

Population dynamics of cyanobacteria in alluvial rice grown soils of lower Brahmaputra floodplain

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Abstract

The present investigation was carried out to study the cyanobacterial diversity and population dynamics during different months in relation to soil physico-chemical parameters in various rice fields in alluvial flood plain of the Brahmaputra River. Altogether 71 species of cyanobacteria under 20 genera and 9 families were recorded. Among the species heterocystous filamentous were dominant (80%) over the other forms and Nostocaceae (54%) was predominant in all the three sites followed by Chroococcaceae (14%) and Rivulariaceae (13%) respectively. In case of population number it ranged in between $0.14 \pm 10^3/g$ to $9.35 \pm 9.68 \times 10^4/g$ of soil. It was recorded highest during July-August (monsoon season) followed by September-October (post monsoon), March-June (pre monsoon) and November-February (winter season) respectively. Principle component analysis (PCA) justified the seasonal pattern and identified close relation to soil parameters like temperature, moisture, pH, phosphorus and sodium. Pearson's correlation analysis also revealed significant negative correlation of cyanobacterial number with soil nitrogen, organic carbon and conductivity in the floodplain soils.

Key words: Alluvial soil, Brahmaputra River, cyanobacteria, PCA ordination, population dynamics, rice fields, soil physico-chemical parameters.

Introduction

Cyanobacteria are the successful phototrophic prokaryotes and most widely distributed group among soil micro-organisms (Stanier and Cohen-Bazire, 1977). They are found in almost every conceivable environment including vast oceanic areas to freshwater lakes, temperate soils to bare rocks and even extreme habitats like arid deserts, frigid lakes, or hot springs (Hoffmann, 1989). Some of the cyanobacterial taxa are capable of fixing the atmospheric nitrogen and hence maintain the nitrogen-level in the soil in which they grow and flourish (Venkataraman, 1993). Their presence in various ecosystems has also been reported to increase the oxygen concentration and improve the physico-chemical properties of the soil (Mandal *et al.*, 1998). Among the diverse habitats rice fields provide a very congenial condition for abundant growth of N₂-fixing cyanobacteria (Nayak *et al.*, 2001; Whitton, 2000). However their diversity, abundance and contribution to the total nitrogen fixed in rice fields are greatly affected by soil, climatic conditions (Pabbi, 2008), different seasons and rice cultivation cycle. So, it becomes utmost important for the algologist to know the factors that allow N₂-fixing cyanobacteria to establish and bloom in rice fields. Considerable amount of research have been carried out on diversity and distribution of the cyanobacterial flora of rice fields of India (Kotle and Goyal, 1986; Mishra and Pabbi, 2004; Digambar Rao *et al.*, 2008; Dey *et al.*, 2010; D'souza *et al.*, 2011). A few attempts have also been made in finding the diversity of cyanobacteria in rice fields of Assam (Saikia and Bordoloi, 1994; Ahmed *et al.*, 1999; Dihingia and Baruah, 2011; Bharadwaj and Baruah, 2013; Deb *et al.*, 2013). Though a little information on the soil physico-chemical parameters modulating variation on cyanobacterial population particularly from rice fields were available till date (Chunleuchanon *et al.*, 2003; Song *et al.*, 2005; Thamizh Selvi and Sivakumar, 2011; Das *et al.*, 2011), no such study has been reported from the alluvial soils of Brahmaputra floodplain. Therefore, the present endeavor had been taken an effort to study the diversity and population dynamics of cyanobacteria in relation to seasonal variation and soil physico-chemical parameters in different alluvial rice field's soils on both the banks of the Brahmaputra River, so that proper management of cyanobacterial population can be maintained for sustainable agro-ecosystem development in the floodplain region.

Materials and methods

Site selection:

A total of nine (9) rice fields were selected from three different sites on both the floodplains banks of the river Brahmaputra in Kamrup district, Assam, which lies between 25°46' to 26°49' N latitudes and 90°48' to 91°50' E longitudes and covers an area of ca. 4345 sq. km. Since the area is blessed with the mighty Brahmaputra River, the valley region is formed with alluvial deposits, and hence is one of the suitable habitat types for growing rice. A group of three rice grown areas located between 26°08' N latitudes and 91°37' E longitudes were selected from the southwest bank of the river; three rice fields (located between 26°12' N

latitudes and 91°34' E longitudes) from northern bank and three rice fields (located between 26°13' N latitudes and 91°54' E longitudes) from the South east bank of the river Brahmaputra.

The climate of Assam in general may be divided into four seasons. The monsoon season which starts from June and continues till September, receives highest rainfall (>79% of annual rainfall) and is characterized by high humidity (80% to 95%) and high temperature (35°C to 39°C). A short post-monsoon with moderate temperature and rainfall falls during the months of October and November. The winter season starts in November and continues till the month of February. It is basically characterized by scanty rainfall with a minimum temperature of 7°C and the maximum of 24°C and with a relative humidity varying from 60% to 90% respectively. Pre monsoon season starts in the early part of March and extends up to May. Temperature starts rising gradually and at the later part of the season there is occasional thunderstorm, often the weather remains windy and dusty (Chowdhury, 2005).

Sample collection and analysis:

Soil samples were collected in two months intervals from 8-10 randomly selected spots to a depth of 10-15 cm by removing the surface debris. Samples were then mixed, air dried, powdered and sieved and were analyzed for various soil physico chemical parameters following the standard protocols as described by Trivedy *et al.* (1987).

Cyanobacterial culture and its enumeration:

Soil samples weighed 0.5 g were inoculated in sterilized nitrogen free BG11 medium in pre-sterilized flasks under optimal growth condition for 20-25 days at 30⁰ ± 2⁰C temperature in 2.3 K lux light intensity. Enumeration of cyanobacterial number was carried out by MPN technique following Pabbi *et al.* (2010). Identifications of cyanobacteria were done by following the keys given by Desikachary (1959).

Statistical analysis:

Principal component analysis (PCA) and Pearson's correlation coefficient was carried out between N₂-fixing cyanobacterial population and physico-chemical parameters of the soil at different months using the statistical programs XLSTAT and SPSS.

Results

Species diversity:

A total of 71 species of cyanobacteria belonging to 20 genera under 9 families were identified from the study sites. Out of these 71 species enumerated, 57 species belonging to 14 genera under 7 families were filamentous heterocystous form, 10 species belonging to 5 genera under 1 family were unicellular and 4 species belonging to single genus were filamentous non-heterocystous form. Filamentous heterocystous families reported were Nostocaceae with 4 genera (*Anabaena*, *Anabaenopsis*, *Aulosira* and *Nostoc*), followed by Rivulariaceae with 3 genera (*Calothrix*, *Gloeotrichia*, *Rivularia*) and Scytonemataceae with 2 genera (*Scytonema*, *Tolypothrix*). Other filamentous heterocystous species represented by only 1 genus each were under families Stignemataceae (*Hapalosiphon*), Mastigocladaceae (*Mastigocladus*), Mastigocladopsidaceae (*Mastigocladopsis*) and Microchaetaceae (*Microchaete*). The Oscillatoriaceae was the single recorded filamentous non-heterocystous family with the genus *Lyngbya*. The Chroococcaceae with 5 genera (*Aphanocapsa*, *Aphanothece*, *Chroococcus*, *Gloeocapsa* and *Synechococcus*) was the only recorded unicellular family of cyanobacteria.

Maximum number of species was reported under family Nostocaceae with 38 species which were of the genera *Anabaena* (22), *Anabaenopsis* (1), *Aulosira* (3) and *Nostoc* (12). It was followed by Chroococcaceae with 10 species comprising of genera *Aphanocapsa* (4), *Aphanothece* (2), *Chroococcus* (1), *Gloeocapsa* (2) and *Synechococcus* (1). Family Rivulariaceae followed next with 9 species which were of the genera *Calothrix* (6), *Gloeotrichia* (2), *Rivularia* (1). Under family Scytonemataceae only 5 species were reported which were represented by 2 genera viz. *Scytonema* (3) and *Tolypothrix* (2) and that of family Oscillatoriaceae with 4 species that belonged to a single genus *Lyngbya*. The family Stigonemataceae was represented by 2 species belonging to genera *Hapalosiphon* (1) and *Westiellopsis* (1). Families Mastigocladaceae, Mastigocladopsidaceae and Microchaetaceae were represented by 1 species each that belongs to genera *Mastigocladus*, *Mastigocladopsis* and *Microchaete* respectively

Population dynamics of cyanobacteria in relation to soil physico-chemical properties:

The population number of cyanobacteria varies in relation to different months in rice fields situated in three different sites (Fig: 1). Highest population number was recorded in the rice fields situated in the SW (4.07±2.98x10⁴/g soil), followed by North (0.94±0.76x10⁴/g soil) and SE bank of the river Brahmaputra (0.52±0.4x10⁴/g soil). In site SW cyanobacterial population number was recorded maximum during the month of September-October (9.35±9.68x10⁴/g soil) and minimum during January-February (0.9±0.56 x10⁴/g soil). In N bank population number recorded maximum during July-August (2.34± x10⁴/g soil) and minimum during November-December (0.15± x10⁴/g soil). In SE bank of the river, maximum number of cyanobacteria was recorded during the months July-August (1.34± x10⁴/g soil) and minimum during January-February (0.14± x10⁴/g soil).

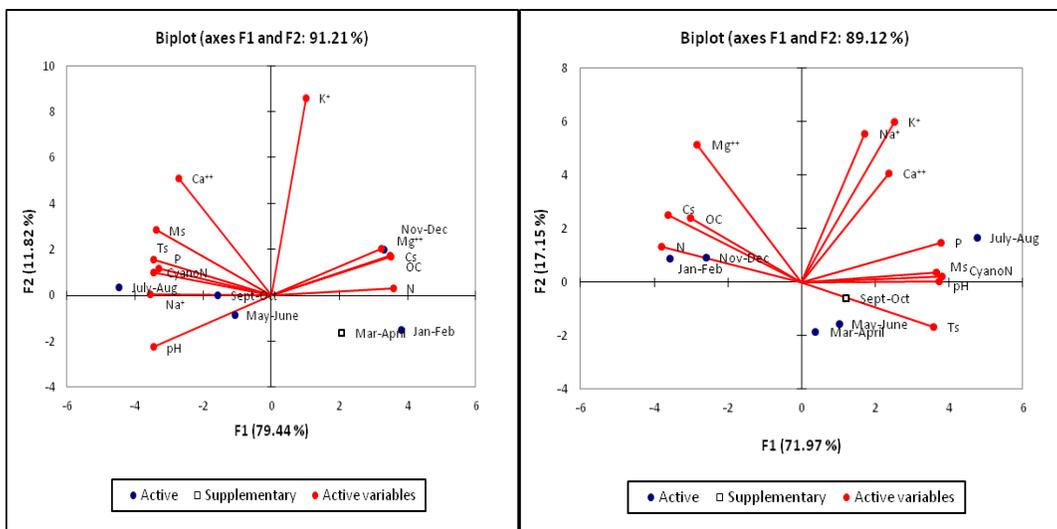


Fig.1: SW bank of Bhramaputra River.

Fig.2: N bank of Brahmaputra River.

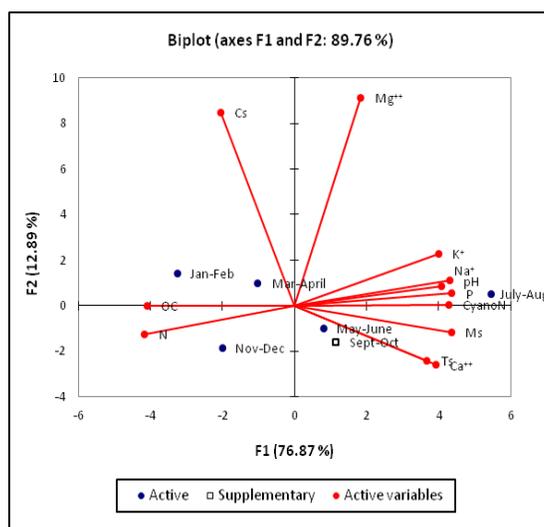


Fig.3: SE bank of Brahmaputra River

Fig 1-3: PCA ordination biplots showing the relationship between soil physico-chemical parameters and cyanobacterial population number in relation to different months.

Soil physico-chemical parameters also varied during different months in the three sites. In SW bank of the river Brahmaputra the soil temperature ranged between 17.6 ± 1.34 to 29.4 ± 0.12 °C, moisture between 18.39 ± 0.49 to 29.91 ± 2.02 %, phosphorus between 70.31 ± 1.56 to 125.84 ± 24.62 mg/100gm, sodium between 0.28 ± 0.023 to 0.86 ± 0.16 meq/100gm and calcium ranged in between 2.28 ± 1.32 to 4.13 ± 0.77 meq/100gm. Maximum values were recorded during July-August and minimum were recorded during January-February. The pH value was varied from 6.30 ± 0.10 to 8.05 ± 0.18 and that of potassium from 0.098 ± 0.12 to 0.39 ± 0.1 meq/100gm respectively. July-August was experienced with highest pH and potassium and overall lowest pH was recorded during November-December and potassium during May-June. Similarly, conductivity, magnesium, organic carbon which varied from 62 ± 21.22 to 105.66 ± 16.2 μ s, 2.386 ± 0.74 to 3.14 ± 0.49 meq/100gm and 1.89 ± 0.66 to 3.39 ± 0.53 % recorded maximum during November-December and minimum during July-August. Nitrogen that ranged between 0.024 ± 0.003 to 0.055 ± 0.001 % recorded maximum level during January-February and minimum during July-August.

In N bank, soil physico-chemical properties like temperature, pH, phosphorus and calcium which varied from 18.5 ± 0.40 to 30.14 ± 0.028 °C, 5.8 ± 0.29 to 7.31 ± 0.57 , 64.93 ± 11.8 to 92.53 ± 4.5 mg/100gm, 1.63 ± 0.44 to 2.77 ± 1.19 meq/100gm

respectively and showed similar trend of fluctuation i.e. maximum during July-August and minimum during January-February. Moisture, potassium and sodium that ranged between 13.62±2.01 to 23.46±0.24 %, 0.16±0.05 to 0.81±0.16 meq/100gm and 0.16±0.08 to 0.51±0.20 meq/100gm recorded maximum during July-August while November-December experienced with minimum moisture, September-October experienced with minimum potassium and May-June with sodium. Conductivity, nitrogen and magnesium that ranged between 50±18.4 to 108.66±34.3 µs, 0.023±0.006 to 0.039±0.009 %, and 1.32±0.37 to 2.12±1.51 meq/100gm showed maximum during January-February while conductivity showed minimum during September-October, nitrogen during July-August and magnesium during May-June respectively. Organic carbon ranged between 1.49±0.12 to 2.67±0.67 % recorded maximum during November-December and minimum during March-April.

In SE bank of the river soil parameters, like temperature ranged between 19.2±0.04 to 29.2±0.05 °C, moisture between 13.33±1.54 to 24.95±1.25 %, pH between 5.45±0.07 to 6.76±0.35, phosphorus between 21.05±3.11 to 65.13±26.9 mg/100gm, calcium between 1.44±0.68 to 2.63±0.15 meq/100gm, and sodium ranged between 0.22±0.10 to 0.62±0.18 meq/100gm. Maximum values were recorded during July-August and minimum were recorded during January-February. Potassium and magnesium that varied from 0.31±0.13 to 1.57±0.45 meq/100gm and 1.42±0.50 to 1.72±0.53 meq/100gm recorded maximum during July-August and minimum during November-December. Conductivity varied from 39.66±9.88 to 73.33±14.14 µs and showed maximum during March-April and minimum during May-June. Both nitrogen (0.067±0.003 to 0.038±0.007 %) and organic carbon (2.41±0.63 to 3.97±0.14 %) showed maximum during January-February and minimum during July-August respectively.

In order to understand the direct or indirect influence of season and soil properties on cyanobacterial population dynamics a PCA ordination diagram is presented. The PCA ordination represented the correlation of all the three variables studied. The bi-plot for the axis (X and Y) was maximum in the rice fields of SW bank (91.21%) followed by N bank (89.12%) and SE bank (89.76%) of the river Brahmaputra within Kamrup district. The PCA ordination of SW bank revealed that cyanobacterial number was highest in the months of September-October and was highly related to phosphorus, soil temperature and soil moisture (Fig 1.). Cyanobacterial number in rice fields situated in the North bank was closely related to pH and soil moisture and was highest in the month of July-August and September-October (Fig 2.). Cyanobacterial number was also highest in the months of July-August in rice fields situated in the SE bank and was closely related to pH, soil moisture, phosphorus, sodium and potassium (Fig 3.). So the PCA ordination of the three sites revealed that cyanobacterial number was recorded maximum during July-August (monsoon season) followed during the months September-October (post monsoon). While comparatively less number of cyanobacteria was recorded during March-June (pre monsoon) and November-February (winter season).

Pearson's correlation coefficient between soil physico-chemical parameters and cyanobacteria revealed significant positive as well as negative correlation. Significant positive correlation was observed between cyanobacterial population and soil physico-chemical properties *i.e.* soil temperature, soil moisture, pH and phosphorus in all the three sites. Significant positive correlation with potassium (SE bank), magnesium (SW bank) and sodium (SW and SE bank) was also seen. Study revealed a strong negative correlation of population number with nitrogen in all the sites. Conductivity (SW bank) and organic carbon (SW and SE bank) too showed significant negative correlation with the population number of N₂-fixing cyanobacteria (Table: 1).

Table 1: Pearson's correlation coefficient between population number of N₂-fixing cyanobacteria and soil physico-chemical in each site.

Sites/soil parametes	Ts	Ms	pH	Cs	N	P	K ⁺	Ca ⁺⁺	Mg ⁺⁺	Na ⁺	OC
Southwest (SW)	0.888**	0.924**	0.855*	-0.894*	-0.952**	0.922**	-0.069	0.698	-0.839*	0.965**	-0.905*
North (N)	0.872**	0.888*	0.935**	-0.673	-0.894*	0.859*	0.658	0.496	-0.686	0.455	-0.715
Southeast (SE)	0.707	0.960**	0.916	-0.483	-0.860*	0.984**	0.915*	0.893*	0.237	0.976	-0.831*

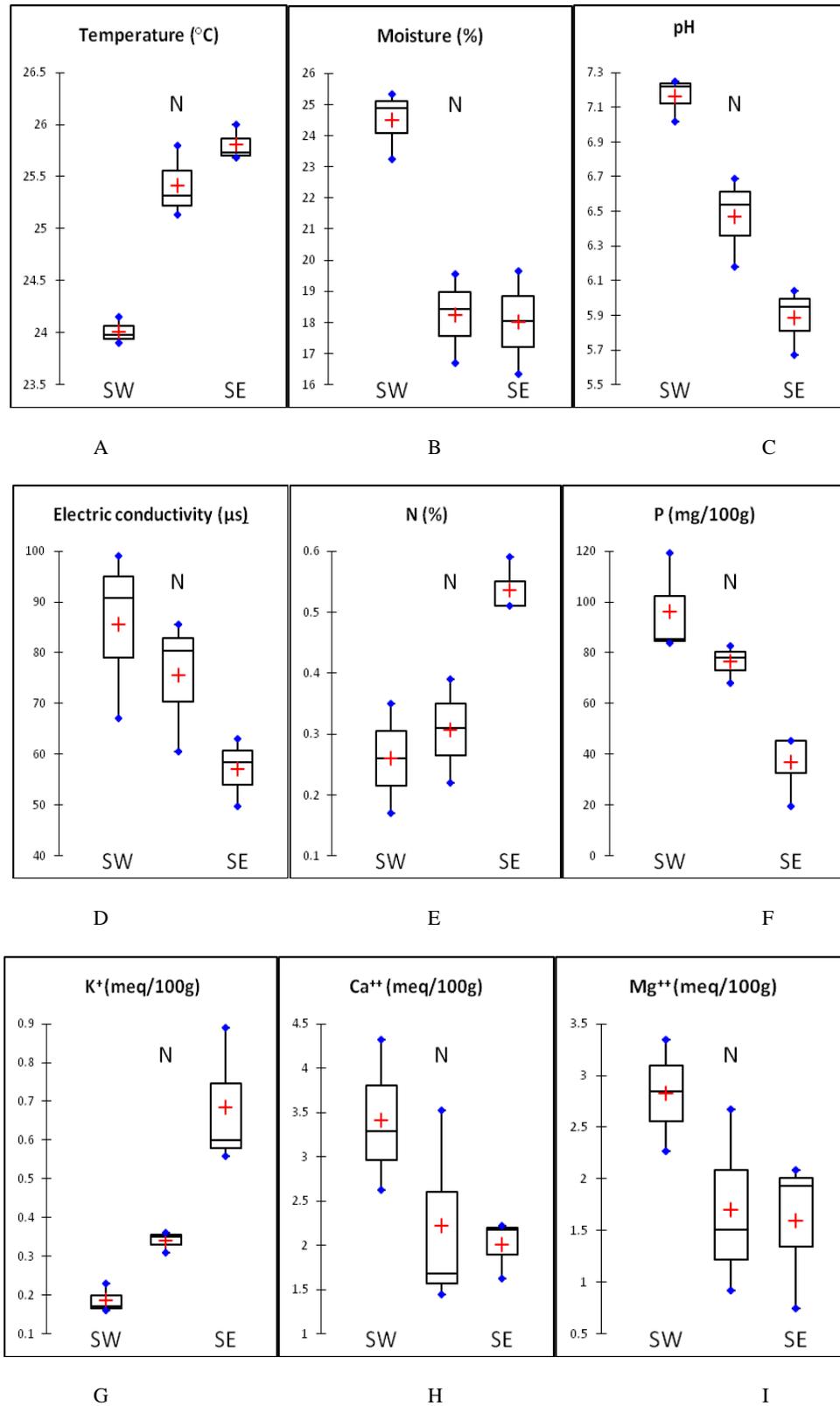
*and** Significant difference at P<0.05 and P<0.01 respectively. Ts = soil temperature, Ms = soil moisture, Cs = soil conductivity, N = nitrogen, P = phosphorus, K⁺ = exchangeable potassium, Ca⁺⁺ = exchangeable calcium, Mg⁺⁺ = magnesium, Na⁺ = exchangeable sodium, OC= organic carbon

Discussion

A rich diversity of 71 species of cyanobacteria under 20 genera and 9 families were recorded in alluvial rice grown soils of lower Brahmaputra floodplain. Among these filamentous heterocystous (80%) cyanobacteria showed clear dominance over the unicellular (14%) and filamentous non-heterocystous (6%) forms. Hazarika (2007) while studying the cyanobacteria of greater Guwahati (Kamrup) reported 20.83% of unicellular/colonial and 30.56% of filamentous non-heterocystous form which was outnumbered by filamentous heterocystous cyanobacteria (48.61%) in rice fields, which was in conformity with the present findings. In term of species richness the family Nostocaceae (54%) was found to be dominated in the rice fields situated in all three banks followed by Chroococcaceae (14%) and Rivulariaceae (13%). The family Scytonemataceae, Oscillatoriaceae and Stignemataceae comprise of only 7%, 6% and 3% of species respectively while Mastigocladaceae, Mastigocladopsidaceae and Microchaetaceae contributed with only 1%. The genera *Nostoc* and *Anabaena* are highly competitive and able to colonize whether as floating assemblages or as edaphic forms in rice fields soil (Prasanna and Nayak, 2007). In the present study also genus *Anabaena* (31%) showed the highest number of species, thereby indicating its dominance in rice growing fields of the district in terms of species richness. Genera *Nostoc* (17%) and *Calothrix* (8%) occupied the second and third position followed by unicellular cyanobacteria *Aphanocapsa* (6%). Similar observations were made in Tripura (Singh *et al.*, 1997a) and Nagaland (Singh *et al.* 1997b) respectively.

The principal component analysis (PCA) indicated that the cyanobacterial number during the months from July to October (monsoon and post monsoon season) was mainly influenced by soil temperature, soil moisture, phosphorus and pH. Positive significant correlation was also established in between cyanobacterial population number and these soil parameters (Table: 1). Soil moisture induced the cyanobacterial population in the monsoon season where soil contained a relatively higher amount of moisture compared to the months during November to February (winter season). Cold inhibited the growth of cyanobacteria as the optimum temperature range for luxuriant cyanobacterial growth in paddy fields of India is about 30-35⁰ C (Subrahmanyam *et al.*, 1965). Maximum cyanobacterial number occurred in the monsoon season when the temperature was highest that ranged from 29.2⁰C-30.14⁰C which was in conformity with the findings of Castenholz and Waterbury (1989). The average high temperature on the other hand increased the rate of decomposition of organic matter which results in a continuous availability of high nutrients supply in rice fields soil. The population number was observed to increase with the increase in pH values. Many evidences are present supporting pH in regulating the increasing number of cyanobacteria which was in conformity with our findings (Mitra, 1951). The amount of phosphorus nutrient which gradually increases during monsoon season and also showed significant positive correlation had a great impact on cyanobacterial population too. De and Sulaiman (1950) and Bisoyi and Singh (1988) and showed much earlier that a good correlation between the level of phosphorus applied and biomass of cyanobacteria. Cyanobacteria were also been reported to excrete organic acids that render phosphorus solubilisation thus contributing available phosphorus to the soil (Fuller and Rogers, 1952; Whitton *et al.*, 1991). Moreover sodium was found to be positively correlated which indicate that this nutrient also played a role in inducing the population number. This was in conformity with the earlier findings of Shubert and Starks (1980) and Issa *et al.* (2000). Positive correlation with potassium, calcium and magnesium has not been properly reported yet, though we found positive correlation with these soil parameters. However, according to Reynaud (1987) mineral nutrients (calcium, potassium, iron and phosphorus) increase with increased relative and absolute heterocystous biomass. Nitrogen, an important nutrient for plants showed significantly strong negative correlation with cyanobacterial population (Table: 1), thus indicating its establishment in particular soil where the concentration of combined nitrogen is low. This corroborates with the findings of Howarth *et al.*, (1988); Nayak and Prasanna (2007). Heavy rainfall during monsoon season might lead to highly mobile nitrate to leach out which results in overall nitrogen deficit. Negative correlation was also observed between cyanobacterial population and conductivity as well as organic carbon which collaborate with the findings of Reynaud (1987) and Dey *et al.*, (2010).

Later, during winter and pre-monsoon, cyanobacterial patches gradually disappear with the concomitant reduction of soil temperature, moisture, pH and with high accumulation of N and organic carbon due to the decomposition of cyanobacterial biomass. Minimum population number recorded in rice fields situated in the SE bank of the River Brahmaputra (Fig:4L) thus, could be attributed to the presence of high level of average nitrogen and low level of phosphorus and soil moisture (Fig:4E, F and B). Soil pH value towards acidic ranges (Fig:4C) too supports the poor growth of cyanobacterial population in these rice fields. (Nayak and Prasanna, 2007). Higher amount of phosphorus and soil moisture (Fig:4F and B) along with lower amount of nitrogen and alkaline pH (Fig:4E and 4C) could be responsible for maximum population number of cyanobacteria in the rice grown soils in SW bank of the River Brahmaputra.



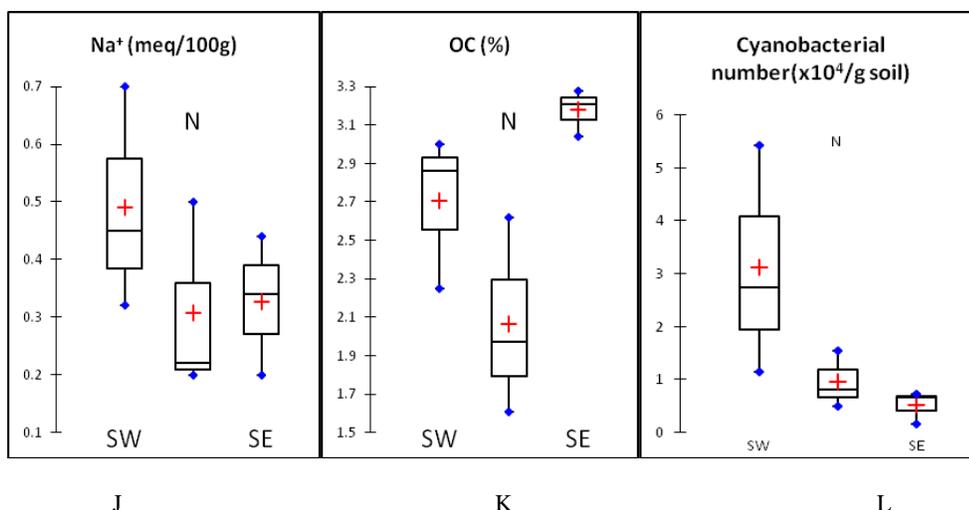


Fig: 4. Box plot graphics displaying the mean (+ sign), maximum (top dot) and minimum values (bottom dot) of the soil physico-chemical parameters (A-K) and cyanobacterial number (L) of each site. Horizontal lines in each box plot are, from top to bottom: third quartile, median and first quartile.

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