

Microbiological Analysis of Drinking Water Sources in Chilakala Gedda Panchayt Ananthagri Mandal, Visakha district, Andhra Pradesh, India

CH. SHANTHI DEVI

Department of Microbiology, Visakha Women's Degree College
Visakhapatnam, Andhra Pradesh, India

M. V. MANIVARMA

School of Distance Education, Andhra University, Visakhapatnam

Abstract:

The quality of potable water and treatment of waterborne diseases are critical public health issues. Microbial contamination of drinking water sources in the most common health risk. The research determines microbiological quality of drinking water sources in Chilakala gedda panchayat, Ananthagiri mandal, Visakhapatnam, India during the period of 2011-2012. A Total of 10 drinking water samples were randomly collected from Bore well, Well, Tap and stream in different places of Chilakala Gedda Panchayat. The water samples were tested using the MPN multiple tube technique on Macconkey broth for Presumptive coliform count followed by Escherichia coli confirmation, Total Plate count and Coliform count were conducted (APHA, 2005). According to the results the MPN count high in well water sample 2400/100ml and Total plate count range between 1.28×10^2 to 3.85×10^2 cfu / 100ml and Fecal coliform count 0.25 to 1.35×10^2 cfu/100ml observed in the study area. The isolates were characterized and identified as E.coli, Enterobacter, Klebsiella, Salmonella and Shigella. The result indicates that microbial values are high according to WHO guidelines. The use of contaminated water in drinking can exposes human body to many water borne diseases hence water treatment and improving quality of water before drinking is required.

Key words: Drinking water, MPN, Coliform count, WHO

INTRODUCTION

Water is one of the most important elements for all forms of life. It is indispensable in the maintenance of life on earth. It is also essential for the composition and renewal of cells. Despite of this, human beings are contaminating tap water sources resulting in provoking water related illnesses [1].

The public health significance of water quality cannot be over emphasized. Many infectious diseases are transmitted by water through the fecal-oral route. Diseases contracted through drinking water kill about 5 million children annually and make 1/6th of the world population sick [2]. Water is vital to our existence in life and its importance in our daily life makes it imperative that thorough microbiological and physico-chemical examinations be conducted on water. Potable water is the water that is free from disease producing microorganisms and chemical substances that are dangerous to health [3]. In India, majority of the rural populace do not have access to potable water and therefore, depend on well, stream and river water for domestic use. The bacterial qualities of ground water, pipe borne water and other natural water supplies in India, have been reported to be unsatisfactory, with coliform counts far exceeding the level recommendation elucidation of important parameters in water quality assessment may be attributed to the fact that in the overall potability of water such parameters should not be ignored [4].

The aim of the study was to determine the microbiological contamination of drinking water sources in Chilakala Gedda panchayat Ananthagiri Visakhapatnam district, A.P. India and compare with studies of BIS and WHO.

MATERIAL AND METHODS:

A survey was conducted for water samples at Chilakala Gedda village of Visakhapatnam district; Andhra Pradesh, India for this water samples were collected every fortnight intervals and method of sample collection and water analysis as follows.

1. Sample Collection:

Sampling was done according to the procedure recommended by American Public Health Association [5]. Water samples were collected for Bacteriological analysis. Samples were collected for container, which is immediately covered tightly after collection of water samples and transported to the laboratory. In Chilakala gedda panchayat, Bore well, well, tap are present as sources of water in the villages and eight samples were collected that are given in the table number 1.



Figure A. Water in the open well



Figure B. A women collecting from the Bore well



Figure C. Tap water

Table 1. Sampling stations

Name of the Village	Source of water	Sampling Code
Chilakala Geeda	Bore	S1
Chilakala Geeda	Tap	S2
Dasavthota	Bore	S3
Dabbala padu	Bore	S4
Venkayya palem	Bore	S5
Vara goddu	Bore	S6
Julaga padu	Well	S7
Settayyathota	Well	S8

2. Microbiological Analysis:

The most probable number (MPN) technique was used to determine the fecal coliform counts of the water samples. This involved the presumptive test using lactose broth and Total coliforms were test using eosin methylene blue (EMB) agar. The total plate count was conducted by pour plate technique on plate count agar (PCA) and counting the colonies developed after the incubation at 37°C for 24 for 24 hours [6]. All colonies with different characteristics on Endo agar, Manital salt agar, SS agar, MacConkey agar, Thiosulphate Citrate Bile salt sucrose agar (TCBS) were sub-cultured onto Nutrient agar (NA) for purification. Enteric bacteria isolated on respective selective or differential media were identified on the basis of their colonial, morphological and Biochemical properties following Bergey's Manual of determinative Bacteriology, 1994.

RESULTS AND DISCUSSION:

A total of eight drinking water samples were collected from different villages of Chilakala gedda Panchatyat, of which 5 were Bore wells, 2 from wells, 1 from tap waters. The test values were then compared with the standard methods for the examination of water [6] and Bureau of Indian Standards (BIS), World Health Organization (WHO).

Table 2. Water analysis of different samples from villages of Chilakala gedda panchayat

Name of the sample water	MPN count/ 100ml	Total Plate Count cfu/100ml	Fecal coliforms cfu /100ml
Bore-1	210	2.51×10^2	0.56×10^2
Tap	64	1.28×10^2	0.25×10^2
Bore-2	150	1.99×10^2	0.52×10^2
Bore-3	120	1.92×10^2	0.55×10^2
Bore-4	93	1.75×10^2	0.48×10^2
Bore-5	240	2.85×10^2	0.61×10^2
Well-1	2400	3.85×10^2	1.35×10^2
Well-2	1100	3.51×10^2	1.28×10^2

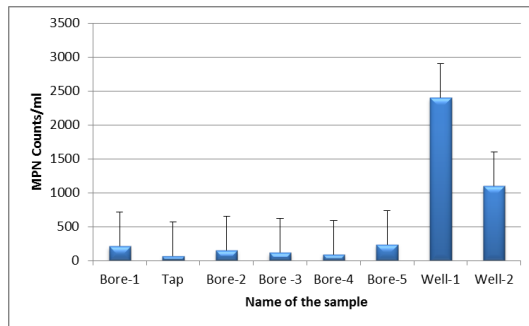


Figure 1. MPN counts of fecal coliform in different water samples

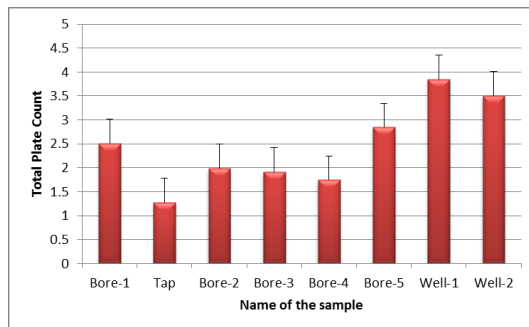


Figure 2. Total bacterial counts from different water samples

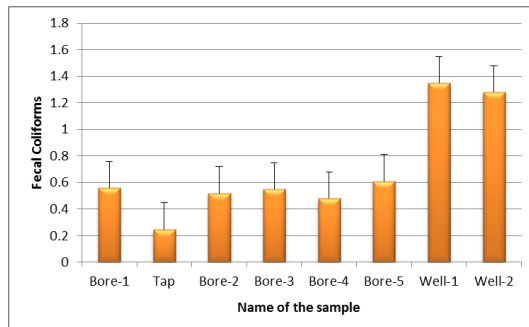


Figure 3. Fecal coliform contents from different water samples

Presence of Coliform group of microbes as a whole is recognized as a suitable indicator for drinking water contamination. Total Coliform counts in Well waters were found to be maximum at 2400 MPN/ 100ml in the well-1 sample (Table 2 and Figure 1). The minimum was in the range 64 MPN/100ml in tap water. Compared with the different Well samples from the above observation that the open well waters had high load of coliforms when compared with that of bore well waters and tap water. It might be due to per collation of water from sanitary land filled areas and leachates from septic tanks (Table-1). The present findings on par with the observation of [7], who have reported high load of coliform counts for open well water. Presence of TC in different drinking water sources indicates inadequate treatment and poor sanitation [8]. Adequate treatment and sanitation is necessary for drinking water.

The total bacterial counts (TVC) for all the water samples were generally high exceeding the limit of 1.0×10^2 cfu/ml which is the standard limit of heterotrophic count for drinking water [9]. A high total heterotrophic count is indicative of the presence of high organic and dissolved salts in the water. The Total Plate Count ranged between 1.28×10^2 to 3.85×10^2 cfu / 100ml (Table 2 and Figure 2).

The Fecal coliform count in any region should be below 10 CFU to consider the water to be safe. The results seen in the

table indicates highly unsafe drinking water, inadequate water treatment, seepage of industrial and domestic pollutants and an overall lacking of infrastructure. The highest counts noticed in S7 open well (well-1) waters were staggeringly high (Table 2 and Figure 3).

In the study area different bacterial species were identified based upon the morphological characteristics of isolates which are obtained from the water samples on Nutrient Agar (NA) and different selective media as shown in table 3. The biochemical characteristics of the isolates which are obtained from these water samples were shown in Table. The isolated enteric bacterial species were identified to be the same as those commonly encountered in water which were also reported in study on river water sources of rural Venda region, South Africa [10] and reviewed by [11].

Table 3. Morphological characteristics of isolates

Isolate	Morphological Characteristics	Organism
1	Non- spore forming and non- motile, gram positive cocci, circular, low convex with entire margin, smooth, medium, opaque colony on Nutrient Agar, Yellow colure colonies on Mannitol Salt Agra Media grown at pH 7 and 37 ^o C	<i>Staphlococcus sp.</i>
2	Gram positive cocci, thin, even, growth on Nutrient Agar, black or brown colure colonies on Bile esilin Agar.	<i>Group D Streptococcus,</i>
3	Gram positive rod, spore forming, abundant, opapue, white waxy growth on Nutrient Agar .	<i>Bacillus sp.</i>
4	Gram negative rod, circular, low convex, with entire margin, mucoid, opaque, growth on Nutrient Agar, green metallic sheen colony on Eosin Methlene Blue (EMB) Agar.	<i>E. coil</i>
5	Gram negative rod, Slimy, white somewhat translucent, raised growth on Nutrient Agar, Dark pink colure colonies on MacConkey Agar.	<i>Klebsiella sp</i>
6	Gram negative rod, thin, blue gray, spreading growth on Nutrient Agar.	<i>Proteus sp.,</i>
7	Gram negative rod, abundant, thin, white medium turns green on Nutrient Agar. pink Colure colonies on Phenothalin diphospate Agar.	<i>Pseudomonas sp.,</i>
8	Gram negative curved rod, abundant, thick, mucous white colure colonies on Nutrient Agar. Yellow colure colonies on TCBS agar	<i>Vibrio cholera</i>
9	Gram negative curved rod abundant, thick, mucous white colure colonies on Nutrient Agar. Green colure colonies on TCBS agar	<i>Vibrio parahaemolytics</i>
10	Gram negative rod, thin even grayish growth on Nutrient	<i>Salmonella sp.,</i>

	Agar	
11	Gram negative rod, thin even grayish growth on Nutrient Agar	<i>Shigella</i>
12	Gram negative rod, abundant thick, white glistening growth on Nutrient Agar	<i>Enterobacter aerogenes</i>

Table 4. Biochemical Characteristics of isolates

Test	W1	W2	W3	W4	W5	W6	W7	W8	W9	W10	W11	W12
Catalase	+	-	+	+	+	+	+	+	+	+	-	-
Oxidase	-	-	-	-	-	-	+	+	-	-	-	-
Motility	-	-	+	+	-	-	+	+	-	-	+	-
Indole	-	-	-	+	-	+	-	+	-	+	-	-
Methyl-red	-	+	-	+	-	+	-	-	+	+	(+)	-
Voge-Proskauer	+	-	+	-	+	-	-	+	-	-	+	-
Citrate Utilization	-	-	-	-	+	-	+	+	+	-	+	+
Urease	+	-	-	-	+	+	-	-	-	-	+	+
Hydrogen sulphide	-	-	+	-	-	+	-	-	+	-	-	-
Starch hydrolysis	-	-	+	-	-	-	-	-	-	-	-	-
Nitrate Utilization	-	-	+	+	+	+	+	+	+	+	+	+
Gelatin liquefaction	-	-	+	-	-	+	-	+	-	-	(+)	+
Lactose fermentation	-	A	-	AG	AG	-	-	AG	-	-	AG	-
Glucose fermentation	A	A	A	AG	AG	AG	-	AG	AG	A	AG	-
Sucrose fermentation	A	A	A	A(+)	AG	AG+	-	AG	AG	A+	-	-

W1-Staphylococcus, W2-Streptococcus, W3- Bacillus Sp., W4- E. coil, W5- Klebsiella Sp, W6-, Proteus Sp., W7- Pseudomonas sp., W8- Vibrio sp., W9- Salmonella sp., W10- Shigella, W11- Enterobacter A-Acid production only; AG - Acid and gas production; +- = Variable reaction; + - Positive; - = Negative ;(+)- Late Positive

Other bacteria isolated from all water samples such as *Staphylococcus*, *Pseudomonas sp.*, *Enterobacter aerogenes*, *Micrococcus sp.*, and *Proteus sp.*, also have public health significance. *Staphylococcus aureus* is known to produce enterotoxin. *Proteus Sp.*, belong to the intestinal flora but is also widely distributed in soil and water [12]. *Enterobacter aerogenes* isolated from water samples is an example of non-

fecal coliforms and can be found in vegetation and soil by which the pathogens enter the water [3].

The presence of opportunistic *Pseudomonas* in the water carries the potential for problems in an immuno compromised population. Shallow groundwater samples commonly contain *Pseudomonas sp.*, *Bacillus sp.*, which occur in both soil and fecal material, and may not be indicative of livestock manure [13].

The presence of *Salmonella* in water samples indicates that the public water supply system is poor and chances of outbreak of water-borne *Salmonella* infection is higher among people consuming the water without proper disinfection. Occurrence of *Salmonella*, *Shigella*, and *Vibrio* in urban water supply. Water distribution systems have been reported to provide unique condition for the development of biofilm [14] water borne pathogens was found only in those samples which were positive for *coliforms*. Similar result was reported by [15].

CONCLUSION:

This study concluded that water quality distributed at tribal area need more effort in limiting the number of microbial organisms present in drinking water sources. The majority of the water sources had unacceptable total coliform count and all the water sources which were positive for presumptive coliform count had *E.coli* showing fecal contamination of water sources, and the present study revealed and recommend regular disinfection of drinking water sources, periodic bacteriological appraisal of drinking water sources, and construction and distribution of piped water, this research also demonstrated the importance of education for the people who use drinking water. Much needs to be done to increase awareness of the hazard of drinking.

REFERENCES

1. WHO, 2008, Guidelines for Drinking water Quality, 3rd edition, Volume 1, Geneva, World Health Organization.
2. WHO, 2004. Guidelines for Drinking –water Quality, 2nd edition. Volume 1, World Health Organization, Geneva, 231-233.
3. Shittu, O.B., J.O. Olaitan and T.S. Amusa, 2008 Physico-chemical and bacteriological analyses of water used for drinking and swimming purposes in Abeokuta, Nigeria. Afr J Biomed Res, 11:285–90.
4. Osuinde, M.I. and N.R. Eneuzie, 1999. Bacteriological analysis of ground water. Nigeria Journal of Microbiology, 13:47-54
5. APHA, 1992. Standard Methods for the Examination of Water and Wastewater, 18th Edition, Washington, D.C. Article URL: http://www.webmedcentral.com/article_view/2084.
6. APHA, 1998. Standards Methods for the Examination of Water and Wastewater. 20th edition, American Public Health Association, Washington, D.C.
7. Kannan, K., J.C. Franson, W.W. Bowerman, K.J. Hansen, P.D. Jones and J.P. Giesy, 2001. Perfluorooctane sulfonate in fish eating water birds including bald eagles and albatrosses. Environmental Science and Technology, 35: 3065-3070.
8. WHO, 1985. Health Hazards from Nitrate in Drinking water. Report on a WHO meeting, Environmental Health Series, No. 1, Copenhagen, 5-9 March 1984. WHO Regional Office for Europe, Copenhagen
9. EPA, 2002. US Environment Protection Agency, Safe Drinking Water Act Ammendment [http:// www.epa. gov/safe water /mcl. Html](http://www.epa.gov/safe_water/mcl.html).
10. Obi, C.L., N. Potgieter, P.O. Bessong and G. Matsaung, 2002. Assessment of the microbial quality of river water sources

in rural Venda communities in South Africa. *Water SA*, 28 (3): 287-291.

11. Okonko, I.O., O.D. Adejoje, T.A. Ogunnusi, E. Fajobi, and O.B. Shittu, 2008. Microbiological and physicochemical analysis of different water samples used for domestic purposes in Abeokuta and Ojota, Lagos Nigeria. *African Journal of Biotechnology*, 7(5) 617-6721.

12. Schlegel, H.G. 2003. *General Microbiology*. 7th. ed. Cambridge. University Press. 480p.

13. Aydin, A. 2007. The Microbiological and Physico-Chemical Quality of Groundwater in West Thrace, Turkey *Polish J. of Environ. Stud*, 16 (3): 377-383

14. Scher, K., U. Romling and S. Yaron, 2005. Effect of heat, acidification and chlorination on *Salmonella enterica serovar Typhimurium* cells in a biofilm formed at the air-liquid interface. *Applied Environmental Microbiology*, 71: 1163–1168.

15. Bhatta, D.R., A. Bangtrakulnonth, P. Tishyadhigama, S.D. Saroj, J.R. Bandekar, R.S. Hendriksen and B.P. Kapadnis, 2007. Serotyping, PCR, phage typing and antibiotic sensitivity testing of *Salmonella serovars* isolated from urban drinking water supply systems of Nepal. *Letters in Applied Microbiology*, 44(6): 588–594.