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RESEARCH ARTICLE

MICROBIAL BIODIVERSITY OF TRIBUTARIES OF RIVER GANGA IN UTTARAKHAND

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ABSTRACT

Water quality assessment conducted in the tributaries of Ganga River for microbial analysis in the year 2012 and 2013 has been done. For the study Devprayag is selected at river Ganga in Uttarakhand at which is the confluence of river Alaknanda and Bhagirathi. All samples were positive for *E. coli*, which indicates fecal pollution of water. The MPN count ranges from 15 to 46 MPN/100 ml for the water samples from rivers Alaknanda and 16 to 35 MPN/100 ml water samples from rivers Bhagirathi. The SPC count ranges from 10 to 49 SPC/ml x1000 for the water samples from rivers Alaknanda and 21 to 49 SPC/ml x1000 water samples from rivers Bhagirathi. The fecal coliform counts also exceeding the standard limit for water. The Isolated organisms were identified to be *Staphylococcus aureus*, *Salmonella* species, *Escherichia coli*, *Pseudomonas aeruginosa*, *Enterobacter aerogenes* and *Shigella* species.

Key words:

Biodiversity, *E. coli*, MPN, SPC

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INTRODUCTION

Water is super abundant on the planet as a whole, but fresh potable water is not always available at the right time or the right place for human or ecosystem use. The importance of water is underscored by the fact that many great civilizations in the past sprang up along or near water bodies. The development of water resources has often been used as a yardstick for socioeconomic and health status of many nations worldwide. However, pollution of waters often negates the benefits obtained from the development of these water resources.

'Coliform' was the term first used in the 1880s to describe rod-shaped bacteria isolated from human faeces. The coliform group of bacteria is a functionally-related group which all belongs to a single taxonomic family (Enterobacteriaceae) and comprises many genera and species. There are other genera in the Enterobacteriaceae family, such as *Salmonella* and *Shigella* that are not considered coliforms. The total coliform group of bacteria was originally used as a surrogate for *E. coli* (the name coming from 'coli-form' or like) which, in turn, was considered to show faecal pollution. As a result, total coliforms were adopted and considered to be equivalent to *E. coli* until more specific and rapid methods became available. Originally total coliform bacteria were considered to be from four genera of the family Enterobacteriaceae that could all ferment lactose. These genera were *Escherichia*, *Klebsiella*, *Enterobacter* and

Citrobacter. Out of the total coliforms present in the human gut, *Escherichia coli* (*E. coli*) represent the majority of the population. Total coliforms represent only about 1% of the total population of bacteria in human faeces in concentrations of about 10⁹ bacteria per gram (Brenner *et al.*, 1982).

Most drinking water guidelines refer to the use of total estimates of bacterial numbers in water. This measure is generally called 'total heterotrophic plate count' (HPC) or 'standard plate count bacteria' (SPC), and is considered to represent the general cleanliness of a drinking water. As the HPC is not considered indicative of a potential health risk, these bacteria are not generally considered as a compliance measure; rather their numbers are monitored to understand changes in a drinking water system over time and to alert operators to increases in general bacterial numbers.

Many infectious diseases are associated with faecally contaminated water and are a major cause of morbidity and mortality worldwide (Leclerc *et al.*, 2002; Theron and Cloete, 2002). Waterborne diseases are caused by enteric pathogens such as bacteria, viruses and parasites that are transmitted by the faecal oral route (Grabow, 1996; Leclerc *et al.*, 2002). Waterborne spread of infection by these pathogenic microorganisms in the water environment, the infectious dose of the microorganism required to cause a disease in susceptible individuals, the microbiological and physio-chemical quality of the water, the presence and absence of water treatment and

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the season of the year (Deetz *et al.*, 1984; Leclerc *et al.*, 2002; Theron and Cloete, 2002). The survival of microorganism such as bacteria in water environments depend on the presence of nutrients and water temperature (Edberg *et al.*, 2000; Leclerc *et al.*, 2002). The infectious dose of some bacteria ranges between 10^7 to 10^8 cells, with some enteric bacteria able to cause infection at doses as low as 10^1 cells (Edberg *et al.*, 2000; Leclerc *et al.*, 2002). Although waterborne pathogens are distributed worldwide, outbreaks of some diseases tend to be subjected to geographical factors. In the last number of years several outbreaks of pathogenic diseases have appeared that cannot be prevented by traditional waste water treatment.

Escherichia coli are the most widely adopted indicator of faecal pollution and they can also be isolated and identified simply, with their numbers usually being given in the form of faecal Coliforms (FC)/100 ml of wastewater (De Boer and Heuvelink, 2000). Outbreaks of these diseases can occur as a result of, drinking water from wells polluted by a combination of different wastewater microorganism species, eating contaminated fish, or indulging in recreational activities in polluted water bodies containing water borne pathogen. *E. coli* cause urinary tract infection and diarrhoea and *Bacillus* can cause the anthrax. *Pseudomonas aeruginosa* is a common bacterium which can cause disease in animals and humans (Balcht, 1994). *Pseudomonas* can, in rare circumstances, cause community-acquired pneumonias as well as ventilator-associated pneumonias, being one of the most common agents isolated in several studies (Fine *et al.*, 1996). *Staphylococcus aureus* is the most common cause of staph infections. It is a spherical bacterium, frequently found in the nose and skin of a person. *S. aureus* can cause a range of illnesses from minor skin infections, such as pimples, impetigo, boils, cellulitis folliculitis, furuncles, carbuncles, scalded skin syndrome and abscesses, to life-threatening diseases such as pneumonia, meningitis, osteomyelitis, endocarditis, toxic shock syndrome, and septicemia. Its incidence is from skin, soft tissue, respiratory, bone, joint, endovascular to wound infections (Fine *et al.* 1996).

Study Area

For the present study three rivers of Uttarakhand was chosen for the study of microbiological analysis of the water samples taken in different seasons such as summer, post monsoon and winter from different spots. The rivers for study were Alaknanda and Bhagirathi at the confluence place Devprayag.

RESULTS AND DISCUSSIONS

Determination of MPN and SPC Devprayag (Alaknanda)

At this sampling station, it is cleared by MPN method in the year 2012-2013 that MPN was highest in the month of June (summer) i.e. 65 MPN/100 ml in 2012 and 60 MPN/100 ml in 2013. The lowest value were observed in the month of October (winter) i.e. 32 MPN/100 ml in 2012 and 28 MPN/100 ml in 2013. From the SPC Method it is found that during 2012 it was highest in the month of June (summer) i.e 65 SPC/ml x1000 and 60 SPC/ml x1000 in 2013 and lowest were observed in the

month October (winter) i.e. 30 SPC/ml x1000 in 2012 and 35 SPC/ml x1000 in 2013. (Table 1 and Figure 1a and 1b)

Table 1 Seasonal variation in MPN and SPC of microorganism of the sampling Station Devprayag at on the river Alaknanda in the year 2012 and 2013

Season	MPN/100ml (2012)	MPN/100ml (2013)	SPC/ml x 1000 (2012)	SPC/ml x 1000 (2013)
Summer	44	46	40	49
Post-monsoon	15	15	18	18
Winter	18	16	15	10

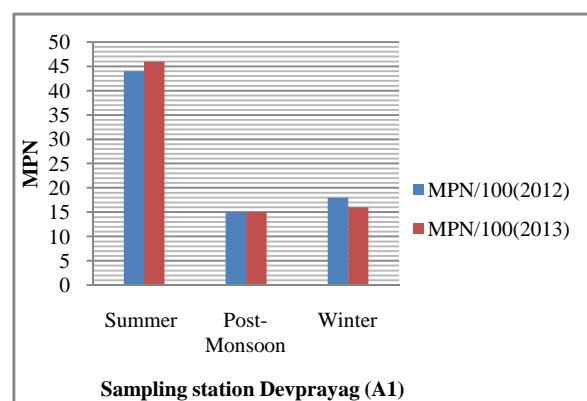


Figure 1a Comparative MPN of the water samples from sampling station Devprayag at on the river Alaknanda during the study period 2012-2013 in different season

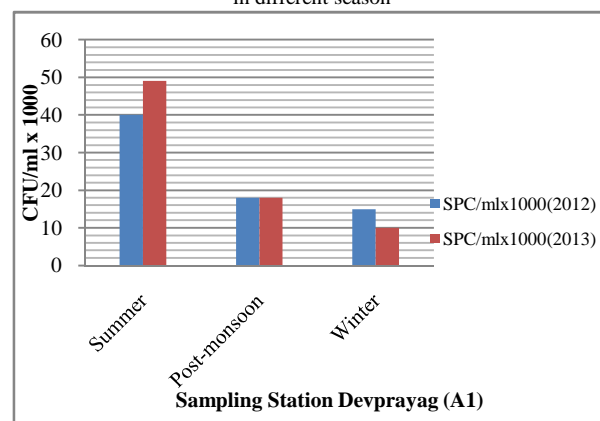


Figure 1b Comparative SPC of the water samples from sampling station Devprayag at on the river Alaknanda during the study period 2012-2013 in different season

Original cell concentration of microorganism at sampling station Devprayag (Alaknanda river)

At sampling station A1 it was found that the original cell concentration of *Escherichia coli* is $2-15 \times 10^3$ /ml in 2012 and $2-16 \times 10^3$ /ml in 2013. The original cell concentration of *Enterobacter aerogenes* was $0-08 \times 10^3$ /ml in 2012 and $2-10 \times 10^3$ /ml in 2013. The cell concentration of *Staphylococcus* was $0-05 \times 10^3$ /ml in 2012 and nil in 2013. The cell concentration of *Shigella* was $0-3 \times 10^3$ /ml in 2012 and $0-05 \times 10^3$ /ml in 2013. *Pseudomonas* was absent in samples during 2012-2013. (Table 2 and Figure 2a and 2b)

Determination of MPN and SPC Devprayag (Bhagirathi)

At sampling station Devprayag (Bhagirathi river), it was observed by MPN method in the year 2012-13 that MPN was highest in the month of June (summer) i.e. 35 MPN/100 ml

during study year 2012 and 34 MPN /100 ml during study year 2013. The lowest value were observed in the month of October (winter) i.e. 18 MPN/100 ml during study year 2012 and 16 MPN/100 ml during study year 2013. From the SPC method it was found that during 2012 SPC of the water sample from sampling station A4 was highest in the month of June (summer) i.e. 15 SPC/ml x1000 during study year 2012 and 16 SPC/ml x1000 during study year 2013 and lowest were observed in the month October (winter) i.e.8 SPC/ml x1000 during study year 2012 and 6 SPC/ml x1000 during study year 2013. All the other related results are represented in the Table 3 and comparative analysis of MPN and SPC is represented in the Figure 3a and 3b respectively.

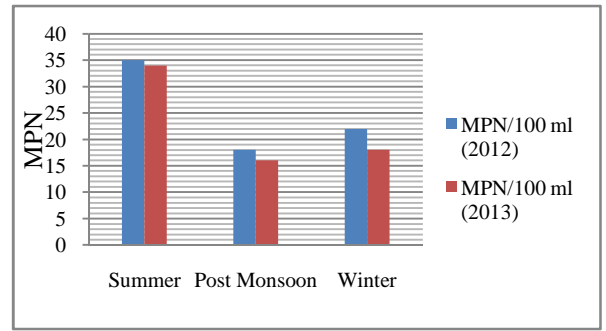


Figure 3a Comparative MPN of the water sample at sampling station Devprayag at river Bhagirathi in different seasons during the study period 2012-2013

Table 2 Original cell concentration of microorganisms in water samples from sampling station Devprayag at on the river Alaknanda during the study period 2012-2013

Year	2012				2013			
	Winter	Post-monsoon	Summer	Average	Winter	Post-monsoon	Summer	Average
Bacteria								
Escherichia coli	08	10	15	11	10	12	16	12.6
Enterobacter aerogenes	05	08	08	7	04	8	10	7.33
Staphylococcus aureus	Nil	Nil	05	1.66	Nil	Nil	Nil	Nil
Salmonella	Nil	04	Nil	1.33	Nil	Nil	04	1.33
Shigella	Nil	03	03	2	Nil	05	Nil	1.2
Psuedomonas	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil

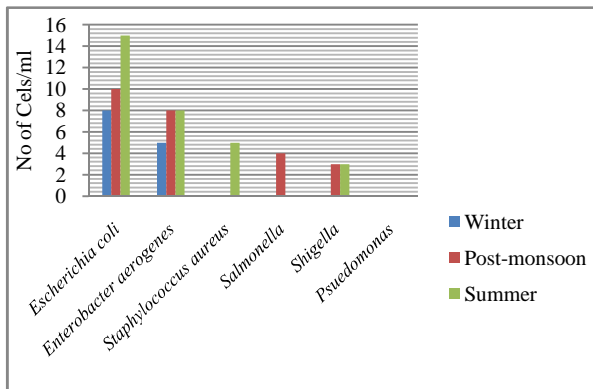


Figure 2a Comparative cell concentration of different microorganisms in water samples from sampling station Devprayag at on the river Alaknanda during the study year 2012 in different season

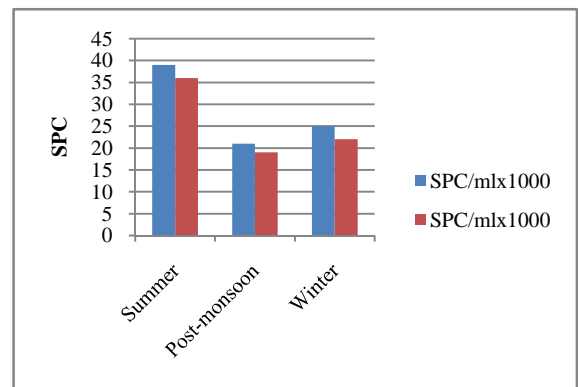


Figure 3b Comparative SPC of the water sample at sampling station Devprayag at river Bhagirathi in different seasons during the study period 2012-2013.

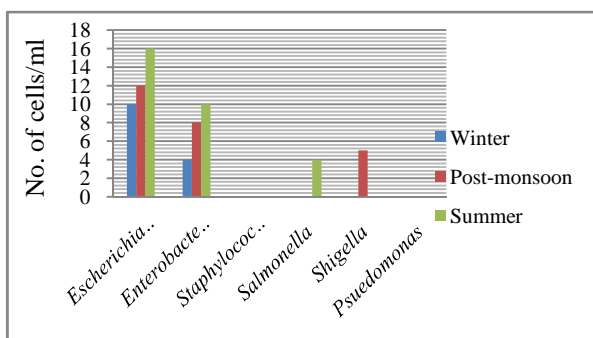


Figure 2b Comparative Diversity Index of different microorganisms in water samples from sampling station Devprayag (A1) during the study year 2013 in different season

Table 3 Seasonal variation in MPN and SPC of microorganism of the sampling Station Devprayag at river Bhagirathi in the year 2012 and 2013

Season	MPN/100ml (2012)	MPN/100ml (2013)	SPC/mlx1000 (2012)	SPC/mlx1000 (2013)
Summer	35	34	39	36
Post-monsoon	18	16	21	19
Winter	22	18	25	22

Original cell concentration of microorganism at sampling station Devprayag (Bhagirathi River)

From the analysis of the water sample from Bhagirathi river it was found that the total concentration of *E. coli* was between 10-19 x 10³/ml during the study year 2012 and between 4-14 x 10³/ml during the study year 2013. The cell concentration of *Enterobacter aerogenes* was between 0-4 x10³/ml during the study year 2012 and between 0-7 x10³/ml during the study year 2013. *Staphylococcus aureus* was absent in all the seasons in the water samples during the study year 2012 as well as also absent in the season of winter and post monsoon during study year 2013. *Staphylococcus aureus* was present in the water samples collected in summer of 2013 showing the cell concentration of 3 x 10³/ml. *Salmonella* was absent in post monsoon season of both the year as well as absent in the winter and summer season of 2013. The Cell concentration was recorded as 4 x 10³ cells/ml in winter and 5 x 10³ cells/ml in the summer of 2012. *Shigella* was also absent in the water samples collected during the winter and post monsoon season of both the year 2012 and 2013.

Table 4 Seasonal variation in the cell concentration of different microorganism from the water samples from River Bhagirathi during the study period 2012-2013

Year	2012			Average	2013			Average
	Winter	Post-monsoon	Summer		Winter	Post-monsoon	Summer	
Bacteria								
Escherichia coli	10	15	19	14.66	10	13	14	13.33
Enterobacter aerogenes	Nil	Nil	04	9.6	05	06	07	9.6
Staphylococcus aureus	Nil	Nil	Nil	10	Nil	Nil	03	7
Salmonella	04	Nil	05	5.33	Nil	Nil		3
Shigella	Nil	Nil	04	1.66	Nil	Nil	04	2.33
Pseudomonas	NIL	NIL	02	0.66	NIL	NIL	03	1

The cell concentration of *Shigella* was observed common as 4×10^3 cells/ml in the summer of both the year 2012 and 2013.

Pseudomonas was absent in winter and post monsoon of both the years. The cell concentration was observed as 2×10^3 cells/ml during study year 2012 and 3×10^3 cells/ml during study year 2013. The related result is also represented in the Table 4 and comparative study is shown in the Figure 4a for 2012 and in Figure 4b for 2013

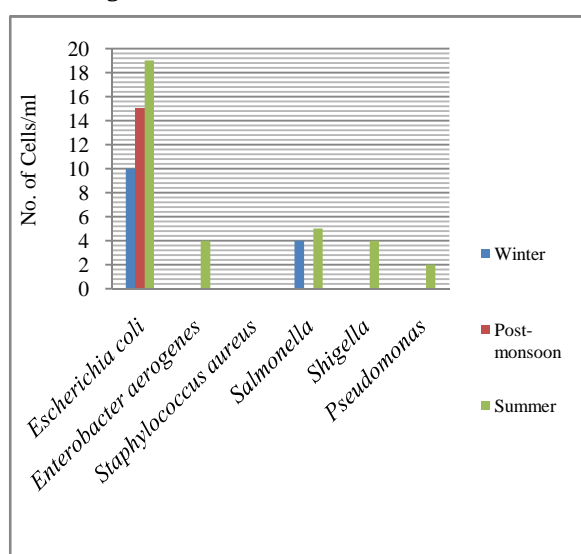


Figure 4a Comparative cell concentration of different microorganisms in the water samples at sampling station Devprayag at river Bhagirathi in different season during the study year 2012

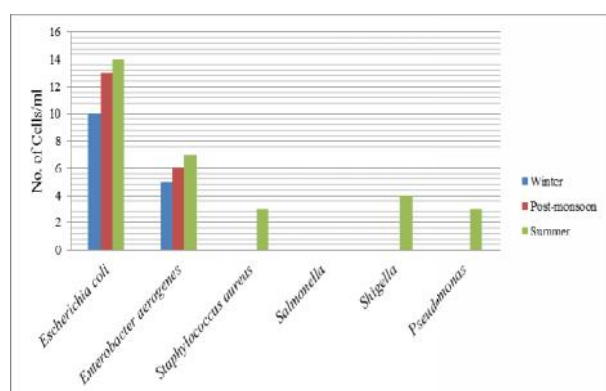


Figure 4b Comparative cell concentration of different microorganisms in the water samples at sampling station Devprayag at river Bhagirathi in different season during the study year 2013

CONCLUSION

Bacteriological analysis showed six bacteria in river water samples and all samples had *E.coli*, as indicator of faecal pollution other bacteria are *Staphylococcus*, *Salmonella*, *Shigella*, *Pseudomonas*, *Enterobacteraerogenes*. During the study, the average values showed that three species i.e. *Escherichia coli*, *Enterobacteraerogenes*, *Staphylococcus* were found to be dominating in river Alaknanda than the river Bhagirathi at Devprayag.

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