Indian Journal of Agricultural Sciences **84** (11): 1316–23, November 2014/Article
https://doi.org/10.56093/ijas.v84i11.44624

Influence of boron and organic matter on the changes in soil biological properties and yield of rape (*Brassica campestris*) in an aeric endoaquept

MITALI MANDAL[1] and DILIP KUMAR DAS[2]

Orissa University of Agriculture and Technology, Bhubaneswar, Odisha

Received: 5 May 2012; Revised accepted: 17 July 2014

ABSTRACT

Field experiments were conducted during winter seasons of 2006 -07 and 2007-08 to study the influence of boron and organic matter application on the changes in soil biological properties in an aeric endoaquept (pH,7.5;organic carbon content, 0.61%; $CaCl_2$–extractable boron 0.32 mg/kg) growing rape (*Brassica campestris* L.) as a test crop. The results reveal that non-symbiotic nitrogen fixing bacteria (NFB), non symbiotic nitrogen fixing capacity, phosphate solubilising micro-organisms (PSM), phosphate solubilising capacity (PSC) and microbial biomass carbon (MBC) have been found to be influenced with different treatments, being recorded highest (90.45, 69.29 × 10^5/g soil; 11.73, 9.63; 80.29, 63.28 mg of N_2 fixed/g of soil/g of sucrose; 0.038, 0.032 mg/g dry soil and 159.48, 149.58 µg/g dry soil respectively) in the treatment T_4 where lower level (0.5 kg/ha) of boron as calbor and organic matter at 5 tonnes/ha was applied along with recommended levels of N, P_2O_5 and K_2O (80:40:40) in both rhizosphere (R) and non-rhizosphere (NR) soils. The yield of rape was also recorded highest (8.95 q/ha) in the treatment T_4 (lower level (0.5 kg/ha) of boron as calbor,organic matter at 5 tonnes/ha and recommended levels of N, P_2O_5 and K_2O (80:40:40). From the stepwise technique of multiple regression it was revealed that the NFB at R and NR, PSC at R and NR, PSM at R and NR and MBC at NR are only important predictors which can explain 99 % of total variance.

Key words: Boron, Microbial properties, Non-rhizosphere, Organic matter, Rhizosphere, Yield

Boron is one of the essential micronutrients required for plant growth. and stimulates microbial activity in both rhizosphere and non-rhizosphere soils. Micro-organisms help to increase the nutrient use efficiency especially boron through integrated boron management practices (Rai 2006). Soil microorganisms directly influence boron content of soil as maximum boron release corresponds with the highest microbial activity. The essentiality of B for growth and development of higher plants has been earlier demonstrated (Marschner 1995). Boron plays an important role in the movement and metabolism of carbohydrate in the plant, synthesis of plant hormones and nucleic acids, pollen germination, flowering and fruiting, water use, nitrogen assimilation and generative growth of plants. The most important functions of boron in plants are thought to be its structural role in cell wall development and stimulation or inhibition of specific metabolism pathways (Ahmad *et al.* 2009). Boron in the soil, an important fraction is associated with organic matter and is released through microorganism activities (Das 2007). Recently, Nelson and Mele (2007) reported that B and sodium chloride are more likely to affect rhizosphere microbial community structure indirectly through root exudates quantity and/or quality than directly through microbial toxicity, and that plant health is a major determinant in rhizosphere microbial community structure and normal N cycle. Several studies have pointed out the essentiality of B for N_2 fixation in the heterocyst of the cyanobacterium *Anabaena* PCC 7119 (García- González *et al.* 1990) and in the vesicles of actinomycetes of the genus *Frankia* (Bolaños *et al.* 2002). Both types of microorganisms require B for the stability of the envelopes that protect nitrogenase from inactivation by oxygen when grown under N_2- fixing conditions. Keeping these in view, the present study was undertaken to study the influence of boron and organic matter on soil biology, growth and yield of rape (*Brassica campestris* L.) in an aeric endoaquept.

MATERIALS AND METHODS

Field experiments were conducted during two winter seasons of 2006-2007 and 2007-2008 with rape (cv B 9) in an Aeric Endoaquept (pH 6.8, organic carbon, 0.52%, CEC 12.5 c mol (p+)/kg, hot calcium chloride (HCC) extractable B 0.23 mg/kg, non-symbiotic nitrogen fixing bacteria 45.32 × 10^5CFU/g soil, non-symbiotic nitrogen fixing capacity 7.92 mg of N fixed/g of soil/g sucrose consumed, P-solubilising microorganisms 40.91×10^5 CFU/g soil; P solubilising capacity, 0.018 mg P per 15 mg insoluble P/kg soil/0.15 g sucrose; microbial biomass carbon, 115.31 µg/g

[1]e-mail: mitali_mandal@rediffmail.com, Department of Soil Science & Agricultural Chemistry, [2]e-mail: dkdas1231@rediffmail.com, Department of Agricultural Chemistry and Soil Science, Bidhan Chandra Krishi Viswavidyalaya, P O Mohanpur, Nadia,West Bengal 741 252

dry soil) at Bidhan Chandra Krishi Viswavidyalaya Instructional Farm, Jagulia, West Bengal, India. In the present study the treatments were used as control, only recommended dose of fertiliser (80:40:40) (T_1); N, P_2O_5 and K_2O as recommended + organic manure @ 5 tonnes/ha (T_2); N, P_2O_5 and K_2O as recommended + B as calbor (1 kg B/ha) (T_3); N, P_2O_5 and K_2O as recommended + B as calbor (0.5 kg B/ha) + organic manure @5 tonnes/ha (T_4); N, P_2O_5 and K_2O as recommended + B as borax (1 kg B/ha) (T_5); N, P_2O_5 and K_2O as recommended + B as borax (0.5 kg B/ha)+ organic manure @5 tonnes/ha (T_6). Experiments were laid out in a randomised block design (RBD), and the respective treatments were applied to each plot. Each treatment was replicated thrice. A basal dose of each P_2O_5 and K_2O was applied at 40 kg/ha and N was applied at 80 kg/ha in two splits, i.e. 40 kg N/ha at basal , 40 kg N/ha at 21 days after sowing. Nitrogen, P_2O_5 and K_2O were applied in the form of urea , single super phosphate and muriate of potash respectively. Both calbor (4.5 % B, 1.7 % K as K_2O, 11% Ca as CaO, 12 % S , 1% Mg) and borax (10.5 % B) as a source of B were applied as basal. Organic matter as farmyard manure (FYM) containing 0.38 mg/kg B was incorporated at the time of final land preparation.

Soil samples were collected periodically at an interval of 30 days from five to six randomly selected locations in each plot from both R and NR regions of each plot up to 90 days after sowing. For counting of total number of non-symbiotic nitrogen fixing bacteria in soil, Jenson's nitrogen free agar medium (Jensen 1930) was used.Non – symbiotic N – fixing power of the soil was examined by estimating nitrogen after incubating 1 g moist soil at 30°C ± 1°C in 50 mL sterile nitrogen free Jenson's broth containing 20 g sucrose in 250 mL conical flask. The idea of providing double amount of energy material was to have profuse growth of aerobic non – symbiotic N – fixing bacteria. Four flasks were kept for each soil, of which two flasks were sterilized after addition of soil. All four flasks were incubated for 15 days. After 15 days of incubation, all flasks were analysed for total nitrogen, following Kjeldahl method (Jackson 1973). The difference in the amount of nitrogen in the non-sterilized and sterilized flasks were the nitrogen – fixing power of soils, expressed as mg of nitrogen fixed/g of soil/g sucrose consumed. For phosphate solubilizing micro-organisms, sucrose-tricalcium phosphate agar media (Pikovskaia 1948) was used. Phosphate solubilizing capacity was determined by estimating soluble phosphorous in 15 ml of sucrose – tricalcium phosphate both containing 1% sucrose after incubating 1 g of soil in culture tubes at 30 ± 1° C for 15 days (Olsen and Dean 1982). For estimating phosphate solubilizing capacity of soil, 1 g soil was added aseptically in each of the four culture tubes containing 15 ml broth having 15 mg insoluble phosphorous in 75 mg of tricalcium phosphate and 0.15 g of sucrose. Two tubes were sterilized immediately after addition of soil and all the four tubes were incubated at 30 ± 1° C for 15 days. After incubation, the tubes were centrifuged at 6000 rpm for 20 minutes. After filtering, the supernatant through Whatman filter paper (No. 42), 5 ml sterilized and 1 ml non-sterilized aliquot were taken separately and water soluble phosphorous was estimated with the help of a double cell photoelectric colorimeter following chloromolybdic stannous chloride method (Olsen and Dean 1982).The difference in the amount of soluble phosphorous in non-sterilized and sterilized tubes would be the phosphate solubilizing capacity of the soil sample.Microbial biomass carbon was determined in soil samples by fumigation extraction procedure described by Joergenson (1995).

Yields were recorded after harvesting the crop. All replicated data for 2 years were pooled statistically. Univariate ANOVA technique for RCBD followed by Duncan's test at 5 % level of significance was used for comparing day-wise mean boron concentration. General Linear Model technique repeated over 4 DAS was used further to compare the main effects. Such study was followed both for R and NR soils. Similar technique was also used to compare the mean difference between R and NR in different DAS and treatments. All main effects means were subjected to Post–hoc tests like Duncan's test and day means along with R and NR means were tested by LSD (Least Significant Difference) to identify the homogeneous means at 5% level of significance.

Principal Component Analysis (PCA) attempts to identify underlying variables, or factors, that explain the pattern of correlations within a set of observed variables. Factor analysis was often used in data reduction to identify a small number of factors that explain most of the variance observed in a much larger number of manifest variables. Principal Component Analysis (PCA) is a factor extraction method which was used to form uncorrelated linear combinations of the observed variables. The first component has maximum variance. Successive components explain progressively smaller portions of the variance and are all uncorrelated with each other. Principal components analysis is used to obtain the initial factor solution. It can be used even when a correlation matrix was singular.

RESULTS AND DISCUSSION

Phosphate solubilising micro-organisms and phosphate solubilising capacity

The results (Table 1) reveal that the population of phosphate solubilising microorganisms (PSM) has been found to be varied with treatments. The PSM was recorded lowest in the control treatment where only recommended NPK was applied. Comparing the results of different treatments, the highest proliferation of PSM was maintained in the treatment T_4 where recommended NPK, boron at 0.5 kg/ha as calbor and organic manure at 5 tonnes/ha was applied compared to other treatments in both rhizosphere (R) and non-rhizosphere (NR) soils which might be explained by the stimulating effect of calcium present in the calbor as well as application of organic manure(Bonilla and Bolanos 2004) who also reported that boron – calcium relationship for biological nitrogen fixation as well as the stimulating effect of phosphate solubilising microorganisms.

Table 1 Effect of organic matter and boron application on the changes of phosphate solubilizing microorganisms (× 10^5/g soil) in soil during the year 2006-2008 (Pooled)

Treatment	Days after sowing								Mean	
	30		50		70		90			
	R	NR	R	NR	R	NR	R	NR	R	NR
T_1: control (only NPK recommended	62.84[e]	48.94[e]	49.63[e]	36.24[f]	42.85[e]	28.92[f]	29.91[e]	19.26[e]	46.30[E]	33.34[M]
T_2: NPK as recommended + organic manure @5 t/ha	75.16[d]	61.26[d]	62.25[d]	52.55[e]	55.64[d]	46.44[e]	44.53[d]	35.57[d]	59.40[D]	48.95[L]
T_3: NPK as recommended + B as calbor (1 kg B/ha)	81.27[bc]	72.77[b]	77.06[b]	60.07[c]	68.45[b]	52.16[c]	61.93[b]	39.18[c]	72.18[B]	56.04[J]
T_4: NPK as recommended + B as calbor (0.5 kg B/ha) + organic manure @5 t/ha	94.70[a]	78.28[a]	82.86[a]	68.57[a]	75.75[a]	57.76[a]	67.84[a]	48.49[a]	80.29[A]	63.28[I]
T_5: NPK as recommended + B as borax (1 kg B/ha)	77.86[cd]	68.17[c]	71.51[c]	57.36[d]	63.94[c]	49.85[b]	58.31[c]	37.77[c]	67.91[C]	53.29[K]
T_6: NPK as recommended + B as borax (0.5 kg B/ha)+organic manure @5 t/ha	84.27[b]	72.08[b]	80.36[a]	63.2[b]	72.55[a]	55.26[d]	62.24[b]	42.48[b]	74.86[B]	58.27[J]
Mean	79.35[P]	66.92[W]	70.61[Q]	56.34[X]	63.19[R]	48.40[Y]	54.13[S]	37.12[Z]	66.92[U]	52.20[V]
SEm (±)	1.153	0.929	0.979	0.795	1.175	0.675	0.763	0.542	0.332	0.332
CD (P = 0.05)	3.63	2.92	3.08	2.50	3.70	2.12	2.40	1.70	1.045	1.045

Within a column :means followed by the same letter are not significantly different at the 0.05 level of probability by Duncan's multiple range test (DMRT). R – Rhizosphere; NR–non-rhizosphere

The results further envisaged that the proliferation of PSM in NR soils was maintained always lower than that of R soils which might be explained by the effect of rhizodeposition where in the former case it is lowest or practically nil. The applied B fertilizer was also helpful in creating a better soil biological environment in the cultivated rapeseed soil which also finds support from the present study by the increased microbial population. The population of bacteria, fungi and actinomycetes significantly increased with B application level (Bharadwaj and Datta 1995).The results (Table 2) reveal that the phosphate solubilising capacity(PSC) in rhizosphere (R) and non-rhizosphere (NR) soils has been found progressively decreased irrespective of treatments. However, comparing the results of different treatments it was observed that the PSC was recorded highest (0.045 mg/g soil) in R and 0.034 mg/g in NR soils under the treatment T_4 where recommended levels of NPK (80:40:40), boron at 0.5 kg/ha as calbor and organic manure at 5 tonnes/ha was applied at the initial 30 days period of crop growth.

Table 2 Effect of organic matter and boron application on the changes in phosphate solubilizing capacity (mg/g dry soil) in soil during the year 2006-2008 (Pooled)

Treatment	Days after sowing								Mean	
	30		50		70		90			
	R	NR	R	NR	R	NR	R	NR	R	NR
T_1: control (only NPK recommended	0.032[f]	0.024[e]	0.026[e]	0.021[d]	0.022[e]	0.015[e]	0.017[e]	0.015[a]	0.024[E]	0.018[N]
T_2: NPK as recommended + organic manure @5 t/ha	0.035[e]	0.026[d]	0.029[d]	0.022[d]	0.025[d]	0.019[cd]	0.020[d]	0.017[a]	0.027[D]	0.021[M]
T_3: NPK as recommended + B as calbor (1 kg B/ha)	0.040[c]	0.039[b]	0.037[b]	0.025[c]	0.032[b]	0.020[c]	0.028[b]	0.023[a]	0.034[B]	0.026[K]
T_4: NPK as recommended + B as calbor (0.5 kg B/ha)+ organic manure @5 t/ha	0.045[a]	0.034[a]	0.040[a]	0.030[a]	0.036[a]	0.027[a]	0.033	0.037[a]	0.038[A]	0.032[I]
T_5: NPK as recommended + B as borax (1 kg B/ha)	0.037[d]	0.027[c]	0.032[c]	0.022[d]	0.028[c]	0.018[d]	0.022[c]	0.014[a]	0.029[C]	0.020[L]
T_6: NPK as recommended + B as borax (0.5 kg B/ha)+ organic manure @5 t/ha	0.042[b]	0.031[b]	0.038b	0.027[b]	0.034[b]	0.023[b]	0.029[b]	0.019[a]	0.035[B]	0.025[J]
Mean	0.039[P]	0.029[W]	0.034[Q]	0.025[X]	0.030[R]	0.020[Y]	0.025[S]	0.032[Z]	0.031[U]	0.023[V]
SEm (±)	0.001	0.001	0.001	0.0009	0.001	0.0009	0.001	0.001	0.0009	0.009
CD (P = 0.05)	0.003	0.003	0.003	0.002	0.003	0.002	0.003	NS	0.002	0.002

Within a column :means followed by the same letter are not significantly different at the 0.05 level of probability by Duncan's multiple range test (DMRT). R – Rhizosphere; NR – non-rhizosphere

The decrease of the same at the latter period of crop growth might be due to its continuous uptake by crops caused by greater biomass production. Comparing the results (Table 2) of R and NR soils, it was found that the amount of phosphate solubilized was far greater in R soils than that of NR soils which possibly due to favourable effect of root exudates for the solubilisation of insoluble phosphatic compounds. Abou-El-Yazeid *et al.* (2007) also reported similarly where they found a positive and significant microbial activity in soil rhizosphere expressing by activity of dehydrogenase, phosphatase and nitrogenase enzymes and available phosphorous reacting a maximum value due to inoculation of *P. polymyxa* and *B. megatarium* and B application.

Non-symbiotic nitrogen fixing bacteria and nitrogen fixing capacity

The amount of non symbiotic nitrogen fixing bacteria

Table 3 Effect of organic matter and boron application on the changes in non symbiotic nitrogen fixing bacteria (10^5/g soil) in soil during the year 2006-2008 (Pooled)

Treatment	Days after sowing								Mean	
	30		50		70		90			
	R	NR	R	NR	R	NR	R	NR	R	NR
T_1: control (only NPK recommended	69.32^{e}	50.32^{e}	53.12^{f}	38.94^{e}	49.13^{e}	31.23^{e}	32.02^{e}	20.11^{e}	50.90^{F}	35.15^{N}
T_2: NPK as recommended + organic manure @5 t/ha	84.19^{d}	62.12^{d}	75.08^{e}	55.40^{d}	62.94^{d}	49.55^{d}	55.93^{d}	39.17^{d}	69.54^{E}	51.56^{M}
T_3: NPK as recommended + B as calbor (1 kg B/ha)	94.48^{bc}	75.83^{b}	86.26^{c}	62.27^{c}	72.45^{c}	54.26^{c}	62.34^{b}	43.28^{c}	78.88^{C}	58.91^{K}
T_4: NPK as recommended + B as calbor (0.5 kg B/ha)+ organic manure @5 t/ha	107.33^{a}	89.47^{a}	98.85^{a}	72.58^{a}	85.56^{a}	65.07^{a}	70.05^{a}	50.03^{a}	90.45^{A}	69.29^{I}
T_5: NPK as recommended + B as borax (1 kg B/ha)	90.92^{c}	70.28^{c}	82.58^{d}	55.56^{d}	65.15^{d}	51.85^{d}	58.04^{d}	40.92^{d}	74.17^{D}	54.65^{L}
T_6: NPK as recommended + B as borax (0.5 kg B/ha)+ organic manure @5 t/ha	96.72^{b}	79.03^{b}	90.04^{b}	67.81^{b}	77.95^{b}	59.76^{b}	67.84^{a}	48.21^{b}	83.14^{B}	63.70^{J}
Mean	90.49^{P}	71.18^{W}	80.99^{Q}	58.76^{X}	68.86^{R}	51.95^{Y}	57.70^{S}	40.29^{Z}	74.51^{U}	55.54^{V}
SEm (±)	1.271	1.026	1.145	0.868	0.993	0.746	0.813	0.574	0.368	0.368
CD (P = 0.05)	4.00	3.23	3.60	2.73	4.14	2.34	2.56	1.80	1.159	1.159

Within a column :means followed by the same letter are not significantly different at the 0.05 level of probability by Duncan's multiple range test (DMRT). R – Rhizosphere; NR- Non-rhizosphere

Table 4 Effect of organic matter and boron application on the changes in nitrogen fixing capacity (mg of N_2 fixed/g of soil/g sucrose) in soil during the year 2006-2008 (Pooled)

Treatment	Days after sowing								Mean	
	30		50		70		90			
	R	NR	R	NR	R	NR	R	NR	R	NR
T_1: control (only NPK recommended	10.45^{c}	8.33^{c}	10.32^{c}	8.12^{c}	10.21^{b}	7.96^{c}	10.07^{c}	7.65^{c}	10.26^{C}	8.02^{K}
T_2: NPK as recommended + organic manure @5 t/ha	11.35^{b}	9.28^{b}	11.24^{b}	9.16^{b}	11.13^{a}	9.02^{b}	10.92^{b}	8.89^{b}	11.16^{B}	9.09^{J}
T_3: NPK as recommended + B as calbor (1 kg B/ha)	11.48^{ab}	9.62^{ab}	11.37^{ab}	9.49^{ab}	11.22^{a}	9.32^{ab}	11.01^{ab}	9.17^{ab}	11.27^{AB}	9.40^{J}
T_4: NPK as recommended + B as calbor (0.5 kg B/ha)+ organic manure @5 t/ha	11.96^{a}	9.89^{a}	11.82^{a}	9.70^{a}	11.63^{a}	9.55^{a}	11.49^{a}	9.39^{a}	11.73^{A}	9.63^{I}
T_5: NPK as recommended + B as borax (1 kg B/ha)	11.42^{b}	9.53^{ab}	11.30^{b}	9.40^{ab}	11.12^{a}	9.26^{ab}	10.99^{ab}	9.10^{ab}	11.21^{AB}	9.32^{IJ}
T_6: NPK as recommended + B as borax (0.5 kg B/ha)+ organic manure @5 t/ha	11.69^{ab}	9.76^{a}	11.60^{ab}	9.58^{a}	11.52^{a}	9.42^{ab}	11.40^{ab}	9.27^{ab}	11.55^{AB}	9.51^{IJ}
Mean	11.39^{P}	9.40^{W}	11.28^{Q}	9.24^{X}	11.14^{R}	9.09^{Y}	10.98^{S}	8.91^{Z}	11.20^{U}	9.16^{V}
SEm (±)	0.154	0.127	0.152	0.123	0.151	0.123	0.149	0.120	0.054	0.054
CD (P = 0.05)	0.485	0.400	0.478	0.387	0.475	0.387	0.469	0.378	0.170	0.170

Within a column :means followed by the same letter are not significantly different at the 0.05 level of probability by Duncan's multiple range test (DMRT). R – Rhizosphere; NR- Non-rhizosphere

Table 5 Effect of organic matter and boron application on the changes in microbial biomass carbon (µg/g dry soil) in soil during the year 2007-2008 (Pooled)

Treatment	Days after sowing								Mean	
	30		50		70		90			
	R	NR	R	NR	R	NR	R	NR	R	NR
T_1: control (only NPK recommended	145.95[c]	121.39[d]	138.20[d]	112.60[d]	122.89[c]	110.32[e]	110.89[d]	103.66[d]	129.48[D]	111.99[L]
T_2: NPK as recommended + organic manure @5 t/ha	165.28[b]	149.72[c]	151.72[c]	142.52[c]	147.20[b]	132.32[d]	122.30[c]	126.60[bc]	146.62[C]	137.79[K]
T_3: NPK as recommended + B as calbor (1 kg B/ha)	167.39[b]	153.42[bc]	153.62[bc]	147.60[abc]	149.42[b]	135.57[cd]	124.52[c]	128.41[b]	148.74[BC]	141.25[JK]
T_4: NPK as recommended + B as calbor (0.5 kg B/ha) + organic manure @5 t/ha	175.90[a]	161.99[a]	164.21[a]	152.81[a]	159.30[a]	147.50[a]	138.50[a]	136.00[a]	159.48[A]	149.58[I]
T_5: NPK as recommended + B as borax (1 kg B/ha)	169.39[ab]	150.31[c]	159.60[ab]	145.99[bc]	151.45[ab]	139.20[bc]	127.49[bc]	121.98[c]	151.98[BC]	139.37[JK]
T_6: NPK as recommended + B as borax (0.5 kg B/ha) + organic manure @5 t/ha	172.11[ab]	158.60[ab]	160.75[ab]	150.21[ab]	153.42[ab]	142.37[ab]	132.80[b]	125.78[bc]	154.77[AB]	144.24[IJ]
Mean	166.00P	149.24[W]	154.68[Q]	141.96[X]	147.28[R]	134.55[Y]	126.08[S]	123.74[Z]	148.51[U]	137.37[V]
SEm (±)	2.240	2.025	2.292	1.911	2.503	1.829	1.730	1.696	0.764	0.764
CD (P = 0.05)	7.04	6.37	6.58	6.02	6.31	5.76	5.44	5.34	2.406	2.406

Within a column :means followed by the same letter are not significantly different at the 0.05 level of probability by Duncan's multiple range test (DMRT). R–Rhizosphere; NR–non-rhizosphere

(NFB) was showed a similar pattern of changes to that of non symbiotic nitrogen fixing capacity (NFC) in soils in both R and NR soils (Table 3).The highest population of bacteria, fungi and actinomycetes and CO_2- C production were observed at 3 kg/ha B level in different growing periods of the plants and in different soil depths (Bilen *et al.* 2011). The results suggested that the proliferation of NFB was recorded always highest in rhizosphere (R) soil compared to non-rhizosphere (NR) soil, which might be explained by stimulating effect of root exudates on bacterial growth.

The results (Table 4) reveal that the amount of NFC was followed a similar pattern of changes with that of PSC in soil. but varied with amounts. At the initial 30 days period of growth , N_2 – fixing capacity has been found to be increased in both rhizosphere (R) and non-rhizosphere (NR) soils, being highest (11.96 mg/N g sucrose) in the treatment T_4, while that of the same was highest (9.89 mg N/g sucrose) in the non-rhizosphere (NR) soil. The results further envisaged that the N_2 – fixing capacity was maintaining a higher value in the T_4 treatment throughout the crop growth period up to 90 days. The highest NFC in the R soil might be partly due to accretion of nitrogen resulting from the secretion of plant roots (root exudates)(Oger *et al*. 2004).

Microbial biomass carbon

The results (Table 5) show that the absolute amount of microbial biomass carbon (MBC) has been found to be consistently lower in both rhizosphere (R) and non-rhizosphere (NR) soils with the progress of crop growth irrespective of treatments. However, the amount of MBC content was maintained higher with an integrated application of boron and organic matter over that of the control. Comparing the results of different treatments it was found that the amount of MBC content was maintained highest in the treatment T_4 throughout the crop growth period in both R and NR soils. Nelson and Mele (2007) also reported that the application of B and NaCl to soil changed rhizosphere microbial community structure (MCS) indirectly through

Table 6 Effect of boron and organic matter application on the changes in seed yield (q/ha) of rape seed

Treatment	Seed yield		
	2006-07	2007-08	Pooled
T_1: control (only NPK recommended	5.60[e]	5.58[e]	5.59[E]
T_2: NPK as recommended + organic manure @5 t/ha	6.80[d]	6.87[d]	6.84[D]
T_3: NPK as recommended + B as calbor (1 kg B/ha)	8.10[b]	8.15[b]	8.12[B]
T_4: NPK as recommended + B as calbor (0.5 kg B/h) + organic manure @5 t/ha	8.90[a]	8.99[a]	8.95[A]
T_5: NPK as recommended + B as borax (1 kg B/h)	7.20[c]	7.31[c]	7.26[C]
T_6: NPK as recommended + B as borax (0.5 kg B/ha) + organic manure @5 t/ha	7.80[b]	7.83[b]	7.82[B]
SE.m (±)	0.105	0.106	0.105
CD (P = 0.05)	0.330	0.333	0.330

Table 7 Regression results following stepwise method of analysis where both criterion measure and predictors are mean results over all four days after sowing with allowed tolerance = 0.0001

Predictors	Equation number	Equations	R^2	Adj. R^2	SE (est)
NFC at Z_1 & Z_2, NFB at Z_1 & Z_2, PSC at Z_1 & Z_2, PSM at Z_1 & Z_2, MBC at Z_1 & Z_2	1	Seed yield =-0.11-0.24NFBZ_1** +27.00PSCZ_2** +0.26PSMZ_2** +0.25PSMZ_1**–0.03MBCZ_2** –0.05NFBZ_2**–7.31PSCZ_1	0.99**	0.99	0.008

**Significant at 1% level, *significant at 5 % level

Table 8 Regression results where predictors are weighted by principal component based component loadings (Principal Component Regression based upon regression factor scores of predictors) #

Predictors	Equation number	Equations	R^2	Adj. R^2	SE (est)
NFC at Z_1 & Z_2, NFB at Z_1 & Z_2, PSC at Z_1 & Z_2, PSM at Z_1 & Z_2, MBC at Z_1 & Z_2	2	Seed yield =7.43+0.70 PSMZ_1**+ 0.39PSMZ_2*	0.97**	0.96	0.209

Note: Component weights used for regression factor scores used for each predictor is listed in Table 9. ** Significant at 1% level, *significant at 5 % level

Table 9 Principal component weights for first component explaining largest variance

Variable	Days After Sowing				Variance
	30	50	70	90	accounted for
BZ_1E_1	1.00	1.00	1.00	1.00	99.58%
BZ_2E_1	1.00	1.00	1.00	1.00	99.58%
NFC at Z_1	1.00	1.00	1.00	1.00	99.69%
NFB at Z_1	0.99	1.00	0.99	0.98	98.03%
PSC at Z_1	0.99	0.99	1.00	0.99	98.34%
PSM at Z_1	0.97	1.00	1.00	0.99	97.52%
MBC at Z_1	0.99	0.99	0.99	0.98	97.86%
NFC at Z_2	1.00	1.00	1.00	1.00	99.77%
NFB at Z_2	0.98	1.00	1.00	0.98	97.90%
PSC at Z_2	0.98	0.95	0.98	-0.58	78.97%
PSM at Z_2	0.99	1.00	1.00	0.99	98.87%
MBC at Z_2	1.00	0.99	0.99	0.97	97.40%

increased soil moisture and subtle changes in root exudate patterns resulting greater rhizo-deposition which favours greater proliferation of microorganisms in the rhizosphere compared to non-rhizosphere soil.

The results further revealed that the application of organic manure at 5 tonnes/ha along with NPK fertilizer increased the MBC content in soil over control, being further enhanced with the application of boron at 0.5 kg/ha as calbor and organic manure at 5 tonnes/ha. This might be due to the presence of calcium and nitrogen in the calbor in addition to boron in both R and NR soils. Boron can inhibit growth of bacteria especially at higher concentration of applied B (Guhl 1996).

As regards to the rhizosphere (R) and non-rhizosphere (NR) soils, it was found that the amount of MBC content was more in the treatment receiving boron as calbor in R soils which might be explained by the greater proliferation of microbial population resulting from the root exudates. Increasing B application has rapidly changed soil microbial biomass and biologically active fraction in soil.The availability of readily mineralised C and N and improvement in the physico-chemical properties of the soil might have improved the microbial population of the soil (Bharadwaj and Datt 1995).

The results (Table 6) show that the yield of rape seed during the year 2006 – 07 and 2007 – 08 showed a significant variation with different treatments. However, the yield of rape seed has been found to be increased due to different treatments in both the years, being recorded highest yield of rape seed (8.90 and 8.99 q/ha) in the treatment T_4 where NPK (80:40:40), B at 0.5 kg/ha as calbor and organic manure at 5 tonnes/ha was applied. Salroo *et al*. (2002) and Zou *et al.* (2008) also reported similarly who showed that the application of boron through integrated system increased the yield and yield components of rape.

Mean values of NFC at R and NR, NFB at R and NR, PSC at R and NR , PSM at R and NR and MBC at R and NR were calculated over four different time period prior to regression analysis as described before. When predictors variables are used to predict the yield (equation 1), the stepwise technique of multiple regression revealed that the NFB at R and NR, PSC at R and NR , PSM at R and NR and MBC at NR are only important predictors which can explain 99 % of total variance. Principal component regression technique was further used following similar technique and same predictor variables but here predictors are the regression factor score resulted by principal component analysis. Each predictor which was measured at 4 different time periods was actually constructed this factor score and the principal component weights will dictate the contribution of variance of each day in such score according to the importance of total variance of this predictors (Table 8). The results revealed that PSM at Z_1 and Z_2 are the only important predictors which can explain 97 % of total variance (equation 2).

The overall results suggest that the application of boron in any source and levels integrated with organic manures increases the microbial proliferation especially in rhizosphere soil of rape with the simultaneous increase in yield.

REFERENCES

Abou El-Yazeid A, Abou-Aly H E, Mady M A and Moussa S A M.2007. Enhancing growth, productivity and quality of squash plants using phosphate dissolving microorganisms (Bio phosphor) combined with boron foliar spray. *Research Journal of Agriculture and Biological Sciences* **3**(4):274–86.

Ahmad W, Niaz A, Kanwal S, Rahmatullah M K and Rasheed M K. 2009. Role of boron in plant growth: A Review. *Journal of Agricultural Research* **47**(3).

Bharadwaj K K R and Datt N. 1995. Effects of legume green manuring on nitrogen mineralization and some microbiological properties in an acid rice soil. *Biology and Fertility of Soils* **19(1)**: 19–21.

Berger K C and Pratt P J .1963. Advances in secondary and micronutrient fertilization. (*In*) *Fertilizer Technology and Use*, pp 281–340. McVickar M H, Bridger G L, Nelson L B (Ed.). Soil Science Society of America, Madison, WI.

Bilen S, Bilen M and Bardhan S. 2011. The effects of boron management on soil microbial population and enzyme activities. *African Journal of Biotechnology* **10**(27):5 311–9.

Bolaños L, Redondo-Nieto M, Bonilla I and Wall L G. 2002. Boron requirement in the *Discaria trinervis* (Rhamnaceae) and *Frankia* symbiotic relationship. Its essentiality for *Frankia* BCU110501 growth and nitrogen fixation. *Physiologia Plantarum* **115(4)**: 563–70.

Bonilla I and Bolanos L. 2004. Boron-Calcium relationship in biological nitrogen fixation under phyisiological and salt stressing conditions. *Production practices and quality assessment of food crops* **2**: 139–70.

Das D K. 2007. *Micronutrients: Their behavior in Soils and Plants*, 2nd Edn. Kalyani Publishers, Ludhiana/New Delhi.

García-González M, Mateo P and Bonilla I .1990. Effect of boron deficiency on photosynthesis and reductant sources and their relationship with nitrogenase activity in *Anabaena* PCC 7119. *Plant Physiology* **93**:560–5.

Guhl W. 1996. Ecological aspects of boron. *SOFW-Journal* **118**:1159–68

Jackson M L. 1973. *Soil Chemical Analysis.* Prentice-Hall of India Private Ltd, New Delhi.

Jensen H L. 1930. Azotobacteriaceae. *Bacteriology Reviews* **189**:195–14.

Joergensen R G. 1995. Microbial biomass. (In) *Methods in Applied Microbiology and Biochemistry*, pp 382–6. Alef K and Nannipieri P (Eds). Academic Press.

Nelson D R and Mele P M. 2007. Subtle changes in the rhizosphere microbial community structure in response to increased boron and sodium chloride concentrations. *Soil Biology and Biochemistry* **39**: 340–51.

Oger P M, Mausouri H, Nesme X and Dessaux Y. 2004. Engineering root exudation of Lotus toward the production of two novel carbon compounds leads to the selection of distinct microbial populations in the rhizosphere. *Miocrobial Ecology* **47**: 96–103.

Olsen S R and Dean L A. 1982. Phosphorous. (In) *Methods of Soil Analysis,* Part 2, pp 1035–49. Black C A, Evans D D, White J L, Ensminger L E , Clark F E and Dinaner R C (Eds). American Society of Agronomy Inc., Madison, Wisconsin, USA.

Pikovskaya R I. 1948. Mobilization of phosphorus in soil in connection with the vital activity of some microbial species. *Microbiologiya* **17**: 362–70.

Rai M K. 2006. *Handbook of microbial biofertilizers.* Food Products Press, an imprint of The Haworth Press, Inc, Binghamton, New York.

Salroo M N Y, Bhat M A, Muski G M and Sheikh F A. 2002. Integrated nutrient management studies in brown sarson (*B Campestis* L.) under temperate conditions. *Plant Archives* **2**(1): 37–9.

Zou J, Lu J W, Liao Z W, Gong X M, Wang H, Zhou Y G and Zhou H.2008. Study on response of rapeseed to boron application and critical level of soil available B in Hubei province. *Scientia Agriculture Sinica* **41**(3): 752–9.