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Effects of (*Azotobacter* and *Azosprillium*) Inoculants and Chemical Fertilizers on Growth and Productivity of Canola (*Brassica napus* L.)

Esmaeil Yasari and A.M. Patwardhan
Department of Environmental Science, University of Pune, Pune 411 007, India

Abstract: To investigate the effects of inoculants (biofertilizer) and chemical fertilizer on the yield, yield components and seed oil content of canola (*Brassica napus* L.), a split-plot experimental design with 20 treatments in 4 replications was carried out during 2004-2005 growing season at the Baiecola Agricultural Research Station in the Mazandaran province, Iran. Canola (cv. Hyola 401 hybrid), a high yielding early maturity variety, was grown in rotation after wheat. In the main plots, the biofertilizer treatments were at two different levels: 1) control (no seed inoculation) and 2) seed-inoculation with a combination of three different strains of bacteria *Azotobacter chrocooccum* and *Azosprillium brasilense* and *Azosprillium lipoferum.* In the two sets of 10 sub-plots chemical fertilizers comprising N, P, K and their combinations, NPKS and NPK Zn were applied. The seed yield touched a high of 3741.5 kg h^{-1} at treatment T_{20} (Bio + NPK Zn), that corresponded to 257.7 pods per plant and maximum CGR (18.3 g m^{-2} day^{-1}). The highest weight of 1000 seeds (4.45 g) was obtained at treatment no T_{19}(Bio + NPK S) which coincided with the maximum TDM (1155 g^{-2}) and maximum LAI (5.06). The maximum branching (4.43 branches per plant) was obtained at treatment T_{20} (Bio + NPK Zn) showing a 46.2% increase over the control. The maximum oil content 47.73% was obtained at T_{16} (Bio + NK). The application of inoculation with *Azotobacter* and *Azosprillium* helped to increase the yield by 21.17% over the control, raised the number of pods per plant (16.05%), number of branches (11.78%), weight of 1000 grain (2.92%) and the oil content of seeds (1.73%) but decreased (-0.24%) the number of seeds per pod.

Key words: *Azosprillium*, *Azotobecter*, biofertilizer, chemical fertilizers, canola

INTRODUCTION

Application of fertilizers has become a necessity in the crop production of oil seeds, especially canola, because of the ever increasing demand of the health conscious population of the world. In Iran also efforts are being made to increase the areas under cover for oil seed crops as much of the edible oil is imported. It is well known that N-P-K fertilizers help in the healthy growth of crops like canola, soybean and also in increasing the yield of food grains like wheat and rice. But recently there have been some reports that excessive and repeated use of chemical fertilizers may spoil the soil, ground water and pollute even the atmosphere (Kennedy and Tchan, 1992; Mytton, 1993). These problems have renewed public interest in exploring alternative or supplementary non-polluting sources of N- fertilizers (Ladha *et al*., 1998), i.e., the biofertilizers. Besides the cost of importing chemical fertilizers is equally prohibitive and necessitates the finding of suitable alternative. Positive reports exist on the use of biofertilizers *Azotobacter* or *Azosprillium* for sorghum (*Sorghum bicolor*), (Singh *et al*., 2005), onion (*Allium cepa*), (Navala *et al*., 2004), wheat and mustard (Gupta and Gupta, 2006; Sharma *et al*., 1997). Rai and Caur (1988) reported that combined application *of Azotobacter chroocuccum* and *Azosprillium lipoferum* resulted in higher increase in seed and stover yields of wheat compared to the application of each bacterium alone. Not much experimental work has been conducted on the use of such N_2 fixing bacteria on the growth and yield of canola. The only attempts made on canola refer to the application of inoculation with *Penicillium bilaji*, *Bacillus thuringiensis* and phosphate solubilizing rhizobacteria for the P-uptake, vegetative growth and grain yield of canola (Gleddie *et al*., 1993; Fretas *et al*., 1997). Therefore the present study on effects of (*Azotobacter* and *Azosprillium*) inoculants and chemical fertilizers together on growth and productivity of canola (*Brassica napus* L.) was planned.

MATERIALS AND METHODS

A split-plot experimental design with 20 treatments and 4 replications was carried out in the period October-May, 2004-2005. The bacterial strains

Corresponding Author: Esmaeil Yasari, Department of Environmental Science, University of Pune, Pune 411 007, India

Azosprillium and *Azotobacter* inoculants were applied in the main plots and a combination of chemical fertilizers, both macro and micronutrients were applied in the sub plots. The experiment was carried out at the Baiecola Agricultural Research Station in Mazandaran province (Iran).

The *Azotobacter* and *Azosprillium* strains were isolated from the different samples of the soils of local area. To facilitate the identification of *Azotobacter sp*., modified mannitol agar medium with 10 g of glucose, mannitol per liter as a carbon source (Thompson and Skerman, 1979) was used. For *Azospirillum* NFb a potato extract media (Baldani and Dobereiner, 1980) was used. The isolates were compared with the reference strains. Combined inoculants of *Azotobacter chroococcum, Azosprillium lipoferum* and *Azosprillium brasilense* strains were applied for the biofertilizer treatments.

The Canola (cv. Hyola 401 hybrid) crop, a high yielding early maturity variety, was taken as a second crop in rotation after wheat. It was grown under rain fed conditions. Soil samples were collected and analyzed to know the composition or the nutrients availability and the crop requirements for the nutrients. The chemical fertilizers were chosen and applied accordingly. The experimental soil was texturally silt-clay, with pH 7.6, 1.3% O.C, 180 ppm of available K, 7 ppm of available P, 18 ppm Mn, 10 ppm Fe, 1.1 ppm B and 0.96 ppm of Zn. The chemical fertilizers consisting N, P, K, S and Zn were applied prior to cultivation, except the nitrogen fertilizer which was applied in split stages, once basal and twice top-dressed.

Treatments consisted of: T_1 = control, T_2 = N, T_3 = P, T_4 = K, T_5 = N.P, T_6 = NK, T_7 = P.K, T_8 = NPK, T_9 = NPKS, T_{10} = N PK, Zn, T_{11} = biofertilizer, T_{12} = biofertilizer + N, T_{13} = biofertilizer + P, T_{14} = biofertilizer + K, T_{15} = biofertilizer + NP, T_{16} = biofertilizer + NK, T_{17} = biofertilizer + PK, T_{18} = biofertilizer +NPK, T_{19} = biofertilizer + NPKS and T_{20} = biofertilizer +NPK Zn.

Five plants were sampled randomly in each plot and averaged for recording the change in dry weight in shoots (above ground), at intervals of 34, 67, 82, 111, 133, 153 and 182 DAS (Days after sowing), relating to different stages of canola growth. The samples were first sun dried and thereafter in oven at 70°C till a constant weight was recorded. The leaf area (only one side) was determined by estimating the leaf weight and comparing it with the weight and surface of a standard paper. The dry matter accumulation rate per unit of land area (CGR), expressed as g $m^{-2}day^{-1}$ was calculated using the formula $CGR = (W_2-W_1)/\{SA\ (t_2-t_1)\}$. W_1 and W_2 are crop dry weights at the beginning and end of the interval t_1 and t_2 and SA is the soil area occupied by the plants at each sampling (Acuqaah, 2002; Gupta and Gupta, 2005). Data were analyzed following the analysis of variance technique (ANOVA) and then the mean differences were adjudged by Duncan's multiple range tests (DMRT, Gomez and Gomez, 1984).

RESULTS

Dry-matter accumulation: The dry-matter accumulation was quite significantly improved by the treatments (Table 1). The treatments promoted a much healthier growth before the commencement of winter. A significantly higher TDM at rosette (68 to 82 DAS) was obtained at combined treatments of NPK, NPK S or NPK Zn along with biofertilizer (T_{18}, T_{19} and T_{20}) when compared to the corresponding non-biofertilizer treatments. A threshold value of about 100 g m^{-2} was obtained in these treatments. This increase in TDM continued till (134-153 DAS), with a threshold value of 1080 g m^{-2}. The exuberant growth rate was observed during the 134-153 DAS, irrespective of the treatments. At maturity (154-182 DAS), the TDM gets lowered in all the treatments. Wysocki *et al*. (2005) have also reported such a decline in TDM after reaching a climax in full bloom.

Leaf Area Index (LAI): The rate of increase in the leaf area determines the photosynthetic capacity of plant. The application of different treatments did influence the LAI (Table 2) significantly. The canola plant growth is critical at the rosette stage and if it is able to produce enough leaves before rosette, the plant may spend a healthy winter. The LAI at rosette stage was higher when NP, NPK NPKS and NPK Zn with or without biofertilizer were applied, a threshold value of 3.4 observed in these treatments. The results showed 3.5% increase in LAI at rosette stage in the presence of biofertilizer. LAI increased further till 111 DAS then declines somewhat at the flowering stage. Correlating LAI with the yield, it observed that the canola yield suffered at treatments whenever the LAI is <4 at the flowering stage. Mendham *et al*. (1990) have also reported that a LAI <4 may results in decrease in the growth and yield of canola.

Crop Growth Rate (CGR): The different treatments resulted also in increasing the CGR (Table 3). During rosette the CGR was low at each treatment except in treatments with NPK, Bio + NPK, Bio + NPKS and Bio + NPKZn. In all the treatments the plants resumed growth rapidly after rosette. During flowering period (112 to 134 DAS) the CGR was the highest, especially in these treatments showing 10-12% increase over the non-biofertilizer treatments.

Table 1: Effects of biofertilizers and chemical fertilizers on dry matter accumulation

Treatments TDM g m^{-2}	0-34 DAS	35-67 DAS	68-82 DAS	83-111 DAS Rosette	112-133 DAS	134-153 DAS flowering	154-182 DAS maturity
T_1 = control	1.72	15.84	42.72	182.00	357.00	512.00	428.05
T_2 = N	2.68	26.316	66.00	257.40	527.80	716.80	487.20
T_3 = P	2.60	26.52	59.40	226.20	485.80	658.00	412.65
T_4 = K	2.42	27.42	48.96	241.80	427.00	603.00	430.50
T_5 = NP	3.14	32.40	84.24	330.20	621.60	820.40	544.60
T_6 = NK	2.88	28.80	72.36	283.40	469.00	601.00	488.60
T_7 = PK	2.46	23.04	53.16	224.90	476.00	594.00	483.35
T_8 = NPK	2.98	32.64	89.88	357.50	695.80	862.40	605.85
T_9 = NPKS	3.23	35.22	94.80	362.70	709.80	914.20	635.95
T_{10} = NPKZn	3.85	32.952	96.60	353.60	712.60	921.20	649.80
T_{11} = Bio	2.15	21.60	60.00	279.50	543.20	683.20	499.00
T_{12} = Bio+N	2.78	30.048	74.64	299.00	611.80	777.00	510.00
T_{13} = Bio+P	2.76	27.912	74.88	274.30	547.40	677.60	423.00
T_{14} = Bio+K	2.40	26.40	73.20	282.10	597.80	744.80	516.00
T_{15} = Bio+NP	3.99	32.424	92.40	370.50	658.00	856.00	576.00
T_{16} = Bio+NK	3.01	30.144	61.20	296.40	597.80	705.00	488.00
T_{17} = Bio+PK	2.96	28.80	82.80	370.50	679.00	791.00	532.00
T_{18} = Bio+NPK	4.20	37.608	98.40	481.00	873.60	1080.00	612.00
T_{19} = Bio+NPKS	4.28	36.408	100.80	484.90	879.20	1155.00	675.00
T_{20} = Bio+NPKZn	4.01	38.40	99.60	488.80	891.80	1138.20	691.00
LSD (0.01)	0.2220	5.089	7.289	34.37	61.87	51.74	30.48

Table 2: Effect of biofertilizers and chemical fertilizer on LAI at different stages of canola growth

Treatments LAI	0-34 DAS	35-67 DAS	68-82 DAS rosette	83-111 DAS	112-133 DAS flowering	134-153 DAS	154-182 DAS maturity
T_1 = control	0.102	0.38	1.04	2.80	2.65	2.38	1.78
T_2 = N	0.138	0.75	2.60	3.90	3.49	3.14	2.39
T_3 = P	0.156	0.713	2.36	3.88	3.44	2.76	2.03
T_4 = K	0.158	0.5233	2.24	3.36	3.58	3.22	2.31
T_5 = NP	0.144	0.76	3.52	3.95	3.90	3.51	3.12
T_6 = NK	0.144	0.7133	2.84	3.80	3.70	3.33	2.86
T_7 = PK	0.144	0.553	2.00	3.50	3.80	3.42	3.13
T_8 = NPK	0.136	0.78	3.44	4.56	4.02	3.61	3.17
T_9 = NPKS	0.18	0.933	3.40	4.61	3.98	3.58	3.40
T_{10} = NPKZn	0.198	0.876	3.58	4.80	4.26	3.83	3.66
T_{11} = Bio	0.12	0.576	1.72	3.20	3.04	2.88	2.45
T_{12} = Bio+N	0.172	0.833	2.62	4.24	4.02	3.81	2.68
T_{13} = Bio+P	0.168	0.7133	2.44	4.04	3.83	3.63	2.99
T_{14} = Bio+K	0.166	0.75	2.28	4.06	3.85	3.65	2.81
T_{15} = Bio+NP	0.172	0.976	3.24	4.80	4.50	4.32	3.35
T_{16} = Bio+NK	0.168	0.72	2.92	4.60	4.37	4.14	3.11
T_{17} = Bio+PK	0.168	0.726	2.68	3.96	3.76	3.56	2.91
T_{18} = Bio+NPK	0.208	0.976	3.52	4.98	4.73	4.48	3.49
T_{19} = Bio+NPKS	0.228	0.94	3.64	5.06	4.80	4.55	3.52
T_{20} = Bio+NPKZn	0.202	0.96	3.60	5.00	4.75	4.50	3.57
LSD (0.01)	0.0222	0.0222	0.1986	0.4441	0.2220	0.4441	0.5439

Unlike the TDM and LAI, the CGR was highest during the flowering period (112 to 134 DAS). It is more likely that the new and actively photosynthesizing tissues of pods might be responsible for the increase in CGR during this phase and the increase in dry weight during 135 to 153 DAS. Similar observation was made by Clarke and Simpson (1978). The CGR then declined eventually between the period 134-153 DAS and even to a negative value at final maturity (after 153 DAS). The steep decline in CGR in the last four weeks is on account of senescence of older leaves. Shukla *et al.* (2002) reported that using sulphur and zinc as supplementary nutrients resulted in 23 and 20.5% increase in CGR value of Indian mustard.

Yield and Yield-components: The yield, yield-components and the seed oil content of canola were all influenced significantly by the treatments (Table 4). The effect on yield was statistically significant at $p<0.01$, the best results again obtained at T_{20} (Bio + NPK Zn), as the yield (3741.5 kg h^{-1}) i.e., five times increase over the control. The yield shot above 3280 kg h^{-1} (almost two times more than average of canola yield in the country) by application of additional S and Zn in the presence of biofertilizer.

The effect on the number of pods per plant was also statistically significant $p<0.01$ (Table 4), showing a definite improvement. The best result was obtained at T_{20} with 257.5 pods per plant, applying biofertilizer along with

Table 3: Effect of biofertilizers and chemical fertilizer on CGR at different stages of plant growth

Treatments	0-34 DAS		68-82 DAS		112-133 DAS		154-182 DAS
CGR	g m^{-2} day^{-1}	35-67 DAS	Rosette	83-111 DAS	Flowering	134-153 DAS	Maturity
T_1 = control	0.050	0.427	1.792	4.802	7.954	7.75	-2.798
T_2 = N	0.078	0.716	2.645	6.60	12.290	9.45	-7.653
T_3 = P	0.076	0.724	2.192	5.751	11.80	8.61	-8.178
T_4 = K	0.071	0.757	1.436	6.649	8.418	8.80	-5.75
T_5 = NP	0.092	0.886	3.456	8.481	13.245	9.94	-9.193
T_6 = NK	0.084	0.785	2.904	7.277	8.436	6.60	-3.746
T_7 = PK	0.072	0.623	2.008	5.922	11.413	5.90	-3.688
T_8 = NPK	0.087	0.898	3.816	9.228	15.377	8.33	-8.551
T_9 = NPKS	0.095	0.969	3.972	9.237	15.777	10.22	-9.275
T_{10} = NPKZn	0.113	0.881	4.243	8.862	16.318	10.43	-9.046
T_{11} = Bio	0.063	0.589	2.56	7.568	11.986	7.00	-6.14
T_{12} = Bio+N	0.081	0.826	2.972	7.736	14.218	8.26	-8.90
T_{13} = Bio+P	0.081	0.762	3.131	6.876	12.413	6.51	-8.486
T_{14} = Bio+K	0.070	0.727	3.12	7.203	14.35	7.35	-7.626
T_{15} = Bio+NP	0.117	0.861	3.998	9.589	13.068	9.90	-9.333
T_{16} = Bio+NK	0.088	0.822	2.070	8.110	13.70	5.36	-7.233
T_{17} = Bio+PK	0.087	0.783	3.60	9.920	14.022	5.60	-8.63
T_{18} = Bio+NPK	0.123	1.012	4.052	13.193	17.845	10.32	-15.60
T_{19} = Bio+NPKS	0.125	0.973	4.292	13.244	17.922	13.79	-16.00
T_{20} = Bio+NPKZn	0.117	1.042	4.080	13.420	18.318	12.32	-14.906
LSD (0.01)	0.02220	0.1570	0.4965	1.210	3.070	4.121	2.001

Table 4: Combined effect of Biofertilizers and chemical fertilizers on canola seed yield and yield

Treatments	Seed yield (kg h^{-1})	Pods/plant	Seeds/pod (of main stem)	Number of branches	1000 grain weight	Seed oil content
T_1 = control	736.3h	106.4g	23.27b-d	3.03g	3.73b	41.84d
T_2 = N	1827.4d-h	131.4d-g	24.80a-d	3.48d-g	4.00ab	43.69a-d
T_3 =P	1718.9e-h	120.3fg	24.17a-d	3.08fg	4.06ab	44.00a-d
T_4 =K	1266.1gh	124.4e-g	23.73b-d	3.50d-g	4.18ab	44.35ab
T_5 =NP	2621.5a-f	156.2b-g	24.85a-c	3.98a-d	4.20a	45.42a-d
T_6 =NK	1936.8c-g	157.6b-g	24.95a-c	3.15e-g	4.18ab	43.99a-d
T_7 =PK	1520.7f-h	134.6d-g	24.73a-d	3.68b-g	4.05ab	45.23a-c
T_8 =NPK	2997.7a-d	168.7b-f	24.63a-d	3.65c-g	4.33a	45.68ab
T_9 =NPKS	3095.3a-c	189.8bc	24.35a-d	3.55c-g	4.38a	44.38a-d
T_{10} =NPKZn	3141.2a-c	203.4b	25.88a	3.73b-f	4.35a	45.83ab
T_{11} =Bio	1668.6f-h	125.1e-g	23.08cd	3.58c-g	4.10ab	43.15b-d
T_{12} = Bio+N	2409.3b-g	140.7c-g	25.17ab	3.65c-g	4.13ab	42.24cd
T_{13} = Bio+P	2303.0b-g	151.8b-g	22.88d	3.75b-e	4.18ab	46.28ab
T_{14} = Bio+K	1662.9f-h	140.3c-g	24.27a-d	3.40d-g	4.15ab	45.15a-c
T_{15} = Bio+NP	2910.6a-e	184.5b-d	24.67a-d	3.95a-d	4.28a	45.52ab
T_{16} = Bio+NK	2318.7b-g	178.2b-e	24.65a-d	4.18a-c	4.33a	46.73a
T_{17} = Bio+PK	1942.5c-g	167.9b-f	24.80a-d	3.83a-d	4.31a	45.09a-c
T_{18} = Bio+NPK	3041.5a-c	194.5b	24.90a-c	3.83a-d	4.35a	45.52ab
T_{19} = Bio+NPKS	3282.1ab	191.8bc	25.27ab	4.33ab	4.45a	46.09ab
T_{20} = Bio+NPKZn	3741.5a	257.5a	25.00a-c	4.43a	4.38a	46.34a
LSD value	1044	46.26	1.671	0.5617	0.3959	2.646

NPK Zn, showing a 142.01% increase over the control. Results also showed that the Bio + NPK Zn (T_{20}) treatment gave a 51% increase, compared to the non-biofertilizer treatment NPK Zn (T_{10}).

The increase in the number of seeds per pod (of main stem) was also significant mainly at T_{10}, T_{19} and T_{20}. Otherwise did not change much from the control (LSD = 1.671). With NPK Zn the best result was obtained with 25.88 seeds per pod showing a distinct increase of 11.21% over the control. However seeds inoculated with the biofertilizer did result in further increasing the number of seeds/pod compared. The results indicated that the increase in the yield was consequent mostly to the proliferation of pods per plant. A similar observation was made by Hocking *et al.* (2003) in experiments using N- fertilizer in canola growing.

The number of branches appears to be statistically significant at $p<0.05$ levels (Table 4), the best result was obtained at T_{20} = Bio + NPK Zn i.e., 46.20% increase over the control.

The effects on 1000 grain weight also was positive but did not vary with different treatments; the best result was at T_{19} = Bio + NPKS showed a 19.30% increase over the control. But the change was not statistically different from the other treatments. Despite the differences in the number of pods and the yield with every treatment,

the 1000 grain weight remained between 4 and 4.45 g. This appears to be the range or limit up to which only the improvement could be achieved using these treatments. The narrow range makes it obvious that the 1000 grain weight did not determine the improvement in the yield ($T_{18, 19, 20}$).

The effects on the seed oil content of seeds was statistically significant at $p<0.01$ level (Table 4), the maximum oil content (46.73%) obtained at T_{16} (Bio + NK) showing a 11.68% increase over the control. The treatments T_{19} = Bio + NPKS and T_{20} = Bio + NPK Zn also resulted in improving the oil content to more than 46%.

DISCUSSION

In managing the crop production, it is important to understand how the yield and the yield components respond to the treatments of only chemical fertilizers and a combination of chemical and biofertilizer. Seed yield of canola was found to be related to certain plant characters which can be called as the yield-attributing characters. In the present experiment, it was found closely related to the total dry matter, LAI and CGR. The higher yield corresponded to an increased number of pods per plant and higher CGR and LAI. The additional application of strains of bacteria *Azotobacter chrocooccum* and *Azosprillium brasilense* and *Azosprillium lipoferum* shot up the yield by 21.17% over the control (chemical fertilizers). This appeared mainly related to the proliferation of pod/plant (16.05%) though simultaneously the number of branches (11.78%) also increased. The weight of 1000 grain (2.92%) and the oil content of seeds (1.73%) also increased with this additional biofertilizer support. The present results are better than those obtained by Sharma *et al.* (1997) and Shukla *et al.* (2002) on Indian mustard, using only *Azotobacter* as biofertilizer.

Azotobacter and *Azosprillium* are free living N_2 fixing bacteria which in the rhizospheric zone have the ability to synthesize and secret some biologically active substances like B vitamins, nicotinic acid, pantothenic acid, biotin, heteroauxins, gibberellins etc. which enhances the root growth (Kader, 2002). The *Azotobacter* and *Azosprillium* association helps the crop improvement also by excretion of ammonia in the presence of root exudates that enhances and regulates the nutrient uptake by plants (Narula *et al.*, 1993; Narula and Yadav, 1989). The higher dry-matter production by the inoculated plant might be because of the augmented uptake of nitrogen, phosphorous and potassium, which in turn was consequent to the root proliferation (Table 5).

Table 5: Gain of biofertilizers on yield, yield contributing characters and oil content of seeds comparing to control

Treatment	Control	Inoculation (Biofertilizer)	Increase over the control (%)
Yield (kg h^{-1})	2086.22	2527.99	21.17
Pods/plant	149.26	173.22	16.05
Seeds/pod (of main stem)	24.53	24.47	-0.24
Number of branches	3.48	3.89	11.78
1000 grain weight (g)	4.143	4.264	2.92
Seed oil Content (%)	44.44	45.21	1.73

REFERENCES

Acquaah, G., 2002. Principals of Crop Production (Theory, Technical and Technology). Prentice-Hall of India, New Delhi, pp: 460.

Baldani, V.L.D. and J. Dobereiner, 1980. Host-plant specificity in the infection of cereals with *Azospirillum* sp. Soil Biol. Biochem., 12: 433-439.

Clarke, J.M. and G.M. Simpson, 1978. Growth analysis of *Brassica napus* cv. Tower. Can. J. Plant Sci., 58: 587-595.

Fretas, J.D.E., M.R. Banerjee and J.J. Germida, 1997. Phosphate solubilizing *Rhizobacteria* enhance the growth and yield but not phosphorus uptake of canola (*Brassica napus*). Biol. Fertility of Soil, 24: 358-364.

Gleddie, S.C., D. Schlechte and G. Turnbull, 1993. Effect of inoculation with *Penicillium bilaji* (provide) on phosphate and yield of canola in western Canada. In Proceedings. Alberta Soil Science Workshop, Edmonton, Alberta. February 19-20, pp: 155-160.

Gomez, K.A. and A.A. Gomez, 1984. Statistical Procedures for Agricultural Research 2nd Edn., John Wiley Sons, New York, Chichester, Brisbane, Toronto, Singapore.

Gupta, N.K. and S. Gupta, 2005. Plant Physiology. Oxford and IBH publishing Co. Pvt. Ltd. New Delhi.

Gupta, P. and V. Gupta, 2006. Studies on efficacy of biofertilizers on yield of wheat (*Triticum aestivum*) and mustard (*Brassica juncea*). J. Microb. World, 8: 51-56.

Hocking, P.J., J.A. Mead, A.J. Good and S.M. Diffey, 2003.The response of canola (*Brassica napus* L.) to tillage and fertilizer placement in contrasting environments in southern New South Wales. Aust. J. Exp. Agric., 43: 1323-1333.

Kader, M.A., 2002. Effects of *Azotobacter* inoculant on the yield and nitrogen uptake by wheat. On Line J. Biol. Sci., 2: 259-261.

Kennedy, I.R. and Y.T. Tchan, 1992. Biological nitrogen fixation in non leguminous field crops: Recent advances. Plant Soil., 141: 93-118.

Ladha, J.K., A.T. Padre, G.C. Punzalan, E. Castillo, U. Singh and C.K. Reddy, 1998. Nondestructive estimation of shoot nitrogen in different rice genotypes. Agron. J., 90: 33-40.

Mendham, N.L., J. Russell and N.K. Jarosz, 1990. Response to sowing time of three contrasting Australian cultivars of oilseed rape (*Brassica napus* L.). J. Agric. Sci. Cambridge, 114: 275-283.

Mytton, L., 1993. Nitrogen Fixation. In Institute of Grassland and Environmental Research Rep. Inst. Grassland and Environ. Res., Aberystwyth, UK., pp: 46-50.

Narula, N. and K.S. Yadav, 1989. Nitrogen Fixation Research in India with *Azotobecter*. In Biological Nitrogen Fixation Research Status in India. Dadarwal, K.R. and K.S. Yadav (Eds.). Indian Society for Plant Physiology and Biochemistry, New Delhi, pp: 88-124.

Narula, N., P.P. Gupta, P.R. Kumar and K. Laxminarayan, 1993. Field response of Indian mustard (*Brassica juncea*) to inoculation of soil isolate and analogue resistant mutant of *Azotobacter chrooccocum*. Ann. Bio. Ind., 9: 144-148.

Navala, A.M., P.P Wani and A.S. Patil, 2004. Effects of VAM and *Azosprillium* inoculation to on onion (*Allium cepa*) cv. B-780 with respect to N, P and micronutrient uptake. The Orissa J. Hortic., 32: 83-88.

Rai, S.N. and A.C. Caur, 1988. Characterization of *Azotobacter* sp. and effect of *Azosprillium lipoferum* on the yield and N-Uptake of wheat crop. Plant and Soil., 109: 131-134.

Sharma, S.K., R. Mohan Rao and D.P. Singh, 1997. Effects of crop geometry and nitrogen on quality and oil yield of *Brassica* species. Ind. J. Agron., 42: 357-360.

Shukla, R.K., A. Kumar, B.S. Mahapatra and K. Basanth, 2002. Integrated nutrient management practices in relation to morphological and physiological determination of seed yield in Indian mustard (*Brassica juncea*). Ind. J. Agric. Sci., 72: 670-672.

Singh, M.M., M.L. Mautya, S.P. Singh and C.H. Mishra, 2005. Effects of nitrogen and biofetilizers inoculation on productivity of forage sorghum (*Sorghum bicolor*). Ind. J. Agric. Sci., 73: 167-168.

Thompson, J.P. and V.B.D. Skerman, 1979. *Azotobacteraceae*: The Taxonomy and Ecology of the Aerobic Nitrogen-fixing Bacteria. Academic Press, Inc., London.

Wysocki, D., S. Nick and S. Ott, 2005. Growth and nutrient uptake on winter canola at Pedelton, Oregon. Dry land Agricultural Research Annual Report.