

## Research Paper

# Microbiological water quality of the sacred River Bhagirathi, Garhwal Himalaya, India

Nidhi Sharma, Rahul Kumar and Ramesh C. Sharma

### ABSTRACT

Water quality of the sacred River Bhagirathi was evaluated by microbiological and physico-chemical characteristics of water. Monthly water samples were collected from the upper zone (1,158–4,100 m a.s.l.) to lower zone (457–1,158 m a.s.l.) of the river for a period of two years during October, 2013 to September, 2015. The data on microbial density revealed that CFU count was minimum (13,185 CFU.m<sup>-1</sup>) in the winter season and maximum (36,410 CFU.m<sup>-1</sup>) in the monsoon season, when the degradation of water quality was maximum due to mixing of a large amount of allochthonous materials from the catchment area. No total coliform (TC) and fecal coliform (FC) was found in the water samples from Gaumukh (4,100 m) to Gangotri (3,140 m) during the two-year sampling period due to minimum anthropogenic pressure. However, total coliform and fecal coliform were recorded downstream of Gangotri (TC: 980 and FC: 120) and Harshil (TC: 1,100 and FC: 200). Microbial density was recorded to be high in the lower stretch: Uttarkashi (TC: 2,850 and FC: 860) Tehri (TC: 5,000 and FC: 4,200), and Deoprayag (TC: 3,800 and FC: 2,700). A total of 14 bacterial, four actinomycetes, and 11 fungal species were found in the Bhagirathi River.

**Key words** | Bhagirathi, Ganga, Garhwal Himalaya, India, microbiological, water quality

#### Nidhi Sharma

Department of Microbiology,  
J.V. College,  
Baraut-Baghat, Uttar Pradesh,  
India

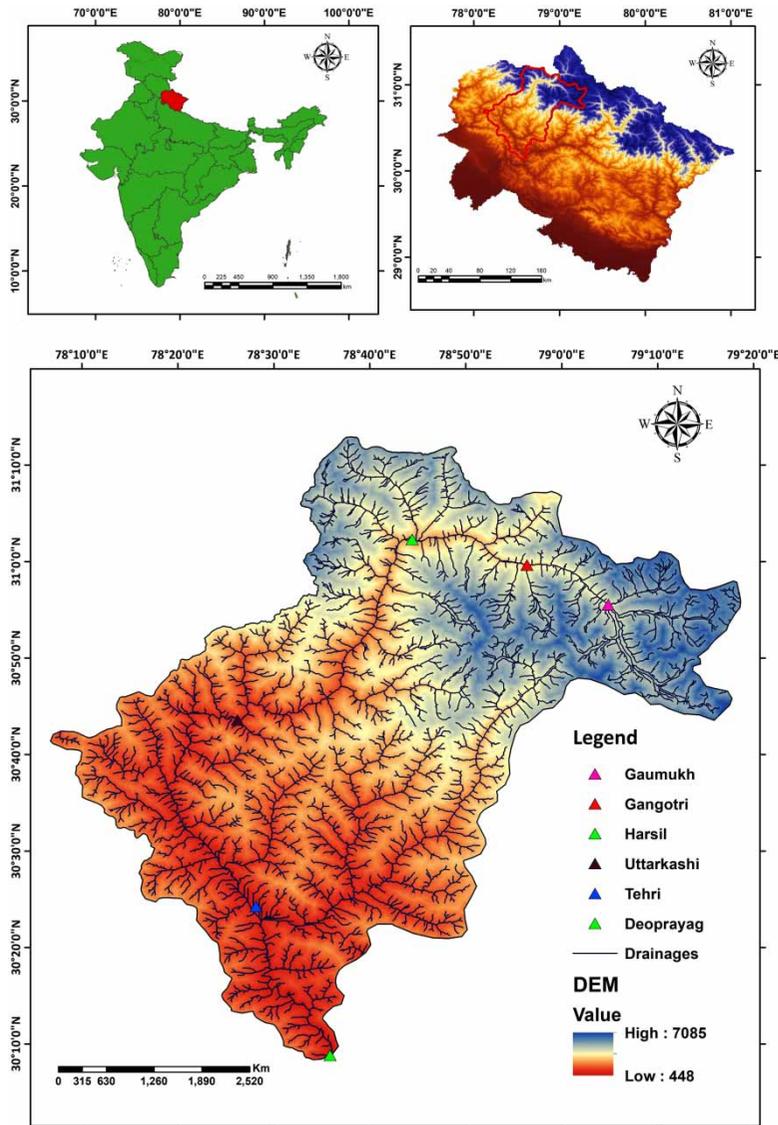
#### Rahul Kumar (corresponding author)

**Ramesh C. Sharma**  
Department of Environmental Sciences,  
H.N.B. Garhwal University (A Central University),  
Srinagar Garhwal 246174, Uttarakhand,  
India  
E-mail: rahul.khadwalia@gmail.com

### INTRODUCTION

The River Bhagirathi is one of the most important parent streams of the Holy River 'Ganga', the National River of India, which is worshipped as 'Mother Ganga' by large number of Hindus. This sacred river is a vital resource for drinking water supplies, irrigation, recreational opportunities, and hydropower generation in the Garhwal Himalaya, Uttarakhand, India (Figure 1). Several cultural and religious activities are associated with this river. More than 0.41 million people (Census 2011) are dwelling in the catchment area of the sacred River Bhagirathi. A substantial number of floating population of pilgrims (0.38 million per annum) visit the world famous Gangotri shrine located in the upper zone of Bhagirathi. Thus, there is an estimated human population (more than 0.8 million) that directly depends on the River Bhagirathi for water supply for

drinking, irrigation, bathing, and taking away the holy water to their homes for performing various worship rituals. There are many places along the stretch of the river where point sources of pollution in terms of discharging untreated sewage and risk of anthropogenic pollution of the sacred River Bhagirathi are common. Pollution of the surface water with organic substances of anthropogenic origin is a global problem (Čučak *et al.* 2016). The River Bhagirathi originates from Gaumukh (4,100 m a.s.l.) and passes through Gangotri (3,140 m a.s.l.), a place dedicated to the world famous temple Gangotri. From Gangotri, the river passes through Harshil (2,620 m a.s.l.), Uttarkashi (1,158 m a.s.l.), and Tehri (640 m a.s.l.), where Asia's highest dam, the Tehri Dam is located. Bhagirathi is joined by the Alaknanda at Deoprayag (457 m a.s.l.) and is finally named as River



**Figure 1** | Sampling sites along the Bhagirathi River.

‘Ganga’ afterwards. The river continues to flow down to Rishikesh and Hardwar, the important religious destinations.

A considerable contribution has been made on the water quality assessment of lower stretches of the Ganga. Sharma & Sharma (2016a) reported microbiological water quality of the lower stretch of the sacred River Alaknanda; another parent stream of Ganga. Bhutiani *et al.* (2016) contributed a study on water quality assessment of Ganga River at Hardwar, and Haritash *et al.* (2016) a water quality assessment of Ganga River at Rishikesh. However, no information is available in the public domain on the water quality of the sacred

River Bhagirathi, the uppermost stretch of the Ganga. Thus, the present contribution on the microbial assessment of water quality of Bhagirathi is of paramount importance for the large number of pilgrims and local inhabitants.

## MATERIALS AND METHODS

The water sampling was undertaken for a period of two years during October, 2013 to September, 2015 from various sites including Gaumukh (4,100 m a.s.l.), Gangotri (3,140 m a.s.l.),

Harshil (2,620 m a.s.l.), Uttarkashi (1,158 m a.s.l.), Tehri (640 m a.s.l.), and Deoprayag (457 m a.s.l.) of the River Bhagirathi. The entire river stretch of Bhagirathi was divided into two zones, the upper zone (1,158–4,100 m a.s.l.) and the lower zone (457–1,158 m a.s.l.). Water samples were collected by dipping autoclaved sample bottles and closing the cap under water to prevent atmospheric exposure for the assessment of water quality of the River Bhagirathi.

The sampling sites were identified based on their specific significance. Gaumukh is the point of origin of the River Bhagirathi. Gangotri is the place where the world famous Hindu shrine Gangotri Temple is located. Harshil is famous for its landscape and apple cultivation, while Uttarkashi is a big town and district headquarters. Tehri is famous for its Tehri Dam and its huge reservoir. Deoprayag is the site of confluence of the Alaknanda and Bhagirathi forming the River Ganga.

Physico-chemical parameters of water like pH, water temperature, free CO<sub>2</sub> and dissolved oxygen were measured at the sampling sites. For the remaining parameters, the water samples were transferred to the Laboratory of Environmental Microbiology, Department of Environmental Sciences, H.N.B. Garhwal University (A Central University), Srinagar Garhwal, Uttarakhand, India. All the physico-chemical parameters, coliform test and microbial diversity were analyzed by following the standard methods outlined in [Golterman et al. \(1978\)](#), [Wetzel & Likens \(1991\)](#), [APHA \(2005\)](#), and [Harley & Prescott \(2002\)](#). Water temperature was measured by carefully dipping the digital thermometer 10 cm below the surface in the river. The temperature range of the digital thermometer was (–50 °C to +300 °C). pH was measured both at the site by using litmus paper and a portable pH meter of Electronics India (Model No. 7011) and in the laboratory by using the Toshcon Bench Top Multiparameter (Model No. TPC-17). Dissolved oxygen was measured by using the Modified Winkler method at the sampling sites. Conductivity and total dissolved solids (TDS) were measured by using the Toshcon Bench Top Multiparameter (Model No. TPC-17). Free CO<sub>2</sub>, total alkalinity, total hardness, calcium hardness, chlorides, and magnesium hardness were measured by following the protocols available in [APHA \(2005\)](#). Nitrates, sulfates, and phosphates were measured by spectrophotometric method

by using the Systronic UV-VIS Spectrophotometer (Model No. 117). Monthly data of physico-chemical parameters of all the sampling sites were pooled seasonally.

Nutrient Agar media (HiMEDIA) was used for the estimation of the numbers of colony forming units (CFUs) of bacteria. Media pH for bacterial isolation was set according to the pH of sampling sites. Sabouraud Dextrose Agar (SDA) and Potato Dextrose Agar (PDA) were used for fungal species. Both media (SDA and PDA) were supplemented with 50 mg.l<sup>-1</sup> each of Streptomycin and Ampicillin to prevent bacterial contamination. Actinomycetes Isolation Agar (AIA) was used for actinomycetes isolation. Specific media, Eosin Methylene Blue (EMB) agar medium was used for the detection of the members of the family, *Enterobacteriaceae* and plates (EMB) were incubated for 24 hrs at 37 °C. *Vibrio* was detected using Trypticase Citric Bile Salts (TCBS) as a selective plating medium. The medium contains sucrose and therefore allows the differentiation of *Vibrio* species such as *Vibrio cholera* (sucrose positive) and *Vibrio haemolyticus* (sucrose negative). To enrich samples for *Vibrio* growth, 1% of Alkaline Peptone Water (APW) was added to the water samples. *Vibrio* species grew on the agar plates as yellowish, round colonies. The numbers of total and fecal coliforms were determined using Most Probable Number (MPN) method. Statistical tables were used to interpret the results of MPN of the bacteria. From each dilution 1 ml was added to each of triplicate tubes containing 5 ml of MacConkey broth. The tubes were incubated at 37 °C for 24 hrs for total coliforms and 44 °C (in water bath) for 24 hrs for fecal coliforms. The positive tubes were streaked on the EMB agar plates and incubated at 37 °C for 24 hrs ([APHA 2005](#)).

To study the morphological characteristics, the purified selected bacterial isolates were Gram stained and observed under the phase contrast microscope (Nikon Eclipse TS100). Moreover, detailed biochemical characterizations were carried out to identify the bacterial isolates up to possible genus or species level. Identification of all the fungal isolates was made by microscopic analysis by using taxonomic keys and standard procedures. Some of the bacterial cultures isolated from the River Bhagirathi were sent to the Microbial Type Culture Collection and Gene Bank, MTCC (Institute of Microbial Technology), Chandigarh for identification.

Mean seasonal/annual variations in fecal coliform and total CFU from both the zones of the river were calculated.

## RESULTS AND DISCUSSION

The monthly data on physico-chemical parameters of water of the River Bhagirathi from all the sampling sites were pooled seasonally (Tables 1 and 2). The water temperature in the upper zone (1,158–4,100 m a.s.l.) of the River Bhagirathi fluctuated from a minimum  $7.00 \pm 1.24$  °C (during winter) to a maximum of  $13.6 \pm 0.007$  °C (during monsoon season). The water temperature during the summer season did not attain peak due to mixing of melting snow causing a rise in the atmospheric temperature. The water was almost clear ( $1.30 \pm 0.007$  NTU to  $2.80 \pm 0.4$  NTU) during autumn and winter seasons in the upper zone of the River Bhagirathi. However, maximum turbid water ( $174.62 \pm 171.83$  NTU) was recorded during the monsoon season due to mixing of heavy silt, sand, and clay caused by heavy precipitation in the catchment area of the river. Transparency in the upper zone of the River Bhagirathi was found to be minimum ( $0.50 \pm 0.01$  m) during the monsoon season and maximum ( $1.80 \pm 0.00$  m) during the winter season. Conductivity of water in the upper zone of the River

Bhagirathi attained peak ( $0.17 \pm 15.0$   $\mu\text{S}\cdot\text{cm}^{-1}$ ) in summer season and a dip ( $0.07 \pm 0.03$   $\mu\text{S}\cdot\text{cm}^{-1}$ ) in the monsoon season. The decreasing trend of conductivity during the winter season may be due to precipitation of calcium carbonates, while its higher value in monsoon and autumn seasons may be due to surface evaporation of water resulting in an increase in salt concentration. The total dissolved solids (TDS) denote mainly various kinds of minerals present in water. The TDS were found to be maximum ( $89.00 \pm 2.84$   $\text{mg}\cdot\text{l}^{-1}$ ) during the monsoon season and minimum ( $7.25 \pm 3.21$   $\text{mg}\cdot\text{l}^{-1}$ ) in the winter season. However, high TDS ( $258.50 \pm 12.02$   $\text{mg}\cdot\text{l}^{-1}$ ) in Baldi River, Doon Valley, Garhwal Himalaya was also recorded in the monsoon season by Singh & Sharma (2016). As both the rivers (Baldi and Bhagirathi) are Himalayan rivers, it is necessary to compare their findings to assess the water quality. It is essential to compare the findings of both the rivers. The increasing pattern of TDS in the monsoon season may be due to addition of inorganic salts and organic matter carried along with rain water and surface run-off. However, the pH ranged from  $7.30 \pm 0.10$  to  $7.79 \pm 0.07$  during the entire period of study, which shows the alkaline nature of the Bhagirathi water. Almost similar results were found by Bora & Goswami (2016) in Kolong River, Assam. Hydrogen ion concentration (pH) is the measure of the intensity of acidity or

**Table 1** | Mean ( $\bar{x} \pm \text{SD}$ ) seasonal variations in physico-chemical environmental variables in upper zone (1,158–4,100 m a.s.l.) of the River Bhagirathi

Parameters	Autumn (Oct–Nov)	Winter (Dec–Mar)	Summer (Apr–Jun)	Monsoon (Jul–Sep)
Water temperature (°C)	$10.61 \pm 1.41$	$7.00 \pm 1.24$	$12.23 \pm 0.63$	$13.6 \pm 0.007$
Turbidity (NTU)	$1.30 \pm 0.007$	$2.80 \pm 0.41$	$25.63 \pm 37.25$	$174.62 \pm 171.83$
Transparency (m)	$1.53 \pm 0.11$	$1.80 \pm 00.00$	$1.38 \pm 0.29$	$0.50 \pm 0.001$
Conductivity ( $\mu\text{S}\cdot\text{cm}^{-1}$ )	$0.14 \pm 0.01$	$0.13 \pm 0.009$	$0.17 \pm 15.00$	$0.07 \pm 0.03$
TDS ( $\text{mg}\cdot\text{l}^{-1}$ )	$19.50 \pm 2.13$	$7.25 \pm 3.21$	$31.35 \pm 20.72$	$89.00 \pm 2.84$
pH	$7.50 \pm 0.04$	$7.30 \pm 0.10$	$7.55 \pm 0.10$	$7.79 \pm 0.07$
Dissolved oxygen ( $\text{mg}\cdot\text{l}^{-1}$ )	$12.80 \pm 0.80$	$16.53 \pm 0.40$	$10.47 \pm 0.057$	$10.30 \pm 0.30$
Free CO <sub>2</sub> ( $\text{mg}\cdot\text{l}^{-1}$ )	$0.014 \pm 0.002$	$0.005 \pm 0.006$	$0.021 \pm 0.002$	$0.030 \pm 0.002$
Phosphates ( $\text{mg}\cdot\text{l}^{-1}$ )	$0.09 \pm 0.01$	$0.06 \pm 0.01$	$0.20 \pm 0.01$	$0.33 \pm 0.035$
Nitrates ( $\text{mg}\cdot\text{l}^{-1}$ )	$0.14 \pm 0.01$	$0.10 \pm 0.13$	$0.30 \pm 0.15$	$0.50 \pm 0.03$
Sulfates ( $\text{mg}\cdot\text{l}^{-1}$ )	$0.60 \pm 0.14$	$0.55 \pm 0.10$	$0.90 \pm 0.05$	$1.0 \pm 0.03$
Chloride ( $\text{mg}\cdot\text{l}^{-1}$ )	$1.30 \pm 0.50$	$0.75 \pm 0.36$	$1.45 \pm 0.15$	$2.30 \pm 0.30$
Sodium ( $\text{mg}\cdot\text{l}^{-1}$ )	$3.21 \pm 0.45$	$2.45 \pm 0.50$	$3.45 \pm 0.12$	$4.14 \pm 1.18$
Potassium ( $\text{mg}\cdot\text{l}^{-1}$ )	$2.45 \pm 0.007$	$1.75 \pm 0.20$	$3.22 \pm 1.20$	$3.78 \pm 1.60$

**Table 2** | Mean ( $\bar{x} \pm SD$ ) seasonal variations in physico-chemical environmental variables in lower zone (457–1,158 m a.s.l.) of the Bhagirathi River

Parameters	Autumn (Oct–Nov)	Winter (Dec–Mar)	Summer (Apr–Jun)	Monsoon (Jul–Sep)
Water temperature ( $^{\circ}\text{C}$ )	15.35 $\pm$ 1.25	11.20 $\pm$ 0.48	17.80 $\pm$ 1.90	17.85 $\pm$ 0.32
Turbidity (NTU)	14.25 $\pm$ 7.11	12.15 $\pm$ 4.17	89.50 $\pm$ 37.25	625.23 $\pm$ 210.12
Transparency (m)	0.39 $\pm$ 0.27	0.65 $\pm$ 0.15	0.19 $\pm$ 0.11	0.03 $\pm$ 0.02
Conductivity ( $\mu\text{S}\cdot\text{cm}^{-1}$ )	0.14 $\pm$ 0.03	0.16 $\pm$ 0.005	0.17 $\pm$ 0.04	0.09 $\pm$ 7.07
TDS ( $\text{mg}\cdot\text{l}^{-1}$ )	36.52 $\pm$ 3.24	22.45 $\pm$ 2.80	40.50 $\pm$ 20.18	100.0 $\pm$ 20.14
pH	7.75 $\pm$ 0.11	7.45 $\pm$ 0.08	7.62 $\pm$ 0.07	7.85 $\pm$ 0.04
Dissolved oxygen ( $\text{mg}\cdot\text{l}^{-1}$ )	10.10 $\pm$ 0.35	11.17 $\pm$ 0.52	9.95 $\pm$ 0.39	9.25 $\pm$ 0.13
Free $\text{CO}_2$ ( $\text{mg}\cdot\text{l}^{-1}$ )	2.10 $\pm$ 0.16	1.72 $\pm$ 0.50	1.80 $\pm$ 0.45	2.53 $\pm$ 0.02
Phosphates ( $\text{mg}\cdot\text{l}^{-1}$ )	0.21 $\pm$ 0.07	0.18 $\pm$ 0.08	0.54 $\pm$ 0.35	0.85 $\pm$ 0.12
Nitrates ( $\text{mg}\cdot\text{l}^{-1}$ )	0.40 $\pm$ 0.04	0.25 $\pm$ 0.10	0.45 $\pm$ 0.08	0.78 $\pm$ 0.04
Sulfates ( $\text{mg}\cdot\text{l}^{-1}$ )	1.62 $\pm$ 0.26	0.97 $\pm$ 0.41	2.19 $\pm$ 0.28	2.53 $\pm$ 1.05
Chloride ( $\text{mg}\cdot\text{l}^{-1}$ )	2.90 $\pm$ 0.28	2.54 $\pm$ 0.25	3.10 $\pm$ 0.12	3.84 $\pm$ 0.22
Sodium ( $\text{mg}\cdot\text{l}^{-1}$ )	4.25 $\pm$ 0.35	3.51 $\pm$ 0.19	5.095 $\pm$ 1.25	6.63 $\pm$ 1.22
Potassium ( $\text{mg}\cdot\text{l}^{-1}$ )	3.25 $\pm$ 0.002	3.21 $\pm$ 0.18	3.58 $\pm$ 0.0	4.26 $\pm$ 0.006

alkalinity in water. The dissolved oxygen is considered as the most important chemical parameter, which reflects the quality of water and its use. The dissolved oxygen recorded from the upper zone of the River Bhagirathi revealed that it was maximum ( $16.53 \pm 0.40 \text{ mg}\cdot\text{l}^{-1}$ ) in winter and minimum ( $10.30 \pm 0.30 \text{ mg}\cdot\text{l}^{-1}$ ) in the monsoon season. Free  $\text{CO}_2$  is an end product of respiration and aerobic decomposition of organic matter (Welch 1952). It was maximum ( $0.030 \pm 0.002 \text{ mg}\cdot\text{l}^{-1}$ ) in the monsoon season and minimum ( $0.005 \pm 0.006 \text{ mg}\cdot\text{l}^{-1}$ ) in the winter season. The concentration of phosphates was recorded as minimum ( $0.06 \pm 0.001 \text{ mg}\cdot\text{l}^{-1}$ ) in the winter season and maximum ( $0.33 \pm 0.035 \text{ mg}\cdot\text{l}^{-1}$ ) during the monsoon season. Phosphates enter fresh water from atmospheric precipitation and from groundwater and surface run-off. Nitrates represent the highest oxidized form of nitrogen. It is the most predominant form of inorganic nitrogen entering freshwater, groundwater, and precipitation. Concentration of nitrates was found to be minimum ( $0.10 \pm 0.13 \text{ mg}\cdot\text{l}^{-1}$ ) in the winter season and maximum ( $0.50 \pm 0.03 \text{ mg}\cdot\text{l}^{-1}$ ) in the monsoon season. The same trend in the concentration of chlorides was recorded. Sodium is a naturally occurring element in water. In natural water, the major source of sodium is from weathering of various rocks. The concentration of sodium was minimum

( $2.45 \pm 0.50 \text{ mg}\cdot\text{l}^{-1}$ ) in the winter season and maximum ( $4.17 \pm 1.18 \text{ mg}\cdot\text{l}^{-1}$ ) in the monsoon season. The same trend in the concentration of potassium was recorded. Potassium is another important cation occurring naturally (Table 1).

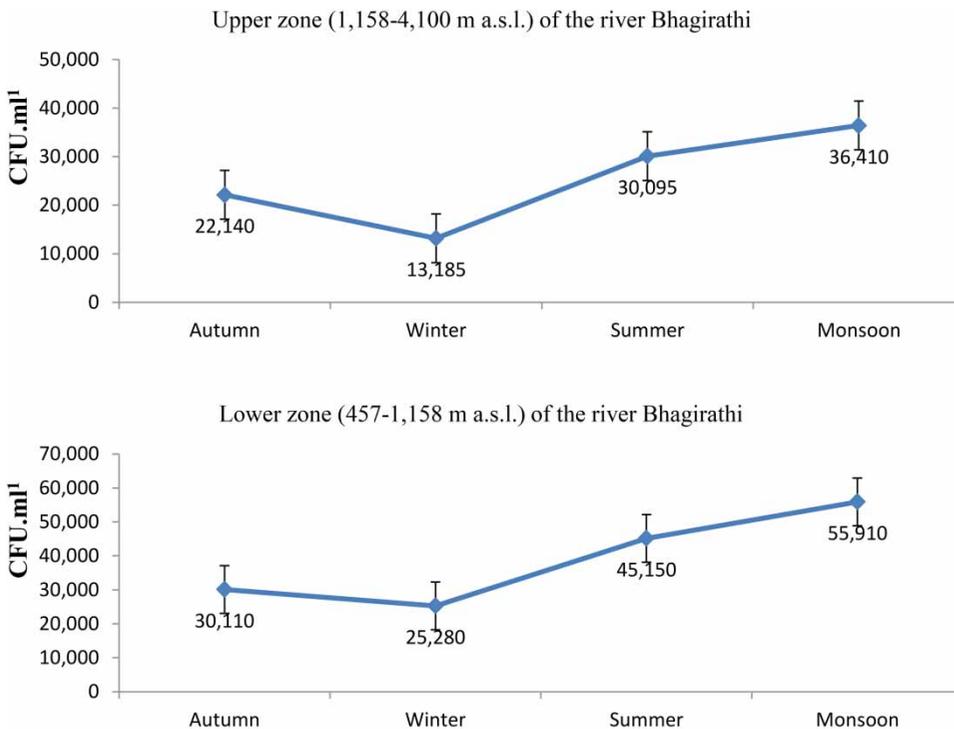
The physico-chemical environmental variables from the lower zone (457–1,158 m a.s.l.) of the River Bhagirathi revealed that the temperature, turbidity, conductivity, total dissolved solids, free  $\text{CO}_2$ , phosphates, nitrates, sodium, and potassium were found to be minimum in the winter season and maximum in the monsoon season (Table 2). However, the dissolved oxygen was found to be maximum in the winter season and minimum in the monsoon season. A similar trend of fluctuation in the concentration of sulfates and chlorides was also recorded in the lower zone of the Bhagirathi.

The data on physico-chemical parameters of River Bhagirathi revealed the poor quality of water in the lower zone in comparison to the upper zone (Tables 1 and 2). Seasonally, the poorest quality of water in both the zones was recorded in the monsoon season. The same findings were reported by Bhutiani et al. (2016) on Ganga River at Harwar, downstream to Deoprayag. Haritash et al. (2016) also reported the degraded water quality of Ganga River at Rishikesh, downstream to Deoprayag.

## Microbial data

The microbial diversity dwelling in the sacred River Bhagirathi is mainly represented by bacteria, fungi, and actinomycetes. Microbial density and diversity were recorded as minimum in the upper zone of Bhagirathi. The number of pathogenic bacteria were high in the upper zone of the River Bhagirathi (Gaumukh, Gangotri, Harshil, Uttarkashi). However, the microbial density and diversity increased from high altitude to lower altitude. Rapid urban growth and sprawling sub-urban development, conversion of forest land to urban land, dam construction activities and other anthropogenic activities including road construction and widening in the catchment area of the river have contributed to degradation of the water quality of this sacred river. A high nutrient load in terms of discharge of untreated sewage, influx of organic matter from the horticulture and agriculture practices in the catchment area has contributed towards the enrichment of microbial growth. Pathogenic bacteria (*Escherichia coli*, *Vibrio cholera*, and *Klebsiella* sp.) were also abundant in the lower altitude stretch of the river. A perusal of data on the density and

diversity of microbes at all the sites of the River Bhagirathi revealed that it increased from high to lower altitude. A similar observation was made by Sharma & Sharma (2016b) on the microbes of the sacred River Alaknanda. Mean seasonal variations in colony forming units (CFU.ml<sup>-1</sup>) of bacterial density recorded at 28 ± 2 °C in upper and lower zones of the River Bhagirathi are presented in Figure 2. A perusal of the data revealed that the minimum (13,185 CFU.ml<sup>-1</sup>) was recorded in the winter season and then it increased during summer (30,095 CFU.ml<sup>-1</sup>) and attained the peak (36,410 CFU.ml<sup>-1</sup>) during the monsoon season, when the maximum degradation in the water quality was observed. Due to the onset of autumn and winter seasons, the quality of water improved substantially and the density of the bacteria decreased (22,140 CFU.ml<sup>-1</sup>) significantly during autumn from the monsoon season. The same seasonal trend with higher values in CFU.ml<sup>-1</sup> of bacterial density was recorded in the lower zone of the River Bhagirathi (Figure 2). The water quality improved during the onset of autumn and the density of bacterial growth also declined due to improved conditions of the aquatic environment.



**Figure 2** | Mean ( $\bar{x} \pm SD$ ) seasonal variations in colony forming units (CFU.ml<sup>-1</sup>) of bacteria in high and lower zones of the sacred River Bhagirathi.

No total coliform (TC) and fecal coliform (FC) bacteria were found at Gaumukh and Gangotri (upstream) during the period of study (October, 2013 to September, 2015), which may be due to the pristine environment and negligible human interference at these sites (Figure 3). The presence of FC bacteria indicates that the water is contaminated with human excreta or animal waste, while the TC counts indicate water contamination due to several anthropogenic activities in the catchment area (Zamxaka et al. 2004). The TC and FC bacterial density were found to be increasing from Gangotri downstream (TC: 980 and FC: 120) to Harshil (TC: 1,100 and FC: 200), Uttarkashi (TC: 2,850 and FC: 860), Tehri (TC: 5,000 and FC: 4,200), and Deoprayag (TC: 3,800 and FC: 2,700). This may be due to dam construction activities, rapid urban growth, discharge of domestic sewages, and several anthropogenic activities in the catchment area. The settlement of people near the river, open defecation, several Hindu rituals (bathing, immersion of ashes after funeral, leftovers of worship, flowers, incense sticks, wrappers, etc., and several other anthropogenic activities might have contributed to the contamination of river water. The presence of pathogenic microorganisms at these sites of the River Bhagirathi may also be due to the mixing of organic matter, discharge of untreated sewage and domestic waste with the Bhagirathi River.

### Microbial diversity in River Bhagirathi

The diversity of the microbes in the River Bhagirathi has shown a definite trend in the presence of specific microbes from high altitude to lower altitude. Overall, 14 species of bacteria (*Bacillus circulans*, *Escherichia coli*, *Enterobacter*

*aerogens*, *Klebsiella* sp., *Micrococcus* sp., *Microbacterium schleiferi*, *Paenibacillus azatofixans*, *Pseudomonas aeruginosa*, *Pseudomonas fluorescens*, *Shigella dysenteriae*, *Staphylococcus aureus*, *Streptococcus fecalis*, *Vibrio cholerae*, and *Yersinia enterocolitica*) were recorded from the sacred River Bhagirathi (Table 3). However, four species (*Streptomyces clavifer*, *Streptomyces rangoon*, *Streptomyces* spp., *Nocardia* spp.) of actinomycetes were recorded from the Bhagirathi. Fungi were represented by 11 species (*Achlya* spp., *Alternaria* sp., *Aspergillus flavus*, *Aspergillus niger*, *Cladosporium* sp., *Curvularia* sp., *Phoma* sp., *Penicillium* spp., *Rhizopus* sp., *Trichoderma* sp., and *Saprolegnia* sp.). Microbial diversity in the River Bhagirathi was found to be minimum ( $\alpha$ -diversity: 08) at Gaumukh and maximum ( $\alpha$ -diversity: 29) in Tehri Dam Reservoir and Deoprayag, where the pressure of anthropogenic activities in the River Bhagirathi is maximum (Table 3). Several sources of point pollution in terms of mixing of domestic sewage and other effluents are common at these places. Several religious rituals like immersion of ashes, funerals, bathing, and dumping of municipal solid waste are also very common at the river bank of Bhagirathi. Some pilgrims offer milk, curds, butter, flowers, ashes of worship, idols, ashes of departed ones, and discharge other worship materials which cause the environmental degradation of the river. During mass bathing of pilgrims, they carry and spread pathogenic microbes into the river. It has been observed that the abundance of all microbes including fungi substantially increased during the rainy season and post-flood periods. Most of the fungi were found to be mesophilic and growing at the temperature of  $28 \pm 2^\circ\text{C}$ . Surface run-off brings a number of geofungi including the species *Alternaria*, *Aspergillus*,

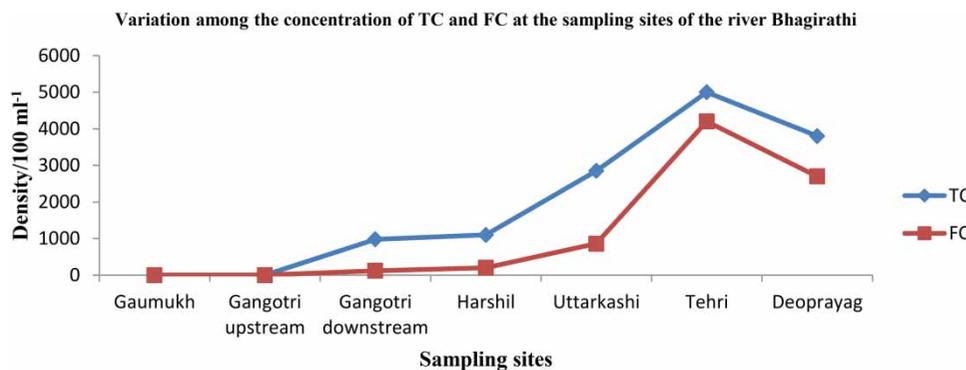


Figure 3 | Annual mean ( $\bar{x} \pm \text{SD}$ ) variations in total coliform (TC) and fecal coliform (FC) bacteria at different sampling sites of the sacred River Bhagirathi.

**Table 3** | Microbial diversity in the sacred River Bhagirathi

S. no.	Microbes	S <sub>1</sub>	S <sub>2</sub>	S <sub>3</sub>	S <sub>4</sub>	S <sub>5</sub>	S <sub>6</sub>
<b>A Bacteria</b>							
1	<i>Bacillus circulans</i>	+	+	+	+	+	+
2	<i>Escherichia coli</i>	+	+	+	+	+	+
3	<i>Enterobacter aerogens</i>	-	+	+	+	+	+
4	<i>Klebsiella</i> sp.*	-	-	-	+	+	+
5	<i>Microbacterium schleiferi</i>	+	+	+	+	+	+
6	<i>Micrococcus</i> sp.*	+	+	-	+	+	+
7	<i>Paenibacillus azatofixans</i>	-	-	-	-	+	+
8	<i>Pseudomonas aeruginosa</i>	-	-	+	+	+	+
9	<i>Pseudomonas fluorescens</i>	-	+	+	+	+	+
10	<i>Shigella dysenteriae</i>	-	-	-	+	+	+
11	<i>Staphylococcus aureus</i>	+	+	+	+	+	+
12	<i>Streptococcus faecalis</i>	-	+	+	+	+	+
13	<i>Vibrio cholera</i>	-	-	-	+	+	+
14	<i>Yersinia enterocolytica</i>	-	-	-	-	+	+
<b>B Actinomycetes</b>							
1	<i>Streptomyces clavifer</i>	-	-	-	-	+	+
2	<i>Streptomyces rangoon</i>	-	-	-	+	+	+
3	<i>Streptomyces</i> spp.*	+	+	+	+	+	+
4	<i>Nocardia</i> spp.*	+	+	+	+	+	+
<b>C Fungi</b>							
1	<i>Achlya</i> spp.*	+	+	+	+	+	+
2	<i>Alternaria</i> sp.*	+	+	-	+	+	+
3	<i>Aspergillus flavus</i>	-	+	+	+	+	+
4	<i>Aspergillus niger</i>	-	+	+	+	+	+
5	<i>Cladosporium</i> sp.*	-	+	+	+	+	+
6	<i>Curvularia</i> sp.*	-	+	+	+	+	+
7	<i>Penicillium</i> spp.*	+	+	+	+	+	+
8	<i>Phoma</i> sp.*	-	+	+	+	+	+
9	<i>Saprolegnia</i> sp.*	-	+	+	+	+	+
10	<i>Rhizopus</i> sp.*	-	-	-	+	+	+
11	<i>Trichoderma</i> sp.*	-	+	+	+	+	+
<b><math>\alpha</math>-Diversity</b>		<b>8</b>	<b>19</b>	<b>19</b>	<b>26</b>	<b>29</b>	<b>29</b>

S<sub>1</sub>: Gaumukh (3,812 m a.s.l.); S<sub>2</sub>: Gangotri (3,140 m a.s.l.); S<sub>3</sub>: Harshil (2,620 m a.s.l.); S<sub>4</sub>: Uttarkashi (1,158 m a.s.l.); S<sub>5</sub>: Tehri (640 m a.s.l.); S<sub>6</sub>: Deoprayag (457 m a.s.l.).

\*Unidentified at species level; +: present; -: absent.

*Cladosporium*, *Penicillium*, *Trichoderma*, *Rhizopus*, etc. The  $\alpha$ -diversity of microbes was found to be highest at Uttarkashi, Tehri, and Deoprayag along with many pathogenic bacteria (*Escherichia coli*, *Vibrio cholera*, and *Klebsiella* sp.) in the lower zone of the River Bhagirathi (Table 3).

## CONCLUSION

It is concluded that the myriad of physico-chemical environmental variables and nutrient load from various sources into the river are responsible for seasonal variations in the density and diversity in the Bhagirathi. The quality of water of the Bhagirathi was poor in the lower zone in comparison to the upper zone. Seasonally, the water quality was most degraded in the monsoon season. The presence of a high density of total and fecal coliforms in the lower stretches of Bhagirathi is also an indication of poor water quality of water of the sacred River Bhagirathi. Minimum  $\alpha$ -diversity (8) was found at Gaumukh and maximum (29) at Tehri and Deoprayag. Pathogenic bacteria (*Escherichia coli*, *Vibrio cholera*, and *Klebsiella* sp.) were also abundant in the lower altitude stretch of the river.

## ACKNOWLEDGEMENTS

The authors gratefully acknowledge the financial assistance given by Department of Biotechnology (DBT), New Delhi, Govt. of India in the form of a major research project (File No. BT/PR/3452/PID/4/153/2002).

## REFERENCES

- APHA 2005 *Standard Methods for the Examination of Water and Wastewater*, 21st edn. American Public Health Association, American Water Works Association, Water Environment Federation, Washington, DC, USA.
- Bhutiani, R., Khanna, D. R., Kulkarni, D. B. & Ruhela, M. 2016 *Assessment of Ganga river ecosystem at Hardwar, Uttarakhand, India with reference to water quality indices. Applied Water Science* **6**, 107–113.
- Bora, M. & Goswami, D. C. 2016 Water quality assessment in terms of water quality index (WQI): case study of the Kolong River, Assam, India. *Applied Water Science* **7** (6), 3125–3135.
- Census 2011 Primary Census Abstracts, Registrar General of India, Ministry of Home Affairs, Government of India. Available at [http://www.censusindia.gov.in/2011census/PCA/pca\\_highlights/pe\\_data.html](http://www.censusindia.gov.in/2011census/PCA/pca_highlights/pe_data.html), accessed on 14 July 2013.
- Čučak, D. I., Marković, N. V. & Radnović, D. V. 2016 *Microbiological water quality of Nišava River. Water Science & Technology: Water Supply* **16** (6), 1668–1673. Doi: 10.2166/ws.2016.089.

- Golterman, H. L., Clymo, R. S. & Ohnstad, M. A. N. 1978 *Method for Physical and Chemical Analysis of Freshwaters*. IBP Handbook No. 8, Blackwell Scientific Publication, Oxford, UK, pp. 1–72.
- Haritash, A. K., Gaur, S. & Garg, S. 2016 [Assessment of water quality and suitability analysis of river Ganga at Rishikesh, India](#). *Applied Water Science* **6**, 383–392.
- Harley, J. P. & Prescott, L. M. 2002 *Laboratory Exercises in Microbiology*, 5th edn. McGraw-Hill Higher Education, New York, pp. 1–466.
- Singh, N. & Sharma, R. C. 2016 Assessment of physico-chemical parameters of mountain River Baldi. *Garhwal Himalaya. International Journal of Fisheries and Aquatic Studies* **4** (2B), 88–93.
- Sharma, N. & Sharma, R. C. 2016a Microbial and physico-chemical assessment of the sacred river Alaknanda at lower stretch, Uttarakhand, India. *Journal of Plant Development Sciences* **8** (6), 285–289.
- Sharma, N. & Sharma, R. C. 2016b Microbiological and physico-chemical assessment of the sacred river Alaknanda, India. *International Journal of Agricultural Invention* **1** (1), 102–107.
- Welch, P. S. 1952 *Limnology*. McGraw-Hill Book Co. Inc., New York.
- Wetzel, R. G. & Likens, G. E. 1991 *Limnological Analysis*, 2nd edn. Springer-Verlag, New York, pp. 1–391.
- Zamxaka, M., Pironcheva, G. & Muijima, N. 2004 [Microbiological and physico-chemical assessment of the quality of domestic water sources in selected rural communities of the Eastern Cape Province, South Africa](#). *Water SA* **30** (3), 333–340.

First received 27 October 2017; accepted in revised form 28 September 2018. Available online 12 October 2018