

## RESEARCH ARTICLE

### OCCURRENCE OF INDICATOR BACTERIA AND PATHOGENS IN THE WATERS OF THE NOYYAL RIVER NEAR ITS SOURCE AND DOWNSTREAM IN COIMBATORE DISTRICT

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#### ABSTRACT

Water, the bloodstream of the biosphere, also acts as a sink, solvent and transport vehicle for domestic, agricultural and industrial waste, causing pollution. The Noyyal River, a tributary of the River Cauvery, was once the lifeline of Coimbatore, but is now being threatened by pollution from various sources. Assessments of microbiological water quality have focused on the presence of Fecal Indicator Bacteria, which indicate that a contamination pathway exists between a bacterial source and the water supply, serving as a route for pathogen entry. Water samples from seven sites (Kooduthurai, Madakkadu, Thombilipalayam, Alandurai, Mathampatti, Perur and Aathuppalam) along the Noyyal, were analyzed for various microbiological characteristics. The results indicated that the river was relatively unpolluted upstream near its sources, but is progressively polluted on moving further downstream towards Coimbatore city. Levels of Total Heterotrophic Bacteria, indicators such as *Enterobacteriaceae*, Total Coliforms, Fecal Coliforms (FC), Fecal Streptococci (FS), pathogens like *Salmonella sp.*, *Shigella sp.*, and *Vibrio sp.* were elevated at downstream sampling sites. Evaluation of the FC:FS ratio revealed that, the water was unfit for drinking at almost all the downstream sampling stations, and could have limited use for irrigation. The release of fecal microorganisms and pathogens from the sediments to the water column could be a major source of bacterial loading. Non-point sources were identified as the major contributors of contaminants. Regular monitoring of the river water and sediment, amelioration of point / non-point sources of pollution, and speedy implementation of remedial measures, may restore this stretch of the Noyyal River to its former pristine state.

**Keywords:** Fecal Coliforms, Fecal Streptococci, *Salmonella sp.*, *Shigella sp.*, *Vibrio sp.*

#### 1. INTRODUCTION

Water is the elixir of life [1], and the bloodstream of the biosphere [2]. Among the surface water resources, rivers are important as sources of drinking water, irrigation and fishing. Rivers represent only about 0.0001 % of the total amount of water in the world [3]. There are about 10,360 rivers in India, and the mean annual flow in all the river basins is estimated to be 1,869 cubic km.

The supply and demand for surface water is widening with increasing population [4]. Water of adequate quantity and standard quality is required to meet household, industrial, and agricultural needs [5]. In addition to fulfilling the basic needs of humans and ecosystems, water is also one of the most delicate parts of the environment [6], acting as a sink, solvent and transport vehicle for domestic, agricultural and industrial waste, causing pollution [7]. Attention on water contamination and its management has become the need of the hour because of its far reaching impact on human health [8]. Pollution of riverine systems could result in the outbreak of diseases like cholera, typhoid fever and diarrhoea [9]. Toxins within water can harm or even kill

aquatic organisms and other animals that may have accidentally fed on the infected organisms.

The widespread degradation of many Indian rivers has been reported by several authors [10-18].

The Central Pollution Control Board (CPCB) in collaboration with State Pollution Control Boards (SPCB) monitors water quality of national aquatic resources. Legislative provisions such as the Water (Prevention and Control of Pollution) Act (1974), and Environment Protection Act (1986), have not been implemented effectively. The Water Cess Act (1977), meant to reduce pollution has also made marginal impacts. The State of Tamil Nadu had tried to combat the alarming rise of aquatic pollution by imposing strict guidelines. However, the effluents often flow through open drains and reach rivers, lakes, etc. Sewage from municipalities and settlements has also increased tremendously due to piped water supply and is contaminating rivers, lakes, tanks, and ground water [19].

The Noyyal or Noi il river, which translates into "devoid of illness" in Tamil, is a tributary of the River Cauvery. It is believed that the water of the Noyyal River contains natural medicines and is therefore good for health [20]. Today, the river has

become "ill" and is one of India's most polluted rivers. The water originating from the hills is tasty and sweet, but soon the quality changes. As it passes through towns and cities, sewage and effluents from several sources are discharged into it. Percolation of the wastes through the soil results in the contamination of the ground water sources [1]. Parts of the Noyyal river basin are "Industrial Wastelands", areas subjected to degradation as a result of a large-scale discharge of industrial effluents [21].

To explore the extent of pollution in the Noyyal river, water samples from the five stations of Boluvampatti, Perur, Alangadu, Kasipalayam and Orathupalayam were analysed [22]. Results showed heavy contamination of the river towards the downstream stations. A biological study on the impacts of effluents in Noyyal River found that the river was highly polluted from Sulur to the Orathupalayam dam and that the plankton species diversity was comparatively low in this part [20] [23]. Geetha *et al.* [24] carried out a systematic study to assess the underground water quality and the effect of textile effluents, on Noyyal river basin at Tirupur. It was found that the underground water was contaminated at Orathupalayam, Karuvapalayam, Kulathupalayam, Uttukuli and Kodumanalpurur, due to the industrial discharge of effluents into the river from Tirupur. Chitradevi and Sridhar [5] demonstrated penetration of polluted river water into adjoining aquifers, rendering it unsuitable for irrigation. The physico-chemical and bacteriological characteristics of the Noyyal river and groundwater quality were assessed at Perur near Coimbatore [1]. The total heterotrophic bacteria, Total Coliforms (TC), Fecal Coliforms (FC), Fecal Streptococci (FS) and FC/FS ratio in the river water samples were found to be greater than the standard WHO limits.

Contamination from human or animal wastes, improperly treated septic and sewage discharges, leaching of animal manure, storm water runoff, wastes of domestic animals or wildlife, are sources of pathogens in water. Bacteria and other harmful microorganisms from any of these sources may be washed into rivers. These pathogens pose a serious health risk to individuals consuming the water, and may even cause epidemics.

Rivers feature multiple pathogen loading inputs from both point and non-point sources along their course such that pathogens may accumulate within the same plume. 'Point' sources are defined by the USEPA as "any discernible, confined, and discrete conveyance, including any pipe, ditch, channel, tunnel, conduit, well, discrete fixture, container, rolling stock, concentrated animal feeding operation, landfill leachate collection system, vessel, or other floating craft from which pollutants may be discharged." 'Non-point' sources, as the

term indicates, are diffuse and caused by rainfall (i.e. stormwater), snowmelt or irrigation flowing above and below ground. Potential nonpoint sources of bacteria may include onsite septic systems, urban stormwater run-off, boats, livestock and wildlife. Both point and nonpoint sources may contain microbes, which may then be deposited into freshwater bodies, wetlands, coastal waters, and drinking water. Furthermore, water withdrawals along the river may remove pathogens while water inputs from uncontaminated tributaries and groundwater seepage may result in dilution.

Difficulties and expenses involved in the testing for specific pathogens have generally led to the use of indicator organisms of enteric origin to estimate the persistence and fate of enteric bacteria in the environment [25]. Traditional assessments of microbiological water quality have focused on the presence of fecal indicator bacteria (FIB). Although FIB are not necessarily pathogenic, they are abundant in human and other animal waste products where pathogenic organisms are found [26]. They also indicate that a contamination pathway exists between a source of bacteria (surface water, septic system, animal waste, etc.) and the water supply. Disease-causing bacteria may use this pathway to enter the water supply.

Total and fecal coliform bacteria have been used extensively for many years as indicators for determining the sanitary quality of surface and recreational water [26], and have been included in water quality standards in different parts of the world [27] [28] [29] [30].

By definition [31], the total Coliform (TC) group comprises all aerobic and facultative anaerobic, gram-negative, non-spore-forming, rod-shaped bacteria that ferment lactose with gas and acid formation within 48 hours at 35°C. The maximum permissible value of total coliforms in drinking water is 1 per 100ml [27], and 10 per 100ml [30]. Fecal Coliforms (FC) are defined as those coliforms which respond at an elevated temperature of 44.5°C. Fecal coliform bacteria are specific to the intestinal tracts of warm-blooded animals, including humans. Fecal coliforms (FC) and *E. coli* (EC) are widely used as indicators of fecal contamination in water [32] [33], and have been shown to be positively correlated with the presence of pathogenic enteric bacteria [33]. Coliform bacteria are indicators of dangerous bacteria such as, *Shigella* or *Salmonella* [34]. The absence of coliform bacteria is indicative of a water supply free of pathogenic bacteria [35].

The fecal Streptococci comprise *Streptococcus faecalis*, *S. bovis*, *S. equinus* and *S. avium*. Since they inhabit the intestinal tract of humans and warm-blooded animals, they are used to detect fecal contamination in water, and survive longer than other bacterial indicators. The fecal coliform to fecal streptococci ratio (FC/FS ratio)

serves as an indicator of the origin (human vs. animal sources) of fecal pollution of surface waters [36].

*Salmonella* and *Shigella*, which are important genera in the *Enterobacteriaceae* family, are not considered coliforms, but their presence is indicated by the coliforms [34]. Geldreich [37] found that for FC concentrations greater than about 1000/dL, the frequency of *Salmonella* occurrence doubled.

*Vibrio sp.* is primarily a water-borne pathogen, but can also be a food-poisoning hazard associated with ingestion of shellfish which become contaminated with the bacteria by filter feeding [38].

The health effects of exposure to pathogenic bacteria in water are varied. The most common symptoms of waterborne illness include fever, nausea, vomiting, and diarrhoea. Outbreaks from *Salmonella sp.* contamination of water have been documented [39]. Symptoms of salmonellosis include nausea, vomiting, cramps, diarrhoea, and, in about 2% of the cases, arthritis [40].

It has been estimated that waterborne diseases kill more than 5 million people annually [41]. The microbial pathogens responsible for most of these deaths originate from human and animal feces. These wastes contaminate water through a variety of means including direct discharge, surface runoff and seepage through groundwater. Simple water treatment procedures such as chlorination inactivate the majority of these pathogens. Therefore, most cases of waterborne disease result from the consumption of inadequately treated water.

Understanding the effects of changing land usage, population density, seasonal variations, sediment-bacteria interactions, and point and non-point sources of contamination, on river microbiology is important for public health officials seeking to predict the presence of microbial pathogens in water resources. Reports on the contamination of the Noyyal River and its degradation are chiefly restricted to the course of the river from Perur in Coimbatore and beyond. Precise data on the microbiological quality of the river, at various sites along its course, from its tributaries progressing downstream to Perur and Aathuppalam will help in tracking the microbial quality and sources of contaminants. The study focuses on filling this lacuna, which will help to identify the point and non-point sources of pollution, and pinpoint threats to human health, thereby aiding in formulating management strategies to combat contamination.

## 2. MATERIALS AND METHODS

### 2.1. Study Area

The study was undertaken at selected sampling stations of the Noyyal River in Coimbatore District. The Noyyal River is a tributary of the River Cauvery and originates from the Vellingiri Hills of the Western Ghats in the Coimbatore district of Tamil Nadu, South India. The river has moderate to good flow for a short period during the North-East and South-West monsoons and flows over a distance of 170 km in an area of 3510 km<sup>2</sup>. The river's basin is 170 km (110 m) long and 25 km (16 m) wide and covers a total area of 3,500 km<sup>2</sup> (1,400 sq. m). It passes through seven taluks (Coimbatore, Tirupur, Avinashi, Palladam, Karur, Erode and Dharapuram) in five districts (Coimbatore, Tirupur, Erode, Karur, and Trichy). The river has a valley fill (made of alluvial kankar soil) over a stretch of 25 km (16 m) and a depth of 198 ft (60 m). It extends from the origin of the river at Kooduthurai (in Madhvarayapuram, 30 km (19 m) west of Coimbatore city) to the Ukkadam Tank on the city border. The area of land irrigated by the Noyyal River in Coimbatore district is approximately 1600 acres [42].

### 2.2. Samples

Water samples were collected at seven sampling stations along the Noyyal River during the period June, 2012 to February, 2013. The first sampling station (A) was Kooduthurai, located near Sadivayal (10°56'36"N 76°43'23"E), where two minor tributaries joined to form a major tributary, which later emptied into the Noyyal at Thombilipalayam. The second sampling station was at another tributary of the Noyyal at Madakkadu (B) tribal village (10°58'41"N 76°44'28"E), which also joins the Noyyal at Thombilipalayam. The third sampling station was near the village of Thombilipalayam (C) (nearly 14 km from Coimbatore city). The fourth and fifth sampling stations were near the villages of Alanthurai (D) (10°57'11"N 76°47'21"E) and Mathampatti (E) (10°58'12"N 76°51'36"E), both harboring rural and agriculture-based populations. The sixth sampling station was near Perur (F) (10°58'N 76°54'E), at the outskirts of Coimbatore city, a heavily polluted stretch of the river, due to the discharge of domestic sewage and municipal waste (Usharani *et al.*, 2010). The seventh sampling station (G) was near Aathuppalam (10°58'27"N 76°57'42"E), in Coimbatore city, where the river was further contaminated by dyeing effluents and sewage.

The water samples were collected at the sampling sites in sterile labeled containers, and brought to the laboratory for microbiological analysis. Bacteria present in the samples were then isolated and enumerated using standard microbiological methods [43].



Plate 1. Sampling Stations of Noyyal River

Sample	Sampling Station
A	Kooduthurai
B	Madakkadu
C	Thombilipalayam
D	Alanthurai
E	Mathampatti
F	Perur
G	Aathuppalam

### 2.3. Media and Chemicals

The various selective media used in the study were designated as: a. Nutrient agar, b. EMB agar, c. Endo agar, d. MFC agar, e. Fecal Streptococci agar, f. Salmonella agar, g. TCBS agar, h. Shigella base. The selective media and chemicals used in the study were procured from Hi Media Laboratories, Mumbai.

### 2.4. Microbiological Analysis

The total heterotrophic bacterial counts (THBC) of the water samples were enumerated on nutrient agar medium using the serial dilution method and spread plate technique. The method of Dubey and Maheswari [44] was adopted for the inoculation of media. EMB Agar [45] [46] was utilised for the isolation, enumeration and differentiation of members of *Enterobacteriaceae* including *Escherichia coli* and *Enterobacter aerogenes*. The dyes in EMB agar, eosin Y and methylene blue are pH indicators and inhibitors of gram-positive bacteria and at an acid pH combine to form a green-metallic precipitate [47]. Endo Agar [48] was used to confirm the detection and enumeration of coliform bacteria, which produce pink colonies on fermentation of lactose while lactose non-fermenters produce colourless colonies on the medium. M-FC Agar Base, designed by Geldreich *et al.* [49] was used for the detection and enumeration of fecal coliforms using the membrane filter technique. M-FC Agar Base is recommended by APHA [50] for detection of fecal coliforms. Bile Esculin Azide Agar, a modification of Bile Esculin Agar [51] [52] as per Isenberg [53], uses esculin hydrolysis in the identification of fecal Streptococci or Enterococci. HiCrome RajHans Medium, Modified, a modification of the original formulation of Rambach [54], was used for

differentiation of *Salmonella* species. The chromogenic mixture incorporated in the medium yields pink to red colonies of *Salmonella*. TCBS Agar was used for the selective isolation and cultivation of *Vibrio cholerae* and other enteropathogenic Vibrios. Strains of *V. cholerae* produce yellow colonies on TCBS Agar because of fermentation of sucrose. A few strains of *V. cholerae* may appear green or colourless on TCBS Agar due to delayed sucrose fermentation. Shigella Base was used for the isolation and cultivation of *Shigella* species.

A minimum of three samples per sampling site was used for microbiological analysis. Triplicates were maintained for enumeration of bacteria in each sample and mean viable counts noted.

The number of bacteria per mL of original culture is calculated by multiplying the number of colonies counted by the dilution factor, which is the reciprocal of the dilution.

No. of colonies per mL of culture = Number of colonies x Dilution factor x mL of dilution plated

### 2.5. Statistical Analysis

Pearson's correlation was computed using the formula:

$$r = \frac{\sum XY - \frac{\sum X \sum Y}{N}}{\sqrt{(\sum X^2 - \frac{(\sum X)^2}{N})(\sum Y^2 - \frac{(\sum Y)^2}{N})}}$$

Where,

N=Number of values or elements, X=First Score, Y=Second Score,  $\sum XY$  = Sum of the product of first and Second Scores,  $\sum X$ =Sum of First Scores,  $\sum Y$ =Sum of Second Scores,  $\sum X^2$ =Sum of square First Scores,  $\sum Y^2$  = Sum of square Second Scores.

## 3. RESULTS AND DISCUSSION

The Noyyal River, whose medicinal waters once rejuvenated Coimbatore district, is being threatened by pollution from anthropogenic sources. To gain a better understanding of the contaminants and the possible hazards to human health, water samples from seven sites were analysed for various microbiological characteristics.

The results of this study indicate that the total, viable, heterotrophic bacterial counts gradually increased from the sources of the river Noyyal, attaining high levels at the last two stations of Perur (F) and Aathuppalam (G). In a study of bacterial populations on the Welsh River Dee, Nuttall [55] recorded a similar progressive increase in heterotrophic bacteria from the source of the river towards the lower reaches.

Further, the survey period i.e., June, 2012 to February, 2013, coincided with an intense dry spell, with little or no rainfall. The volume of water in the Noyyal River was reduced, the river shallow,

with significant input from terrestrial run-off. This may account for the moderate to high bacterial counts observed in the study. Such sporadic

increases in THBC of the water column, due to run-off, were reported in a narrow, shallow creek of the Rhode River by Carney *et al.* [56].

**Table 1. Microbial Contamination in Water of Noyyal River**

Type of Microbe	Sample A	Sample B	Sample C	Sample D	Sample E	Sample F	Sample G
MVC of bacteria (THBC)	0.3 x 10 <sup>5</sup>	1.33 x 10 <sup>9</sup>	5.17 x 10 <sup>4</sup>	1.67 x 10 <sup>4</sup>	0.57 x 10 <sup>5</sup>	0.87 x 10 <sup>10</sup>	1.88 x 10 <sup>15</sup>
<i>Enterobacteriaceae</i>	0.15	0.62	0.95	1.36 x 10	0.57	1.67 x 10 <sup>6</sup>	1.61 x 10 <sup>16</sup>
Total Coliforms	0.25	0.33	0.62	3.85 x 10 <sup>5</sup>	1.05	1.88 x 10 <sup>15</sup>	1.38 x 10 <sup>15</sup>
Fecal Coliforms	0.88	3.50	0.40	0.85	0.88	1.09 x 10 <sup>6</sup>	1.12 x 10 <sup>5</sup>
Fecal <i>Streptococci</i>	0.88	0.15	0.38	1.63	1.22	0.22 x 10 <sup>5</sup>	0.22 x 10 <sup>5</sup>
<i>Salmonella sp.</i>	0.72	0.20	0.52	2.75	3.53	1.44 x 10	1.83 x 10 <sup>16</sup>
<i>Vibrio sp.</i>	0.17	0.16	0.18	0.10	0.18	4.72	0.83 x 10 <sup>5</sup>
<i>Shigella sp.</i>	0.78 x 10 <sup>5</sup>	0.65	0.77	1.0	1.23 x 10 <sup>10</sup>	1.95 x 10 <sup>6</sup>	3.23 x 10 <sup>5</sup>

MVC – Mean Viable Count

**Table 2. Fecal Coliform: Fecal Streptococci (FC:FS) Ratio**

FC:FS Ratio	Sample A	Sample B	Sample C	Sample D	Sample E	Sample F	Sample G
< 0.7	66.67	100	33.33	66.67	33.33	0	0
0.7 – 4.0	33.33	0	66.67	33.33	66.67	0	33.33
> 4.0	0	0	0	0	0	100	66.67

< 0.7 – Animal Origin, 0.7 – 4.0 – Mixed Origin, > 4.0 – Human Origin

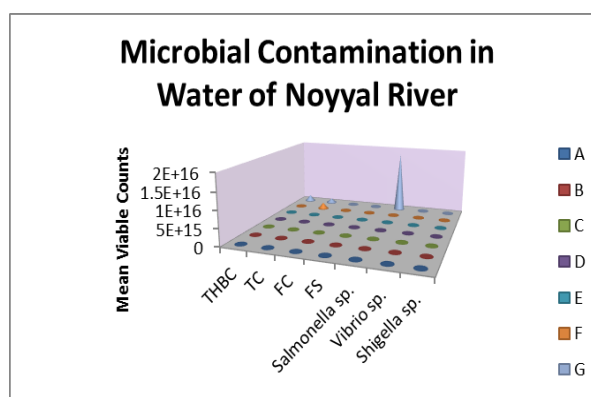
The quantitative abundance of heterotrophic microbes is a direct indication of the rich availability of organic nutrients. The rich availability of organic nutrients can sustain relatively higher bacterial populations. According to USEPA [57], high THBC may be an indicator of poor general biological quality of river and sediments. Large sediment populations at both upstream and downstream stations in the eutrophic Shetucket River basin in north-eastern Connecticut revealed that sediments were an important reservoir of pollution indicator bacteria [58].

Important pollution indicators are the family *Enterobacteriaceae*, which comprises *Escherichia coli* and *Enterobacter sp.* Of these *E. coli* are specific to the intestinal tract of mammals.

*Escherichia coli* can be identified with eosin methylene blue (EMB) agar.

Their numbers increased from sampling stations A to G, with the lowest counts being recorded at site A. Fecal indicators have been reported from both pristine, non-agricultural watersheds [59], and agricultural watersheds [60]. Their results concur with our findings, since the

sampling stations were located in both agriculture-based and pristine areas. Other sources would include surface run-off, sewage, feces of domestic and wild animals. The possibility that indicator bacteria are capable of multiplying in the aquatic sediments has been suggested. This is based upon the general finding that indicator bacterial densities in sediments are 2-3 logs greater than in overlying waters [61]. Their populations maintained themselves at high densities in the presence of constant predation and competition/antagonism effects and without significant external supplementation [62].



**Table 3. Water Quality Evaluation at the Sampling Sites in Noyal River**

Micro-organism	No. of bacteria	Water quality <sup>3</sup>	Site A	Site B	Site C	Site D	Site E	Site F	Site G
TVC <sup>1</sup>	- 500000	1	100	0	100	100	100	0	0
	>500000 – 1000000	2	0	0	0	0	0	0	0
	>1000000 – 10000000	3	0	0	0	0	0	0	0
	>10000000 – 50000000	4	0	0	0	0	0	0	0
	>50000000 – 100000000	5	0	0	0	0	0	0	0
	>100000000 – 750000000	6	0	33.33	0	0	0	0	0
	>750000000	7	0	66.67	0	0	0	100	100
FC <sup>2</sup>	- 1000	1	100	100	100	100	100	0	0
	>1000 – 10000	2	0	0	0	0	0	0	33.33
	>10000 – 100000	3	0	0	0	0	0	0	0
	>100000 – 500000	4	0	0	0	0	0	0	66.67
	>500000 – 1000000	5	0	0	0	0	0	33.33	0
	>1000000 – 10000000	6	0	0	0	0	0	66.67	0
	>10000000	7	0	0	0	0	0	0	0

<sup>1</sup> – Total no. of heterotrophic bacteria; <sup>2</sup> – No. of fecal coliforms; <sup>3</sup> – Degree of loading with easily decomposable organic substances and feces; 1 – very little, 2 – little, 3 – moderate, 4 – moderately high, 5 – high, 6 – very high, 7 – extremely high

Members of the *Enterobacteriaceae* including *E. coli*, are collectively known as total coliforms. The *Enterobacteriaceae* were enumerated on EMB agar and further confirmed by the test for Total Coliforms on Endo agar. The coliform test is a reliable indication of the possible presence of fecal contamination and is consequently, correlated with pathogens [63].

The total coliform counts were very high at sampling stations F (Perur) and G (Aathuppalam), and moderately high at site D (Alanthurai). The first three sampling stations (A, B and C) occurring in closer proximity to the source of the river, and the fifth sampling site E (Mathampatti) had low TC counts. The levels of total coliforms exceeded the maximum permissible values of 1 per 100ml [27] in drinking water and 10 per 100ml [30]. Because

of their known associations with plants and soils [64], total coliforms are a reflection of urban runoff [65]. This is reflected in our results, since the sites with increased TC counts are suburban areas contributing large quantities of sewage and

effluents, in addition to agricultural run-off. The total coliform counts recorded here are higher than the fecal coliform counts. According to Goyal *et al.* [66], this is not a surprising finding, since total coliforms can originate from non-fecal sources.

**Table 4. Correlation Matrix of Microbiological Parameters in Water of Noyyal River**

	MHC	TC	FC	<i>Salmonella sp.</i>	<i>Vibrio sp.</i>	<i>Shigella sp.</i>
Total Coliforms	0.499					
Fecal Coliforms	- 0.065	0.833				
<i>Salmonella sp.</i>	0.999	0.499	- 0.065			
<i>Vibrio sp.</i>	0.999	0.499	- 0.065	0.999		
<i>Shigella sp.</i>	- 0.167	- 0.254	- 0.186	- 0.167	- 0.167	
FS	0.645	0.984	0.720	0.645	0.646	- 0.258

MHC - Mean Heterotrophic Count, TC – Total Coliforms, FC – Fecal Coliforms, FS – Fecal *Streptococci*.

The presence of Coliform bacteria in sediment and hence, in water, can lead to symptoms like diarrhoea, nausea, vomiting, cramps or other gastro intestinal distress, and in severe cases prove fatal [67]. Ingestion of *E. coli* may result in hemolytic uremic syndrome. *E. coli* and *Klebsiella* may also cause urinary tract infections [3]. The chief organisms implicated as threats to public health are *E. coli*, parasites or fecal coliforms.

Fecal coliforms find widespread use as indicators of fecal contamination in water [32] [33]. Analysis of the water of the Noyyal River indicated that the FC counts became higher on progression from the source towards downstream areas. The last two sampling stations (G and F) downstream (Aathuppalam and Perur) exhibited the greatest abundance, while the upstream sampling sites showed relatively lower counts. Hyland *et al.* [68] also reported that waters with the lowest FC counts were nearest the river source, and those with the highest were downstream of areas with substantial agricultural activity. Sampling stations C (Thombilipalayam), D (Alanthurai) and E (Mathampatti) are predominantly agriculture-based. Doran and Linn [69] found FC counts 5 to 10 times higher in runoff from grazed lands than in ungrazed lands. High FC levels at agricultural sites may be due to a number of factors such as domestic or wild animals defecating in or near the surface waters, runoff

from manure applied to lands adjacent to surface waters and liquid effluent entering these waters from animal rearing operations [70]. Septic systems from densely clustered residences have a substantial impact on bacterial loading [59] in the river and sediments.

Water samples from the source tributaries (A and B) adjacent to forested areas also contained fecal coliforms. Animal waste deposited in water or carried in runoff contributes to high concentrations of dissolved nitrogen, biochemical oxygen demand and bacterial pollution [71]. Migratory herbivores cause elevated dung deposition in different watersheds [72], leading to surface-water contamination [73], and seasonal fecal bacterial pollution [74].

In a rural mixed land use watershed, sources could consist of point (domestic septic systems, manure storages and animal defecation directly into streams) and non-point (sediments and manure application) sources [59]. Doran and Linn [69] monitored surface runoff from grazed and ungrazed pasture land over a three-year period in Nebraska. Fecal coliform counts were 5 to 10 times greater in grazed areas. Patni *et al.* [75] observed that FC bacteria were almost always present in runoff from non-manured cropland, presumably due to non-livestock sources. Land use was found to have the most significant effect on concentrations of bacterial indicators [76].

The water samples from the last two sampling stations of this study, namely, G (Aathuppalam) and F (Perur) exhibited extremely high to moderate counts of the third group of indicator bacteria, the fecal *Streptococci* (FS). These regions are the most populated among the seven sampling stations, generating large quantities of sewage, effluent and surface runoff. The samples from the first four sampling sites contained substantially lower numbers of FS. High frequencies of fecal *Streptococci* have been reported from populated areas and areas receiving pasture land runoff [56].

The ratio of fecal coliform (FC) to fecal *Streptococcus* (FS) is considered an indicator of the sources of the fecal bacteria, whether human or animal. Human fecal material contains a much higher FC:FS ratio than fecal material from other mammals and birds [77]. Geldreich and Kenner [36] suggested that ratios greater than 4.0 indicated sewage effluents of human origin and those lower than 0.7 suggested non-human, warm-blooded animal sources. FC:FS ratio from 0.7 – 4.0 was of mixed origin. In natural waters, therefore, the higher the FC:FS ratio, the greater the probability of human origin [78].

The results of this study indicated that all the water samples from site B (Madakkadu), 66.67% of samples from site A (Kooduthurai) and site D (Alanthurai), 33.33% of samples from site C (Thombilipalayam) and site E (Mathampatti), had a FC:FS ratio < 0.7, and it is inferred that the fecal contamination in these sites is of animal origin. 33.33% of samples from site A (Kooduthurai), site D (Alanthurai) and site G (Aathuppalam), as well as 66.67% samples from site C (Thombilipalayam) and site E (Mathampatti) displayed a FC:FS ratio from 0.7 – 4.0, indicating that contamination was from mixed sources of both human and animal origin. In 100% of water samples from site F (Perur), and 66.67% of samples from site G (Aathuppalam), the FC:FS ratio was > 4.0, showing that fecal contamination was exclusively of human origin.

Kavka [79] and Kohl [80] proposed bacteriological criteria for estimation of water and bottom sediments, which was later modified by Albinger [81]. It is possible to estimate the extent of contamination by decomposable organic matter, and human and animal related feces, based on the total number of heterotrophic bacteria (TVC) and fecal coliforms (FC).

Applying these criteria, the sampled section of the Noyyal River was perceived to be loaded with easily decomposable organic matter and feces to a wide range between an 'extreme high' to 'very little' extent. This is consistent with the findings of other authors [82] [83].

Fecal indicator bacteria such as fecal *Streptococci*, total and fecal coliforms, show the presence of fecal contamination, and are positively

correlated with the presence of pathogenic enteric bacteria [84].

Water samples from the seven sampling sites on the Noyyal River were assessed for the presence of three important pathogenic bacteria – *Salmonella sp.*, *Shigella sp.*, and *Vibrio sp.* Extremely high counts of *Salmonella sp.* were observed in the last two sampling stations downstream, with lower counts upstream. Very high numbers of *Vibrio sp.* were recorded in the last two sampling stations G (Aathuppalam) and F (Perur), while the highest number of *Shigella sp.* were observed in the last three sampling stations E (Mathampatti), F and G. The other sampling stations showed moderate to low counts of pathogens.

Large volumes of raw sewage and urban runoff could introduce pathogens into rivers [85]. Animal manure also contains many types of pathogens. Manure pathogen levels depend on the source animal, the animal's state of health, and how the manure was stored or treated before use. Rainfall may result in pathogen spread by runoff from stored or unincorporated manure or by leaching [86].

The presence of *Salmonella* in the water might be due to contamination from domestic sewage, agricultural waste and storm water runoffs. Reports of WHO [87] and Arvanitidou *et al.* [88] supports this hypothesis. Another source of *Salmonella* would be from feces of wild and domestic animals in the watershed [89]. *Salmonella typhi* is responsible for salmonellosis, especially typhoid and gastroenteritis. This implies that controlled sewage water systems and personal hygiene will reduce the incidence of gastroenteritis and typhoid fever [90] that might result from contamination.

The presence of *Shigella dysenteriae* might be due to unsanitary conditions and secondary fecal contamination from intermediary sources that entered the river [91]. The implication of this is the risk of possible outbreak of shigellosis or bacillary dysentery when drinking water and food is contaminated [92].

Several studies have reported the isolation of pathogenic bacteria, including *Vibrio parahaemolyticus* [93], non-O-group 1 *V. cholerae* [94], and *V. cholerae* serotype O1 [95] from oysters. Thus, bivalves, which are filter feeders, can accumulate microorganisms from the environment and, thereby transmit these agents to consumers and cause disease.

Aquatic pollution has far-reaching consequences, threatening the habitat and survival of organisms, ultimately affecting man. Pollutants influence various physico-chemical factors, thereby affecting microbial populations, which in turn, interact and respond to changes in these variables, determining occurrence and spread of pathogens. In light of the burgeoning population and increasing demand for clean water, identifying the



source of contamination and minimizing it assumes great significance. Correlation studies and correlation coefficient values can help in selecting treatments to minimize contaminants [96]. Correlation analysis determines the strength of relationship between two variables. In this study, the correlation between various bacteriological parameters was determined.

A strong correlation was observed between the the mean heterotrophic bacterial counts (MHC) and the numbers of *Salmonella sp.* and *Vibrio sp.* The Total Coliform (TC) counts were positively correlated with both Fecal Coliforms (FC) and Fecal *Streptococci* (FS). The counts of *Salmonella sp.* also exhibited a strong positive correlation with counts of, *Vibrio sp.*

Indicators of fecal contamination in water, the fecal coliforms (FC) and *E. coli* (EC) have been shown to be positively correlated with the presence of pathogenic enteric bacteria [84]. In urban watersheds, fecal indicator bacteria are significantly correlated with human density [97]. Mallin *et al.* [98] found that fecal coliform densities were strongly correlated with turbidity (positively) and salinity (negatively).

#### 4. CONCLUSION

From the results of this study it is evident that the river Noyyal is relatively unpolluted upstream near its sources, but is progressively polluted on moving further downstream towards Coimbatore city. Levels of THBC, microbiological indicators such as *Enterbacteriaceae*, TC, FC, FS, pathogenic bacteria like *Salmonella sp.*, *Shigella sp.*, and *Vibrio sp.* were elevated at downstream sampling sites. The release of fecal microorganisms and pathogens from the sediments to the water column during both low and high flow conditions could be a major source of bacterial loading. Evaluation of the quality of water, with respect to TVC, FC and the FC:FS ratio revealed that, the water was unfit for drinking at almost all the downstream sampling stations, and could have limited use for irrigation purposes. Non-point sources were identified as the major contributors of contaminants. Domestic sewage, wastewater runoff, human excrement, wastes of wild / domestic animals, runoff from agricultural land (livestock waste, manure leachate), proximity of cremation grounds, washing of vehicles, etc., are important non-point sources of pollution. The point sources of pollution include, failing septic systems and combined sewer overflows (CSOs).

Improved garbage collection and disposal, construction of sewerage system, setting up sewage treatment plant to treat sewage before discharge into the river, discouraging grazing near the river, and creating public awareness about hygienic sanitary practices, will greatly enhance the water quality of the Noyyal, and reduce incidence of water-borne diseases. The inherent

potential for self-purification by the river and changing hydrological variables will lend a helping hand. Residential, agricultural and industrial development in the area should be carefully monitored along with land use management practices, until a blueprint for improving water quality is formulated. Regular monitoring of the river water and sediment, amelioration of point / non-point sources of pollution, and speedy implementation of remedial measures, may restore this stretch of the Noyyal River to its former pristine state.

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