Case Study
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MICRIBIOLOGICAL ASSESSMENT OF DRINKING WATER AND PERVASIVENESS OF WATER BORN DISEASES IN DUNGARPUR, RAJASTHAN

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ABSTRACT

Introduction: This study examined E. coli numbers in drinking water from different water bodies and illustrates the high prevalence of gastrointestinal diseases in Dungarpur districts of southern Rajasthan, India.

Methodology: The microbiological parameters studied were pathogen detection, total coliform count, and total viable count. Microbiological examination of sampling water was described as in APHA (2005) Standard Methods for the Examination of Water and Wastewater Chhabra (2008). Total coliform count performed by the Most Probable Number (MPN) method, which is commonly used as an indicator of potability of water. Presence of the Enterobacteriacae was confirmed by urease analysis. Waterborne disease patients (Indoor and Outdoor patients) figures were collected.

Result And Discussion: Seasonal fluctuation in coliform counts was found to be prominent and varied from over 200 MPN/ 100 ml in winter to more than 1600 (over the measurable limit) in rainy season in three water bodies. E. coli numbers growth from 17 to 500 MPN/100 ml in the selection site Total coliform, fecal coliform and E. coli were highest in rainy season. Results of IMViC test indicates that in rainy season except for Margia Dam all the water bodies were laden with all five categories of enteric bacteria namely E. coli, Shigella, Citrobacter and Klebsiella/ Enterobacter. Increase in pathogens numbers were excessive during the rainy season compared to summer season, because of WBD patients does exhibit a trend that in late summer and rainy month's incidence of the disease symptoms intensifies.

Conclusion: Dungarpur district was analyzed and same pattern of seasonal variation in microbial counts of water samples and similar hospital data trend was observed. So, somehow the microbial contamination of surface water and run-off reaches the ground-water table. Therefore, hospital data confirms that the source of drinking water plays a strong, positive and significant association with waterborne diseases.

Keywords: Drinking Water, Microbial Load, MPN (Most Probable Number), Escherichia Coli, Klebsiella Pneumoniae, Imvic (Indole, Methyl-Red, Vogus-Proskauer, And Citrate Utilization), Citrobacter Freundii, Shigella

1. INTRODUCTION

The availability of freshwater especially for purpose of drinking will be a one of the most serious resource and environmental issue for long time to come. Poor water quality spreads disease, causes death and affects socio-economic progress. We need water every day for drinking, agriculture and in other activities. Pathogenic microorganisms contaminate the water and are now a major global problem. The

main causes of bacteria in the aquatic environment are the disposal of human waste and municipal wastewater through sewage and drainage discharge systems. Human pathogenic bacteria, particularly members of the coliform can inhabit on fishes and thrive in aquatic environment Saha et al. (2012).

Water become unsafe for human consumption or usage when it contains pathogenic or diseases causing microorganisms. The consumption of unhygienic drinking water and uses of unsafe water for daily purposes lead to the prevalence of diseases among the population Moe and Rheingans (2006). Total coliforms (TC) comprise bacterial species of faecal origin as well as other bacterial groups (e.g., bacteria commonly occurring in soil). Consumption of water with high TC counts in water are usually manifested in the form of diarrhoea and sometimes as fever and other secondary complications.

Dungarpur district population is increased every year but water sources are not much enough. However, microbial contamination in the drinking waterbodies is districts problem that warrants a proper attention. It is evident from hospital data and its correlation with microbial burden of water that the issue is of prime health concern in district and elsewhere. In view of this, an attempt has been made to assess the impact of drinking water on human health in prevailing conditions of its availability and contamination in this district.

2. MATERIAL AND METHODS

2.1. SAMPLING

Samples were collected in sufficient volumes from five sites in early hours of the day during every season sterile sample bottles in the pre-monsoon (February to May), post-monsoon (October to January) and monsoon (June to September) seasons in year 2015-2016.

2.2. MICROBIOLOGICAL ANALYSIS

MPN for Coliform Bacteria

Escherichia coli are isolated by inoculating the sample in Bismuth green bile broth. Enteric bacteria isolated on respective selective or differential media were identified on the basis of their colonial, morphological and Biochemical properties Table 1 following Bergey's Manual of Determinative Bacteriology, 1994.

This is the test for differentiate between related genera or between species within a genus and get information about particular species.

Table 1

Table 1 Biochemical Test

S. No.	Biochemical Testing	Inferences	Type of Bacteria
1	Indole test	Appearance of pink coloured ring positive	*E.coli.* present
2	Methyl red test:	positive	*E.coli* and Citrobacter freundii present
3	Citrase utilization test:	green colour or blue colour Green Negative. Blue- Positive	Absence or presence of Citrobacter freundii.
4	Urease test:	yellow colour - negative. pink colour - positive.	Citrobacter freundii. and Klebsiella pneumoniae present

5	Oxidase reaction:	Appearance of purple colour within 30 minutes.	P. aeruginosa.
6	Fermentation and gas production test:	Change of colour from blue to yellow.	Presence of fermenting and gas producing bacteria.

Escherichia coli were identified using MacConkey and Brilliant green blue broth as total coliform units in the samples.

IMViC Test

Differentiation of principal groups of enteric bacteria can be accomplished on the basis of their biochemical properties and enzymatic reactions in presence of specific substrates. So, the IMViC test was adopted for discerning the types of bacterial contaminants in water.

Characteristic based on IMViC pattern

Organism	Indole	Methyl Red	Voges Proskauer	Citrate
Escherichia coli	+	+	-	-
Shigella sp	+ or -	+	-	-
Citrobacter freundii	-	+	-	+
Citrobacter diversus	+	+	-	+
Klebsiella, Enterobacter and Aerobacter group	+ or -	-	+	+

The IMViC tests stands for four different tests: a) Indole test; b) Methyl-red test; c) Voges-Proskauer test; and d) Citrate utilization test. The letter "i" included for pronunciation. For this test, methodology suggested by Cappuccino and Natalie (2004) was adopted.

3. RESULT AND DISCUSSION

In this study two indicators of the microbial contamination were analyzed. Total Coliform organism and *E. coli* count per 100 ml of sample was estimated. so high microbial load can already be expected in data. Total coliform value as illustrated in **Figure 1** was the real matter of concern. It was from over 200 MPN/ 100 ml in winter to more than 1600 (over the measurable limit) in rainy season in three water bodies. Least coliform value was measured in winter and highest in rainy season.

3.1. TOTAL COLIFORM

Figure 1

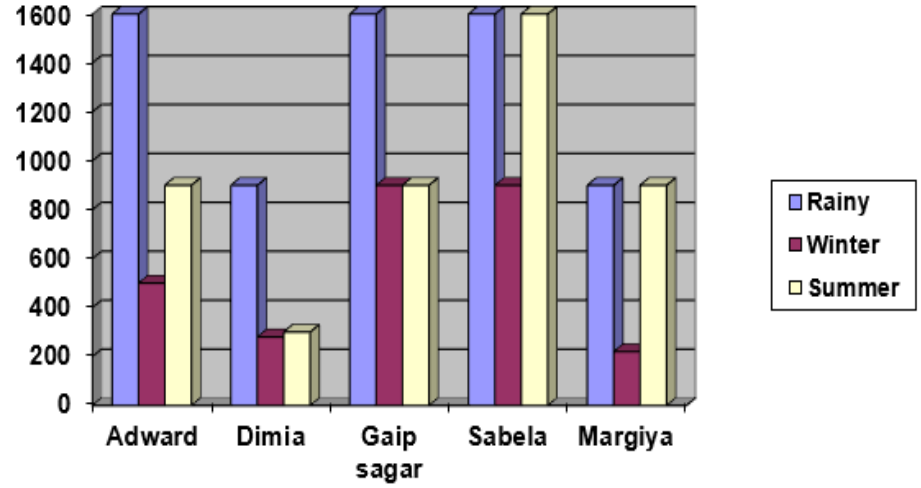

Figure 1 Graphical Representation of Total Coliform in the Water Bodies in Three Seasons

Total Coliform - In Adward samand total coliform was >1600 in rainy season, 500 in winter and 900 in summer. This count in Dimia talab was 900, 280 and 300 in Gapsagar 1600, 900 and 900, in Sabela pond >1600, 900 and 1600 and in Margia dam 900, 220 and 900 in respective seasons. So, there was difference in the microbial load of different water bodies, and drastic seasonal variation is also apparent.

Coli Count

The count was dependent on the location of reservoirs, waste and sewerage entry and the inflow of the water during rains from surrounding area. The count of > 1600 / 100 ml is maximum measurable and this value virtually indicates any count above 1600. So, in rainy season coliform count of Adward samand and Sabela may appear same, but their actual microbial burden may be different.

Figure 2

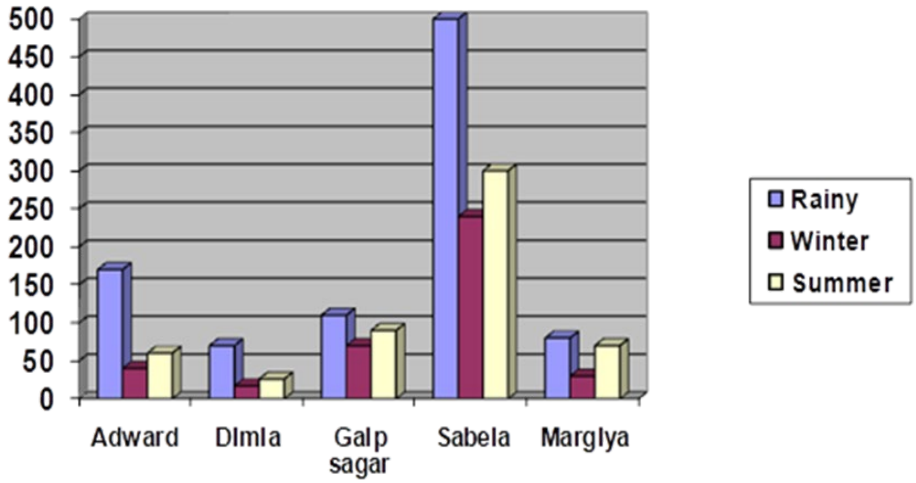

Figure 2 Graphical Representation of E. Coli Organism in the Water Bodies in Three Seasons

In rainy season the coliform count in Adward samand may be the result of the inflow of runoff water from surrounding area. Gap sagar and Sabela are polluted from city wastage (as mentioned in previous section of this chapter), so in these two water bodies high bacterial count is owing to incessant contamination. Margia dam exhibited lowest bacterial count in all seasons, still it is very high. As far as seasonal variation is concerned coliform counts were lowest in winter and highest in rainy months. This explains the high coliform count during monsoon months. The temperature also influences the trend in variation of density of bacterial population Patralekha (1992). Hence, coliform burden during summer was also high. The result of the seasonal variation and counts of total coliform in this study matches with the work of Latha and Ramachandra (2013) conducted in Bangalore, Karnataka. Identical pattern of seasonal variation has been recorded in reports of Paille et al. (1987), Ouma et al. (2016) and Pande et al. (1983). Guideline of water quality standard BIS (2012) stipulates that there should be no detectable coliform in water sample. Therefore, proper treatment of these reservoirs water before using it for drinking purpose is inevitably required. The surge of microbial count in rainy season reflects in hospital data and socioeconomic survey also as the incidences of gastrointestinal infections.

***E. coli* organism –** Faecal contamination magnitude could be ascertained by *E. coli* estimation Figure 2 in water sample. In our samples data repeat pattern with Coliform counts. *E. coli* value as illustrated in Figure 2 count was highest in rainy month, lesser in summer and least in winter in all five water bodies. The count in three seasons (in rainy, winter and summer season) as organism/ 100 ml was 170, 40 and 60 in Adward samand; 70, 17 and 26 in Dimia talab; 110, 70 and 90 in Gap sagar; 500, 240 and 300 in Sabela pond; and 80, 30 and 70 in Margia dam. Hence, though the coliform count was high in Adward samand actual faecal contamination is low, and it is same with Dimia Pond and Margia dam. It is evident that despite the high counts of total coliform the faecal contamination level in these three reservoirs is low. Sabela pond is explicitly the most polluted among all and after that Gap sagar is also highly contaminated. Actually, the *E. coli* count produces the real picture of faecal and sewage contamination of reservoirs.

3.2. IMVIC TEST

Table 2

Table 2 Result of IMViC Test Reactions of Water Samples in Different Seasons of Year

S. No.	Water body	Date/ Season of Collection	IMViC test reaction			
			Indole	Methyl - red	Voges-Proskauer	Citrate utilization
1.	Adward samand	05/10/2015; Rainy season	+	+	+	+
		02/02/2016; Winter season	+	+	–	–
		07/05/2016; Summer season	+	+	–	+
2.	Dimia Talab	05/10/2015; Rainy season	+	+	+	+
		02/02/2016; Winter season	+	–	–	–
		07/05/2016; Summer season	+	+	–	–
3.	Gap Sagar	05/10/2015; Rainy season	+	+	+	+
		02/02/2016; Winter season	+	+	–	–
		07/05/2016; Summer season	+	+	–	+
4.	Sabela Talab	05/10/2015; Rainy season	+	+	+	+
		02/02/2016; Winter season	+	+	+	–
		07/05/2016; Summer season	+	+	+	+
5.	Margiya Dam	05/10/2015; Rainy season	+	+	–	+
		02/02/2016; Winter season	–	+	–	+
		07/05/2016; Summer season	+	–	+	–

Perusal of the results of IMViC test Table 2 reveals that all the surveyed water-bodies were categorically contaminated with enteric bacteria. And, as exhibited data calculated to previous table *coli* and Total coliform contamination was highest in rainy season. But, in other seasons of year also microbial load of enteric pathogen existed with severe magnitude. This kind of microbial load indicates the influx of sewer line or other kind of contaminating drainage into the water body. Open defecation in around and in catchment area may also be a reason of this kind of contamination burden.

3.3. HOSPITAL DATA OF THE PATIENTS REGISTERED WITH WATERBORNE DISEASE SYMPTOMS IN DUNGARPUR

The data of patients registered with symptoms of water-borne disease was collected from Community or Primary health Centers (CHC or PHC) and hospitals from all blocks of Dungarpur district. There are two major Hospitals, seven Community Health Centre (CHC), forty-two Primary Health Centre and 302 Sub Centers in the District. The data was compiled block-wise from health-centers and sub-centers. Altogether there are five blocks in district and named as Dungarpur, Simalwara, Bicchiwara, Aaspur and Sagwara. Hospital, health centers and sub centers of all the blocks were covered in data collection. Only the figure of the patients registered or admitted with symptoms of water-borne disease (WBD) was collected. Since, the problem being addressed in this study is related to drinking water, so other types of water-borne disease like Malaria etc. was not taken into account. Number of the patients reporting common symptoms of WBD like nausea & vomiting, acute Diarrhoea, Gastroenteritis, Dysentery, Abdominal pain & cramps, Typhoid etc. were noted and compiled. Patients with hepatitis A were not reported in hospital during the period of data collection. The data of Indoor patients admitted in hospitals and those who reported in OPD both were collected.

3.4. OUT PATIENT DEPARTMENT (OPD) REGISTRATION DATA

It may be noted that in the span of November 2016 to April 2017 the maximum registered patients in any given month were 1413 (in April, 2017), and minimum was recorded the month of February 2017 that was 1147. Maximum number of OPD patients was registered in the month of August, 2016. Surprisingly in the month of January, 2016 to April, 2016 the figure of OPD patients is 1910, 1521, 1833 and 1704 respectively. These figures are substantially higher than the corresponding months of 2017.

However, the general trend noted in the data of all five blocks and the entire district is that the number of patients suffering from the said symptoms was lesser in the months of winter, more in the latter half of summer and maximum during rainy season. Data of January 2016 seem aberrant from that trend. Besides, it was also observed that in all the months of 2016 the number of patients were higher than corresponding months of 2017.

Figure 3

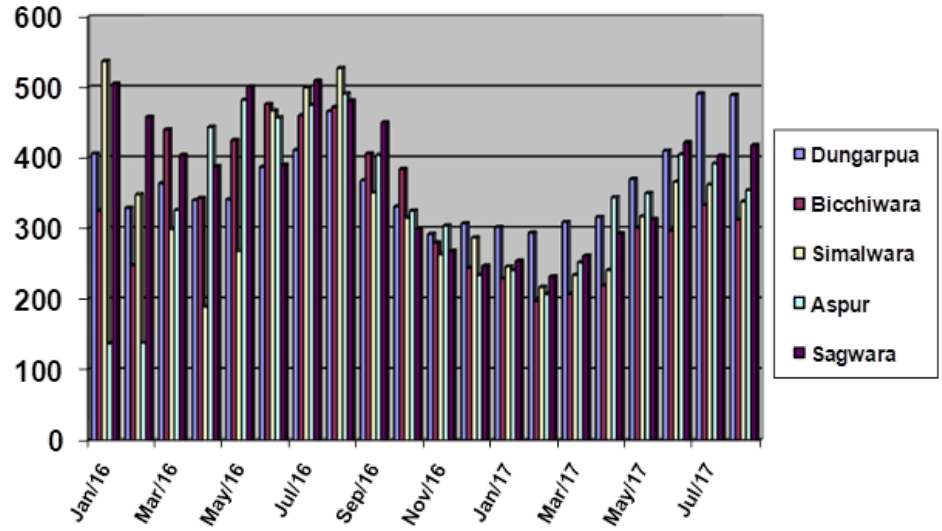

Figure 3 Graphical Representation of the Number of OPD Patients in Different Blocks of Dungarpur District During January 2016 to Aug 2017

3.5. INDOOR PATIENT ADMISSION DATA

Perusal of indoor patients' data also exhibits a seasonal pattern in the upsurge of common WBD symptoms in the population. From November 2016 to February 2017 the number of admitted patients was minimal. Maximum admission was again recorded in summer and rainy months. Figures of months January-February 2016 is again deviant from the general pattern. This is because of the recorded admissions more than in corresponding months of 2017 in Sagwara, Simalwara and Aaspur blocks. With onset of the month of summer reports of WBD symptoms starts increasing it reaches to maximum in months of June to August.

Graphical illustration of the indoor patient's data from all the five blocks in district is being given in Figure 4. The graph clearly indicates a seasonal pattern of upswing in the number of patients with WBD symptoms in summer and rainy months. In the graphical presentation, it also appears that there is maximum registration of patients in Dungarpur block. As the main district hospital is located in Dungarpur city, so many referral cases from health centers of other blocks of district are also registered there.

Figure 4

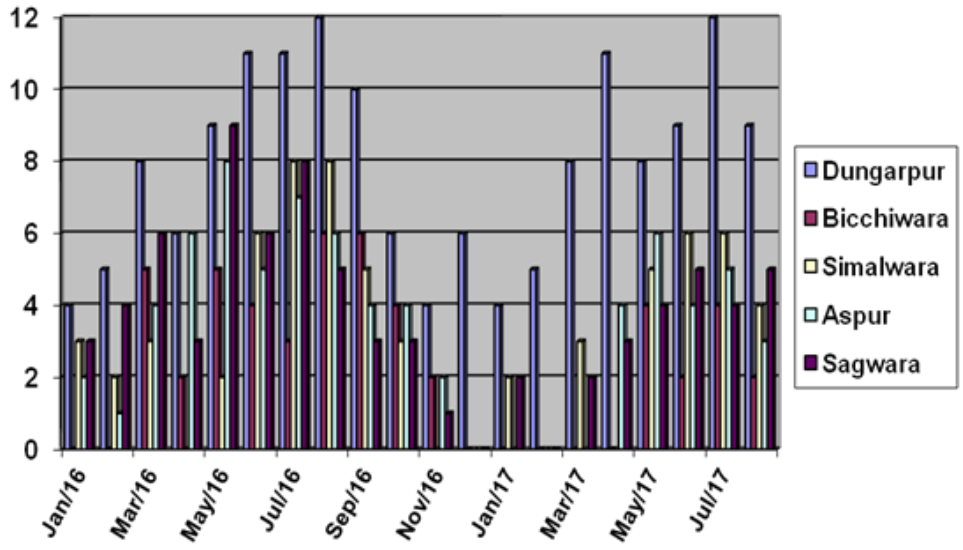

Figure 4 Graphical Representation of the Number of Indoor Patients in Different Blocks of Dungarpur District During January 2016 to July 2017

3.6. TOTAL NUMBER OF OPD AND INDOOR PATIENTS DATA

The trend of seasonal variation in WBD patients registering in hospital is similar to the indoor and outdoor patients' data shown earlier. Data of Simalwara, Dungarpur, Sagwara and even in Bicchiwara blocks in the month of January 2016 is much higher in comparison to that of January 2017. This drastic difference in the number of patients in the same month of consecutive two years is something intriguing. It may be associated with some other reason that affected the surge in the number of patients in the month of January 2016. In the month of April 2016 again the number of Simalwara block was lowest in the entire duration of data collection and much lower in comparison to the numbers from other blocks too.

It is apparent from the scrutiny of hospital data that seasonal variation in the number of WBD patients does exhibit a trend that in late summer and rainy month's incidence of the disease symptoms intensifies and figures in the month of winter are lower. Needless to mention that in late summer the source of drinking water dries up enhancing the chance of consuming contaminated water by the rural populace. It has been noted already that microbial load of water bodies goes up and this attribute of rainy season contamination of water seem to be associated with the trend that appears in hospital data.

The graphical representation of the data of total WBD patients in different blocks and in entire district of Dungarpur Figure 5 presents a distinct picture of the seasonal variation in numbers.

Figure 5

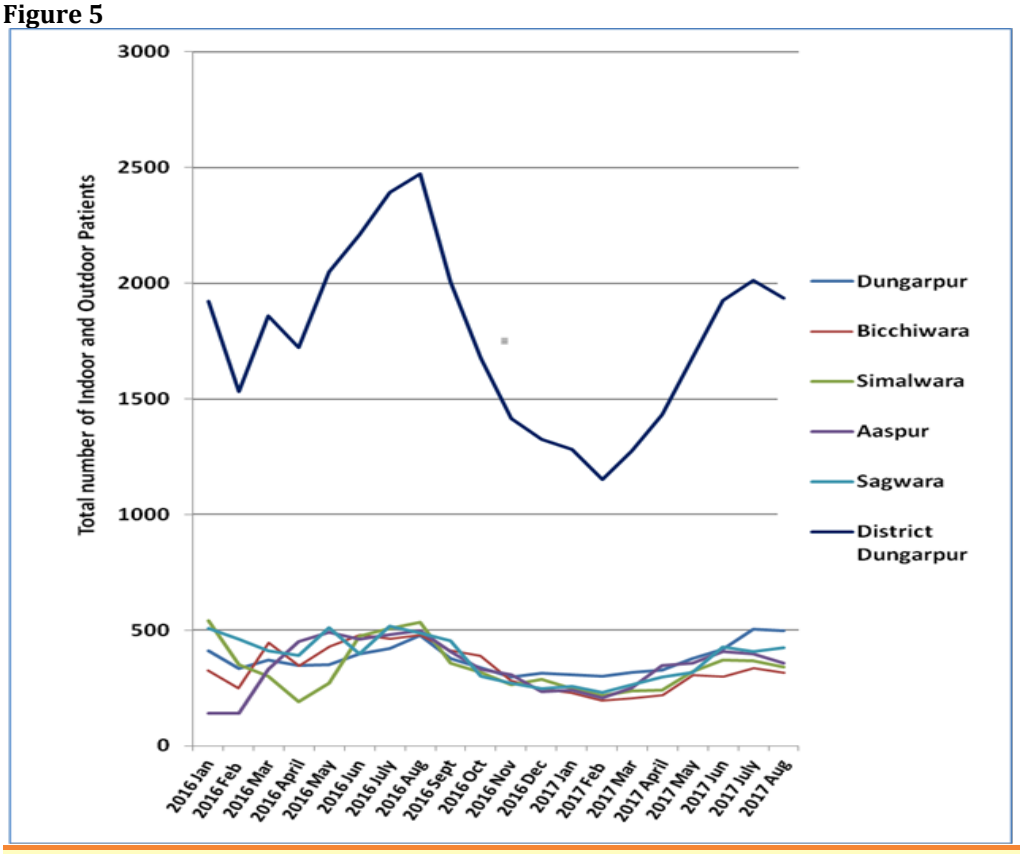

Figure 5 Graphical Representation of the Total Number of Indoor and Outdoor Patients in Different Blocks of Dungarpur District During January 2016 to July 2017

The experimental and field survey data presented in this chapter signifies some interesting findings related to the sources of drinking water with physico-chemical cum microbial properties, their contamination potential and correlation of the incidence of water-borne disease with socio-economic and educational profile of the community in study area. In the graphs depicted in Figures III -IV this data trend is clearly discernable. Therefore, the hospital data confirms the assumption that upsurge in microbial load in water-bodies is reflected in escalation of gastro-intestinal problems in the community.

This problem of microbial contamination of drinking water and consequential symptoms of GI epidemic gets more compounded when the sanitary conditions are poor due to economic backwardness and unawareness towards observing proper hygiene in daily life, and Dungarpur district falls in such a category. Hence, studying the problem of microbial assessment in this district is very relevant particularly in socio-economic prospective.

4. CONCLUSION

The aspects of the problem as discussed above are upsurge in microbial contamination during late summer and monsoon in waterbodies of the district. For the rise of microbial load in water resources rainfall and runoff may be the reason but it is compounded by reckless and unrestrained sewage, fecal waste and garbage dumping in the waterbodies and open defecation. Local population particularly people from lower economic strata are compelled to use unsafe water and ignore

sanitation or hygiene. Hence Awareness campaigns Monitoring the water-sources in disease-prone season and strengthening and maintaining the filtration system by PHED is recommendable.

CONFLICT OF INTERESTS

None.

ACKNOWLEDGMENTS

None.

REFERENCES

APHA, AWWA, WEF. (2005). Standard Methods for the Examination of Water and Wastewater, 21st Edition. Washington, DC.

BIS (2012). Indian Standard: Drinking Water- Specification, 2nd revision. IS10500:2012; 1-18.

Cappuccino, James G. and Sherman Natalie (2004). Microbiology- A laboratory Manual. Pearson Education (Singapore) Pvt. Ltd., Indian Branch, 482 FIE Patparganj, New Delhi. pp. 153-156.

Chhabra, V. K. (2008). A manual on Water, Waste Water and Material Analysis, Indian Water Works Association (IWWA), Jaipur.

Latha, N. and Ramachandra Mohan, M. (2013). Microbial Pollution – Total Coliform and Fecal Coliform of Kengeri Lake, Bangalore Region, Karnataka, India. Int. J. of Scientific and Res. Pub. 3(11), 1-3.

Moe, C.L. and Rheingans, R.D. (2006). Global Challenges in Water, Sanitation and Health. J Water Health. 4:41-57. https://doi.org/10.2166/wh.2006.0043

Ouma, S. O., Ngeranwa, J. N., Juma, K. K. and Mburu, D. N. (2016). Seasonal Variation of the Physicochemical and Bacteriological Quality of Water from Five Rural Catchment Areas of Lake Victoria Basin in Kenya. J. Environ. Anal. Chem. 3(1), 1-7.

Paille, D., Hackney, C., Cole, L. R. M. and Kilgen, M. (1987). Seasonal Variation in the Fecal Coliform Population of Louisiana Oysters and its Relationship to Microbiological Quality. J. Food Protec., 50 (7), 545-549. https://doi.org/10.4315/0362-028X-50.7.545

Pande, J., Das, S. M. and Misra, D. S. (1983). Studies on Bacteriological Quality of Nainital Lake water in Different Seasons. Water, Air and Soil Pollution. 19, 149-154. https://doi.org/10.1007/BF00211801

Patralekha, L. N. (1992). Bacterial Density in the Ganges at Bhagalpur, Bihar. J. Eco. Bio. 3(2), 102-105.

Saha, M.L., Nessa, M., Khan, M.R., Islam, M.N., Hoque, S. (2012). Bacteriological and Physicochemical Water Quality of Four Ponds of Dhaka Metropolis. Bangladesh J. Bot. 41:55-60. https://doi.org/10.3329/bjb.v41i1.11083