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Incidence of Water Borne Protozoans and Their Correlation With Faecal Indicator Bacteria in Drinking Water

KEYWORDS: Clostridium, Cryptosporidium, Faecal coliforms, Giardia, Indicator bacteria, Water borne protozoa

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ABSTRACT *A survey was conducted in rural and urban areas of district Ludhiana, Punjab (India) to find out the current status of water borne protozoans (Giardia and Cryptosporidium) and their incidence in relation to faecal indicator microbial agents (coliforms and Clostridium) in drinking water. In total 100 water samples were collected from various sources like submersible pumps, handpumps, municipal corporation taps and water filters. Giardia was not detected from any of the collected water samples. Results indicated incidence of Cryptosporidium, coliforms and Clostridium in water samples collected from handpump > submersible pumps and municipal corporation taps > water filters. Correlation of Cryptosporidium with Clostridium was also higher in handpump water followed by submersible pumps, while its correlation with coliforms was higher in submersible pumps and similar in water samples collected from other sources.*

INTRODUCTION

Safe drinking water is a basic human right and its adequate supply is one of the major prerequisites for a healthy life, but waterborne diseases are still a major cause of death in many parts of the world, particularly in children (Fawell and Nieuwenhuijsen, 2003). There are three main types of microorganisms that can be found in drinking water viz., bacteria, viruses and protozoa. These can exist naturally or can occur as a result of contamination of water by sewage, unhygienic habits or by person to person contact or food borne. The protozoan parasites like Giardia and Cryptosporidum are responsible for waterborne outbreaks of gastroenteritis via contaminated drinking water and recreational waters (Giangaspeco et al 2007) and cause diarrhoeal diseases in humans and animals (Lalancette et al 2010). The coliforms and Clostridium bacteria are world widely used as indicators of faecal contamination and also for the presence of parasitic protozoans like Giardia and Cryptosporidium oocysts (Briancesco and Bonadonna 2005). The analysis for the presence of indicators is a short cut attempt to determine the microbiological quality and public health safety of water. But the current drinking water treatment processes successfully eliminate the indicator microorganisms. They do not adequately reflect the occurrence of pathogens in disinfected wastewater effluent due to their relatively high susceptibility to chemical disinfection and failure to correlate with protozoan parasites (Tyagi et al 2006). So, to reduce the risk of illness from pathogens in drinking water and to ensure the safety of the consumers, it is imperative that multi-barrier approach should be used. The microbiological water quality guidelines are mainly followed to assess the portability of water, but protozoa have become the leading cause of water borne gastroenteritis illness. Therefore, the detection of protozoans of public health significance is needed prior to declaring water fit for human consumption.

MATERIAL AND METHODS

A total of 100 water samples were collected from urban (n=63) and rural (n=37) areas of Ludhiana district (Punjab) from different sources like submersible pumps, municipal corporation taps, water filters and handpumps. Giardia was detected from the collected water samples by sucrose floatation method (Koompapong et al 2009) and Cryptosporidium by calcium carbonate flocculation method (Shepherd and Wyn-Jones 1996). After staining with Lugol's iodine for Giardia and with Ziehl-Nelson acid fast stain for Cryptosporidium, these were identified under microscope on the basis of their morphological characteristics. Faecal coliforms was detected by MPN (Most Probable Number) method (APHA 1989). The presence of Clostridium was determined by using cooked meat medium having 10 ml of water. Test tubes were sealed with cotton plugs and autoclaved. Water samples to be tested were added in test tubes, then melted wax was poured over it. Test tubes were again sealed with cotton plug and incubated at 37°C for 48 hrs. Tubes were observed, if wax plug was forced upside, it indicated the production of gas due to the presence of Clostridium.

RESULT AND DISCUSSION

A total of 100 samples were analyzed from different sources of drinking water from district Ludhiana (rural as well as urban) and out of these, 30 samples were positive for Cryptosporidium indicating its average incidence 30%. Their comparative study mentioned in table 1 showed that water samples collected from handpumps were found to have the highest incidence of Cryptosporidium (42.8%) > submersible pumps (32.3%) > water filters (27.3%) > municipal corporation tap water (27.1%). Cryptosporidium has been considered as one of the most frequently identified etiologic agent associated with 23.7% of drinking water-borne illness outbreaks worldwide (Lalancette et al 2010). In the present study, Giardia was not detected from any of the water samples, it may be due to chlorination of water, as Giardia protozoans are susceptible to chlorination. Rose et al (1988) have also observed almost negligible contamination of water with Giardia as compared to Cryptosporidium.

Coliforms were found in 48 samples, out of the total tested 100 water samples, hence its 48% incidence and Clostridium was found in 40 samples with incidence of 40%. Maximum level of incidence of both coliforms and Clostridium was found in rural handpump water i.e 71.4% and 57.1% respectively. Water filter water samples showed equal levels of incidence for both coliforms and Clostridium (45.5%). Water samples from submersible pumps showed 47.1% incidence for coliforms and 44.1% incidence for Clostridium. Municipal corporation tap water showed 45.8% level of incidence for coliforms and 35.4% for Clostridium. Thus order of incidence of coliforms in water samples from different sources of Ludhiana district was handpumps > submersible pumps > municipal corporation taps > water filters. However, the pattern for the % infestation of Clostridium

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in the water samples was handpumps > water filters > submersible pumps > municipal corporation taps (Table 1) Faecal coliforms act as indicator organisms for determination of water quality for drinking (Oku et al 2012) and Clostridium spores also appear to be sensitive indicator for contamination of water with faecal waste (Sorensen et al 1989). A study conducted by Welch et al (2000) reported 79% incidence of total coliforms and 61.1% incidence of faecal coliforms in the stored household water. In India, a survey conducted from different rural areas to determine the quality of drinking water from various water sources like ground water, piped supplies and surface water indicated the incidence of 75.1% of thermotolerant coliforms (Ramteke et al 1992).

Out of the total tested 100 samples, both Cryptosporidium and coliforms were found in 22 samples and 25 samples were positive for both Cryptosporidium and Clostridium. Correlation incidence of Cryptosporidium with Clostridium was found to be 100% in handpump water samples and 90% in submersible water and in rest of the water samples taken from different sources, correlation ranged from 66.7% to 77.8%. Overall results indicated the higher correlation between Cryptosporidium and Clostridium i.e 83.3% as compared to that between Cryptosporidium and coliforms i.e. 73.3% (Table 2). In an epidemiological study of water contaminants done by Briancesco and Bonadonna (2005) in Italy, revealed a correlation between the incidence of Cryptosporidium and Clostridium and found that Cryptosporidium has to be determined in water intended for human consumption if Clostridium is detected. The coliform bacteria are used as indicators of faecal contamination and also correlated with the possible presence of disease causing protozoans. Even the presence of Clostridium also indicate the contamination with protozoans like Giardia and Cryptosporidium (Tyagi et al 2006).

Results revealed high incidence of Cryptosporidium protozoa and microbial agents (coliforms and Clostridium) in water samples collected from handpumps > submersible pumps and municipal corporation taps, while filter water showed their lowest incidence. Correlation of incidence of Cryptosporidium with occurrence of coliformis and Clostridium was also observed.

Table 1. Comparative account of incidence of indicator protozoans and bacteria from water samples of Ludhiana district

S. No.	Source of water		No. of samples surveyed	Indicator protozoans (% Incidence)		Indicator bacteria (% Incidence)	
				Crypto-sporidium	Giardia	Coliforms	Clostridium
1.	Submersible Pumps	Rural	29	34.5	ND	42.4	41.9
		Urban	5	20.0	ND	33.3	66.7
		Total	34	32.4	ND	47.1	44.1
2.	Municipal Corporation Taps	Rural	20	30.0	ND	50	28.6
		Urban	28	25.0	ND	42.8	32.1
		Total	48	27.1	ND	45.8	35.4
3.	Water filters	Rural	7	42.8	ND	42.8	42.8
		Urban	4	ND	ND	50	50
		Total	11	27.3	ND	45.5	45.5
4.	Hand-pumps	Rural	7	42.8	ND	71.4	57.1
		Urban	Nil	--	--	---	--
	GRAND TOTAL		100	30	ND	48	41

ND- Not detected

Table 2. Correlation of incidence of Cryptosporidium with the occurrence of coliforms and Clostridium from different sources of water in Ludhiana district

Source of water	Total number of samples surveyed	Total number of non-potable samples	No. of samples positive with					% Correlation of Cryptosporidium with	
			Cryptosporidium	Coliforms	Clostridium	Cryptosporidium + Coliforms	Cryptosporidium + Clostridium	Coliforms	Clostridium
Submersible pumps	34	23	11	16	15	8	10	72.7	90.0
Municipal Corporation taps	35	23	9	15	12	6	7	66.7	77.8
Water filters	11	8	3	5	5	2	2	66.7	66.7
Hand-pumps	7	6	3	5	4	2	3	66.7	100.0
Total	100	60	30	48	41	22	25	73.3	83.3

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