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Enhancement of Growth and Nutrient Uptake of Rapeseed (*Brassica napus* L.) by Applying Mineral Nutrients and Biofertilizers

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Abstract: For investigating the effect of chemical fertilizer as well as biofertilizers on seed yield and quality i.e. oil, protein and nutrients concentration of rapeseed (*Brassica napus* L.), a split-plot fertilizers application experimental design in 4 replications was carried out during the 2005-2006 growing season, at the Gharakheil Agricultural Research Station in the Mazandaran province of Iran. Rapeseed was grown as a second crop in rotation after rice. Biofertilizers treatments were two different levels: control (no seed inoculation) and seeds inoculation with a combination of *Azotobacter chroococcum* and *Azosprillum brasilense* and *Azosprillum lipoferum*, as main plot and chemical fertilizers comprised N, P, K and their combinations, NPKS and NPK Zn as sub plots. The maximum value of seed yield obtained at (BF+NPK Zn) 3421.2 kg h^{-1} corresponding to 244.5 pods per plant and maximum concentration of Zn in leaves as well as seeds. The highest weight of 1000 seeds (4.45 g) happened to obtain at (BF+NPK S) which coinciding with the maximum K levels in leaves. The highest number of branches was obtained at (BF+NPK Zn) with 4.43 branches per plant i.e., 46.2% increase over the control. The maximum value of rapeseed oil content 47.73% obtained at T_{16} (BF+NK) but maximum protein concentration of seed obtained at T_{12} (BF+N). Overall the results indicated that inoculation resulted in increase in seeds yield (21.17%), number of pods per plant (16.05%), number of branches (11.78%), weight of 1000 grain (2.92%), oil content of seeds (1.73%) and protein (3.91%) but decrease (-0.24%) in number of seeds per pods comparing to non-Biofertilizers treatments. Irrespective to the treatments, results showed that application of Biofertilizers coincided with 3.86, 0.82, 2.25, 0.75 and 0.91% increase in concentrations of N, P, K, S and Zn in the seeds over the non-Biofertilizers treatments.

Key words: *Azotobacter*, *Azosprillum*, nutrient concentration, rapeseed

INTRODUCTION

Amongst the factors that influence rapeseed growth and yield, the choice of fertilizers and provision of conditions for optimum nutrient availability is of paramount importance since rapeseed plant is very efficient at taking up nutrients. This efficiency is reflected in the high nutrient contents in rapeseed leaf and seed relative to other crops. While on one hand it means that the vegetative matter and the seed are healthy for animal feed, taking a rapeseed crop as a rotation crop depletes the soil nutrients. Problems arising out of such depletion are overcome to a great extent by returning dry matter to the soil. Rapeseed is also viewed as a cleaning crop as it provides a break in the cycle of the pests and diseases hosted by other crops and for managing grass weeds. Using a judicious combination of chemical fertilizers as well as biofertilizers to obtain the best results can be strategic step.

Positive reports of application of biofertilizers (*Azotobacter* and *Azosprillum* and other bacteria) on yield are available on crops like: Indian mustard (Suneja and Lakshminaraya, 2001), cotton (Yue *et al*., 2007), corn (Albrecht *et al*., 1981), sorghum (Singh *et al*., 2005), wheat (Cecilia *et al*., 2004), mustard (Sharma *et al*., 1997), wheat and mustard (Gupta Pramila and Gupta Vishal, 2006), tobacco (Li *et al*., 2007) and barley (Ozturk and Sahin, 2003), which are mainly attributed to the enhancement of factors like N_2-fixation nitrate reductase activity, intake of NO_3, NH_4, H_2PO_4, K and Fe, plant water status and production of phytohormones such as indol acetic acid (Antoun *et al*., 1998; Arshad and Frankenberger, 1998; Wani *et al*., 1998).

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Since attention has seldom been given to the nutrient requirement of the soil, rapeseed crop has suffered in most cases, either from lack or excess of these nutrients. Hence, the necessity to evaluate the best nutrient uptake for improving rapeseed yield, quality and fortification in Mazandaran crops, based on soil test results.

MATERIALS AND METHODS

In order to evaluate the rapeseed growth and nutrient uptake a split-plot experimental design with 20 treatments and 4 replications was carried out in 2005-2006, where *Azosprillum* and *Azotobacter* inoculants were as main plot and combination of chemical fertilizers including macro and micronutrients were as sub plots in Gharakheil Agricultural Research Station in Mazandaran province (Iran). *Azotobacter* and *Azosprillum* strains were isolated from the different samples of the soils of local area. To facilitate the recognition of *Azotobacter* sp., modified Mannitol agar medium with 10 g of glucose, Mannitol per liter as a carbon source (Thompson and Skerman, 1979) and for recognition of *Azosprillum* NFb and potato extract media were used. The isolates were compared with reference strains. Combined inoculants of *Azotobacter chroococcum*, *Azosprillum lipoferum* and *Azosprillum brasilense* strains were used as Biofertilizers treatments.

Rapeseed (cv. PF 7045.91 or Sarigol), was grown under rain fed condition. The experimental soil was texturally silt-clay, with pH 7.2, 1.5% OC, 193 ppm of available K, 8.7 ppm of available P, 19 ppm Mn, 11 ppm Fe, 1 ppm B and 1 ppm of Zn. The chemical fertilizers consisting N, P, K, S and Zn were incorporated with soil prior to cultivation, except nitrogen fertilizer which was applied in split into basal and top-dressed in two times. Treatments were consist of: T_1 = control, T_2 = N, T_3 = P, T_4 = K, T_5 = N.P, T_6 = N.K, T_7 = P.K, T_8 = N.P.K, T_9 = N.P.K.S, T_{10} = N.P.K.Zn, T_{11} = Biofertilizers, T_{12} = Biofertilizers+N, T_{13} = Biofertilizers+P, T_{14} = Biofertilizers +K, T_{15} = Biofertilizers+NP, T_{16} = Biofertilizers +NK, T_{17} = Biofertilizers+PK, T_{18} = Biofertilizers+NPK, T_{19} = Biofertilizers +NPKS and T_{20} = Biofertilizers+NPKZn.

RESULTS

Yield and yield components: The yield and yield components were all influenced significantly by the treatments (Table 1). The effect on rapeseed yield was statistically significant at $p<0.01$, the best results obtained at (BF +NPK Zn) touching 3421.2 kg h^{-1} i.e., almost five times increase over the control.

The effect on the number of pods per plant was also statistically significant $p<0.01$ (Table 1), showing a definite improvement. The best result obtained at (BF+NPK Zn) with 244.5 pods per plant, applying Biofertilizers along with NPK Zn, showing a 138% increase over the control. Result also showed that BF+NPK Zn coinciding with 51% increase comparing to non-Biofertilizers treatment of NPK Zn.

The increase in the number of seeds per pod (of main stem) was also significant. The number of seeds per pod did not change much from the control with the different

Table 1: Effect of Biofertilizers and chemical fertilizers on rapeseed yield, yield component and seed oil and protein contents

Treatments	Seed yield (kg h^{-1})	Pods/plant	Seeds/pod (of main stem)	No. of branches	1000 seed weight	Seed oil content (%)	Seed protein content (%)
T_1 (control)	736.3h	106.4g	23.27bcd	3.03g	3.73b	41.84d	21.20d
T_2 (N)	1827.4defgh	131.4defg	24.80abcd	3.48defg	4.00ab	43.69abcd	23.42abcd
T_3 (P)	1718.9efgh	120.3fg	24.17abcd	3.08fg	4.06ab	44.00 abcd	22.95abcd
T_4 (K)	1266.1gh	124.4efg	23.73bcd	3.50efg	4.18b	44.35ab	22.75bcd
T_5 (NP)	2621.5abcdef	156.2bcdefg	24.85abc	3.98abcd	4.20a	45.42abcd	23.82abc
T_6 (NK)	1936.8cdefg	157.6bcdefg	24.95abc	3.15efg	4.18ab	43.99abcd	23.17abcd
T_7 (PK)	1520.7fgh	134.6defg	24.73abcd	3.68cdefg	4.05ab	45.23abc	22.50cd
T_8 (NPK)	2997.7abcd	168.7bcdef	24.63abcd	3.65defg	4.33a	45.68ab	23.50abc
T_9 (NPKS)	3095.3abc	189.8bc	24.35abcd	3.55cdefg	4.38a	44.38abcd	24.25abc
T_{10} (NPKZn)	3141.2abc	203.4b	25.88a	3.73bcdef	4.35a	45.83ab	24.02abc
T_{11} (BF)	1668.6fgh	125.1efg	23.08cd	3.58cdefg	4.10b	43.15bcd	22.55cd
T_{12} (BF+N)	2409.3bcdefg	140.7cdefg	25.17ab	3.65cdefg	4.13b	42.24cd	25.22a
T_{13} (BF+P)	2303.0bcdefg	151.8bcdefg	22.88d	3.75bcde	4.18b	46.28ab	23.22abcd
T_{14} (BF+K)	1662.9fgh	140.3cdefg	24.27abcd	3.40defg	4.15b	45.15abc	24.07abc
T_{15} (BF+NP)	2910.6abcde	184.5bcd	24.67abcd	3.95abcd	4.28a	45.52ab	24.52abc
T_{16} (BF+NK)	2318.7bcdefg	178.2bcdes	24.65abcd	4.18abc	4.33a	46.73a	23.45abcd
T_{17} (BF+PK)	1942.5cdefg	167.9bcdef	24.80abcd	3.83abcd	4.31a	45.09abc	23.12abcd
T_{18} (BF+NPK)	3041.5abc	194.5b	24.90abc	3.83abcd	4.35a	45.52ab	24.95ab
T_{19} (BF+NPKS)	3282.1ab	191.8bc	25.27ab	4.33ab	4.45a	46.09ab	24.65abc
T_{20} (BF+NPKZn)	3421.2a	244.5a	25.00abc	4.43a	4.38a	46.34a	24.90ab
LSD value	1044.0	46.26	1.671	0.5617	0.3959	2.646	1.919

N: Nitrogen fertilizer (Urea at 250 kg h^{-1}), P: Phosphorus fertilizer (Triple super phosphate at 150 kg h^{-1}), K: Potassium fertilizer (Potassium sulfate at 150 kg h^{-1}), S: Sulphur fertilizer at 100 kg h^{-1}), Zn: Zinc sulfate at 40 kg h^{-1}), BF: Biofertilizers (combination of *Azotobacter chroococcum* and *Azosprillum brasilense* and *Azosprillum lipoferum*), LSD: Least significant differences at 0.05 level, Values with different letter(s) indicate significant difference from each other treatments (LSD = 1.671) except at NPK, BF + NPKS and BF+NPK Zn. With the application of Zn along with NPK, the best result with 25.88 seeds per pod showed a distinct increase of 11.21% over the control. However seeds inoculation with the Biofertilizers did resulted in more increase in the number of seeds per pod compared to the chemical fertilizer treatments. The results indicated that an increase in the yield achieved was consequent mostly to the proliferation in the number 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 1), the best result was obtained at BF+NPKZn i.e., 46.20% increase over the control.

The effects on 1000 grain weight thought positive but did not show much difference between different treatments. The best result was at the BF+NPKS i.e., 19.30% increase over the control, but was not statistically different from the other treatments except the control. 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 only up to which the improvement could be achieved by the treatments. Considering this narrow range of variation, it is obvious that the change in the 1000 grain weight was not determining the improvement in the yield at high yielding treatments (BF+NPK, BF+NPKS and BF+NPK Zn).

Seed quality

Seeds oil and protein contents: Oil and protein content of the rapeseed seeds also significantly were influenced by the treatments (Table 1). The effects of oil content of seeds were statistically significant at $p<0.01$ level, the maximum seed oil content was obtained at BF+NK showing a 46.73% i.e., 11.68% increase over the control. The treatments BF+NPKS and BF+NPK Zn also resulted in the oil contents of more than 46%.

The results showed that the maximum protein contents of the seed resulted at BF+N (25.22%) i.e., 18.96% increase over the control, which was followed by BF+NPK, BF+NPKS and BF+NPK Zn. The results indicated the role of nitrogen in increasing the protein content of the seed which is supported by those observed by Ahmad *et al*. (2007).

Nutrient contents of leaves and seeds

Nitrogen: The application of N fertilizer, either as chemical fertilizer or Biofertilizers resulted in a substantial increase in N content of the leaves and seeds. The leaves maximum N concentration at BF+NP (4.24%) was about 45.20% more over the control (Table 2). The maximum N concentration reported in leaves at flowering stage of rapeseed plant is 3-4.5% (Holmes, 1998). The result showed thereby the capacity of the rapeseed plant to extract N from the soil, which was more efficient at the presence of *Azotobacter* and *Azosprillum* showing their ability to make available more N for the rapeseed. There were 14, 29 and 34% more

Table 2: Effect of Biofertilizers and chemical fertilizers on nutrients contents of leaves at flowering stage

Treatments	N in leaves (%)	P in leaves (%)	K in leaves (%)	S in leaves (ppm)	Zn in leaves (ppm)
T_1 (control)	2.92d	0.157d	1.237f	92.83c	21.50c
T_2 (N)	3.09cd	0.200bcd	1.353cdef	97.50c	22.00c
T_3 (P)	2.89d	0.243abc	1.333def	90.33c	19.42c
T_4 (K)	2.99d	0.163d	1.803abcd	97.33c	21.33c
T_5 (NP)	3.15cd	0.210bcd	1.263ef	100.5c	22.67bc
T_6 (NK)	3.45bcd	0.187cd	1.747abcde	114.7abc	22.17c
T_7 (PK)	2.94d	0.230bc	1.713abcdef	99.67c	22.50bc
T_8 (NPK)	3.85abc	0.210bcd	1.567abcedf	104.83abc	23.00bc
T_9 (NPKS)	3.17cd	0.257ab	1.853abc	130.00a	20.33c
T_{10} (NPKZn)	3.63abcd	0.213bcd	1.727abcdef	105.33abc	27.17ab
T_{11} (BF)	3.34bcd	0.190cd	1.417bcdef	100.00c	20.83c
T_{12} (BF+N)	3.99ab	0.204bcd	1.467bcdef	98.33c	23.17bc
T_{13} (BF+P)	3.33bcd	0.243abc	1.430bcdef	96.00c	19.67c
T_{14} (BF+K)	3.06d	0.195cd	1.867ab	95.33	21.67c
T_{15} (BF+NP)	4.24a	0.287a	1.733bcdef	107.67abc	22.33bc
T_{16} (BF+NK)	3.50abcd	0.230bc	1.790abcd	99.00c	22.50bc
T_{17} (BF+PK)	3.18cd	0.205bcd	1.783abcd	90.83c	19.50c
T_{18} (BF+NPK)	3.67abcd	0.237abc	1.920ab	105.17abc	23.00bc
T_{19} (BF+NPKS)	3.50abcd	0.233abc	2.043a	127.17ab	23.50abc
T_{20} (BF+NPKZn)	3.66abcd	0.230bc	1.920ab	102.33bc	27.83a
LSD value	0.6540	0.04965	0.4328	22.84	4.193

N: Nitrogen fertilizer (Urea at 250 kg h^{-1}), P: Phosphorus fertilizer (Triple super phosphate at 150 kg h^{-1}), K: Potassium fertilizer (Potassium sulfate at 150 kg h^{-1}), S: Sulphur fertilizer at 100 kg h^{-1}), Zn: Zinc sulfate at 40 kg h^{-1}), BF: Biofertilizers (combination of *Azotobacter chroococcum* and *Azosprillum brasilense* and *Azosprillum lipoferum*), LSD: Least significant differences at 0.05 level. Values with different letter(s) indicate significant difference from each other

N concentration in leaves in non-Biofertilizers treatments of control, N and NP comparing to BF, BF+N and BF+NP respectively. However in the presence of other nutrient components the effectiveness of the bacteria for nutrient availability was less.

Application of N fertilizer resulted also in an increase in N concentration in seeds at all the treatments (Table 3), the higher levels found at BF+N (4.04%) showing 19.17% increase over control which statistically was similar to the rest of the treatments except control. Nitrogen content of seed in the present study was more than the range of N concentrations (2.7 to 3.84%) recorded by Hocking and Strapper (2001). Kakati and Kalita (1996) reported 3.69% N in the grains of Indian mustard (*Brassica juncea*) by applying 100 kg ha^{-1} N.

Phosphorus: The application of P fertilizer individually and in combination with N and K resulted in increase in P concentration in leaves at treatments: P, NPKS, BF+K, BF+NP and BF+NPK. However maximum P concentration in the leaves (Table 2) obtained at T_{15} when the treatment included Biofertilizers along with NP (0.287%) showing 82.80% increase over the control.

The results showed that P concentration on seeds was also affected by the treatments. The maximum P concentration was at PK and BF+P (0.69%) having 18.96% increase over the control. Ahmad and Abdin (2000) reported that rapeseed seed P concentration was ranged between 0.2 and 0.6%, although Holmes (1998) suggested that seed P concentration was within 0.68-0.81% of the TDM (Total dry Matter). In the present study (Table 3), however, the seed P level ranged between 0.51 to 0.69%. Together with nitrogen fertilizer, phosphorus plays an important role in rapeseed yield and seed oil content. Its concentration in the seed composition is an obvious value addition to the animal feed.

Thakur and Chanel (1998) reported that application of 120 kg N and 40 kg P ha^{-1} significantly increased the seed yield along with the uptake of 56% N and P 12% respectively in gobhi sarson (*Brassica napus*).

There results showed that there was 21% more P concentration in leaves in non-Biofertilizers treatments of control comparing to Biofertilizers. However in the presence of P there was no increase in P concentration in BF +P. It was concluded that in the presence of the specific nutrient in soil solution the activity and ability of the bacteria in making available nutrient decreased.

Potassium: The application of K fertilizer individually or in combination with N, P, S and Zn in the presence or absence of Biofertilizers showed an overall increase in the concentration K in leaves (Table 2), in treatments of chemical fertilizers the most significant being at treatments NPKS (1.853%) showing 49.79% increase over the control but it was not statistically different from the rest of the treatments except with control, N, P and K. application of Biofertilizers resulted in higher increase relative to the corresponding treatments without Biofertilizers. The maximum concentration K in leaves obtained at BF = NPKS (2.043%) showing 65.15% increase over the control. Increase in K concentration of leaf have already been observed (Kopsell *et al*., 2004) in Leafy *Brassica oleracea*

Table 3: Effect of Biofertilizers and chemical fertilizers on nutrients contents of seeds at maturity

Treatments	N contents of seed (%)	P contents of seed (%)	K contents of seed (%)	S contents of seed (ppm)	Zn contents of leaves (ppm)
T_1 (Control)	3.39b	0.58abcd	0.56c	0.320abc	28.90bc
T_2 (N)	3.75ab	0.54cd	0.57bc	0.317abc	31.30bc
T_3 (P)	3.67ab	0.67ab	0.57bc	0.313bc	28.23bc
T_4 (K)	3.64ab	0.57bcd	0.65abc	0.338abc	31.10bc
T_5 (NP)	3.81ab	0.63abcd	0.61abc	0.305c	32.18b
T_6 (NK)	3.69ab	0.57abcd	0.65abc	0.310bc	28.48bc
T_7 (PK)	3.61ab	0.69a	0.66abc	0.325abc	30.70bc
T_8 (NPK)	3.76ab	0.63abcd	0.65abc	0.310bc	28.47bc
T_9 (NPKS)	3.88a	0.63abcd	0.64abc	0.370a	29.98bc
T_{10} (NPKZn)	3.84ab	0.57bcd	0.66abc	0.362ab	55.03a
T_{11} (BF)	3.61ab	0.60abcd	0.61abc	0.328abc	28.75bc
T_{12} (BF+N)	4.04a	0.51d	0.58c	0.345abc	32.13b
T_{13} (BF+P)	3.67ab	0.69a	0.59abc	0.322abc	32.98b
T_{14} (BF+K)	3.85ab	0.58abcd	0.69a	0.303c	29.85bc
T_{15} (BF+NP)	3.92a	0.61abcd	0.61abc	0.317abc	29.45bc
T_{16} (BF+NK)	3.75ab	0.54cd	0.66abc	0.338abc	30.28bc
T_{17} (BF+PK)	3.70ab	0.66bc	0.66ab	0.320abc	28.80bc
T_{18} (BF+NPK)	3.99a	0.66ab	0.66ab	0.320abc	27.23bc
T_{19} (BF+NPKS)	3.94a	0.60abcd	0.65abc	0.363ab	25.60c
T_{20} (BF+NPKZn)	3.98a	0.65abc	0.66abc	0.345abc	56.88a
LSD value)	0.4136	0.1034	0.8443	0.04483	6.568

N: Nitrogen fertilizer (Urea at 250 kg h^{-1}), P: Phosphorus fertilizer (Triple super phosphate at 150 kg h^{-1}), K: Potassium fertilizer (Potassium sulfate at 150 kg h^{-1}), S: Sulphur fertilizer at 100 kg h^{-1}), Zn: Zinc sulfate at 40 kg h^{-1}), BF: Biofertilizers (combination of *Azotobacter chroococcum* and *Azosprillum brasilense* and *Azosprillum lipoferum*), LSD: Least significant differences at 0.05 level. Values with different letter(s) indicate significant difference from each other cultivars, the leaf K levels ranged between 2.1 and 3.5% while in the present study, leaf K levels ranged from 1.2 to 2.04%. Holmes (1998) reported the level of K in the plant between 1-2.2% of TDM in leaves at flowering. Asare and Scarisbrick (1995) also reported that N and S concentration of oilseed rape (*Brassica napus* L.) improved considerately in treated plots as compared to control plots.

K concentration of seeds was also affected by the treatments. The maximum value obtained at BF+K resulting in 0.69% (23.21%) increase over the control. It was observed that application of K fertilizer along with Biofertilizers resulted in 7% more increase as compare with the application of K alone.

Sulfur: Application of S fertilizer in non Biofertilizers plots showed an increase in the concentration of S in leaves only at treatments NPKS and NK, while in the rest of the treatments it was lower and statistically similar. The results indicated that N and K have been supportive for mobilization of S to the leaves as at P the concentration was less than the control. The application of Biofertilizers resulted in higher increase relative to the corresponding treatments of non-Biofertilizers plots. The maximum concentration of S in leaves obtained at BF+NPKS (127.17 ppm) showing 37% increase over the control.

S concentration of seeds was also affected by the treatments. The maximum value obtained at (NPKS) resulting in 0.370 ppm (15.62%) increase over the control which was similar with those of NPK Zn and BF+ NPK S.

Zinc: Application of Zn fertilizer in non-Biofertilizers plots showed an increase in the concentration of Zn in leaves only at treatments NPK Zn 27.17 ppm showing 26.37% increase over the control. In Biofertilizers treatments at T_{20} the maximum concentration of Zn in leaves (29.44 ppm) was resulted which showed that application of Biofertilizers resulted in 3% more increase in Zn concentration of leave as compare to the corresponding treatment with no Biofertilizers treatment.

The Zn concentration in the seed was highest at NPK Zn showing >90% increase over the control. However in treatment at presence of Biofertilizers i.e. at BF + NPK Zn (56.88 ppm) resulted in 96.81% increase over the control. The result obtained in the present study was comparable with the Holmes (1998) suggesting 70 ppm of Zn concentration in seeds at the maturity. Malewar *et al.* (2001) reported an increase of 15% in yield of Indian mustard by application of Zn as $ZnSO_4$ (30 kg ha^{-1}), which also resulted in high uptake of Zn in stover and seed.

DISCUSSION

Seeds inoculation with *Azotobacter* and *Azosprillum* resulted in 21.17% increase in seed yield over the control (chemical fertilizers), which was mainly due to increase in number of pod/plant (16.05%). Increase in number of branches (11.78%) and weight of 1000 grain (2.92%), oil content of seeds (1.73%) and protein content of seeds (3.91%) was also observed with application of biofertilizers (Table 4). However, a decrease in the number of seeds per pod in inoculated seeds was observed. The results of the present experiment was fairly comparable with those of the earlier workers, Shukla *et al.* (2002) report that application of *Azotobacter* resulted significantly in higher number of seeds per siliquae, TDM, branches/plant and the length of siliquae in Indian mustard. Sharma *et al.* (1997) suggested that oil and protein production increased significantly when nitrogen applied either through *Azotobacter* or urea.

Azotobacter and *Azosprillum* 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, pentothenic acid, biotin, heteroauxins, gibberellins etc. which enhances the root growth (Kader, 2002; Khalid *et al*., 2004). The *Azotobacter* and *Azosprillum* 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 (Chandra *et al*., 2007). 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.

The results showed that although application of chemical fertilizers have definite increase especially in the corresponding element but seed inoculation with *Azotobacter* and *Azosprillum* resulted even more in

Table 4: Gain of Biofertilizers on yield, yield contributing characters and oil content of seeds comparing to control

Treatments	Control	Inoculation (biofertilizers)	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
No. 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
Seed protein content (%)	23.16	24.067	3.91
N concentration of seeds (%)	3.703	3.846	3.86
P concentration of seeds (%)	0.607	0.612	0.82
K concentration of seeds (%)	0.622	0.636	2.25
S concentration of seeds (ppm)	32.192	32.435	0.75
Zn concentration of seeds (ppm)	0.327	0.330	0.91

accumulation of the element either in leaves during flowering period or in seeds at maturity. Irrespective to the treatments, results showed that application of Biofertilizers coincided with 3.86, 0.82, 2.25, 0.75 and 0.91% increase in concentrations of N, P, K, S and Zn in the seeds over the non-Biofertilizers treatments.

Cheema *et al*. (2001) suggested that the high yield of rapeseed depended on an optimum concentration of all elements in plant. Enhancing the yield is the prime objective for agronomical research while an enriched animal feed, available after seed oil extraction, also indirectly benefits the human health. The human need of quality food and proteins is fulfilled to a great extent by the animal production industry which has a synergistic relationship with oilseed production and processing. The economic sustainability of both depends on this mutually beneficial relationship and is critical to any evaluation of the role of nutrients in producing enriched oilseed and animal meal.

In rapeseed cultivation equally important are the TDM and protein rich seed, the two useful by-products, which depend upon the mobility of nutrients to the leaves and seeds. These two treatments promoted a healthy plant growth and resulted also in concentrating the nutrients in leaf as well as in seed. This enrichment and fortification of seed will ultimately result in improving the rapeseed yield and quality in successive seasons. Care must be taken however to ensure that the levels of S in seed after oil extraction does not exceed a limit beyond which it may be harmful (Glucosinolate) as direct animal meal. The dry matter by-product also makes the fertilizer application cost-effective in returning the nutrients back to the soil for other crops like soybean, rice and wheat.

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