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Response of different wheat varieties towards *Azospirillum* and phosphate solubilizing bacteria (PSB) seed inoculation

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Abstract: Present investigation was undertaken to utilize *Azospirillum* and Phosphate Solubilizing Bacteria (PSB) to avail atmospheric nitrogen and soluble phosphate in soil for the wheat crop. The sonalika variety (V_1) responded best towards seed inoculation by *Azospirillum* and Phosphate Solubilizing Bacteria (PSB) for the character speed of germination which is regarded as very important indicator of seed vigour. Germination percentage for untreated control was 86.6% after one month of storage and found to be reduced to 76.67% after three month of storage but for treated *Azospirillum* in case of sonalika it was retained at 83.33%. V_1T_1, V_2T_1 and V_3T_1 recorded improved germination even after six month of storage. The seedling characters like shoot and root length was greatly improved with T_1 and T_2 for almost the varieties included in this programme. The character vigour index greatly enhanced in case of V_1T_1, V_1T_2, V_3T_1 and V_3T_2 after one month and six months of storage. This work reported utilization of *Azospirillum* and PSB as seed inoculation for wheat varieties to enhance seed storability and seedling growth parameter. It can be concluded that, *Azospirillum* and PSB seed inoculation can be recommended for wheat for better seedling growth storability of seed

Keywords: *Azospirillum,* PSB, Seed inoculation, Varieties, Wheat

INTRODUCTION

Wheat (*Triticum aestivum L.*) can be classified as winter or spring growth habit based on flowering responses to cold temperatures. Biofertilizer like *Rhizobium, Azotobacter, Azospirillum* and blue green algae have been used for many years. *Azospirillum* inoculants are recommended mainly for wheat, sorghum, millets, maize, sugarcane and vegetable crops. Biofertilizer is defined as a substance which contains living organisms that when applied to seed, plant surface, or soil, colonize the rhizosphere or the interior of plant and promote growth by increasing supply or availability of primary nutrients to the host plant (Vessey, 2003). Biofertilizers are well recognized as an important component of integrated plant nutrient management for sustainable agriculture and hold a great promise to improve crop yield (Narula *et al*., 2005). Plant growth promoting rhizobacteria are free living microorganisms having beneficial effects on plants by colonizing their roots. They include such effects as the production of phytohormones; auxin, cytokinins and gibberellins (Garcia *et al*., 2001), enhancing release of the nutrients (Nautiyal *et al*., 2000). *Azotobacter* and *Azospirillum* have previously significantly increased wheat and barley yield in irrigated as well as in rainfed crops (Pauw De *et al*., 2008). To maintain the production potential as well as seed or grain quality of wheat, a concrete production technique is essential. One of the major essential elements for growth of plants is nitrogen. The Green Revolution (GR) technology adoption between 1960 to 2000 has increased wide varieties of agricultural crop yield per hectare which increased 12-13% food supply in developing countries. Nitrogen is required in large quantities for plants to grow, since it is the basic constituents of proteins and nucleic acids). Bio-fertilizers are the formulation of living organisms, which are able to fix atmospheric Nitrogen in the available from plants either by living frequency in soil or being associated symbiotically with plants (Subba Rao *et al*., 1993). *Azospirilum*, a bacterial fertilizer is highly beneficial micro-organism for cereals, cotton, plantation crops and other crops. The main function of *Azospirillum* inoculation is to assimilate atmospheric nitrogen and fix in soil and finally providing the growing plant. The aim of present investigation was to study the effect of *Azospirillum* and PSB inoculation on different seed quality parameters like speed of germination, storability and different seedling parameters like fresh and dry weight, seedling length and vigour index.

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MATERIALS AND METHODS

An experiment was carried out during the year of 2012 -2013 to 2013-2014 in the department of Seed Science and Technology, Faculty of Agriculture, Bidhan Chandra Krishi Viswavidyalaya, Mohanpur, Nadia, West Bengal. The experiment was done with three treatments and three replications with four varieties. The varieties were: V_1 (Sonalika), V_2 (PBW 443), V_3 (HD 2821) and V_4 (K 9107) and the treatments were- T_1 (*Azospirillum*), T_2 (PSB) and T_3 (Control) with thrice replicates. The data so obtained as described earlier were subjected to statistical analysis by Analysis of Variance method (Gomez and Gomez, 1984). The Standard Error mean (SEm±) and the value of critical difference (CD) were computed to compare the difference between means have been provided in the tables of results. The data on germination and field emergence were transformed into angular (arcsine) values (Snedecor and Cochran, 1967) transformed values are given in the parenthesis and then subject to statistical analysis.

The statistical calculations were prepared by Factorial Design for analysis of data linked to varietal consequence and Completely Randomized Design (CRD) for laboratory data. All statistical analyses were done using SPSS (version 10.0, 1990) on a desktop Computer. The initial seed moisture content was 12%. The seeds were stored in paper packet for one, three and six months of storage. The following seed quality parameters are recorded in the lab like Germination Percentage, Speed of germination, Root length, Shoot length, Seedling Fresh Weight, Dry Weight and Vigour index. The analysis of variance method (Cochran and Cox, 1963) was followed to analyze various data statistically. The significance of different sources of variation was tested by "Error Mean Square Method" of Fisher Snedecor's 'F' test at probability level 0.05. Standard error of Mean (SEm±) and critical difference (C.D.) have been estimated to compare the differences between means.

RESULTS AND DISCUSSION

The influence of different seed treatment on different varieties of wheat, particularly on the speed of germination which is regarded as very important indicator of seed vigour, found that the variety sonalika (V_1) was the best towards *Azospirillum* treatment (T1) and in treatment in PSB (T_2), V1 was best (Table 1). Speed of germination after one month of storage recorded highest volume in V_1T_1 and V_1T_2 than control. After three months of storage V_1T_1 and V_3T_2 responded better than control (Table 1). Response of different varieties after six months of storage was greatly pronounced in V_1T_1, V_1T_2, V_2T_2 and V_3T_2 for the character speed of germination (Table 1).

Germination percentage for control, after one month of storage was 86.67% which was found to be reduced to 76.67%, after three months of storage in case of V_1 but for T_1 it was retained at 83.33% after three months of storage (Table 2). V_1T_1, V_2T_1 and V_3T_1 recorded improved germination percentage than control. After six months of storage, control (T_3) exhibited loss of germination percentage to the extent of 63.33% in case of V_3 whereas the V_3T_1 and V_3T_2 maintained at 73.33% (Table 2). In case of soybean after 30 days plant maximum seed germination observed by biofertilizer treated plant (82.85%) and minimum by chemical fertilizer treated plants (0%) stated by Javed and Panwar (2013). Similar results were also reported by Strelec *et al*. (2010) to indicate significant influence of storage conditions on moisture content, germination and vigour changes during storage of wheat seeds, as well as varietal dependence of seed viability recorded. Response of different wheat varieties towards the character shoot length recorded high value for V_1T_1, V_1T_2 than control (Table 3). Similarly response was also obtained for V_2T_1, V_2T_2, V_3T_1 and V_3T_2 after three months of storage. After six months of storage, treatment effect towards shoot length was better for V_1T_1, V_1T_2, V_2T_1, V_2T_2, V_3T_1, V_3T_2 and V_4T_1 than control. Indicating *Azospirillum* and PSB seed treatment has the ability for improving seedling shoot length. Similar higher values were also recorded for V_1T_1, V_3T_1, V_3T_2 than control (Table 3). The biofertilizer resulted in maximum reduction of seed rot and foot/root rot (*Fusarium oxysporum*) of bushbean stated by Khalequzaman and Hossain (2008).

The character root length was greatly improved by T_1 and T_2 for V_1 but for V_3T_1 and V_4T_2 the results were lower than control. Enhanced root length was also found after six months of storage. Higher values were recorded for V_1T_1, V_1T_2 and V_4T_1 than control (Table 4). Chandrasekhar (2003) observed that the plant growth parameters viz., root length and number of leaves per plant in green gram plants at 45 Days were significantly increased due to inoculation of P-solubilizing fungal strains along with rock phosphate application as compared to rock phosphate alone (control).

Wheat varieties responded better towards seed treatment for the character fresh seedling weight after one month of storage for V_1T_1, V_1T_2, V_2T_1, V_2T_2, V_3T_1, V_3T_2 and V_4T_1, V_4T_2 than control (Table 5). After six months of storage, the seed treatment effect was observed for V_1T_1, V_1T_2, V_3T_1, V_3T_2 and V4T_1, V_4T_2 but slightly reduced in V_2T_2. The variety-treatment interaction was greatly positive in V_1T_1, V_1T_2, V_3T_1, V_3T_2 and V_4T_1 and V_4T_2 (Table 5). Mahfouz and Sharaf eldin (2007) have suggested the use of biofertilizers to increase the fresh weight of corn. The seedling dry weight, which is regarded as a great contributer towards seed vigour was also greatly enhanced by seed treatment. After one month of storage, V_1T_1, V_1T_2 recorded such type of enhancement response of variety was for to be best both after three and six months of

Table 1. Biofertilizers effect on speed of germination at after one, three and six months of storage (Three replicates).

Speed of germination												
V/T	After one month of storage				After three months of storage				After six months of storage			
	T1	T2	T3	Mean	T1	T2	T3	Mean	T1	T2	T3	Mean
V1	2.26	2.33	2.20	2.26	2.20	2.06	2.07	2.11	2.08	1.93	1.82	1.94
V2	1.78	2.27	2.30	2.12	1.63	2.14	2.29	2.02	1.49	2.08	1.94	1.84
V3	2.07	2.23	1.75	2.02	1.80	2.29	1.84	1.97	1.45	1.90	1.63	1.66
V4	2.07	2.30	2.45	2.27	2.00	2.09	2.29	2.13	1.66	2.04	2.06	1.92
Mean	2.04	2.28	2.18		1.91	2.14	2.12		1.67	1.99	1.86	
	V	T	VXT		V	T	VXT		V	T	VXT	
S.Em(±)	0.072	0.063	0.125		0.028	0.024	0.049		0.023	0.020	0.039	
CD(P=0.05)	0.211	0.183	0.366		0.082	0.071	0.143		0.066	0.058	0.115	
CD(P=0.01)	0.836	0.724	1.447		0.326	0.283	0.565		0.263	0.228	0.455	

V_1-Sonalika, V_2- PBW 443, V_3, HD- 2821 and V_4- K 9107 and the treatments were- T_1-*Azospirillum*, T_2- PSB and T_3- Control.

Table 2. Biofertilizers effect on germination percentage at after one, three and six months of storage (Three replicates).

Germination %												
V/T	After one month of storage				After three months of storage				After six months of storage			
	T1	T2	T3	Mean	T1	T2	T3	Mean	T1	T2	T3	Mean
V1	96.67	96.67	86.67	93.33	83.33	83.33	76.67	81.11	76.67	73.33	70.00	73.33
V2	96.67	86.67	96.67	93.33	83.33	73.33	83.33	80.00	76.67	56.67	73.33	68.89
V3	96.67	93.33	93.33	94.44	83.33	83.33	76.67	81.11	73.33	73.33	63.33	70.00
V4	80.00	83.33	90.00	84.44	70.00	70.00	76.67	72.22	56.67	56.67	63.33	58.89
Mean	92.50	90.00	91.67		80.00	77.50	78.33		70.83	65.00	67.50	
	V	T	VXT		V	T	VXT		V	T	VXT	
S.Em (±)	2.222	1.925	3.849		2.003	1.735	3.469		2.079	1.800	3.600	
CD (P=0.05)	6.486	5.617	11.234		5.847	5.063	10.127		6.067	5.254	10.509	
CD (P=0.01)	25.656	22.219	44.437		23.126	20.028	40.055		1.468	1.271	2.542	

V_1-Sonalika, V_2- PBW 443, V_3, HD- 2821 and V_4- K 9107 and the treatments were- T_1-*Azospirillum*, T_2- PSB and T_3- Control.

Table 3. Biofertilizers effect on shoot length (cm) at after one, three and six months of storage (Three replicates).

Shoot length (cm)												
V/T	After one month of storage				After three months of storage				After six months of storage			
	T1	T2	T3	Mean	T1	T2	T3	Mean	T1	T2	T3	Mean
V1	15.47	11.37	10.17	12.34	14.43	10.47	9.53	11.48	10.62	5.89	6.00	7.50
V2	15.67	14.99	12.27	14.31	12.93	12.50	11.73	12.39	8.72	8.20	7.22	8.05
V3	15.07	16.07	10.30	13.81	13.67	13.83	10.43	12.64	9.45	9.85	5.99	8.43
V4	14.00	9.34	9.09	10.81	12.30	8.90	9.00	10.07	8.19	5.37	5.33	6.30
Mean	15.05	12.94	10.46		13.33	11.43	10.18		9.25	7.33	6.14	
	V	T	VXT		V	T	VXT		V	T	VXT	
S.Em(±)	0.282	0.245	0.489		0.125	0.109	0.217		0.127	0.110	0.220	
CD(P=0.05)	0.825	0.714	1.428		0.366	0.317	0.634		0.371	0.321	0.643	
CD(P=0.01)	3.261	2.824	5.649		1.447	1.253	2.506		1.468	1.271	2.542	

V_1-Sonalika, V_2- PBW 443, V_3, HD- 2821 and V_4- K 9107 and the treatments were- T_1-*Azospirillum*, T_2- PSB and T_3- Control.

Table 4. Biofertilizers effect on root length (cm) at after one, three and six months of storage (Three replicates).

Root length (cm)												
V/T	After one month of storage				After three months of storage				After six months of storage			
	T1	T2	T3	Mean	T1	T2	T3	Mean	T1	T2	T3	Mean
V1	13.36	13.43	11.38	12.72	11.93	12.53	10.83	11.77	10.87	11.47	9.00	10.44
V2	15.03	13.83	14.87	14.58	12.80	12.80	13.33	12.98	11.67	11.80	12.03	11.83
V3	12.42	13.47	13.00	12.96	12.20	12.93	12.27	12.47	10.30	10.33	10.67	10.43
V4	15.40	10.17	11.09	12.22	13.80	9.93	9.97	11.23	12.60	8.30	8.33	9.74
Mean	14.05	12.73	12.58		12.68	12.05	11.60		11.36	10.48	10.01	
	V	T	VXT		V	T	VXT		V	T	VXT	
S.Em(±)	0.207	0.179	0.358		0.165	0.143	0.286		0.124	0.107	0.214	
CD(P=0.05)	0.604	0.523	1.045		0.482	0.418	0.835		0.361	0.313	0.626	
CD(P=0.01)	2.387	2.068	4.135		1.907	1.652	3.303		1.428	1.237	2.474	

V_1-Sonalika, V_2- PBW 443, V_3, HD- 2821 and V_4- K 9107 and the treatments were- T_1-*Azospirillum*, T_2- PSB and T_3- Control.

storage (Table 6). The similar work was done in case of corn and showed the biofertilizers increased the dry weight proposed by Mahfouz and Sharaf eldin (2007). The study revealed that maximum germination (92%), viability (95%), germination speed (1.58), germination energy (70.0%) were recorded in polybags under complete darkness in *Aconitum heterophyllum* wall ex. Royle while minimum were recorded under partial light in cloth bags. Similarly, in case of *Podophyllum hexandrum* same results were obtained stated by

Table 5. Biofertilizers effect on fresh weight (g) at after one, three and six months of storage (Three replicates).

Fresh weight (g)												
V/T	**After one month of storage**				**After three months of storage**				**After six months of storage**			
	T1	**T2**	**T3**	**Mean**	**T1**	**T2**	**T3**	**Mean**	**T1**	**T2**	**T3**	**Mean**
V1	1.21	1.30	0.86	1.12	1.07	1.17	0.79	1.01	1.06	1.14	0.83	1.01
V2	0.74	0.73	0.71	0.73	0.69	0.66	0.67	0.67	0.67	0.65	0.67	0.66
V3	1.15	1.13	0.88	1.05	1.17	1.17	0.82	1.05	1.10	1.07	0.85	1.01
V4	0.93	0.90	0.80	0.88	0.92	0.85	0.76	0.85	0.85	0.83	0.74	0.81
Mean	1.01	1.01	0.81		0.96	0.96	0.76		0.92	0.92	0.77	
	V	T	VXT		V	T	VXT		V	T	VXT	
S.Em(±)	0.027	0.024	0.047		0.015	0.013	0.027		0.021	0.018	0.036	
CD(P=0.05)	0.080	0.069	0.138		0.045	0.039	0.078		0.060	0.052	0.104	
CD(P=0.01)	0.316	0.273	0.547		0.178	0.154	0.309		0.238	0.206	0.411	

V_1-Sonalika, V_2- PBW 443, V_3, HD- 2821 and V_4- K 9107 and the treatments were- T_1-*Azospirillum*, T_2- PSB and T_3- Control.

Table 6. Biofertilizers effect on dry weight (g) at after one, three and six months of storage (Three replicates).

Dry weight (g)												
V/T	**After one month of storage**				**After three months of storage**				**After six months of storage**			
	T1	**T2**	**T3**	**Mean**	**T1**	**T2**	**T3**	**Mean**	**T1**	**T2**	**T3**	**Mean**
V1	0.18	0.19	0.13	0.17	0.17	0.17	0.10	0.15	0.14	0.15	0.07	0.12
V2	0.12	0.13	0.41	0.22	0.11	0.10	0.10	0.10	0.06	0.06	0.04	0.05
V3	0.17	0.13	0.13	0.14	0.17	0.12	0.12	0.14	0.13	0.09	0.07	0.09
V4	0.13	0.11	0.13	0.12	0.12	0.10	0.13	0.12	0.06	0.05	0.07	0.06
Mean	0.15	0.14	0.20		0.14	0.12	0.11		0.09	0.08	0.06	
	V	T	VXT		V	T	VXT		V	T	VXT	
S.Em(±)	0.049	0.042	0.084		0.002	0.002	0.003		0.009	0.008	0.016	
CD(P=0.05)	0.142	0.123	0.247		0.005	0.004	0.009		0.027	0.024	0.047	
CD(P=0.01)	0.563	0.488	0.975		0.020	0.017	0.035		0.108	0.094	0.187	

V_1-Sonalika, V_2- PBW 443, V_3, HD- 2821 and V_4- K 9107 and the treatments were- T_1-*Azospirillum*, T_2- PSB and T_3- Control.

Table 7. Biofertilizers effect on Vigour index at after one, three and six months of storage.

Vigour index												
V/T	**After one month of storage**				**After three months of storage**				**After six months of storage**			
	T1	**T2**	**T3**	**Mean**	**T1**	**T2**	**T3**	**Mean**	**T1**	**T2**	**T3**	**Mean**
V1	2785	2402	1868	2352	2198	1915	1562	1892	1648	1270	1049	1322
V2	2965	2498	2624	2696	2146	1856	2088	2030	1563	1132	1409	1368
V3	2660	2757	2172	2530	2154	2230	1739	2041	1449	1482	1053	1328
V4	2347	1631	1812	1930	1821	1318	1454	1531	1179	774	865	939
Mean	2689	2322	2119		2080	1830	1711		1459	1165	1094	
	V	T	VXT		V	T	VXT		V	T	VXT	
S.Em (±)	64.507	55.865	111.730		46.73	40.47	80.94		36.57	31.67	63.35	
CD (P=0.05)	188.28	163.06	326.12		136.4	118.1	236.2		106.8	92.45	184.9	
CD (P=0.01)	744.75	644.97	1289.94		539.5	467.2	934.4		422.3	365.7	731.4	

V_1-Sonalika, V_2- PBW 443, V_3, HD- 2821 and V_4- K 9107 and the treatments were- T_1-*Azospirillum*, T_2- PSB and T_3- Control.

Verma *et al*. (2011). A storage experiment was conducted to understand the effect of organic and integrated management practices of seed production and storage containers along with organic (insecticide and fungicide) and organic (botanicals) as seed treatments on seed viability of scented rice cv. Mugad sugandha for 20 months reported by Raikar *et al*. (2011). Chattha *et al*. (2012) observed the effect of different packing materials (metal bin, earthen bin, plastic bag, cloth bag and gunny bag) and grain moisture content at packing (10% and 16%) on viability of wheat seed for ten months of storage.

The seedling quality parameter, vigour index, where two important seed quality parameters like germination percentage and seedling length was taken into account to find out the response of varieties under study towards maintaining or enhancing vigour status after six months of storage. It is noted that, vigour index was greatly enhanced in case of V_1T_2, V_1T_2, V_3T_1, V_3T_2 after one, three and six months of storage. After six months of storage, in comparison to control, all the treatments recorded maximum value except V_4T_2 and V_2T_2. Other V_1T_1, V_1T_2, V_2T_1, V_3T_1, V_3T_2 and V_4T_1 all recorded better value than control. Similar results were reported by Albrecht *et al*. (1981), Mishra *et al*. (1998) and Rout *et al*. (2001) in case of maize. The highest germination, normal seedlings and vigour index which were followed by polythene bag, where Gunny bag showed the lowest germination, normal seedlings and vigour index upto 60 days after storage by Khalequzzaman *et al*. (2012).

Conclusion

From findings it was revealed that, *Azospirillum* and PSB seed inoculation can be recommended for wheat for better seedling growth storability of seed. It should also be noted that, these bio-fertilizers are less expensive than inorganic manures and they are also eco-friendly. More number of wheat genotypes may be used in future for recommendation of actual bio-

fertilizers dose for yield enhancement.

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