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Cyanobacterial Occurrence and Diversity in Seagrass Meadows in Coastal Tanzania

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Abstract: We report on the occurrence and diversity of cyanobacteria in intertidal seagrass meadows at Ocean Road and Mjimwema, Dar es Salaam, Tanzania. Nutrients, temperature and salinity were measured as comparative environmental factors. A total of 19 different cyanobacteria taxa were encountered, out of which eight were found exclusively in Mjimwema, four exclusively in Ocean Road and seven were common to both sites. *Oscillatoria, Lyngbya* and *Spirulina* were the dominant cyanobacterial genera. Cyanobacterial coverage was higher in Mjimwema (31–100%) than in Ocean Road (0–60%). The levels of nutrients in tidal pool waters at Ocean Road ranged from 0.45–1.03 µmol NO_3^-N/l, 0.19–0.27 µmol NO_2^-N/l and 0.03–0.09 µmol PO_4^-P/l. At Mjimwema the nutrient concentration ranges were 0.14–0.93 µmol NO_3^-N/l, 0.20–0.30 µmol NO_2^-N/l and 0.01-0.07 µmol PO_4^-P/l . The nutrient levels were significantly higher at Ocean Road than at Mjimwema ($P = 0.001$ for nitrate and $P = 0.025$ for phosphate). There was no significant difference in nitrite levels between the study sites ($P = 0.83$). The low cyanobacterial diversity and coverage in Ocean Road is related to the high levels of nutrients and physical disturbance from sewage discharge and the harbour in the area.

INTRODUCTION

Globally, seagrasses, together with mangroves and coral reefs, are amongst the most productive systems in the marine environment (Capone & Taylor, 1980).

Seagrasses are found in the subtidal zone of many coastal waters and support a rich diversity of organisms, ranging from epiphytic algae to a variety of fish and other fauna (Tomasko & Lapointe, 1991; Welsh et al., 2000). Seagrasses provide breeding, nursery and feeding grounds for these organisms (Short & Wyllie-Echeverria, 1996). In addition, they trap suspended sediments and nutrients entering the coast, thus protecting coral reefs. They also buffer wave action, which allows development of mangroves that protect beaches from erosion (UNEP, 1989).

The high productivity in seagrass ecosystems is contributed not only by the seagrass component but also by a variety of epiphytic organisms (Hemminga & Duarte, 2000) that live there. Epiphytes may provide as much as 46% of the total autotrophic production in seagrass meadows (Pinckney & Micheli, 1998). Epiphytic microalgae are known to support much herbivory in the seagrass beds and the abundance of epiphytes also influences meiofaunal abundance (Pinckney & Micheli, 1998).

Seagrasses are not known to produce any toxic substances, and lack physical mechanisms for controlling the development of epiphytes, hence epiphytes may be found on all parts of the seagrasses and also on the adjacent sediments (Borowitzka & Lethbridge, 1989). The distribution of epiphytes on seagrasses beds depends mainly

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on the morphology and life span of the portion colonized by epiphytes (Semesi, 1988; Uku, 1996). Cyanobacteria have an ability of forming macroscopic, coloured, vertical, stratified microbial communities known as microbial mats, which may vary from a mucilaginous coating on sand, mud or organic debris to well-developed, thick 'carpets' (Paerl et al., 1993). The microbial mats are usually dominated by filamentous *Oscillatoria* spp., *Spirulina* spp. and *Microcoleus* spp. (Bergman et al., 1997; Stal, 1984).

Cyanobacteria may play a double role related to productivity, first by fixing CO_2 and second by fixing atmospheric nitrogen, which often limit primary production in many ecosystems. It has been proposed that cyanobacteria are important food sources for heterotrophs and that the inorganic nitrogen released, through breakdown of cyanobacteria by heterotrophs, supports continued production in seagrasses (Yamamuro, 1999).

Diversity and abundance of cyanobacteria may be controlled by various factors, such as competition, grazing pressure and levels of nitrogenous nutrients. Uku & Björk (2001) observed higher numbers of epiphytic cyanobacteria in seagrass beds with lower nutrient levels than in beds with high nutrient loading along the Kenyan Coast. However, there are a few studies on epiphytes on plants in Tanzanian coastal waters (Lugendo et al., 1999; Semesi, 1988; Leliaert et al., 2001) and the emphasis has been on macroalgae. The only study on epiphytic cyanobacteria in seagrass meadows along the Tanzania coast is that of Lugomela (2002), carried out at the Zanzibar Island and investigating the cyanobacterial diversity and nitrogen fixation.

The aim of our research was to provide basic information on the occurrence and species diversity of cyanobacteria in seagrass meadows subjected to different human impacts in the western Indian Ocean coast.

MATERIALS AND METHODS

Study area

Two stations, Ocean Road and Mjimwema, were chosen in Dar es Salaam, Tanzania. Ocean Road receives sewage from the city centre through pipes and the Msimbazi River, and also faces heavy sedimentation due to harbour activities. In contrast, Mjimwema beach lacks major inflows and is also protected from wave action by Kendwa, Makatumbe and Sinda Islands.

Ocean Road is located at 06° 48.3' S, 39° 18' E, just outside the entrance of the Dar es Salaam harbour on the northern side. This beach is characterized by sandy/muddy habitats and seagrasses occur as small patches from the lower littoral zone and extend towards the sublittoral zone on a harder substrate of calcareous gravel. Mjimwema beach is located at 06° 50' S, 39° 21' E 4 km south of the Dar es Salaam harbour. It is characterised by a short, steep sandy slope, below which are sandy/muddy intertidal flats with seagrass beds extending to the Kendwa Islands.

Cyanobacteria identification and coverage

Triplicate samples of seagrasses and sediments were taken from intertidal seagrass meadows at monthly intervals between September 2002 and August 2003. Four seagrass genera were sampled, namely *Thalassodendron, Thalassia, Cymodocea* and *Halodule*. Sediments and any associated mats were also collected and analysed. If not immediately examined samples were preserved in a 4 % formaldehyde–seawater solution. To remove epiphytes, seagrass surfaces were scraped in filtered seawater and debris removed under a dissecting microscope. Samples were then examined microcopically as wet mount slides using an Olympus BHT microscope fitted with a BX 50 camera. The works of Desikachary (1959); Lyimo (1995); Silva & Pienaar (2000) and Lugomela (2002) were used to identify the cyanobacteria. Chlorophyll *a* was measured as described by Parsons et al. (1989). All microalgal cells were counted and the percentage of the cyanobacterial cells was calculated as:

$$\frac{\text{Total cyanobacteria cells}}{\text{Total microalgae cells}} \times 100.$$

Physico-chemical parameters

Triplicate measurements of seawater temperature and salinity were made using a mercury thermometer and a salt refractometer 300011 (SPER SCIENTIFIC, China) respectively. Nitrates, nitrites and phosphates were measured from tidal pool and pore waters. Tidal pool water samples were collected and placed in 250-ml plastic bottles and then filtered using GF/C microfibre filter papers. Pore water samples were extracted from the sediments collected within 5 cm depth using a core of 6 cm diameter. The extraction was done in the laboratory using a suction pump equipped with Whatman filter papers GF/A. Both pore water and tidal pool water were analyzed as described by Parsons et al. (1989).

Data analysis

Data were analysed using an unpaired *t*–test. For cyanobacterial species diversity, the Shannon–Weaver diversity index (H') was calculated. All the analyses were done as described by Zar (1999) and Graph Pad Instant t_m 1990 – 1993 software. P-values less than 0.05 ($P < 0.05$) were considered to be significanty different.

RESULTS

Cyanobacterial diversity and coverage

The highest diversity and percentage cover was encountered in microbial mats found on both seagrasses and sediments. The encountered cyanobacteria were described based on their morphology (Table 1). Nineteen cyanobacteria taxa belonging to 11 genera were recorded; among them three were heterocystous. Eight were found exclusively in the seagrass meadows of Mjimwema, four in Ocean Road and seven occurred at both sites (Table 2). The commonest cyanobacteria in the seagrass meadows belonged to the genera *Oscillatoria, Lyngbya* and *Spirulina*. The members of the genus *Oscillatoria* occurred at Mjimwema seagrass beds throughout the year and seemed to be the most dominant with the highest diversity, followed by the genus *Lyngbya*. At Ocean Road, the dominant cyanobacterial genera were *Oscillatoria* and *Spirulina* while other cyanobacteria occurred occasionally. Generally, the cyanobacterial diversity was higher ($P = 0.02$) in Mjimwema, with a Shannon–Weaver diversity index H' ranging from 0.28–0.98, while at Ocean Road H' was between 0.0 and 0.45 (Fig. 1a).

Mjimwema seagrass meadows had significantly higher ($P = 0.03$)cyanobacteria coverage (31–100%) than those of Ocean Road (0–60%), throughout the study period (Fig. 1b). However, Ocean Road had a higher diatom and other microalgal than cyanobacteria coverage. The microalgal biomass based on chlorophyll *a* measurements was higher at Ocean Road than at Mjimwema. The mean values ranged from 17.6–99 and 29–166.7 μg chlorophyll a/m^2 for Mjimwema and Ocean Road, respectively.

Seasonal cyanobacterial variation was also noted during the study (Fig. 1). At Mjimwema the cyanobacterial percentage coverage was significantly higher during the SE monsoon (June–October) than during the NE monsoon (December-April), ($P = 0.02$). At Ocean Road, there was no significant difference in cyanobacterial diversity and percentage coverage between the two monsoons. During the period with higher cyanobacterial diversity and percentage coverage, prominent thick cyanobacteria mats with green, brown and red pigment covered the Mjimwema intertidal seagrass bed. The mats colonised both sediments and the seagrasses (Plate A & B).

The sediments were dominated by one or more *Oscillatoria* spp., the most common being *Oscillatoria limosa* (Plate C); Oscillatoria sp. 6 (Plate D) was rarely found. The seagrasses were dominated by two *Lyngbya* spp. (E and F). However, at Ocean Road only patches of similar cyanobacterial mats were observed. The pigmentation of the cyanobacterial mats is presumably in response to changes in environmental conditions (Desikachary, 1959).

The highest cyanobacterial diversity (H' = 5.01) occurred in sediments, and the lowest (H' = 0.73) was found on *Thalassodendron* (Table 2). H' values showed that cyanobacterial diversity was significantly higher in sediments within the seagrass meadows than on seagrasses ($t = 3.8$, $P = 0.02$). It was observed that cyanobacteria, especially *Oscillatoria* spp. and *Lyngbya* spp.,

Table 1. Characterisation of various cyanobacteria encountered on seagrasses beds off the Dar es salaam Coast, based on their morphology and mat appearance. ND = not determined, Ns = mixed sample, Hc = heterocyst, Nhc = non heterocyst cells, and Tc = terminal cell, Mc = Middle of the filament Se = in sediment, Ep = epiphytic on seagrass. Letters in parentheses represent micrograph plate label.

Species	Size (μm)		Description	
	Cell width	Cell length	Habitat and mats	Microscopy
NON-HETEROCYSTOUS				
Oscillatoria limosa	19.2–21.05	3.0 – 3.94	Thick, Golden, pale/dark red; Se & Ep	Tc round **(A)**
Oscillatoria sp. 2	13.0–13.2	2.85 – 2.92	-do- constricted walls	Tc round with
O. proboscidea	Mc 20.0–21.0 Tc 7.0 – 7.89	ND	-do-	Tc pointed, constricted
Oscillatoria sp. 4	ND	ND	-do-	Tc flat
Oscillatoria sp. 5	7.9 – 8.0	ND	-do-	Tc curved at one end.
Oscillatoria sp. 6	Mc 18.4–19.0 Tc 4.8 – 5.2	2.6	-do-	Tc curved both ends **(B)**
Lyngbya sp. 1	40.0–44.7	1.15 – 1.37	Dark green-black filamentous, Ep	Green filament, Tc flat **(C)**
Lyngbya sp. 2	26.1–26.3	ND	Dark brown filamentous Se & Ep	Colourless filament, Tc round
L. confervoides	Mc ~ 56 Tc ~ 15		-do-	Tc pointed, constricted **(D)**
Spirulina gigantea	~13.0	ND	Green/brown mats; Se & Ep	Spiral not close to each other ~13.1 apart
S. labyrinthiformis	~7.9	ND	-do-	Spiral very close to each other ~ 6.5
Microcoleus chthonoplastes	15.5 – 50.0 (bundle size)	ND	Olive green thin mats; Se	Bundle of trichomes in sheath
Synechococcus sp 1	ND	ND	Ns; Se	Spherical cell jointed to form chain
HETEROCYSTOUS				
Calothrix contarenii	ND	ND	Green filaments; Se	The Tc is a heterocyst
Anabaena sp. 1	ND	ND	Slimy green mats; Se	Heterocyst in centre large & ovoid
Nodularia sp. 1	Nh_c 10.5 Hc 13.5	Nh_c 7.8 Hc 10.5	Ns; Se	Heterocyst in centre, large & spherical
UNICELLULAR				
Chroococcus minutus	ND	–	Ns; Se	Ovoid cells
Merismopedia elegans	ND	–	Ns; Se	Two spherical cells occurring together
Aphanocapsa sp. 1	ND	ND	Ns; Se	Group of spherical cells

sometimes occurred with diatoms, mostly *Climacosphenia moniligera* (Plate H), which were firmly attached to the cyanobacterial filaments. The association may be parasitic, as the core of some of the cells of cyanobacteria seemed empty when attached to these diatoms.

Table 2: The occurrence of various cyanobacteria species as epiphytes to the selected seagrass species, or attached on sediments of the Mjimwema and Ocean Road seagrass meadows*

	MM	OR	Th	Ts	Cy	Hd	Sd	Period
NON HETEROCYSTOUS								
Oscillatoria limosa	+	+	+	+	+	+	+	Sept – June
Oscillatoria sp. 2	+	+	+	+	+	+	+	Sept –March
O. proboscidea	+	-	+	-	+	+	+	May – June
Oscillatoria sp. 4	+	-	+	-	+	+	+	Sept –Feb
Oscillatoria sp. 5	+	-	+	-	+	+	+	May – June
Oscillatoria sp. 6	+	-	+	-	+	+	+	Oct
Lyngbya sp. 1	+	+	+	-	+	+	-	Oct –Nov
Lyngbya sp. 2	+	-	+	-	+	+	+	Sept –Dec
L. confervoides	+	-	-	-	-	+	+	Oct – Nov
Spirulina gigantea	+	+	-	-	+	+	+	Oct –Dec, May–June
S. labyrinthiformis	-	+	-	-	+	+	+	Oct –Dec,May –June
Microcoleus chthonoplastes	+	+	-	-	-	-	+	March – June
Synechococcus sp 1	-	+	-	-	-	-	+	Oct
HETEROCYSTOUS								
Calothrix contarenii	-	+	-	-	-	-	+	Oct
Anabaena sp. 1	+	+	-	-	-	-	+	Oct
Nodularia sp. 1	+	+	-	-	-	-	+	Nov
UNICELLULAR								
Chroococcus minutus	+	-	-	-	-	-	+	Oct – Nov
Merismopedia elegans	+	-	-	-	-	-	+	Nov
Aphanocapsa sp. 1	-	+	-	-	-	-	+	April –May
Total number of cyanobacteria spp. found	15	11	8	2	10	11	18	

*MM, Mjimwema; OR, Ocean Road; Th, *Thalassia;* Ts, *Thalassodendron;* Cy, *Cymodocea*; Hd, *Halodule;* Sd, Sediments; Period, Duration of occurrence; +, Present; -, Absent

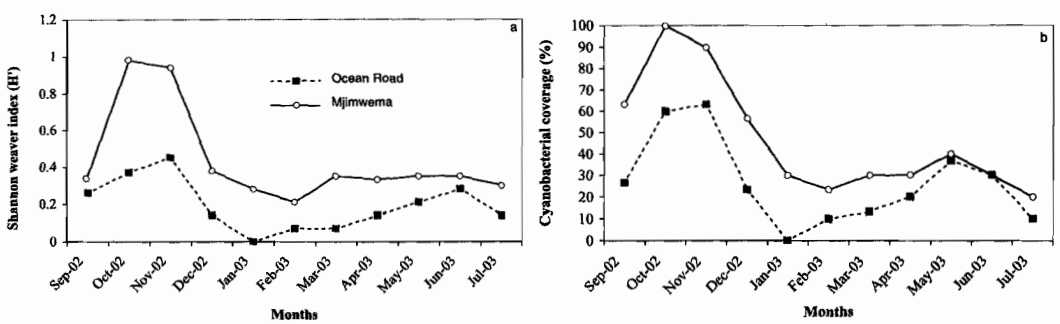
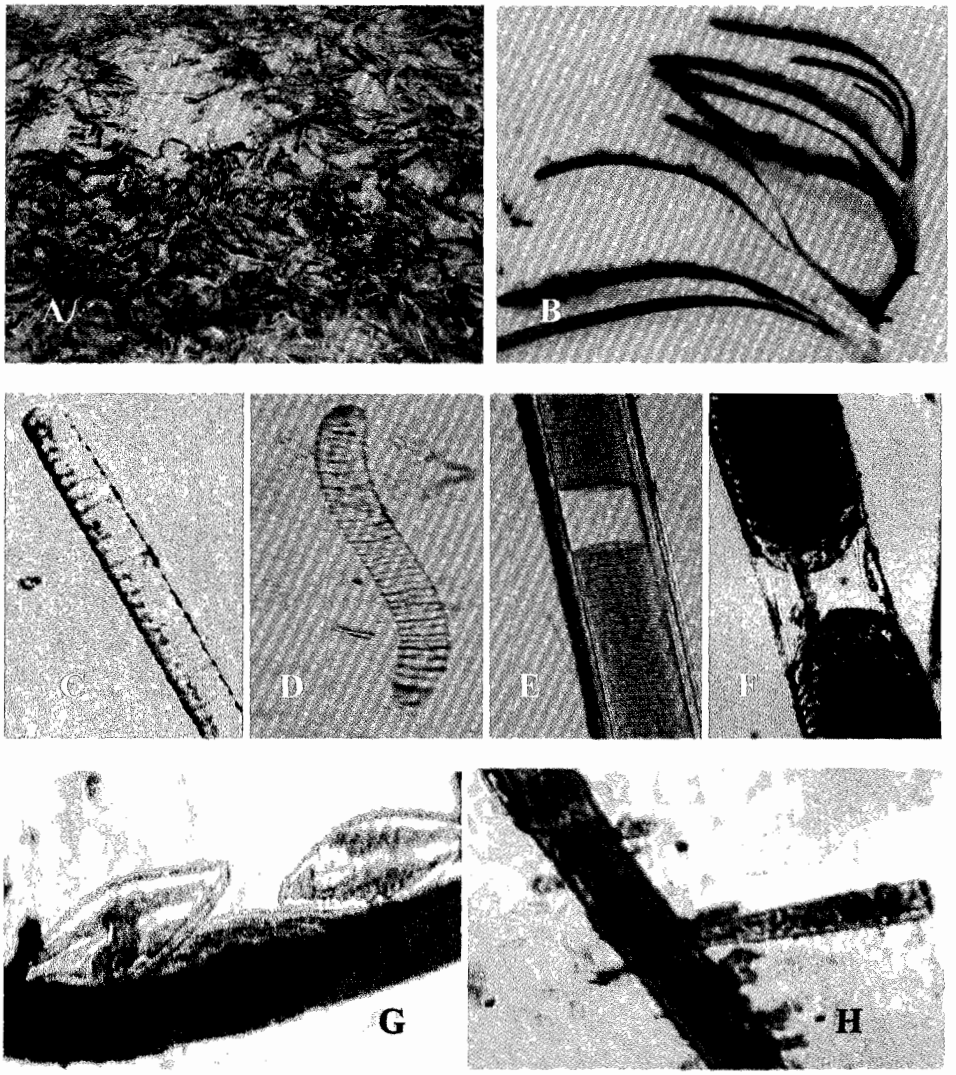


Fig. 1. Seasonal variation of cyanobacteria diversity and abundance in Mjimwema and Ocean Road. (a) Diversity (b) percentage coverage as a measure of relative abundance: Ocean Road (Dashes lines); Mjimwema (solid lines)



Plates. (A) Seagrass meadows dominated by the *Thalassia* sp. at low tide at Mjimwema showing the epiphytic cyanobacteria mats covered the seagrasses dominant cyanobacteria was *Oscillatoria* spp. (B) Epiphytic cyanobacteria as brownish mats attached to the *Cymodocea* sp., this mats was dominated by *Lyngbya* spp.(C) *Oscillatoria limosa* (D) *Oscillatoria* sp. 6 (E) *Lyngbya* sp. 1 (F) *Lyngbya confervoides* (G) Diatoms (*Pleurosigma* sp.) occurred together with the cyanobacteria (*Oscillatoria* sp.) (H) *Lyngbya* sp. 1 attached with the diatom (*Climacosphenia moniligera*). Scale (size bar):_____ Represents 50 μm in all micrographs.

Physico-chemical parameters

Nutrient levels in the tidal pool waters and the sediment pore waters of the two sites are shown in Figure 2a–c. The nutrient levels in seawater (tidal pool) ranged from 0.45–1.03 μmol NO_3^-NL/l, 0.19-0.27 μmol NO_2^-N/l, and from 0.03-0.09 μmol PO_4^-P/l for Ocean Road while at Mjimwema nutrient levels ranged from 0.14-0.93 μmol NO_3^-N/l, 0.20-0.30 μmol NO_2^-N/l and from 0.01–0.07 μmol PO_4^-P/l. Nitrate and phosphate levels were significantly higher ($t = 2.75$; $P = 0.001$ and $t = 2.34$; $P = 0.025$ respectively) at Ocean Road than at Mjimwema. However, there was no significant difference in nitrite levels between the study sites. The values for the nutrient levels in sediment pore waters were significantly higher compared to those of the tidal pool waters.

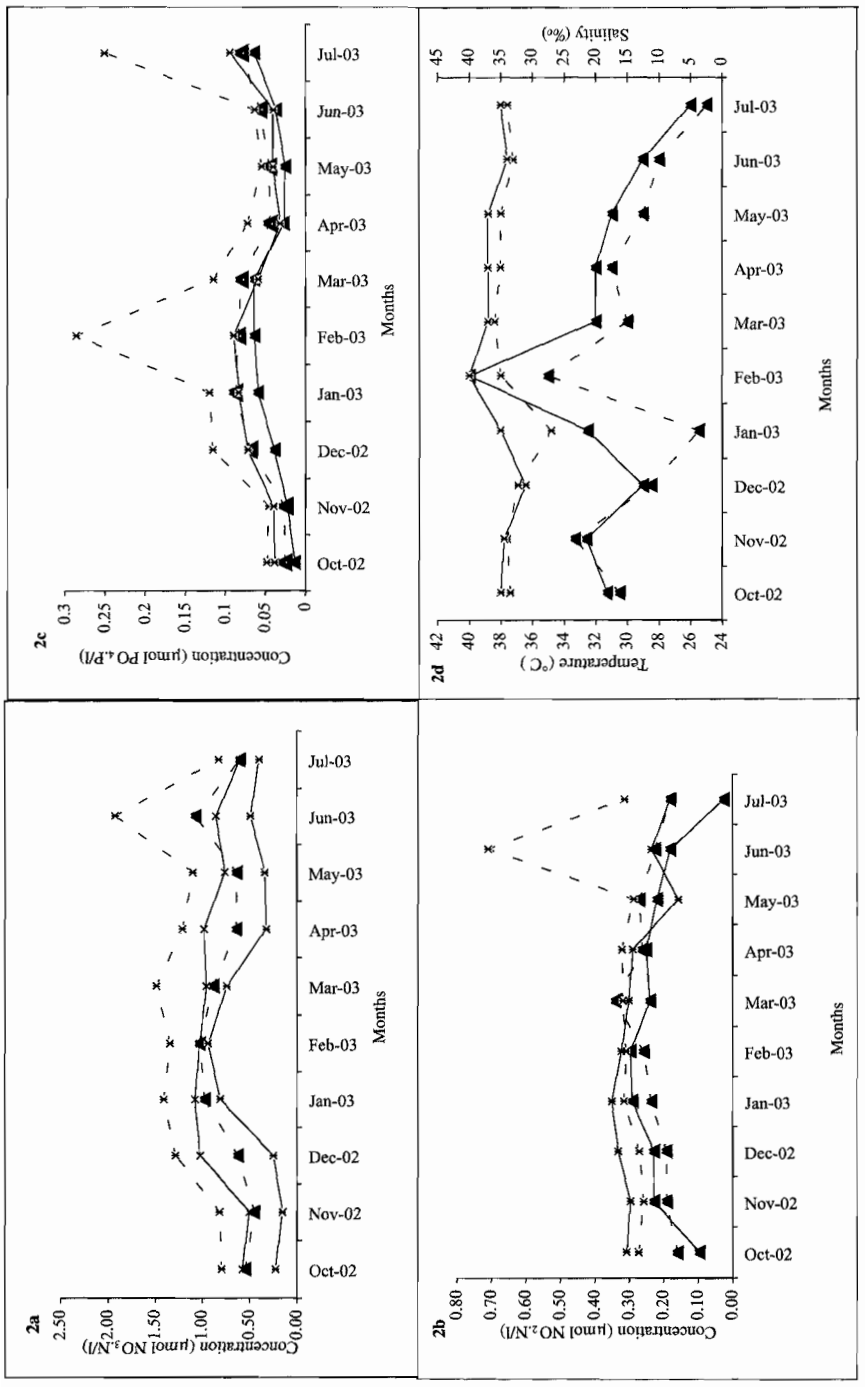


Fig. 2. Seasonal variation of the physical chemical parameters. (a) nitrate, (b) nitrite, (c) phosphate, (d) salinity and temperature: Dashed lines represent Ocean Road, Solid lines represent Mjimwema, triangles represent tidal pool waters, and stars sediment pore waters. In 2d, stars represent salinity and triangles represent temperature.

Temperature and salinity ranged from 25–40 °C and 27–40‰, respectively (Fig. 2d). There were no significant differences in salinity and temperature between the two sites or seasons. The Spearman correlation test showed that there was a significant negative correlation between the cyanobacterial cover and nitrate and phosphate levels ($r = -0.47$, $P = 0.047$ and $r = -0.55$, $P = 0.017$, respectively). However, no significant correlation was observed between cyanobacterial cover and other measured physico-chemical parameters.

DISCUSSION

Generally non-heterocystous *Oscillatoria* spp., *Lyngbya* spp. and *Spirulina* spp. were the dominant cyanobacteria found on the seagrass meadows. This might be because heterocystous forms have a glycolipid cell envelope that acts as an oxygen barrier, and limits their competitive advantage for survival in tropical sea waters. Thus, heterocystous cyanobacteria might be out-competed by non-heterocystous forms (Staal et al., 2003). It is also reported that heterocystous cyanobacteria are less resistant to turbulence of the ocean, which might break the weaker connections between heterocysts and vegetative cells (Stal, 1995). This may also explain why the heterocystous cyanobacteria were not found as epiphytes on seagrass leaves.

There were differences in the cyanobacterial composition between the two studied sites, with Ocean Road being dominated by *Spirulina* spp. and Mjimwema by *Oscillatoria* spp. and *Lyngbya* spp. The differences in species composition could perhaps be attributed to differences in physico-chemical parameters between the two sites. *Spirulina* spp. occur in environments with higher nutrient levels and varying temperature and salinity (Vonshak & Tomaseli, 2000). Thus, the domination of *Spirulina* spp. in Ocean Road is likely to be influenced by the higher nutrient levels, particularly nitrate and phosphate, as a result of sewage discharge.

While the cyanobacterial diversity and percentage coverage was higher at Mjimwema than at Ocean Road, the latter had a higher percentage of diatoms. This observation might be related to higher nutrient levels at Ocean Road, which in turn might affect competition and grazing (Stal, 1995). The association between these diatoms and cyanobacteria (Plate G & H) remains a subject for future research.

Some of the identified cyanobacteria, such as *Oscillatoria* spp., *Microcoleus chthonoplastes*, *Lyngbya* spp. and *Synechococcus* spp., are known to fix nitrogen gas (Bergman et al., 1997), thus they do not depend on exogenic nitrogen sources for growth (Pinckney et al., 1995). This may be an added advantage for cyanobacteria in Mjimwema, with lower nutrient levels as compared to Ocean Road. Other factors that would influence variability in cyanobacterial diversity and percentage coverage between the two study sites are the physical disturbance of the substratum, and sedimentation, both of which were higher at Ocean Road.

The higher species diversity and percentage coverage of cyanobacteria at Mjimwema coincide with a distinct formation/development of coloured microbial mats on the whole seagrass meadow. This type of mat was likely to be associated with low nutrient concentrations (especially nitrate) in the water column during the SE monsoon. This season is characterized by low rainfall, resulting in decreased runoff from rivers and hence low supplies of nutrient ions and organic metabolites (Bryceson, 1982). This trend is the opposite of that reported by Lugomela, (2002) in which planktonic cyanobacteria were observed to flourish during the NE monsoon. The lack of differences between the two seasons observed at Ocean Road may be due to the disturbance of substratum and high sedimentation as a result of anthropogenic effects such as harbour activities, sewage discharge and high inflow throughout the study period. Similar observations were reported previously by Lugendo et al. (1999).

The higher species diversity observed in sediments may be a result of the protective cover afforded by the seagrasses. However, the reason for the variation in cyanobacterial diversity within the studied seagrass genera was unclear. It may be attributed to specific seagrass zonation, as trends showed a higher diversity in seagrass species in the upper zone of the intertidal seagrass meadows, decreasing seaward.

CONCLUSION AND RECOMMENDATIONS

The generally observed elevated levels of nutrients, as a result of sewage discharge and other physical disturbances, may have a direct influence on the seagrass ecosystem, resulting in changes in and/or loss of epiphytic biodiversity. Sewage treatment, which reduced nutrient runoff, is clearly recommended. Considering the global ecological role of the seagrass ecosystems, periodic studies should be undertaken to assess changes in the seagrass ecosystem and its productivity. More

research is also needed to elucidate the variation in the cyanobacterial flora on the seagrass species, as well as to develop better methods for cyanobacterial identification, which might include molecular techniques.

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