



RESEARCH ARTICLE

BLOOM FORMING TOXIC CYANOBACTERIA FROM MAHANADI RIVER NEAR HIRAKUI RESERVOIR OF WESTERN ODISHA

Niamen-Ebrottie J.E.¹, Bhattacharyya S.², Deep P.R.², and Nayak B^{2*}

¹Laboratoire d'Environnement et de Biologie Aquatique, Université Nangui Abrogoua, Côte d'Ivoire, 02 BP 801 Abidjan 02,

²School of Life Sciences, Sambalpur University, Jyoti Vihar, Burla, Odisha, Pin-768019, India

ARTICLE INFO

Article History:

Received 2nd, June, 2015
Received in revised form 10th,
June, 2015
Accepted 4th, July, 2015
Published online 28th,
July, 2015

Key words:

Cyanobacteria, Cyanotoxin,
Blooms, Mahanadi River,

ABSTRACT

Cyanobacteria are widely distributed in all natural ecosystems. These organisms form blooms under condition of nutrient over-enrichment, high temperature and low velocity. These blooms are harmful to the environment, animals and human health. The present study involves bloom forming cyanobacterial flora from Mahanadi River Near Hirakud Reservoir located in Sambalpur district, Odisha, India and from lower part of the hirakud dam. From the water sample, 37 species of cyanobacteria belonging 17 genera were recorded. Of these species, 9 were unicellular, 9 non-heterocystous filamentous and 19 heterocystous filamentous forms. Genus *Calothrix* with 7 species, *Nostoc* with 5 species, *Cylindrospermum* and *Aphanocapsa* with 4 species each were recorded. According to literature, 7 genera of them are potential toxin producer (*Anabaena*, *Calothrix*, *Cylindrospermum*, *Hapalosiphon*, *Microcystis*, *Nostoc* and *Phormidium*).

Copyright © Nayak B et al., This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution and reproduction in any medium, provided the original work is properly cited.

INTRODUCTION

Hirakud reservoir of the Mahanadi River is the main and ultimate water source for Agriculture and drinking purpose of livelihood of Sambalpur, Bargarh and Jharsuguda district. Due to massive industrialization (Deep et al., 2013), domestic and other sewages the river is going polluted day by day. The pollutants change the water quality which acts as catalyst for harmful cyanobacterial bloom (Paerl and Huisman, 2009). Cyanobacteria are naturally found in soils, rocks, lakes, streams, ponds, oceans and other surface waters and also found in extreme ecosystems such as hot springs, desert and polar regions (Adhikary 2002, Bhatnagar et al., 2008, Palleyi 2011, Aharon et al., 2009, Muthukumar et al 2007, Deepa et al., 2011) . Eutrophication is caused by nutrient over-enrichment because of human activities such as use of pesticides and fertilizers in agriculture, urbanisation and industrialization. In such condition cyanobacteria can rapidly multiply in surface water and cause "blooms." Cyanobacterial blooms can be harmful to environment, human being and animals as reported by Carmichael, 1992; Falconer, 2005. Factors responsible for cyanobacterial bloom formation are light intensity, total sunlight duration, nutrient availability (especially phosphorus), water temperature, pH, increase in precipitation events, water flow (whether water is calm or fast-flowing) and water column

stability (Carmichael, 1992). After full grown, the blooms decay, consumes oxygen and create hypoxic conditions that result in plant and animal die-off (Dadheech et al., 2001). Toxin producing cyanobacteria under favourable conditions of light and nutrients, are species of *Nostoc*, *Nodularia*, *Anabaena*, *Oscillatoria*, *Aphanizomenon*, *Microcystis*, *Anabaenopsis*, *Planktothrix*, *Cylindrospermopsis*, *Lyngbya*, *Raphidiopsis*, *Umezakia*, *Synechococcus*, *Hapalosiphon* and *Schizothrix* as reported by Dadheech et al., 2001; Oberhaus et al., 2007; Briand et al., 2005; Codd et al., 1999; Agrawal et al., 2006. Both nontoxic and common toxin-producing varieties cyanobacteria exist, and it is impossible to tell whether a species is toxic or not toxic by looking at it (United States Environmental Protection Agency, 2012). In most cases, the cyanobacterial toxins such as anatoxin-a and the microcystin are found intracellularly (approximately 95%) in the cytoplasm and are retained within the cell. These toxins are found during the growth stage of the bloom (United States Environmental Protection Agency, 2012). For those species, when the cell dies or breaks, the cell membrane ruptures and the toxins are released into the water (Bagchi, 1999). However, *Cylindrospermopsis*, *Aphanizomenon* and *Umezakia* produce cylindrospermopsin, a significant amount of the toxin may be naturally released to the water by the live cyanobacterial cell; the ratio is about 50% intracellular and 50% extracellular

*Corresponding author: **Nayak B**

Laboratoire d'Environnement et de Biologie Aquatique, Université Nangui Abrogoua, Côte d'Ivoire, 02 BP 801 Abidjan 02

(United States Environmental Protection Agency, 2012, Griffiths and Saker, 2003). The extracellular toxins may be absorbed by clays and organic materials dissolved in the water column and are generally more difficult to remove than the intracellular toxins.

MATERIAL AND METHODS

Samples Collection

Samples were collected from water bodies and rocks of Mahanadi River, situated 3 km away from Sambalpur University, Burla, Odisha (Figure. 1) from four different sites namely site-1 (lower part of Mahanadi bridge), site-2 (just below the Hirakud Dam), site-3(near the Hirakud Reservoir) and site-4 (rocks from Mahanadi river). Water samples are collected in 250 ml air tight plastic jars and few rocks were collected to examine attached cyanobacteria (Deep et al.2013).



Figure 1 Samples collection from Mahanadi River.

Isolation And Purification Of Cyanobacteria

One ml water samples were added to 10 ml of sterilized BG 11±N medium in petridishes (Rippka et al., 1979). The dishes were kept under 7.5 W/m² light intensity at 25±0.5°C in a culture room. After 10-12 days of incubation, algal colonies appeared on the agar plates. The numbers of colonies of each species were recorded (CFU). Microscopic observation of each colony was done, then isolated and spread on to fresh agar plates. Colonies appearing in fresh agar plates were examined microscopically and transferred to agar slants. This process was repeated till pure colonies were obtained.

Microscopic Analysis

Morphological identification of cyanobacteria species was performed under microscope Magnüs MLX-TR. For morphometric analysis camera lucida drawings were done. Basing upon trichome shape, filament colour, akinete and heterocyst (shape, size, position and number) were recorded. Identification of cyanobacteria was done using the keys given by Desikachary (1959); Komárek and Anagnostidis (2005, 2008).

RESULTS AND DISCUSSION

37 species of cyanobacteria belonging to 17 genera were identified from the four sites. Of these 37 species, 9 were

unicellular, 9 non-heterocystous filamentous and 19 heterocystous filamentous forms. The different genus identified were *Calothrix* (7 species), *Nostoc* (5 species), *Cylindrospermum* and *Aphanocapsa* (4 species each), *Jaaginema* (3 species), *Geitlerinema* (2 species), *Anabaena* (2 species) and ten genera with only one species were *Aphanothece*, *Chroococcus*, *Coelosphaerium*, *Cyanobacterium*, *Hapalosiphon*, *Heteroleibleinioides*, *Leptolyngbya*, *Microcystis*, *Phormidium* and *Pseudoanabaena* (Figure 2).

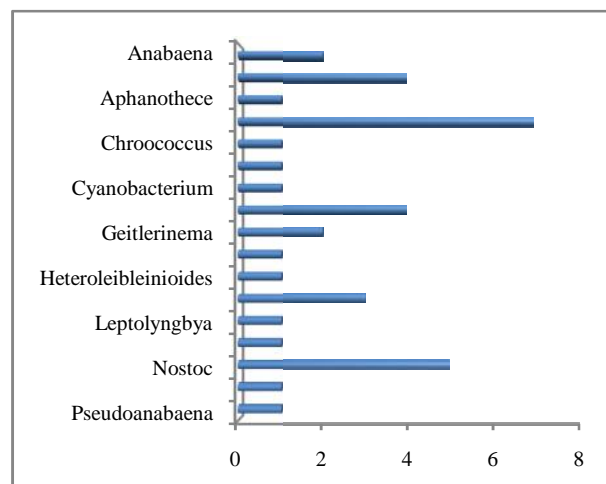


Figure 2 Number of genera and frequency of occurrence

In nitrogen free BG-11 medium species richness shows high value at site 4 with 10 species, site 1 with 9 species, site 2 with 6 species and site 3 with 4 species (Figure 3). In the BG-11 medium with nitrogen, species richness show high value at site 1 with 6 species, site 2 with 2 species; site 3 with 3 species and site 4 with 2 species. The table 1 show the abundance and total species of cyanobacteria present in different sampling sites. From the table it is observed that certain species are growing both in nitrogen supplement and nitrogen free media.

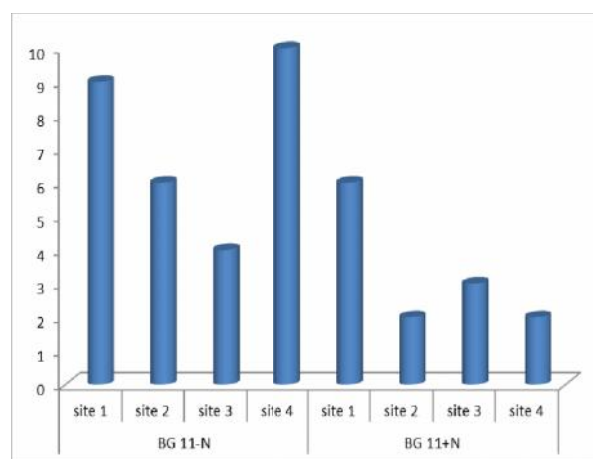


Figure 3 Species richness at four different sites. Site-1 (lower part of Mahanadi bridge), site-2 (just below the hirakud dam), site-3(near the hirakud reservoir) and site-4 (rocks from Mahanadi river)

The proliferation of a cyanobacterial population determines the quality of water bodies hence it is necessary to identify the cyanobacterial species of these water bodies. Good identification of cyanobacteria species can only be done after isolation and culturing under laboratory conditions (Komárek

and Anagnostidis, 2008). Cyanobacteria species can be divided in two groups, heterocystous species and non-heterocystous species. Non-heterocystous species grow well in BG-11 medium and heterocystous species grow well in nitrogen free BG-11 medium. In fact, the heterocystous cyanobacteria have the ability to fix atmospheric nitrogen and develop high biomass even when dissolved inorganic nitrogen is depleted (Komárek and Anagnostidis, 2008).

CONCLUSION

In this study, 37 taxa of cyanobacteria are encountered belonging to 17 genera and 7 genera are potential toxin producer (*Anabaena*, *Calothrix*, *Cylindrospermum*, *Hapalosiphon*, *Microcystis*, *Nostoc* and *Phormidium*). As the water of the Mahanadi River is used by local people for drinking and other purposes, hence immediate measures should

Table 1 Occurrence of cyanobacterial strains form water samples of different sites under laboratory condition

Taxa	1				2			
	1	2	3	4	1	2	3	4
	BG11-N				BG11+N			
<i>Anabaena fertilissima</i> Rao			1					
<i>Anabaena oryzae</i> Fritsch	1							
<i>Aphanocapsa biformis</i> A. Br.								1
<i>Aphanocapsa grevillei</i> (Berkeley) Rabenhorst						1		
<i>Aphanocapsa incerta</i> (Lemmermann) Cronberg & Komarek					1			
<i>Aphanocapsa orae</i> (Kosinskaja) Komarek & Anagnostidis				1				1
<i>Aphanothece stagnina</i> (Sprengel) Braun in Rabenhorst				1				
<i>Calothrix castelli</i> var. <i>somastipurensis</i> Rao			1					
<i>Calothrix clavatoidea</i> Ghose								
<i>Calothrix fusca</i> (Kütz.) Born. & Flah.			1					
<i>Calothrix javanica</i> de Wilde			1					
<i>Calothrix marchica</i> var. <i>intermedia</i> Rao			1					
<i>Calothrix viguieri</i> Frémy								
<i>Calothrix wembaerensis</i> Hieron. & Schmidle								
<i>Chroococcus distans</i> (Smith) Komarkova-Legnerova & Cronberg						1		
<i>Coelosphaerium limnicolum</i> Lund					1			
<i>Cyanobacterium minervae</i> (Copeland) Komarek et al.		1						
<i>Cylindrospermum licheniforme</i> Kütz. ex Born. & Flah.								
<i>Cylindrospermum marchicum</i> Lemmermann								
<i>Cylindrospermum musicola</i> Kütz. ex Born. & Flah.	1							
<i>Cylindrospermum musicola</i> var. <i>kashmiriensis</i> Bharadwaja			1					
<i>Geitlerinema amphibium</i> (Agardh ex Gomont) Anagnostidis	1	1			1	1		
<i>Geitlerinema thermale</i> Anagnostidis			1					
<i>Hapalosiphon flagelliformis</i> (Schmidle) Forti				1				
<i>Heteroleibleinioides fontana</i> (Hansgirg) Anagnostidis & Komarek			1					
<i>Jaaginema geminatum</i> (Meneghini ex Gomont) Anagnostidis & Komarek	1							
<i>Jaaginema pseudogeminatum</i> (Schmidle) Anagnostidis & Komarek						1		
<i>Jaaginema quadripunctulatum</i> (Brühl & Biswas) Anagnostidis & Komarek					1			
<i>Leptolyngbya</i> sp.	1							
<i>Microcystis flos-aquae</i> (Wittrock) Kirchner			1		1			
<i>Nostoc ellipsosporum</i> (Desm.) Rabenh. ex Born. & Flah.			1					
<i>Nostoc humifusum</i> Carmichael ex Born. & Flah.	1							
<i>Nostoc piscinale</i> Kütz. Ex Born. & Flah.	1							
<i>Nostoc punctiforme</i> (Kütz.) Hariot						1		
<i>Nostoc spongiaeforme</i> Agardh ex Born. & Flah.						1		
<i>Phormidium tergestinum</i> (Kütz.) Anagnostidis & Komarek						1		
<i>Pseudoanabaena catenata</i> Lauterborn			1					
Total	9	6	4	10	6	2	3	2

It has been reported that some species of cyanobacteria have the ability to produce toxin, which is their secondary metabolites (Oberhaus et al., 2007). Out of the 17 genus reported in this study the potential toxin producing genera are *Anabaena*, *Calothrix*, *Cylindrospermum*, *Hapalosiphon*, *Microcystis*, *Nostoc* and *Phormidium* as reported earlier also by Carmichael, 1992, 1997; Codd, 1995, 1998; Sivonen, 1996; Dadhech et al., 2001; Lopes and Vasconcelos, 2011. According to literature, these genera can produce microcystin, anatoxin-a, anatoxin-a(s), and saxitoxin. These toxins are dangerous for human health and animal. These are classified according to their effects on health, neurotoxins (on nervous system), hepatotoxins (on the liver) and dermatotoxins (on skin) (Bagchi, 1999). Species *Geitlerinema amphibium* recorded in this study is a potential saxitoxins producer (Borges et al., 2015), while *Aphanocapsa incerta* and *Microcystis flos-aquae* produces microcystin (Chorus, 2012; Mowe et al., 2014).

be taken to eradicate these harmful organisms from these water bodies for maintaining the health risk of livelihood.

Acknowledgement

First Author thanks DST, Govt. Of India For financial support as C.V. Raman International Fellow for African researchers Under Visiting Fellowship Programme, Author also thanks Head, School of Life Sciences for giving facilities.

References

- Adhikary SP(2002) Utilization of region specific cyanobacteria as biofertilizer for rice-a case study from Orissa; Conference paper.Biotechnol Micr Sustainable Utilization, 47-56.
- Agrawal M.K., Ghosh S.K., Bagchi D., Weckesser J., Erhard M. and Bagchi S., (2006). Occurrence of microcystin-

- containing toxic water blooms in Central India. J. Microbiol. Biotechn., 16:212-218.
- Aharon O, Danny I, Hindiye Muna Y, Malkawi Hanan I (2009) Morphological, phylogenetic and physiological diversity of cyanobacteria in the hot springs of Zerka Ma'in Jordan. *BioRisk*, 3:69-82.
- Bagchi S.N., (1999). Cyanobacterial toxins. J. Sci. and In. Res., 55: 715-727.
- Bhatnagar A, Makandar MB, Garg MK, Bhatnagar M, (2008) Community structure and diversity of cyanobacteria and green algae in the soils of Thar Desert (India). *J Arid Environ*, 72:73-83.
- Borges H.L.F., Branco L.H.Z., Martins M.D., Lima C.S., Barbosa P.T., Bittencourt-Oliveira M.C., Molica R.J.R. & Lira G.A.S.T., (2015). Cyanotoxin production and phylogeny of benthic cyanobacterial Strains isolated from the northeast of Brazil. *Harmful Algae* 43: 46–57. doi:10.1016/2015.01.003.
- Briand J.-F., Jacquet S., Flinois C., Avois-Jacquet C., Maissonette C., Leberre B. and J.-F. Humbert, (2005). Variations in the Microcystin Production of *Planktothrix rubescens* (Cyanobacteria) Assessed from a Four-Year Survey of Lac du Bourget (France) and from Laboratory Experiments. *Microbial ecology*, 50: 418–428.
- Carmichael W. W., (1992), Cyanobacteria secondary metabolites-the cyanotoxins. J. Appl. Bacteriol. 72:445–459.
- Carmichael W.W., The cyanotoxins. In *Advances in Botanical Research*, vol. 27 (Callow, J.A., editor), 211-256. Academic Press, London (1997).
- Chorus I., (2012). Current approaches to Cyanotoxin risk assessment, risk management and regulations in different countries. Federal Environment Agency (Umweltbundesamt) Wörlitzer Platz 1, 151p.
- Codd G.A., (1995). Cyanobacterial toxins : occurrence, properties and biological significance. *Water Sci. Technol.*, 32: 149±156.
- Codd G.A., Bell S., Kaya K., Ward C., Beattie K. & Metcalf J., (1999). Cyanobacterial toxins, exposure routes and human health. *European Journal of Phycology*, 34:4, 405-415.
- Codd G.A., (1998). Cyanobacterial blooms and toxins in fresh-, brackish and marine waters. In *Harmful Algae. Proceedings of the VIII International Conference on Harmful Algae*, Vigo, Spain, 1997 (Reguera, B., Blanco, J., Fernandez, M.L. & Wyatt, T., editors), 13±17. Xunta de Galicia and Intergovernmental Oceanographic Commission of UNESCO, Grafisant, Santiago de Compostela, Spain.
- Dadheech, P.K., Raisinghani G. and Trivedi, P.C., (2001). Phycotoxins- a status report. In: *algal biotechnology*. F.C. Trivedi (ed.), popular printers, Raj., India, pp 383-389.
- Deep, P.R., Bhattacharyya, S., and Nayak, B. (2013), Cyanobacteria in wetlands of the industrialized Sambalpur District of India, *Aquatic Biosystems*, 9:14 doi:10.1186/2046-9063-9-14
- Deepa P, Jeyachandran S, Manoharan C, Vijayakumar S (2011) Survey of Epilithic Cyanobacteria on the temple walls of Thanjavur District, Tamilnadu, India. *World J Sci Technol*, 1(9):28-32.
- Desikachary T. V., Cyanophyta. Indian Council of Agriculture research, New Delhi, 689p.(1959)
- Falconer I.R., (2005). Cyanobacterial toxins of drinking water supplies: cylindrospermopsins and microcystins. CRC, BocaRaton.
- Griffiths D.J. and Saker M.L., (2003). The Palm Island mystery disease 20 years on: a review of research on the cyanotoxin cylindrospermopsin. *Environ. Toxicol.*, 18:78-93.
- Komárek J. & Anagnostidis K., Cyanoprokaryota. 1. Teil : Chroococcales in Sü wasserflora von Mitteleuropa 19/1. H. Ettl, G. Gärtner, H. Heynig and D. Mollenhauer, Spektrum Akademischer Verlag Heidelberg, 548p. (2008).
- Komárek J. and Anagnostidis K., Cyanoprokaryota -2. Teil/ 2nd Part: Oscillatoriales. In : Büdel B., Krienitz L., Gärtner G., Schagerl M. (eds), Süswasserflora von Mitteleuropa 19/2, Elsevier/Spektrum, Heidelberg, 759 p.(2005).
- Lopes V. R. and Vasconcelos V. M., (2011). Planktonic and benthic cyanobacteria of European brackish waters: a perspective on estuaries and brackish seas, *European Journal of Phycology*, 46:3, 292-304, DOI:10.1080/09670262.2011.602429.
- Mowe M. A. D., Mitrovic S. M., Lim R. P., Furey A. and Yeo D. C. J., (2014). Tropical cyanobacterial blooms: a review of prevalence, problem taxa, toxins and influencing environmental factors. doi: 10.4081/jlimnol.2014.1005.
- Muthukumar C, Muralitharan G, Vijayakumar R., (2007). Cyanobacterial biodiversity from different freshwater ponds of Thanjavur, Tamilnadu (India). *Acta Botanica Malcitana*, 32:17-25.
- Oberhaus L., Gelinas M., Pinel-alloul B., Ghadouani A. and Humbert J-F., (2007). Grazing of two toxic *Planktothrix* species by *Daphnia pulex*: potential for bloom control and transfer of microcystins. *Journal of Plankton Research*, 29 (10): 827–838.
- Paerl H.W. and Huisman J., (2009). Climate change: a catalyst for global expansion of harmful cyanobacterial blooms. *Environmental Microbiology Reports* 1, 27-37.
- Palley S, Kar R N, Panda C R J., (2011). Influence of water quality on the biodiversity of phytoplankton in Dhamra river Estuary of Odisha Coast, Bay of Bengal. *Appl Sci Environ Management*, 15(1):69-74.
- Poste A. E., Hecky R. E. and Guildford S. J., (2013). Phosphorus enrichment and carbon depletion contribute to high *Microcystis* biomass and microcystin concentrations in Ugandan Lakes. *Limnol. Oceanogr.*, 58:1075-1088.
- Sivonen, K., (1996). Cyanobacterial toxins and toxin production. *Phycologia*, 35 (Suppl.): 12-24.
- United States Environmental Protection Agency, Cyanobacteria and Cyanotoxins: Information for Drinking Water Systems. Office of Water 4304T Report, 9p, 2012.
