

## STUDY ON THE ANALYSIS OF PHYSICOCHEMICAL PARAMETER, HEAVY METAL AND MICROBIAL DIVERSITY OF WATER SAMPLES FROM UYYAKONDAN CANAL

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### ABSTRACT

The present study focuses on microbial pollution and antibiotic sensitivity profiling of the microbes like fecal coliforms from River Cauvery along Uyyakondan canal, a major drinking water source in Tiruchirappalli, India. Water samples were collected from hotspots during the year 2019. The physicochemical characteristics and microbial count of water samples collected from most of the hotspots exhibited greater biological oxygen demand and bacterial count especially coliforms in comparison with control samples ( $p \leq 0.01$ ). The antibiotic sensitivity testing was performed using 4 antibiotics against the bacterial isolates with disk-diffusion assay. The current study showed that out of 5 bacterial isolates, 70% ( $n = 3$ ) of the isolates were found to be multidrug-resistant to most of the current generation antibiotics. Among the major isolates, 85% ( $n = 4$ ) of the isolates were found to be multidrug-resistant to 3 antibiotics and they were identified to be gram negative. Almost all bacteria are resistant to at least two antibiotics. Analogously, most of the other Gram-negative bacteria were found to be multidrug-resistant and the Gram-positive bacteria, isolated from the water samples were found to be Ampicillin and Chloramphenicol-resistant. This is probably the first study elucidating the bacterial pollution and antibiotic sensitivity profiling of fecal coliforms isolated from Uyyakondan canal, Tiruchirappalli, India. Compared with all, samples of Kailash Nagar and Periyakulam make pink colour that indicates their pH near 8.2 or above 8.2. Similarly compared with others they have good content such EC (Electrical conductivity), Salinity and TDS (total dissolved salt) in the value of 632 and 834 S/m, 358 and 479 % and 448 and 594 ppm, respectively. In heavy metal analysis, four metals were observed by AAS. Palakarai and Palpannai have higher content of heavy metals other sample.

**Keywords:** Antibiotic sensitivity, Bacteria, Heavy metals, Multidrug Resistance, Uyyakondan canal, Water samples,

### INTRODUCTION

#### Freshwater Ecosystem

The freshwater ecosystem comprises probably less than one percent of the total surface of the earth and is produced by gigantic distillation processes in nature. Water evaporates from the ocean and land surface and is carried up into the atmosphere and precipitates as rain or snow on the Earth's Surface. A portion of the rain water on the land is absorbed into soil, some part of it evaporates

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and excess water is either drained off into the lakes and ponds or flows back into the sea through a riverine system. Water locked up in lakes and ponds forms 'lentic bodies' and the running water of rivers and streams forms the 'lotic bodies'.

### **Components of the freshwater ecosystem:**

The freshwater ecosystem consists of three components:

- 1) The autotrophs, producers that fix energy of the sun;
- 2) The heterotrophs, consumers and decomposers that utilize the energy and nutrients fixed by the producers and return nutrients to the system
- 3) The dead organic material and inorganic substrate that act as short term nutrient pools and maintain cycling of nutrients within the system.

### **Role of microbes in the aquatic ecosystem:**

Decomposition of organic matter in fresh water is brought about by diverse aquatic biota namely bacteria, fungi, nematodes and worms. Aquatic microbes have been reported to improve the palatability and nutrient content of the plant and animal remains. The survival and success of microbes as decomposers are largely dependent on their ability to adapt to the environment immediately surrounding the substratum and to produce viable reproductive units. Along with nature of substrate, ever-changing factors like light, temperature, oxygen and hydrogen ion concentration act either singly or in concert to influence substrate colonization, growth and reproduction of a fungus. In the natural environment, substrate degradation is achieved by association of succession of taxonomically unrelated microbes and other organisms adapted to, or tolerant to the special environmental conditions associated with the fluid medium. Most of the water systems like seas, rivers, streams and lakes are the final repositories of all effluents, impurities, refuse, wastes and throw offs of all the industries, mills, factories, tanneries, and distilleries located around them. The pollution of the aquatic environment greatly affects the population of aquatic organisms including microbes.

In general, the distribution of the fungal components of the microflora of the riverine systems and the impact of pollutants on their distribution have not been worked out in detail. Almost all major rivers in India including the river Kaveri are getting polluted due to urbanization and industrialization in the riverine belt and these adversely affect the physical, chemical and biological characteristics of the aquatic systems. Therefore a study on microbes and their ecology in the Uyyakondan canal has been taken up for present investigation. Among the decomposers, the fungi and bacteria are of equal importance in performing the vital role of reducing organic matter to inorganic form which may then be used again by the producers.

Our nature is made up of five basic elements: earth, water, fire, air, and space. If any element is impure or out of balance with another it leads to many diseases. Each element is responsible for different structures in the body. Water is one of most important commodities, which man has exploited than any other resources for the sustenance of his life. Water is not only the basic need for human existence but also a vital input for all development activities. The use of water for drinking and other domestic purposes is generally conceded to be its highest and most essential purpose. Pollution of water bodies is increasing steadily due to rapid population growth, industrial proliferation, urbanization, increasing living standards, and wide sphere of human activities. Kaveri which is also known as "Dakshin Ganga" is one of the major rivers of Tamilnadu. The river basin covers an area of about 87,900 km<sup>2</sup>, spread over the states of the Western Ghats at an elevation of 1345 m above mean sea level and extends approximately 800 km to the Bay of Bengal (Radhakrishna 1993).

The area mentioned above, taken up for the present study is unique in the following aspects;

The river has supported irrigated agriculture for centuries and served as the lifeblood of the ancient kingdoms and modern cities of South India. The canal, which originates from the Cauvery near Pettavaithalai and traverses about 70 km up to Valavanthankottai tank, irrigates about 32,000 acres. However, in the fallout of urbanization, the 7-km city stretch has become an

eyesore with several opendrain and sewage from houses being let out directly into it. As part of the rehabilitation, Public Works Department (PWD) sources say that it would take up bed lining and construction of a retaining wall for a stretch of 1,250 meters in the Palakkarai area, where the canal was subject to the heaviest pollution. Statistical analyses and experimental data revealed that the Uyyakondan river may cause severe health risks to the recreational users and fisher folk, ultimately warning environmental quality management to control heavy metal contamination. In the recent past, the biological oxygen demand (BOD) and heavy metal contents in the river line water bodies have increased dramatically. The river, being a natural dumping ground for the industries situated along the bank, discharge of sewage from various industries, hospitals, and pharmaceutical sectors to the water bodies leads to high turbidity, reduced transparency, and increased suspended solids and heavy metals. Such environments might have a direct influence on the proliferation of fecal coliforms which cause several health hazards. (Gazzaz, 2012; Umamaheswari and Saravanan, 2009).

The enormous accumulation of domestic and industrial wastes along with pesticide residues resulted in the massive multiplication of various pathogenic microorganisms in River Cauvery. This paves way for many waterborne diseases (Sharma, 2012) such as typhoid, diarrhea, cholera, dysentery, and gastroenteritis (Moon *et al.*, 2014). Recent reports have revealed that many bacteria mutate and acquire genes responsible for multiple drug resistance (MDR) to the present generation antibiotics and have emerged as “superbugs.” At present, many freshwater ecosystems have turned into reservoirs for antibiotic-resistant bacteria (Xu *et al.*, 2014). Hence, there is an urgent need to assess the physicochemical and bacteriological status of the parent river which acts as the major source of drinking water and agriculture for the urban and rural residents in Tiruchirappalli, India. As the information available about the microbial flora present in River Cauvery is sparse and limited and it is important to analyze the microbial populations, especially with the prevalence of multidrug-resistant bacteria such as fecal coliforms, fecal *Streptococcus*, and *Staphylococcus*. This is probably the first study reporting on the physicochemical characteristics and fecal microbial population, predominance of multidrug resistance

and heavy metal in the water samples collected from the Uyyakondan canal. Besides, fencing would be done to prevent the dumping of solid wastes into the canal. The present study focuses on the prudent elucidation of microbial pollution and antibiotic sensitivity profiling of the fecal coliforms isolated from River Cauvery along the Uyyakondan canal, a major drinking water source in Tiruchirappalli, India.

## Materials and Methods

### Study area

Uyyakondan canal covering a length of 60 km, is an important irrigation canal, originating at Mayanur from Kaveri and completing its course at Valavanthankottai (about 20 km east of Tiruchirappalli town). This canal irrigates vast stretches of agricultural lands used for cultivating rice, banana and sugarcane. This canal receives water for irrigation almost throughout the year. At Tiruchirappalli town, this canal receives pollutants from domestic, agricultural and industrial sources. Study area is the riverine system of Tiruchirappalli district. Tiruchirappalli is a significant city in Tamil Nadu, southern India. Ten different sampling sites have been selected from the study area.

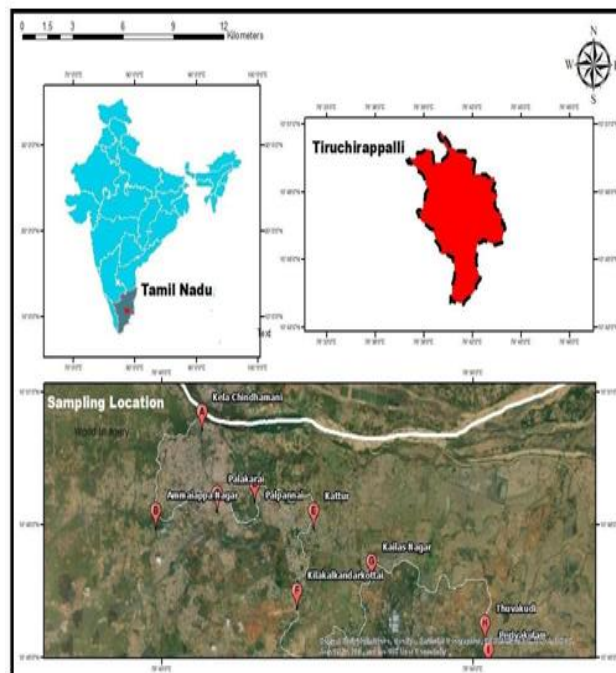


Fig. 1. Shows the sampling location for the study

**Table 1- Sampling sites with their longitude and latitude**

S.No	Place	Longitude	Latitude
1	Kila Chinthamani	78°41'17.09"E	10°50'25.19"N
2	Ammayappan nagar	78°39'47.61"E	10°48'12.32"N
3	Palakarai	78°41'45.82"E	10°48'34.48"N
4	Palpannai	78°42'57.81"E	10°48'47.61"N
5	Kattur	78°44'52.15"E	10°48'11.62"N
6	Kailash nagar	78°46'43.85"E	10°47'2.47"N
7	Kilakalkondankotai	78°44'19.66"E	10°46'23.85"N
8	Thuvakudi	78°50'22.89"E	10°45'39.40"N
9	Periyakulam	78°50'29.08"E	10°45'3.27"N

### Sampling:

In December 2019, the sample collections were done. Physicochemical parameters were analyzed at the spot by Winkler's method. We collected 1 liter of water sample from each location for heavy metal analysis in a Screw capped polythene bottle and also 10 ml of samples were collected from each site for biological analysis by us that had been kept in an ice bag.

### Physico-chemical analysis of water samples

Physicochemical characterization was through estimation of biological oxygen demand. The time, temperature, pH, and other physiological parameters during the time of sample collection were recorded by Multi-Parameter Analyzer ST400M-B, UTECH. Further, the dissolved oxygen (DO) and BOD were estimated by Winkler's method (Shriwastav *et al.*, 2010) for 5 days.

### Isolation and counting of bacteria

One-milliliter water from each of the sample was serially diluted and plated in nutrient agar, MacConkey's agar (Hi-Media, India) by standard plate techniques (Geldreich *et al.*, 1972). These plates were incubated at 37 °C for 24 h. The experiment by nutrient agar was replicated as three independent trials. The morphological characteristics of each colony were analyzed, and the viable bacterial count (colony-forming units

(CFU) per milliliter) was enumerated by digital colony counter (Labtronics, India).

### Microbial characterization of isolated bacteria and fungi

The morphological characteristics of isolated bacteria were studied by Gram staining (Beveridge, 2001). The isolates were further characterized by standard biochemical tests. The biochemical tests performed were MR- VP, oxidase (Gordon and McLeod, 1928), catalase (Kubica *et al.*, 1966) triple sugar iron (Sulkin and Willett, 1940), and Simmon citrate agar (Hedberg, 1969).

### Antibiotic susceptibility test of isolated microbes

The antibiotic sensitivity testing was performed on isolated bacteria using Mueller-Hinton agar (Hi-Media, India) by disk-diffusion method (Bauer *et al.*, 1966). The total numbers of antibiotics are 4 which are Ampicillin, Chloramphenicol, Gentamicin and Polymyxin B. Mueller-Hinton agar plates were aseptically inoculated with the test organism by lawn culture technique and filter paper disks, impregnated with a specific concentration of antibiotic, was placed on the medium. The plates were incubated at 37 °C for 24 h. The procedure was performed as three independent replicates, and the mean value of diameter of the zone of inhibition was measured and compared with standard (Clinical and Laboratory Standards Institute, 2013) and their resistant, intermediate, and sensitive patterns were recorded. The activity of anti-fungi was determined by following the same procedure, but small change especially, potato dextrose agar was used to the culture medium and also plate was incubated at 28±2°C for 72 hours.

### Heavy metal analysis of water samples

The collected water samples were filtered through Whatman filter paper No. 42 (2.4-µm pore size). The filtered water sample was taken in an acid-cleaned separating funnel, added the 10 ml of 1 % ammonium pyrrolidine dithiocarbamate (APDC). The chelates were extracted by using methyl isobutyl ketone (MIBK) after rinsing for 15 min. The aqueous phase was removed and the organic phase was collected. The metal present in the organic phase was back extracted with 50 % HNO<sub>3</sub> and making up 25 ml with high purity water

(Jonathan *et al.*, 2008). The metals present in extracts of water and sediment were analysed by flame Atomic absorption spectrometer (GBC HG 3000; Sens AA, Australia; 2009). The working wavelength for the heavy metals are 213.9 nm for Zn, 324.8 nm for Cu, 228.8 nm for Cd and 283.31 nm for Pb. Quality assurance testing relied on the control of blanks and yield for chemical procedure. For quality assurance, replicate samples, blanks and standardized reference materials were used during analysis. The standards of the metal samples were prepared in deionized water using the serial dilution in the range 0.5–7 ppm and were run to check the precision of the instrument throughout the analysis.

## Results and Discussion

### Physicochemical analysis

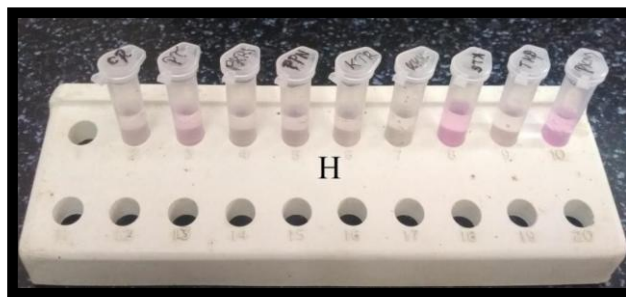
The physicochemical parameters recorded at the time of sample collection are given in Table 2. The temperature and pH were found to be ideal for the bacterial growth in most of the sampling spots. The BOD was found to be greater in most of the sampling areas ranging from 1.6 to 2.8 mg/L in January 2020 in comparison with the control water samples. The control water samples showed the BOD values within the permissible limits. The samples collected from Pettavaithalai (2.8 mg/L), Palakarai (1.6 mg/L), Palpannai (1.65 mg/L), Kattur (1.8 mg/L), and Periyakulam (2.0 mg/L) showed increase in the BOD content at each time of sample collection (Table 1). The statistical analysis indicated that variations observed in the mean BOD values of the sample collections were found to be significant. This increase in the biological oxygen demand was probably due to the sewage accumulated from temples, tourists, organic and agricultural farms, residents, hospitals, and municipal and industrial sectors along the river basins during the month of December and January which was visible during the survey of this study. These 2 months are the post monsoon season in Tiruchirappalli which are expected to contribute high loads of waste materials from all these sectors compared to other seasons. However, the waste garbage generated during the other three seasons is also contributing to high levels of pollutants. The mean BOD levels in most of the water samples

collected were exceeding the permissible limits suggested by European Union for aquatic life (Chapman, 1996). The BOD values of polluted water systems are estimated to be approximately 2 mg/L, and such BOD values were observed in many places along the banks of Uyyakondan canal in this study. Such kind of bulk liquid BOD concentration has direct influence over the microbial community in the water bodies especially the growth of coliforms (Downing and Nerenberg, 2008).

**Table 2- Physicochemical parameter result of water sample**

Places	TDS	Salinity	pH	Temperature °C	EC
Kilachindhamani	248 ppm	200 g/kg	7.52	29.3	548 S/m
Ammaiyappan Nagar	446 ppm	375 g/kg	7.32	28.8	678 S/m
Palakarai	322 ppm	255 g/kg	7.17	29	453 S/m
Palpannai	346 ppm	275 g/kg	6.9	28.9	483 S/m
Kattur	352 ppm	280 g/kg	6.86	29.1	496 S/m
Kilakalkondan kottai	360 ppm	286 g/kg	6.86	29	508 S/m
Kailash Nagar	448 ppm	358 g/kg	8.4	29.7	632 S/m
Thuvakudi	594 ppm	479 g/kg	6.82	29.9	837 S/m
Periyakulam	435 ppm	348 g/kg	8.22	29.7	613 S/m
RO (Reverse osmotic)	33.5 ppm	33.6 g/kg	7.2	30.6	47.5 S/m
DD water	46.8 ppm	42.8 g/kg	7.2	29	64.4 S/m
Kaveri river	368 ppm	294 g/kg	7.4	30.2	501 S/m
Pettavaithalai	382 ppm	304 g/kg	7.95	31.3	539 S/m

Ammaiyappan Nagar, Kailash Nagar and Thuvakudi are gave high values for TDS, EC and salinity.

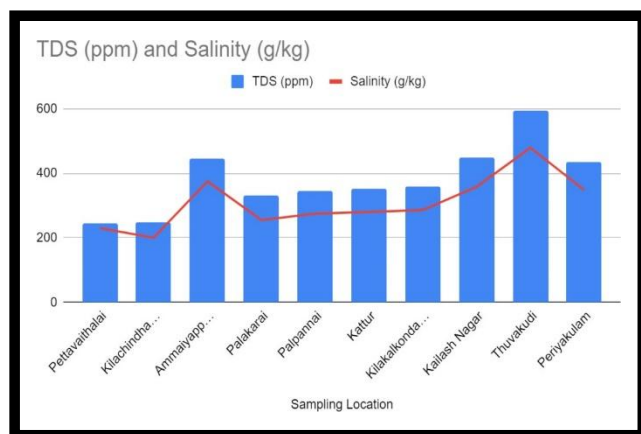


**Figure 2. pH of the sample tested with Phenolphthalein indicator**

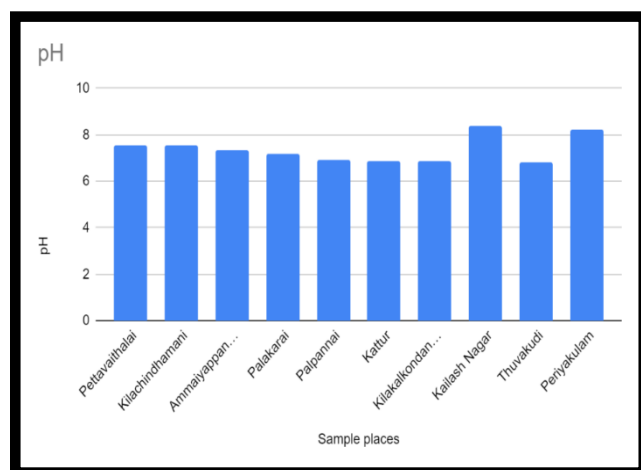
Phenolphthalein reacts with hydroxyl groups and produces pink colour when sample pH above 8.2. Sample of Kailash Nagar (KLS) and Periyakulam (PKM) produce pink colour and so their pH is near 8.2.



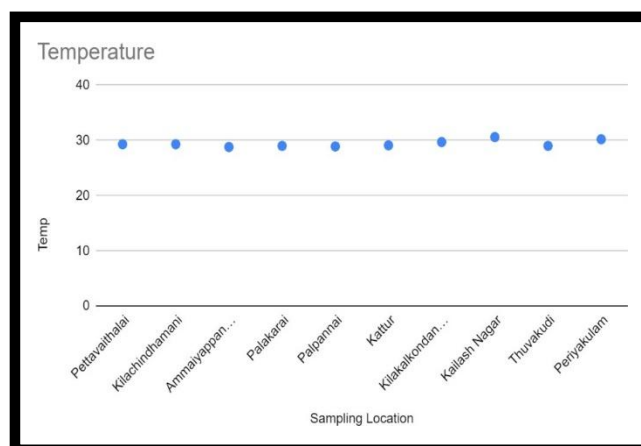
**Figure 3.** TDS of the samples Ammaiappan Nagar, Kailash Nagar and Thuvakudi have given high values of TDS in water samples.



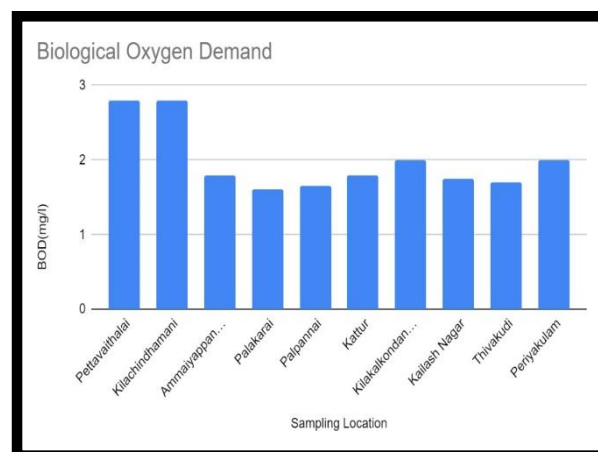
**Figure 4.** pH of the samples as a result, Kailash Nagar and Periyakulam have above 8 in pH



**Figure 5.** Temperature of the samples. All samples are commonly defined a temperature at 30°C.

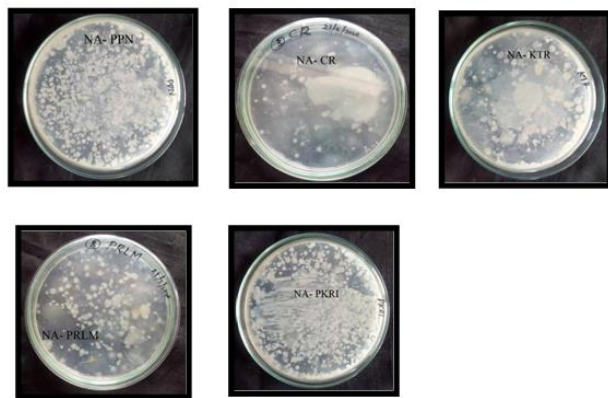


**Figure 6.** BOD of the samples.

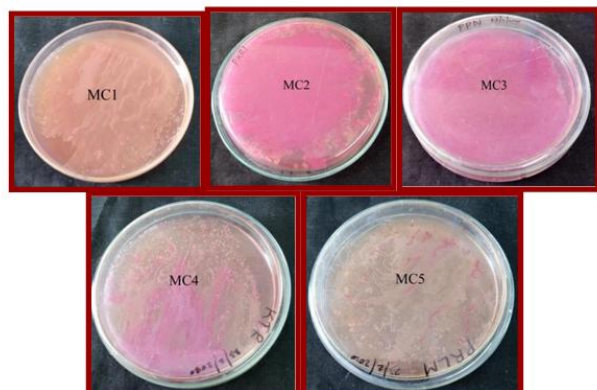


### Isolation and counting of bacteria

The number of viable bacterial counts (CFU/ml) estimated from most of the sampling sites were high when compared with the quality of drinking water prescribed by WHO (Aryal *et al.*). The viable bacterial counts estimated were found to be  $1.7 \times 10^5$  to  $6.8 \times 10^5$  CFU/ml during January 2020. The total bacterial count from the control water samples were found to be within the permissible range as per American Public Health Association (2005). Among the five sampling areas studied, Pettavaithalai ( $1.7 \times 10^5$  CFU/ml), Palpannai ( $6.8 \times 10^5$  CFU/ml), Palakarai ( $6.2 \times 10^5$  CFU/ml), Periyakulam ( $3.3 \times 10^5$  CFU/ml), and Kattur ( $5.2 \times 10^5$  CFU/ml) displayed bacterial count. The variations observed in the bacterial count during the sampling from hot spots were found to be significant. This study suggests that the water samples collected from Palpannai showed maximum bacterial count in comparison with other sampling spots. It is clear that there was a high relation with flow gradient towards the total count as Palpannai is at the lowest sampling point in the study. The current study suggests that the bacterial counts in these hot spots are increasing with respect to time. These observations were made based on one time samplings. Hence, further studies for a longer term need to be performed to conclude the relationship with increasing mode of bacterial count, BOD and heavy metal.



**Figure 7.** Nutrient agar plates Show the presence of different species of bacteria in water samples. Palakarai and Palpannai have more bacterial content than other sites.



**Figure 8.** (MC)-Shows different bacteria from the sample in MacConkey agar.

MacConkey agar is the selective agar for gram negative bacteria. Enteric bacteria that have the ability to ferment lactose can be detected using the carbohydrate lactose, and the pH indicator neutral red. The plate of Palakarai (MC3) and Palpannai (MC2) have good growth of lactose fermentative bacteria than others.

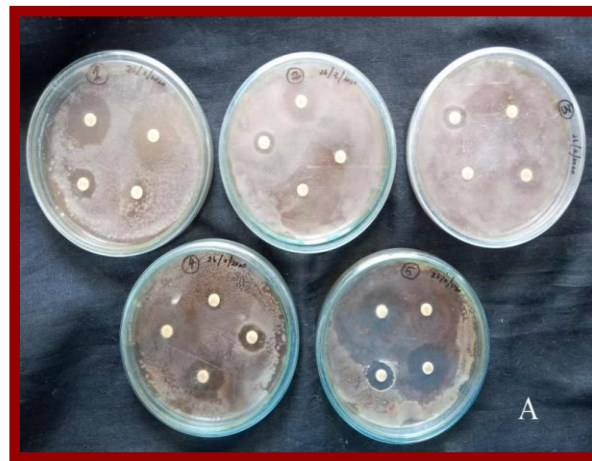
### Antibiotic susceptibility test of isolated microbes

The present study demonstrated that 93.51 % ( $n = 4$ ) of the bacterial isolates were found to be drug resistance to most of the current generation antibiotics. The study revealed that two main isolates were found to be drug resistant to 4 different antibiotics. From the antibiotic sensitivity testing, it is clear that isolates, such as *gram positive* and *gram negative* have acquired drug resistance to most of the carbapenem groups which includes

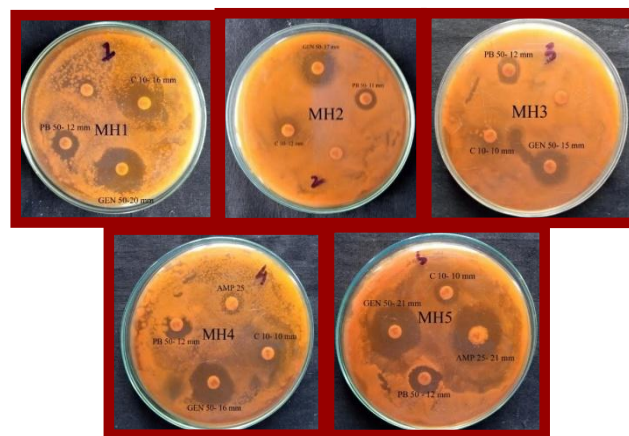
*Ampicillin, Chloramphenicol, Gentamicin and Polymyxin B*. These antibiotics are regarded as the strongest  $\beta$ -lactams and last line drugs currently available against these Gram-negative enteric pathogens.

### Bacteria:

We have identified only five different bacteria based on their morphological variation. So those are tested by antibiotics.



**Figure 9.** comparison of the different types of bacterial reaction against antibiotics



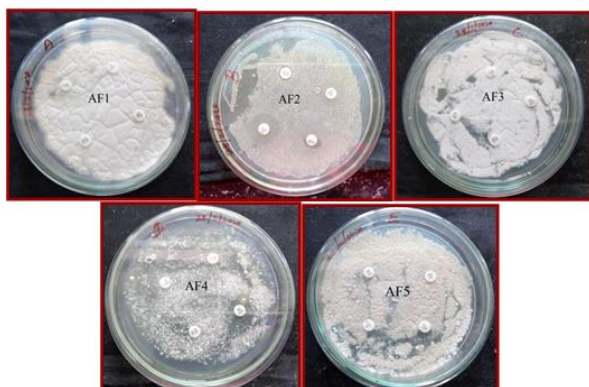
**Figure 9-B.** Individual bacterial susceptibility tests

Gentamicin only gives the good sensitive activity to bacteria. MH5 bacteria are only sensitive to all test drugs. In antibiotics resistance tests, MH3 bacteria is a good result. The antimicrobial resistance patterns of the minor bacterial isolates obtained from various hotspots of Uyyakondan canal during jan-2020. The assay was performed as three independent replicates, and the mean value of

the diameter of the zone of inhibition was compared with standard chart (CLSI, 2013). Figure shows that the isolates of bacteria showed resistance to antibiotics, in which most of them are expected to be acquired resistance.

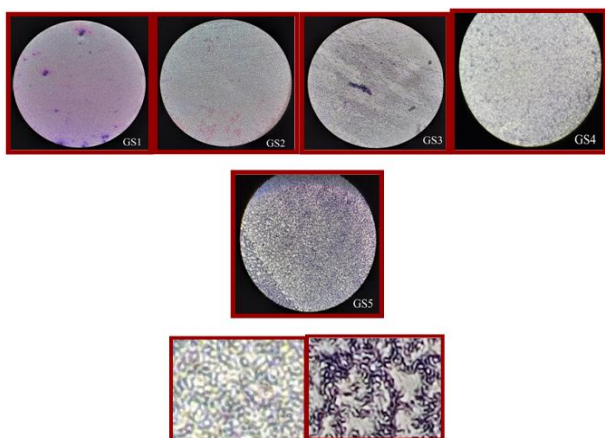
## Fungi

We have selected only five different bacteria based on their morphological variation. So those are tested to determine the resistance by using antibiotics.



Different fungal colonies were isolated by their morphology and then examined their antibiotics sensitivity. All fungi are resistant to tested antibiotics such as Gentamicin, Ampicillin, Polymyxin B and Chloramphenicol.

## Microbial characterization of isolated bacteria & fungi



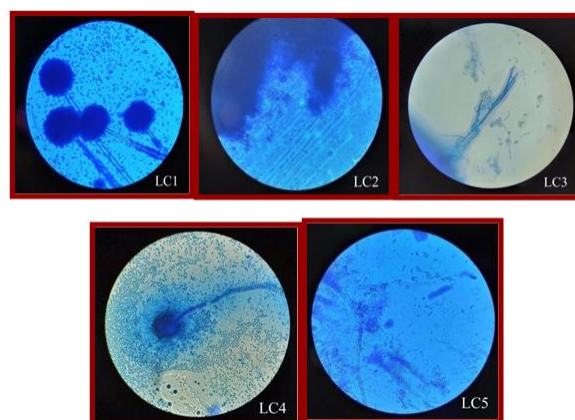
## Bacteria

Five different species of bacteria were isolated based on their morphological visual observation.

Bacteria were isolated by their different morphological colour and classified based on their shape by gram stain with the help of microscope. From Gram's staining, GS1 and GS2 appear in pink colour. GS3 and GS5 are in rod shape. As a result, coliform microbes may be higher in water.

## Fungi

Five different species of fungi were isolated based on their morphological visual observation.



**Figure 11.** Staining for Fungi (lactophenol cotton blue stain)

Fungal isolates were stained by Lactophenol cotton blue to analyze their phenotypic structure. From microscopic observation, they related to *Penicillium* sp, *Geotrichum* sp, *Mucor* sp, and *Aspergillus* sp.

## Biochemical test

The enzyme, catalase, is produced by bacteria that respire using oxygen, and protects them from the toxic by-products of oxygen metabolism. In the catalase test, all bacteria react with hydrogen peroxide and make foam that is positive. The oxidase test is a test used in microbiology to determine if a bacterium produces certain cytochrome c oxidases. 3<sup>rd</sup> and 4<sup>th</sup> tube of bacteria negatively respond to oxidase. Phenol red indicator present in media that used to indicate media whether acid or alkaline. If red colour formed, alkaline level. Hydrogen sulphite makes the black colour formation in the media. It is useful

**Figure 10.** (GS)- Gram's staining result of isolated bacteria



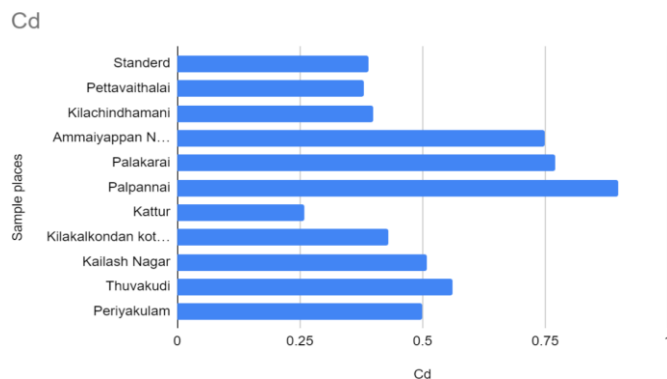
for selecting organisms that use citrate as its main carbon and energy source. Every bacteria gives a positive result.

Methyl Red (MR) test determines whether the microbe performs mixed acids fermentation when supplied glucose. Methyl red (p-dimethylamino aeo benzene-TO-carboxylic acid), which is yellow above pH 5.1 and red at pH 4.4. In 4 and 5th tubes have red colour on the top layer that is positive and others are negative. They first observed the red color reaction produced by appropriate culture media after treatment with potassium hydroxide. It was later discovered that the active product in the medium formed by bacterial metabolism is acetyl methyl carbinol, a product of the butylenes glycol pathway. All bacteria give negative results. VP2 has dark red colour in their top and so to be positive.

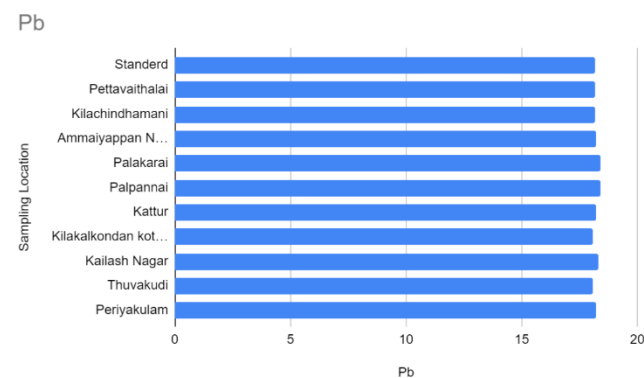
## Heavy metal analysis of water samples

**Figure 12.** Cd presence level in each site

Approximately 0.75 ppm of Cadmium in water sample of Ammaiyappan Nagar, Palakarai and Palpannai.

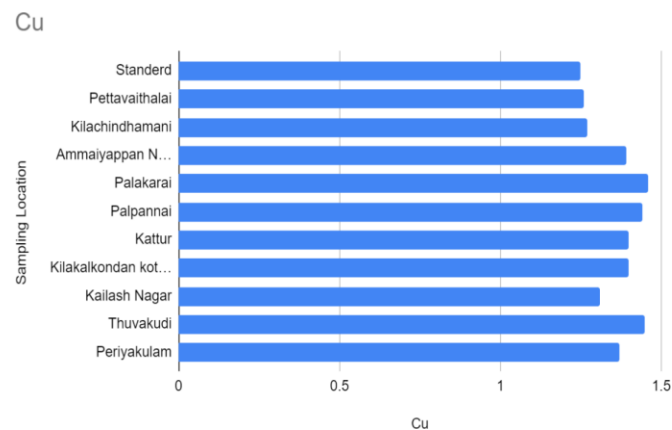


**Figure 13.** Pb presence level in each site  
Approximately 18 ppm of Lead in all water sample



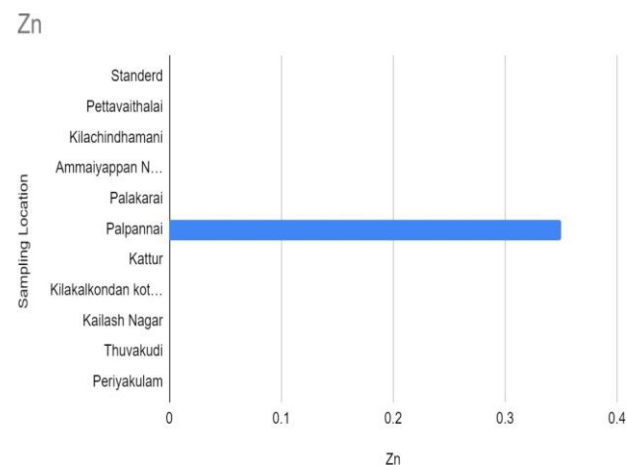
**Figure 14.** Cu presence level in each site

Compare with other, Palakarai, Palpannai and Thuvakudi give the high result in above 1.38 ppm



**Figure 15.** Zn presence level in each site

Palpannai only have Zinc metal. Other site doesn't have result



The chart shows the positive result that detected metal in water. Palpannai (VY11) and Palakarai (VY12) have huge amount of heavy metal than other place, Cd-  $1.44 \pm 0.02$  &  $1.46 \pm 0.03$  ppm and Cu-  $0.90 \pm 0.10$  &  $0.77 \pm 0.54$  ppm. Kilachindamani (VY2), Rockfort (VY5) and Thiruvanaikovil (VY) better water quality compare with other sites, Cd-  $1.27 \pm 0.08$ ,  $1.36 \pm 0.01$  &  $1.31 \pm 0.07$  ppm and Cu-  $0.40 \pm 0.02$ ,  $18.15 \pm 0.08$  &  $0.46 \pm 0.02$  ppm. Commonly, Metal Pb is present in every site at above 18 ppm.

Recent reports have revealed that many pathogenic *E. coli* can survive in soil environments which resulted in the contamination of fresh water systems in East China (Wang *et al.*, 2014). Studies from California and Arizona, USA showed the ecological habitat and persistence of

Enterohemorrhagic *E. coli* in leafy green producing soil (Ma *et al.*, 2012). The major portion of the Cauvery river covers leafy green producing soils which could be one of the reasons for predominance of these bacteria in the banks of the River. Recently it was reported that *E. coli* isolated from selected surface water sources in Zaria, Nigeria was found to be multidrug-resistant to most of the  $\beta$ -lactams, fourth generation cephalosporins and fluoroquinolones. The antimicrobial resistance, virulent factors and genetic diversity of different isolates of *E. coli* from household water supply in Dhaka, Bangladesh (Talukdar *et al.*, 2013) and San Pedro River, Mexico were also reported (Ramírez Castillo *et al.*, 2013). The current study revealed that most of the *Shigella* spp., isolated from River Cauvery has become extremely resistant to even fourth generation cephalosporins and aminoglycosides. Similar types of extended spectrum of  $\beta$ -lactamase mediated third generation cephalosporin resistance were reported in Southern Vietnam (Vinh *et al.*, 2009), Bangladesh (Rahman *et al.*, 2004), Israel (Vasilev *et al.*, 2007), Iran (Tajbakhsh *et al.*, 2012) and Spain (Arias *et al.*, 2006). The emergence of MDR *Enterobacter cloacae* against carbapenem and related antibiotics are considered as major public health concerns in Spain (Fernández *et al.*, 2011) and Northern Iran (Bayani *et al.*, 2013). This study also emphasizes the massive occurrence of MDR in Uyyakondan canal which can become a critical health issue in India in the near future.

The massive discharges of sewage from pharmaceutical industries, chemical and electroplating industries, animal farm houses, hospitals, and temples have drastically polluted the river and contributed to the high content of organic matters in the water leading to the change in the physico chemical status of water and in turn favored the growth of coliforms. The aquatic ecosystems such as rivers and streams are ideal reservoirs for antibiotic resistant bacteria (Ramírez Castillo *et al.*, 2013). The misuse and overuse of antibiotics contribute to the selection and persistence of antibiotic resistant bacteria (Amabile-Cuevas, 2010). The acquisition of resistance genes in the bacteria by various methods minimize therapeutic options and lead to frequent treatment failures. The prevention and control of MDRs have become a global priority and one that requires all healthcare facilities and agencies to assume

responsibilities. By considering the entire medical, social, economic, and environmental relevance, there is a necessity to understand the antimicrobial resistance profile and undertake all precautionary measures to maintain the natural status of the Uyyakondan canal.

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