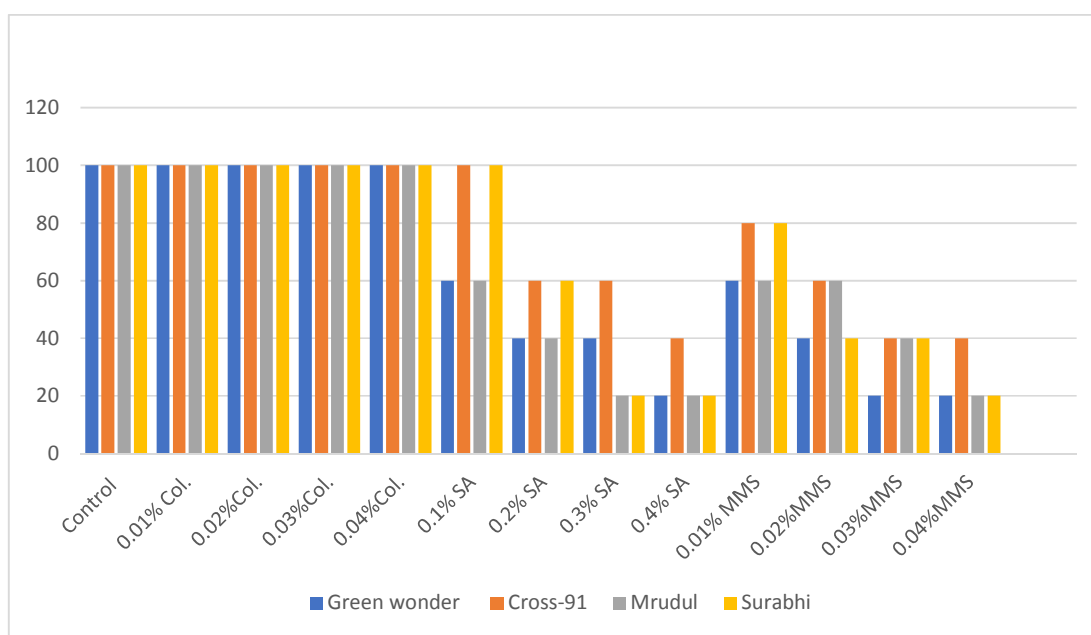


Fig. 2. Comparative effect of mutagen on seed germination in field

3.2 Comparative Effect of Mutagens on Plant Height (cm) at 50 DAS in Field

The effect of different mutagens on plant height at 50 DAS in field are shown in Table 3 and Fig. 3 respectively. At colchicine concentration 0.01 to 0.04% the height ranges from 7.12 cm to 8.7 cm in Green wonder, 15.12 cm to 8.0 cm in Cross-91, 13.90 cm to 8.20 cm in Mrudul and 11.72cm to 9.44 cm in Surabhi variety. In between them highest height was found in Cross-91(15.12 cm at 0.01 % Col.), Mrudul (13.92 at 0.02% Col.) and Surabhi (11.72 at

0.01% Col.) which was higher than Control. Sodium azide (SA) concentration shows variable result in plant height in field highest height was found in Cross-91 12.04 cm at 0.1% SA and lowest plant height was observed in Mrudul 4.2 cm at 0.4% SA. Similar mutagenic effect of sodium azide on growth and yield characteristics was observed in wheat [27,28,29,12,30,31]. In all MMS treatment it was observed that as concentration of MMS increases the plant height decreases. Concentrations above 0.02% are found to be lethal to plant height.

Table 3. Comparative effect of mutagens on plant height (cm) at 50 DAS in field

Treatment	Coriander varieties (Plant height cm)			
	Green wonder	Cross-91	Mrudul	Surabhi
Control	12.8	14.6	10.7	10.08
0.01% Col.	7.12	16.12	13.9	10.6
0.02%Col.	12.5	11.88	13.92	11.36
0.03%Col.	6.4	9.3	9.1	13.3
0.04%Col.	8.7	8	8.2	13.44
0.1% SA	8.56	15.04	8.6	9.8
0.2% SA	6.24	11.88	6.2	7.8
0.3% SA	7.92	10.96	6.9	8.4
0.4% SA	6.35	9.71	4.2	4.6
0.01% MMS	8.3	9.2	7.56	8.43
0.02%MMS	6.56	6.24	5.36	6.16
0.03%MMS	5.3	4.9	5.88	3.18
0.04%MMS	3.9	4.2	4.34	2.46

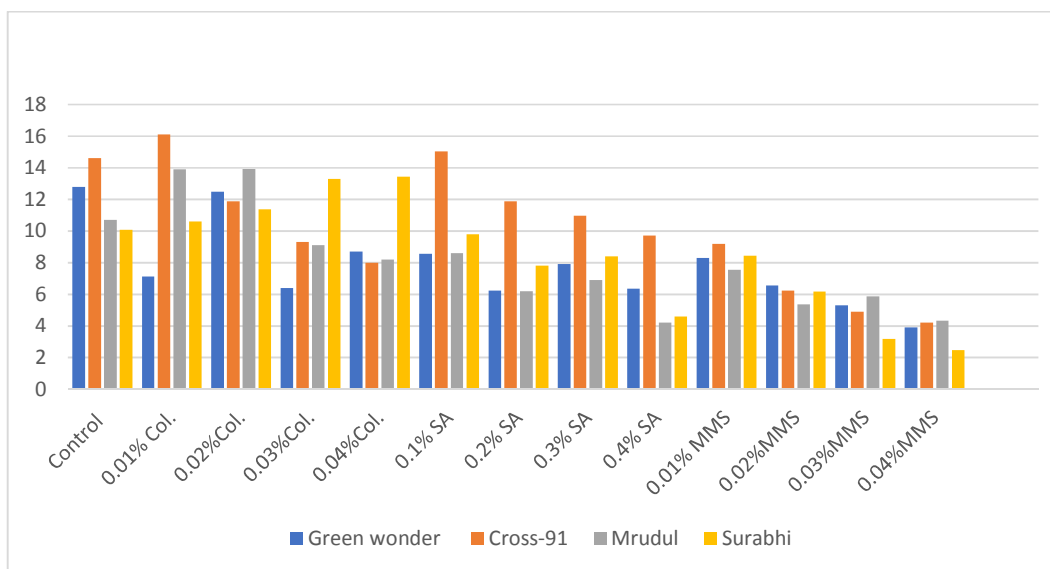


Fig. 3. Comparative effect of mutagens on plant height (cm) 50 DAS in field

3.3 Comparative Effect of Mutagens on Number of Primary Branches at 50 DAS in Field

In all the concentrations of Colchicine, Sodium azide and Methyl methane sulphonate (MMS) it was noticed that increase in concentrations of mutagens result in decrease in number of primary branches as compared to control. Among the treatments higher number of primary branches was observed in Cross-91 (15.24) treated with 0.02% Colchicine. After that Green wonder shows highest number of primary branches which was ranged from 11.24 at 0.01% to 9.88 at 0.04% Colchicine. Among the Sodium azide treatments 0.1% SA had recorded more

number of primary branches in Cross-91(13.18%) and 0.4% SA recorded lower number of primary branches in Green wonder (6.47). Many workers reported the adverse effect of physical and chemical mutagen on growth and yield [32,33,34]. Similar result of different concentration of Sodium azide on number of primary branches was observed in groundnut [35,12,15]. The significant difference was observed in all four varieties when treated with MMS. Only 0.01 and 0.02% MMS concentrations are good for seed treatment. Concentration above 0.02% shows the reduction in number of primary branches which was lowest in Green wonder (4.52) at 0.04% MMS.

Table 4. Comparative effect of mutagen on number of primary branches at 50 DAS in field

Treatment	Coriander varieties (no. of primary branches)			
	Green wonder	Cross-91	Mrudul	Surabhi
Control	12.55	15.48	13.2	12.82
0.01% Col.	11.24	13.2	9.16	11.83
0.02%Col.	12.18	15.24	9.44	10.47
0.03%Col.	11.36	14.54	10.56	11.22
0.04%Col.	9.88	13.44	8.34	8.36
0.1% SA	11.28	13.18	12.77	10.24
0.2% SA	10.37	10.48	10.23	9.88
0.3% SA	8.28	9.77	7.68	7.43
0.4% SA	6.47	8.56	7.23	8.54
0.01% MMS	11.18	11.56	10.36	9.46
0.02%MMS	9.66	9.77	7.28	6.26
0.03%MMS	8.23	7.18	6.47	5.48
0.04%MMS	4.52	6.26	5.32	5.14

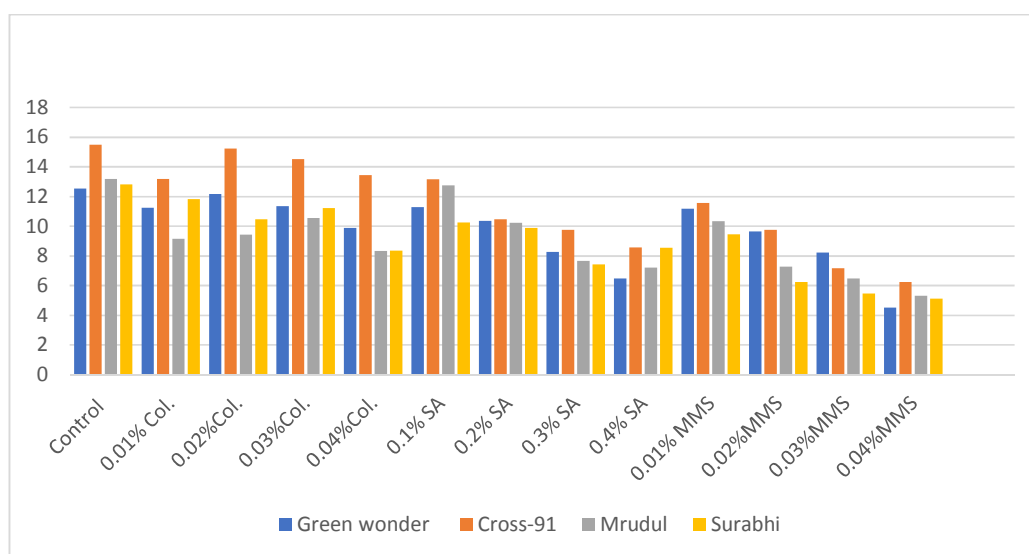


Fig. 4. Comparative effect of mutagen on number of primary branches at 50 DAS in

3.4 Comparative Effect of Mutagens on Number of Leaves at 50 DAS in Field

Among the different treatments under study the number of leaves ranged from 12.60 to 11.40 in Green wonder, 13.76 to 10.96 in Cross-91, 10.80 to 10.12 in Mrudul and 11.40 to 9.03 in Surabhi when treated with 0.01% to 0.04% Colchicine Table 5 and Fig. 5. Among all these varieties as like highest no.of primary branches Cross-91 also had highest number of leaves 13.76 at 0.01% Colchicine. Variation in number of leaves was observed in Sodium azide treatment. Green wonder shows highest number of leaves 12.96 at 0.1% SA. Whereas lower number of leaves

observed in Mrudul (4.72) at 0.4% SA. Different doses of sodium azide influenced the performance of chilli [12]. Significant differences was observed in MMS treatment for number of leaves only concentration 0.01% and 0.02% MMS are suitable for treatment whereas concentration above 0.02% is found to be lethal which shows lower number of leaves in all four varieties (Table 5 and Fig. 5). Mutagenic effectiveness and efficiency generally increased with increase dose of mutagen [36]. The effect of mutagens on morphology of plant reported in cowpea [37,38,39]. Dose dependent decrease in biological parameters has also been observed in chilli [40].

Table 5. Comparative effect of mutagens on number of leaves at 50 DAS in field

Treatment	Coriander varieties (number of leaves)			
	Green wonder	Cross-91	Mrudul	Surabhi
Control	13.16	14.77	10.73	12.38
0.01% Col.	12.6	13.76	10.8	11.4
0.02%Col.	10.93	12.07	11.63	10.36
0.03%Col.	12.78	11.88	9.78	9.47
0.04%Col.	11.4	10.96	10.12	9.63
0.1% SA	12.96	11.37	11.23	12.3
0.2% SA	11.42	10.28	9.48	10.54
0.3% SA	9.56	9.68	8.37	9.84
0.4% SA	5.63	6.36	4.72	5.42
0.01% MMS	10.32	10.92	9.27	10.12
0.02%MMS	8.18	9.08	8.66	7.37
0.03%MMS	4.88	3.96	5.32	4.58
0.04%MMS	2.44	3.27	3.2	2.77

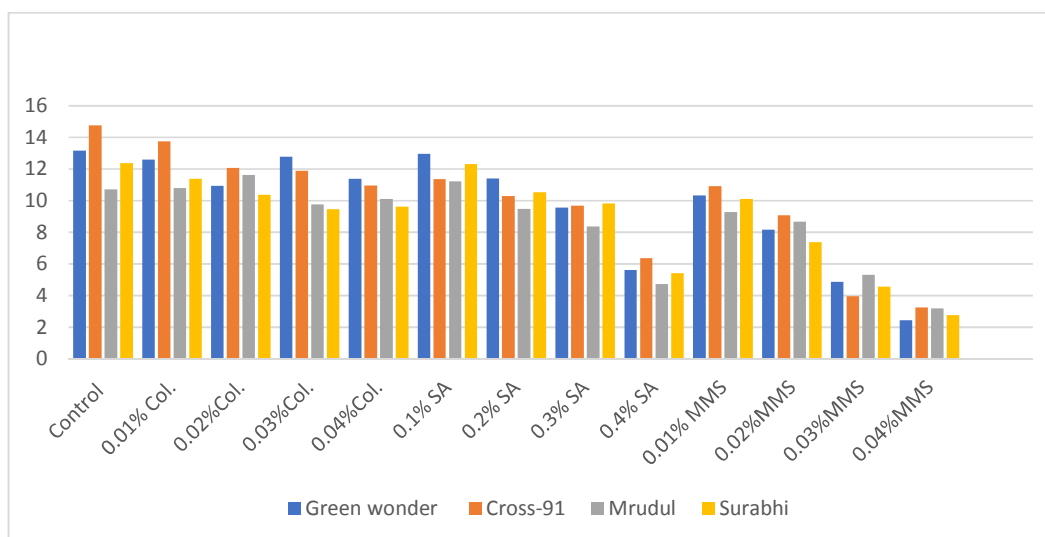


Fig. 5. Comparative effect of mutagens on number of leaves at 50 DAS in field

4. CONCLUSION

In the present study from all the observations recorded on different biological parameters we can concluded that Colchicine is better chemical mutagen for induction of mutation. After that sodium azide had good result for seed treatment. Some of the concentrations of MMS 0.01% and 0.02% had good results; in general reduction in seed germination seedling growth occurs due to increased dose/ concentration of mutagen. Among all the four varieties of coriander, Cross-91 had good result for all observations. The effectiveness of mutagen for induction of mutation was MMS>SA>Colchicine.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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