DENSITY AND BIODIVERSITY OF BGA IN RICE FIELDS OF GOA.

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Abstract: Cyanobacteria forms a large group of structurally complex and ecologically significant gram negative prokaryotes which flourish in rice fields and also known to sustain the fertility of this ecosystem. This study is aimed to characterize the abundance of cyanobacteria in various habitats of rice field areas in Goa i.e. Khazan lands, Coastal areas, Hinterlands and Mining areas for khariff and Rabi seasons. A total of 16 genera and 90 species of heterocystous, non- heterocystous and unicellular BGA forms were recorded. The diversity of all the three types of algae was higher in the hinterlands as compared to the other habitats and also the diversity was more in Rabi season than khariff. The density of heterocystous forms was most abundant followed by non-heterocystous and unicellular forms respectively. The results were analysed statistically using standard statistical package.

INTRODUCTION:

Blue green algae possess an autotrophic mode of growth like eukaryotic plant cells, metabolic system like bacteria and occupy an unique position. They possess chlorophyll 'a' and are gram negative which carry out oxygenic photosynthesis. They exhibit a great morphological diversity and their broad spectrum of physiological properties reflects their widespread distribution and tolerance to environmental stress. (Tandeau de Marsac and Howard 1993). Interesting results are obtained from detailed studies carried out on the distribution and periodicity of BGA from various parts of India. (Venkataraman, 1975; Kolte and Goyal, 1985; Singh, 1985). Several reports have indicated a widespread distribution of forms like *Oscillatoria, Nostoc, Anabaena, Phormidium and Aphanothece* (Gupta, 1975; Sinha and Mukherjee, 1975; Paul and Santra, 1982). Singh (1950) and Talpasayi (1962) made a systematic enumeration of cyanobacteria collected from moist soils and rocks. Research has also shown the occurrence of mostly hrterocystous forms due to their competitive ability in comparison to non heterocystous forms (Garcia-Pichel and Belnap 1996). The dominating heterocystous nitrogen fixing blue green algal species of *Aluosira, Cylindrospermum, Nostoc, Anabaena, Tolypothrix* and *Calothrix* were reported from soils of Cuttack and Orissa (Singh, 1961). Distributional profiles of cyanobacterial isolates from soils of Bhubaneshwar, Cuttack and Howrah indicated the predominance of heterocystous strains. (Sudhir Saxena et al., 2007). Paddy field ecosystem provides a unique aquatic-terrestrial habitat for the favourable growth and nitrogen fixation by cyanobacteria meeting their requirements for light, water and higher temperature thus maintaining the stable yield of rice under flooded conditions and also the productivity of soils (Roger et al. 1993).

Goa is a coastal region with a hot, humid and tropical climate with about 54 thousand hectares of land under rice cultivation, which is the staple food of Goans (Sakshena, 2003).

Rice ***(Oryza sativa L)**,* the staple food of Goans is being cultivated over an area of 54,000 hectares both in Kharif (44,000 ha) and Rabi *(*10,000 ha)*.* This cereal crop accounts for 31% of the total cropped area and 86% of the food grain production. It is cultivated on three different land types viz. Kher lands (rainfed lowlands), Morod lands (rain fed uplands) and Khazan lands (coastal saline lands). Our investigation was directed towards evaluating the density and diversity of BGA in four different types of rice fields in Goa which are influenced by different environmental conditions.

MATERIALS AND METHODS:

The four different fields selected were

1. khazans lands: which are salty lands with regular influx of saline sea water and are used for rice cultivation by farmers.
2. Coastal fields: which are in the vicinity of beach area
3. Mining area fields: rice fields which are situated close to the mines and which receive runoff and rejects of mines.
4. Hinterlands: are remote interior areas where rice cultivation is carried out and is free of coastal influence.

The physicochemical characteristics of the sampling sites were analysed with respect to their EC and Ph range following the methodology outlined by Black (1992).

The investigation on the density and diversity of cyanobacteria from the four selected habitats of Goa was carried out for a period of three years from June 2006 to June2009.

Collection was done from the paddy fields of four habitats both in khariff and rabi seasons of rice cultivation. Collection was made in fixed spots marked for collections in a 1L capacity wide mouthed bottle. The phytoplanktons were immediately preserved in 1% lugols iodine (1ml/100ml). This sedimented the phytoplanktons. After all phytoplanktons are settled the supernatant is siphoned out and the remaining sample is concentrated by centrifugation in a centrifuge at 1500 r.p.m. The total concentrated volume made was 100ml. Initial study for taxonomic identification was carried out using light binocular microscope. The algae were then identified using keys given by Desikachary (1959), Anand (1989) and Santra(1993).

For quantitative analysis the sample was analyzed by lackey's drop method (1938) as mentioned in APHA (1985), The formula used was:-

Phytoplankton unit per liter= $\frac{n \times c}{V} \times 1000$

Where n= number of phytoplanktons counted in 0.1ml. (1 drop of concentrate)

c= total volume of concentrate in ml.

V=total volume of water filtered in liters.

A colony is considered as one individual. Filament more than ¾ as one individual.

Data on density is recorded in Table 1.

STATISTICAL ANALYSES:

The data collected for three years of study period was statistically analyzed using PAST statistical package. Shannon (H), Simpson (1-D) and Margalef diversity indices were analyzed for the three types of cyanobacteria namely Heterocystous, Non-heterocystous and Unicellular during khariff and Rabi seasons of paddy cultivation.

RESULTS AND DISCUSSION:

Rice fields are temporary wetland ecosystems, with variable biodiversity and cyanobacteria are known to be an integral component of waterlogged rice fields. The rice field ecosystems with its optimum levels of light, water, temperature, humidity and nutrient availability provide a favorable environment for the luxuriant growth of cyanobacteria. The favourable balance of soil nitrogen of rice fields wherein rice can be grown on the same land even without any addition of fertilizers and without any reduction in yield, confirms to the significance of cyanobacterial nitrogen fixation (Venkatraman, 1972; Nayak et al., 2001; Nayak et al., 2004; Song et al 2005). It is also a well known fact that besides contributing to soil nitrogen and improvement in yield of rice, cyanobacteria also produces agronomically significant changes in the physical, chemical and biological properties of soil and soil-water interface of rice fields (Mandal et al. 2001; Nayak et al 2004). This may account for the higher abundance of cyanobacteria in paddy fields than in cultivated soils as reported under widely different climatic conditions of India (Mitra, 1951). In tropical rice fields, biological nitrogen fixation is mainly contributed by indigenous population of cyanobacteria or application of biofertilizers (algalisation), which meets the nitrogen demand of the rice crop. Therefore efforts need to be focused towards enrichment of indigenous population of cyanobacteria, which are better adapted to the specific niche, through development of multiple inocula preparations on regional basis. The present study was undertaken to evaluate cyanobacterial diversity and density in diverse rice soil ecologies of Goa during the two growing seasons of khariff and Rabi. The physicochemical environment of the sampling sites show variations in recordings of EC and pH (Table 1). The slightly alkaline pH of 7.3 was recorded in the hinterland paddy fields of Quepem with an EC of 3.5d sm^{-1} whereas coastal region of Utorda recorded a pH of 6.8 with a moderate EC of 3.0 d sm^{-1}. The other two sampling sites recorded acidic Ph in the range of 5 to 6 with a high EC of 7.6 d sm^{-1}in Quelossim khazans and a low EC of 0.5 d sm^{-1}in Velguem mining area fields.

Density data shows the highest a density of 640cells/ml in khariff season of heteroystous forms in the hinterlands of Quepem in the year 2006-2007 followed by coastal region of Utorda which recorded 540cells/ml and the least were recorded in mining fields 320cells/ml followed by khazan lands with 500cells/ml. But however Rabi season recorded comparatively less than khariff in hinterlands (610 cells/ml) and khazan lands (470 cells/ml) and comparatively more in the remaining two habitats. Overall the data indicates the predominance of heterocystous forms in all the 4 habitats followed by non-heterocystous and unicellular forms (Table 2). Similar results were obtained in a study where 166 cyanobactarial isolates were purified which included maximum heterocystous genera followed by non heterocystous forms in diverse rice soil ecologies

(Prassanna and Saswati, 2007). Earlier studies on cyanobactarial diversity of rice fields at the Indian Agricultural Research Institute have shown the predominance of heterocystous forms irrespective of chemical/biofertilizers treatments and stage of crop growth (Nayak et al. 2001, 2004). In the present study, paddy fields of hinterlands are richest in cyanobacterial density followed by coastal fields and khazans and least in mining fields. Although cyanobacteria are ubiquitous in their distribution, it is well established that they prefer neutral to slightly alkaline pH. A study on biodiversity and seasonal variation of cyanobacterial strains in rice paddy fields in Fujian China showed that cyanobacterial diversity in deeper soil fractions was higher than the upper soil fractions, and in addition the highest diversity was found in the middle of growth season and the lowest after harvest (Song et al, 2005).

In the present investigation Shannon's diversity indices in all the habitats of all groups of BGA was in the range 1.5-1.6. The highest Shannon's diversity indice of 1.608 was recorded in khariff season during the study period 2008-2009 in coastal and khazan lands for heterocystous forms. Whereas lowest Shannon's diversity index of 1.587 was recorded in khariff season during the study period 2008-2009 in mining area rice fields for unicellular forms (Table 3).

Simpson diversity indices in all the habitats of all groups of BGA range from 0.79-0.80. The highest Simpson diversity index of 0.80 was recorded during the study period 2008-2009 in rabi season for non heterocystous in the Quepem hinterland, whereas lowest Simpsons diversity index of 0.790 was recorded in khariff season during the study period 2008-2009 in mining area rice fields for unicellular forms (Table 3). In a similar study Shannon's diversity index was indicative of extensive diversity of cyanobacteria within the rhizosphere of rice cultivars. Simpson's diversity index (1-D) which takes into accounts both richness and evenness also showed significant values (Prassanna et al. 2009).

The Margalef's diversity indices in all the habitats of all three groups of BGA were in the range of 0.9-1.2. The highest Margalef's diversity index of 1.294 was recorded during the khariff season of study period 2007-2008 and rabi season of 2008-2009 for unicellular forms in mining area rice fields, whereas lowest Margalef's diversity index of 0.9618 was recorded in khariff season during the study period 2006-2007 in hinterlands rice fields for heterocystous forms in khariff season (Table 3). The present study indicates a moderately rich but variable diversity of BGA in Goan rice fields. It is evident from the results that though the count of heterocystous BGA was the highest in all rice fields but the diversities of all groups are moderately high in all the four habitats. Thus the present investigation throws light on the density and diversities of BGA in Goan rice fields especially with regard to the indigenous species which could help in development of niche specific inocula for Goan rice fields.

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Table 1: Details of physicochemical characteristics of soil and water of the sampling area during study period of 2006-2009

Location	Soil		Water	
parameters	EC dsm^{-1} ± SD	pH ± SD	EC dsm^{-1} ±SD	pH ± SD
Quepem hinterlands	3.5 ± 0.1	7.3 ± 0.2	3.2 ± 0.1	7.0 ± 0.1
Utorda coastal	3.0 ± 0.2	6.8 ± 0.1	2.8 ± 0.2	6.9 ± 0.2
Quelossim khazans	7.68 ± 0.1	5.0 ± 0.1	7.5 ± 0.1	5.3 ± 0.1
Velguem mines	0.5 ± 0.2	5.2 ± 0.2	0.48 ± 0.2	4.9 ± 0.1

Table 2: Density of BGA at different habitats during the study period 2006-2009.

Group/place		2006-2007 seasons		2007-2008 seasons		2008-2009 seasons	
Quepem Hinterlands		**khariff**	**rabi**	**khariff**	**rabi**	**khariff**	**rabi**
Heterocystous	number/ml	640	610	620	600	510	560
Non-Heterocystous	number/ml	470	400	430	440	440	480
Unicellular	number/ml	480	500	510	480	440	440
Utorda-coastal							
Heterocystous	number/ml	540	560	530	550	600	550
Non-Heterocystous	number/ml	540	540	530	500	530	540
Unicellular	number/ml	440	470	460	500	530	540
Quelossim-Khazans							
Heterocystous	number/ml	500	470	450	460	420	390
Non-Heterocystous	number/ml	430	450	440	440	460	470
Unicellular	number/ml	350	380	360	390	340	400
Velguem mines							
Heterocystous	number/ml	320	340	340	320	350	310
Non-Heterocystous	number/ml	310	290	290	310	340	330
Unicellular	number/ml	260	270	220	290	250	220

Table 3: Comparative diversities of BGA of different habitats.

Year of study	2006-2007						2007-2008						2008-2009					
Season	Khariff			Rabi			Khariff			Rabi			Khariff			Rabi		
Diversity indices	Shannon	Simpson	Margalef	Shannon	Simpson	Margalef	Shannon	Simpson	Margalef	Shannon	Simpson	Margalef	Shannon	Simpson	Margalef	Shannon	Simpson	Margalef
Hetererocystous																		
Hinterlands	1.597	0.7949	0.9618	1.608	0.7992	0.973	1.606	0.799	0.9692	1.602	0.7972	0.977	1.601	0.797	1.017	1.604	0.798	0.9937
coastal	1.605	0.7984	1.003	1.601	0.7966	0.9937	1.607	0.799	1.007	1.596	0.7947	0.9982	1.608	0.799	0.977	1.593	0.793	0.9982
khazans	1.604	0.7976	1.022	1.608	0.7995	1.039	1.607	0.799	1.051	1.606	0.7987	1.045	1.608	0.799	1.07	1.605	0.798	1.092
mining	1.607	0.7988	1.154	1.603	0.7976	1.134	1.603	0.798	1.134	1.607	0.7988	1.154	1.601	0.797	1.125	1.602	0.797	1.165
Non-heterocystous																		
Hinterlands	1.606	0.7986	1.039	1.6	0.7962	1.084	1.605	0.798	1.063	1.603	0.7975	1.057	1.606	0.799	1.057	1.608	0.8	1.033
coastal	1.605	0.7984	1.003	1.598	0.7956	1.003	1.605	0.798	1.007	1.607	0.7992	1.022	1.603	0.797	1.007	1.6	0.796	1.003
khazans	1.593	0.7939	1.063	1.602	0.797	1.051	1.606	0.799	1.057	1.6	0.7965	1.057	1.606	0.799	1.045	1.604	0.798	1.039
mining	1.602	0.7971	1.165	1.601	0.7967	1.188	1.601	0.797	1.188	1.597	0.795	1.165	1.608	0.799	1.134	1.607	0.799	1.144
Unicellular																		
Hinterlands	1.606	0.7986	1.033	1.604	0.7976	1.022	1.603	0.797	1.017	1.608	0.7995	1.033	1.606	0.799	1.057	1.606	0.799	1.057
coastal	1.606	0.7986	1.057	1.606	0.7986	1.039	1.606	0.799	1.045	1.607	0.7992	1.022	1.603	0.797	1.007	1.6	0.796	1.003
khazans	1.601	0.7967	1.125	1.607	0.7992	1.1	1.604	0.798	1.116	1.605	0.7982	1.092	1.603	0.798	1.134	1.606	0.799	1.084
mining	1.599	0.7959	1.228	1.592	0.7929	1.214	1.603	0.798	1.294	1.601	0.7967	1.188	1.587	0.79	1.243	1.603	0.798	1.294