

## QUANTITATIVE ASSESSMENT OF CYANOBACTERIA AND THEIR RELATIONSHIP WITH SOIL FERTILITY IN SOME REPRESENTATIVE LOW LYING RICE FIELDS OF BANGLADESH

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Quantitative abundance of cyanobacteria in rice fields of eleven districts encompassing a wide range of soil fertility belonging to five agro ecological zones (AEZ) of Bangladesh showed a significant variation among the fields irrespective of districts in most of the cases. The range of cyanobacteria within four fields of each district varied from  $14.6 \times 10^4$  to  $18.6 \times 10^4$  and  $81.8 \times 10^4$  to  $141.0 \times 10^4$  g<sup>-1</sup> soil in Manikgonj and Brahmanbaria, respectively. Similarly, the enumerated quantity of cyanobacteria among the AEZ, the values ranged between  $24.5 \times 10^4$  to  $48.78 \times 10^4$  g<sup>-1</sup> soil in Ganges Tidal Floodplain and Meghna River Floodplain, respectively. Numerical variation assessed by correlation and regression analysis suggests that growth of cyanobacterial population was positively stimulated in soil having higher pH, available phosphorus and available sulphur while depressed by higher content of nitrogen and organic matter in the rice fields. However, multiple correlation coefficient revealed that variation of cyanobacteria in rice field ecosystem is due to joint contribution of all the fertility variables.

**Keywords:** Cyanobacteria; Low lying rice fields; Quantitative assessment; Soil fertility.

### Introduction

The modern methods of biotechnology may improve cyanobacterial biofertilizer technology, provided the relationship of cyanobacteria with soil is well understood through coordinated research in the laboratory and field. It is an established fact that cyanobacteria play an important role in improving the soil physical property particularly the soil structure and fertility of the rice field soils<sup>1</sup>. Moreover, cyanobacteria not only enrich the soil with nitrogen but also secrete some growth promoting substances<sup>2</sup> which, in addition, promotes the physico-chemical and nutritional status creating an over all congenial ecological environment of the rice growing soils. It has been, undoubtedly, recognized that amount of nitrogen fixed by cyanobacteria is a function of efficacy of individual cyanobacterial species and environmental conditions prevailing in the rice fields. Begum and Mandal<sup>3</sup> and Mandal *et al.*<sup>4</sup> reported that N-fixing capacity of cyanobacteria bears a significant relationship with soil properties. Thus, an attempt, has been made to assess the relationship between cyanobacteria and some soil fertility variables of some representative soils collected from rice fields encompassing different agro ecological zones of Bangladesh.

### Material and Methods

Soil samples were collected from some representative districts of Bangladesh namely, Brahmanbaria, Comilla, Dhaka, Faridpur, Gazipur, Khulna, Kishoregonj, Manikgonj, Munshigonj, Narayangonj and Narshingdi. During sampling, the fields were covered with rice mostly in tillering stage and in a condition of fifty percent to maximum water holding capacity. Four fields were selected in each district. Five subsamples were collected diagonally from each field depending on the nature of topography. Each sample was a composite sample of five subsamples. *Isolation, Identification and Enumeration of Cyanobacteria*- Soil samples were collected from the rice fields by scrapping the surface (0-0.5 cm) carefully to avoid any damage of algal material for quantitative assessment of cyanobacteria. In the laboratory, the soil samples were kept in a cool atmosphere. Isolation and enumeration of cyanobacteria were done by growing the cyanobacteria in nitrogen free Fogg's medium. Identification of cyanobacterial taxa was achieved by using standard texts following their growth both in the conical flasks containing cultural solution and agar plates. MPN technique was employed for quantification of cyanobacteria.

**Table 1.** Certain chemical properties and number of cyanobacteria ( $\times 10^4$  g<sup>-1</sup> soil) in the soil samples examined.

Name of Districts	pH	Organic C	Total N	Available		No. Cyanobacteria	LSD P=0.05
				P	S		
		Percent		$\mu\text{g g}^{-1}$ soil			
Dhaka (Demra)	5.2-6.9	1.31-1.79	0.18-0.24	2.05-2.08	11.50-21.40	14.6-21.2	1.81
Faridpur (Goalchamot)	7.4-7.6	0.51-0.96	0.08-0.15	2.40-4.50	35.20-46.80	38.1-43.5	NS
Gazipur (Tangi)	6.5-7.2	0.42-0.75	0.03-0.05	4.68-16.00	30.00-40.60	48.3-81.8	4.12
Kishoregonj (Bhairab)	5.3-6.5	0.26-1.56	0.04-0.22	2.20-4.20	7.00-21.88	16.6-21.7	0.82
Manikgonj (Singair)	5.5-5.9	0.76-0.83	0.12-0.16	2.00-4.50	25.50-37.50	14.6-18.6	1.55
Munshigonj (Muktarpur)	5.4-5.9	1.21-1.60	0.16-0.23	7.00-17.31	10.00-15.60	38.8-47.4	1.22
Narayangonj (Sadar)	5.9-7.0	0.96-2.44	0.11-0.22	2.20-5.41	18.10-65.60	21.0-42.5	NS
Narshingdi (Gouripur)	7.1-7.3	0.72-0.88	0.05-0.07	6.68-14.68	12.00-13.75	47.4-70.0	3.45
Brahmanbaria (Nabinagar)	6.8-7.5	0.44-1.11	0.05-0.18	12.42-28.50	19.30-62.50	81.8-141.0	5.65
Comilla (Muradnagar)	5.5-6.1	0.33-0.62	0.03-0.08	2.00-2.06	9.00-12.13	12.8-21.2	NS
Khulna (Teligati)	6.5-7.5	0.95-1.50	0.10-0.21	4.00-5.00	21.80-46.80	16.6-31.6	2.85

**Table 2.** Soil fertility variables and cyanobacterial population (on the basis of Agro Ecological Zones).

Agro Ecological Zones	pH	Organic C	Total N	Available Nutrients		Cyanobacteria ( $10^4$ g <sup>-1</sup> soil)
				P	S	
		Percent		$\mu\text{g g}^{-1}$ soil		
Madhupur Tract (Dhaka, Gazipur)	6.33	1.09	0.12	7.08	26.16	41.53
Ganges River Floodplain (Faridpur)	7.50	0.71	0.11	3.35	41.06	41.42
Brahmaputra Floodplain (Kishoregong, Manikgonj, Narshingdi)	6.17	0.91	0.12	5.49	18.47	32.11
Meghna River Floodplain (Munshingonj, Naryangonj, Brahmanbaria, Comilla)	6.16	1.04	0.12	10.11	21.61	48.78
Ganges Tidal Floodplain ( Khulna)	7.10	1.23	0.14	4.32	32.27	24.05

**Chemical Analyses** - For chemical analysis, soil samples were collected from a depth of 0-15 cm and air-dried in the laboratory, ground and passed through 100 mesh sieve and preserved in polyethylene bags. Determinations were made of pH by electrochemical method, organic carbon by wet oxidation method<sup>5</sup>. Total N following conc. H<sub>2</sub>SO<sub>4</sub> digestion and available N extracted by 2MKCl solution<sup>6</sup> were estimated by Kjeldhal distillation. Content of 0.5M acetic acid extractable P<sup>7</sup> was measured spectrophotometrically by developing phosphomolybdic acid blue complex and that of 500 ppmP<sup>8,9</sup> extractable S by forming turbidity complex with BaCl<sub>2</sub>, respectively.

### Results and Discussion

Results outlined in Table 1 reflect that the soils collected from rice fields of different districts varied in pH ranging from 5.2 to 7.6 and poor in organic carbon and total nitrogen (0.26 to 2.44 %, 0.03 to 0.24%) content. However, the soils were assessed to be low to moderate in fertility status with respect to available nutrient content.

**Abundance of cyanobacteria in different districts**- Results

showed a significant variation in cyanobacterial population among the 4 fields of each district. Of the 11 districts, the soils of Brahmanbaria district showed significantly higher range of cyanobacterial population. The maximum value i.e.  $141.0 \times 10^4$  was enumerated in field IV and that of minimum was found to be  $81.8 \times 10^4$  in field III with an average of  $109.62 \times 10^4$  g<sup>-1</sup> soil. This average value is about 7 times higher than the lowest average value recorded in the soil samples of Comilla ( $15.82 \times 10^4$  g<sup>-1</sup> soil) and Manikgonj ( $16.12 \times 10^4$  g<sup>-1</sup> soil) districts. The second highest range of cyanobacteria was estimated in Gazipur where the value was between  $48.3 \times 10^4$  in field I and  $81.8 \times 10^4$  in field IV with an average of  $64.72 \times 10^4$  g<sup>-1</sup> soil.

Number of cyanobacteria ranged from  $12.8 \times 10^4$  in field IV to  $21.2 \times 10^4$  in field I of Comilla district with an average value of  $15.82 \times 10^4$  g<sup>-1</sup> soil. Comparatively, lower range of cyanobacteria was recorded varying from  $21.2 \times 10^4$  in field IV to  $14.6 \times 10^4$  in field I with an overall mean of  $18.35 \times 10^4$  in Dhaka district. However,

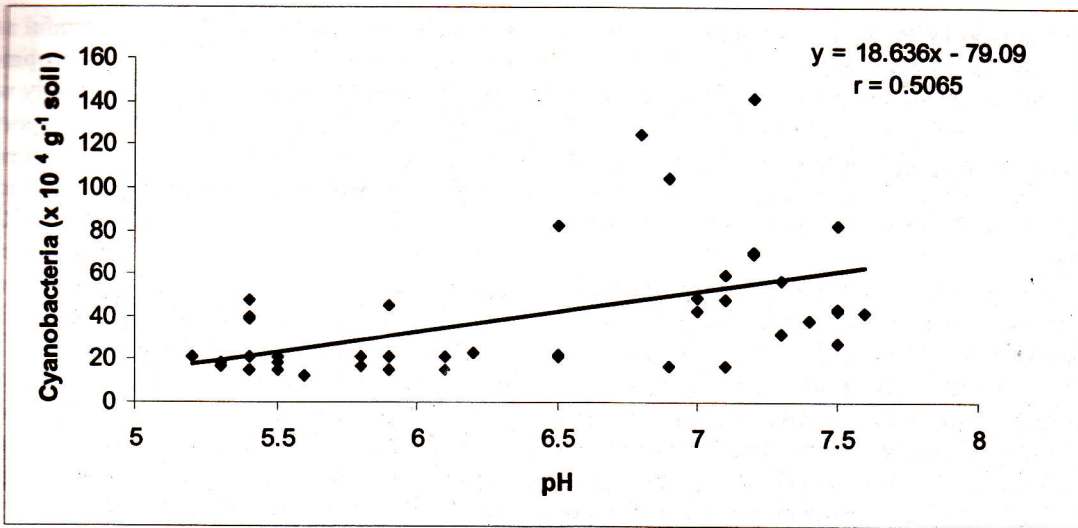


Fig.1. Scattered diagram between cyanobacterial population and soil pH.

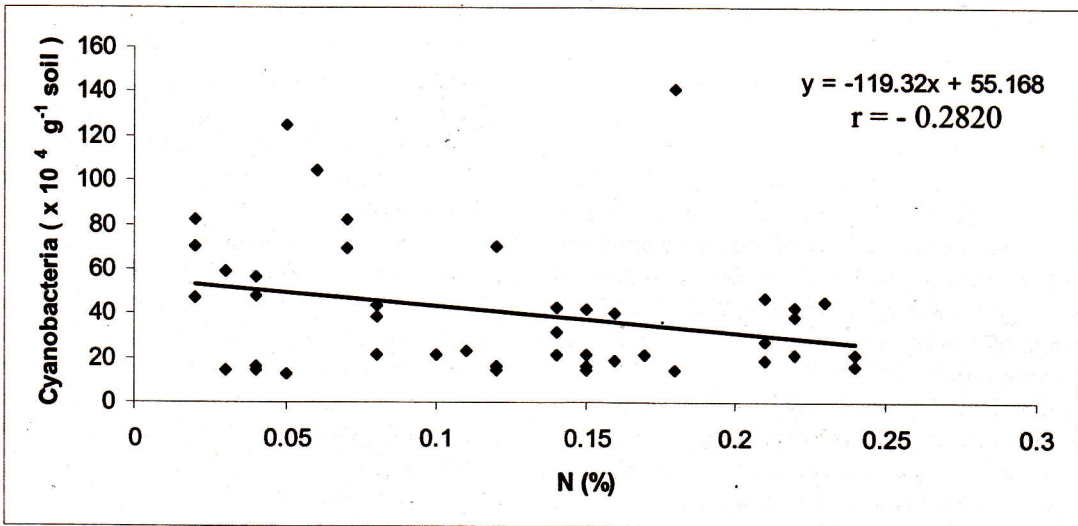


Fig.2. Scattered diagram between cyanobacterial population and total nitrogen of soil.

the variation in number of the same was very narrow in the soil of Faridpur district, i.e. from  $38.1 \times 10^4$  in field III to  $43.5 \times 10^4$  in field IV with a mean of  $41.42 \times 10^4 \text{ g}^{-1} \text{ soil}$ .

The prevalence of cyanobacterial population in the rice fields of Khulna district, accounted the maximum value of  $31.6 \times 10^4$  in field I and that of minimum  $16.6 \times 10^4$  in field IV with an overall mean of  $24.05 \times 10^4 \text{ g}^{-1} \text{ soil}$ . In Kishoregonj district, the highest and the lowest numbers were accounted to be  $21.7 \times 10^4$  in field IV and  $16.6 \times 10^4$  in field I, respectively, with a mean of  $19.5 \times 10^4 \text{ g}^{-1} \text{ soil}$ . The range of cyanobacterial population measured was comparatively in the lower level, i.e.

between  $14.6 \times 10^4$  in field I and  $18.6 \times 10^4$  in field II with an average of  $16.12 \times 10^4 \text{ g}^{-1} \text{ soil}$  in Manikgonj district. In Munshigonj, the maximum number of cyanobacteria was estimated to be  $47.4 \times 10^4$  in field II and that of the second highest was observed to be  $45.0 \times 10^4$  in field IV while the lowest number was being  $38.8 \times 10^4$  in field III with an overall mean value of  $42.77 \times 10^4 \text{ g}^{-1} \text{ soil}$ . The values reported from Narayanganj district, was enumerated to be  $42.5 \times 10^4$  in field I which was nearly double to the values recorded in fields II and III i.e.  $21.2 \times 10^4$  and  $21.0 \times 10^4 \text{ g}^{-1} \text{ soil}$ , respectively, with an average value of  $26.92 \times 10^4 \text{ g}^{-1} \text{ soil}$ . Relatively higher values were found in Narshingdi district covering from

$70.0 \times 10^4$  in field IV to  $47.4 \times 10^4$  in field I with an average of  $60.72 \times 10^4$  g<sup>-1</sup> soil.

The order of sequence of abundance in cyanobacterial population among the rice fields of 11 districts may be arranged as follows: Brahmanbaria > Gazipur > Narshingdi > Munshiganj > Faridpur > Narayangonj > Khulna > Kishoregonj > Dhaka > Manikgonj > Comilla.

*Abundance of cyanobacteria in different Agro Ecological Zones* - The soil samples collected from 11 districts of Bangladesh were partitioned to five agro ecological zones (Table 2). The results ascertained that abundance of cyanobacterial population in rice field soils of various agro ecological zones showed a relative variation ranging from  $32.11 \times 10^4$  to  $48.78 \times 10^4$  g<sup>-1</sup> soil (Table 2). The highest number of cyanobacterial population was found to be enumerated in Meghna River Floodplain accounting  $48.78 \times 10^4$  g<sup>-1</sup> soil. Contrary to this, the lowest number of the same organisms was recorded to be evident in Ganges Tidal Floodplain attaining upto  $24.05 \times 10^4$  g<sup>-1</sup> soil. Madhupur Tract and Ganges River Floodplain accounted almost identical number of cyanobacteria i.e  $41.53 \times 10^4$  and  $41.42 \times 10^4$  g<sup>-1</sup> soil, respectively. Brahmaputra Floodplain ranked next to these agro ecological zones recording about  $32.11 \times 10^4$  g<sup>-1</sup> soil of the organism. The variation in abundance of cyanobacterial population can be ranked as follows: Meghna River Floodplain ( $48.78 \times 10^4$  g<sup>-1</sup> soil) > Madhupur Tract (  $41.53 \times 10^4$  g<sup>-1</sup> soil) > Ganges River Floodplain (  $41.42 \times 10^4$  g<sup>-1</sup> soil) > Brahmaputra Floodplain ( $32.11 \times 10^4$  g<sup>-1</sup> soil) > Ganges Tidal Floodplain ( $24.05 \times 10^4$  g<sup>-1</sup> soil).

*Relationship of cyanobacterial population with soil fertility variables* - Assessment of the cyanobacterial population showed a significant variation over a wide range from rice field to rice field not only among the districts but also within the districts. Since, most of the members of cyanobacteria are soil borne microorganisms so their quantitative variation in soil is quite obvious because of variation in both macro and micro environmental conditions prevailing around them as soil is being a system of dynamic change.

In order to assess the relationship, simple and multiple correlation studies have been done to evaluate the contribution of soil fertility variables (pH, organic carbon, total nitrogen, available phosphorus and available sulphur) on the propagation of cyanobacterial flora in soil.

The positive value of co-efficient of correlation ( $r = 0.5065$ ,  $P = 0.001$ ) indicates that cyanobacterial population increases with the increase of pH or decrease in soil acidity and vice-versa (Fig. 1). This fact suggests

that higher concentration of H<sup>+</sup> ion is harmful and toxic for the normal growth and development of cyanobacteria. The findings agreed favourably well with the reports advanced by Pandey<sup>10</sup> who reported that propagation of nitrogen fixing blue-green algae (cyanobacteria) is favoured by higher pH values and under neutral conditions. Mandal *et al.*<sup>4</sup> and Roger and Reynaud<sup>11</sup> also observed that blue-green algae grow preferentially in environments that are neutral to alkaline in reaction. Holm-Hansen<sup>12</sup> proposed an optimum alkaline range of pH from 7.5 to 10.0 and that of acidic to neutral range of pH 6.5 to 7.0 for the growth and nitrogen fixation by cyanobacteria. In contrast, Garcia *et al.*<sup>13</sup> reported a lower range of pH from 4.0 to 6.8 and found positive relationship between pH of paddy soils and the number of species of nitrogen fixing cyanobacteria in the soil during dry season.

Co-efficient of single correlation further showed that cyanobacteria bear an inverse significant ( $r = -0.2820$ ,  $P = 0.05$ ) relationship with total nitrogen content of the soil (Fig. 2). The negative value of  $r$  explains the fact that growth of cyanobacteria is greatly favoured in soils deficient in nitrogen content. The inhibitory effect of applied nitrogen fertilizer on nitrogen fixing blue-green algae in rice field have also been previously demonstrated<sup>4,14-16</sup>.

As regard to organic carbon content of soil, an inverse medium relationship was observed with cyanobacterial population of the soil though not significantly ( $r = -0.0970$  NS, Fig. 3). This is quite obvious because organic matter contains a significant fraction of nitrogen in organic combination which possessed a significant indirect relationship with cyanobacterial population (Table 1, Fig. 3). Similar opinion was also proposed by Mandal *et al.*<sup>4</sup>.

Available phosphorus revealed a very strong positive relationship with cyanobacterial population (Fig. 4). The value of  $r$  (0.8414) is significant at 0.1% ( $P = 0.001$ ) level. The coefficient of correlation suggests that availability of phosphorus significantly promoted the growth of cyanobacteria in rice field ecosystems. The stimulative impact of phosphorus might be due to its association in the sensitive energy yielding reactions involved in the metabolic processes of the living systems. The favourable effect of phosphorus on the multiplication of cyanobacteria in the rice field have also been postulated by other investigators<sup>4,17-18</sup>. Earlier, Okuda and Yamaguchi<sup>19</sup> also noted that growth of blue-green algae in moist soil was closely related to the available phosphorus content of the soil and was poor at 0 to 5  $\mu\text{g g}^{-1}$  but vigorous above 6  $\mu\text{g g}^{-1}$  soil.

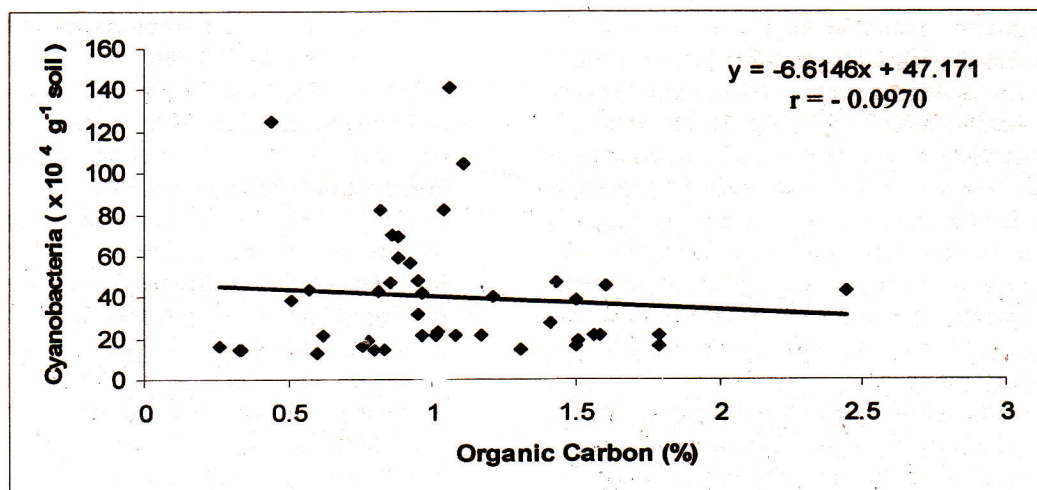


Fig.3. Scattered diagram between cyanobacterial population and soil organic carbon.

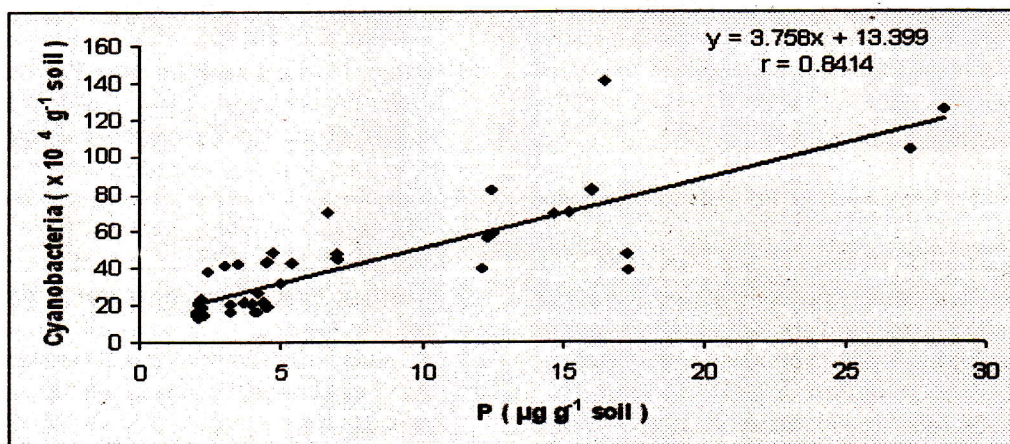


Fig. 4. Scattered diagram between cyanobacterial population and available P of soil.

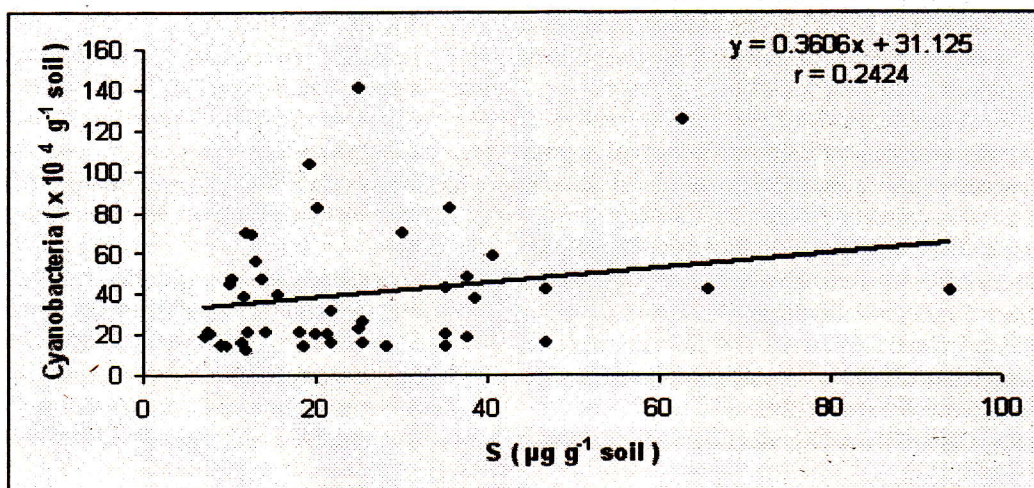


Fig.5. Scattered diagram between cyanobacterial population and available S of soil.

Similarly, available sulphur also showed its significant sensitive impact ( $r = 0.2424$ ) on the growth of cyanobacteria resulting a positive relationship between them at probability level  $P = 0.1$  (Fig. 5). The value of  $r$  indicates that with the increase in available sulphur, there will be a quantitative increase in cyanobacterial population in the rice field soils under investigation. Variation in monopoly of cyanobacterial population was again found to be relatively higher in agro ecological zones having ecosystem with almost neutral to alkaline reaction and high content of available phosphorus and available sulphur but low in available nitrogen.

Co-efficient of multiple correlation suggests that the growth of cyanobacteria is not controlled by one or the single factor of soil fertility. The result showed that joint contribution of the variables through interaction is significantly better to modify the quantitative promotion of the cyanobacterial population in the soils ( $R = 0.8789$ ,  $P = 0.001$ ). The reason might be due to the cumulative effect of all the variables interacted together. Some soil properties played positive role while others play negative role either significantly or non significantly.

#### References

- Fay P 1983, *The blue-greens*. Edward Arnold, London pp 88.
- Subrahmanyam R, Relwani LL and Manna GB 1965, Nitrogen enrichment of rice soils by blue-green algae and its effect on the yield of paddy *In: Symposium on land fertility improvement by blue-green algae Proc. Natl Acad. Sci. India Sect. A* 35(3) 382-386.
- Begum ZN Tahmida and Mandal R 1997, Nitrogen fixing capacity of some cyanobacterial strains from Bangladesh. *J. Phytol. Res.* 10(1-2) 67-73.
- Mandal R, Begum ZN Tahmida, Khan Zahed UM and Hossain MZ 1993,  $N_2$ -fixing blue-green algae in rice fields and their relationship with soil fertility. *Bangladesh J. Bot.* 22(1) 73-79.
- Walkley A and Black CA 1934, An examination of the Deglizeroff method for determining soil organic matter and a proposed modification of the chromic acid titration method. *Soil Sci.* 37 29-38.
- Bremner JM 1965, Inorganic forms of nitrogen. *In: Methods of Soil Analysis* (Black, C.A ed). Agron. No. 9. *Amer. Soc. Agron.* Madison, Wisconsin pp 1179.
- Williams EG and Stewart AB 1941, The colorimetric determination of readily soluble phosphorus in soils. *J. Soc. Chem. Indust.* 60 291.
- Fox RL, Olsom RA and Rhodes HF 1964, Evaluation of sulphur status of soils by plants and soil tests. *Soil Sci. Amer. Proc.* 28 243-246.
- Sakai H 1978, Some analytical results of sulfur deficient plants, soil and water. *Workshop on Sulphur Nutrition in Rice*. December, *BRRRI, Dhaka, Bangladesh*. Publication No. 41 35-59.
- Pandey DC 1965, A study of the algae from paddy soils of Ballia and Ghazipur districts of Uttar Pradesh, India. II(A): Taxonomic considerations - Cyanophyceae. *Nova Hedwigia Z. Kryptogamenk.* 10(1/2) 177-209.
- Roger PA and Reynaud PA 1978, La numeration des algues en soil submerge; ioi de distribution et problems dechantillonage (Enumeration of the algae in submerged soil: low of the distribution of organisms and the density of sampling). *Rev. Ecol. Biol. Sol.* 15(2) 219-234.
- Holm-Hansen O 1968, Ecology, physiology and biochemistry of blue-green algae. *Annu. Rev. Microbiol.* 22 47-70.
- Garcia JK, Raimbault M, Jacq V, Rinaudo G and Roger PA 1973, Microbial activities in Senegal rice soils in relation to soil properties and the rhizosphere. *Rev. Ecol. Biol. Sol.* 11(2) 169-185.
- Rinaudo G 1974, Fixation biologique de l'azote dans trios de sols de riziere de cote d'Ivoire (Biological nitrogen fixation in three types of rice soil in Ivory Coast) [in France, English summary]. *Rev. Ecol. Biol. Sol.* 11 149-168.
- Rinaudo G, Balandreau J and Bomnergues, Y. 1971, Algal and bacterial non-symbiotic nitrogen fixation in paddy soils. *Plant Soil Sci.* 47 471-479.
- Aiyer RS, Salahuddeen S and Venkaraman GS 1972, Long term algalization field trial with high yielding varieties of rice (*Oryza sativa* L.). *Indian J. Agric. Sci.* 42 380-383.
- Than Tun 1969, Effect of fertilizers on the blue-green algae of the soil of the paddy fields of Mandalay Agricultural Station. Union Burma. *J. Life Sci.* 2(3) 257-258.
- Srinivasan S 1978, Use of chemical fertilizers along with blue-green algae. *Aduthurai Rep.* 2(12) 145-146.
- Okuda A and Yamaguchi M 1952, Algae and atmospheric nitrogen fixation in paddy soils. II: Relation between the growth of blue-green algae and physical or chemical properties of soil and effect of soil treatments and inoculation on the nitrogen fixation. *Mem. Res. Inst. Food Sci.* 4 1-11.