

Assessment of physiochemical and microbial concentrations in different fresh waters in Tiruchirappalli city: A special emphasize on their correlation

M. Anitha* and S. Ravikumar

Department of Microbiology, Ponnaiyah Ramajayam College, Vallam, Thanjavur, Tamil Nadu – 613 403, India.

Abstract

The six fresh water samples belonging to three different sources (Cauvery river water, ground water and bottle drinking water) from Tiruchirappalli city were collected (bi-monthly sampling) during April - June 2014 for physiochemical and microbiological study. Bacterial and physiochemical parameters levels were two- to 10-fold higher in river water sample than in ground and bottle water due to discharges of municipal sewage and human activities. The elevated bacterial communities in May month indicated that which received waste materials from landside and more visits due to the lack of water scarcity. The higher pollution index (PI) ratio (>1) were observed in river water samples (than the ground and bottle water) in all the three months indicate the human fecal matters were responsible for water pollution. The order of decreasing microbial and physiochemical values were: April month > May month > June month. The correlation proved that physicochemical parameters were strongly supported to microbial communities due to addition of rich organic content in the water bodies from different pollution sources. Based on the report, this study was suggesting that throughout impoundment is needed to protect fresh water sources.

Keywords: Bacteriological parameters, Physiochemical parameters, Cauvery river water, Tiruchirappalli city, Bottle water

1. Introduction

The Rivers are important freshwater resources and most developmental activities are still dependent upon them which used in every sector of development like agriculture, industry, transportation, aquaculture, public water supply, etc. Naturally, the freshwater resources have also been used for cleaning and disposal purposes¹. Visits by people and livestock to surface water systems are common in developing countries and most of the peoples lack access to portable clean water². Ground water is one of the aquatic biotope that harbours varied microflora. The evolution of the microflora depends on several factors such as solid particles mobility, hydrodynamics, hydrochemistry, etc³. Unfortunately, freshwater are polluted by indiscriminate sewage discharge, massive industrial waste, agricultural practices and human activities, which affect its microbiological quality. Human health risks associated with microbial vectors are a serious problem in aquatic areas and may increase dramatically in the future^{4, 5}. Pathogenic bacteria and pollution indicators have been used worldwide to indicate if human wastes have contaminated a water body and are found in elevated concentrations in human feces⁵. Unsanitary means of disposing human waste and fecal droppings from livestock/birds are routes through which fecal matter may enter aquatic systems. It degrades water quality due to the possible introduction of pathogens, nutrients and organic matter^{6, 2}. Billions of people especially children are worldwide suffer from diseases due to the water pollution⁷. Safe drinking

water would prevent around 2.5 million deaths from diarrheal diseases, 150 million cases of schistosomiasis and 75 million cases of trachoma every year⁸. The freshwater quality is not only assessed by the physicochemical characteristics which is also conforms to microbiological water quality⁹. The aim of this research was to determine the concentration of bacterial and physiochemical parameters from three different water samples in the populous city in order to find their sources such as fecal/sewage/industrial contributions. Further, the statistical approach helps to understand the relationship between the parameters with respect to pollution.

2. Materials and methods

2.1 Sampling site

The Tiruchirappalli is the fourth largest municipal corporation and the biggest urban agglomeration in the state. Tiruchirappalli sits almost at the physiographic centre of the state. It covers 167.23 sq km (64.57 sq mi) and is completely surrounded by agricultural fields. Densely populated industrial and residential areas have recently been built in all part of the city and about 1 million people are living in this city. As Tiruchirappalli is on the Deccan Plateau the days are extremely warm and dry; evenings are cooler because of cold winds that blow from the south-east. From June to September, the city experiences a moderate climate tempered by heavy rain and thundershowers. Rainfall is heaviest between October and December because of the north-east

monsoon winds, and from December to February the climate is cool and moist. The average annual rainfall is 841.9 mm (33.15 in), slightly lower than the state's average of 945 mm (37.2 in). Fog and dew are rare and occur only during the winter season.

2.2 Sampling

The six water samples from three different sources in Tiruchirappalli city were collected during April–June 2014 (Sampling dates are: 7th and 22nd of April, May and June) for bacteriological (pollution indicators and pathogenic bacteria), and physiochemical analysis. The three different water samples were Cauvery river water, ground water and packed/bottle drinking water and were collected from Chinthamani region, Chattram bus stand and super market, respectively. The samples such as river water, ground water and bottle drinking water (commercial) were marked as S1, S2 and S3, respectively. The sampling was regularly carried out in 15 days interval. The river water samples were collected from 0 to 20 cm below the surface^{9, 10}. The ground and bottle water were collected from municipal hand pumps (the depth was unknown) and super market. The 2000 mL of water samples were collected with a 2500 mL sterile container in each location at all the period.

2.3 Physiochemical analysis

The physiochemical parameters, i.e., pH, electrical conductivity (EC) and total dissolved solids (TDS) were measured using field kit (Thermo Orion 5-Star pH Multi-Meter) on the site and the concentrations of soluble cations and anions (Ca²⁺,

Mg²⁺, Na⁺, K⁺, CO₃⁻, HCO₃⁻, Cl⁻ and SO₄²⁻) were determined according to the method described by¹¹. All samples were collected with precautions required for microbiological analysis, held on iceboxes and processed within 12 h of collection.

2.4 Bacteriological analysis

All the specific/selective media were prepared with the addition of double distilled water and autoclaved properly. The bacterial populations in different samples were estimated by pure culture technique (spread plating method) on selective medium plates with 100 µL of suitable dilutions. All the media plates were incubated at 37°C ± 1°C for 24–48 h, except M-FC agar plates. The M-FC agar plates were incubated at 44.5°C ± 1°C for 24–48 h. After incubation, the final counts of colonies were noted and all trials were performed in triplicate. On the basis of media manufacturer's guide, typical colony morphology characteristics of different bacterial groups were recognized and initial enumeration of pathogenic pollution indicator bacteria was completed. Since recommended selective media were used for all organisms, specific biochemical tests were performed for identification and they are therefore referred to as like organisms (LO)^{1,5}. For confirmation of the pathogens, typical colonies were inoculated into Rapid Microbial Limit Test kits recommended for diagnostic microbiology supplied by Hi-media Laboratories Limited^{9,12}. Typical colony characteristics of each bacterial group and specific media used for enumerating them are listed in Table 1.

Table 1: Details of specific culture media used for quantitative bacterial analysis

S. No	Bacterial Indicators	Culture Medium	Positive Colonies	References
1.	Total Viable Count (TVC) ^a	Nutrient Agar	All colonies counted	Nagvenkar and Ramaiah (2009)
2.	Total Coliforms (TC) ^a	MacConkey Agar	All colonies counted	Clark et al. (2003)
3.	Total <i>Streptococci</i> (TS) ^a	M <i>Enterococcus</i> Agar	All colonies counted	Vignesh et al. (2013)
4.	<i>Vibrio</i> Count (VC) ^a	TCBS Agar	All colonies counted	Vignesh et al. (2012)
5.	Fecal Coliforms (FC) ^b	M FC Agar	Blue colonies counted	Vignesh et al. (2012)
6.	Fecal <i>Streptococci</i> (FS) ^a	KF <i>Streptococcus</i> Agar	Red colonies counted	Vignesh et al. (2014)
7.	<i>Salmonella</i> sp. (SA) ^a	XLD Agar	Black colonies counted	Clark et al. (2003)
8.	<i>Shigella</i> sp. (SH) ^a	XLD Agar	Pink colonies counted	Clark et al., (2003)
9.	<i>Pseudomonas</i> sp. (PC) ^a	Cetrimide Agar	Green colonies counted	Kumarasamy et al. (2009)

^a Media plates were incubated at 37°C ± 1°C for 24–48 h;

^b Media plates were incubated 44.5°C ± 1°C for 24–48 h

2.5 Statistical analysis

Pearson correlation coefficient was employed for the better understanding of relationship between the concentration of multiple variables (physiochemical and bacteriological parameters) by using statistical package of ORIGIN8.0.¹² The ANOVA was employed (ORIGIN8.0) to understand the variation in the variables between different stations, different locations and their interactions.

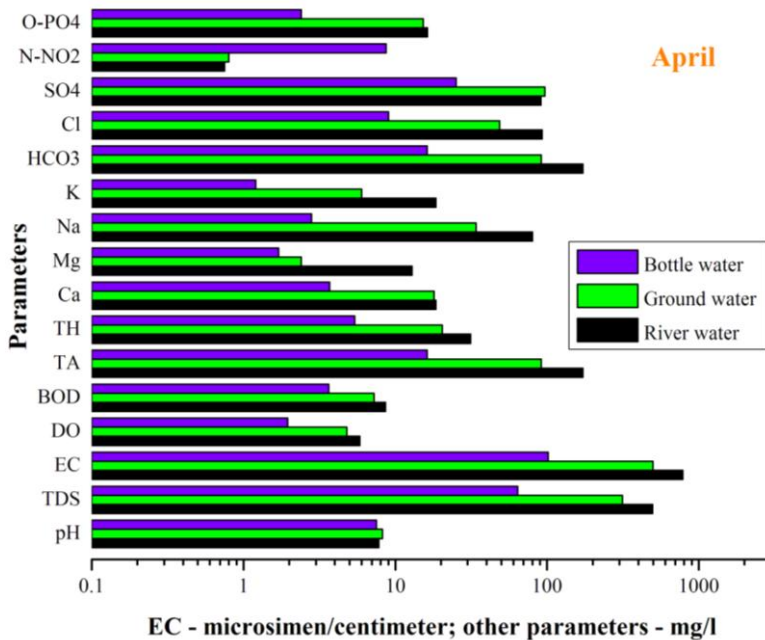
3. Result and discussion

In this study, the physiochemical and microbial levels were high in the water and it can be classified either polluted or unpolluted based on Bureau of Indian Standards (BIS) and World Health Organization (WHO) guidelines. In this study, the range of pH, EC and TDS in sample were 7.11–7.84, 51.4–828.6 µS/cm and 32.4–52.2 mg/L, respectively (Figure 1). Enormous amount of waste from industries, domestic sewage and agricultural practices find their way into rivers and percolated into ground water aquifers, resulting in large scale deterioration of

the water quality. In river water at the month of April, the mean values of pH, TDS, EC, DO, BOD, TA, TH, Ca, Mg, Na, K, HCO₃, Cl, SO₄, N-NO₂ and O-PO₄ were 7.78, 494.9, 785.55 (µS/cm), 5.85, 8.6, 172, 31.4, 18.5, 12.9, 80.1, 18.55, 172, 92.8, 91.2, 0.75 and 16.3 mg/L, respectively. The physiochemical

parameters were high in April and followed by May and June and were two- to five-fold higher than the May and June month. Similar pattern were also observed in ground and bottle waters. Interestingly, the similar levels of physiochemical parameters were observed in all the months.

Figure 1: Concentration of physiochemical parameters in three different water samples during April month



In ground water at the month of May, the mean values of pH, TDS, EC, DO, BOD, TA, TH, Ca, Mg, Na, K, HCO₃, Cl, SO₄, N-NO₂ and O-PO₄ were 7.77, 199, 315.85 (µS/cm), 3.85, 6.8, 52.85, 18.2, 15, 3.2, 18.2, 4.75, 52.85, 35.9, 54.05, 0.8 and 11.75 mg/L, respectively (Figure 2). The river and ground water results were indicated that few parameters were crossing the BIS and WHO guidelines. But in bottle waters at June, the mean

values of pH, TDS, EC, DO, BOD, TA, TH, Ca, Mg, Na, K, HCO₃, Cl, SO₄, N-NO₂ and O-PO₄ were 6.98, 36.85, 58.45 (µS/cm), 2.7, 5.1, 7.5, 4.45, 3, 1.45, 2.45, 1.45, 7.5, 4.15, 10.65, 3.1 and 2 mg/L, respectively (Figure 3). The results from bottle waters at June months were closely similar to April and May month because it was the treated by reverse osmosis process very well before marketing.

Figure 2: Concentration of physiochemical parameters in three different water samples during May month

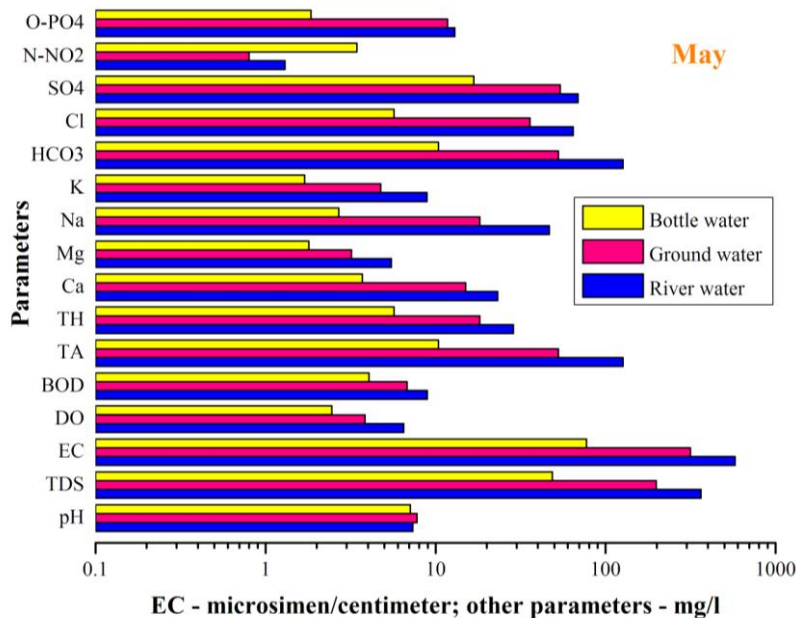
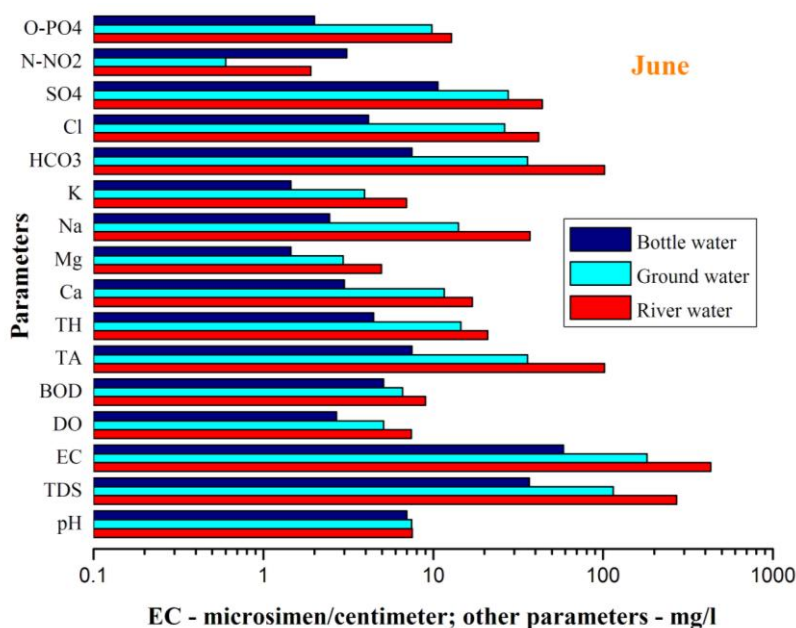


Figure 3: Concentration of physiochemical parameters in three different water samples during June month



Most of the parameters in the river and ground waters showed marked variation at each samples (month-wise and with-in month). The river and ground water is able to recover from the inorganic/organic/trace metal pollution stress only after the diffusion and probably through their self-purification system⁹. The physiochemical and bacterial parameters were higher in Gola river¹³ at state of Uttaranchal, India when compared to the Cauvery river water at Tiruchirappalli. The growing problem of degradation of our river ecosystem has necessitated the monitoring of water quality of various rivers all over the country to evaluate their production capacity, utility potential and to plan restorative measures¹⁴. The ranges of DO and BOD in bottled water were 1.8–2.1 mg/L and 3.6–3.7 mg/L, 2.3–2.6 mg/L and 3.9–4.2 mg/L, and 2.1–3.3 mg/L and 4.4–5.8 mg/L in April, May and June, respectively.

The ranges of TA and TH in bottled water were 16–17 mg/L and 5–6 mg/L, 9.2–12 mg/L and 5–7 mg/L, and 6.6–8.4 mg/L and 4–5 mg/L in April, May and June, respectively. But in the ranges of TA and TH in ground water were 67–116 mg/L and 18–22 mg/L, 43–63 mg/L and 18–18 mg/L, and 31–41 mg/L and 13–16 mg/L in April, May and June, respectively. The TA and TH results indicated that the ground waters contained higher levels of TA and TH than bottled water. The ranges of TA and TH in river water were 165–179 mg/L and 26–37 mg/L, 120–134 mg/L and 25–32 mg/L, and 99–105 mg/L and 16–26 mg/L in April, May and June, respectively. The TA and TH were one- to three-fold higher than ground water which denoted that the Ca, Mg and HCO₃ were also contributing higher pollution rate in river and ground waters. Chloride (Cl) is the second major

anions after HCO₃ in the aquatic environment and is also present in all samples. Minimum amount of nutrients like Nitrate, potassium and ortho-phosphate were present in most of the samples and is higher in the month of April. Commonly, the anions were highly found in the water and sediment column than cations. Similarly, the same pattern was also reported in our study. The physiochemical parameters were easily dissolve in water and were ability to exchange the materials between water and sediment column. Apart from this process, it was easily observed by microorganisms for their growth.

The overall mean value of physiochemical and microbial parameters were presented in Table 2. Conformation with microbiological standards is of special interest because of the capacity of water to spread diseases within a large population¹². Fecal material from human, domestic animals (dogs, cattle, and horses), as well as birds/waterfowl (geese, gulls, and ducks), all lead to increases in bacterial/pollution indicators loading in aquatic regions. Monitoring of physicochemical characteristics is not only decided the quality of water but the microbiological studies are also an important analysis for assessment of water quality¹¹. Total viable counts (TVC) were in the order of enormity above 10²/mL for all the three samples during all months. The bacteriological parameters were higher during May month followed by April month and June month. However, the river and ground water samples contained higher pollution indicators during all the months while the bottle waters were also found to have minimum level of total coliforms (TC) (<100 CFU/mL) (Figure 4). Sewage contamination in aquatic environments is commonly detected and quantified by enumerating the coliforms bacterial groups¹⁵.

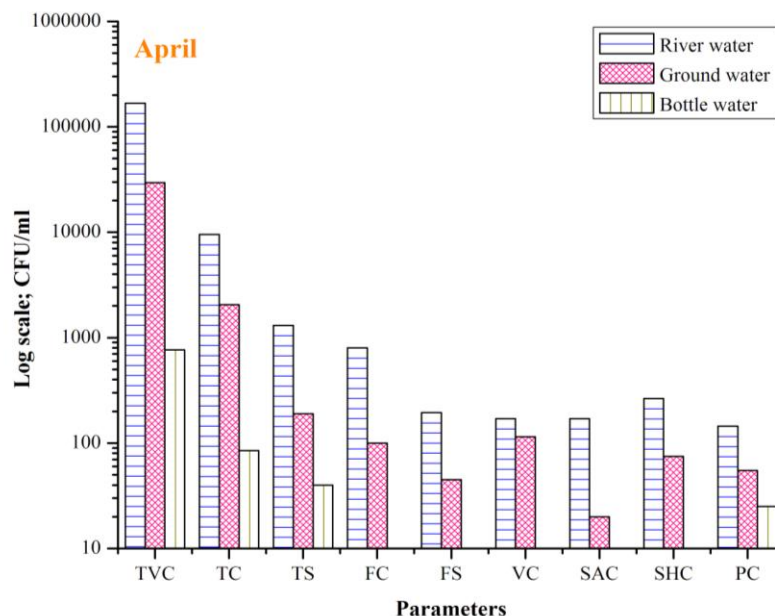
Table 2: Descriptive statistics of physiochemical and microbiological parameters in water samples

Variables	N	Mean	SD	Sum	Min	Max
pH	18	7.53	0.39	135.49	6.92	8.33
TDS	18	211.92	157.27	3814.5	32.4	522
EC	18	336.36	249.63	6054.5	51.4	828.6
DO	18	4.51	1.99	81.24	1.8	8.1
BOD	18	6.67	2.03	120	3.6	9.8
TA	18	68.34	57.11	1230.2	6.6	179.3
TH	18	16.64	10.15	299.5	3.9	37.2
Ca	18	12.63	7.55	227.4	2.8	26.2
Mg	18	4.09	3.69	73.7	1.1	16.2
Na	18	26.46	25.57	476.2	2.1	89.2
K	18	5.94	5.43	106.9	0.9	22.1
HCO ₃	18	68.34	57.11	1230.2	6.6	179.3
Cl	18	36.55	29.11	657.9	3.2	97.4
SO ₄	18	48.36	32.09	870.4	9.8	110.8
N-NO ₂	18	2.38	2.54	42.8	0.4	8.9
O-PO ₄	18	9.46	5.83	170.3	1.7	16.8
TVC	18	64068.89	86430.82	1.15E+06	380	246000
TC	18	1904.44	3092.78	34280	30	11900
TS	18	371.67	483.17	6690	0	1700
FC	18	177.22	299.62	3190	0	1200
FS	18	62.22	78.03	1120	0	270
VC	18	65.56	71.06	1180	0	200
SAC	18	46.67	72.84	840	0	210
SHC	18	98.89	125.18	1780	0	370
PC	18	85.00	83.47	1530	0	280

N – Numbers; SD – Standard Deviation; Sum – Summation; Min – Minimum; Max – Maximum

TDS – Total dissolved solids; EC – Electrical conductivity; DO – Dissolved oxygen; BOD – Biological dissolved oxygen; TA – Total alkalinity; TH – Total hardness; Ca – Calcium; Mg – Magnesium; Na – Sodium; K – Potassium; HCO₃ – Bicarbonate; CO₃ – Carbonate; Cl – Chloride; SO₄ – Sulphate; N-NO₂ – Nitrite; O-PO₄ – Ortho-phosphate; TVC – Total viable count; TC – Total coliforms; TS – Total *Streptococci*; FC – Fecal coliforms; FS – Fecal *Streptococci*; VC – *Vibrio* count; SAC – *Salmonella* sp. Count; SHC – *Shigella* sp. Count; PC – *Pseudomonas* sp. count

Figure 4: Counts of bacterial assemblage in three different water samples during April month



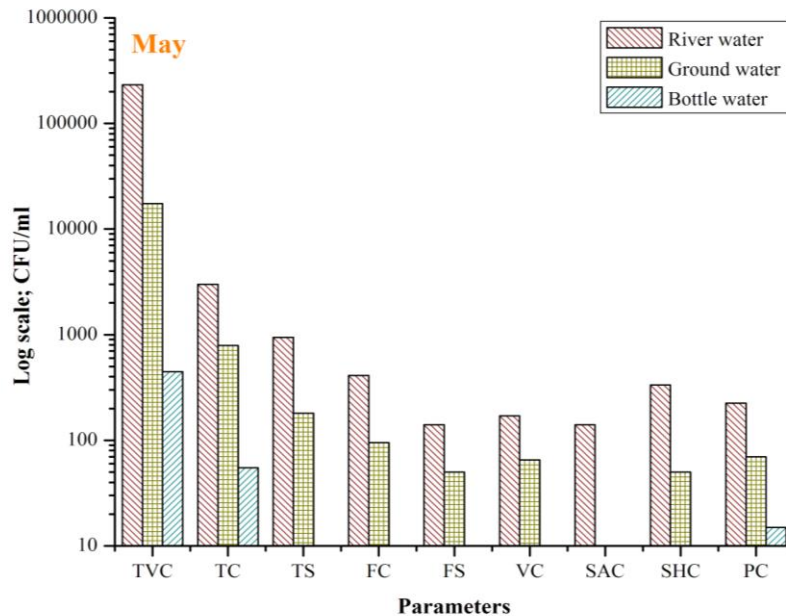
Interestingly, apart from TVC and TC, the other bacterial parameters were not observed from the bottle water in all the months, except *Pseudomonas* sp. count (PC). The PC was not found in June and May month (7th). In river water, counts of TVC, TC, TS, FC, FS, VC, SAC, SHC and PC were in the range of 117,000–233,000/mL, 1,200–9,500/mL, 600–1,300/mL, 160–800/mL, 110–190/mL, 70–170/mL,

90–170/mL, 160–330/mL and 140–190/mL, respectively (Figure 5). But in ground water, counts of TVC, TC, TS, FC, FS, VC, SAC, SHC and PC were in the range of 10,000–29,500/mL, 360–2,100/mL, 90–190/mL, 30–100/mL, 20–50/mL, 0–110/mL, 0–20/mL, 0–80/mL and 40–70/mL, respectively (Figure 5). Kistemann *et al.*¹⁶ observed that during rainfall, the microbial loads of running

water may suddenly increase and reach reservoir bodies very quickly. However, most of the pollution indicators and human pathogenic bacteria counts are

lower than those reported from the Cauvery river¹, Cauvery river and its estuary⁹, Mondovi and Zuari estuary waters¹⁷ and Ganga waters¹⁸.

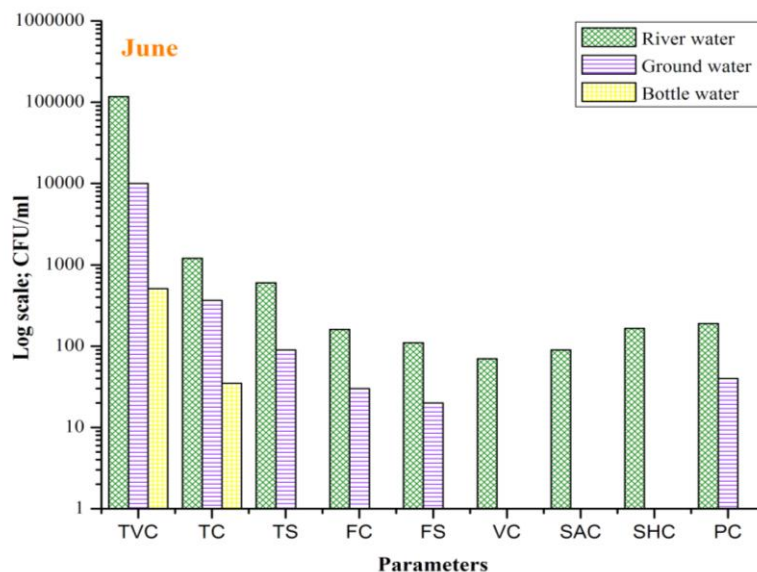
Figure 5: Counts of bacterial assemblage in three different water samples during May month



In April month, the mean value of TVC, TC, TS, FC, FS, VC, SAC, SHC and PC in river water were 168,000/mL, 9,500/mL, 1,300/mL, 800/mL, 190/mL, 170/mL, 170/mL, 260/mL and 140/mL, respectively. But in ground water at April month, the mean count of bacterial parameters such as TVC, TC, TS, FC, FS, VC, SAC, SHC and PC were 29,500/mL, 2,100/mL, 190/mL, 100/mL, 40/mL, 110/mL, 20/mL, 80/mL and 50/mL, respectively. Similar pattern were obtained in the May and June month. In this study, the counts of bacterial assemblage in river water were two- to five-fold higher than ground water while two- to 100-fold higher than bottle waters. The values of bacterial parameters were found to be higher during

the May month, and the similar findings were reported from Cauvery waters⁹, Cauvery water¹ and Ganga waters¹⁸. The nil counts of TS, FC, FS, VC, SAC and SHC were observed in bottle waters. The total viable counts were not above 10³/mL for all the samples during all months. But in river water, there is no nil count was observed in all the months in all samples. Interestingly, in June month, nil count was observed only in VA, SAC and SHC at ground water samples (Figure 6). Variations in bacterial counts were observed largely in both month-wise and sample-wise. In comparison with the Mondovi and Zuari estuary waters¹⁷, pollution indicators were lower in this study such as Tiruchirappalli waters.

Figure 6: Counts of bacterial assemblage in three different water samples during June month



3.1 Correlation matrix

Pearson coefficient correlations between the variables can provide interesting information on the relationship of the factors studied and their sources. The coefficient correlations between the microbiological and physiochemical parameters are listed in Table 3. The microbial and physiochemical parameters from the fresh waters of Tiruchirappalli city showed high positive correlations between them which indicated that those were strongly associated with each other statistically due to same origin. Moderate positive correlation relationships, such as

DO—SO₄, TC, FC & VC; BOD—Mg & TC; SO₄—TVC & PC may also be seen. Interestingly, negative correlation was also shown only between nitrite (N-NO₂) to all other physiochemical and microbial parameters. Commonly, the significant (p) value of two-tailed analysis of variance (ANOVA) infers the statistical significance on the correlation between microbial and physiochemical parameters¹². Insignificant p values (>0.5) were obtained between the correlations of pH with other parameters, except few parameters. While, there was no insignificant p value shown between microbial parameters.

Table 3: Coefficient correlation between microbial and physiochemical parameters in fresh waters of tiruchirappalli city

Variables	pH	TDS	EC	DO	BOD	TA	TH	Ca	Mg	Na	K	HCO ₃	Cl	SO ₄	N-NO ₂	O-PO ₄	TVC	TC	TS	FC	FS	VC	SAC	SHC	PC	
pH	1																									
TDS	0.57	1																								
EC	0.31	0.73	1																							
DO	0.36	0.83	0.73	1																						
BOD	0.49	0.98	0.83	0.93	1																					
TA	0.53	0.94	0.98	0.83	0.84	1																				
TH	0.59	0.88	0.94	0.98	0.86	0.93	1																			
Ca	0.25	0.81	0.88	0.88	0.86	0.86	0.95	1																		
Mg	0.46	0.81	0.88	0.88	0.86	0.86	0.95	0.59	1																	
Na	0.39	0.97	0.98	0.98	0.93	0.93	0.93	0.91	0.91	1																
K	0.49	0.98	0.98	0.98	0.86	0.86	0.86	0.86	0.82	0.97	0.97	1														
HCO ₃	0.51	0.97	0.97	0.76	0.84	0.84	0.93	0.85	0.84	0.97	0.97	0.97	1													
Cl	0.77	0.87	0.87	0.69	0.80	0.80	0.93	0.85	0.84	0.84	0.84	0.84	0.81	1												
SO ₄	0.77	0.87	0.87	0.69	0.80	0.80	0.93	0.85	0.84	0.84	0.84	0.84	0.81	0.81	1											
N-NO ₂	-0.29	-0.54	-0.54	-0.60	-0.70	-0.51	-0.61	-0.65	-0.57	-0.51	-0.48	-0.51	-0.56	-0.49		1										
O-PO ₄	0.72	0.88	0.88	0.79	0.86	0.84	0.90	0.91	0.62	0.82	0.74	0.84	0.84	0.88	-0.69		1									
TVC	0.09	0.81	0.81	0.73	0.78	0.84	0.84	0.78	0.74	0.82	0.77	0.84	0.79	0.55	-0.35	-0.35		1								
TC	0.35	0.84	0.84	0.47	0.57	0.84	0.84	0.57	0.74	0.92	0.97	0.84	0.87	0.66	-0.35	-0.35	0.61		1							
TS	0.23	0.88	0.88	0.69	0.75	0.91	0.89	0.74	0.93	0.94	0.94	0.88	0.88	0.63	-0.37	-0.37	0.67	0.92		1						
FC	0.24	0.80	0.80	0.55	0.61	0.80	0.81	0.63	0.95	0.88	0.93	0.80	0.80	0.61	-0.33	-0.33	0.61	0.79	0.92		1					
FS	0.32	0.87	0.87	0.72	0.74	0.88	0.88	0.63	0.88	0.90	0.90	0.84	0.84	0.66	-0.42	-0.42	0.61	0.86	0.94	0.91		1				
VC	0.44	0.93	0.93	0.58	0.75	0.91	0.88	0.63	0.70	0.88	0.81	0.84	0.84	0.66	-0.51	-0.51	0.61	0.86	0.94	0.91	0.78		1			
SAC	0.21	0.83	0.83	0.70	0.70	0.85	0.85	0.72	0.85	0.88	0.86	0.85	0.79	0.60	-0.32	-0.32	0.90	0.84	0.94	0.91	0.78	0.75		1		
SHC	0.20	0.87	0.87	0.73	0.79	0.89	0.87	0.72	0.74	0.86	0.86	0.85	0.79	0.65	-0.39	-0.39	0.90	0.84	0.94	0.91	0.78	0.75	0.91		1	
PC	0.24	0.76	0.76	0.83	0.82	0.89	0.84	0.82	0.62	0.75	0.66	0.80	0.71	0.54	-0.36	-0.36	0.91	0.84	0.94	0.91	0.78	0.75	0.91	0.91		1

Negative correlation value shown in bold form

TDS – Total dissolved solids; EC – Electrical conductivity; DO – Dissolved oxygen; BOD – Biological dissolved oxygen; TA – Total alkalinity; TH – Total hardness; Ca – Calcium; Mg – Magnesium; Na – Sodium; K – Potassium; HCO₃ – Bicarbonate; CO₃ – Carbonate; Cl – Chloride; SO₄ – Sulphate; N-NO₂ – Nitrite; O-PO₄ – Ortho-phosphate; TVC – Total viable count; TC – Total coliforms; TS – Total *Streptococci*; FC – Fecal coliforms; FS – Fecal *Streptococci*; VC – *Vibrio* count; SAC – *Salmonella* sp. count; SHC – *Shigella* sp. count; PC – *Pseudomonas* sp. count

4. Conclusion

The findings of the present study identified the relationship between the microbial and physiochemical parameters by Pearson coefficient correlation method and was also indicated the sources of pollution. River water samples were found to have higher indicator bacterial levels than those allowed by the WHO guidelines. The bacterial and physiochemical concentrations were high in river water followed by ground and bottle water. The human activities and sewage waste could be the main contributing sources for the pollution. Among the three months, May month received higher pollution load followed by April and June which was studied based on the microbial and physiochemical level. The correlation indicated that microbial accumulation is depending on the physiochemical concentration in the fresh waters. The efforts need to be made to raise awareness among the public about the risks that occur in their environment and are due to their activities.

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