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The bacteriological quality of drinking water in Golaghat Sub-division of Golaghat District, Assam, India

Madhab Borah¹, Joydev Dutta^{2*}, Abani Kumar Misra¹

¹Department of Chemistry, Gauhati University, Guwahati, Assam, India, 781 014 ^{2*} Department of Chemistry, Chaiduar College, Gohpur, Sonitpur, Assam, India, 784168

*Corres.author : joydevghp@rediffmail.com, Tel. +91(9435383570), Tel. + 91(9864507603)

Abstract: The drinking water quality with respect to bacteriological examination by quantitative determination of total coliform and fecal coliform count (MPN) and presence or absence for E.coli were done for 32 numbers of drinking water (well, T.E. supply, pond and tube well) samples from tea estate areas where cases of dysentery and diarrhea were found to be maximum. Standard methods were used for analysis of total coliform and fecal Coliform bacteria. The number of total coliform and fecal coliform bacteria was determined on Mac Conkey broth by using most probable number method (MPN), presence of E.coli was determined using EMB Agar¹. The water samples were taken from Sept 2007 to Nov 2007, examined and found bacterial levels were failed to meet water quality standards. Coliform contamination far exceeds the WHO standards in most cases, the highest number of total coliform bacteria was found on 30th Nov,2009. After that date the number of total coliform and fecal coliform decreased. Water samples contained varying levels of fecal coliform bacteria² ranging from a Most Probable Number (MPN) of 10 to 2.8 x 10³ cfu/100mlThe presence absence test for E-coli was also done for same samples. 78.1% of samples have E-coli contaminated.. The required data are found at Joint Director of Health services, Golaghat district, Assam, India.

Keywords: Bacteriological quality, drinking water, tea estates, diarrhea, Golaghat Subdivision.

Introduction

When we are sick and take antibiotics, the goal is to kill the "bad" bacteria that have caused the disease. With no bacteria, the planet would be covered with dead plants and animals. They make it possible for ruminant animals (cows, sheep, goats) to digest plant cellulose and for some plants, (soybean, peas, alfalfa) to convert nitrogen to a more usable form¹. Again, drinking water may be contaminated by the bad bacteria resulting health problems. The World Health Organization (WHO) reported that nearly half of the population in developing countries suffers from health problems associated with lack of drinking water or water². microbiologically with contaminated Groundwater is an important source of drinking water and its quality is currently threatened by the

combination of chemical pollution and microbiological contamination, especially microbes of sewage origin³. High incidence of diarrhea, helmenthiasis, trachoma and the overall high mortality rates are associated with poor environmental sanitation^{4,5}. Sanitation with good hygiene, acts as a fundamental 'Primary barrier' by ensuring that fecal matter is disposed of safely, and does not spread in the environment. Once in the environment, however, here are many ways in which infected fecal matter can be spread. Safe water supply can support a number of can act as 'secondary barrier', which prevents further spread of contamination and infection to new hosts^{5,6} Currently, about 20% of the world's population lacks access to safe drinking water. and more than 5 million people die annually from illness associated with safe drinking water or inadequate sanitation¹. The World Health Organization estimated that up to 80% of all sicknesses and diseases in the world is caused by inadequate sanitation, polluted water or unavailability of water'. Approximately three out of five persons in developing countries do not have access of safe drinking water and only about one in four has any kind of sanitary facilities. The World Health Organization (WHO) reported that nearly half of the population in developing countries suffers from health problems associated with lack of drinking water or with microbiologically contaminated water. In Golaghat district, rural and urban population receives potable water from a variety of sources, including groundwater, through the different supply systems, or independently processed and maintained water wells. Thus, the potential risk from water contamination varies widely.

The U.S. Environmental Protection Agency (EPA) requires all drinking water systems to monitor for total coliforms in distribution systems. The EPA states that no more than 5.0% of samples can test positive for total coliform in a month. (For water systems that collect fewer than 40 routine samples per month, no more than one sample can be total coliform-positive). Coliform bacteria are the standard used for bacterial quality in drinking water. Coliform bacteria is not a single bacteria species, rather it is a grouping of several different bacterial species. The presence of water sources indicates that coliform bacteria in sewage or some type of surface water is entering and contaminating the water supply. Along with coliform bacteria, other disease causing organisms may be present, and these can cause diseases such as dysentery, typhoid and hepatitis. A water source contaminated with coliform bacteria requires immediate attention. Coliform bacteria are often used as indicator of sanitary quality of foods and water. Coliform bacteria are defined as rod-shaped Gram negative organisms which ferment lactose with the production of gas when incubated⁸ at 35° C. These organisms are normally found in the aquatic

environment and on vegetation. The presence of coliform bacteria in drinking water indicates that the water was not properly treated to eliminate pathogens, or that it got contaminated somewhere in the distribution system. *Escherichia coli*, a member of the coliform group can ferment lactose at 44° C as well. The origin of *Escherichia coli* is almost exclusively of fecal origin, thus, if it is found in water or food, it indicates fecal contamination, and an imminent health danger, as other fecal pathogens such as viruses or parasites may be also present. The majority of test for bacteria depend on using three indicator bacterial types. They are the total coliform group, the fecal coliform group, and *E. coli*.

Total coliforms include bacteria that are found in the soil, in water that has been influenced by surface water, and in human or animal waste.

Fecal coliforms are the group of the total coliforms that are considered to be present specifically in the gut and feces of warm-blooded animals. Because the origins of fecal coliforms are more specific than the origins of the more general total coliform group of bacteria, fecal coliforms are considered a more accurate indication of animal or human waste than the total coliforms.

Escherichia coli (E. coli) is the major species in the fecal coliform group. Of the five general groups of bacteria that comprise the total coliforms, only E. coli is generally not found growing and reproducing in the environment. Consequently, E. coli is considered to be the species of coliform bacteria that is the best indicator of fecal pollution and the possible presence of pathogens. *E. coli*, is becoming more well known in the public lexicon for "bad bacteria". (E. coli is the popular way of referring to the common bacterium, *Escherichia coli*.) The irony is that *E. coli* itself is not toxic – it is one of the required inhabitants of man and beast, as noted above – *E. coli* is simply an "indicator" of the possible presence of other, toxic, bacteria.

Present work represents the total coliform bacteria(CFU/100ml), most probable number of fecal coliform bacteria and presence absence test for E-coli⁹.

Table I.: Guideline value/ permissible limits for Microbiological quality parameters in drinking water (For untreated water entering distribution system) :

(I OI anticated mater e	(i of undered water entering distribution system) :							
Parameters	Unit	WHO	Guideline	ISI tolerance limit	EPA standards			
		value		desirable value				
Coliform organisms	MPN/100 ml	0		10	0			
Fecal coliform	MPN/100 ml	0		0	0			
E-coli	MPN/100 ml	0		0	0			

Present work also represents the New diarrheal disease, New cumulative diarrheal disease, and total diarrheal death of Golaghat District(Tea Estate area) from October to November 2007 by the general survey at these places and the data found at Joint Director of Health services, Golaghat district.

Experimental

The study was conducted in Golaghat sub-division of Golaghat district, from September to November 2007.A total of 32 water samples were taken for bacteriological analysis. The sampling strategy was based on capturing all types of water sources used by the community. The method of sample collection at each source was according to the WHO Guidelines for Drinking water quality assessment¹⁰. Five hundred ml of water sample from each source was collected, labeled and kept in icebox during transportation and analyzed in the laboratory. Samples were analyzed using standardized bacteriological methods for water quality analysis¹¹ to determine the degree of contamination. All Samples were analyzed for total bacterial count, fecal coliform and E. Coli in the Laboratory of botany department, Gauhati university. Water samples were processed in accordance to Standard Methods for the Examination of Water and Wastewater¹². Upon sample collection, a small amount of Na₂SO_{3.5}H2O solution (18 mg/L) was added to the test bottles in order to block the continuous disinfectant action of chlorine in water. The samples were kept in a portable refrigerator and were taken to the laboratory (ambient temperature of 25°C) for bacteriological analysis.

The standard plate count method consists of diluting a sample with sterile saline or phosphate buffer diluent until the bacteria are dilute enough to count accurately. That is, the final plates in the series should have between 30 and 300 colonies. Fewer than 30 colonies are not acceptable for statistical reasons (too few may not be representative of the sample), and more than 300 colonies on a plate are likely to produce colonies too close to each other to be distinguished as distinct colony-forming units (CFUs). The assumption is that each viable bacterial cell is separate from all others and will develop into a single discrete colony (CFU). Thus, the number of colonies should give the number of bacteria that can grow under the incubation conditions employed. A wide series of dilutions (e.g., 10^{-4} to 10^{-10}) is normally plated because the exact number of bacteria is usually unknown. Greater accuracy is achieved by plating duplicates or triplicates of each dilution, although we will not be doing that in this exercise.

Materials needed for plate count method: 4 sterile 99-ml saline blanks,1-ml pipettes with pi-pump, 6 petri plates, 6 agar pour tubes of nutrient agar (plate count agar), 48 to 50°C water bath, boiling water bath, Bunsen burner, 6 micro-cuvettes and rack,1 microcuvette holder.

In the MPN method, a "presumptive coliform test" is performed first. The presumptive coliform test is used to detect and estimate coliforms population of a water sample. The test used known as presumptive because of the development of a positive result in a Mac conkey broth, inoculated with water may very occasionally be due to non-coliform organisms. In this test known volumes of water (dilution) are added to lactose fermentation tubes and production of acid and gas from the fermentation of lactose is a positive test of coliform bacteria. The lactose broth used in the test is selective for the isolation of coliform because of the addition of bile and lauryl sulphate or brilliant green(used in present test). A pH indicator is also added to lactose broth for the detection of acid . A statistical method is used to estimate the population of coliforms which means that the result of obtained is expressed as the most probable number (MPN) of coliforms. A count of number of lactose fermentation tubes showing production of gas following the incubation period is taken.

Results and Discussion

The results of bacteriological examination are given on table IV where the water samples were collected from sept 2007 to nov 2007 .From table IV it is observed that the highest most probable number of total coliform count was found 1,11,022 cfu/100 ml in Sockieting T.E(T.E supply water sample). Again highest MPN of fecal coliform count was found 28000 cfu/100 ml in Kakotibari TE and Sockieting TE respectively. It was also found that about 78.1% sample sources were E coli contaminated. The results from present study indicate that about 0.7 % of population of the study area was affected by the diarrheal and dysentery disease. The minimum value of MPN of fecal coliform was found in Radhabari T.E(10 cfu/100 ml). High level of microbial contamination of water supply in tea estates, tube well, well and pond within Tea Estates could be due to water distribution network as well as much body contact with the water. This suggests the reason for high prevalence of water-borne diseases such as Typhoid fever, diarrhea, dysentery and few incidences of cholera on the TE of Golaghat subdivision. Also, inadequate protection for the water source which leads to seepage from sewage lines and other waste disposal facilities into bore holes might be responsible. High coliform counts were the most common reason for the failure of potable water to meet acceptable standards¹³. Relative lower loads of E. coli in water samples serial no 5,6,7,8,11,14,24 from tube wells and well samples (Table IV), may be due to increased depth of these

sources (>100 meters deep) as compared to other sample sources 14 .

All water sources were grossly polluted. The type of coliform exhibited is a fecal type specifically of human origin. The effect, therefore, is attributed to constructional defects, poor sanitation, low level of hygiene education, poor supervision and maintenance and irregular disinfection¹⁵.

Table II & III represents the cases of diarrhea due to contamination of fecal coliform bacteria and e-coli.(The data for diarrheal cases and diarrheal death, were collected from Joint Director of Health services, Golaghat district and distance of various tea estates from HQ of Golaghat district and population of affected area were collected from Office of deputy director, economics and statistics, Golaghat district as per 2001 census). From Table II it is seen that the highest new diarrheal disease was found in the week of D (821) whereas total diarrheal death of Golaghat District(basically Tea Estate area) from October 2007 to November 2007 was 40. Again, total new cumulative diarrheal disease was 5464 ,whereas highest number of diarrheal death was found 5 and 4 in Kakotibari T.E and Sockieting T.E respectively. Figure 1 represents a graph plotted between New diarrheal cases of Golaghat district vs weeks of the month

October 2007 to November 2007 and Figure 2 represents a bar diagram plotted between New diarrheal cases of Golaghat district vs weeks of the month October 2007 to November 2007. From figure 1 and 2, it was found that the highest number of new diarrheal cases was 821 and lowest number of diarrheal cases was found in the week of A(68) i.e. at the initial stage the new cases of diarrheal disease was minimum. From figure 1 &2 it was also found that the number of new diarrheal cases sharply increases with highest value in the week D and sharply decreases until November last week, 2007. Water treatment was done by the various health departments including Joint director of health services, Golaghat during that period. The degree of bacteriological contamination decreases from week D ,the reason may be due to treatment of water done by health departments. It can be observed that maximum number of diarrheal cases(821) was found in the forth week of October 2007. Where maximum number of diarrheal disease was found at Kamarbandha ali. Mean, media, mode, std. deviation , varience shown in the table IV, the positive skewness shows the long right hand tail and all the cases shown the positive kurtosis value. 25%,50% and &75% confidential level shown.

 Table II.: Representation of New diarrheal disease, New cumulative diarrheal disease, and total diarrheal death of Golaghat District(Tea Estate areas) from October 2007 to November 2007.

Weeks	Abbreviation for weeks	New cumulative diarrheal cases	New diarrheal cases	Total cumulative diarrheal (from sept to Nov '07) death
1.10.07 to 7.10.07	А	2323	69	-
8.10.07 to4.10.07	В	2664	341	-
15.10.07 to 21.10.07	С	3367	703	-
22.10.07 to 28.10.07	D	4188	821	-
29.10.07 to 04.11.07	Е	4705	517	-
05.11.07 to 11.11.07	F	4951	246	-
12.11.07 to 18.11.07	G	5183	232	-
19.11.07 to 25.11.07	Н	5386	203	-
26.11.07 to 30 .11.07	Ι	5464	78	40

Sl No.	Affected area/tea estate/village/Block PHE/CHE	Population (As per 2001 census)	Total diar	rheal cases	Total diarrheal death	
			Oct '07	Nov'07	Oct '07	Nov'07
1	Abhoijan TE	549	23	3	1	0
2	Rungajan TE	3475	103	31	1	0
3	Forkating TE	380	50	8	0	0
4	Koomtai TE	2200	86	60	0	0
5	Oating TE	1638	11	2	0	0
6	Rungamatty TE	1292	14	0	0	0
7	Radhabari TE		4	0	0	0
8	Lokhoujan TE		10	0	0	0
9	Borkatonee TE	100	20	134	0	0
10	Basakumargaon area		5	0	1	0
11	Boruah gaon area	966	2	0	0	0
12	Golaghat TE	234	70	89	1	0
13	Goranja TE	281	0	33	0	0
14	Dergaon CHC		54	20	0	0
15	Diffloo TE /area	4131	13	0	0	0
16	Dooria TE	3500	243	21	1	0
17	Doyang TE	2385	36	18	0	0
18	Ghiladhary TE	4500	165	17	0	0
19	Halmira TE		119	1	1	0
20	Kakotibari TE	450	31	0	5	0
21	Mockrong TE	2377	123	5	1	0
22	Woka TE	4361	119	21	1	0
23	Sockieting TE	3300	101	2	4	0
24	Jamuguri TE	2842	99	10	1	0
25	Borjan TE	1500	52	25	0	0
26	Dokhinhengra TE	4500	139	61	0	0
27	Bokakhat PHC	146229	51	39	0	0
28	Charingia PHC	134334	359	288	1	1
29	Kamabandha Ali	306558	972	260	2	0
30	Missamara PHC	95643	71	15	0	0
31	Morongi TE	321676	144	48	2	0
32	KK civil Hospital		494	266	8	0
33	Hautley TE	727	0	39	0	1
34	Letekujan TE	1039	0	13	0	0
35	Majikuchi TE	1794	4	3	0	0
36	Thorajan TE	970	88	5	1	0
37	Missamara TE	3185	16	12	0	0

Table III: Names of affected tea estates(TE), population, total diarrheal cases and total diarrheal Deaths.

_	Population	Direa1	Direa2	Death1	Death2
Mean	34100.5484	105.1622	41.8649	.8649	.0556
Median	2377.0000	52.0000	15.0000	.0000	.0000
Mode	4500.00	.00	.00	.00	.00
Std. Deviation	83694.22054	178.63674	74.59973	1.63575	.23231
Variance	7.005E9	31911.084	5565.120	2.676	.054
Skewness	2.783	3.684	2.569	3.047	4.051
Kurtosis	7.175	15.856	5.824	10.467	15.260
Range	321576.00	972.00	288.00	8.00	1.00
Minimum	100.00	.00	.00	.00	.00
Maximum	321676.00	972.00	288.00	8.00	1.00
Percentiles 25	966.0000	12.0000	2.0000	.0000	.0000
50	2377.0000	52.0000	15.0000	.0000	.0000
75	4361.0000	119.0000	39.0000	1.0000	.0000

Table IV.: Descriptive statistics of population, total diarrheal cases and total diarrheal death.

Table V: Bacteriological examination of 32 numbers of drinking water samples of Tea Estate areas of Golaghat District and their distance from the district HQ are given below.

		Nature of	MPN of total	e-coli	MPN of
Sl No. /	Affected area/tea	sampling source	coliform	present	fecal
sample No	estate/village/Block		cfu / 100 ml	/absent	coliform
sample No	PHE				cfu/
					100 ml
1	Abhoijan TE	well	88,600	Present	2900
2	Rungajan TE	Pond	96,020	Present	3400
3	Forkating TE	Tube well	60,440	Present	2100
4	Koomtai TE	Tube well	70,320	Present	2900
5	Oating TE	Tube well	13,660	Absent	28
6	Rungamatty TE	well	17,280	Absent	39
7	Radhabari TE	Tube well	8,450	Absent	10
8	Lokhoujan TE	Tube well	11,340	Absent	20
9	Borkatonee TE	Tube well	71,220	Present	2100
10	Basakumargaon area	pond	74,440	Present	2900
11	Boruah gaon area	Tube well	16,840	Absent	110
12	Golaghat TE	Tube well	73,320	Present	2900
13	Goranja TE	Tube well	60,480	Present	210
14	Diffloo TE	Tube well	22110	Absent	20
15	Dooria TE	Tube well	88,422	Present	3400
16	Doyang TE	Tube well	72,024	Present	2100
17	Ghiladhary TE	Tube well	70,282	Present	2100
18	Halmira TE	Tube well	86,028	Present	11000
19	Kakotibari TE	T.E. Supply	116088	Present	28000
20	Mockrong TE	Tube well	80,046	Present	3400
21	Woka TE	T.E. Supply	76,0098	Present	3400
22	Sockieting TE	T.E. Supply	1,11,022	Present	28000

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23	Jamuguri TE	Tube well	90,046	Present	1100
24	Borjan TE	Tube well	13,408	Absent	29
25	Dokhinhengra TE	pond	74082	Present	340
26	Kamabandha Ali	Ring well	92,018	Present	2400
27	Morongi TE	Tube well	86,082	Present	1100
28	Hautley TE	Tube well	88,088	Present	1100
29	Letekujan TE	Tube well	60,464	Present	3400
30	Majikuchi TE	Tube well	62,242	Present	2900
31	Thorajan TE	Tube well	64,064	Present	2900
32	Missamara TE	Tube well	66,012	Present	2900

Table VI.:Bacteriological examination of 32 numbers of drinking water samples of Tea Estate areas of Golaghat District and their distance from the district HQ are given below.

		Nature of	MPN of total	e-coli	MPN of
	Affected area/tea	sampling source	coliform	present	fecal
Sl No. /	estate/village/Block	1 0	cfu / 100 ml	/absent	coliform
sample No	PHĚ				cfu/
					100 ml
1	Abhoijan T.E	well	88,600	Present	2900
2	Rungajan T.E	Pond	96,020	Present	3400
3	Forkating T.E	Tube well	60,440	Present	2100
4	Koomtai T.E	Tube well	70,320	Present	2900
5	Oating T.E	Tube well	13,660	Absent	28
6	Rungamatty T.E	well	17,280	Absent	39
7	Radhabari T.E	Tube well	8,450	Absent	10
8	Lokhoujan T.E	Tube well	11,340	Absent	20
9	Borkatonee T.E	Tube well	71,220	Present	2100
10	Basakumargaon area	pond	74,440	Present	2900
11	Boruah gaon area	Tube well	16,840	Absent	110
12	Golaghat TE	Tube well	73,320	Present	2900
13	Goranja T.E	Tube well	60,480	Present	210
14	Diffloo T.E	Tube well	22110	Absent	20
15	Dooria T.E	Tube well	88,422	Present	3400
16	Doyang T.E	Tube well	72,024	Present	2100
17	Ghiladhary T.E	Tube well	70,282	Present	2100
18	Halmira T.E	Tube well	86,028	Present	11000
19	Kakotibari T.E	T.E. Supply	116088	Present	28000
20	Mockrong T.E	Tube well	80,046	Present	3400
21	Woka T.E	T.E. Supply	76,0098	Present	3400
22	Sockieting T.E	T.E. Supply	1,11,022	Present	28000
23	Jamuguri T.E	Tube well	90,046	Present	1100
24	Borjan T.E	Tube well	13,408	Absent	29
25	Dokhinhengra T.E	pond	74082	Present	340
26	Kamabandha Ali	Ring well	92,018	Present	2400
27	Morongi T.E	Tube well	86,082	Present	1100
28	Hautley T.E	Tube well	88,088	Present	1100
29	Letekujan T.E	Tube well	60,464	Present	3400
30	Majikuchi T.E	Tube well	62,242	Present	2900
31	Thorajan T.E	Tube well	64,064	Present	2900
32	Missamara T.E	Tube well	66,012	Present	2900



Figure II.: A bar diagram is plotted between New diarrheal cases of Golaghat district vs weeks of the month October 2007 to November 2007.



Conclusion:

The study areas were highly polluted with respect to bacteriological. Water of present study areas were not fit for drinking. Insects or other media may carry bacteria to enter the well, tube well, pond or supply water. The source of contamination may be septic system, too close to the well or the well casing isn't deep enough to assure that recharge water receives sufficient filtration to remove bacteria. The Newly made wells or tube wells often show contamination because the drill hole was contaminated by dirty tools, pipe or drilling water . Contaminated surface water or groundwater can enter an improperly constructed well. The e-coli contaminated water can be treated using chlorine, ultra-violet light, or ozone, all of which act to kill or inactivate E. coli. Systems using surface water sources are required to disinfect to ensure that all bacterial contamination is inactivated, such as E. coli. If any water sample test is positive for E. coli, it should not drink the water unless boil it for at least one minute at a rolling boil, longer anyone live at high altitudes. We would like to recommend the following important points: proper sanitary survey, design and implementation of water and/or sanitation projects; regular disinfections, maintenances and supervisions sources; and regular of water bacteriological assessment of all water sources for drinking should be Planned and conducted. Statistical observation shown that all the observation exhibits an asymmetric distribution with a long asymmetric tail on the right of the median. Difference between mean and madian in each case, high standard deviation and positive kurtosis in most of the cases indicate that the distribution of observation is widely off normal. Wide

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data range in each observation indicates the presence of extreme values in the form of outliers, which is bias the normal distribution statistics.

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