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DISTRIBUTION OF EPIPHYTIC CYANOBACTERIA ON BROWN MACROALGAL SPECIES OCCURRING AT A ROCKY SHORE (BULEJI), KARACHI, PAKISTAN

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ABSTRACT

The present research was conducted to elucidate biodiversity of cyanobacteria associated with brown macroalgal species distributed on rocky coast of Buleji, Karachi. In total 48 species of cyanobacteria were identified and grouped into four orders and fifteen genera. Most of the species were recognized in the orders Nostocales (27 species) and Chroococcales (14 species). Whereas a number of species were also identified in the orders Chamaesiphonales (5 species) and Pleurocapsales (2 species). Marine cyanobacterial species have capability to grow in different type of media. The media used were: ASNIII, MN and Miquel's made up of both natural or artificial seawater and different nutrient composition.

Key-words: Buleji, Cyanobacteria, Epiphytic, Marine Habitat.

INTRODUCTION

A number of cyanobacterial species dwell in nature as epiphytes. Epiphytes grow on other plants saprophytically and derive moisture and nutrients from host (Yamamuro1999; Fong *et al.,* 2006.). Plants which live inter or intra-cellularly in some other plants or in cavities of glands cells, etc. are referred as epiphytes (Krings *et al*., 2009; Kaplan *et al.,* 1986; Lindblad, *et al.,* 1985; Stewart *et al.,* 1983; Desikachary, 1959).

In nature, a variety of cyanobacteria attach themselves to submerged plants. Some of the epiphytic species are included in taxonomic work from India (Desikachary 1959). Most of these species were from terrestrial and freshwater habitats. Comparatively a little work has been done on the occurrence of cyanobacteria associated particularly with marine plants. Most epiphytic cyanobacteria are briefly mentioned in the literature. For example, endosymbiosis relationship has been established for water fern *Azolla* (Stewart *et al.,* 1980; Singh and Singh, 1988). Cycads (Peters *et al.,* 1986; Millbank 1974), lichens (Fogg *et al*., 1973; Rai 1990a,b; Desikachary 1959) and an angiosperm *Gunnera* (Silvester, 1976). An endophytic cyanobacteria *Richellia intracellularis* is also reported in a microalgal species *Rhizosolenia styloformis* inhabiting in tropical ocean. *Richellia*- *Rhizosolenia* association is probably the only known endosymbiosis established from marine environment. The epiphytic cyanobacterial species on marine plants are comparatively better described. Cyanbacteria has been shown associated with seagrass, *Zostera marina* (Penhale 1977; Penhale and Smith 1977; Sand- Jansen 1977), mangrove barks and pneumatophores (Potts 1980), macroalgae (Carpenter1972; Cattaeno and Kalff 1979, 80; Desikachary 1959) etc.

From Pakistan epiphytic cyanobacteria are also reported from freshwater habitats (Shameel 1987). A few reports are available marine environments (Shameel and Butt 1984); Shameel and Tanaka 1992; Saifullah and Taj 1995; Nizamuddin and Gessner 1970).

There has been no dedicated work as regard to epiphytic cyanobacteria associated with marine algae, particularly from Pakistan. Since marine epiphytic cyanobacteria from Pakistani waters are scarcely known, studies on the epiphytic cyanobacteria on rocky shore of Karachi were carried out. The present research was undertaken with a view to expand the pool of knowledge regarding epiphytic cyanobacteria, particularly those growing on macroalgal species inhabiting intertidal zone of Buleji coast. Endophytic cyanobacteria are also known from terrestrial and freshwater environments.

MATERIALS AND METHODS

Cyanobacterial association with marine flora

A total of 6 species of brown seaweeds were randomly collected from Buleji, a rocky coast of Karachi. List of the species is set out in Table (1 and 3). Seaweeds species were kept separately in polythene bags and brought to the laboratory. The seaweeds were gently washed with sterilized seawater to remove loosely attached epiphytic species and extra organisms. The seaweeds were separated into different portions, for example, frond, utricle and rhizoids,

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all portions were individually inoculated in tubes containing three diverse types of media ASNIII, MN and Miquel's media and incubated under cool florescent light (12L/ 12D) at 30 ± 2^{o}C.

All seaweeds were inoculated in triplicates and in some cases where growth was either low or unobservable experiments were repeated more rigorously. The field materials and growth obtained in culture tubes were observed under the light microscope. The taxonomic assessments were made following Rippka *et al.,* (1979); Desikachary, (1959); Anagnostidis and Komarek, (1985, 1988), Komarek and Anagnostidis, (1986, 1989).

RESULTS

Composition and diversity of cyanobacteria growing as epiphytes on 6 Phaeophytes (Brown algal species) are documented in Tables 1 and 2. A total of 48 cyanobacterial species were identified belonging to 4 orders and 15 genera (Table 1). Most cyanobacteria observed were classified under Nostocalean genera (27 species), and some 14 Chroococcalean species were also registered. A fewer species were identified in orders Chamaesiphonales (5 species), and Pleurocapsales (2 species). It may be noted that brown algae *I. stellata, C. sinuosa* and *P. pavonia* had no cyanobacteria species that belong to orders Pleurocapsales and Chamaesiphonales (Table 1 and 2). The most interesting and conspicuous feature of distribution of cyanobacteria on brown algae is the one species *Stoechspermum marginatum* did not show any epiphytic cyanobacteria attached to it (Table 1 and 2). Some (7 species) of cyanobacteria were more common *i.e.*, attached with three or more species of Phaeophytes (Table 1). The abundance and distribution of cyanobacteria on each of the Phaeophytic species are shown in Table 1. *Sargassum tenerrimum* (27 species), and *Stokeyia indica* (19 species) had higher number of associative species. *Iyengaria stellata, Colpomenia sinuousa* and *Padina pavonia* represented only 16, 8 and 6 attached species, respectively (Table 1 and 2).

Table1. Distribution of cyanobacterial species on different species of phaeophytes inhabiting a rocky shore of Buleji near Karachi.

Cyanobacterial species	**Medium***	***Padina pavonia***	***Iyengaria stellata***	***Colpomenia sinuosa***	***Stoechospermum marginatum***	***Sargassum tenirrimum***	***Stokeyia indica***
Chroococcales							
Synechocystis pevalekii	MM	-	-	-	-	+	-
S. aquatilis	MM	-	-	-	-	+	-
Gloeocapsa kuetzingianum	ASNIII	-	-	+	-	-	-
Chroococcus cohaerence	ASNIII, MM	-	+	-	-	-	+
C. turgidus	MM	-	-	-	-	-	+
C. minutus	ASNIII, MM	+	+	-	-	+	-
C. minor	MM	-	+	+	-	-	-
C. indicus	MM	-	+	-	-	-	-
C. gomontii	ASNIII	-	-	-	-	+	-
Gloeothece rupestris	ASNIII	-	-	+	-	-	-
G. rhodochlmys	ASNIII, MM	-	-	+	-	+	-
Aphanocapsa littoralis	MN	-	-	-	-	+	-
Aphanothece nidulans	ASNIII	-	-	-	-	-	+
Merismopedia elegans	MM	+	-	-	-	-	-
Chamaesiphonales							
Dermocarpa leibleiniae	ASNIII, MN, MM	+	-	-	-	+	+
D. olivacea	MN	-	-	-	-	+	-
D. flahaultii	MM	-	-	-	-	-	+

D. clavata	MN, MM	-	-	-	-	+	-
D. sphaerica	MN	-	-	-	-	+	-
Pleurocapsales							
Myxosarcina spectabilis	ASNIII, MN, MM	-	-	-	-	+	+
M. burmensis	ASNIII	-	-	-	-	-	+
Nostocales							
Komvophoron anabaenoides	MN	-	-	-	-	+	-
K. schmidlei	ASNIII, MN	-	-	-	-	+	+
K. minutum	ASNIII, MN, MM	-	+	-	-	+	+
K. crassum	ASNIII	-	-	-	-	-	+
Psuedoanabaena galeata	ASNIII, MN	-	+	-	-	-	+
P. lonchoides	MN, MM	-	-	-	-	+	+
P. catenata	MN	-	-	-	-	+	-
Lyngbea polysiphoniae	ASNIII	-	-	-	-	-	+
Oscillatoria lemmermanii	MN, MM	+	-	-	-	+	-
O. pseudogaminata	ASNIII, MN	+	-	-	-	+	-
Spirulina labyrinthiformis	ASNIII, MM	-	+	-	-	-	-
Phormidium purpurascence	ASNIII, MM	-	+	+	-	+	-
P. subincrustatum	ASNIII	-	+	+	-	-	-
P. tenue	ASNIII, MM	-	+	+	-	+	+
P. uncinatum	ASNIII	-	-	+	-	-	-
P. angustissimum	ASNIII, MM	-	+	-	-	+	+
P. faveolarum	MM	-	+	-	-	-	-
P. ambiguum	ASNIII, MM	-	+	-	-	-	+
P.laminosum	ASNIII	-	+	-	-	-	-
P. amplivaginatum	ASNIII, MN, MM	-	-	-	-	+	+
P. fragile	ASNIII, MN, MM	-	+	-	-	+	+
P. breve	MN	-	-	-	-	+	-
P. incrustatum	ASNIII, MN	-	-	-	-	+	+
P. rimosum	MN	-	+	-	-	-	-
P. okenii	MN	-	-	-	-	+	-
P. luteum	MN	-	-	-	-	+	-
P. papyraceum	ASNIII	+	-	-	-	-	-

SNIII, MN (Rippka *et al.*, 1979), MM (Imai, 1977)

Table 2. Total number of species observed on surface of phaeoophyte species. Species are also grouped according to their taxonomic orders.

Cyanobacterial orders	***Padina pavonia***	***Iyengaria stellata***	***Colpomenia sinuosa***	***Stoechospermum marginatum***	***Sargassum tenirrimum***	***Stockeya indica***
Chroococcales	2	4	4	-	6	3
Chamaesiphonales	1	-	-	-	4	2
Pleurocapsales	-	-	-	-	1	2
Nostocales	3	12	4	-	16	12
Total	6	16	8	-	27	19

Most of the cyanobacteria were classified in the genus *Phormidium* (16 species) (Table 1). A fewer species were recognized in the following genera: *Dermocarpa* (5 species), *Komvophoron* (4 species), *Pseudoanabaena* (3 species), *Oscillatoria* (2 species), *Synechocystis* (2 species), *Gloeothece* (2 species) and *Myxosarcina* (2 species) (Table 1). The genera appear in single are *Gloeocapsa*, *Aphanocapsa*, *Aphanothec*, *Merismopedia*, *Spirulina* and *Lyngbea* (Table 1). Among these 26 species of cyanobacteria showed an algal species-specific cyanobacterial association with brown algae (Table 1). *Sargassum tenerrimum* showing greater number of species-specific associates. *Padina pavonia* (2 species), *Iyengaria stellata* (5 species), *Colpomenia sinuousa* (3 species) and *Stokeyia indica* (6 species) showed relatively lower number of such cyanobacteria (Table 1).

To obtain as many species as possible from the environment three types of media were used (Table 1). Only 28, 21 and 24 species were appeared in ASNIII, MN and Miquel's media respectively (Fig. 2). Five cyanobacterial species grew commonly in all three media (Table 1). There are some species which grew only in one medium. Number of exclusively growing species in ASNIII, MN and Miquel's media were 11, 9 and 8 species respectively ((Table 1 and Fig. 2).

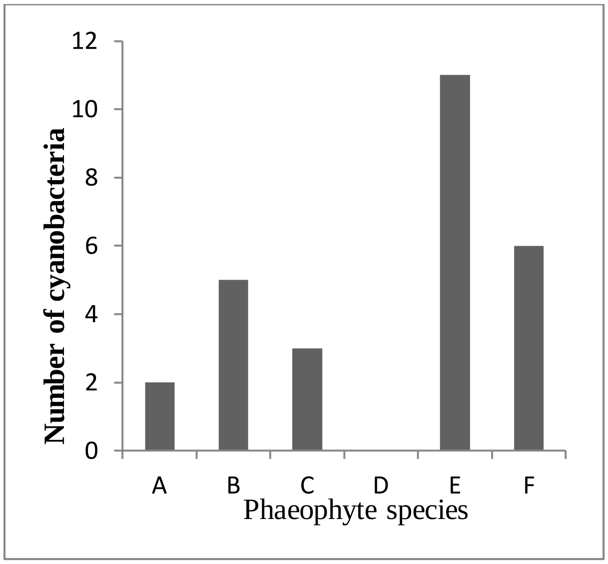

Fig. 1. Species-specific cyanobacteria associated with A- *Padina pavonia*, B-*Iyengaria stellata*, C-*Colpomenia sinuosa*, D- *Stoechospermum marginatum*, E-*Sargassum tenirrimum*, F-*Stokeyia indica.*

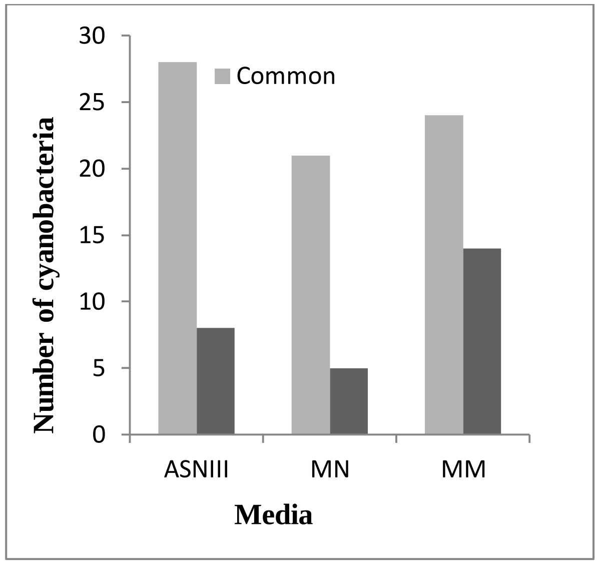

Fig. 2. Cyanobacterial species were common in all three media but some species showed up only in one specific medium.

DISCUSSION

There have been no attempts to identify the marine epiphytic flora in Pakistan. The data presented here contributes to the regional diversity data pool, and at the same time confirms the presence of many species reported from the other part of the world in the northern Arabian Sea bordering Pakistan.

Only a few reports are available on epiphytic cyanobacteria from coastal zones of Pakistan. These study reports only a few species of macroalgae (Nizamuddin and Gessner 1970; Shameel 1987) and on mangrove plants (Saifullah and Taj 1995).

There are some 475/255 species of marine macroalgae known to grow in the intertidal zone (Shameel and Tanaka 1992). Nutritional quality of coastal waters is high and thus support the high level of primary productivity in the region, and cyanobacteria may also an important role in this regard.

Despite its importance in the aquatic ecosystem, marine cyanobacteria has not been studied in detail. Growth of cyanobacteria on the group of macroalgae may suggest its possible role in the physiology of the algal species. The epiphytic cyanobacterial relationship may be symbiotic relationship. It has previously been reported that an exchange of carbon and nitrogen compounds exist between epiphytic cyanobacteria and the marine plants (Mohamed, and Al Shehri, 2010; Smith, and Russell, 1982; Harlin and Craigie 1975; Wetzel and Penhale 1979; Penhale and Thayer 1980; Harlin 1973; McRoy and Georing 1974). For example, diazotrophic epiphytes have been reported from a few marine algae (Moisander, *et al* 2005; Hoffman 1999; Steppe, *et al* 1996; Smith, and Russell 1982; Capone 1977; Capone *et al.*, 1977; Carpenter 1972; Hansen 1977; Head and Carpenter 1975).

Some cyanobacterial species encountered in the study as epiphytes are also present in many other niches of the rocky shore and are also reported from other habitats as well (Bano and Siddiqui, 2015; Hameed, 2009; Saifullah, and Ahmed 2007; Saifullah, *et al.*, 1997. Thajuddin and Subramanian 1992, 94; Santra and Pal 1988; Santra *et al.*, 1988; Desikachary 1959). These organisms therefore, appeared to be cosmopolitan and marine plant surface may simply be a substrate for attachment. However, there are some species of cyanobacteria found exclusively on macroalgae, and hence suggested a possibility of the existence of a symbiotic relationship. Only a variation in the diversity of species associated with different seaweed species, suggest that there may be a specific relationship between macroalgae and cyanobacteria.

The variation in cyanobacterial assemblages on algae may be due to the fact that cyanobacteria with different nutritional and environmental requirements occupy algal species growing at different tidal levels on a rocky shore. The extracellular exudates of different algae as may differ in nutrition and therefore support different cyanobacteria. Similarly, the exudates may carry deterrent compound, such as antibiotics which do not allow cyanobacteria to grow. A classical example may be a species of brown algae *Stoechospermum marginatum,* which was studied for epiphytic cyanobacteria but showed no attached cyanobacteria. A complete absence of cyanobacteria on *S. marginatum* indicates the possible secretion of some antibiotic compounds by this seaweed. This species has been shown to have antibacterial activity (Shaikh *et al.*, 1990; Rizvi, 2010). An absence of heterocystous epiphytic cyanobacteria is recorded as it is registered in the population of cyanobacteria inhabiting other niches of rocky shore (Howsley and Pearson, 1979).

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