

## Detection of Some Water Borne Zoonotic Pathogens in Untreated Ground Water and its Impact on Human and Animal Health in Kalyoubia Province (Rural Areas)

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**Abstract:** The present study was carried out to evaluate the frequency of water borne zoonotic pathogens in untreated ground water used by humans and animals in some rural areas at Kalyoubia province, also to isolate and identify some pathogenic microorganisms that have public health importance. A total of fifty untreated ground water samples (From hand pumps) were collected and examined for detection of bacterial and protozoal zoonotic pathogens, as well as one hundred and twenty fecal samples were collected from humans with enteric disease problems attended to health units of rural areas, moreover thirty fecal samples were collected from different animal species which were in contact with or drinking from water sources and were also analyzed for the above pathogens. The results recognized Salmonella spp, Shigella spp, Escherichia coli, Entamoeba histolytica and Giardia lamblia at percentages of 28.6%, 7.1%, 64.3%, 20% and 80% respectively from water; while they were detected with percentages 30%, 7.5%, 62.5%, 30% and 70% respectively in humans. Mean while, they were 25%, 12.5%, 62.5%, 0% and 100% respectively in different animal species. The most predominant serotypes of Salmonellae were S.typhimurium and S.enteritidis. Shigella dysenteriae is the only isolated serotype of Shigella species, meanwhile, the most predominant serotypes of *E. coli* were O<sub>128</sub>: K<sub>67</sub> (B<sub>12</sub>), O<sub>111</sub>: K<sub>58</sub> (B<sub>4</sub>) O<sub>119</sub>: K<sub>69</sub> (B<sub>14</sub>) and O<sub>126</sub>: K<sub>71</sub> (B<sub>16</sub>) in all examined samples. The clinical complaints related to enteric diseases were more evident in females (56%) than males (44%) and the majority of cases existed among children less than 5 year. It was apparent that the majority of people who reported enteric disease problems used pit (non-piped) as sewage disposal system (58%) and with a history of consumption of stored water (60%). In addition to the main symptoms reported by patients were diarrhea (30%), followed by fever (22%), abdominal cramps (20%), vomiting (16%) and nausea (12%). The results confirm human bio-hazards through rural water supplies and reflected the need of public health education toward the accurate use of drinking ground water only after perfect treatment. The public health importance of pathogens was discussed and suggestive recommendations were recorded.

**Key words:** Untreated Ground Water • Zoonotic Pathogens • Public Health • Rural Areas • Egypt

### INTRODUCTION

Water is an important resource that supports life of all living organisms and is used for different purposes including drinking, cooking, washing and crop cultivation. Polluted water is an important vehicle for spread of diseases. Most outbreaks of water borne diseases are caused by the contamination of drinking water with feces of infected animals or people [1].

Sharing of water sources between humans and live stock is common in rural areas in developing countries and this may be associated with contamination of water. Uses of water sources contaminated with human and animal excreta containing pathogenic organisms pose health risks to the population because of increased incidence of zoonotic water borne diseases [2]. The ground water represented by means of wells is often of better quality than surface water. This is true only if

the soil or rock is fine grained and doesn't have cracks, crevices and bedding plants, which permit free passage of polluted water [3], but ground water in some areas becomes contaminated by fecal material of human and animals. Fecal contamination can reach the ground water from the land surface or from under ground sources of contamination. Major surface sources include feces from animals and other livestock operations, improperly constructed sanitary landfills where trash and garbage are disposed, seepage from contaminated lakes or shallow ponds, but the under ground sources, such as improperly functioning septic tank systems, underground reservoirs for liquid household sewage (cesspools), or leaking under ground sewer lines [4].

Many places in rural areas of Egypt depend on ground water of domestic water supply and irrigation. Most people in rural areas receive water through hand pumps without adequate protection or effective treatment and don't adhere to good hygienic measures of disease prevention such as use of latrines and boiling of drinking water, so there is increased predisposition to infection with water borne diseases such as Amebiasis, Salmonellosis, Giardiasis, Colibacillosis, .... etc [5].

For people who get sick from water borne diseases, the symptoms vary depending on the infectious agent. For many water borne diseases, symptoms begin from two to ten days after drinking the contaminated water and may include diarrhea, stomach cramp, nausea, vomiting and fever [6].

This study was conducted in different rural Egyptian localities in Kalyoubia province after considering that unsafe drinking water is a risk factor of gastro enteritis and that rural communities have a larger percentage of unsafe water supply and unimproved sanitation facilities compared to urban communities. The objectives of this study were intended to detect some zoonotic water borne pathogens in untreated ground water, animals and humans and to assess the public health risks associated with drinking, using or sharing of water sources between animals and humans, in addition to conclude the available preventing measures to overcome such problem.

## MATERIALS AND METHODS

The present study was carried out in the laboratory of zoonoses department, faculty of veterinary medicine, Benha university, Egypt.

### Collection of Samples

**Water Samples:** A total of fifty untreated ground water samples (1 liter each) was collected using sterile bottles

under complete aseptic precautions from different hand pumps (27-30 meters deep) that are used by humans and animals and located at different rural localities in Kalyoubia province. Each sample was labeled, identified and transferred as soon as possible to laboratory for examination.

**Human Samples:** A total of 120 fresh stool specimens was collected from persons, attended to health units of the same rural localities and were suffering from gastro intestinal illness or enteric problems and the food histories revealed no common food ingested or exposure but all persons had drunk water. A series of questionnaires were administered to all persons including (sex, age, consumption of water as soon or stored, disposal facilities and occurrence of enteric problems including diarrhea, vomiting, fever, nausea and abdominal cramps). The stool specimens were collected in clean sterile disposable plastic containers, labeled and transferred as soon as possible for examination.

**Collection of Animal Samples:** Fecal samples were randomly collected from 30 different animal species (cattle, sheep, goats and dogs) which live in contact or drink from the same water sources. The samples were labeled and transferred as soon as possible for examination.

### Laboratory Analysis of Water Samples

**Bacteriological Analysis of Water Samples:** This was done according to APHA [7] as 100 ml of each water sample were filtered through membrane filters (0.45 µm pore size) and these membrane filters were inoculated onto the surface of plates of different selective and differential media then identified chemically according to schemes described by Finegold and Martin [8] and Quinn *et al.* [9] and serological identification according to Edwards and Ewing [10] which was carried out at Food Analysis Lab. (Fac. vet. Med. Benha Univ.).

**Parasitological Analysis of Water Samples:** Enteric protozoa (Giardia and Entamoeba cysts) were demonstrated from water samples on the basis of their morphology according to Levine [11], Fleck and Moody [12] and Garcia and Bruckner [13].

**Laboratory Analysis of Human and Animal Samples:** Stool specimens collected from humans and fecal samples collected from animals were examined bacteriologically and parasitologically according to schemes as mentioned previously for detection of enteric pathogens.

**RESULTS AND DISCUSSION**

The data presented in Table (1) clarified types and percentages of the isolated zoonotic pathogens from examined water samples. It is evident that the bacterial pathogens were detected with a percentage of 28%. Regarding types of isolated pathogenic bacteria, Salmonella spp. Shigella spp. and E.coli were representing 28.6%, 7.1% and 64.3% respectively of bacterial pathogens. These results substantiate what had been reported by Barwick *et al.* and Kotb *et al.* [14, 15]. The detection of these bacterial pathogens in water may be due to fecal contamination from human being in rural areas where use of latrines is low. It may also be due to contamination by animal excreta through sharing of water sources contamination by run off from catchments and grazing areas near the water sources, perhaps may be a pathway exists between land surface and the aquifer in addition to an illegal sewage disposal site on the center of some villages was uncovered, where untreated sewage from residential homes had been dumped into sinkhole leading to the ground water aquifer and polluted water. This provides some indications that there is a risk for people living in these areas to acquire these pathogens from water sources used for domestic purpose and live stock [2]. The primary risk of consuming untreated water is the transmission of communicable diseases, however, the water which is not suitable for drinking, may be usable

for irrigation or for other domestic purposes. Thus it can be seen that each use of water imposes its own limits on the degree of pollution that can be considered acceptable [3]. Concerning the pathogenic protozoa detected in water, E. histolytica and G. lamblia represented 20% and 80% respectively of protozoal organisms. These results were in accordance with Salata and Aucott [16] who recorded that transmission of Amebiasis, Giardiasis included water borne route. Moreover, parasitic infestation might be due to the contamination of ground water in rural localities via human and animal excreta and wastage. Also, E. histolytica and G. lamblia are both persistent environmentally and extremely resistant to chlorine [15].

The presence of these protozoa in water albeit at low prevalence, coupled with the observation that most of people don't boil drinking water, suggests that the parasites may be play an important role in diarrheic syndromes in human beings [17].

Results recorded in Table (2) illustrated the percentage distribution of bacterial pathogens among humans and domestic animals, they were detected with a percentage of 33.3% and 26.7% respectively. Salmonella spp, Shigella spp and E. coli were representing 30%, 7.5% and 62.5% respectively of bacterial pathogens among humans and they were 25%, 12.5% and 62.5% respectively of bacterial pathogens among domestic animals. Regarding to protozoal pathogens, the

Table 1: Types and percentages of isolated zoonotic pathogens detected in untreated ground water

No of water samples	Types of pathogens	No. of positive	%	Strains	No
50	Bacteria				
	Salmonella spp.	4	28.6	S. typhimurium	3
				S. enteritidis	1
	Shigella spp.	1	7.1	Sh. dysenteriae	1
	E. coli	9	64.3	O <sub>128</sub> :K <sub>67</sub> (B <sub>12</sub> )	2
				O <sub>111</sub> :K <sub>38</sub> (B <sub>4</sub> )	3
				O <sub>119</sub> :K <sub>69</sub> (B <sub>14</sub> )	2
				O <sub>126</sub> :K <sub>71</sub> (B <sub>16</sub> )	2
	Total	14	28		
	Protozoa				
Entamoeba	1	20	E. histolytica	1	
Giardia	4	80	G.lamblia	4	
Total	5	10			

Table 2: Summarized results of isolated zoonotic pathogens from humans and animals

Isolated from	No. of samples	Bacteria								Protozoa					
		Salmonella spp.		Shigella spp.		E. coli		Total		E. histolytica		G. lamblia		Total	
		No	%	No	%	No	%	No	%	No	%	No	%	No	%
Humans	120	12	30	3	7.5	25	62.5	40	33.3	3	30	7	70	10	8.3
Animals	30	2	25	1	12.5	5	62.5	8	26.7	0	0	3	100	3	10

Table 3: Types and frequency of isolated zoonotic pathogens from humans and animals

Isolates	strain	Humans	Animals
Salmonella	S.typhimurium	7	1
	S. enteritidis	5	1
Shigella	Sh. dysenteriae	3	1
E. Coli	O <sub>128</sub> : K <sub>67</sub> (B <sub>12</sub> )	8	2
	O <sub>111</sub> :K <sub>58</sub> (B <sub>4</sub> )	6	1
	O <sub>119</sub> : K <sub>69</sub> (B <sub>14</sub> )	7	2
	O <sub>126</sub> : K <sub>71</sub> (B <sub>16</sub> )	4	0
E. histolytica	E. histolytica	3	0
G. lamblia	G.lambliia	7	3

corresponding percentages 8.3% and 10%.E. histolytica and G.lambliia were representing 30% and 70% among humans and 0% and 100% among domestic animals respectively. These results were similar to those mentioned by Kusiluka *et al.* and Haggag *et al.* and Lobna and Nagwa [17-19] who recorded that the importance of animals in spreading or contamination of water with intestinal pathogens. Concerning the identification of the common bacterial pathogens isolated from water, humans as well as domestic animals, Table (1& 3) revealed that the most predominant serotypes of Salmonellae was S. typhimurium and S. enteritidis. While the most predominant serotype of Shigella was Sh. dysenteriae. Also the most predominant serotypes of E. coli were O<sub>128</sub>: K<sub>67</sub> ( B<sub>12</sub>), O<sub>111</sub>: K<sub>58</sub> (B<sub>4</sub>), O<sub>119</sub>: K<sub>69</sub>: (B<sub>14</sub>) and O<sub>126</sub> K<sub>71</sub>: (B<sub>16</sub>). Such serotypes have a great pathogenicity in the intestinal tract and cause gastroenteritis and implicated in food poisoning out breaks [20].

The previously mentioned results showed that the bacterial pathogens were the same in water, humans and domestic animals. This probably indicates that there is cross infection of bacterial pathogens between water, humans and animals. As water treatment facilities were not available in rural areas and ground water wells were found to harbor multiple bacterial indicators of fecal contamination, implying that there is a risk of human infection if drinking water is not boiled. Moreover, people don't consistently use latrines, contamination of the environment with human or animal excreta is high.

The public health importance of E. coli has been emphasized by many authors as these enteropathogenic serotypes have been implicated in cases of gastroenteritis, sporadic summer diarrhea in children as well as in cases of food poisoning. Salmonellosis as a zoonotic disease has become a world wide problem and its incidence was increasing in both man and animals. Human salmonella infection is most commonly caused by drinking water contaminated by secretion and excretion of

diseased animals. The Signs of Salmonellosis in man include fever, headache, abdominal pain, nausea, vomiting, diarrhea, gastroenteritis and septicemia [21].

Regarding to parasitic pathogens of zoonotic importance, E. histolytica and G. lamblia were demonstrated in water, humans and domestic animals. The results confirm human bio-hazards through rural water supplies and reflect the need of public health education toward the accurate use of drinking ground water only after treatment. E. histolytica and G. lamblia have become significant water borne pathogens in the developed world as they are indigenous infection with low infectious dose; densities of environmental contamination with infective cyst is sufficient to pollute the aquatic environmental and cysts are small enough to penetrate water treatment processes, in addition to less sensitive to commonly disinfectants used in water treatment [22].

Table (4) demonstrated the frequency distribution of detected enteric pathogens (bacterial and protozoal) among 50 positive cases, it is evident that the occurrence of enteric disease problems was more in female (56%) than in males (44%). Similar findings were reported by Kusiluka *et al.* and Payment *et al.* and El- Sherbini [17, 23, 24]. It appears to be associated with more active and out door wandering nature of women as they are responsible for obtaining water for family. Concerning the age distribution among 50 cases, 15 were less than 5 years, 12 were of 6-20 years old, 11 were of 21-49 years old and 12 were 50 years old or more with the corresponding percentages of 30%, 24%, 22% and 24%. These findings indicated that all age groups were affected with enteric disease problems and slightly higher in less 5 years age group. This may be attributed to the poor defense mechanisms against infection in this age group.

Regarding water consumption, it was clear that infection among people who stored water is higher (60%) than people who used or consumed water as soon as in the same day (40%). This may be explained that the stored water could be contaminated at several points either from

Table 4: Demographic and clinical informations of 50 positive cases

Characters	Exam. No	Positive cases	
		No	%
Sex			
Males	60	22	44
Females	60	28	56
Total	120	50	41.7
Age			
<5 years	32	15	30
6-20	30	12	24
21-49	28	11	22
50+	30	12	24
Total	120	50	41.7
Water consumption			
In the same day	80	20	40
stored	40	30	60
Total	120	50	41.7
Sewage System			
Pipe	70	21	42
Pit	50	29	58
Total	120	50	41.7
Symptoms			
Diarrhea	30	15	30
Fever	27	11	22
Abdominal cramps	25	10	20
Vomiting	22	8	16
Nausea	16	6	12
Total	120	50	41.7

source contamination, excreta of animals and birds freely living in houses, presence of rodents or arthropods especially flies which acting as mechanical vector of organisms [25]. It was apparent that the majority of people (58%) who reported enteric disease problems used pit (non-piped) as sewage disposal system and in such case it could actually have adverse effect on the underground water. These observations could explain the persistence of enteric disease problems in the studied areas and this is agree with El- Sherbini [24] who recorded that the large proportion of children dying from enteric diseases lived in houses without latrines or without hygienic sewage disposal.

It is noticed that the main symptoms reported by the patients were diarrhea (30%), followed by fever (22%), abdominal cramps (20%), vomiting (16%) and lastly nausea (12%). This is in agreement with that reported by O'Reilly *et al.* and Kusiluka *et al.* and Kakakhel *et al.* [6, 17, 26]. It is evident that contamination of water sources with human and animal excreta is known to have serious consequences on public health and the clinical symptoms may be associated with water borne diseases, with the most common ones being diarrhea, vomiting, nausea and fever [27].

On the basis of our findings, It could be concluded that the untreated ground water samples (from hand pumps) are exposed to different sources of pollution and the consumers in the villages receiving water through hand pumps are often exposed to risk of water borne disease due to inadequate treatment, so that water constitutes a variety of disease hazards to man and animals. Wells (hand pumps) for public water supplies must be located as far as possible from all sources of pollution including septic tanks, drainage system, barns, human dwellings and other activities that responsible for biological contamination of water sources. Strict hygienic measures should be applied to improve water quality and to avoid deleterious effects on the public health and safe guard the water user against infection. A adequate sanitation, proper disposal facilities and raise awareness among the residents about good personal hygiene practices should be emphasized a long with the need for safe water supply in any preventive program employed to reduce the incidence of enteric disease problems in rural communities as majority of people were not aware of water borne zoonoses and non of the villages had a comprehensive and specific public health program addressing issues pertaining to water borne zoonoses.

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