A STUDY ON MICROBIAL DIVERSITY OF KONDAKARLA AVA LAKE, VISAKHAPATNAM.

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Abstract:

Kondakarla Ava lake (Lat.17^0 35' 30"N and 17^0 36' 02"N and Long.82^0 59' 27" E and $83^0$01'02" E)is the Second largest (6.5km^2) fresh water lake of Visakhapatnam in Andhra Pradesh state and the lake is also famous as bird resort, with it's rich biodiversity of flora and fauna. The lake is at about 40 K.M. from Visakhapatnam and 7 K.M. from Anakapalle.

It is also recognized as an Eco Tourism destination. It is surprising to see that even though the pollution is high, the ecology of the lake is not much disturbed, that attracted the attention of the author to assess the quality of the watersduring the period of 2016-2018. The eutrophic condition of the lake is being addressed by the microorganisms by way of bioremediation. Most of the toxicants released are being processed by the plant communities present in the lake. A detailed study of the composition of the Microorganisms in the lake was attempted in the present study.The Natural Bioremediation that is taking place in the ambient waters by the micro organisms is highlighted in the study.

Key words : Bioremediation, Pathogens. Growth media, Nutrients,Most Probable Number.

I. Introduction :

Freshwater lakes are the best wetlands which have great economic, ecological and cultural importance. All the surrounding communities depend directly on them for drinking water, food and their livelihoods. Most lakes loose their "self-cleaning mechanism" due to several anthropogenic activities and tend to have more complex situation and become fragile.

These freshwater environments have very different physical and chemical characteristics, and correspondingly different microbial communities and activities.(Virgin I. Rich, Raina M. Maier, 2015).

Freshwater lakes contain diverse and dynamic microbial communities that play key roles in maintaining water quality, cycling nutrients, and regulating carbon uptake and storage. The nutrient supplements of a lake are affected by biological, geological, and anthropogenic activities and even the amount of oxygen and the pH level can also affect a lake's chemistry.

Microbial organisms address the biogeochemical (C, N, P, S) pathways of deep reservoirs and shallow lake ecosystems. The biogeochemical importance of bacteria in freshwater ecosystems was first recognized in the 1940s. The aquatic bacteria drive transformations and the cycling of most biologically active elements in these ecosystems. (Ryan J. Newton et.al, 2011). Apart from their microbial and algal populations, streams, rivers and lakes also contain fungi, protozoa and viruses, which interact and contribute to the functioning of the food web.

These water bodies host distinct water, sediment and surface-associated microbial communities and their populations may be largely driven by changes in the nutritional status of the water bodies due to variations in the hydrological regime (e.g., thermal stratification and water level) and climate change. Many Microorganisms are found naturally in fresh and saltwater. These include bacteria, Proteobacteria, Actinobacteria, cyanobacteria, Bacteroidetes, protozoa, algae, and tiny animals such as rotifers.

Common freshwater lake genera include *Microcystis*, *Anabaena*, *Rhodospirillum*, *Chlorobium*, *Desulfovibrio*, *Proteobacteria*,*Aphanizomenon*, *Oscillatoria*, *Planktothrix*, *Synechococcus*, and *Cyanothece*.

The present study lake, Kondakarla Ava lake (Fig. 1) is maintained mainly by the Water Resources Department (Irrigation) of Andhra Pradesh, while Forest Department, Fisheries department and Tourism Development Corporation also service the lake use. Therefore, the lake is used as a livelihood opportunity for several fishermen, farmers of over 17 villages besides being a bird resort for several migratory species and tourism opportunities.

Fig:1 Satelliete image of kondakarla ava lake

II. Methodology:

Identification of Microbial composition of water is analysed by Most Probable Number (MPN) method (APHA, 1995). The water samples were collected using 1.5l plastic samplers. The water samples were initially passed through a mesh sieve (40mm pore size) to remove course particle. Most Probable Number (MPN) is a method used to estimate the concentration of viable microorganisms in a sample by means of replicate liquid broth growth in ten-fold dilutions. The result was expressed in MPN/100 *ml*. The diluted samples were cultured on different Media for identification of Microbes, standard media like Nutrient Agar, Mac Conkey agar, etc. are used.

III. Results and Discussion:

There are many groups of bacteria that are observed by cultural methods and some of microbial organisms identified in Kondakarla lake water samples and their morphology are furnished below in the table no.1

Table no. 1 : List of microbial organisms identified in kondakarla ava lake

S. no	Microorganisms	Morphology	Source	Symptoms	Remarks
1	*Vibrio cholerae*	Gram-Negative, Comma-Shaped bacterium	Consuming contaminated food and water after entering into the host it infects the intestines	Watery diarrhea (a grey and cloudy liquid), occasional vomiting and abdominal cramps etc.	Pathogenic organisms.
2	*Bacillus flexus*	Gram variable, rod shaped, endospore forming	Non pathogenic	Non pathogenic	Use in Food, Pharmaceutical and detergent industries
3	*Brevibacterium sp.*	Gram - Positive	Fresh and salt water, Found in dairy products,	Unpleasant body odors.	Antimicrobial substances, which inhibit growth of

			insects and decaying organic matter.		pathogenic bacteria, yeasts and molds
4	*Bacillus aerius*	Gram Positive, Motile rods	Contaminated animal carcasses in the water	Food borne illness	Pathogenic organisms.
5	*Bacillus vallismortis*	Gram Positive, Motile Rods	Contaminated water	Skin infections	Antifungal activity
6	*Serratia nematodiphila*	Gram Negative rod shaped	Lives in the intestine of nematodes	Infect urinary and respiratory tracts	Pathogenic organisms.
7	*Entamoeba coli*	Motile trophozite and a cyst	Contaminated food and water and even insects and rodents carry the parasite to cause infection in the food and drinks	Amoebiasis, dyspepsia, hyperacidity, gastritis and indigestion	Pathogenic organisms.
8	*Escherichia coli*	Gram negative, rod shaped	Lives in human intestines and also found in the gut of some animals	Diarrhea , Vomiting , fever etc	Used for removal of Heavy metals from water bodies, Pathogenic organisms.
9	*Cryptosporidium sp.*	Oocysts appear as small round structures (4to6µm) similar to yeast	Contaminated food or water (drinking or recreational)	Gastrointestinal illness	Pathogenic organisms.

3.1 Vibrio cholerae:-

Vibrio cholerae grow at temperature 37^0C on Nutrient Agar . The colonies are moist, round and translucent. They are Gram - negative , comma shaped bacterium. Vibrios are one of the most common pathogenic water borne organisms in surface waters of the world, they occur in both marine and freshwater habitats and along with aquatic animals. *V. cholerae* can be spread by consuming contaminated food and water after entering into the host it infects the intestines and shows Symptoms which include abrupt onset of watery diarrhea (a grey and cloudy liquid), occasional vomiting, and abdominal cramps etc.

3.2 Bacillus flexus:-

Bacillus flexus grow at temperature 17 to 37^0C , colonies are opaque and smooth. They are Gram - variable , rod shaped bacterium. The isolated *Bacillus flexus* strain produces the lipase which is thermo stable, alkaliphilic and has potential to use in food, pharmaceutical and detergent industries. (**Alkaliphilic** *Bacillus Flexus***: A Potential Source Of Lipase Producer For Industrial And Medical Application;** D. H. Tambekar and et.al; *IJPSR, 2017.)*

3.3 Brevibacterium sp:-

*Brevibacterium sp*are observed as opaque, small and convex , smooth surface colonies on nutrient agar.*Brevibacterium.sp*. are Gram Positive and found in fresh and salt water, dairy products, marine organisms, insects and decaying organic matter and may occur as spoilage organisms in foods and in the case of human skin surfaces. Brevibacterieae probably contribute to unpleasant body odors. These Species are known to produce Antimicrobial substances, which inhibit the growth of many food poisoning and pathogenic bacteria as well as several yeasts and molds (Bikash, Ghosh, Sienkiewicz, & Krenkel, 2000;Jones & Keddie, 1986;Onraedt et al., 2005;Rattray & Fox, 1999) .

3.4 Bacillus aerius :-

Bacillus aerius grow at temperature 8 to 37^{0}C , white, irregular raised colonies are observed on nutrient agar. The Gram - Positive , motile rod species are found in contaminated water and animal carcasses, they cause food borne illness in human beings.

3.5 Bacillus vallismortis:-

Bacillus vallismortis grow at temperature 5-50 ^{0}C on Nutrient Agar . The colonies are opaque, smooth, circular, gram - positive , motile rods (Fig:2). Bacillus vallismortis pathogenic and can be observed rarely in the lake samples and if abundant quantity of bacteria is present in lake it may cause skin infections. *Bacillus vallismortis* showed strong growth inhibition activity *in vitro* against the phytopathogens.(Study of the antifungal activity of *Bacillus vallismortis* ZZ185 *in vitro* and identification of its antifungal components;ZhenzhenZhao and et.al; Bioresource Technology, January 2010,)**.**

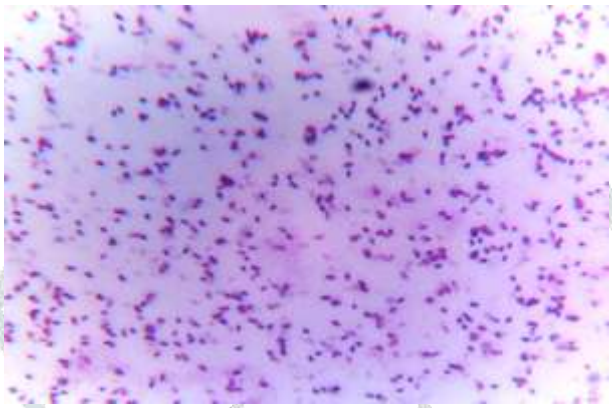

Fig. 2: Bacillus vallismortis

3.6 Serratia nematodiphila :-

Serratia nematodiphila (Zhang et al,2009) grow at temperature 20-35 ^{0}C on Nutrient Agar. The colonies are circular , opaque, cream - white and smooth , gram negative rod shaped .*S. nematodiphilia* is pathogenic bacteria which infect urinary and respiratory tracts. *Serratia nematodiphila*with single flagellum,lives in the intestine of nematodes - Heteroorhabditidoides-*changmingensis* has been known to have symbiotic-pathogenic life cycle on the multilateral relationships with entomopathogenic nematode and insect pest. (*Serratia nematodiphila* sp. nov., associated symbiotically with the entomopathogenic nematode Heterorhabditidoides *chongmingensis* (Rhabditida: Rhabditidae). Zhang CX and et.al; Int J Syst Evol Microbiol. 2009.)

3.7 Entamoeba coli :-

Entamoeba species all come in monogenetic forms, or having one generation lifecycles. *E. coli* has "three distinct morphological forms exist airing the life cycle-Trophozoite, Pre-cystic stage and Cystic stage". Excess quantity of *E.coli* may lead to "dyspepsia, hyperacidity, gastritis, and indigestion"; these are common problems of most intestinal parasites. The cysts cause infection by consuming contaminated food and drinks like waste water and even insects and rodents carry the parasite to cause infection in the food and drinks. Excystation happens once the cysts are ingested, and travel to the large intestine. The pathogenic intestinal protozoan causes amoebic dysentery in humans(amoebiasis).

3.8 Escherichia coli:-

Escherichia coli growth was observed on Mac Conkey agar with bright pink colonies,gram negative, rod shaped(Fig.3;).*E. coli* (*Escherichia coli*), normally lives in human intestines and also found in the gut of some animals. Most types of *E. coli* are harmless and they even keep the human digestive tract healthy. But some strains can cause diarrhea , Vomiting , fever etc. by taking contaminated food or by taking contaminated water. *E.Coli* species are used for removal of Heavy metals from water bodies (Bioremediation of Polluted Waters Using Microorganisms by Luciene M. Coelho and et.al.2015.)

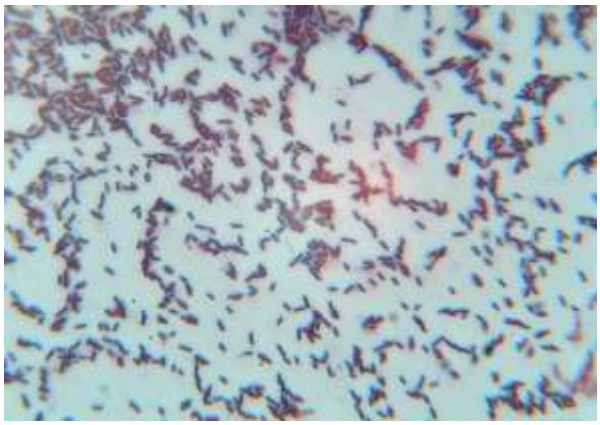

Fig.3:- Escherichia coli

3.9 Cryptosporidium sps:-

*Cryptosporidium sps*can be seen through bright-field microscopy using differential interference contrast (DIC), oocysts appear as small round structures (4 to 6 µm) similar to yeasts. They do not autofluoresce. Oocysts (4 to 6 µm) often have distinct oocyst walls and stain from light pink to bright red , can be observed by Acid fast Staining technique.

Numerous outbreaks of cryptosporidiosis is due to contaminated food or water (drinking or recreational) have been reported in several industrialized nations, and studies have sometimes identified water as a major route of *Cryptosporidium* transmission in areas where the disease is endemic. Gastrointestinal illness (cryptosporidiosis) that primarily involves watery diarrhea (intestinal cryptosporidiosis) with or without a persistent cough (respiratory cryptosporidiosis) in both immunocompetent and immunodeficient humans. (*Cryptosporidium* Taxonomy: Recent Advances and Implications for Public Health; Lihua Xiao, Ronald Fayer, Una Ryan, Steve J. 2004)

The Kondakarla ava lake established a platform for number of mostly pathogenic micro organisms as shown above. The reasons for the occurrence of these organisms in quite good numbers is mostly due to anthropogenic activities in and around the lake. Eco tourism is in the development stage people who come here for recreation leave lots of food waste near the banks of the lake that attracts the attention of most of the domestic as well as wild animals that visit the lake for shelter and water.

The rate of contamination is increasing alarmingly in recent times as more and more human habitations are developing around the lake causing a threat to the very existence of the lake. The organic contamination into the lake has decreased the average depth of the lake from 12 feet earlier to 7 feet in recent times as per the observations.

Conclusions:

The Kondakarla ava lake water is now not safe for drinking as most of the water bodies got contaminated with different kinds of microbial organisms (may be in less numbers but there is a threat).The Ecotourism department must focus its attention to address the extent of pollution in the lake and measures to mitigate the damage, ultimately top priority is to be given to conserve the lake ecosystem which is the second largest fresh water lake in Andhra Pradesh.

Apart from biological pollution, there is also bioremediation taking place in the Kondakarla ava lake ecosysytem, most of the toxicants released and the eutrophic condition in the lake'secosystem is being mitigated by the microbial organisms in the ambient environment, this is very clearly seen in this particular ecosystem, inspite of heavy pollutant load due to Industries around the lake, the waters are still eco friendly and not much affected. The reason is that the microbes in the water bodies are taking care of the bioremediation process by reducing the nutrient levels in the ecosystem to a considerable level.

There is immediate urgency in restoring and establishing pollution free environment in the Lake.Since most of the lake waters are loaded with heavy amount of pollutants as mentioned,such as Industrial and Agricultural waste, there is also report of trace elements in the ambient environment. The most convenient method of addressing the pollution in the lake is encouraging natural bioremediation by the micro organisms, which is cost effective and ecofriendly.Since the lake eco system is land locked, natural bioremediation process can take less time to clean the lake.

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References:-

1. American Public Health Association (1965) - Standard Methods for Examination of Water and Wastewater Including Bottom Sediments and Sludges, 12th Ed. American Public Health Association Inc., New York.

2. Andersen, F.Ø.; Olsen, K.J. (1994) Nutrient cycling in shallow, oligotrophic Lake Kvie, Denmark. Hydrobiologia 275/276: 267–276; 1994.

3. Angela J. Ramsay (1972) - Studies on the Microorganisms of a Freshwater Lake - Ph.D Thesis in the University of Canterbury, Christchurch, New Zealand.

4. D. H. Tambekar and et.al; - Alkaliphilic ***Bacillus Flexus***: A Potential Source Of Lipase Producer For Industrial And Medical Application;*IJPSR, 2017; Vol. 8(10): 4313-4317*

5. Freshwater - www.newworldencyclopedia.org

6. Freshwater Ecology - www.waterencyclopedia.com

7. Hongchen Jiang and et.al (2006) - Microbial Diversity and Sediment of Lake Chaka, an Athalassohaline Lake in Northern China- Applied and Environmental Microbiology, 72(6) pg 3832-3845.

8. Joao P.S.Cabral (2010) - Water Microbiology. Bacterial Pathogens and Water; International Journal of Environmental Research and Public Health(MDPI)- 7(10) pg 3657-3703.

9. Luciene M. Coelho and et.al. - Bioremediation of Polluted Waters Using Microorganisms

10. Lihua Xiao, Ronald Fayer, Una Ryan, Steve J.- *Cryptosporidium* Taxonomy:Recent Advances and Implications for Public Health, **DOI:** 10.1128/CMR.17.1.72-97.2004

11. Ryan J. Newton and et.al (2011) - A Guide to the Natural History of Freshwater Lake Bacteria; Microbiology And Molecular Biology Reviews, Mar. 2011, p. 14–49 Vol. 75, No. 1.

12. Sravani Pericharla (2017) - A study on the awter Quality of Kondakarla Awa Lake, Visakhaptanm , Andhra Pradesh, India; International Journal of Innovative Research and Creative Technology ; Volume - 2 ; Issue -6; pg -223-225.

13. Zhang CX and et.al(2009) - Serratia nematodiphila sp.nov.; associatedsymbiotically with the entomopathogenic nematode Heterorhabditidoides chongmingensis (Rhabditida:Rhabditidae). Int J Syst Evol Microbiol.2009 Jul; 59 (Pt 7): 1603 -8. doi: 10.1099/ijs.0.65718-0.

14. Zhenzhen Zhao and et.al; - Study of the antifungal activity of Bacillus Vallismortis ZZ185 in vitro and identification of its antifungal components; Bioresource Technology , Volume 101, Issue 1, January 2010.Pages 292-297.