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Bacteriological Biodiversity and its Relationship with the Quality of Water in Panvel Lakes (Vishrale, Krishnale and Dewale Lake) at Raigad (Maharashtra), India

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Abstract: This research is carried out to study the bacteriological biodiversity and quality of water during pre-monsoon, monsoon and post-monsoon in Panvel lakes, Maharashtra, India. Most Probable Number (MPN) count was carried out by using Mac Conkey's broth in order to classify the lakes into different categories. Pathogenic bacteria colonies were isolated from water samples collected from different lakes, by spread plate method. After incubation, different colonies from incubated plates were picked; purified by repeated sub-culturing before being examined for gram reaction, microscopically bacteria were identified into positive or negative, cocci or bacilli, rod or spiral. Biochemical analysis was carried out using different tests. The results were analyzed using Bergey's manual of systematic bacteriology. Many different types of bacteria were isolated from water samples that helped in classifying the lakes and knowing the bacterial loads of each lake.

Keywords: Bacteria, Biochemical test, MPN count, Lake, Panvel

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Introduction

Water is the basic requirement of all living organisms. It receives microorganisms from air, sewage, soil and other organic wastes. Many people in the world suffer from water

borne diseases as contaminated water causes direct danger to health. Manja and Kaul (1992) stated that in developing countries like India, about 50 per cent of the deaths among

the children are due to unhygienic and water born diseases. Hence, issues related to polluted water must be recognized as early as possible so that steps should be taken to treat the water (Anonymous, 1992). Lots of researches were performed on bacterial community throughout the world of the freshwater lakes like Lake Baikal, Russia (Khakhinov *et al.*, 2012), Dianchi Lake, China (Bai *et al.*, 2012), Chandra Tal, India (Yadav *et al.*, 2016) and Dashair Lake, India (Yadav *et al.*, 2017).

Bacteriological water analysis is a method of analyzing water to estimate the number of bacteria present and find out what sort of bacteria they are, and draw inference about the suitability of the water. Thus, present study was aimed to investigate the quality of water and presence of pathogenic and disease causing indicator organism in the lakes of Parnel city which may be pathogenic to human life. Analysis is usually performed using culture, biochemical and sometimes optical method.

Materials and Methods

Sampling for Bacteriological Analysis:

Water samples (during pre-monsoon, monsoon and post-monsoon) of three different lakes Vishrale, Krishnale and Dewale were collected in sterile leak proof glass bottle following the standard method of water collection for microbiological analysis.

Culture Media:

All the chemicals used were of analytical grade. Composition of nutrient Agar (pH 7) for 100 ml are: Peptone 1 g, NaCl 0.5 g, Meat Extract 0.3 g, DW 100 ml, Agar 3 g.

Biochemical Test:

MPN count was carried out by using MacConkey's broth. Results obtained were compared with the standard chart and recorded as total MPN/100 ml. *Escherichia coli* were estimated by pour plate technique on MacConkey Agar incubated at 37 C (APHA, 2005).

Pathogenic bacterial colonies were isolated from water samples collected from different lakes, by spread plate method. After incubation, different colonies from incubated plates were picked; purified by repeated sub-culturing before being examined for gram reaction, microscopically bacteria were identified into positive or negative, cocci or bacilli, rod or spiral (Claus, 1992). Biochemical analysis included -- Sugar fermentation test, Methyl red test, Voges-Proskauer test, Citrate utilization test, Indole test, Urease test, Catalase test, Oxidase test, Gelatinase test, Triple Sugar Iron agar, lysine decarboxylase test, Ornithine test and H₂S production. The results were analyzed using Bergey's manual of systematic bacteriology (Sneath *et al.*, 1986).

Results and Discussion

Seasonal variation in MPN count in the three lakes is represented in Tables 1-3 and Fig. 1. During monsoon season more MPN count is observed as 540/100 ml, 220/100 ml and 170/100 ml in Vishrale, Krishnale and Dewale Lake, respectively. Bright pink colonies on incubated MacConkey plates were recognized as *Escherichia coli* (Harrigan and McCance, 1976).

The bacteria which were isolated from water samples include *Escherichia coli*,

Table 1: Seasonal MPN index and 95% confidence limits for Vishrale lake during 2010 – 2011

Seasons	Combination of positives	MPN index/100 ml	Confidence limits	
			Low	High
Pre – monsoon	5-5-1	350	100	1100
Monsoon	5-5-2	540	150	1700
Post – monsoon	5-4-3	280	100	710

Table 2: Seasonal MPN index and 95% confidence limits for Krishnale lake during 2010 – 2011

Seasons	Combination of positives	MPN index/100 ml	Confidence limits	
			Low	High
Pre – monsoon	5-3-3	170	70	400
Monsoon	5-4-2	220	70	440
Post – monsoon	5-4-0	130	36	400

Table 3: Seasonal MPN index and 95% confidence limits for Dewale lake during 2010 – 2011

Seasons	Combination of positives	MPN index/100 ml	Confidence limits	
			Low	High
Pre – monsoon	5-3-2	140	52	400
Monsoon	5-3-3	170	70	400
Post – monsoon	5-4-0	130	36	400

Staphylococcus aureus, *Klebsiella* sp., *Proteus* sp., *Salmonella* sp., *Shigella* sp., *Vibrio* sp., *Bacillus* sp., *Staphylococcus* *cholerae*, *Pseudomonas aeruginosa*, *Enterobacter* sp. and *Bacillus* sp.

Table 4: List of isolated micro-organisms from the three lakes

Isolates	Description of isolates
<i>Bacillus</i>	Gram positive, spore forming rods, Catalase positive, creamy colony on Nutrient Agar
<i>Escherichia coli</i>	Gram negative rod, heavy lactose fermenter, does not utilize citrate, indole positive test, ferments mannitol, pinkish colony
<i>Salmonella</i>	Dark centered gram negative, Non-lactose fermenting, urease negative, utilize citrate, produces H ₂ S
<i>Enterobacter</i>	Gram negative rod, lactose fermenting, utilize citrate
<i>Shigella</i>	Non-lactose fermenting, does not utilize citrate, produces H ₂ S
<i>S. aureus</i>	Opaque, golden yellow colony on nutrient agar, Gram positive cocci, Catalase positive, mannitol fermenter
<i>Pseudomonas aeruginosa</i>	Colourless colony on Nutrient Agar, Gram negative rod, Catalase positive, negative lactose fermentation, positive H ₂ S production, positive Oxidase
<i>Vibrio cholerae</i>	Gram negative rod, Oxidase positive, glucose fermenter
<i>Klebsiella</i>	Gram negative rod, Oxidase negative, indole negative, MR negative, VP positive, lysine positive
<i>Proteus</i>	Translucent colony with serrated margins, Gram negative, glucose positive, Gelatinase positive

aureus and *Bacillus* species are gram positive and rest are gram negative bacteria. Characterization of bacteria is shown in Table 4.

Some degree of contamination is always present in surface water bodies due to animal and human activities that results in increased bacterial concentration of lake water. In the present study, maximum MPN count is revealed during monsoon and minimum during post-monsoon season. Similar observations were documented by Agarwal and Rajwar (2010) and Singh and Sharma (2016). Presence of coliform is the primary

bacterial indicator for faecal pollution in water as they are unable to survive in clean water (Mishra *et al.*, 2009). Based on studies it was confirmed that majority of the coliform bacteria were found to be gram negative. The highly polluted surface water is abundant in various types of bacteria.

Total bacterial load of Vishrale lake was higher than other lakes. With respect to water quality for the best use, we categorized the water body depending on the values of coliforms (Table 5). Thus, in the present investigation we can place Vishrale lake in III category as "Satisfactory", whereas Krishnale

Lake and Dewale lake is categorized in II category as “good”. Thus the water of these

lakes can be used for the purpose of bathing and swimming (Pandey and Sharma, 1999).

Fig. 1: MPN count

Table 5: Suggested values of Coliform/100 ml for beneficial uses of water (Pandey and Sharma, 1999)

Sr. No.	Water Quality	Grade	Bathing & Swimming	Public Water Supply
1.	Excellent	I	< 10	< 100
2.	Good	II	4	200
3.	Satisfactory	III	250	800
4.	Poor	IV	1500	1000
5.	Unacceptable	V	> 6000	> 8000

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