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# Microbiological Analysis of Municipal Supply Water in Allahabad City

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***Abstract:*** ***The bacteriological analysis was carried out on the available water from municipal supply sources in the city of Allahabad, Uttar Pradesh, in order to check the water quality and evaluate the awareness of people for maintaining cleanness and hygiene conditions for water which is being provided to them. A total of 20 samples from municipal water supply were collected from random spots throughout the city which were being used by the community for drinking and other household purposes. The water samples were tested using serial dilution pour plate technique and all of the samples were found to have contamination more than the permissible range. Among all the samples analyzed none complied with the standard provided by Indian Standard IS 1622 (1981), according to which no sample should contain E. coli as well as any other coliform organisms more than 10 per 100ml. The presence of pathogens in supply water for drinking and other purposes is of public health significance considering the possibility of the presence of other bacteria, protozoa and enteric viruses that are involved in gastro-intestinal water-borne diseases and the low infective dose for these waterborne pathogens.***
***Keywords: E. coli, bacteria, protozoa, enteric viruses***

## I. INTRODUCTION

It is a common saying that there is No life without water depending upon the fact that water is one of the naturally occurring essential requirements of all life functions. It is an important solvent, and all metabolic reactions of living beings depend on its presence. The human body has 55% to 78% water depending on the size of the body. Thus, potable water is always a necessity for living organisms. Water is a resource that has many uses, including recreational, transportation, and hydroelectric power, domestic, commercial, and industrial uses. Water covers 70.9% of the Earth's surface and is essential for all known forms of life. On Earth, it is spotted mostly in oceans and other large water bodies, with 1.6% of water found below the ground in aquifers and 0.001% in the air as vapour, clouds, and precipitation. Water is also present in the atmosphere in liquid, solid, and vapour state. Water plays an important role in the world economy, as it functions as a solvent for a plethora of chemical substances and facilitates industrial cooling and transportation. Approximately 70% of freshwater is utilized by agriculture. (Baroni et al., 2007). Freshwater is a limited resource, necessary for agriculture, industry and even human existence, without fresh water of appropriate quality and quantity, sustainable development will not be feasible.

As we know, human populations living in towns, cities, etc. depends upon the municipal supply of water. The most prevalent and broad danger associated with drinking water is contamination, either directly or indirectly, by sewage, other wastes or animal and human excreta. About 25 years ago, authoritative estimates indicated that each year some of 500 million people are affected by water-borne or hater associated disease, and as many as 10 million of these die. In a recent estimate based on WHO reports suggest that 80% of all human illnesses in the developing world are caused by biological contamination. Faecal pollution of drinking water may introduce a variety of intestinal pathogen. Their presence being related to microbial diseases and carriers present in the community, which may cause diseases from mild gastroenteritis to severe and sometimes fatal dysentry, cholera or typhoid.

Ideally, drinking water should not contain any microorganisms which are pathogenic. It should be free from bacteria indicatory of pollution with excreta. Detection of faecal indicator bacteria in drinking water lay out a very delicate method of quality analysis and it is not possible to examine water for every possible pathogen that might be present (WHO, 1993). The most important pathogenic bacteria transmitted by the water route are *Salmonella typhi*, the organism causing typhoid fever, and *Vibrio cholerae*, the organism responsible for causing cholera. The majority of the population in developing countries is not supplied with potable water, and thus are obliged to use unsafe water for drinking and domestic purposes.

Hence, it is essential to check the quality of the available supplied water from various sources. In view of the above-mentioned facts the present study entitled "Microbiological analysis of municipal Water supply in Allahabad city" was designed to analyze the microbial estimate of the available water from various Municipal supply sources across the Allahabad city to facilitate the examination of the level of contamination.

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## II. MATERIALS AND METHOD-

The study was designed to analyze the microbial quantity of the drinking water supplied by the municipal corporation in different regions of the city of Allahabad in order to check the level of contamination. Accurate and reliable laboratory methods and analyses are a prerequisite to ensure potable drinking water characteristics and thereby following experimental plan was undertaken.

### *A. Place Of Work*

The present study entitled "Microbiological analysis of municipal Water supply in Allahabad city" was carried out in Post Graduate Department of Botany, Ewing Christian College, Allahabad, Uttar Pradesh.

### *B. Study Sample*

A Total of 20 Water samples were collected from different public municipal water sources across the different locations of Allahabad city.

### *C. Collection And Transportation Of Samples*

Water samples were collected in sterile sampling bottles. A total number of 20 samples were collected randomly from the city area as per APHA (1992) Standard protocol and transported to the laboratory of mentioned college in sterile and safe condition.

### *D. Isolation Of Target Organism*

To study the microbiological quality of collected water samples selected bacteria namely (*Escherichia coli, Staphylococcus aureus,* and faecal *Streptococci* ) were isolated on their respective selective media as per the Standard protocol. The test was done to isolate the number of Colony Forming Units (CFU) of target bacteria present in 1ml of the water sample with a dilution factor of $10^{-6}$ on selective media Eosin Methylene Blue agar, Mannitol Salt Agar and MacConkey Agar respectively.

The Bacteriological analysis of Municipal water sample was done within 48 hours of collection. Water samples were analyzed for Colony Forming Units (CFU) using the serial dilution Pour Plate technique ($10^{-6}$ being the dilution chosen) as well as the prevalence of various waterborne pathogen and Indicator organisms examined using selective media and tests for identification of bacteria via Gram Staining technique (Differential Staining).

The serial dilution method is used to obtain a pure culture of the target organism. Pour plate method is the method of choice for counting the number of colony-forming bacteria present in a liquid specimen.

### *E. Isolation OF Escherichia coli*

One ml water sample was subjected to a series of dilution to reach the desired dilution unit ($10^{-6}$) using sterile water blanks. 1ml of this diluted inoculum was placed in the center of a sterile Petri dish using a sterile micropipette. Molten cooled selective cultures medium, Eosin Methylene Blue, (approx.15ml) is then poured into the Petri plates containing the inoculum and mixed well. After the solidification of the media, the Petri plates were inverted and incubated at 37°C for 24-48 hours using incubator (Sanders,2012). Then after which the results were analyzed and photographs were taken.

### *F. Isolation OF Staphylococcus aureus*

Mannitol salt agar was chosen as the selective media for isolation of bacteria; Rest of the methodology remains the same as mentioned above.

### *G. ISOLATION OF faecal Streptococcus*

MacConkey Agar was chosen as the selective media for isolation of bacteria; the rest of the methodology remains the same as mentioned above.

Microorganisms will grow both on the surface as well as inside the medium. Colonies that grow inside the medium are generally smaller in size and may be merged; the few that grow on the agar surface are of the similar size and appearance as those on a streak plate. Each ( large and small) colony is carefully counted (using magnifying colony counter). Each colony represents a "colony forming unit" (CFU). The number of microorganisms present in the test sample is determined using the formula:

### *H. Cfu/Ml= Cfu * Dilution Factor * 1/Aliquot*

For accurate counts, the most appropriate count should be within the range of 30-300 colonies/plate.

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### *I. Cultural And Morphological Examination*

The characteristics of colonies such as colony shape, colour, elevation etc. were studied after incubating different sample plates for 24-48 hours. The morphological examination of isolated bacteria was done by Gram's Staining method as per the procedure given by Dr. R. P. Singh in his book Microbiology 2nd edition, 2008.

## III. RESULT AND DISCUSSION

The present study was conducted to comparatively analyze the municipal supply water for quantitative and qualitative estimation of the target organism. The study reveals that none of the samples collected from four zones of the city comply with the standards. According to the Indian standard IS-1622 (1981), not a single sample should contain *E. coli* as well as other coliform organisms more than 10 per 100 ml. Bacteriological quality of all the samples are shown in Table II.

After incubation of Petri plates for 24-48 hours, the morphological characteristics of colonies isolated on selective media were studied and tabulated in table I. The colonies obtained on EMB were Flat, circular, purple in colour with dark centers. When incubated for more than 48 hours the colonies showed a distinctive metallic green sheen. The colonies obtained on MSA were round, opaque, convex, golden-yellow with yellow zones and entire margin. Some other coagulase stains were also obtained that produced small pink or red colonies with no colour change to medium. The colonies obtained on MacConkey Agar were round, convex, and light pink coloured with an entire margin.

The colonies were picked from the culture plates in a sterilized condition and identified via the gram staining technique.

Table I: Colony Characteristics

| Media | Elevation | Shape | Colour | Cellular arrangement | Gram reaction |
|---|---|---|---|---|---|
| Emb | Flat | Circular, entire | Purple with dark centre | Isolated rods | -ve |
| Msa | Convex | Circular, entire | Golden yellow with yellow zones | Cocci in grape-like clusters | +ve |
| Mac | Convex | Circular, entire | Light pink | Cocci arranged in chain like fashion | +ve |

Table II: Sample Wise Distribution Of Target Organism From Different Areas Of City(In Cfu/Ml)

| SAMPLE NO. | PLACE | E. coli ($No.x10^6$) | S. aureus ($No.x10^6$) | Faecal Streptococci ($No.x10^6$) |
|---|---|---|---|---|
| 1 | Dariyabad | 52 | 239 | 170.5 |
| 2 | ECC | 362 | 2 | 46 |
| 3 | Kareli | 25.5 | 73.5 | 90.5 |
| 4 | University Road | 65 | 55 | 21 |
| 5 | Katghar | 48.5 | 46 | 53.5 |
| 6 | Bethany Convent, Naini | 14 | 27.5 | 8.5 |
| 7 | Attarsuiya | 52 | 18.5 | 4 |
| 8 | ADA, Naini | 87 | 107 | 110 |
| 9 | Railway Station | 494 | 388 | 584 |
| 10 | Teliyerganj | 16.5 | 13.5 | 20.5 |
| 11 | Zero Road | 1.5 | 1 | 0.5 |

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| | | | | |
|---|---|---|---|---|
| 12 | South Malaka | 35 | 31 | 68.5 |
| 13 | Mevalal bagia, Naini | 13.5 | 3.5 | 9.5 |
| 14 | Daud nagar, Naini | 8 | 1 | 7.5 |
| 15 | PAC Colony, Naini | 19 | 10 | 3 |
| 16 | Laxmi talkies, Katra | 46.5 | 9 | 21 |
| 17 | Muirabad | 25 | 2.5 | 58 |
| 18 | Bus station, Civil lines | 136 | 116 | 120 |
| 19 | Kydganj | 9 | 20 | 5 |
| 20 | Allahpur | 288.5 | 321 | 501 |

TABLE III: Zone-wise distribution of target organisms (in CFU/ml)

| ZONE | AREA COVERED | E. coli ($No.x10^6$) | S. aureus ($No.x10^6$) | Faecal Streptococci ($No.x10^6$) |
|---|---|---|---|---|
| I | Dariyabad, ECC, Kareli, Katghar, Attarsuiya | 108 | 75.8 | 72.9 |
| II | Railway Station, Teliyerganj, zero road, Muirabad, Bus Stand | 134.6 | 104.2 | 156.6 |
| III | University Road, South Malaka, Katra, Kydganj, Allahpur | 88.5 | 87.2 | 123.3 |
| IV | Bethany convent, ADA colony, Mevalal ki Bagiya, chak-Daud Nagar, PAC colony | 28.3 | 29.8 | 27.7 |

TABLE IV: Average Distribution of Target organism throughout Allahabad City (in CFU/ml)

| AREA | E.COLI ($No.x10^6$) | S.AUREUS ($No.x10^6$) | FAECAL STREPTOCOCCI ($No.x10^6$) |
|---|---|---|---|
| ALLAHABAD CITY | 96.05 | 74.25 | 95.125 |

The bacteriological analysis of water ensures the potability of water. According to Indian standard (BIS, 1981) throughout the year 95% of samples should not contain any coliform organisms or should not be detectable in 100 ml of any two consecutive samples and no sample contains *E. coli* in 100ml. The agreeable limit of coliform in water is 10 MPN/100ml (ISI). The result shows that all the water samples in the above four zones, i.e. Zone I, Zone II, Zone III, and Zone IV were contaminated with a high amount of bacterial population than acceptable Indian limit. Zone II and Zone III were found to be highly polluted with faecal contamination and the reason for the high number of bacterial colonies might be due to inadequate maintenance of water reservoirs or the mixing of sewage into the reservoirs and insufficient management of faecal waste by the community and responsible government authorities. The potentiality of drinking water to carry microbial pathogens to large numbers of people, causing subsequent illness, is well reported in countries at all levels of economic development. It has been noted that most sporadic cases of waterborne intestinal ill-

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ness will not be detected, or, if detected, may not be recognized to be water related. Several researchers have attempted to estimate the total burden of waterborne diseases, which might account for one- third of intestinal infections worldwide.

From the figure, it was noted that the contamination from *E. coli* and faecal *Streptococcus* are almost equal (36.187% and 35.839% respectively). Sources of such bacterial contamination include surface runoff, pasture, and other land areas where animal wastes are deposited. Additional sources include seepage or discharge from sewage treatment facilities, septic tanks, and natural plant /soil bacteria. The same results of the high number of total coliforms were observed by different authors in different water bodies in India during pre-monsoon and post-monsoon seasons (Rajurkar et al., 2003; Radha Krishnan et al., 2007). Bacteriological study of water during an epidemic of cholera in Delhi was done by Baveja et al. (1989). Sixty samples from various sources like hand pumps, taps, and tube wells and from different areas of Delhi during epidemics of Cholera and Gastroenteritis were collected which only 27 (45%) of them were found to be suitable for human consumption and the rest indicate the presence of coliform organisms with MPN values from 10 to 1800 + per 100 ml. In a related work in Malawi, Taulo et al (2008), detected 13% faecal coliforms from 60 water source samples; whereas a long time monitoring of 76 well samples showed a contamination of 12% Total coliform and 9% Faecal coliform in Gaza town (Amr and Yassin, 2000, 2008). In Scotland, McDonald et al (2008), reported the Faecal coliform count of 0.57 CFU/100ml from well-protected source water. If we compare the findings of this study with a study conducted in Jimma town in 2005, it showed that 95.8% of samples were unacceptable or extremely contaminated. The finding of this study (100%) was higher. This difference in percentage might be due to variation in methods used in both the studies. The presence of fecal coliforms and E. coli in all of the water sources were demonstrated in this study. Accordingly, the potability and safety of these sources were dubious. As it is shown in a study performed in Lesotho Highlands, adequate protection of water sources could improve the hygienic quality of water sources (Kravitz et al., 1999).

The microbiological quality of supply water is a concern to consumers, water suppliers, regulators, and public health authorities alike. The number of outbreaks that have been reported throughout the world demonstrates that the transmission of pathogens by drinking water remains a significant cause of illness.

A large proportion of the population in developing countries does not have access to sanitary toilets and is thus forced to defecate in the open, leading to contamination of water resources. According to a recent World Bank report, the sanitation coverage in India is only 68%. Though India as an emerging economic superpower in the world, open defecation still remains a major public health concern, with 6% of its gross domestic product (GDP) wasted annually due to lost productivity, healthcare provision, and other consequences of poor sanitation.

Against the studies discussed above, present study analyses the sources of drinking water from supply, among the 20 samples analyzed, sample no. 9 which was collected from Railway station of the city was found to be most contaminated and sample no. 11 which was collected from zero road area had very less contamination comparatively. However, none of the samples complies to the Indian standard IS 1622 (1981). All taken together, the overall picture showed that the supply water sources are not free from bacterial contamination. Water quality is a growing concern, and the availability of safe drinking water is still out of reach for the majority of the people in developing countries.

This research measures only microbial water quality by isolating the mentioned target organisms. As a limitation, the physiochemical analysis was not done due to logistics limitation. However, I believe that the information obtained about contamination of the water sources in Allahabad city is the first in its kind and revealed the hygienic condition of water sources which are used by the community.

## IV. CONCLUSION

In conclusion, the control of human activities to prevent sewage from entering water body is the keys to the avoiding bacteria contamination of drinking water. It is evident that water-borne diseases are due to improper disposal of refuse, contamination of water by sewage, surface runoff, therefore programmes must be organized to educate the general population on the proper disposal of refuse, treatment of sewage and the need to purify our water to make it suitable for drinking because the associable organisms are of public health significance being involved in one form of infection or the other. Simultaneously, awareness among the people towards sanitation and hygienic conditions for storage of drinking water is needed to encourage the use of contamination-free water.

In the contrast of study, it is also recommended to the municipality to ensure the regular maintenance of water supply pipelines, monitoring of leakage of sewage lines to avoid faecal contamination and related disease in communities. Proper storage and disinfection methods should be applied along with the usage of flocculants such as sodium aluminate & sodium silicate to eliminate the suspended particles and microbes during the water treatment process.

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## V. ACKNOWLEDGEMENT

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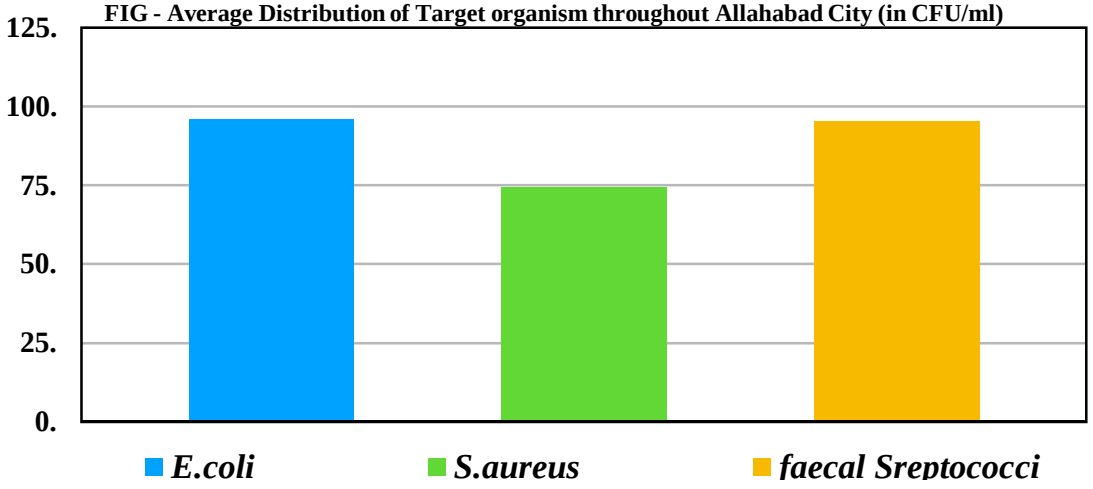
**FIG - Average Distribution of Target organism throughout Allahabad City (in CFU/ml)**

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