

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

AZRA BANO¹ AND PIRZADA. J. A. SIDDIQUI²

¹Lasbela University of Agriculture Water and Marine Sciences, Uthal-90150, Baluchistan,

²Centre Of Excellence in Marine Biology, University of Karachi, Karachi-75270, Pakistan

* Corresponding author e-mail azrabano2006@yahoo.com

خلاصہ

موجودہ مطالعہ کے دوران cyanobacteria کی نوع بھولے جی کراچی کے ساحل سے معلوم ہوا کی گئی۔ کل 67 انواع سرخ الجی کے ساتھ چپکی ہوئی حالت میں اس گروہ کی پائی گئی ہیں۔ رودافانی سی انواع ہیں Melanothamnus somaliensis, Laurencia pinnatifida, ceramium monorencse, centroceras clavulatum, Hypxeapannusa, calliblepharis fimbriata, Galaxaures oblongata موجود تھیں۔ Cyanobetesia کی انواع کی جماعت بندی 4 ادرز میں کی گئی یعنی اتر Nostocales میں 41 Chroococcales میں 17 Hypnea اور 6 Chamaesiphonales میں 6 اور pleurocapsales اتر میں دو انواع رکھیں گئیں۔ سب سے زیادہ افراد یعنی 23 انواع Hypnea 17 پاننوسا پر جبکہ سب سے کم یعنی چھ انواع Calliblepharis fimbriata پر چپکی حالت میں پائی گئی۔ تین مختلف اقسام کے میڈیا (Miquels, MN, ASN) یا تو قدرتی تور پر تیار کیئے گئے یا مصنوعی سمندر کے پانی کو استعمال کیا گیا موجودی تجربات کے دوران

Abstract

In the present research the epiphytic species of cyanobacteria were recorded in the coastal region of Buleji, Karachi. In total 67 species of cyanobacteria were found attached to the surface of red algae. The rhodophycean species were *Galaxaura oblongata*, *Calliblepharis fimbriata*, *Hypnea pannosa*, *Centroceras clavulatum*, *ceramium manorencse*, *Laurencia pinnatifida* and *Melanothamnus somaliensis*. The species of cyanobacteria were classified in the four orders Nostocales, (41 species) Chroococcales (17 species), Chamaesiphonales (6 species) and Pleurocapsales (2 species). The highest number of cyanobacteria were attached with *Hypnea pannosa* (23 species) and the lowest number with *Calliblepharis fimbriata* (6 species). Three different media (ASNIII, MN and Miquel's) made of either natural or artificial seawater were used in this experiment.

Introduction

The epiphytic relationship of cyanobacteria appears to be symbiotic (Meeks and Elhai, 2002; Carpenter and Foster, 2002; Braun-Howland and Nierzwicki, 1990). The cyanobacterial associates receive carbon, nitrogen and other inorganic nutrients from the organisms it is attached to. For example, transfer of photosynthetic products, nitrogen and phosphorous has been shown (Yamamuro, 1999; Wetzel and Penhale, 1979; Penhale and Thayer, 1980; Harlin and Craigie, 1975; McRoy and Georing, 1974; Harlin, 1973). In return, plants are protected from the grazers (O'Neil and Roman, 1992; Hutchinson, 1975). Plants also obtain organic material from symbiotic cyanobacterial counterparts. Endosymbiotic cyanobacteria have been shown to fix dinitrogen and its products are used up by the plants in exchange of carbon and photosynthesis reductants supply to cyanobacteria (Lesser, *et al.*, 2004; Lesser, *et al.*, 2007). Such endosymbiosis is known for a variety of systems, viz, Ascomycetes (Gehrig *et al* 1996; Fogg *et al*, 1973), diatoms (Carpenter *et al.*, 1999; Villareal, 1989; Martinez *et al.*, 1983), bryophytes from *Azolla* (McCowen, *et al.*, 1981; Kaplan and Peter, 1998), gymnosperm species of cycads (Thajuddin *et al.*, 2010; Stewart *et al.*, 1980) and angiosperm *Gunnera* (Johansson and Bergman, 1994; Rai, 1990a). Epiphytic cyanobacterial diazotrophs have also been reported from macroalgal species (Capone 1977; Capone *et al.*, 1977). Nitrogen fixing bacteria and cyanobacteria are, for example, reported on *Codium* and *Sargassum* species (Hamersley *et al.*, 2015; Gerard *et al.*, 1990; Philips, *et al.*, 1986; Carpenter, 1972; Hanson, 1977; Head and Carpenter, 1975). In nature, a variety of cyanobacteria attach themselves to submerged plants. Some of the epiphytic species are included in taxonomic work from India (Thajuddin and Subramanian, 2005; Desikachary, 1959).

From Pakistan epiphytic cyanobacteria are also reported from freshwater habitats (Malik *et al.*, 2012; Ahmed *et al.*, 2010; Shameel, 1987). A few reports are available marine environments (Uzair *et al.*, 2012; Javed *et al.*, 2011; Bano and Siddiqui, 2004. Shameel and Butt 1984; Shameel and Tanaka, 1992; Saifullah and Taj, 1995; Nizamuddin and Gessner, 1970). As marine epiphytic cyanobacteria from Pakistani waters is scarcely known, studies on the epiphytic cyanobacteria from rocky shore of Karachi was conducted. 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.

Materials & Methods

A total of 7 seaweeds were randomly collected from Buleji, a rocky coast of Karachi. List of the species is set out in Table (1). 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 other organisms. The seaweeds were divided into different portions, for example, frond, utricle and rhizoids. Each portions was separately inoculated in tubes containing three different types of media ASNIII, MN and Miquel's media and incubated under cool florescent light (12L/ 12D) at $30 \pm 2^\circ\text{C}$.

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 Rippka *et al.*, 1979; Desikachary, 1959; Anagnostidis and Komarek, 1985, 1988, Komarek and Anagnostidis, 1986, 1989.

Results

Epiphyte species of cyanobacteria growing on red seaweeds (Rhodophyte) were observed and results are expressed in (Tables 1 and 2). A total of comprising 67 cyanobacterial species pertaining to 21 genera distributed among 4 orders (Chroococcales, Chamaesiphonales, Pleurocapsales and Nostocales) identified from seven species of red algae (Tables 1 and 2). Species in the order Stigonematales were not observed (Tables 1 and 2). The observed species mostly belongs to the orders Chroococcales (17 species) and Nostocales (42 species). The orders Chamaesiphonales and Pleurocapsales represented only 6 and 2 species respectively. (Table 1 and 2). Table 1 also shows that the Nostoccalean genera *Phormidium* (17 species) and *Oscillatoria* (8 species) were dominating among cyanobacterial flora. The other genera represented only a few species were *Dermocarpa* (5 species), *Lyngbea* (5 species), *Gloeocapsa* (3 species), *Chroococcus* (3 species), *Merismopedia* (3 species), *Komvophoron* (3 species), *Psuedoanabaena* (3 species), *Spirulina* (3 species), *Synechocystis* (2 species), *Gloeotheca* (2 species), and *Planktothrix* (2 species) (Table 1). A few genera (eight) represented by one species (Table 1). No cyanobacterial species belonging to orders Chamaesiphonales and Pleurocapsales were observed with associated with some red algae species (*Calliblepharis fimbriata*, *Galaxaura oblongata*, *Centroceras clavulatum*, and *Laurencia pinnatifida*) (Table 1 and 2). Table 1 and 2 also illustrate the abundance of cyanobacteria on each species of red algae. A large number of cyanobacteria species were guested on *Hypnea pannosa* (23 species) and *Ceramium manorense* (20 species). The other rhodophytic species had variable numbers of cyanobacteria, such as, *Galaxaura oblongata* (23 species), *Laurencia pinnatifida* (17 species), *Centroceras clavulatum* (15 species), *Melanothamnus somaliensis* (15 species) and *Calliblepharis fimbriata* (6 species). A species-specific association also observed between cyanobacteria and rhodophytic species. Out of 67 species of cyanobacteria, over 50% species (39 species) were showed species-specific association i.e., attached to any one species of red algae (Table 1 and Fig.1). *Ceramium manorense* (9 species) and *Hypnea pannosa* (8 species) have greater number of species-specific cyanobacteria, whereas, *Galaxaura oblongata* (7 species) *Centroceras clavulatum* (5 species), *Melanothamnus somaliensis* (4 species) and *Laurencia pinnatifida* (6 species) had low numbers of associative cyanobacteria (Table 1 and Fig.1).

Epiphytic cyanobacteria were grown in all three media and result are described (Table 1). A large number of cyanobacteria (45 species) were grown in ASNIII medium. There were 33 and 30 species of cyanobacteria observed in Miguel's and MN media, respectively. 9 species were common in all three media while 17, 10 and 8 species appeared only in ASNIII, MN and Miquel's media (Table 1 & Fig.2).

Discussion

The information presented here put in to the regional wide range of records, and at the same time provide evidence the presence of various species cyanobacteria details from the other part of the globe in the northern Arabian Sea bordering Pakistan.

In all marine environments, available surfaces are quickly colonized by a diversity of life forms. However, these organisms grow on plants and macroalgae they are described epiphytes. Symbioses among cyanobacteria and marine organisms are sufficient and common among marine plants and animals (Rosenberg and Paerl, 1980). In the marine environment, symbioses are well-known to occur between cyanobacteria and sponges, Ascidians (sea squirts), and Echuroid worms in the benthos, and diatoms, dinoflagellates and a protozoan with the plankton (Carpenter and Foster 2002).

The macroalgal species provide excellent substratum (stratum, nutrient and current) for the attachment and growth of various cyanobacterial species. In the current investigation, a number of the species of cyanobacteria among 67 species of cyanobacteria may be mandatory to be epiphytic in the marine environment. The species of macroalgae collected in this study were commonly distributed at the intertidal zone of rocky coast (Buleji). Cyanobacterial growth was observable on most of the species by naked eye. It is interesting to note that various

species of cyanobacteria observed as epiphytic, which have previously been reported the same as edaphic, epilithic and planktonic from India (Desikachary, 1959), and also as epizoic from freshwater habitats from Pakistan (Khalid *et al.*, 2014; Saifullah *et al.*, 1997; Hussein and Anjum, 1982; Anjum *et al.*, 1980). The observed species have been reported from marine as well as freshwater habitats from different parts of the world (Ulcaiy *et al.*, 2015; Aleem, 1980; Desikachary, 1959; Thajuddin and Subramanian, 1990, 1992, 1994; Santra and Pal, 1988; Shameel and Butt, 1984).

The observed cyanobacterial species, such as *Gloeocapsa compacta*, *Gloeothece rhodochlmys*, *Dermocarpa leibleinia*, *Oscillatoria pseudogaminata*, *Phormidium angustissimum*, and *P. fragile* were attached on most of macroalgal species. This could be due to species preference of the habitat. Most of the cyanobacterial species reported in the study as epiphytic forms were also reported from various types of soil and hard substrata in terrestrial and aquatic habitats, such as, river soil, cultivated and non-cultivated soil, mud mangrove swamps, pneumatophores of mangrove, beach rocks, stones surfaces, associated with sea grasses and on macroalgae (Rigonato *et al.*, 2012; Larkum, *et al.*, 2012; Vizzini, *et al.*, 2002; Borowitzka, *et al.*, 2007; Saifullah and Ahmed, 2007; Bano and Siddiqui, 2004; Toledo *et al.*, 1995; Santra and Pal, 1988; Santra *et al.*, 1988; Maity *et al.*, 1987; Aleem, 1980; Desikachary, 1959). *Phormidium fragile* and *P. tenue* have also been described as epizoic on freshwater turtle and snail shells (Hussein and Anjum, 1982; Anjum *et al.*, 1980). The occurrence of *Chl* d-containing cyanobacteria in epiphytic biofilms on a red alga (*Gelidium caulacanthum*) occupied the pneumatophores of a temperate region mangrove (*Avicennia marina*) (Larkum, *et al.*, 2012).

In the marine environment spongy surface of macroalgae provide a favorable habitat for attachments of cyanobacteria. Attachment of cyanobacteria and variability of species composition of associative cyanobacteria may simply be a function of variation in the habitat of macroalgae as noted above. It seems true for many species growing on surface macroalgae are also observed in other niches of Buleji coast (Bano and Siddiqui, 2017 a, b, 2004, 2003.). Furthermore, the taxonomy and morphology of epiphytic algae in the Indus-Delta were reported (Tanaka and Shameel 1992).

In addition, a lot of species recorded here have been reported from marine and freshwater habitats from India (Desikachary, 1959; Aleem, 1980; Santra and Pal, 1988; Santra *et al.*, 1988; Maity *et al.*, 1987; Anand and Hooper, 1987; Thajuddin and Subramanian, 1990, 1992, 1994), and other parts of the world (Hussein and Khoja, 1993; Bauld, 1981; Hogdson and Abbot, 1992; Khoja, 1987).

The present study provide a basic information about epiphytic cyanobacteria on a variety of macroalgal species inhabiting the rocky shore of Buleji and confirms the presence of many species reported from other parts of the world in the Northern Arabian Sea bordering Pakistan.

Thus, advance research work has to be undertaken to obtain information about the diversity of epiphytic cyanobacteria in the marine environment.

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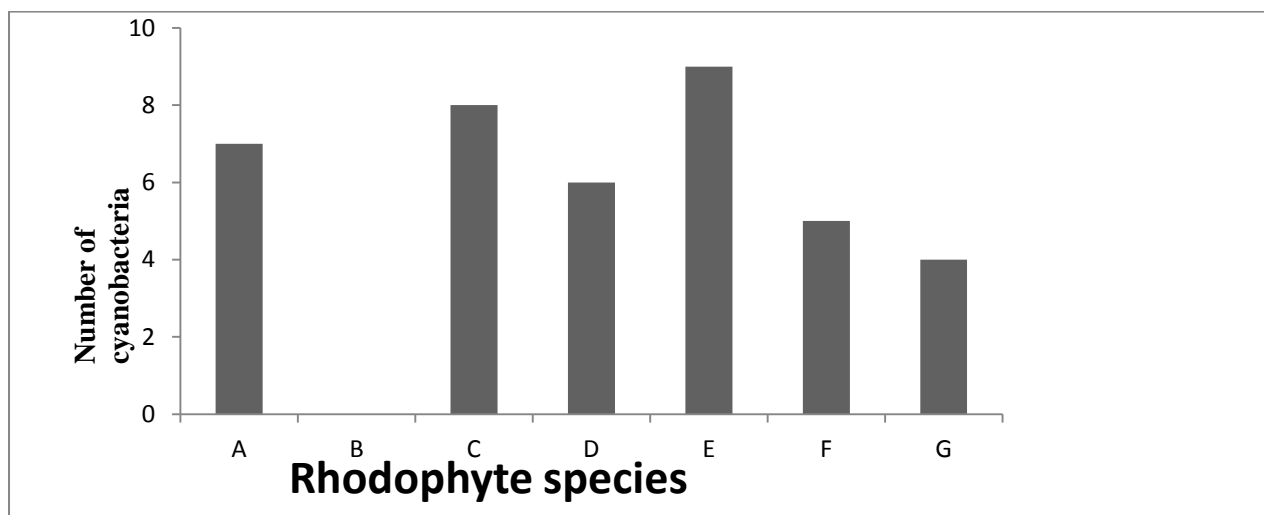


Fig. 1. Species-specific cyanobacteria associated with A- *Galaxaura oblongata*, B- *Calliblipharis fimbriata* C- *Hypnea pannosa*, D- *Centroceras clavulatum*, E- *Ceramium manorense*, F- *Laurencia pinnatifida*, G- *Melanothamnus somaliensis*

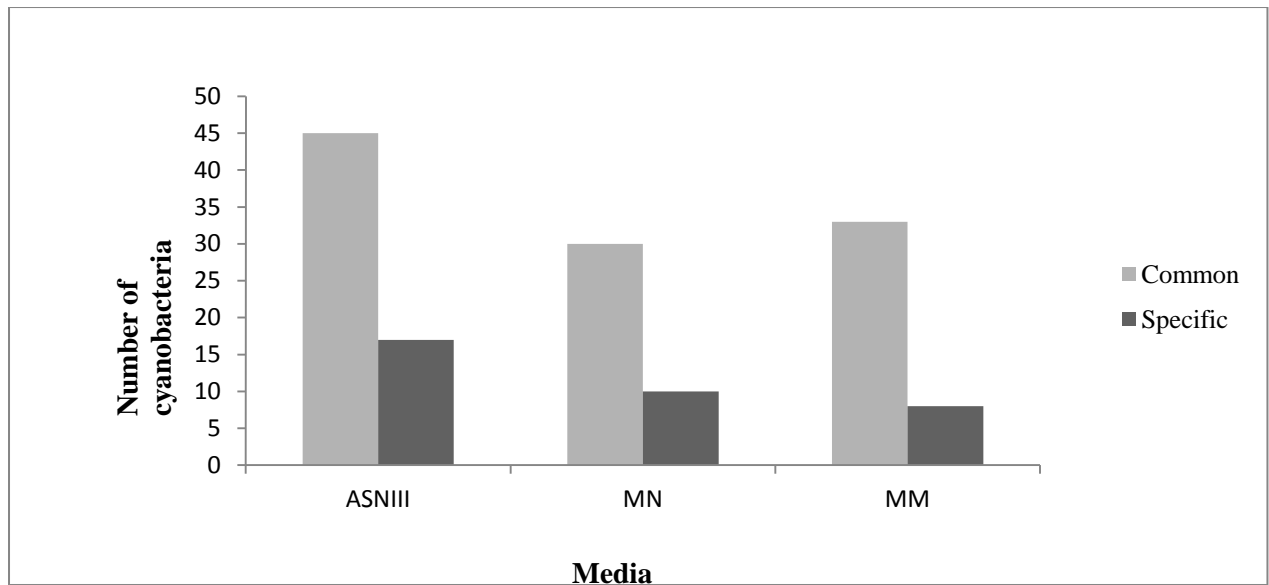


Fig. 2. Cyanobacterial species were common in all three media but some species showed up only in one specific medium.

Table1. Distribution of cyanobacterial species on seven different species of rhodophytes inhabiting a rocky shore of Buleji, near Karachi.

Cyanobacterial species	Medium*	<i>Galaxaura oblongata</i>	<i>Callibipharis fimbriata</i>	<i>Hypnea pamosa</i>	<i>Centroceras clavulatum</i>	<i>Ceramium manorense</i>	<i>Laurencia pinnatifida</i>	<i>Melanothamnus somaliensis</i>
Chroococcales								
<i>Synechocystis pevalekii</i>	ASNIII, MN	-	-	-	-	-	+	+
<i>S. aquatilis</i>	MN	-	-	-	-	-	+	-
<i>Gloeocapsa compacta</i>	ASNIII, MM	+	+	+	-	-	-	-
<i>G. cripidinum</i>	ASNIII	+	+	-	-	-	-	-
<i>G. punctata</i>	MN, MM	-	-	+	-	-	-	-
<i>Chroococcus cohaerence</i>	MN	-	-	+	-	-	-	-
<i>C. minutus</i>	MN, MM	-	-	+	-	+	-	-
<i>C. minor</i>	ASNIII, MM	+	-	-	-	+	-	-
<i>Synechococcus cedrorum</i>	MN	-	-	-	-	+	-	-
<i>Gloeotheca samoensis</i>	MN	-	-	-	-	-	-	+
<i>G. rhodochlmys</i>	ASNIII, MM	+	-	+	-	+	+	-
<i>Aphanocapsa littoralis</i>	ASNIII, MM	+	-	+	-	-	-	-
<i>Aphnothece bullosa</i>	ASNIII	+	-	-	-	-	-	-
<i>Merismopedia elegans</i>	MN	-	-	+	-	-	-	-
<i>M. glauca</i>	MN	-	-	-	+	-	-	-
<i>M. tenuissima</i>	MM	-	-	+	-	-	-	-

<i>Coelosphaerium kuetzingianum</i>	MN	-	-	+	-	-	-	-	
Chamaesiphonales									
<i>Chroococidiopsis indica</i>	MN	-	-	+	-	-	-	-	
<i>Dermocarpa leibleiniae</i>	ASNIII, MN, MM	-	-	+	+	+	+	+	
<i>D. clavata</i>	MN	-	-	-	-	-	+	-	
<i>D. flahaultii</i>	MM	-	-	-	+	-	-	-	
<i>D. versicolor</i>	MM	-	-	-	-	+	-	-	
<i>D. olivacea</i>	ASNIII, MN, MM	-	-	-	-	-	+	-	
Pleurocapsales									
<i>Myxosarcina burmensis</i>	ASNIII, MN, MM	-	-	+	-	-	-	+	
<i>Hyella caespitosa</i>	ASNIII	-	-	-	+	-	-	-	
Nostocales									
<i>Borzia susedana</i>	ASNIII, MM	-	-	-	-	-	+	-	
<i>Komvophoron schmidlei</i>	ASNIII, MN, MM	-	-	-	-	+	-	+	
<i>K. pallidum</i>	MM	-	-	-	-	-	+	-	
<i>K. minutum</i>	MN, MM	-	-	-	-	-	+	+	
<i>Psuedoanabaena galeata</i>	ASNIII, MN, MM	-	-	-	-	-	+	+	
<i>P. catenata</i>	ASNIII, MN	-	-	+	+	-	-	-	
<i>P. papillaterminata</i>	MN	-	-	-	-	+	-	-	
<i>Lyngbea gardnerii</i>	ASNIII	-	-	-	-	+	-	-	
<i>L. nordgarddhii</i>	ASNIII, MM	+	+	-	-	-	-	-	
<i>L. allogei</i>	ASNIII	+	-	-	-	-	-	-	
<i>L. cryptovaginata</i>	ASNIII	-	-	-	-	-	-	+	
<i>L. borgertii</i>	ASNIII	-	-	-	-	-	-	+	
<i>Oscillatoria acuminata</i>	ASNIII	+	-	-	-	-	-	-	
<i>O. fremyii</i>	ASNIII, MN	-	-	-	-	-	+	-	
<i>O. tenuis</i>	ASNIII	+	-	-	-	-	-	-	
<i>O. okenii</i>	MM	-	-	-	-	+	-	-	
<i>O. limosa</i>	ASNIII	-	-	-	-	+	-	-	
<i>O. schultzii</i>	ASNIII, MN	-	-	+	-	-	-	-	
<i>O. quadripunctata</i>	ASNIII	-	-	-	+	-	-	-	
<i>O. pseudogaminata</i>	ASNIII, MN, MM	+	+	+	+	+	+	-	
<i>Spirulina labyrinthiformis</i>	ASNIII, MM	+	-	+	-	-	-	-	
<i>S. subsalsa</i>	ASNIII	+	-	-	-	-	-	-	
<i>S. major</i>	ASNIII	-	-	-	+	-	-	-	
<i>Planktothrix compressa</i>	MM	-	-	-	+	-	-	+	
<i>P. clathrata</i>	ASNIII, MM	-	-	-	+	-	+	+	
<i>Phormidium purpurascence</i>	ASNIII, MM	+	+	-	-	-	-	-	
<i>P. molle</i>	ASNIII	+	-	-	-	-	-	-	
<i>P. tenue</i>	ASNIII, MM	-	-	+	+	-	-	-	
<i>P. insigni</i>	ASNIII	-	-	+	-	-	-	-	
<i>P. angustissimum</i>	ASNIII, MN, MM	-	-	+	+	+	-	+	
<i>P. corium</i>	ASNIII, MM	+	-	-	-	-	-	-	
<i>P. mucosum</i>	MN, MM	-	-	+	+	+	-	-	
<i>P. africanum</i>	ASNIII, MN	-	-	+	-	-	+	-	
<i>P. amplivaginatam</i>	ASNIII, MN	-	-	-	-	-	+	+	

<i>P. fragile</i>	ASNIII, MN, MM	+	+	+	+	+	+	-
<i>P. ambiguum</i>	ASNIII, MN	-	-	+	-	+	-	-
<i>P. retzii</i>	ASNIII, MM	-	-	+	-	+	-	-
<i>P. mucicola</i>	ASNIII, MN, MM	+	-	-	-	-	+	+
<i>P.laminosum</i>	MM	-	-	-	+	-	-	-
<i>P. papyraceum</i>	ASNIII	-	-	-	-	+	-	-
<i>P. lucidum</i>	ASNIII	-	-	-	-	-	-	+
<i>P. valdrrianum</i>	MM	-	-	-	-	+	-	-

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

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

Cyanobacterial orders	<i>Galaxaura oblongata</i>	<i>Callitripharis fimbriata</i>	<i>Hypnea pannosa</i>	<i>Centroceras clavulatum</i>	<i>ceramium manorensense</i>	<i>Laurencia pinnatifida</i>	<i>Melanothamnus somaliensis</i>
Chroococcales	6	2	9	1	4	3	2
Chamaesiphonales	-	-	2	2	2	3	1
Pleurocapsales	-	-	1	-	1	-	1
Nostocales	12	4	11	12	13	11	11
Total	18	6	23	15	20	17	15

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