



Subaerial cyanobacteria on the monuments and exterior surface of building facades of Odisha state, India

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ABSTRACT: Subaerial cyanobacteria on exterior surface of monuments and building facades have great importance under extreme conditions to enhance their colonization and diversity. Work was undertaken to assess the cyanobacteria diversity of Western Odisha especially concerning to monuments and building facades, and their comparative analysis. In total fifty subaerial cyanobacterial taxa belonging to 21 different genera of five orders were documented. Out of these 19 taxa were documented as new distributional records from the subaerial habitats of Odisha. Morphologically, 50% of taxa were filamentous heterocystous forms, followed by 26% colonial forms and 24% filamentous non-heterocystous forms. Diversity analysis revealed that cyanobacteria diversity was relatively higher in building facades rather than the monuments which may be resulted due to variation in substrate composition and exposure to climatic factors. A comparative distributional analysis of so far documented cyanobacteria from various subaerial habitats of Odisha revealed a habitat specificity is observed among the cyanobacteria from the monuments which represent biofilms with less common taxa.

KEY WORDS: Subaerial biofilms, buildings and stone monuments, cyanobacteria diversity, diversity indices.

INTRODUCTION

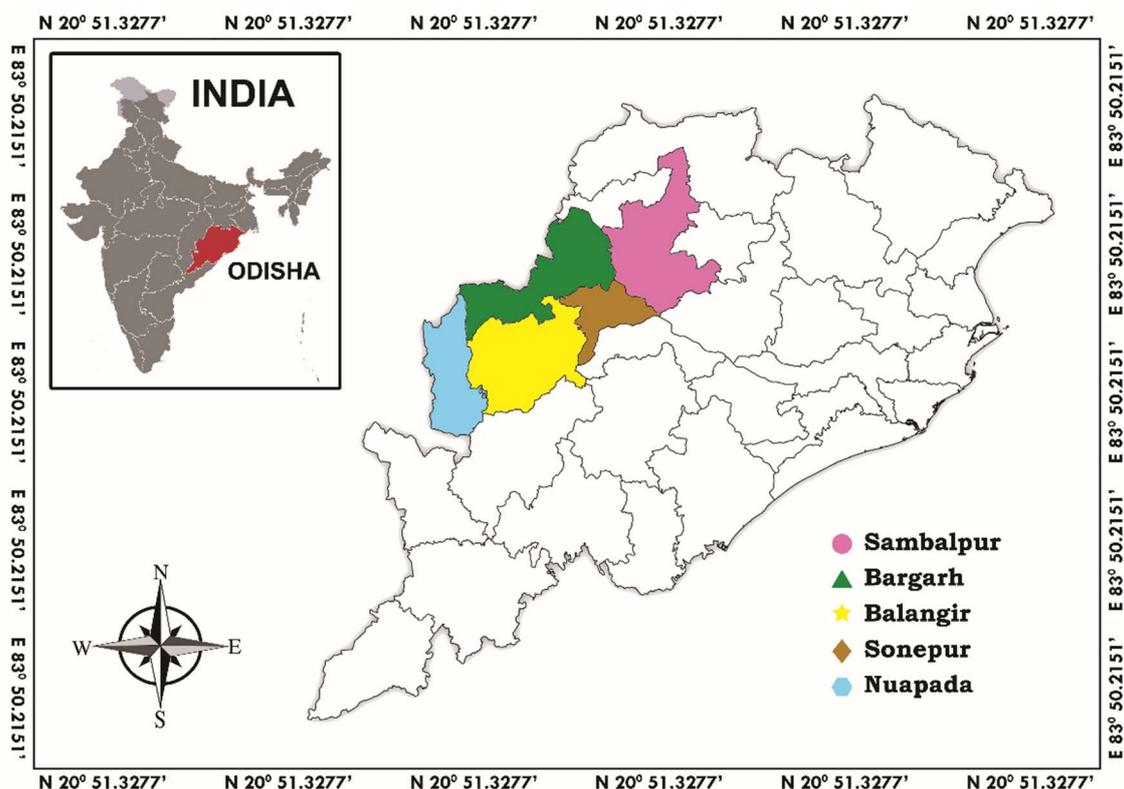
Odisha is located in the eastern part of India, houses a bountiful of culturally and archaeologically significant rock/stone monuments scattered all across the state. Cyanobacteria form multi-species consortium along with other microbes, i.e., algae, fungi and lichens, and thrive on the exterior of these monuments and sometimes cause biodeterioration. Since last few decades, these photosynthetic bacteria grab attention of researchers and their diversity was documented periodically (Tripathy *et al.*, 1997; Roy *et al.*, 1997; Samad and Adhikary, 2008; Sahu *et al.*, 2011; Keshari and Adhikary, 2014; Adhikary *et al.*, 2015; Pradhan *et al.*, 2018). These organisms were not only identified through polyphasic characterization, but also screened for the presence of UV sunscreen pigments. Their unique survival and drought/heat stress tolerance ability enabled them to colonize for years by formation of 'biofilms'. The organisms growing there-in biofilms can be called subaerial or extremophiles due to enduring extreme environments (Gaylarde, 2020; Pinna, 2021). Apart from the monuments, these subaerial cyanobacteria also colonize exposed building facades, rock surfaces, and other lime washed surfaces (Samad and Adhikary, 2008). Environmental factors, like illumination, atmospheric humidity, and rainfall regulate the cyanobacterial distribution in various biofilms. Geological aspects, like the surface roughness, porosity, and mineralogical nature of the rock substrate, also influence the cyanobacterial diversity. Several studies (Gaylarde and Gaylarde, 2005; Barberousse *et al.*, 2006; Samad and Adhikary, 2008; Macedo *et al.*, 2009;

Karande *et al.*, 2012; Adhikary *et al.*, 2015; Villa *et al.*, 2016; Ogawa *et al.*, 2017; Pradhan *et al.*, 2018; Ortega-Morales *et al.*, 2019) from tropical as well as temperate regions reveal their findings that the subaerial biofilms on building facades and monuments varied greatly in their biodiversity by the influences of environmental factors basing on their substrates which included cement, mortar, brick, concrete, rock, granite, lime-washed wall, stone carving, sandstone, limestone, marbles etc., between the hot summer and the rainy seasons. Most of the reports cited that subaerial biofilm on exposed substrata appears with bluish-green, brownish or blackish colours due to the assemblages of microorganisms in which cyanobacteria are the dominant colonizers (Keshari and Adhikary, 2014).

The presence of pigments in the cells and their ability to produce variable secondary metabolites ensured their adaptability in extreme conditions (Singh *et al.*, 2016; Pattnaik *et al.*, 2021). Therefore, in recent years, the cyanobacteria from subaerial habitats gaining great interest with the hope of yielding promising results and make an innovative contribution as a potential source of various secondary metabolites and their biotechnological applications (Jeong *et al.*, 2020). Many studies have reported cyanobacteria as a good source of various pigments used in diagnostic, fluorescent probes or in oxidative stress and metabolites with diverse biological activities in pharmaceutical sciences and industries (Wada *et al.*, 2013; Saini *et al.*, 2018; Tiwari and Tiwari, 2020). This prompted us to carry out this investigation to survey the cyanobacteria communities from two different subaerial habitats i.e., Monuments and Building facades

**Table 1.** The location with GPS, number of sampling sites and their environmental conditions of five districts of Western Odisha.

Sl. No.	District	Geographic location		Sampling substrates	Temp (°C)	RH (%)	Annual average Rainfall (mm)
		Latitude	Longitude				
1.	Balangir	20°96.8'N	85°74.5'E	10 Building facades 05 Monuments	27 - 28°C 31 - 45°C	82 - 85% 65 - 77%	1229.4
2.	Bargarh	21°38.1'N	83°59.6'E	06 Building facades 05 Monuments	27 - 30°C 29 - 35°C	80 - 85% 72 - 82%	1337.5
3.	Nuapada	20°80.7'N	82°53.4'E	05 Building facades 03 Monuments	27 - 28°C 33 - 38°C	77 - 82% 70 - 80%	1378.2
4.	Sambalpur	21°50.0'N	83°94.5'E	09 Building facades 05 Monuments	28 - 33°C 38 - 45°C	80 - 85% 70 - 78%	1587.9
5.	Sonepur	24°49.2'N	86°70.0'E	05 Building facades 02 Monuments	26 - 27°C 33 - 45°C	80 - 85% 75 - 77%	1443.5

**Fig 1.** Map of Odisha state showing the sampling districts of western Odisha, India.

from five different localities of Western Odisha i.e., Balangir, Bargarh, Nuapada, Sambalpur and Sonepur. We also made a comparative distributional analysis of subaerial cyanobacteria from other regions of Odisha to understand the guiding force behind their distribution.

MATERIALS AND METHODS

Sampling

A total of 150 subaerial biofilm samples were collected from 55 different sampling sites including 35 building facades and 20 stone monuments, covering five districts of Western Odisha namely Balangir, Bargarh, Nuapada, Sambalpur and Sonepur (Table 1; Fig.1-2). The two different habitats have chosen as a study area based on their construction materials to assess the diversity of the

cyanobacteria growth. Most of the sampling was made during post-monsoon period, as the biofilms flourish maximum diversity after getting soaked with monsoon rain. The biofilms were collected by gentle scraping the surface with sterile scalpel and following non-destructive method using adhesive tape strips (La Cono and Urzi, 2003) for ensuring to remove a portion of the substrate to study thoroughly of the biofilm and were stored in the zipped polythene bag, and brought to the laboratory for further analysis. The geographical locations of the study sites and environmental parameters like temperature, rainfall and relative humidity are broadly studied and represented in (Table 1). The atmospheric temperature and relative humidity were measured on the spot. Average annual rainfall data of the sampling districts were collected from Odisha Rainfall Monitoring System.



Fig 2. Photographs show the microbial biofilms on the exterior surface of monuments and building facades. **A.** on brick; **B–C.** on cement; **D.** on concrete; **E.** on limewashed; **F–G.** on painted surface and **H–I.** on stones. Arrow (↑) marked indicates to show the growth of cyanobacterial biofilms.

Microscopy and identification of cyanobacteria

The collected microbial biofilms or crusts were soaked in sterile distilled water and incubated under the fluorescent light for up to 72 hours and observed microscopically. Since, no morphological features were distinct even after prolonged soaking (up to 72hrs), a small amount of each sample was transferred to agar plates (1.5% agar) with BG-11 medium with or without nitrogen (Rippka *et al.*, 1979). Cultures were incubated at $28 \pm 1^\circ\text{C}$ under the continuous white fluorescent light at an intensity of 3000 lux for up to 14 days (Ferris and Hirsch, 1991). Microscopy and photography of the organisms were made using a Lawrence & Mayo trinocular research compound microscope fitted with a camera. The morphology of all the organisms was studied thoroughly at different growth stages. Basing on their phenotypic characterization, the cyanobacteria were identified

following standard monographs and literature (Desikachary, 1959; Komárek and Anagnostidis, 1986, 1988, 1989, 1999, 2005; Komárek and Hauer, 2013).

Calculation of biodiversity indices

Different biodiversity indices such as biodiversity percentage, species diversity, species richness, dominance and evenness were calculated following standard methods.

Percent abundance (Verberk, 2011)

The percent abundance of species was calculated as follow - $\frac{Y}{X} \times 100$

(Where, X = Total number of isolates, Y = Number of isolates belonging to a particular).

Species richness index/Margalef's index (Margalef, 1958)

Margalef's index was calculated as follow –

$$\frac{(S - 1)}{\ln N}$$

(Where, S = total number of species, N = total number of individuals in the sample and \ln = natural logarithm)

Species diversity index

Species diversity is the number of different species that are represented within a community or in each habitat (in this investigation, building facades and monuments).

Different diversity indices were studied as follows -

(i) Shannon-Weiner Index (Shannon and Wiener, 1949)

$$H = -\sum p_i \ln p_i$$

(Where $p_i = S/N$, S = number of individuals of one species, N = total number of all individuals in the sample and \ln = logarithm to base)

(ii) Simpson's Diversity Index (Simpson, 1949)

$$1 - D = 1 - \sum (p_i)^2$$

(Where, p_i = total number of strains of genus i /total number of all strains)

Species Dominance index/Simpson's Dominance (Simpson, 1949)

Dominance indices is a measure towards the abundance of the commonest species within a community or a given habitats. The species dominance indices in the study sites were studied as follow -

Simpson's Dominance (D)

$$D = \sum (p_i)^2$$

(Where, D = Simpson dominance index, p_i = proportion of species in a community or habitats)

Species evenness index/Pielou's evenness index (Pielou, 1966)

Pielou's evenness index: $e = \frac{H}{\ln S}$ (Where, H = Shannon - Wiener diversity index, S = total number of species in the sample and \ln = natural logarithm)

Similarity index/Sorensen's coefficient (Sorensen, 1948)

The Sorensen's coefficient, for two different sites (site A: Building facades, site B: Monuments) were studied as follow:

$$\text{Sorensen's coefficient index: } SC = \frac{2a}{2a+b+c}$$

(Where, a = number of species common in two habitats, b = number of species present in habitat B but absent in habitat A, c = number of species present in habitat A but absent in B)

RESULTS AND DISCUSSION

A total of fifty cyanobacterial taxa were identified from the subaerial samples collected from 55 localities comprised of 35 building facades and 20 monuments from five districts, Balangir, Bargarh, Nuapada, Sambalpur, Sonepur of western Odisha which are

represented as microphotographs in fig. 3–4 and details are documented as asterisk (*) in table 2. While studying the environmental factors like temperature, relative humidity including annual rainfall (Table 1), it observed to be with temperature range optimum between 26–33°C and relative humidity $\geq 77\%$ in building facades, whereas, in monuments temperature range within 29 - 45°C and relative humidity $\leq 65\%$. The overall rainfall of five districts studied range within 1229.4–1587.9mm (Table 1). Morphologically, we studied that order Nostocales showed highest percent abundance of species in both the different substrata (Fig. 5). A majority number of taxa (nearly 50%) colonizing on both the substrates were filamentous heterocystous forms (25 taxa), followed by coccal forms (13 taxa) and filamentous non-heterocystous cyanobacteria (12 taxa) are depicted in Fig. 6. However, these results highlighted the growth and distribution pattern of cyanobacteria in various substrata shield against external stress proving to be a decisive evolutionary selection advantage. Moreover, the presence of higher coccoid and filamentous forms of cyanobacteria with thick sheath layer/or mucilaginous sheaths around their trichomes and densely pigmented cells, and secondary metabolites further emphasis the fact that these forms are more adjustable in extreme environments as it serves to protect the cells from desiccation, high solar irradiation, invasion of pathogens and toxic or harmful substances, as the stress-tolerant dominant colonizers on subaerial biofilms (Drovac-cik, *et al.*, 2007; Ferrari *et al.*, 2015; Pinna, 2021). Out of these, 17 taxa were recorded from the monuments and 45 taxa from the building facades. However, only 12 taxa occur commonly on both the substrates (Table 2). Our findings support the earlier hypothesis (Tripathy *et al.*, 1999; Samad and Adhikary, 2007, 2008; Sahu *et al.*, 2011; Rossi *et al.*, 2012; Keshari and Adhikary, 2014; Adhikary *et al.*, 2015; Pradhan *et al.*, 2018) of differential microbiota between stone monuments and building facades.

During the study of different diversity indices such as, Margalef's index (Species richness), Shannon-Weiner's index (species diversity), Simpson's diversity index (community diversity), Simpson's dominance index (Dominance), Pielou's index (Evenness) and Sorensen's coefficient (Similarity index) etc., along with the impact of two environmental factors such as temperature and relative humidity were analyzed. Different diversity indices were analyzed to know the distribution patterns of species in two distinct substrata (Monuments and Building facades) are graphically represented in the (Fig. 7). The results based on Margalef's index showed that the monuments with the highest species richness value 16.6, while building facades showed the lowest value with 12.89 in richness. Similarly, based on Shannon-Weiner and Simpson's diversity indices in building facades showed the highest diversity index (3.80, 0.98) and the lowest was found in monuments (2.78, 0.96). Whereas,

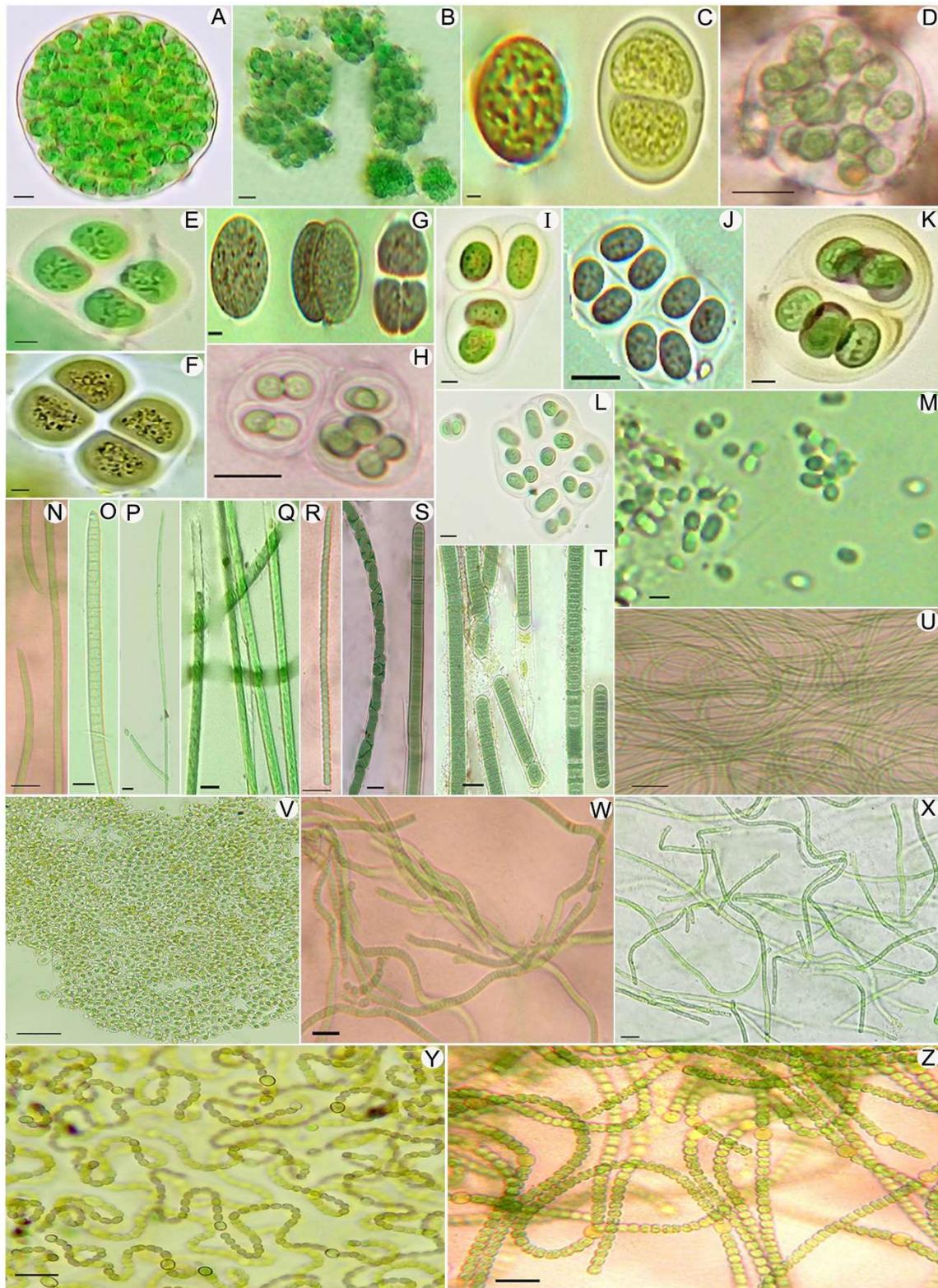


Fig 3. Photomicrographs of various subaerial cyanobacteria strains. **A.** *Chroococcidiopsis kashayi*; **B.** *Gloeopcapsopsis crepidinum*; **C.** *Chroococcus indicus*; **D.** *Limnococcus limneticus*; **E-F.** *Chroococcus minor*; **G.** *Chroococcus turgidus*; **H.** *Cyanosarcina spectabilis*; **I.** *Chroococcus schizodermaticus*; **J.** *Gloeocapsa biformis*; **K.** *Gloeotheca rupestris*; **L.** *Aphanothece pallida*; **M.** *Gloeocapsa kuetzingiana*; **N.** *Kamptomena animale*; **O.** *Microcoleus autumnalis*; **P.** *Phormidium corium*; **Q.** *Phormidium mollis*; **R.** *Phormidium retzii*; **S.** *Phormidium stagninum*; **T.** *Lyngbya calcarea*; **U.** *Leptolyngbya tenuis*; **V.** *Gloeotheca palea*; **W.** *Leptolyngbya nostocorum*; **X.** *Plectonema putaele*; **Y.** *Nostoc commune* and **Z.** *Nostoc linckia*. Scale bars (A–Z) - 10µm.

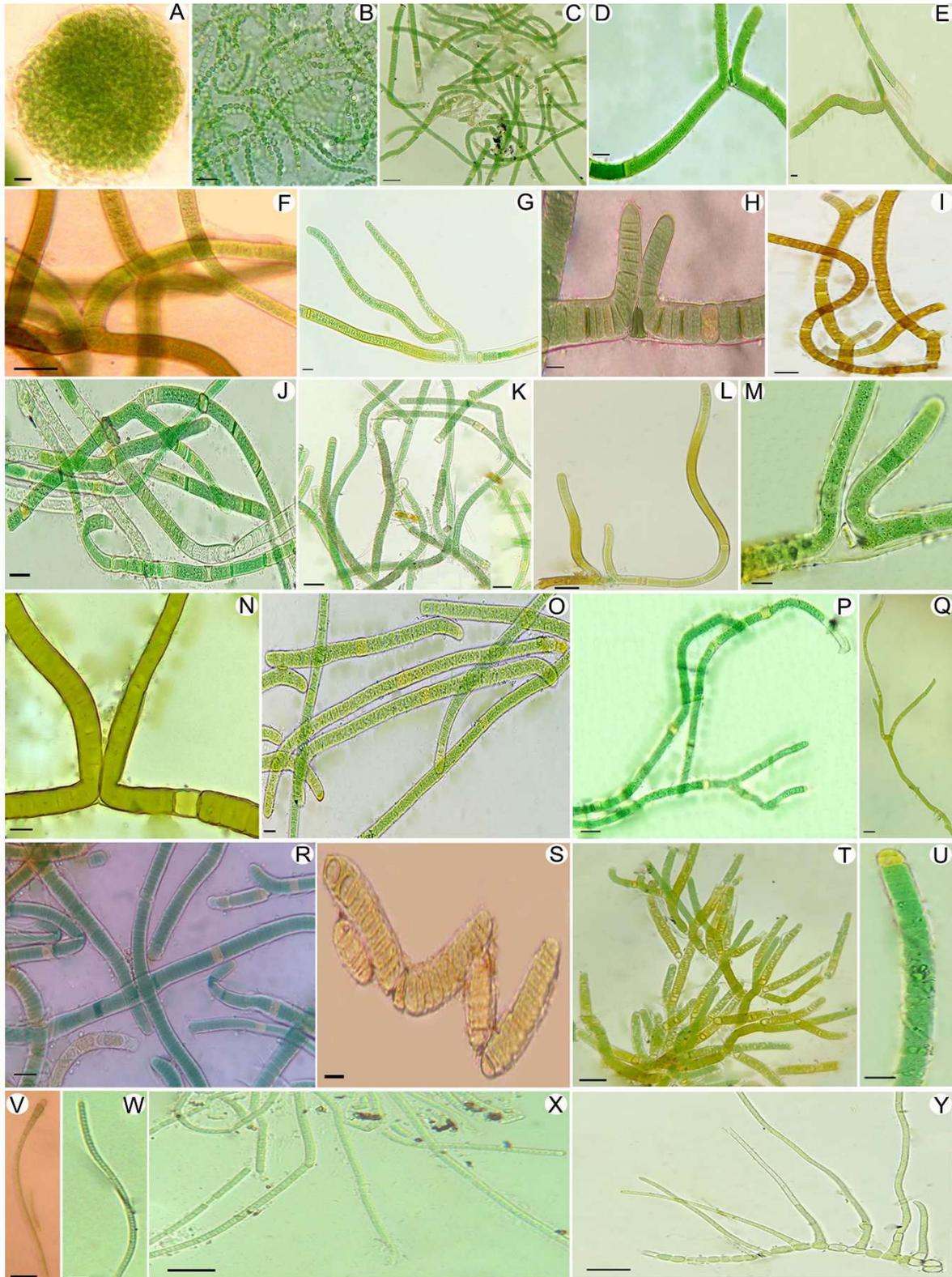


Fig 4. Photomicrographs of various subaerial cyanobacteria strains. **A.** *Nostoc microscopicum*; **B.** *Nostoc punctiforme*; **C.** *Scytonema crispum*; **D.** *Scytonema geitleri*; **E.** *Scytonema guyanense*; **F.** *Scytonema hofmanni*; **G.** *Scytonema hyalinum*; **H.** *Scytonema millei*; **I.** *Scytonema multiramosum*; **J.** *Scytonema wolleanum*; **K.** *Scytonema ocellatum*; **L.** *Scytonema punctatum*; **M.** *Scytonema rivulare*; **N.** *Scytonema tolypothrichoides*; **O.** *Tolypothrix distorta*; **P.** *Tolypothrix rechingeri*; **Q.** *Tolypothrix tenuis*; **R.** *Tolypothrix scytonematoides*; **S.** *Hassallia byssoidea*; **T.** *Hassallia bouteilli*; **U.** *Calothrix aequalis*; **V.** *Calothrix fusca*; **W.** *Leptolyngbya fragilis*; **X.** *Leptolyngbya notata* and **Y.** *Hapalosiphon welwitschii*. Scale bars (A–Y) - 10µm.



Table 2. Comparative analysis of the occurrence of different genera of cyanobacteria in the subaerial biofilmson exterior surface of buildings and temples in four different zone of tropical region of Odisha, India. (* Taxa documented in the present study)

Sl. no.	Cyanobacterial taxa	Monuments	Building facades	Distribution in different parts of Odisha				References
				Eastern	Western	Northern	Southern	
Order: Chroococciopsidales								
1	<i>Chroococciopsis</i> cf. <i>crepidinum</i>	+		+				7
2	<i>Chroococciopsis cubana</i> Komárek & Hindák	+			+			6, 8
3	<i>Chroococciopsis indica</i> Desikachary	+		+				12
4	<i>Chroococciopsis kashayi</i> Friedmann*	+	+		+		+	10
5	<i>Gloeocapsopsis crepidinum</i> (Thuret) Geitler ex Komárek*		+	+	+			1, 6, 10
6	<i>Gloeocapsopsis dvorakii</i> (Nováček) Komárek & Anagnostidis ex Komárek	+		+				1, 1
7	<i>Gloeocapsopsis pleurocapsoides</i> (Nováček) Komárek & Anagnostidis ex Komárek	+			+			6
Order: Pleurocapsales								
8	<i>Chroococcopsis fluviatilis</i> (Lagerheim) Komárek & Anagnostidis		+				+	10
Order: Chroococcales								
9	<i>Asterocapsa divina</i> Komárek	+	+	+	+			6, 8, 10
10	<i>Chroococcus indicus</i> Zeller*	+	+		+		+	6, 10
11	<i>Chroococcus lithophilus</i> Ercegović		+	+				10
12	<i>Chroococcus minor</i> (Kützing) Nägeli*		+	+	+			6, 10
13	<i>Chroococcus pallidus</i> Nägeli		+	+				10
14	<i>Chroococcus schizodermaticus</i> West*		+	+	+			10
15	<i>Chroococcus turgidus</i> (Kützing) Nägeli*		+		+			8
16	<i>Chroococcus varius</i> A. Braun		+	+				10
17	<i>Limnococcus limneticus</i> (Lemmermann) Komárková et al.*	+	+		+		+	10
18	<i>Cyanosarcina burmensis</i> (Skuja) Kováčik	+			+			6
19	<i>Cyanosarcina parthenonensis</i> Anagnostidis		+				+	10
20	<i>Cyanosarcina spectabilis</i> (Geitler) Kováčik*		+	+	+		+	6, 10, 12
21	<i>Cyanosarcina thermalis</i> (Hindák) Kováčik	+		+				1
22	<i>Gloeocapsa atrata</i> Kützing		+	+				10
23	<i>Gloeocapsa biformis</i> Ercegović*	+	+		+			@
24	<i>Gloeocapsa caldariorum</i> Rabenhorst	+		+				1
25	<i>Gloeocapsa kuetzingiana</i> Nägeli ex Kützing*		+	+	+			1, 6, 7, 10
26	<i>Gloeocapsa livida</i> (Carmichael) Kützing		+	+				10
27	<i>Gloeocapsa muralis</i> Kützing		+	+				10
28	<i>Gloeocapsa punctata</i> Nägeli	+	+	+				1, 10
29	<i>Gloeocapsa rupestris</i> Kützing		+				+	10
30	<i>Gloeocapsa sanguinea</i> (C. Agardh) Kützing		+				+	10
31	<i>Gloeotheca palea</i> (Kützing) Nägeli*		+		+			@
32	<i>Gloeotheca rhodochlamys</i> Skuja	+	+	+	+		+	5, 6, 10
33	<i>Gloeotheca rupestris</i> (Lynýbye) Bornet*		+	+	+			6, 10
34	<i>Aphanothece pallida</i> (Kützing) Rabenhorst*		+		+			@
35	<i>Aphanothece Saxicola</i> Nägeli	+			+			6
36	<i>Aphanothece stagnina</i> (Sprengel) A. Braun		+	+				10
Order: Oscillatoriales								
37	<i>Arthrospira massartii</i> Kufferath	+			+			6
38	<i>Cyanothece aeruginosa</i> (Nägeli) Komárek		+	+				10
39	<i>Geitlerinema numidicum</i> (Gomont) Anagnostidis		+				+	4,
40	<i>Kamptomena animale</i> (C. Agardh ex Gomont) Strunecký et al.*		+		+			@
41	<i>Microcoleus autumnalis</i> (Gomont) Strunecký et al.*	+		+	+			1, 10
42	<i>Microcoleus paludosus</i> Gomont		+	+				10
43	<i>Phormidesmis mollis</i> (Gomont) Turicchia et al.*	+	+		+			@
44	<i>Phormidium corium</i> Gomont ex Gomont*	+	+		+			@
45	<i>Phormidium insigne</i> Anagnostidis	+		+				1
46	<i>Phormidium kuetzingianum</i> (Kirchner ex Hansgirg) Anagnostidis & Komárek	+		+				2



47	<i>Phormidium retzii</i> Kützing ex Gomont*	+	+	+	+	+	1, 9
48	<i>Phormidium roseum</i> (Batters) Anagnostidis	+		+			1
49	<i>Phormidium stagninum</i> Anagnostidis*		+		+		@
50	<i>Phormidium truncicola</i> S. L. Ghose	+		+			1, 12
51	<i>Plectonema puteale</i> Kirchner ex Hansgirg*		+	+	+		10, 12
52	<i>Plectonema tomasinianum</i> Bornet ex Gomont	+		+			1
53	<i>Plectonema</i> sp.	+		+			7
54	<i>Potamolinea aerugineocaeerulea</i> (Gomont) M. D. Martins & L. H. Z. Branco	+	+	+			1, 2, 10
55	<i>Pseudophormidium batrachosperma</i> (Starmach) Anagnostidis & Komárek	+		+			1
56	<i>Pseudophormidium hansgirgii</i> (Schmidle) Anagnostidis & Komárek	+		+			12
57	<i>Pseudophormidium hollerbachianum</i> (Elenkin) Anagnostidis	+		+			1
58	<i>Pseudophormidium radiosum</i> (Gomont) Anagnostidis & Komárek	+		+			1
59	<i>Porphyrosiphon ceylanicus</i> (Wille) Anagnostidis & Komárek	+		+			1, 10
60	<i>Lyngbya calcarean</i> (Tilden) Symoens*		+		+		@
61	<i>Lyngbya corticicola</i> Brühl & Biswas	+		+			1, 12
Order: Nostocales							
62	<i>Nostoc calcicole</i> Bréb. ex Kützing		+			+	4
63	<i>Nostoc carneum</i> C. Agardh ex Bornet & Flahault	+		+			1, 2, 12
64	<i>Nostoc commune</i> Vaucher ex Bornet & Flahault*		+	+	+	+	1, 2, 3, 10, 11
65	<i>Nostoc ellipsosporum</i> Rabenhorst ex Bornet & Flahault	+		+			1, 12
66	<i>Nostoc linckia</i> Bornet ex Bornet & Flahault*		+	+	+		1, 2, 10
67	<i>Nostoc microscopicum</i> Carmichael ex Bornet & Flahault*	+	+	+	+		1, 10
68	<i>Nostoc pruniforme</i> C. Agardh ex Bornet & Flahault		+			+	4
69	<i>Nostoc punctiforme</i> Hariot*		+	+	+	+	1, 2, 3, 8, 10, 11, 12
70	<i>Nostoc verrucosum</i> Vaucher ex Bornet & Flahault	+		+			1
71	<i>Desmonostoc muscorum</i> (Bornet & Flahault) Hrouzek & Ventura	+		+			1, 12
72	<i>Dolichospermum circinale</i> (Rabenhorst ex Bornet & Flahault) Wacklin <i>et al.</i>		+			+	11, 13
73	<i>Scytonema bohneri</i> Schmidle		+	+			10
74	<i>Scytonema coactile</i> Montagne ex Bornet & Flahault	+				+	9
75	<i>Scytonema crispum</i> Bornet ex De Toni*	+	+		+		@
76	<i>Scytonema geitleri</i> Bharadwaja*	+	+	+	+		10
77	<i>Scytonema guyanense</i> Bornet ex Flahault*		+		+		@
78	<i>Scytonema hoffmannii</i> Agardh*		+	+	+	+	1, 10
79	<i>Scytonema hyalinum</i> Gardner*	+			+		@
80	<i>Scytonema millei</i> Bornet ex Bornet & Flahault*		+	+	+		10
81	<i>Scytonema multiramosum</i> Gardner*		+	+	+		1, 10
82	<i>Scytonema ocellatum</i> (Dillwyn) Lyngbye ex Bornet & Flahault*		+	+	+		1, 10
83	<i>Scytonema pseudoguyanense</i> Bharadwaja	+		+			1, 10
84	<i>Scytonema pseudopunctatum</i> Skuja*		+		+		@
85	<i>Scytonema rivulare</i> Borzi ex Bornet & Flahault*		+		+		@
86	<i>Scytonema tolypothrichoides</i> Kützing ex Bornet & Flahault*		+		+		@
87	<i>Scytonema wolleanum</i> Forti*		+		+		@
88	<i>Tolypothrix arenophila</i> West & G. S. West	+		+			5
89	<i>Tolypothrix campylonemoides</i> S. L. Ghose	+		+			12
90	<i>Tolypothrix crassa</i> West & G. S. West	+		+			12
91	<i>Tolypothrix distorta</i> Kützing ex Bornet & Flahault*		+		+		@
92	<i>Tolypothrix fragilis</i> (N. L. Gardner) Geitler		+	+			10
93	<i>Tolypothrix penicillate</i> Thuret ex Bornet & Flahault	+		+			1, 2, 12
94	<i>Tolypothrix rechingeri</i> (Wille) Geitler*	+	+	+	+		12



95	<i>Tolypothrix scytonematooides</i> Gardner*		+	+	+			1, 2, 12
96	<i>Tolypothrix tenuis</i> Kützing ex Bornet & Flahault*	+			+	+		5
97	<i>Hassallia bouteillei</i> Bornet & Flahault*			+		+		@
98	<i>Hassallia byssoidea</i> Hassall ex Bornet & Flahault*		+		+			1, 2, 12
99	<i>Calothrix braunii</i> Bornet & Flahault			+			+	4
100	<i>Calothrix fusca</i> (Kützing) Bornet & Flahault*	+			+	+		10
101	<i>Calothrix ghosei</i> Bharadwaja	+			+			12
102	<i>Calothrix marchica</i> Lemmermann*			+		+		@
103	<i>Calothrix marchica</i> var. <i>crassa</i> C. B. Rao	+			+			1, 12
104	<i>Calothrix parietina</i> Thur. ex Bornet & Flahault			+			+	4
105	<i>Calothrix scytonemicola</i> Tilden	+			+			12
106	<i>Dichothrix baueriana</i> Bornet & Flahault	+			+			1, 2
107	<i>Hapalosiphon hibernicus</i> West & G. S. West	+				+		6
108	<i>Hapalosiphon stuhlmannii</i> Hieronymus	+			+			12
110	<i>Hapalosiphon welwitschia</i> West & G. S. West*		+			+	+	4, 6
111	<i>Fischerella</i> sp. 1	+				+		6
112	<i>Fischerella</i> sp. 2	+				+		6
113	<i>Fischerella major</i> Gomont	+				+		6
114	<i>Fischerella muscicola</i> Gomont	+			+			12
115	<i>Fischerella tenuis</i> De Toni	+	+		+			5, 11
116	<i>Westiellopsis iyengarii</i> Jeeji Bai	+				+		6
117	<i>Westiellopsis prolifica</i> Janet	+	+		+	+		6, 9
118	<i>Stigonema tomentosum</i> Hieronymus	+			+			1, 10
Order: Synechococcales								
119	<i>Aphanocapsa benaresensis</i> Bharadwaja	+				+		6
120	<i>Leptolyngbya</i> sp.	+			+			7
121	<i>Leptolyngbya boryana</i> (Gomont) Anagnostidis & Komárek	+	+		+			1, 10
122	<i>Leptolyngbya foveolara</i> (Gomont) Anagnostidis & Komárek			+			+	4
123	<i>Leptolyngbya fragilis</i> (Gomont) Anagnostidis & Komárek*	+	+		+	+		1, 8, 10
124	<i>Leptolyngbya gracillima</i> (Hansgirg) Anagnostidis & Komárek	+			+			12
125	<i>Leptolyngbya nostocorum</i> (Bornet ex Gomont) Anagnostidis & Komárek*		+		+	+		1, 10
126	<i>Leptolyngbya notata</i> (Schmidle) Anagnostidis & Komárek*	+			+	+		10
127	<i>Leptolyngbya tenuis</i> (Gomont) Anagnostidis & Komárek*	+	+			+		@
128	<i>Schizothrix fragilis</i> Gomont	+			+			1
129	<i>Schizothrix lateritia</i> Gomont	+	+		+			1, 10
130	<i>Synechococcus elongatus</i> (Nägeli) Nägeli	+				+		6

*+ in bold indicates the cyanobacteria species documented for the first time from the subaerial habitats of Odisha. **Reference:** 1. Adhikary *et al.* 2015. 2. Keshari & Adhikary 2014. 3. Keshari *et al.*, 2015. 4. Kumar *et al.*, 2018. 5. Pattanaik & Adhikary, 2002. 6. Pradhan *et al.*, 2018. 7. Rossi *et al.*, 2012. 8. Sahu *et al.*, 2011. 9. Samad & Adhikary, 2007. 10. Samad & Adhikary 2008. 11. Sethi *et al.*, 2012. 12. Tripathy *et al.*, 1999. 13. Samad *et al.*, 2008 @: Documented for the first time from the subaerial habitats of Odisha

species dominance was found to be highest in monuments and lowest in building facades (0.04, 0.02). However, the evenness was recorded lowest in monuments and highest in building facades (0.71, 0.97). Further, the Sorensen's coefficient (SC) for similarity index value of two different substrata (0.438) revealed that there was low similarity index. In agreement to our observations, many studies (Cámara *et al.*, 2014; Ferrari *et al.*, 2015; Noeiaghahi *et al.*, 2017; Del Mondo *et al.*, 2018; Jacob *et al.*, 2018; Urzi *et al.*, 2018) have been focused on porosity, surfaces roughness and chemical composition of the building construction's material determined

bioreceptivity with response to external environmental factors like light intensity, temperature and relative humidity. Recent studies (Gaylarde, 2020; Pinna, 2021) have shown that the substrata of buildings are more influenced by its high porosity and surface roughness due to direct exposed to sunlight's, high level of relative humidity and optimum temperature range, which provides a preferential site for the growth and abundance of phototrophic organisms. Whereas in case of stone substrata, due to seasonal variation altogether show low porosity and relative humidity with extreme temperature make it less favourable. Ortega-Morales *et al.*, (2019)

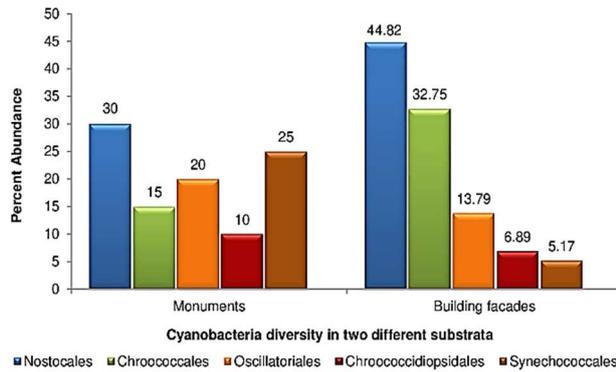


Fig. 5. Percent abundance (%) of species under different orders of subaerial cyanobacteria in two different substrata from western Odisha.

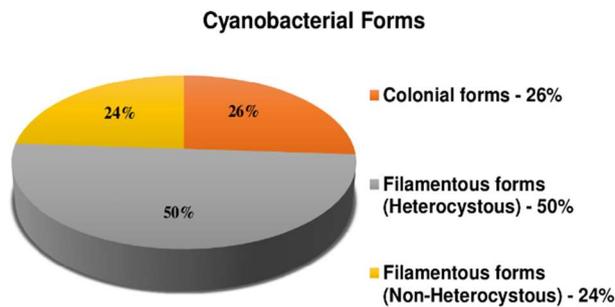


Fig. 6. Micrographs showing the different morphological forms of subaerial cyanobacterial studied and recorded from western Odisha.

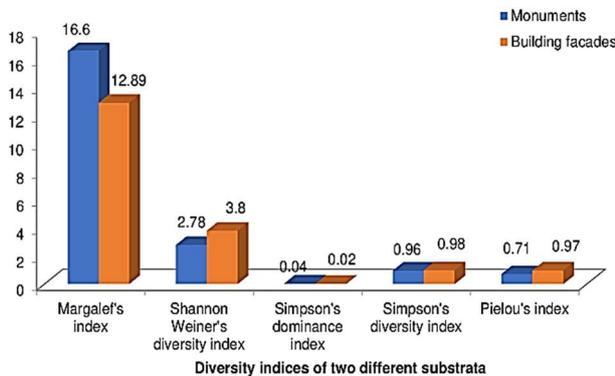


Fig. 7. Micrographs showing the diversity indices of the occurrence of subaerial cyanobacterial studied and recorded from western Odisha.

concludes that cyanobacteria growth and abundance are seen within the annual range of rainfall 1281.7mm per year. Most of the cyanobacteria biomass on different substrata is attributed to the more or less content of water absorption and the level of relative humidity (Miller *et al.*, 2009). Therefore, it can be inferred that the ideal and unique physical properties like high light intensity and relative humidity, optimum temperature, surface structure and rain fall regulates the colonization and diversity of cyanobacteria on building facades and stone monuments corroborates with our observations during this

investigation. All these parameters have been overall broadly studied as described in methodology section and represented in table 1.

Comparative distributional analysis of subaerial cyanobacteria from Odisha

The exploration of subaerial cyanobacteria from the heritage and cultural monuments along with the building facades of Odisha is only two decades old. So far, 130 cyanobacterial taxa (including 129 species and 1 infra-specific taxon) belonging to 38 genera and 6 orders were documented from these specialized substrates including the present study (Table 2). Out of these, the majority of taxa was documented from various monuments of Odisha (90 taxa) and the survey of building facades identified 78 taxa. For our comparative analysis we have divided Odisha into 4 different zones, i.e., eastern, western, northern and southern and the distribution of subaerial cyanobacteria in these zones are presented in Table 2. The analysis corroborates the pattern of common occurrence as that in western Odisha. Nearly half of the taxa were substrate specific, i.e., 56.6% of the cyanobacteria from monuments and 50% from building facades. Only 30 % of taxa exhibited commonality in their occurrences. These were exclusively subaerial species found in terrestrial habitats in India, like *Chroococcidiopsis kashayi*, *Asterocapsa divina*, *Gloeocapsa kuetzingiana*, *Nostoc commune*, *Nostoc linckia*, *Nostoc microscopicum*, *Nostoc punctiforme*, *Scytonema hoffmannii*, *Scytonema ocellatum*, *Tolypothrix tenuis*, *Hassallia byssoidea*, *Westiellopsis prolifica*, etc. The zone-wise distributional pattern of the cyanobacterial diversity in subaerial habitats revealed the sparse exploration in both northern and southern districts of Odisha. The monuments and building facades of eastern and western Odisha were surveyed by multiple researcher's time to time. However, it is noteworthy to mention that the present study identified 18 cyanobacterial taxa as new distributional record from sub-aerophytic habitats of Odisha, sampled from western Odisha (Table 2). The diversity in eastern zone districts is represented by 81 taxa, followed by 65 from western, 14 from northern and 10 from southern Odisha. No commonality in distribution was observed among all these zones. *Cyanosarcina spectabilis* and *Gloeotheca rhodochlamys* were the only species with common occurrence in eastern, western and northern Odisha. Likewise, *Phormidium retzii*, *Nostoc commune*, *Nostoc punctiforme*, *Scytonema hoffmannii* and *Westiellopsis prolifica* were commonly distributed in eastern, western and southern Odisha.

The lithobiontic cyanobacteria forming biofilms on the light exposed surfaces of monuments and building facades are resistant to high irradiance, UV radiation, high temperature, drought and desiccation stresses. Further eco-physiological assessment of these organisms will provide more insight into their stress tolerance and



adaptability. So, the prime objective of this study was not only to identify extremophilic cyanobacteria with further biotechnological potential, but also to develop cognizance among researchers and policy makers for conservation of these culturally important monuments from biodeterioration.

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