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Influence of different seed pre -treatments on germination attributes of common bean (*Phaseolus vulgaris*), spinach (*Spinacea oleracea*) and okra (*Abelmoschus esculentus*)

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Abstract

Present studies entitled "Influence of different seed pre-treatments on seed germination attributes of common bean (Phaseolus vulgaris), spinach (Spinacea oleracea) and okra (Abelmoschus esculentus)" was carried out in the Department of Horticulture, Lovely Professional University, Phagwara, Punjab, during 2017-2018. The experiment wasconducted under laboratory condition in petri plates. The seeds of common bean, spinach and okra were treated with different pre-treatments agents viz. T_1 (GA_3@25ppm), T_2 (Vermiculite), T_3 (Distilled Water), T_4 (Cow urine @200ml/l) and T_5 (Control). All the germination parameters were observed maximum viz; speed of germination (13.78), germination percentage (72.88%), seedling length (8.53cm), seedling fresh weight (0.093mg), seedling dry weight (0.010mg), moisture contents (90.67%) and seed vigor index (535.1) when seeds were treated with GA_3 @ 25 ppm followed by T_4 (Cow urine @200ml/l). Minimum values for all the germination parameters were recorded in control seeds.

Key words: *Cow urine, germination, gibbrellic acid, seed pre-treatments, vegetable crops*

Introduction

Vegetables contribute majorly in human diet as major nutrition component with abundant phytochemicals; vitamins, minerals, dietary fibers as well as aminoacids and proteins (Quebedeaux and Eisa, 1990). Among all the vegetables consumed worldwide, green leafy vegetables are widely consumed in every household as a good sources of iron, phosphorous, calcium, carotene, folate and vitamin A. In India, spinach (*Spinacea oleracea*) is one of the most nutritious leafy vegetables available and affordable by common people.Spinach is extensively

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cultivated throughout North America, South America, Europe and temperate regions of Asia (Dhaliwal, 2017).Other economically important vegetables in India is okra(*Abelmoschus esculentes*)which is widely cultivated in different parts of the country as well in other countries like Nigeria, Sudan, Pakistan, Egypt and Saudi Arabia. Okra has a high nutritional value and grows very quickly at high temperatures, which lends its production to more tropical parts of the world (Costa *et al.,* 1981).Other important group of vegetable widely consumed in India is 'legumes' among which common bean (*Phaseolus vulgaris*) is widely cultivated in different parts of the world and represents one of the largest food components in Latin America and Africa. This crop is economically important for these countries as a major income source for small farmers (Pachico 1989). Common bean is grown worldwide as an important vegetable for its high quality, nutritional and sensory properties and as a grain legume for its major protein source and economic value (Martins and Silva, 2004; Sarikamis *et al.,* 2009).One of the concerning impediment related to cultivation of these major crops is poor seed germination. Hard seediness has been reported in okra which leads to its poor field establishment. Similarly, germination rates of spinach are often low (<60%) due to hard seededness induced by long storage periods.The bean seedsalso show loss in viability when stored at room temperature for five years along with lower rates of germination in remaining viable seeds. Further seedling growth is poor with smaller leaves and retarded chlorophyll development (Grubben, 2004).Therefore, to solve the problem concerning germination of these nutritionally important vegetable crops, certain techniques need to be employed that guarantees their seed viability for longer time, improve emergence and enhance vigor. Seed pre-treatments with certain chemical and biological agents are widely employed in many crops which refers to the exposure of seeds to certain physical, chemical or biological agents which enhance the pre germination metabolic events that leads to improvement in germination and also makes the seeds less susceptible to pests and diseases (Forsberg *et al.,* 2003).Seed treatment with GA_3 @100ppm for 3-4 hours increases the germination percentage and seedling length in okra crop (Bello, 2005). Whereas,in common bean, seed soaking with 10% cow urine for 24 hours increased the germination percentage, seedling length, seedling fresh and dry weight as compared to control(Rao, 1975). In spinach, seed germination has been improved applying different treatments like $NaOCl/H_2O_2$ (Katzman *et al*., 2001), leaching in water (Atherton and Farooque, 1983). So, keeping in mind

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the commercial importance of these vegetables and potential of seed pre-treatments inalleviating germination constraints, present studies were planned and conducted.

Materials and methods

The experiment was conducted in laboratory of the vegetable science, Lovely Professional University, Phagwara, Punjab to evaluate the effect of seed pre-treatments on seed germination attributes of common bean, spinach and okra. The experiment consisted of five different seed pre-treatments viz., GA_3@ 25ppm (T_1), Vermiculite (T_2), Distilled water (T_3), Cow urine@ 200ml/l (T_4) along with Control (T_5) for different vegetable crops viz., common bean, spinach and okra.

Seed pre-treatment: Seeds were placed in single layer of filter paper in petri plates. The filter paper was moistened with about 5 ml solution of different agents (GA_3@ 25ppm (T_1), Distilled water (T_3), Cow urine@ 200ml/l (T_4))ensuring that the seeds were immersed with solution allowing one surface absorption. Solid matrix carrier i.e. vermiculite (T_2) was thoroughly mixed with one part of water and then seeds wrapped in muslin cloth are kept in layers of vermiculite as per treatment. Petri plates with seeds immersed in different agents are kept in seed germination chamber at a temperature 20^0C to 25^0C with relative humidity 50% to 65% for 24 hrs as per treatments and replications. After 24 hrs seeds were taken out and washed with distilled water and left for drying at room temperature in dried petri plates. For control no treatment is given, purchased seeds were directly sown for germination studies.

Germination test: The germination experiment was conducted in growth chamber (seed germinator) at temperature 25^0C + 60% RH for okra and 15^0C + 50% RH for common bean and spinach.Already treated seeds with different seed treatment agents were placed for germination in petri plates containing single layer of filter paper with 5 ml of distilled water (that is enough to moisten the filter paper) according to the treatments and replication. Fifty seeds were kept per replication. The seeds were considered to have germinated after radicle emergence. Germination count were taken for 12 days in okra (*Abelmoschus esculetus*) (V_1), 09 days in spinach (*Spinacea oleracea*) (V_2) and 08 days in common bean (*Phaseolus vulgaris)* (V_3).Observations were recorded for all the germination parameters viz; speed of germination, germination percentage, seedlings length, seedlings fresh weight, seedlings dry weight, moisture contents and vigor

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index.The experiment was carried out in completely randomized design with three replications. Data collected under laboratory conditions (in petri plates) wasanalyzed using OPSTAT software.

Results and discussion

The findings of the present study indicated that differentseed pre-treatments had significant effects on the germination attributes of okra, spinach and common bean viz., speed of germination, germination percentage, seedling length, seedling fresh weight, seedling dry weight, moisture contents and vigor index. Among different seed pre-treatments (Table 1), T_1(GA_3@25ppm) resulted in maximum values for all the germination attributes viz., speed of germination (13.78), germination percentage (72.88%), seedling length (8.53cm), seedling fresh weight (0.093mg/seedling), seedling dry weight (0.010mg/seedling), moisture contents (90.67%), and vigor index (535.1)followed by T_4 (cow urine@200ml/l) whereas all the germination attributes were recorded minimum in T_5 (control). Improved germination GA_3 @25 ppm might be due to fact that gibberellic acid increases the activity of hydrolytic enzymes during germination process which gets diffused into endosperm, where they catalyze the digestion of stored food material into sugars, amino acids which ultimately fasten the process of germination or emergence in treated seeds. GA_3 enhances the water uptake of the seedling which might have activated enzymes with an accompanying mobilization of reserve materials in the embryo and stronger seedlings were obtained as a result of better embryo growth. Similar observations were recorded by Gallardo *et al.* (2002) andZhao*et al.* (1995). Along with gibberellic acid cow urine has also been proved to be better in enhancing germination in all the crops studied. Physiologically active factors responsible for activity of cow urine are growth regulators, nutrients and trace elements. Cow urine is composed mainly of water (95%), urea (2.5%) along with some proportion (2.5%) of salts, hormones, enzymes, and minerals and its growth promoting effects has been reported by Amit *et.al.*(2017) in papaya seedlings. On soaking the seeds in cow urine solution, cholophyll contents and soluble proteins were found to be increased in *Albizzia lebbeck* seeds (Illango*et. al*., 1999). Cow urine @ 2% was found effective in improving germination parameters in cluster bean (Tagore *et. al*., 2017).

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Table 1: Effect of different seed pre-treatments on seed germination attributes of common bean, spinach and okra

Seed pre-treatments	Speed of Germination	Germination percentage (%)	Seedling length (cm)	Seedling fresh weight (mg/seedling)	Seedling dry weight (mg/seedling)	Moisture content (%)	Vigor index
T_1	13.78	72.88	8.53	0.093	0.010	90.67	535.1
T_2	11.33	66.33	5.00	0.043	0.009	86.12	320.7
T_3	12.18	66.44	5.51	0.059	0.009	89.30	350.4
T_4	13.08	71.22	6.14	0.068	0.009	90.14	500.8
T_5	11.05	64.22	4.38	0.055	0.007	89.75	324.1
S.E. (m.)±	0.187	0.873	0.044	0.004	0.001	0.682	1.999
C.D. at 5%	**0.542**	**2.535**	**0.126**	**0.011**	**0.002**	**1.978**	**5.801**

*T_1 (GA_3@ 25ppm), T_2 (Vermiculite), T_3 (Distilled water), T_4 (Cow urine), and T_5(Control)

Among different crops(Table 2), germination percentage (90.40%), seedling length (8.74cm) and vigor index (842.4) were found to be maximum in V_1(okra) crop, while speed of germination (14.59), and moisture contents (92.67%) found to be maximum in V_2 (spinach) crop whereas seedling fresh weight (0.14 mg/seedling) and seedling dry weight (0.026 mg/seedling) were found to be maximum in V_3 (common bean) crop. The differences in all germination attributes might be due to the varying seed physiological activity and genetic variation among the crop seeds.Similar observation were recorded byOjeifo*et al.* (2008) andKhan *et al.* (2015).

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Table 2: Effect of different vegetable crops (common bean, spinach and okra)on theirgermination attributes

Crops	Speed of Germination	Germination percentage (%)	Seedling length (cm)	Seedling fresh weight (mg/seedling)	Seedling dry weight (mg/seedling)	Moisture content (%)	Vigor index
V_1	14.49	90.40	8.74	0.13	0.018	86.63	842.4
V_2	14.59	80.80	7.38	0.01	0.001	92.67	591.7
V_3	14.54	80.80	8.51	0.14	0.026	82.06	692.0
S.E. (m.)±	0.145	0.677	0.034	0.003	0.000	0.528	1.548
C.D. at 5%	**N/A**	**1.595**	**0.252**	**0.007**	**0.001**	**1.606**	**36.305**

*V_1 (Okra), V_2 (Spinach) and V_3 (common bean)

The interaction of different seed pre-treatments and crops (Table 3) showed non- significant results for germination percentage whereas for speed of germination, seedling length, seedling fresh weight, seedling dry weight, moisture contents and vigor index,results were significant. Maximum speed of germination (15.25) was recorded in T_1V_1(okra seeds treated with GA_3@25ppm); germination percentage (77.33%), seedling length (8.94cm), seedling fresh weight (0.0263 mg/seedling) and seedling dry weight (0.029 mg/seedling) was recorded maximum in T_1V_3 (common bean seeds treated with GA_3 @25ppm) whereas,maximum moisture contents (94.03%) were recorded in T_4V_1 (okra seeds treated with distilled water); vigor index (597.7) was recorded maximum in T_1V_2 (spinach seeds treated with GA_3 @25ppm). Results of interaction between treatments and crops are varying due to different physiological and inherent genetic potential to respond to different stimulus but overall maximum germination parameters were found maximum in treatments with gibberellic acid followed by cow urine.

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Table 3: Interaction effect of different seed pre-treatments and crops on germination attributes of common bean, spinach and okra

Seed pre-treatments	Speed of Germination	Germination percentage (%)	Seedling length (cm)	Seedling fresh weight (mg/seedling)	Seedling dry weight (mg/seedling)	Moisture content (%)	Vigor index
T_1V_1	15.25	71.00	8.12	0.008	0.001	91.92	439.5
T_1V2	12.15	70.33	8.54	0.008	0.001	90.93	597.7
T_1V3	13.95	77.33	8.94	0.263	0.029	89.17	568.2
T_2V1	13.14	64.00	3.13	0.007	0.001	90.72	201.2
T_2V2	9.92	69.33	5.31	0.006	0.001	90.55	382.1
T2V3	10.93	65.66	6.56	0.116	0.026	77.10	378.8
T3V1	14.16	65.00	4.03	0.007	0.000	94.03	233.8
T3V2	10.76	67.00	6.21	0.008	0.001	89.54	394.8
T3V3	11.62	67.33	6.28	0.162	0.025	84.34	422.6
T4V1	14.86	70.66	5.01	0.009	0.001	92.93	394.9
T4V2	11.58	71.66	6.34	0.007	0.001	90.42	579.3
T4V3	12.78	71.33	7.07	0.189	0.024	87.07	528.4
T5V1	13.05	62.00	2.75	0.006	0.000	92.55	200.6
T5V2	10.47	64.66	5.11	0.008	0.001	90.04	378.0
T5V3	9.63	66.00	5.28	0.150	0.019	86.64	393.7

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S.E. (m.)±	0.323	1.513	0.075	0.006	0.001	1.181	3.462
C.D. at 5%	**0.938**	**N/A**	**0.219**	**0.019**	**0.003**	**3.426**	**10.047**

*T-Treatments (T_1-GA_3@ 25ppm, T_2-Vermiculite, T_3-Distilled water, T_4-Cow urine, and T_5-Control); V-Vegetable crops (V_1-Okra,V_2-Spinach and V_3- Common bean)

Conclusions

From the present studies, it can be concluded that seed treatments has the potential to overcome germination barriers. On commercial scale Gibberellic acid treatment can be used for improving germination in common bean, spinach and okra however, farmers can also practice on farm seed treatment with cow urine owing to its better effects on germination along with cost effectiveness easy affordability and eco-friendly organic approach.

Authors' Contributions

Gurpreet Saini has done the actual experiment in laboratory, recorded observations and prepared the initial manuscript. Rishu Sharma has planned and guided the work. Thaneshwari, Neelam Thakur and HomrajAnandraoSahare have helped in guidance and preparation of final manuscript.

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