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EFFECT OF CCC AND IAA ON VIABILITY EXTENSION OF LENTIL SEEDS

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ABSTRACT

The present investigation was carried out by using two plant growth regulatory substances (PGRs) where one is plant growth promoter (IAA) and one is plant growth retardant (CCC) on lentil seeds to evaluate the efficacy of the chemicals in respect to germinability and metabolism of the experimental seeds. The major problem of the lentil is its poor seed germination capacity under natural condition which leads to impaired plant growth due to loss of seed vigour and viability status. Initially, percentage germination was recorded by using different concentrations of IAA and CCC, where CCC shows inhibitory effect on percentage germination of lentil seeds. Metabolic status of seeds pre-soaked with the CCC shows reduced status of the dehydrogenase activity, amino acid contents and soluble carbohydrate levels of Lens culinaris. Thus, PGRs potentially modulate the activities of the beneficial enzymes of the seeds over control samples.

KEY WORDS

CCC, IAA, seed viability, Lens culinaris, PGRs, seed germination

INTRODUCTION

In this present investigation, chemical manipulation technique is employed to lentil (*Lens culinaris* L.) seeds by using one growth promoter indole acetic acid (IAA) and one growth retardant chlorocholine chloride (CCC). Lentil seeds exist some problems related to seed germinability, poor seedling establishment and loss of vigour and viability under storage as well as ambient environmental condition. To overcome this impairment of germinability of lentil, growth promoter IAA was employed and growth retardant CCC was treated to the seeds for healthy plant establishment.

Seed vigour and viability is an important index for plant health and thus various physiological and biochemical parameters like percentage germination, TTC stainability, some metabolic status of seeds as well as growth parameters was analysed using chemically hardened lentil seeds. This comprehensive work on chemical-induced modulation of seed vigour and viability status to alleviate the specific problems and improve metabolic status of seeds leads to enhanced growth of the experimental plant.

In fact, reports available in the literature where growth promoters and retardants potentially enhance productivity of many crop plants (1,2,3,4,5,6). Some reliable physiological and biochemical parameters was critically analysed to get an insight of the metabolic status of seeds which includes free amino acid leaching, soluble carbohydrate level, dehydrogenase activity of lentil by chemically treated seeds. Metabolic status of seeds reveals a clear concept about the efficacy of the chemicals as well as the storage potentiation of seeds in ambient environmental condition.

MATERIALS AND METHODS

Experiments of the present investigation were carried out with the fully viable, freshly harvested lentil seeds (*Lens culinaris* L.) are collected from local market of Darjeeling, West Bengal. The chemical manipulating

agents are indole acetic acid (IAA) and chlorocholine chloride (CCC). The concentrations of each chemical manipulating agents used in the experiments are 100 and 500 (µg/ml).

Keeping in mind, the effect of these two chemical agents (IAA and CCC) on lentil seeds with respect to its germination behaviour and metabolism following experiments are as follows:

Germination percentage of seeds:

Seeds were pre-soaked with IAA, CCC at concentrations 100 and 500 (µg/ml) each and distilled water for 24 hours. Percentage germination of seeds was recorded after 48 and 96 hours of seed soaking in distilled water. To analyse the percentage germination, the individual seed lots of 100 seeds of each treatment were transferred to separate Petri dishes containing filter paper moistened with 10 ml distilled water. Germination data were recorded every 48 hours intervals upto 96 hours of seed soaking following the International Rules for Seed Testing (7).

T_{50} values of germination of lentil seeds:

T_{50} values of seeds were recorded after 6 days of seed soaking in distilled water. The time (hours) for 50% germination of seeds (T_{50}) was determined following the method described by Coolbear *et al*. (8).

TTC stainability of lentil seeds:

For analysis of TTC-stainability, Lentil seeds (100 Nos.) of each treatment were allowed to imbibe 0.5% TTC (2, 3, 5-triphenyl tetrazolium chloride) solution (w/v) in Petri dishes for 24 h in dark condition. The percentage TTC-stained (red coloured) seeds were calculated from the total number of seeds of each treatment (9).

Dehydrogenase activity of lentil seeds:

To analyse dehydrogenase activity 20 dehusked seeds were imbibed in distilled water with two replicates for 24 hours at 28^0c in sterilized conditions. Then 20 dehusked imbibed seeds of each treatment including control were immerged in 0.5% TTC solution in test tube and incubated for 12 hours in dark. This method was adopted after Rudrapal & Basu (10).

Free amino acids (from seed leachate):

Free amino acid levels from the seed leachates of each treatment were analysed after immersing 10g seed sample of Lentil in 100 ml distilled water for 24 h. From the leachate stock, free amino acid level was quantified following the method of Moore and Stein (11).

Soluble carbohydrates:

Soluble carbohydrate contents from seed leachate was the same as done in case of leachable amino acids, and from the same leachate stock, soluble carbohydrate level was determined following the method of McCready *et al.* with slight modification (12).

Statistical analyses:

Statistical analysis of the data was done in terms of least significant difference (LSD) which was calculated at 95% confidence limits and as per the method of Panse and Sukhatme (13)

RESULT AND DISCUSSION

The present investigation is an attempt to obviate some specific problems of lentil seeds and to get rid of such problems, chemical manipulative agents IAA and CCC (100 and 500 µg/ml each) is used. Chemical manipulative techniques are adopted which is reported to play important role for overcoming some undesired features of plants (14).

Growth promoter IAA and growth retardant CCC positively influences germination behaviour of lentil seeds by enhancing vigour and viability status of seeds. Influence of seed pre-treatment with IAA (500 µg/ml) shows enhanced germination behaviour whereas CCC (500 µg/ml) retards percentage germination of lentil seeds (Table 1). In case of TTC stainability of lentil seeds, fully viable, partially viable, non-viable percentage and in case of CCC (500 µg/ml), lesser number of fully viable seeds are found (Table 2). It is found that profuse leaching found in case of IAA (500 µg/ml) treated lentil seeds and CCC (500 µg/ml) treated seeds show lesser amount of free amino acid leaching than control (Table 4). In case of dehydrogenase activity and soluble carbohydrate levels of *Lens culinaris* seeds, CCC shows inhibitory effect (Table 3 and Table 5).

Table 1: Influence of seed pretreatment with IAA and CCC (100 and 500 µg/ml each) on percentage germination (%) of *Lens culinaris* seeds.

Treatments (µg/ml)		TTC stainability (%)		
		FV	PV	NV
Control		85	10	5
IAA	100	85	15	0
	500	90	10	0
CCC	100	80	20	0
	500	65	35	0

FV: Fully Viable; PV: Partially Viable; NV: Non-Viable

Table 2: Influence of seed pre-treatment with IAA, CCC (100 and 500 µg/ml each) on TTC stainability (%) of Lens culinaris seeds

Treatments (µg/ml)		Percentage of germination after hours (h)		
		0	48	96
Control		0	72.8	100
IAA	100	0	77.0	100
	500	0	85.2	100
CCC	100	0	47.5	100
	500	0	35.6	100
LSD (P=0.05)		-	2.96	-

Table 3: Influence of seed pre-treatment with IAA and CCC (100 and 500µg/ml each) on the changes of dehydrogenase (ΔOD/g wet wt./5ml) of *Lens culinaris* seeds.

Treatments (µg/ml)		Dehydrogenase activity (ΔOD/g wet wt./5ml)
Control		2.157
IAA	100	1.957
	500	1.412
CCC	100	1.345
	500	1.282
LSD (P=0.05)		0.85

Table 4: Influence of seed pre-treatment with IAA and CCC (100 and 500 µg/ml each) on the changes of leaching of free amino acid contents (µg/ml) of *Lens culinaris* seeds.

Treatments (µg/ml)		Free amino acids (µg/ml)
Control		195.02
IAA	100	122.97
	500	296.79
CCC	100	192.08
	500	151.23
LSD (P=0.05)		9.56

Table 5: Influence of seed pre-treatment with IAA, CCC (100 and 500 µg/ml each) on the changes of leaching of soluble carbohydrate contents (µg/ml) of *Lens culinaris* seeds.

Treatments (µg/ml)		Soluble carbohydrates (µg/ml)
Control		146.4
IAA	100	105.8
	500	154.2
CCC	100	95.5
	500	73.6
LSD (P=0.05)	-	5.85

To overcome the vigour viability status of lentil seeds under storage, the chemical CCC hardened the seeds that's why reduced germination behaviour and metabolic activity leads to plant growth is found in case of CCC treated seeds. Reports available in literatures on plant growth retardant-induced enhanced storage potentiation of many crop seeds (15, 16). The experimental result is also in conformity with the reported observations of some previous workers. Thus, a conclusion can be drawn from the entire investigation is that PGRS (IAA and CCC) can potentially enhance seed viability under storage.

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