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Bacteriological Study of Different Water Sources during Outbreaks of Diarrhoea in Ganjam District, Odisha

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Abstract

Background: Water for human consumption must be free from chemical substances and pathogenic microorganisms. Infectious diseases are transmitted primarily through consuming water contaminated with human and animal excreta. Provision of good quality household drinking water is an important way of uplifting the public health.

Objectives: To investigate the bacteriological contamination of different water sources during outbreaks of diarrhoea in rural and urban areas of Ganjam district.

Methods: Over a period of 3 years a total of 433 numbers water samples were analysed from different water sources. These were tested using commercially available K056 Hi Water testing kit and Multiple tube fermentation method for presumptive coliform count followed by Eijkman test for confirmation of *Escherichia coli*

Results: Out of 433 water samples received during outbreaks 265 (61.2%) water samples found to be not potable. The numbers of samples received were more during pre monsoon and monsoon period.

Conclusion: The current study showed maximum number of water sources not potable for human consumption which led to outbreaks of diarrhoea. There is a necessity of proper sanitation and disinfection measures of the sources of water supply prior to human consumption.

Keywords: Bacteriological study, water contamination, most probable number (MPN), coliforms**.**

Key message: Water borne disease like diarrhoea, cholera occur every year during summer and rainy seasons in india due to poor quality drinking water supply and sanitation .Water sources should be protected from contamination and routine microbiological analysis of drinking water should be carried out. Timely disinfection shouLd be done to prevent outbreaks of diarrhoea.

Introduction

Diarrhoea is one of the major public health problems in developing countries. Access to safe drinking water still remains a challenge in this century. It was observed by the United Nations that annually more than one lakh people die due to water borne diseases in India. Most of the water supply including the ground water is not fit for drinking purpose. As per UN reports the water quality in India is poor and ranks 120th out of 122 nations.[1] Approximately 65% of rural and 36% of urban India do not have proper access to safe drinking water as reported by WHO.[2]

In this region of Odisha in summer there is scarcity of water as most of the streams, ponds, wells and rivers dry up and the people have to depend on the limited water sources which are consumed by both humans and animals. Even in the rainy season people depend on non-public water supply and consume the contaminated water from different sources. Most of the water bodies and river

waters are contaminated due to dumping of waste, open defecation adjacent to water sources and discharging of industrial waste or sewage into them. The people affected by diarrhoea are usually those not having proper hygiene or sanitation and are usually of low socioeconomic status. Most of the cases in developing countries involve children less than 5 years.[3]

Nearly 1.1 billion people in the world consume unsafe water and this also attributes to 88% of diarrhoeal diseases in the world and about 1.7 million deaths annually as per World Health Organisation estimates.[4]

John Snow with his pioneering work in epidemiology of cholera has shown that water contaminated with pathogens can cause waterborne diseases like diarrhoea.[4] The pathogens that can contaminate drinking water are human and animal excreta (faeces). Bacteriological parameters, especially *Escherichia coli* and total coliform count have been used to detect faecal contamination of water. Drinking water samples obtained from different sources from areas of outbreak were analysed for bacteriological contamination.

Materials and Methods

A total of 433 water samples were collected from various water sources like ponds, tube wells, bore wells, dug wells, streams, rivers, over head tanks, hand dug wells in dry river beds in summer (*chua*) and municipality supply during periods of outbreak of diarrhoeal diseases from rural and urban areas in the district of Ganjam, Odisha during the period from April 2014 to March 2017.

Sample Collection – About 250 ml of the water sample which is to be tested was collected in a clean sterile bottle as per the guidelines and transported to the laboratory for bacteriological analysis. Into each bottle 0.25ml of fresh 1.8% aqueous solution of Sodium thiosulphate was added prior to sterilisation. In case of delay for more than 3 hours the water samples were transported in ice box.[5]

Bacteriological Analysis – All the water samples were subjected to bacteriological testing by two methods – commercially available K056 Hi Water testing kit (HiMedia laboratories, India) for primary detection of *Escherichia coli*, *Salmonella*, and *Citrobacter* based on H_2S production and detection of total and fecal coliforms using MPN(Most Probable Number) technique.

1. **K056 Hi WaterTesting Kit** : Water was added to the vial up to the arrow level mark and gently shaken to allow the medium from the rolled strip inside the bottle to be released. The vials were incubated at 37°C for 24-48 hours. If the colour of the water turns black the water was considered not fit for drinking (as per the manufacturer's guidelines K056 Hi WaterTesting Kit -Hi media,Mumbai,India)

2. **MPN method**: Measured volumes of water are added to a series of tubes/bottles containing liquid indicator growth medium. From the number of positive and negative reactions the most probable number (MPN) of indicator organisms in the water sample was estimated using reference statistical tables.[5]

Presumptive Coliform Count

Fifty millilitres of double strength MacConkey broth was added to a blood culture bottle and 10 ml into 5 test tubes. Into another 5 test tubes 5 ml of single strength MacConkey broth was added. Durham tubes were placed in all the tubes and sterilised. Then 50ml of the water sample was added to the bottle containing 50 ml of double strength MacConkey broth, 10 ml of water was added to the tubes containing 10 ml of double strength MacConkey broth and 1ml of water was added to the tubes containing 5ml of single strength MacConkey broth. All the inoculated media were incubated at 37°C aerobically. After 24-48 hours of incubation the bottles and tubes showing change of colour and presence of gas in Durham tube were considered presumptive positive for coliform

bacilli (*Escherichi coli, Klebsiella spp or Citrobacter spp*) and noted. Cultures showing no acid or gas production after 48 hours of incubation were considered negative. The MPN of coliform in 100 ml of water was calculated referring to Macrady table.[5]

Confirmed Test

Confirmation of presence of true (faecal) coliform bacilli showing positive presumptive test is done by Eijkman test. This test is done by incubating subcultures from the positive presumptive tests at 44° C and 37°C in a Brilliant -green bile lactose broth .The presence of coliform bacilli is confirmed by the production of gas from lactose at 37°C .The presence of Escherichia coli is confirmed by the production of gas from lactose and indole from tryptophan at 44°C .[5]

Results & Discussion

Fig-1 Seasonal distribution of water samples received during outbreaks of diarrhoea

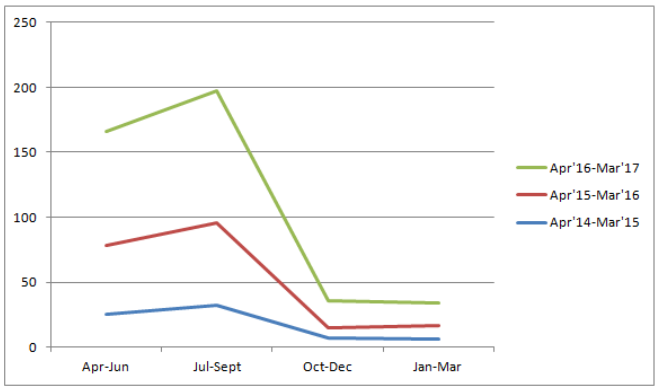

Table-1: Year wise comparision of various water sources by Hi WaterTesting Kit

Type of water source	Apr-2014-Mar-15		Apr-15-Mar-16		Apr-16-mar17	
	Total tested	Not potable	Total tested	Not potable	Total tested	Not potable
Dug well	21	17	33	16	64	30
Bore well	5	1	10	3	27	13
Tube well	13	6	54	31	61	27
Municipality supply water	15	14	27	22	57	45
Over head storage/domestic	6	4	4	3	8	6
Pond	4	4	4	4	5	5
River/Spring water	6	6	3	2	3	3
Hand dug wells in dry river beds in summer(chua)	0		1	1	2	2
Total	70	52	136	82	227	131

Table-2: Year wise comparison of water samples by Hi Water Testing Kit and MPN method

Year	Samples tested	Not potable by Hi Water Testing Kit	Not potable by MPN method
2014-15	70	52(74.3%)	50
2015-16	136	82(60.3%)	78
2016-17	227	131(57.7%)	123
Total	433	265(61.2%)	251

Fig-2 .Detection and confirmation of *Escherichia coli* by Eijkmen test

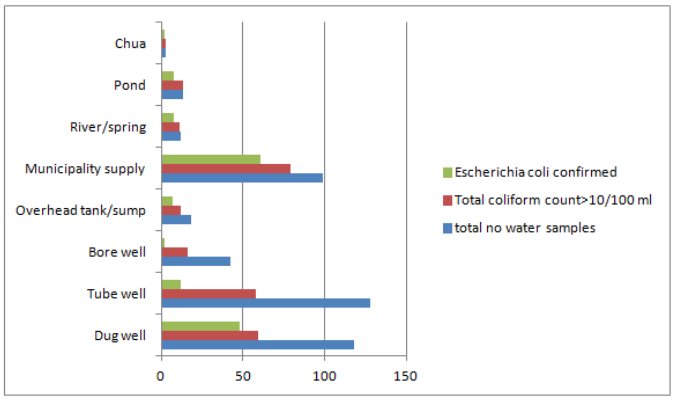

Table-3 : Total number of Escherichia coli isolated from different water sources

Source of water	Total number tested	Total coliform count >10/100 ml by MPN method	*Esch coli* confirmed by Eijkmen test
Dug well	118	59	48
Tube well	128	58	12
Bore well	42	16	2
Over head tank/sumps	18	12	7
Muncipality supply	99	79	61
River/spring	12	11	8
Pond	13	13	8
Chua	3	3	2
Total	433	251	148

A total of 433 water samples from various sources were received during outbreaks of diarrhoea from April 2014 to March 2017. Out of which 118 samples were from dug wells, 128 from tube wells, 42 from bore wells, 18 from over head tank & sumps,99 from municipality supply,12 from river&spring,13 from ponds and 3 no were from chua. In the year 2014-15 seventy water samples were received, 2015-16 a total of 136 and during 2016-17 two hundred twenty seven water samples received. Water samples were received throughout the year. But during the months from July to September in all the three years, there were more number of samples received showing a seasonal pattern.(**Figure 1**) A study by Diwakar et.al showed more contamination of drinking water in the pre-monsooon season.[6]

By Hi water testing kit 265 out of 433 water samples were unsuitable for drinking purpose. Among the different water sources received pond and chua water were 100% contaminated followed by river and spring water and municipality water supply (**Table-1**).By multiple tube fermentation method 251 water samples showed total coliform count (MPN) of >10/100 ml of water. In comparison to MPN (57.9%)method Hi Water testing kit (61.2%)showed a higher value of detection of coliforms in water samples.(**Table 2**). Out of the 433 water samples, *Escherichia coli* were detected in 148(34.2%) samples by Eijkman test.(**Figure 2, Table 3**). Kumar et al in their study also found a very high MPN from municipality tap water.[7]

As most of the people in this district have poor hygiene practices and outdoor defecation is still prevalent, the water sources usually become contaminated by rain water draining these areas. There is also flooding of the drains and as in some areas the sewage pipes and water supply pipes are in close proximity leakage may cause contamination of supply water. During summer there is scarcity of water as most of the streams, wells and ponds dry up and people have to depend on the available sources which are consumed both by humans and animals. One of the major public health problems in this region is diarrhoea in the summer &rainy season; especially children less than five years of age are affected most.

Conclusion

One of the major challenges of the 21st century is to provide safe and clean drinking water with routine monitoring of bacteriological quality.[8] Water sources should be protected from contamination with human and animal excreta and timely disinfection especially during summer and rainy season to prevent outbreaks of diarrhoea. Strict control measures should be taken to reduce the contamination of drinking water. Integrated approach of improvement in water quality supply combined with importance of proper sanitation and hygiene education definitely help to solve this problem.

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