

Biodiversity of Cyanoprokaryota from Monuments of Western Odisha, India-I (Chroococcales and Stigonematales)

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Abstract

Monuments are remarkably adequate sample of unbelievable artistry and guardian pillars of India's cultural heritage. Western Odisha, India has several incredible monuments like temples, palaces, forts built during the 7th -17th century now became discoloured and deteriorated due to colonisation of different group of microorganisms. In this study an attempt has been taken to find out the diversity of Cyanoprokaryota Chroococcales and Stigonematales. 14 popular temple were chosen as study sites. Cyanobacterial samples were collected seasonally, cultured and identified. Among 22 isolated cyanobacterial taxa, 15 are of Chroococcales and 07 from Stigonematales were found to occur on lithic surfaces of temples.

Keywords: Cyanoprokaryota, diversity, Chroococcales, Stigonematales monuments

Introduction

Orissa, a land of great cultural heritage is home to numerous monuments of historical importance more than two thousand years back. Stone monuments, statues and historic buildings are exposed to the effects of physical, chemical and biological deteriorating factors. Stone works of art can be colonized by different groups of micro-organisms, including bacteria, cyanobacteria, algae and fungi. According to several authors, cyanoprokaryota (cyanobacteria) and chlorophyta (green algae) are the pioneering inhabitants in the colonization of stone (Ortega-Calvo *et al.*, 1991; Tiano *et al.*, 1995;; Lamenti *et al.*, 2000; Cecchi *et al.*, 2000; Tomaselli *et al.*, 2000b; Crispim & Gaylarde, 2005).

Cyanoprokaryota forms blackish brown colouration to the exposed rocks (Tomaselli *et al.*, 2000; Crispim *et al.*, 2003; Welton *et al.*, 2003; Barberosse *et al.*, 2006; Crispim *et al.*, 2006; Cappitelli *et al.*, 2012). In the tropical countries epilith and/or endolith communities form pre-dominant flora throughout the year including midsummer months (Anagostidis *et al.*, 1983; Ortega-Calvo *et al.*, 1993b;). Reports are also there about the cyanobacterial growth inside desert rocks, in both hot and cold deserts (Friedmann and Ocampo, 1976; Friedmann, 1982). Lithophytic strains of cyanoprokaryota were reported from many regions of world (Ortega-Calvo *et al.*, 1993a, 1993b; Ortega-Morales *et al.*, 2005; Adhikary and Kovacic, 2010; Rossi *et al.*, 2012). Generally they grow on bare rock surfaces or within the rock substrates, expanding to a few mm below the rock surface. They occur in extreme climate like hot and cold deserts, semi-deserts and also in temperate climates. They are seen in extreme micro-habitats like steep rock surface in the high alpine regions. In this manuscript we present a systematic account of observed unicellular (Chroococcales) and branching Cyanoprokaryota (Stigonematales) occurring on the rock surface of 14 different temples and ancient monuments of western Odisha, India

Materials and Methods

Study sites

Samples were collected from 14 historically important temples (Fig.2.) of 8 districts of Western Odisha (Table 1)

Table 1. Temples from which samples were collected

STUDY SITE	NAME OF THE TEMPLE	GEOGRAPHIC LOCATION	DESCRIPTION
I	Sri NrusinghaNath Temple	20.92°N 82.81°E	This great historic temple built by the king of Patnagarh, Baijal Singha Dev in 13 century A.D. situated at the foot hills of Gandhamardan Hill near Paikmal of Bargarh district. This temple is built of sand stone but the statues are of chlorite stone.
II.	Gandharadi temple	84.01°N 20.52°E	The Gandharadi twin temple is made up of sand stone and has a wheel of blue-chlorite at the top (Sikhara), situated at a distance of about 16km from Boudh at Jagati. This temple is also locally known as "Chari Sambhu Mandir", constructed under the patronage of the Bhanja ruler of Khinjali Mandala in 9 th century A.D.
III	Ramanath temple/Rameswar temple	20.83°N 84.31°E	A mid-9th century A.D. temple at Boudh town built of red sandstone & is profusely carved.
IV	Chausathi Yogini Temple	20.39°N 83°E	This is a circular open air shrine (also, known as Soma-Tirtha) made up of sand stone at Ranipur-Jharial in the south Koshala region, 35 km away from Titilagarh of Bolangir district. This shrine has been built in between 9 th -10 th century A.D. by Somabanshi ruler King Sameswar Gupta. This sand stone temple is carved with 64 niches in its interior wall.
V	Padmasini Temple	21.69°N 83.63°E	This temple is situated in the Hirakud submerged area of Padmapur. This temple was built in 7th century A.D. by a Chalukya King of South and reconstructed by the Chauhan King of 16th century. This temple is an unique example of chalukya art and architecture. From here two silalekha has been recently discovered.
VI	Indralath Temple	20.28°N 82.97°E,	Also in Ranipur-Jharial there is another temple (tallest structure in the whole of South Koshal) of red bricks. It is the only surviving brick structure of Tel basin.
VII	Kedarnath temple	30.73°N 79.07°E	An ancient stone temple of Chauhan architecture, constructed between 11th- 12th century is situated at Ambabhona, about 40 km away from Bargarh town.
VIII	Maa Lankeswari temple	19. 86°N 82.94°E	This temple is situated at Junagarh, Kalahandi district of Odisha state. This ancient temple is worshiped by Junagarh naga clan, having legendary deity of war, Lankeswari inside. Temple deity is made of black chlorite stone.
IX	Bhalugarh, Shiva temple	22.51°N 83.55°E	Temple with fine architect carved over stone, geographically situated towards northeast of Sundergarh.

X	Balunkeswar temple	21.26°N 83.66°E	It is an ancient shrine situated at Gaisima, on the bank of Jira river, about 13 km away from Bargarh town.
XI	Visweswar Temple	21.39°N 83.81°E	This temple is situated at Soranda of Bargarh District.
XII	MandhataBaba Temple	21.41°N 84.02°E	This temple is situated at Maneswar, 8 km away from Sambalpur city.
XIII.	Rambha temple	21.53°N 84.71°E	A small cave temple of goddess Rambha Devi, positioned at 8 km away from Deogarh town.
XIV	Bimaleswar temple	21.29°N 83.92°E,	This Leaning Temple built in 17th century is situated at Huma, about 25 km away from the Sambalpur city The main structure of the temple is leaned at 47 degree to the west of the main temple.

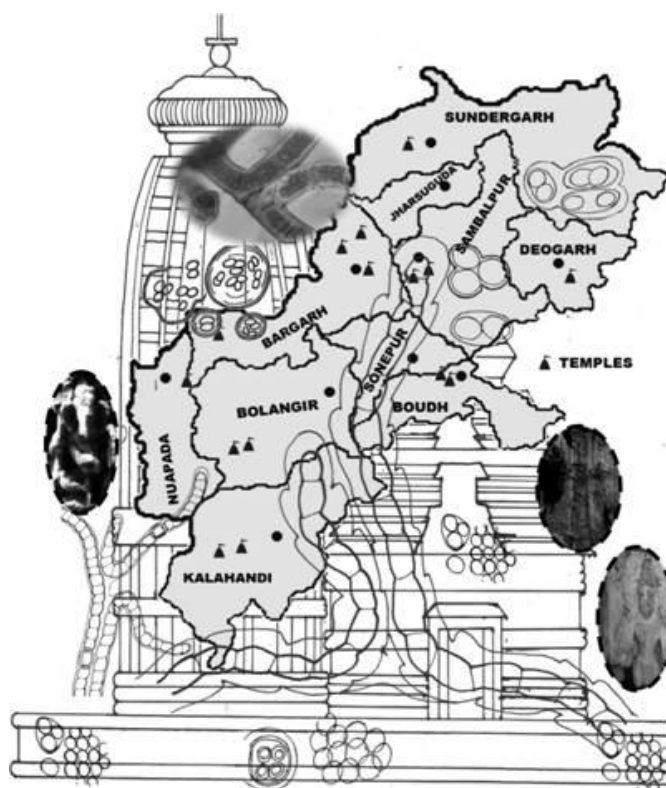


Fig 1: Location map of western Odisha in Odisha state of India

Collection, Observation, Culture and identification:

Samples were collected by scrapping from the exposed stone surface of temples and were placed in pre-sterilized screw cap Tarson bottles. Crust samples were collected from the upper surface of the stone and brick objects from varied locations (temples) of western part of Odisha during winter (September-December) and again at summer (March-May). Broken stone pieces from various study sites were put into BG 11 medium both, with and without nitrogen for emergence of endolithic cyanoprokaryotal flora. Water soaked crust samples were brought for examination under light microscope. Cyanoprokaryotal filaments could be visualized after 12-24 hours of wetting. However, for identification of cyanoprokaryota, isolation and culturing under defined conditions was done. A pinch of crust material was transferred to BG-11_{0/+} semisolid medium in parafilm-sealed Petri dishes and incubated at 25±2°C under 7.5W/m² continuous light from fluorescent tubes till visible growth of the organisms was observed (Rippka *et al.*, 1979). Unialgal

strains of cyanoprokaryota were used for detail morphometric analysis. Cyanoprokaryota were identified according to literature of Desikachary, 1959, Komarek and Anagnostidis, 1998, 2005 and Komarek, 2008.

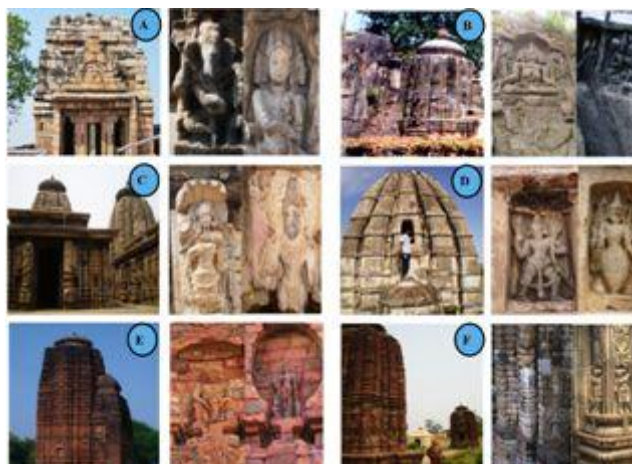


Fig 2. A. Chausathi Yogini temple; B. Sri Nrusinghanath temple; C. Gandharadi temple (Chari Sambhu); D. Padmasini temple; E. Indralath temple; F. Ramanath temple

RESULTS:

In this work 15 taxa of Chroococcales and 7 taxa of Stigonematales were found from the exposed surface of temple wall.

Systematic Enumeration of the Taxa

Chroococcales

1. *Aphanothece saxicola* Nägeli, Neue Denkschr. Allg. Schweiz. Ges. Gesamten Naturwiss. 10 (7): 60. 1849. (Plate 1, Fig. c)

Yellowish, mucilaginous thallus; cells cylindrical, 1-1.6 μm broad and 2.2-4.5 μm long,. Occurred as brown crust on sandstone and lime washed walls.

Documented from site II, V, IX, XII, XIII and XIV.

2. *Synechococcus elongatus* (Nägeli) Nägeli, Gatt. Einzell. Alg.: 56. (Plate 1, Fig. l)

Cells single or 2-4 cells together; cylindrical cells containing blue-green contents; 1.5 μm broad, 2.5-5.2 μm long. Occurred as blackish crust on bricks.

Documented from site IV, VI and XI.

3. *Cyanothece* sp. (Fig. 3 c)

Cells solitary or in groups, 6-14 μm broad, up to 24 μm long, cell content blue green.

Occurred as brownish crust on moist walls.

Documented from site I, III, V, VI, VIII, X and XIV.

4. *Gloeothece rhodochlamys* Skuja, Nova Acta R. Soc. Sc. Upsal., ser. 4, 14 (5): 18. 1949. (Pl. 1, Fig. f)

Cells rounded, 2.2-3.5 μm broad and 3-6.2 μm long; distinct envelopes, 4-16 cells forming colonies in each envelope, with envelope 9-18.5 μm broad.

Occurred as blackish crust on brick surface.

Documented from site I, V, VI and XII.

5. *G. rupestris* (Lyngbye) Bornet in Wittrock & Nordstedt, Algae aquae dulcis exsiccatae, Fasc. 7: 339. 1880. *Palmella rupestris* Lyngbye, Tent. 207, pl. 69. 1819. (Pl.1, Fig. e)

Cells ellipsoidal to cylindrical, without envelop 4-6 μm broad and 8-9 μm long, with envelop 9-13 μm broad and 14-17 μm long, blue green, normally 2-4, often 8 cells forming oval or sub-globose colony; colonies 22-36 μm in diameter, distinctly lamellated, colourless envelope.

Occurred on lime washed wall.

Documented from site II, III, V, VIII, X, XIII and XIV.

6. *Aphanocapsa banarensis* Bharadwaja, Proc. Indian Acad. Sc., sect. B, 2: 96. 1935. (Fig.3 a)

Colonies more or less spherical up to 1.5 cm in diam., spherical, 4-5.5 µm in diameter; sheath up to 1 µm thick .

Occurred as greenish patch on moist rock surface.

Documented from site I, IV, VI, IX, X and XIII.

7. *Asterocapsa divina* Komárek, Bull. Natl. Sci. Mus. Tokyo, ser. B (Bot.), 19: 33. 1993. (Fig. 3 d & Pl.1, Fig. a)

Colonies having 2-4 cells, spherical delimited by mucilaginous thick and colourless sheath, cells 6-9 µm broad and 9-11µm long.

Occurred as bluish green streak inside the sand stone and as blackish crust on cemented wall.

Documented from site II, IV, VII and X.

8. *Chroococcus indicus* Zeller, J. Asiatic Soc. Bengal 42 (2): 176. 1873. (Pl.1, Fig. d)

Thallus brownish; cells single, subspherical, 4-6.5 µm in diameter, with transparent sheath.

Occurred as dark greyish crust on cement wall and rock surface.

Documented from site I, III, V, VII, VIII and IX.

9. *C. minor* (Kütz.) Nägeli, Neue Denkschr. Allg. Schweiz. Ges. Gesamten Naturwiss. 10 (7): 46. 1849. *Protococcus minor* Kütz., Phycol. Germ.: 144. 1845. (Pl.1, Fig. g)

thallus gelatinous, cells spherical 2.8-3.5 µm in diameter, present singly or in pairs; faintly visible sheath.

Occurred as dark greenish crust on lime-washed wall and sandstone.

Documented from site I, II, V, VI, VIII, X, XI XII and XIV.

10. *Cyanosarcina burmensis* (Skuja) Kovacik, Arch. Hydrobiol. Suppl. 80 (Algol. Stud. 50 – 53): 176. 1988. *Myxosarcina burmensis* Skuja, Nova Acta R. Soc. Upsal., ser. 4, 14 (5): 21. 1949. (Pl.1, Fig. k)

Cells angular or with rounded corners, colonies in transverse or vertical series, 1.5-3.5 µm in diameter, young colonies having 4 cells, 30 µm in diameter.

Occurred as greenish crust on sandstone and cemented walls.

Documented from site III, VIII, X and XIII.

11. *C. spectabilis* (Geitler) Kovacik, Arch. Hydrobiol. Suppl. 80 (Algol. Stud. 50 – 53): 176. 1988. *Myxosarcina spectabilis* Geitler, Arch. Hydrobiol. Suppl. 12: 624. 1933. (Pl.1, Fig. i)

Colonies spherical, 9-17 µm in diameter, divided in three planes; colony consisting of variously pressed cells, 6-9.5 µm broad, distinct thin hyaline colonial sheath.

Occurred as dark greyish crust on bricks.

Documented from site II, III, IV and VI.

12. *Gloeocapsopsis pleurocapsoides* (Nováček) Komárek & Anagnostidis ex Komárek, Bull. Natl. Sci. Mus. Tokyo, ser. B (Bot.) 19: 24. 1993. *Gloeocapsa pleurocapsoides* Nováček, Arch. Svazu Ochr. Přír. a Domov. Zemi Moravskoslezské 3a: 93. 1934. (Fig. 3 b)

Brownish colonies consisting of aggregation of sheathed cells; cells sub-spherical to ellipsoidal, 4.5 -14.6 µm in diameter, lamellate, yellowish envelope.

Occurred as blackish patch on wet rocks.

Documented from site I, V, VII, VIII and XIV.

13. *Gloeocapsa kuetzingiana* Nägeli ex Kütz., Sp. Alg.: 224. 1849. (Pl.1, Fig. j)

Brownish, thallus, colonies up to 130 µm diameter, cells without sheath 3-4 µm in diameter, with sheath 4.5- 7 µm in diameter, sheath yellow to brown.

Occurred as dark brown crust on wet cement walls and rock surface.

Documented from site I, III, V, XI and XII.

14. *G. rupestris* Kütz., Tab. Phyc. 1: 17. 1847. (Pl.1, Fig. h)

Brownish thallus; cells single or in group of 2-4, cells without sheath 6-10 µm in diameter and with sheath 11-15 µm in diameter, yellowish brown sheath, colonies 17-40 µm in diameter.

Occurred as blackish crust on cement wall.

Documented from site I, IV, VIII, IX and XIV.

15. *Chroococcidiopsis cubana* Komárek & Hindák, Arch. Hydrobiol. Suppl. 46: 320. 1975. (Pl.1, Fig. b)

Small cells in groups, surrounded by colourless sheath; cells blue green in colour, spherical 3.5 µm long and 3.7 µm broad.

Occurred as bluish green crust on lime washed cement wall.

Documented from site III, VII, IX, XI, XII and XIII.

Stigonematales

1. *Hapalosiphon hibernicus* West & G.S. West, J. Roy. Microscop. Soc. [London] 1896: 163. 1896. (Fig. 3 e)

Single flexuous filamentous branched, main filament 7-8.5 µm broad, cells quadrate to sub cylindrical, lateral branches thinner than the main filament, 4.2-4.8 µm broad, cylindrical heterocyst; 4.5-5 µm broad, 7-9 µm long.

Occurred as bluish green crust on wet rocks.

Documented from site I, III, VI and XIV.

2. *Hapalosiphon welwitschii* West & G.S. West, J. Bot. [London] 35: 242. 1897. (Pl.1, Fig. m)

Thallus of closely entangled filaments, cells spherical to subquadrate, 3-7 µm broad, 4-13 µm long, short branches narrower than the main filaments, 3-6 µm broad, 2-15 µm long, colourless, thin sheath, heterocyst long, cylindrical, 4-6.5 µm broad, 7-10 µm long, intercalary heterocyst, cylindrical, 5-6 µm broad, 6-7.6 µm long.

Occurred as dark green crust on sand stone and lime washed wall.

Documented from site III, V, IX and X.

Family – Fischerellaceae

3. *Fischerella* sp. (Pl. 1, Fig. l)

Main filaments creeping, variously bent and densely interwoven, most of the filaments have cells in many rows, filaments about 7-9 µm broad within thick brown sheath, cells erect branches only on one side; lateral branches with long and narrow cells, main filaments with large spherical cells, 2.5-4 µm broad; heterocyst intercalary.

Occurred as dark green crust on sandstone and cemented walls.

Documented from site I, III, IV, V, VI and VIII.

4. *Fischerella* sp. (Fig 3 i)

Pale green thallus, main filament 12-16 µm broad, moniliform, with uniseriate filament, occasionally multiseriate, barrel shaped cells, in branches cylindrically elongated cells, longer than broad, thin colourless sheath, narrow cylindrical heterocyst.

Occurred as yellowish green patch on bricks and lime washed walls.

Documented from site II, IV, VII, IX, XII and XIII.

5. *Fischerella major* Gomont in Beck & Zahlbruckner, Ann. K. K. Naturh. Hofmus. [Wien] 1: 455. 1898. (Fig.3 f)

Brownish-green prostrate thick thallus, main filament creeping, entangled, torulose, 8-14.5 µm wide, branches cylindrical, erect, 6-11 µm wide, trichomes constricted at the cross-walls, 4-8.5 µm wide, cells cylindrical to subspherical, variably long, 6-7 µm wide, in branches barrel-shaped or isodiametric, thick lamellated sheath, heterocyst intercalary, cylindrical to oval.

Occurred as greyish black crust on moist rocks and cemented walls.

Documented from site III, VIII, IX and XI.

6. *Westiellopsis iyengari* Jeeji Bai in Desikachary, Taxon. Biol. Blue-green Algae: 64. 1972. (Fig. 3 h)

Thallus with true branching filaments of two types, main filaments torulose, up to 3- seriate, constricted at the cross-walls, with barrel-shaped cells, 4-11 μm broad and 4.5-15 μm long; trichomes in branches usually constricted with cells oval to long cylindrical, 2.5-17 μm long, 2.5-6 μm broad; thin and firm sheath, short quadrate to long cylindrical intercalary heterocysts, 4-18 μm long and 3.5-10.5 μm broad.

Occurred as blackish green patch on moist rocks, bricks and cemented wall.

Documented from site I, III, V, VI, VIII, X, XII and XIV.

7. *W. prolifica* Janet, Ann. Bot. n.s. 5 (17): 170. 1941. (Fig. 3 g)

Main filaments with short barrel- shaped cells, 7.5-11 μm broad, 8-12 μm long, branch filaments thinner with elongated cylindrical cells, 4-6 μm broad, heterocyst intercalary, oblong to cylindrical, 5-6 μm broad and 10-17.5 μm long.

Occurred as dark green crust on sand stone, cemented and lime washed walls.

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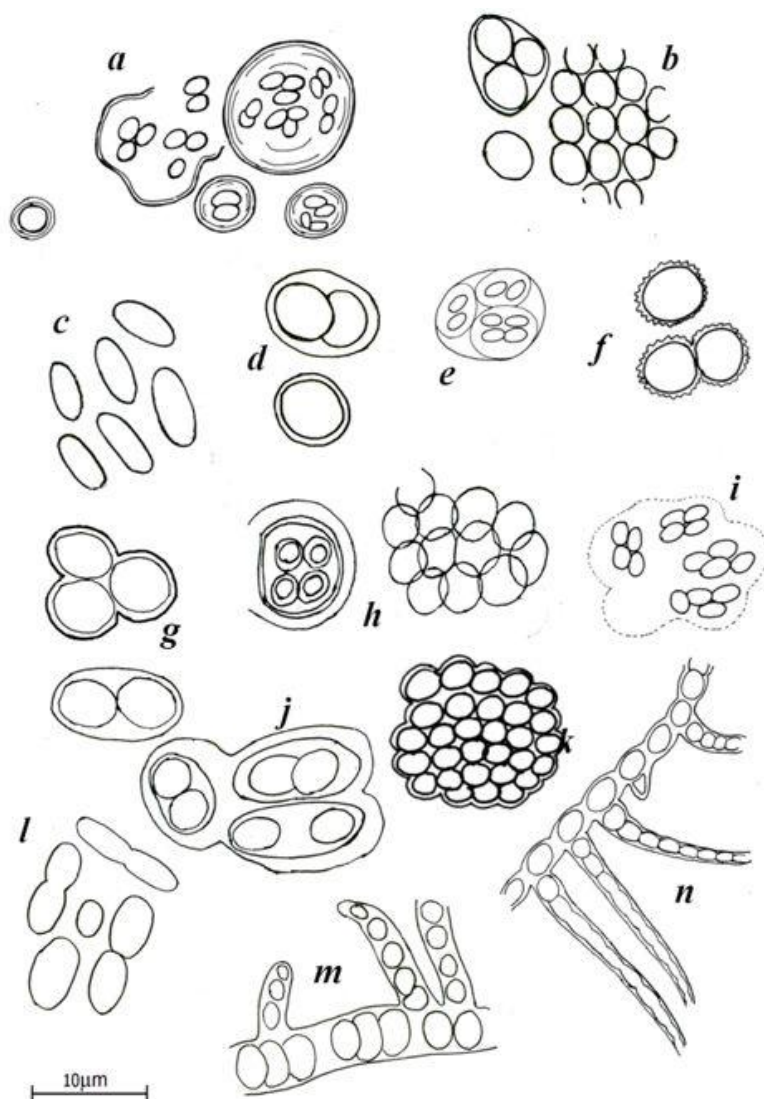


Plate1 Fig. a. *Asterocapsa divina*; b. *Chroococcidiopsis cubana*; c. *Aphanothece saxicola*; d. *Chroococcus indicus* e. *Gloeothece rupestris*; f. *Gloeothece rhodochlamys*; g. *Chroococcus minor*; h. *Gloeocapsa rupestris* i. *Cyanosarcina spectabilis*; j. *Gloeocapsa kuetzingiana*; k. *Cyanosarcina burmensis*; l. *Synechococcus elongates*; m. *Fischerella* sp; n. *Hapalosiphon welwitschii*;

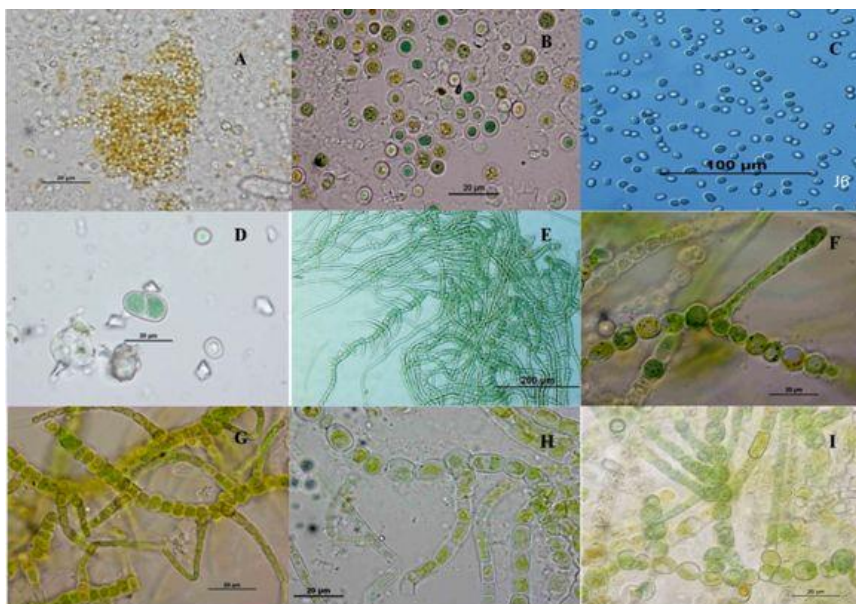


Fig 3: A. *Aphanocapsa banaresensis*; B. *Gloeocapsopsis pleurocapsoides*; C. *Cyanotheca* sp; D. *Asterocapsa divina*; E. *Hapalosiphon hibernicus*; F. *Fischerella major*; G. *Westiellopsis prolifica*; H. *Westiellopsis iyengari*; I. *Fischerella* sp.

A wide range of stone monuments from western Odisha are colonized by Chroococales and Stigonematales showing notable biodiversity. A total of 15 taxa from Chroococales and 7 taxa of Stigonematales were found. The most widespread commonly reported taxa in the stone cultural heritage in this region are *Chroococcus minor* and *Fischerella* sp, These genera were found associated with all lithotypes.

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