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RESEARCH ARTICLE

AN ASSESSMENT OF BACTERIOLOGICAL AND PHYSICOCHEMICAL QUALITIES OF GROUNDWATER IN THE RURAL ENVIRONS OF CHELEKERE, INDIA

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ABSTRACT

Groundwater is normally considered a "safe-source" of drinking water as it is usually having a low microbial load that could be consumed without treatment. However, groundwater sources are often vulnerable to contamination, thus lowering their quality. This study was conducted to determine the groundwater quality in Chelekere, Karnataka State. The study was designed using standard methods to obtain the physical properties [Temperature, pH, Total Dissolved Solids, Total Solid and Total Suspended Solid] and biological properties [Biochemical Characterization Tests, Biological Oxygen Demand, Chemical Oxygen Demand, Dissolved Oxygen, Gram Staining, Most Probable Number and Motility] of the water. All the physical properties were within acceptable limits conversely the biological properties were higher than the WHO, BIS and CPCB Guidelines and Standards for Water. The detection of *Enterobacter* spp., *Escherichia* spp., *Klebsiella* spp., *Proteus* spp., *Pseudomonas* spp., *Shigella* spp. and *Yersinia* spp. from water suggests that these sources could pose severe health hazards to consumers and is inappropriate for direct human ingestion without treatment. The research recommends the utilization of on-site treatment interventions to safeguard the households from further possible consequences of using the water.

INTRODUCTION

The quality of drinking water is closely associated with human health, ordinarily providing of drinking water is one of the important public health priorities. The impact of water on health derives principally from the consumption of water, containing pathogenic organisms or toxic chemicals (Sarker *et al.*, 2016). In rural areas, many people collect water of doubtful quality from unprotected wells or surface water sources, often at a great distance from their homes, discouraging them from collecting sufficient quantities (Ibrahim, 2016). Generally, groundwater quality varies from place to place, sometimes depending on seasonal changes, the types of soils, rocks, and surfaces through which it moves (Palamuleni and Akoth, 2015). As the population, pollution and environmental degradation increases, the drinking water sources face increasing threats from contamination of both chemical and microbiological origins (Malhotra *et al*., 2015). The majority of the populations in developing countries are inadequately supplied with potable water and are thus bound to consume water from sources like shallow wells and boreholes that have a high potential for contamination and provide the unsafe water for domestic and drinking purposes (Sudhakar and Hemalatha, 2015). The World Health Organization Guidelines for Drinking-Water Quality recommends that Faecal Indicator Bacteria (FIB), preferably *E. coli* or alternatively Thermo-Tolerant Coliform (TTC), should not be detectable in any 100-ml drinking water sample (World Health Organization [WHO], 2004); the Bureau of Indian Standards [BIS] (2012) follows the similar guideline to WHO. However, the Central Pollution Control Board [CPCB] (2008) uses a different guideline: The present study was aimed to investigate the Physical and Biological water quality from the various sources in the Chelekere district.

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MATERIALS AND METHODS

Chelekere is the surrounding areas near the Chelekere Lake; often, considered part of Kalyan Nagar which it is located in the Hebbal Valley, near to the Hennur, Kammanahalli, Babusapalya, and Meganahalli. Samples of groundwater water were collected at different locations in Chelekere, Karnataka State India. Total of 20 water sample points was used; water was collected in three different sterile one-liter containers and was taken to the laboratory. Water samples were analyzed for Physical (Temperature, pH, Total Suspended Solid [TSS], Total Dissolved Solid [TDS], and Total Solid [TS]) and Biological (Most Probable Number [MPN], Gram Staining, Motility, Biochemical characteristics, Dissolved Oxygen [DO], Biological Oxygen Demand [BOD], and Chemical Oxygen Demand [COD]) qualities using standard methods from 'Standard Methods for the Examination of Water and Wastewater' (Clesceri *et al*., 1998). The media used for the bacteriological analysis include; for isolation [Brilliant Green Lactose Broth (BGLB) and Eosin Methylene Blue Agar EMB)] and confirmatory [Lactose broth (LB) and MacConkey Agar (MAC)].

Designated best use	Quality Class	Primary Water Quality Criteria
Drinking water source without conventional treatment but with chlorination	A	➢ Total coliform organisms (MPN*/100 ml) shall be 50 or less ➢ pH between 6.5 and 8.5 ➢ Dissolved Oxygen 6 mg/l or more, and ➢ Biochemical Oxygen Demand 2 mg/l or less
Outdoor bathing (organized)	B	➢ Total coliform organisms(MPN/100 ml) shall be 500 or less ➢ pH between 6.5 and 8.5 ➢ Dissolved Oxygen 5 mg/l or more, and ➢ Biochemical Oxygen Demand 3 mg/l or less
Drinking water source with conventional treatment	C	➢ Total coliform organisms(MPN/100 ml) shall be 5000 or less ➢ pH between 6 and 9 ➢ Dissolved Oxygen 4 mg/l or more, and ➢ Biochemical Oxygen Demand 3 mg/l or less
Propagation of wildlife and fisheries	D	➢ pH between 6.5 and 8.5 ➢ Dissolved Oxygen 4 mg/l or more, and ➢ Free ammonia (as N) 1.2 mg/l or less
Irrigation, industrial cooling, and controlled disposal	E	➢ pH between 6.0 and 8.5 ➢ Electrical conductivity less than 2250 micro mhos/cm, ➢ Sodium Absorption Ratio less than 26, and Boron less than 2 mg/l.

* MPN: Most Probable Number

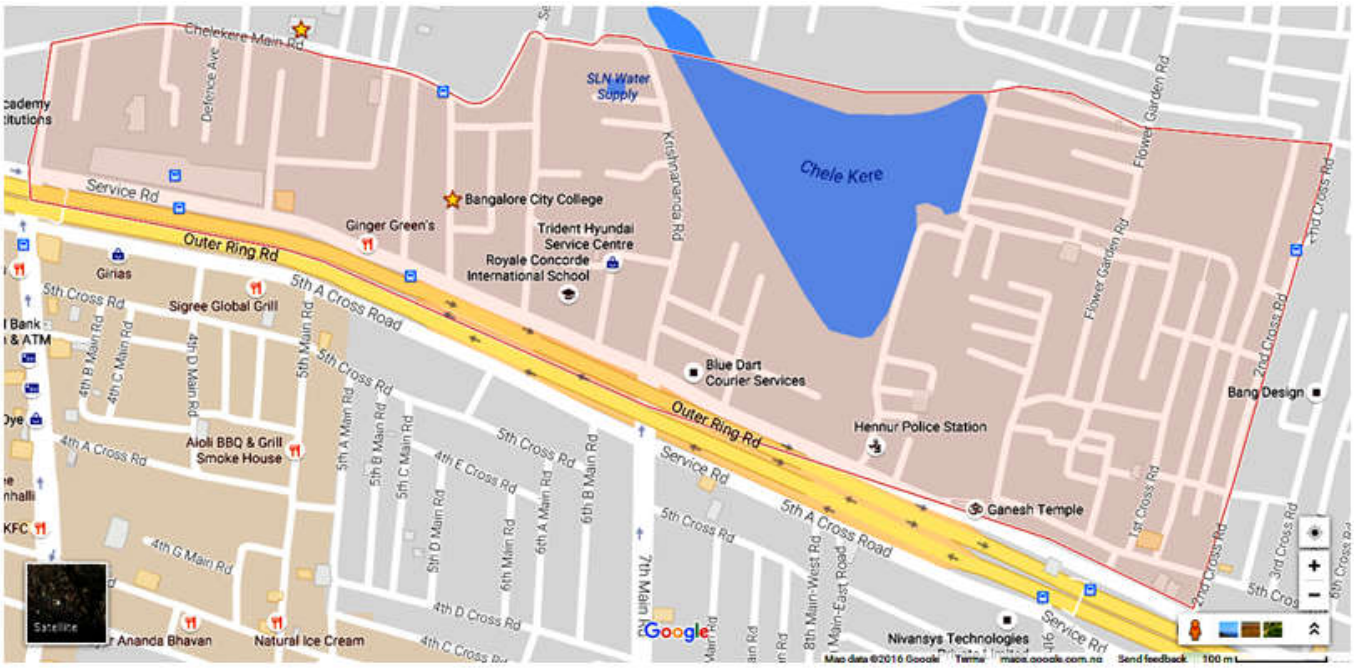

Figure 1. Map of Chelekere

All the media used were weighed out and prepared according to the manufacturer's specification, with respect to the given instructions and directions.

Data Analysis: Data for physiochemical and biological components in water samples were analyzed and documented. Mean and standard deviations were calculated from the analysis of the three samples per sampling point for the physical parameters.

The MPN result was compared with FDA's Bacterial Analytical Manual (Blodgett, 2006). The pure bacterial isolates cultures were identified by subjecting to numerous morphological and biochemical classification tests recommended by the Bergey's Manual of Determinative Bacteriology (Holt *et al*., 1994). Analysis of water quality was compared with the accepted standards provided by WHO (2011), BIS (2012) and CPCB (2008).

RESULTS

Physical properties: Table 1 shows the recorded values obtain for The Temperature, pH, TSS, TDS, and TS. The Temperature of the sample at study area varied between 27.57 ± 0.12 - 29.60 ± 0.52 ^{O}C. The pH value of the sample varied ranged 6.53 ± 0.06 - 8.33 ± 0.06. The values of TSS in the studied area ranged between 059.33 ± 1.15 - 262.33 ± 4.04 mg/L, The TDS values in the studied area stretched between 238.67 ± 1.52 - 497.67 ± 3.22 mg/L, while the Total Solid values ranged from 341.67 – 666.66 mg/L.

Biological properties: The positive results of MPN (Table 2) revealed by samples that produced gas bubbles and which significantly varied in their MPN values. The water in the studied area was found to be in the range of 0 (<1.8) – 43 MPN per 100 ml. The cultural characteristics on the EMB and MAC are shown in Table 3. The Biochemical Characterization identified is shown in Table 4 and shows the seven (7) bacterial strains were isolated; Gram-staining showed Gram-negative with rod-shaped and Motility was seen in 4 isolates while rest were non-motile. Table 5 shows DO, BOD and COD values of the water sample used.

Table 1. Physical Parameters

Sample No.	Temperature (^{0}C)	pH	Total Suspended Solids (TSS) (mg/L)	Total Dissolve Solids (TDS) (mg/L)	Total Solids (TS) (mg/L)
S01	27.27± 0.15	7.87 ±0.06	181.00 ± 1.83	382.00 ± 3.46	563.00
S02	29.43 ± 0.21	6.77 ±0.06	059.33 ± 1.15	331.33 ± 2.31	390.66
S03	27.57 ± 0.25	7.63 ±0.06	262.33 ± 4.04	258.67 ± 2.30	521.00
S04	26.37 ± 0.45	6.87 ±0.06	116.67 ± 5.77	318.00 ± 2.65	434.67
S05	27.50 ± 0.27	7.63 ±0.06	081.67 ± 2.89	477.33 ± 2.52	559.00
S06	28.63 ± 0.25	6.97 ±0.06	142.00 ± 3.46	317.00 ± 2.64	459.00
S07	27.03 ± 0.30	6.63 ±0.06	161.33 ± 2.31	401.00 ± 3.60	562.00
S08	26.27± 0.45	7.77±0.06	202.33 ± 2.31	448.67 ± 3.21	651.00
S09	27.57 ± 0.12	6.53 ±0.06	101.67 ± 2.89	240.00 ± 2.00	341.67
S10	27.40 ± 0.36	6.77 ±0.06	060.33 ± 1.15	389.33 ± 2.08	449.66
S11	28.87 ± 0.32	7.83 ±0.06	121.00 ± 1.73	238.67 ± 1.52	359.67
S12	28.60 ± 0.36	6.73 ±0.06	217.33 ± 4.62	397.67 ± 3.00	615.00
S13	27.43 ± 0.40	7.83 ±0.06	120.67 ± 1.15	360.33 ± 4.51	481.00
S14	26.67 ± 0.25	8.17 ±0.06	083.67 ± 6.35	344.00 ± 5.29	427.67
S15	28.23 ± 0.23	6.67 ±0.06	202.00 ± 3.46	497.67 ± 3.22	620.67
S16	26.90 ± 0.26	7.17 ±0.06	198.33 ± 2.08	368.33 ± 1.15	566.66
S17	27.63 ± 0.35	6.57 ±0.06	239.67 ± 1.15	468.33 ± 2.88	559.00
S18	27.93 ± 0.32	7.63 ±0.06	198.33 ± 2.08	283.33 ± 5.77	666.66
S19	29.60 ± 0.52	7.93 ± 0.06	150.33 ± 2.51	283.33 ± 5.77	433.66
S20	28.83 ± 0.29	8.33 ± 0.06	220.67 ± 1.52	432.00 ± 3.46	652.67
WHO	30.0	6.5-8.5		<500	
BIS		6.5-8.5		500-2000	
CPCB		6.5-8.5		500-2000	

WHO (World Health Organisation); BIS (Bureau of Indian Standards) – (IS: 10500: 2012); CPCB (Central Pollution Control Board)

Table 2. Most Probable Number (MPN)

Sample No.	MPN Index (Per 100 ml) *	95% Confidence Range*	
		Low	High
S01	03.6	00.70	010.0
S02	06.1	01.80	015.0
S03	0 (<01.8)	-	006.8
S04	21.0	06.80	040.0
S05	01.8	00.09	006.9
S06	06.8	01.80	017.0
S07	04.0	00.70	012.0
S08	03.7	00.70	010.0
S09	07.8	02.10	022.0
S10	0 (<01.8)	-	006.8
S11	17.0	05.90	036.0
S12	02.0	00.10	010.0
S13	0 (<01.8)	-	006.8
S14	43.0	14.00	100.0
S15	06.1	01.80	015.0
S16	03.6	00.70	010.0
S17	0 (<01.8)	-	006.8
S18	04.5	00.79	015.0
S19	0 (<01.8)	-	006.8
S20	27.0	09.90	070.0
Total	158		
Mean	7.9		
Median	3.85		
Mode	0		
Standard Deviation	11.0606		
Variance	122.33684		
Confidence Interval (95%)	± 04.85	03.05	012.75
WHO	0		
BIS	0		
CPCB	<50		

Number of tubes giving a positive reaction out of five (5) tubes, * FDA's Bacterial Analytical Manual

Table 3. The Cultural Characteristics

Media	Eosin Methylene Blue Agar [EMB]		MacConkey Agar [MAC]	
Organism	Colour	Remarks	Colour	Remarks
Enterococcus spp.	Red	Minute, round	Pink	Acid Precipitated Bile
Escherichia spp.	Red/pink	Non-Mucoid	Pink	Acid Precipitated Bile
Klebsiella spp.	Red/pink	Mucoid, round	Pink	Acid Precipitated Bile
Proteus spp.	Colourless	Mucoid	Colourless	No Precipitated Zone
Pseudomonas spp.	Green-brown	Fluorescent growth	Colourless	No Precipitated Zone
Shigella spp.	Colourless	Mucoid	Colourless	No Precipitated Zone
Yersinia spp.	Red	Minute, round	Colourless	No Precipitated Zone

Table 4. Gram Staining and Biochemical Reaction

Isolate	01	02	03	04	05	06	07
Gram-Stain	Rod (-)	Rod (-)	Rod (-)	Rod (-)	Rod (-)	Rod (-)	Rod (-)
Motility	+	+	+	-	+	-	-
Indole Test	(+)	-	-	+	+	+	-
Methyl Red Test	+	-	-	+	+	+	-
Voges Proskauer	(+)	-	+	-	-	-	+
Catalase Test	+	+	+	+	+	+	+
Citrate Test	+	+	+	-	-	-	+
Gelatinase Test	-	+	-	-	-	-	+
Oxidase Test	-	+	+	-	-	-	-
Urease Test	+	-	+	-	-	+	+
TSI	$^{K}/_{K}H_2S$	$^{K}/_{K}$	$^{K}/_{A}G$	$^{A}/_{A}$	$^{A}/_{A}G$	$^{K}/_{K}$	$^{A}/_{K}G$
Inference	*Proteus* spp.	*Pseudomonas* spp.	*Enterobacter* spp.	*Shigella* spp.	*Escherichia* spp.	*Yersinia* spp.	*Klebsiella* spp.

Key: + = Organism was Positive for the Test, - = Organism was Negative for the Test, (+) = Organism was Weakly Positive for the Test, A = Acidic, K = Alkaline, G = Gas Production, H_2S = H_2S Production.

Table 5. BOD, COD and DO

Sample No.	Dissolved Oxygen (DO) (mg/L)	Biological Oxygen Demand (BOD) (mg/L)	Chemical Oxygen Demand (COD) (mg/L)
S01	5.10 ± 0.10	3.13 ± 0.12	33.50 ± 0.20
S02	6.53 ± 0.15	4.13 ± 0.12	33..77 ± 0.21
S03	6.03 ± 0.15	3.50 ± 0.17	12.27 ± 0.37
S04	7.23 ± 0.15	3.13 ± 0.12	12.57 ± 0.42
S05	7.47 ± 0.12	3.90 ± 0.17	34.00 ± 0.61
S06	5.03 ± 0.15	2.53 ± 0.12	17.37 ± 0.29
S07	8.40 ± 0.35	5.13 ± 0.12	19.677 ± 0.21
S08	7.60 ± 0.20	5.27 ± 0.23	10.00 ± 0.12
S09	6.63 ± 0.15	3.13 ± 0.12	42.37 ± 0.55
S10	4.77 ± 0.25	1.93 ± 0.12	10.07 ± 0.12
S11	8.60 ± 0.17	4.70 ± 0.17	24.17 ± 0.29
S12	6.67 ± 0.11	2.27 ± 0.23	25.00 ± 0.20
S13	7.00 ± 0.17	1.87 ± 0.23	27.50 ± 0.26
S14	5.83 ± 0.15	2.27 ± 0.23	17.77 ± 0.21
S15	7.73 ± 0.12	5.13 ± 0.12	27.87 ± 0.15
S16	5.63 ± 0.25	4.13 ± 0.12	28.13 ± 0.23
S17	7.43 ± 0.25	2.70 ± 0.17	10.07 ± 0.21
S18	7.80 ± 0.20	3.13 ± 0.12	09.00 ± 0.20
S19	6.70 ± 0.10	4.27 ± 0.23	12.97 ± 0.06
S20	7.07 ± 0.15	2.13 ±0.12	14.17 ± 0.21
WHO	5.0	6.0	
BIS	6.0	3.0	
CPCB	6.0	2.0	

WHO (World Health Organisation), BIS (Bureau of Indian Standards) – (IS: 10500: 2012), CPCB (Central Pollution Control Board)

The DO values varied between 4.77 ± 0.25 - 8.60 ± 0.17 mg/L. BOD readings ranged between 1.87 ± 0.23 - 5.27 ± 0.200 mg/L. COD values ranged between 9.00 ± 0.20 - 42.37 ± 0.55 mg/L.

DISCUSSION

Physical properties

The temperature of the sample was lower than the prescribed limit by WHO. Pianetti *et al.* (2005) conducted a study in Italy and showed that the survival curves of *Aeromonas* spp. decline rapidly at low temperature ($5^{O}C$), whereas survival at temperatures greater than $20^{O}C$ increases. The pH value in the studied area was within the prescribed limit by WHO, BIS and CPCB.

Byamukama *et al.* (2000) observed that some open springs and dug wells had low pH values which might be due to saturated with carbon dioxide. The pH of water is a very important property because it will decide whether the water is suitable for drinking purpose. The pH of the water samples also signifies that it can be used as drinking water. Although pH usually has no direct impact on water consumers, it is one of the most important operational water-quality parameters. Careful attention to pH control is necessary at all stages of water treatment to ensure satisfactory water clarification and disinfection. For effective disinfection with chlorine, the pH should preferably be less than 8 (Krishnan *et al*., 2007). S02, S05, S10, S14 contain had the lowest amount of TSS below 100 mg/L; 059.33 ± 1.15, 081.67 ± 2.89, 060.33 ± 1.15 and 083.67 ± 6.35 [mg/L] respectively. S03, S08, S12, S15, S17,

S20, contained high amount of TSS above 100 mg/L; 262.33 ± 4.04, 202.33 ± 2.31, 217.33 ± 4.62, 202.00 ± 3.46, 239.67 ± 1.15, and 220.67 ± 1.52 [mg/L] respectively. None of the samples exceeded the prescribed TDS limit required by WHO (2011), but they are lower than the permissible limit according to BIS (2012) & CPCB (2008). High values of TDS in groundwater are generally not harmful to human beings but a high concentration of these may affect persons who are suffering from kidney and heart diseases. A high content of dissolved solids elevates the density of water, influences osmoregulation of freshwater organism, reduces the solubility of gases [like oxygen] and reduces the suitability of water for drinking, irrigation, and industrial purposes (Trivedi and Vediya, 2011).

Biological properties

The statistical estimate of the MPN of the study area was calculated, an MPN Mean Value of 7.9 and Confidence Interval of 4.85 was obtained which indicates an overall low MPN value in the study area. However, according to WHO (2011) and BIS (2012) standards, only five (5) water sample (S03, S10, S13, S17, and S19) can be deemed as safe to be used as drinking water (i.e. <1.8 or 0). The rest cannot be used as drinking water as the MPN value ranged from 1.8 – 43 MPN per 100 ml, but according to CPCB (2008), they can be considered for use as drinking water with proper treatment such as chlorination and these samples belong to Class A. The finding of microbes of fecal source in the current study revealed that the water was not secure and might serve up as a latent basis for the conduct of these microbes to persons who drink this water and the sanitation around the water sources were not maintained adequately or properly these conditions may pose potential health problems to those using the water from such unhygienic surroundings (Sudhakar and Hemalatha, 2015). All 20-sampling point showed higher DO values than the prescribed limit by WHO (2011). While the CPCB (2008) states: Drinking Water Source without conventional treatment but after disinfection should have DO of 6mg/L or more; or Drinking water source after conventional treatment and disinfection should have DO of 4mg/L or more. The concentration of dissolved oxygen indicates the distribution of flora and fauna (Naveen *et al*., 2017).

The high concentration of DO imparts good taste to water. In the study conducted by Krishnan *et al.* (2007) around Sivakasi found that dissolved oxygen of their water samples to be 8.33mg/ml and 7.41mg/ml. Similarly, suggested the reason for the low dissolved oxygen content was due to decomposition of organic matter, which can indicate a pollution load in the water. Accordingly, the deficiency of the oxygen in the water is a shelter for bacteria and other pathogens, which are anaerobic. All sampling point showed that the BOD values within the prescribed limit by WHO (2011). While according to the CPCB (2008) also stated, Drinking Water Source after conventional treatment and disinfection should have Biochemical Oxygen Demand 5 days (BOD_5) at 20°C, 2mg/L or less, or drinking water source without conventional treatment but after disinfection should have BOD_5 days, 20°C, 3mg/L or less. BOD indicates the organic load in water samples. Nevertheless, the higher BOD values are found in the polluted water (Naveen *et al*., 2017). Water with high COD values indicates that there is inadequate oxygen available in the water sample (Narasimha *et al*., 2010). Correspondingly, the high chemical oxygen demand levels indicate that there is inadequate oxygen available in the water samples. The depletion of oxygen in the water samples is due greatly to microbial activities related to the dumpsites (Akudo *et al*., 2010).

Conclusion

As Ramachandra and Aithal (2015) reported that the loss of ecologically sensitive wetlands is due to the uncoordinated pattern of urban growth; urbanization and the consequent loss of lakes have led to a decrease in catchment yield, water storage capacity, wetland area, the number of migratory birds, flora and fauna diversity, and the groundwater table. In addition Palamuleni and Akoth (2015), states that human activities can alter the natural composition of groundwater through the disposal or dissemination of chemicals and microbial matter on the land surface and into soils; or can accumulate and migrate to the water tables thus affecting both the physical, chemical and microbial quality of water. Consequently, groundwater is a precious natural resource which forms an important part of the hydrologic cycle. In comparison with the surface water pollution, the groundwater contamination is difficult to control (Prakash and Somashekar, 2006). According to Ramachandra *et al.* (2016), in the Hebbal Valley, 72% of lakes belong to class E and 28% belongs to class D and E of CPCB (2008). Furthermore, the present investigation on the Physical and Biological quality of water around Chelekere showed that some of the water sources are not suitable for domestic purposes as specified by the WHO (2011) and BIS (2012) guidelines and standards. About 75% of water samples were found beyond the acceptable limits of WHO (2011) and BIS (2012) with at least one of the parameters is more than the acceptable limit of WHO (2011) and BIS (2012). The MPN value ranged between 1.8 – 43 MPN per 100 ml but based on the MPN Mean Value (7.9) and Confidence Interval (4.85) which indicates an overall low MPN value in the study area. Conversely, the CPCB (2008) limit for MPN index of <50 can be acceptable if the further treatment like Shock Chlorination Treatment is applied to these sources. From the present study, it is evident that groundwater quality is progressively getting deteriorated and it may worsen further with time. So, the public should be made aware of the water quality importance and hygienic conditions before use. Also, it is necessary to implement certain remedial measures such as Shock Chlorination Treatment, commercially available filtration setups, etc. Just like Malhotra, Sidhu and Devi (2015) stated the bacteriological assessment of all sources of drinking should be planned and conducted on regular basis to prevent waterborne dissemination of diseases; as a matter of public health importance.

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