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ANTIMUTAGENIC EFFECTS OF L-CYSTEINE AGAINST DIETHYL SULPHATE INDUCED TOXICITY IN *TRIGONELLA FOENUM-GRAECUM* L.

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ABSTRACT

Antimutagenicity of L-cysteine has been determined against the mutagenicity of diethyl sulphate (DES; pH 7.41) in Trigonella foenum-graecum L. using toxicity to seed germination and seedling growth as the criteria. Diethyl sulphate (mutagenic agent) induces inhibitory effects on the seed germination and led to the formation of abnormal seedlings significantly with increasing concentrations. Diethyl sulphate mutagenized seeds of T. foenum-graecum were post-treated with three different aqueous concentrations of L-cysteine (0.0010M, 0.0050M and 0.0100M) which exhibited antimutagenic activity by way of promotory effects on seed germination as well as on seedling growth as compared to control.

Key words: Antimutagenicity, L-cysteine, diethyl sulphate, *Trigonella foenum-graecum* L.

1. Introduction

Discharge of toxic elements like carcinogens and mutagens from industries, landfills and diffused sources of pollution like fertilizers and pesticides over the year has been resulted in high levels of contamination in food, fodder, vegetables and ground water. By using of food, fodder, vegetables and ground water having carcinogens or mutagen by living beings including human beings over a long period cause mutations. There are considerable evidence that the effects of mutagenic and carcinogenic agents can be altered by many antimutagens. The term 'antimutagen' was used originally to describe those agents which reduce the frequency or rate of spontaneous or induced mutation independent of the mechanism involved [1]. Studies on antimutagenic factors (about 200 compounds) were initially carried out of microbial genetics [2].

Against this background, in the present study, antimutagenicity of L-cysteine has been determined against diethyl sulphate (DES; pH 7.41) induced toxicity to germination and seedling growth as the parameters in *Trigonella foenum-graecum* L.

2. Materials and Methods

Dry pure line viable seeds of *T. foenum-graecum* were surface sterilized with 0.1% (w/v) mercuric chloride ($HgCl_2$) solution for 3 min. The seeds were thoroughly washed with sterilized distilled water so as to remove the traces of mercuric chloride and were presoaked in distilled water for 4 h at 25±1℃.

The experiments were designed to have the following three sets-

(1) Control or Untreated-In this set, some of the presoaked seeds were kept in distilled water for 20 h at 25±1℃.

(2) Treated with diethyl sulphate (DES; pH 7.41) alone- In this set, some of the presoaked seeds were treated with freshly prepared four different aqueous concentrations of diethyl sulphate (0.50%, 1.00%, 2.00% and 2.50%) for a period of 20 h at 25±1°C.

(3) Post-treated with L-cysteine - In this set, some of the treated seeds (seeds treated with each concentration level of diethyl sulphate) were post treated with freshly prepared three different aqueous, L-cysteine concentration (0.0010M, 0.0050M and 0.0100M) for a period of 20 h at 25±1°C.

For each set, 30 seeds were used and were replicated thrice. Seeds of all the experimental sets were transferred to the sterilized petriplates containing moist filter papers. Observations on seed germination, seedling growth and abnormal seedlings were made regularly for a continuous period of 10 days.

3. Results and Discussion

Table-1 shows that the treatment of seeds of *T. foenum-graecum* with different concentrations of diethyl sulphate (0.50%, 1.00%, 2.00% and 2.50%) resulted in varying degrees of reduction in germination percentage and decreases the formation of normal seedlings. Maximum toxicity was recorded in 0.0100% DES treated series where no germination was observed. Approximately 50% lethality in seed germination was recorded in the 1.00% DES treated series. Maximum reduction of normal seedlings as well as reduction in root and shoot length was observed in 2.00% DES treated series. Further, the values of Karl Pearson's coefficient of correlation (r) depicted in Table-1, clearly indicate that decrease in the seed germination, decrease in the formation of normal seedlings and declining root and shoot length were highly dose dependent.

Diethyl sulphate mutagenized seeds, post-treated with L-cysteine separately, led to the improvements in seed germination, formation of normal seedlings and declining in root/shoot injury. Seeds treated with only 2.50% concentration of diethyl sulphate did not show any germination but surprisingly, after post-treatment with L-cysteine separately, showed germination upto a maximum of 16% as well as formation of normal seedlings upto a maximum of 30%, respectively. Maximum promotion in seed germination (99%) was observed in 0.50% DES + 0.0100M L-cysteine treated series. However, minimum recovery in the seed germination (06%) was observed in 2.50% DES+ 0.0010M L-cysteine treated series. The calculated 'r' values at a significance level p=0.05 for using parameters depicted in Table-1 were comparable with the tabulated 'r' values for 3 or 4 degrees of freedom (p=0.05) except 1.00% DES+L-cysteine treated series for seed germination and shoot length in seedlings, hence indicating a very strong correlation between the used concentrations of diethyl sulphate (mutagen) or L-cysteine and effects on the seed germination and growth of seedlings.

Several researchers have carried out similar studies in prokaryotic system by assessing the potential of various mutagens before and after antimutagenic treatments [3, 4 and 5]. Rawat and Mahna [6] have reported antimutagenic/anticarcinogenic activity of Amla and Myroblan fruit extracts in petriplate experiment of *T. foenum-graecum*. Thus it could be concluded that L-cysteine has the potentiality to stimulate the retarded seed germination and seedling growth in mutagenized seeds of *T. foenum-graecum*.

4. Conclusion

Post-treatment of Diethyl sulphate mutagenized seeds of *T. foenum-graecum* with three different aqueous concentrations of L-cysteine (0.0010M, 0.0050M and 0.0100M) showed promotory effects on seed germination as well as on seedling growth which suggests antimutagenic properties of L-cysteine against mutagenicity induced by Diethyl suphate.

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Table 1: Effect of Diethyl sulphate (DES; pH 7.41) alone and in combination (post-treatment) with L-cysteine in *Trigonella foenum-graecum* L.

DES concentrations (concentrations in molars)	Germination (%)	Abnormal seedlings (%)	Shoot length (cm) Mean±SE	Root length(cm) Mean±SE
Control	100	-	4.5±0.52	3.6±0.68
0.50	80	58	3.0±0.65	1.5±0.13
1.00	49	79	2.1±0.59	0.9±0.07
2.00	10	85	0.5±0.029	0.4±0.01
2.50	-	-	-	-
'r' Value	-0.998	- 0.600	-0.994	-0.997
DES (concentrations in %) + L-cysteine (concentrations in molars)	Germination (%)	Abnormal seedlings (%)	Shoot length (cm) Mean±SE	Root length(cm) Mean±SE
0.50%+No L-cysteine	80	58	3.0±0.65	1.5±0.13
0.50%+0.0010	80	51	4.0±0.31	3.5±0.72
0.50%+0.0050	85	42	4.5±0.25	3.7±0.17
0.50%+0.0100	99	25	4.6±0.39	3.8±0.35
'r' Value	0.972	-0.992	0.809	0.670
1.00+ No L-cysteine	49	79	2.1±0.59	0.9±0.17
1.00+0.0010	52	75	2.8±0.39	1.5±0.01
1.00+0.0050	58	70	2.3±0.42	1.7±0.09
1.00+0.0100	50	78	2.2±0.45	1.5±0.07
'r' Value	0.104*	-0.976	0.307*	0.547

2.00+ No L-cysteine	10	85	0.5±0.29	0.4±0.01
2.00+0.0010	14	85	0.7±0.21	0.4±0.09
2.00+0.0050	20	80	0.9±0.25	0.5±0.07
2.00+0.0100	25	78	1.0±0.17	0.8±0.05
'r' Value	0.976	-0.967	0.924	0.969
2.50+ No L-cysteine	-	-	-	-
2.50+0.0010	06	75	0.1±0.08	0.01±0.01
2.50+0.0050	10	80	0.1±0.01	0.05±0.01
2.50+0.0100	16	70	0.2±0.09	0.08±0.01
'r' Value	0.957	0.532	0.892	0.986

Tabulated 'r' for 3 d.f. at p=0.05 is 0.878

Tabulated 'r' for 4 d.f. at p=0.05 is 0.811

Reported values are mean ±SE of 3 replicates

* Non Significant

r= Karl Pearson's coefficient of correlation.