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INFLUENCE OF NITRATE SALTS ON SEED GERMINATION AND SEEDLING GROWTH OF *WITHANIA SOMNIFERA* (ASHWAGANDHA) FROM THE INDIAN THAR DESERT

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Withania somnifera is an important medicinal plant of the Indian Thar desert. In the present study one year storage effect of seeds on germination behaviour, seedling growth, root: shoot ratio (R/S ratio), germination value (GV) and vigour index (VI) in comparison with fresh ones have been reported. Seeds were presoaked for 24 h duration with different concentrations (0.25, 0.50, 0.75, 1.00 and 1.25%) of nitrate solutions such as KNO_3, $NaNO_3$, NH_4NO_3, $CoNO_3$ and $ZnNO_3$ to evaluate their effects on above-mentioned various parameters. Results revealed that cent percent germination along with maximum root and shoot lengths in fresh seeds were observed in 0.75% NH_4NO_3 solution. . Maximum root and shoot lengths were observed in one-year-old seeds with the treatments of 0.25% $ZnNO_3$ and 1.0% KNO_3. The maximum R/S ratio in fresh and one-year-old seeds was observed in 0.50% and 0.25% $ZnNO_3$, respectively, while GV in 0.75% $CoNO_3$.

Key words: *Withania somnifera*, nitrate solutions, seed germination, seedling growth, germination value, vigour index GV, VI.

Withaniasomnifera (Linn.) Dunal (Ashwagandha) is an important medicinal plant of Solanaceae family, which grows wild in arid and semi-arid regions. It is widely used for the treatment of several chronic diseases such as arthritis, bronchitis, female sexual disorders, stomach and lung inflammations. It also improves potency and provides sound sleep due to its anti-stress properties. Roots contain withasomine and visamine alkaloids, which have sedative, anti-inflammatory and anti-spasmodic properties (Negi *et al*. 2006). Roots are considered alternative, aphrodisiac, tonic, deobstruent, diuretic, narcotic, abortifacient and used in rheumatism, consumption, debility from old age, etc. They are also prescribed for hiccup dropsy and as a sedative in case of senile debility (Bhakuni and Jain 1995).

The seed has a major role to play in agricultural production. Germination may be defined as an emergence of embryo from the seed by starting a variety of anabolic and catabolic activities, including respiration, protein synthesis and mobilization of food reserves after it has absorbed water. To the seed analyst, germination means the emergence and development from the seed embryo of the essential structures that indicate the seed's ability to produce a normal plant under favourable conditions (Desai *et al*. 1997). A large number of chemical substances such as various nitrate solutions have been reported by various workers for breaking dormancy of seeds, enhancing their permeability, inducing and hastening the germination and thereby acting as chemical regulators for seed germination. Chemical substances may behave as germination stimulators or inhibitors and their effects on imbibitions and germination may vary (Sen 1977).

Thus, in the present investigations an attempt

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Table-1: Effects of different nitrate solutions on seed germination, seedling growth, R/S ratio, germination value (GV) and vigour index (VI) in fresh and one-year-old seeds of *W. somnifera* (values are the mean of six replicates)

Nitrate solutions	Conc. (%)	Germination (%)		Seedling growth (cm)				R/S ratio		GV		VI	
				Root		Shoot							
		Fresh	Old	Fresh	Old	Fresh	Old	Fresh	Old	Fresh	Old	Fresh	Old
Control		93.33	43.33	1.76	0.59	2.22	1.22	0.798	0.506	124.43	14.44	371.45	78.42
KNO_3	0.25	100.00	40.00	1.39	0.99	2.16	3.38	0.648	0.294	125.00	12.30	355.00	174.80
	0.50	100.00	50.00	1.50	1.01	2.21	3.13	0.679	0.325	125.00	20.83	371.00	207.00
	0.75	100.00	56.66	2.22	1.72	2.70	2.99	0.824	0.585	125.00	29.18	492.00	266.86
	1.00	100.00	26.66	2.13	1.08	2.44	3.53	0.888	0.305	125.00	4.73	457.00	122.90
	1.25	100.00	6.66	2.01	1.04	2.32	2.51	0.886	0.416	125.00	0.31	433.00	23.64
CD		NS	12.278**	0.285**	0.222**	0.216**	0.249**	0.152**	0.105**	-	-	-	-
NH_4NO_3	0.25	96.66	73.33	1.87	1.11	2.15	2.71	0.872	0.411	116.78	35.84	388.57	280.12
	0.50	100.00	66.66	1.11	0.96	2.27	2.58	0.498	0.375	111.11	37.02	338.00	235.97
	0.75	100.00	40.00	2.61	1.08	2.88	2.12	0.904	0.514	125.00	12.30	549.00	128.00
	1.00	100.00	16.66	2.12	0.91	2.35	2.00	0.907	0.458	125.00	2.52	447.00	48.48
	1.25	53.33	3.33	2.08	1.06	2.44	2.11	0.867	0.504	31.60	0.07	241.05	10.55
CD		9.723**	19.139**	0.265**	NS	0.225**	0.165**	0.124**	0.075**	-	-	-	-
$ZnNO_3$	0.25	56.66	13.33	1.97	2.30	2.19	2.70	0.910	0.858	108.88	1.61	421.85	66.65
	0.50	93.33	30.00	1.92	1.84	2.09	2.98	0.924	0.621	45.86	7.50	227.20	144.60
	0.75	100.00	23.33	2.27	1.16	2.59	2.52	0.879	0.470	125.00	4.94	486.00	85.85
	1.00	96.66	16.66	1.60	1.03	2.07	1.73	0.775	0.608	103.81	2.52	354.74	45.98
	1.25	90.00	13.33	1.37	0.91	1.80	1.61	0.778	0.565	90.00	1.48	285.30	33.59
CD		8.420**	9.722*	0.174**	0.167**	0.189**	0.237**	0.120*	0.101**	-	-	-	-
$CoNO_3$	0.25	93.33	3.33	1.24	0.86	1.48	2.13	0.843	0.406	124.43	0.12	253.85	9.95
	0.50	100.00	53.33	1.51	0.93	1.97	2.24	0.771	0.421	125.00	18.96	348.00	169.05
	0.75	100.00	86.66	1.58	1.36	2.00	3.04	0.794	0.454	166.66	57.76	358.00	381.30
	1.00	13.33	36.66	1.34	0.93	1.71	2.50	0.790	0.373	2.22	12.21	40.65	125.74
	1.25	10.00	36.66	1.34	1.32	1.60	2.62	0.847	0.505	1.11	8.95	29.40	144.44
CD		13.963**	17.358**	0.198**	0.171**	0.124**	0.221**	NS	0.077*	-	-	-	-

NS - Non-significant; and * & ** = Significant at P = 5 &1%, respectively.

have been made to asses the effect of different concentrations of various nitrate salt solutions such as potassium nitrate (KNO_3), ammonium nitrate (NH_4NO_3), sodium nitrate ($NaNO_3$), zinc nitrate ($ZnNO_3$) and cobalt nitrate ($CoNO_3$) on seed germination, seedling growth, R/S ratio, germination value (GV) and vigour index (VI) in *W. somnifera* under controlled laboratory conditions.

MATERIAL AND METHODS

The seeds of *W. somnifera* were collected from the Nagaur (135 km away from JN Vyas University Campus, Jodhpur in north direction) in March-April during 2008 and 2009. For seed collection, mature red fruits ready for dispersal were collected and stored in paper bags. Dry seeds were cleaned and stored in plastic containers with insecticide/parad tablets to protect them from insects.

Seeds germination behaviour was studied in fresh and one-year-old seeds. For germination studies, seeds were placed in sterilized Petri dishes lined with single layer of filter paper, moistened with distilled water as and when needed. The experimental Petr dish contained 10 seeds in triplicate and the experiments were repeated twice. The germination experiments were performed in alternate white light and dark (12 h) obtained from 3 fluorescent tubes of 40 watts each fitted at a height of half meter from the Petri dishes (1000 lux) at 28º C in seed germinator. After 10 days of setting the experiments, seed germination (%) and root & shoot length of seedlings were measured.

Seeds were presoaked for 24 h with different concentrations (0.25, 0.50, 0.75, 1.00 and 1.25%) of nitrate solutions, *viz.* KNO_3, $NaNO_3$, NH_4NO_3, $CoNO_3$ and $ZnNO_3$. The seeds treated with nitrate solutions were kept for germination studies. Germination values of

seeds were calculated for each treatment as per Czabator (1962); GV = PV × MDG

Where, PV= Peak value of germination; and MDG = Mean daily germination

PV is calculated with following formula:

$$PV = \frac{\text{Final germination percentage}}{\text{No. of days that took to reach the peak germination}}$$

MDG is calculated as follows:

$$MDG = \frac{\text{Total germination percentage}}{\text{Total number of days}}$$

The vigour index (VI) was derived from the formula given by Abdul-Baki and Anderson (1973), which is as follows:

VI= Percentage of germination × Seedling length (cm)

Where, seedling lengths are the sum of root and shoot lengths.

The mean values of data obtained from six replicates during the two years (2004 & 2005) for each parameter were executed using CRD design and subjected to analysis of variance (ANOVA) as suggested by Gomez and Gomez (1984).

RESULTS AND DISCUSSION

The data on effect of the presoaked seeds with different nitrate salt solutions on germination percentage, seedlings growth and various parameters are given in Table 1. Freshly harvested seeds showed cent percent germination along with maximum root and shoot lengths in 0.75% concentration of NH_4NO_3, $ZnNO_3$ and $CoNO_3$ solutions. Among all nitrate solutions, the maximum root (2.61 cm) and shoot (2.88 cm) lengths were observed in 0.75% NH_4NO_3. In case of one-year-old seeds, maximum germination (86.66%) was observed in 0.75% of $CoNO_3$ solution. Maximum root (2.30 cm) and shoot (3.53 cm) lengths were recorded in 0.25% $ZnNO_3$ and 1.0% KNO_3, respectively. This may be due to combination of these nitrate salts with an osmotic role on water uptake, which exerted a nutritional effect on protein synthesis as suggested by Mc Intyre *et al.* (1996).

The highest R/S (root/shoot)length ratio in fresh (0.924) and one-year-old seeds (0.858) was observed in 0.50 and 0.25% $ZnNO_3$, respectively. The maximum GV (166.66) and VI (549.00) were obtained in 0.75% $CoNO_3$ and NH_4NO_3 treatments, respectively in fresh seeds. In one-year-old seeds, GV was maximum (57.76) in 0.75% $CoNO_3$ and minimum (0.07) in 1.25% NH_4NO_3 solutions.

The fresh and one-year-old seeds did not germinate at all in $NaNO_3$ pretreatment, which probably may be due to toxicity of this salt to the embryo.

Data were significant at 5 and 1% probability levels, except for germination percentage and R/S ratio in fresh seeds and root length in one-year-old seeds, which were non-significant.

Numerous workers have investigated the effect of KNO_3 and similar substances on the germination of a variety of seeds and have obtained conflicting results, stimulation or inhibition being obtained depending on the concentration of nitrogen sources and the type of seed used. Nitrogen sources such as KNO_3 and NH_4NO_3 when applied to seeds, promotes seed germination and seedling growth in *Plantago ovata* at lower concentrations, but inhibits at higher concentrations (Choudhary and Kumar 2003). KNO_3 and NH_4NO_3 favoured seed germination and seedling growth in *Plumbago zeylanica* (Sharma and Kumar 2003). KNO_3 significantly stimulated seed germination, seedling vigour and reduced mean germination time in *Angelica glauca* (Butola and Badola, 2004). In the present studies, seed germination and seedlings growth in *W. somnifera* enhanced differentially by application of various concentrations of nitrogen sources. Lower concentrations

promoted the germination and seedling growth, while higher ones retarded them. So finally, it is concluded from the present findings that for large-scale multiplication, freshly collected presoaked seeds of *W. somnifera* with 0.75% solutions of KNO_3, NH_4NO_3 and $CoNO_3$ should be used.

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