

## Drinking water quality and public health in Southwestern Saudi Arabia: The need for a national monitoring program

Jobran M. Alqahtani, Ahmed M. Asaad<sup>1</sup>, Essam M. Ahmed<sup>1</sup>, Mohamed A. Qureshi<sup>2</sup>

Departments of Pediatric and <sup>1</sup>Microbiology, College of Medicine, <sup>2</sup>Department of Microbiology, College of Applied Medical Sciences, Najran University, Najran, Kingdom of Saudi Arabia

**Address for correspondence:** Dr. Ahmed Morad Asaad, College of Medicine, Najran University, P.O. Box 1988, Najran, Kingdom of Saudi Arabia. E-mail: ahmedmoradasaad@hotmail.com

ABSTRACT

**Aim of the Study:** The aim was to investigate the bacteriological quality of drinking water, and explore the factors involved in the knowledge of the public about the quality of drinking water in Najran region, Saudi Arabia. **Study Design:** A cross-sectional descriptive study. **Materials and Methods:** A total of 160 water samples were collected. Total coliforms, fecal coliform, and fecal streptococci were counted using Most Probable Number method. The bacterial genes *lacZ* and *uidA* specific to total coliforms and *Escherichia coli*, respectively, were detected using multiplex polymerase chain reaction. An interview was conducted with 1200 residents using a questionnaire. **Results:** Total coliforms were detected in 8 (20%) of 40 samples from wells, 13 (32.5%) of 40 samples from tankers, and 55 (68.8%) of 80 samples from roof tanks. Twenty (25%) and 8 (10%) samples from roof tanks were positive for *E. coli* and *Streptococcus faecalis*, respectively. Of the 1200 residents participating in the study, 10%, 45.5%, and 44.5% claimed that they depended on municipal water, bottled water, and well water, respectively. The majority (95.5%) reported the use of roof water tanks as a source of water supply in their homes. Most people (80%) believed that drinking water transmitted diseases. However, only 25% of them participated in educational programs on the effect of polluted water on health. **Conclusions:** Our results could help health authorities consider a proper regular monitoring program and a sustainable continuous assessment of the quality of well water. In addition, this study highlights the importance of the awareness and educational programs for residents on the effect of polluted water on public health.

**Key words:** Coliforms, drinking water, fecal streptococci, *Pseudomonas aeruginosa*, polymerase chain reaction, wells

### INTRODUCTION

The quality and safety of drinking water remain as an important public health issue. Contamination of drinking water has frequently been blamed for the transmission of infectious diseases that have caused serious illnesses with associated mortality worldwide.<sup>[1,2]</sup> Each year, an estimated 1.9 million deaths, primarily of children under 5 years of age, result from unsafe drinking water and

inadequate sanitation and hygiene.<sup>[3]</sup> The WHO estimates that improving water, sanitation, and hygiene could prevent approximately 9.1% of the global burden of disease and 6.3% of all deaths.<sup>[4]</sup> One of the targets of the WHO Millennium Development Goals is reducing by half the proportion of people without sustainable access to safe drinking-water and basic sanitation by 2015. At the end of 2010, 783 million people still lacked access to improved water sources, and over 2.5 billion people had no access to basic sanitation.<sup>[5]</sup>

Water may be contaminated in many ways. The different forms of contamination originate from a variety of sources and are dealt with in different ways. The three main forms of water contamination are physical, bacterial, and chemical.<sup>[6]</sup> However, microbiological quality is the most important aspect of drinking water with respect to waterborne diseases. Bacteriological

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assessments, particularly for total coliforms, fecal coliforms, and fecal streptococci are recognized as the main indicators of the presence of pathogenic enteric bacteria in water sources.<sup>[7]</sup>

In the past century, the evaluation of the bacteriological quality of drinking water has been performed through the analysis of fecal pollution indicators in finished drinking water, which is expected to predict the potential presence of pathogenic microorganisms in the water.<sup>[8]</sup> However, previous studies have reported cases in which the indicators have been present in the water when it was served to consumers.<sup>[2,4,8,9]</sup> Therefore, WHO has developed several guideline documents for the sole purpose of checking the quality of finished drinking water.<sup>[10,11]</sup>

Groundwater is still and will continue to be the main source of safe and reliable drinking water in arid regions like Saudi Arabia, where surface water is scarce, rainfall is irregular and rates of evaporation are very high. Hence, groundwater is a key source for urban and rural supplies and is considered the only source to meet domestic and agricultural needs in towns and villages.<sup>[12]</sup> In 2003, there were 5661 government wells and 106,370 multipurpose private wells assigned for municipal purposes.<sup>[13]</sup> Currently, there is a heavy reliance on tube-wells to meet the daily demands in most Saudi regions. The contribution of wells toward satisfying the water demand is on the high side and should be reduced in order to promote a sense of better water management in the Kingdom of Saudi Arabia.<sup>[14]</sup> In Najran, which is located in Southwestern of Saudi Arabia, many wells scattered throughout the region are used as the source of drinking water, but this water is untreated. The chances for poor sanitary conditions to evolve are high because of the large number of small units that produce drinking water, spillage from the tankers that transport the water and distribute it to houses and the improper drainage system.<sup>[15]</sup>

The aims of this study were: (1) To investigate the quality of drinking water from source (including wells, tankers, and house tanks) to the point-of-use in Najran region, Southwestern Saudi Arabia, using bacteriological and molecular techniques, and (2) To explore the factors involved in public knowledge, self-described behaviors, and perception of the quality and risks of drinking water in the study area. The findings could be useful in deciding appropriate remedial measures for preventing contamination of drinking water and help as a basis for decisions on water health policy at different administrative levels in different Saudi regions.

## MATERIALS AND METHODS

Water samples were collected in accordance with the standard methods for the examination of water and wastewater.<sup>[16]</sup> A total of 160 water samples (40 samples from wells, 40 samples from tankers and 80 samples from roof tanks) were collected from both urban and rural areas of the Najran region, Southwestern Saudi Arabia, from October, 2012 to June, 2013. Samples were collected in 250 ml sterile glass bottles. They were kept in ice boxes and sent to the Microbiology Department of the Najran University College of Medicine for bacteriological and molecular examination.

Total coliforms, fecal coliforms, and fecal streptococci were counted using Most Probable Number method as previously described.<sup>[16]</sup> Coliform bacteria were determined by incubation of samples into tubes of lactose broth (Difco Laboratories, Detroit, USA) at 35°C for 48 hrs. Fecal coliforms were detected by subculture into brilliant green bile broth 2% (Difco) and incubation at 37°C for 24–48 hrs. Positive samples were further inoculated into *Escherichia coli* broth (Difco) and incubated at 44°C for 24–48 hrs. *E. coli* was identified by the standard biochemical tests. Fecal streptococci were detected by inoculation of water samples into azide dextrose broth (Difco) and incubation at 37°C for 24–48 hrs.

The detection of 876-bp and 147-bp target sequences within the *lacZ* and *uidA* genes specific to coliform bacteria and *E. coli*, respectively, was performed by multiplex polymerase chain reaction (PCR) using specific primers (Qiagen, USA), as previously reported.<sup>[17]</sup>

The sequences of the primers were 5'-ATGAAAGCTGGCTACAGGAAGGCC-3' and 5'-CACCATGCCGTGGGTTTCAATATT-3' for *lacZ* gene and 5'-TGGTAAATTACCGACGAAAACGGC-3' and 5'-ACGGGTGGTTACAGTCTTGCG-3' for *uidA* gene. Purification of bacterial DNA directly from water samples was achieved using a Genomic DNA purification kit (Fermentas, Germany), according to the manufacturer's instructions.

The PCR amplification mixture consisted of pure Taq ready-to-go PCR beads (Amersham Bioscience, UK), 25 pmol/μl of each primer and 10 ng of each DNA extract in a total volume of 25 μl. The amplification was performed using a thermal cycler (Cyclogene, Techne, UK). The reaction mixtures were heated to 94°C for 10 min, followed by 36 amplification cycles, each consisting 60 s at 94°C, 60 s at 55°C, and 60 s at 72°C. A final extension cycle of 72°C for 10 min was included. The amplified products were electrophoresed through 1.5% agarose gels,

which were stained with ethidium bromide and visualized under an ultraviolet transilluminator (Cole-Parmer, USA). The presence of obvious bands of 876-bp or 147-bp was considered a positive result.

The total viable bacteria (TVB) were enumerated using the TVB pour plate count method.<sup>[16]</sup> All water samples were first inoculated into water plate count agar media (Oxoid, Hampshire, UK). Two sets of plates were used for all samples. One set was incubated aerobically at 37°C for 48 hrs and the other set at 22°C for 72 hrs. All colonies were counted as colony forming unit per ml of the water sample. Then, the suspected colonies were sub-cultured into MacConkey and blood agar media and incubated at 37°C for 24 hrs. The organisms which were presumed to be pathogenic bacteria were identified at the genus and/or species level by Gram-staining, culture characters, and biochemical tests using API 20S, API 20E, and API 20NE (BioMerieux, Marcy l'Etoile, France).

The target population was the residents of Najran region. The sample size was determined in order to have 95% confidence limits of 5% maximum error of the estimate.<sup>[18]</sup> The minimal sample size required for the study was calculated as 1090 residents to represent the entire population on a statistical basis. To avoid a no-response expectation, the sample size was increased to 1200 residents. The questionnaire included questions related to the following: Personal profile of the study population (age, occupation, and level of education), various aspects of domestic water supply for the people who live in the study area (source of drinking water and age of water network and the use of roof water tanks and information about them), situation of wastewater networks system (connection to sewage network, age of sewage network in the area and seasons of sewage flood) and knowledge of the study population of drinking water contamination in Najran region.

## RESULTS

The conventional culture technique showed that 6 (15%) of 40 samples from wells, 12 (30%) of 40 samples from tankers, and 50 (62.5%) of 80 samples from roof tanks were positive with total coliforms, while 18 (22.5%) and

8 (10%) samples from roof tanks were positive for *E. coli* and *S. faecalis*, respectively. The results of the PCR technique showed that the *lacZ* gene specific to total coliforms was detected in 8 (20%), 13 (32.5%) and 55 (68.8%) samples from wells, tankers and roof tanks, respectively. The PCR amplification of *uidA* gene specific to *E. coli* was positive in 20 (25%) samples from roof tanks [Table 1].

The number and percentages of isolated organisms from water samples are presented in Table 2. *Pseudomonas aeruginosa* was the most frequently (32.1%) isolated organism followed by *E. coli* (23.8%), *Klebsiella* spp. (9.5%), *S. faecalis* (9.5%), *S. aureus* (5.9%), and *S. epidermidis* (5.9%).

In this study, the mean age of participants was  $32.8 \pm 8.9$  years. More than half of those interviewed (57%) had a university degree, indicating a well-educated community. The response of the study population on various aspects of drinking water supply is summarized in Table 3. Only 120 (10%) participants said that they drank municipal water. However, 546 (45.5%) claimed that they depended on bottled water followed by 534 (44.5%) who depended on well water. Most people (95.5%) reported that the source of their water supply in their homes was roof water tanks and the majority (89.5%) of them used white plastic tanks. Although 60.7% of interviewees saw sediments in the tanks, 42% of people did not clean them. Most people (67.5%) said that their homes were not connected to the sewage network system. The majority (70%) of people did not know how old the sewage network was. All the study participants reported that the sewage got flooded, and 44% said that the flood occurred in the summer while 44% reported flooding in both summer and winter. Most people (80%) believed that drinking water transmitted diseases. Furthermore, 66% of the people believed that this was true of roof tanks. However, about half (50.5%) of the people thought that the water in Najran was suitable for drinking. Only 25% of the people participated in educational programs on the effect of polluted water on health [Table 3].

## DISCUSSION

Detection of bacterial indicators in drinking water suggests the presence of pathogenic organisms that are the source

**Table 1: Bacteriological and molecular analysis of the different drinking water sources in Najran**

Sample source (n)	Total coliforms (%)		<i>Escherichia coli</i> (%)		<i>Streptococcus faecalis</i>
	Culture positive	PCR positive	Culture positive	PCR positive	
Wells (40)	6 (15)	8 (20)	0 (0)	0 (0)	0 (0)
Tankers (40)	12 (30)	13 (32.5)	0 (0)	0 (0)	0 (0)
Roof tanks (80)	50 (62.5)	55 (68.8)	18 (22.5)	20 (25)	8 (10)

PCR: Polymerase chain reaction

**Table 2: Number and percentage of isolated organisms from different water sources in Najran**

Organisms	Wells	Tankers	Roof tanks	Total (%)
<i>Pseudomonas aeruginosa</i>	5	7	15	27 (32.1)
<i>Escherichia coli</i>	0	0	20	20 (23.8)
<i>Klebsiella</i> spp.	2	1	5	8 (9.5)
<i>Aeromonas</i> spp.	0	2	1	3 (3.7)
<i>Acinetobacter</i> spp.	0	0	2	2 (2.4)
<i>Alcaligenes</i> spp.	0	1	3	4 (4.8)
<i>Salmonella</i> spp.	0	0	2	2 (2.4)
<i>Staphylococcus aureus</i>	1	1	3	5 (5.9)
<i>Staphylococcus epidermidis</i>	0	1	4	5 (5.9)
<i>Streptococcus faecalis</i>	0	0	8	8 (9.5)

of water borne diseases.<sup>[8]</sup> In this study, the results of total coliform count showed that 20% of the samples from wells exceeded the guideline values recommended by national and international standards of drinking water.<sup>[11,19]</sup> In a previous large investigation of the quality of water samples from 1062 wells from seven regions in Saudi Arabia, fecal streptococci were detected in 8% of samples.<sup>[20]</sup> In another recent study which evaluated the bacteriological characteristics of drinking water in Khamis Mushait Governorate, Southwestern Saudi Arabia, fecal coliform, and fecal streptococci were detected in 87.9% and 57.6% of 33 well water samples.<sup>[12]</sup> Our result was expected since the wells are not given any chlorine disinfection treatment before the water contained is consumed. The presence of coliform may be attributed to contamination of the hoses used by humans, including farmers and livestock owners; and the exposure of these delivery hoses to dust storms.<sup>[21]</sup> Previous studies have indicated that dust storms and livestock activity in the vicinity of surface wells increase microbial levels and bacterial input.<sup>[22-24]</sup> Our finding is worrisome, as well water is still the main source of drinking water in Najran. According to the responses to the questionnaire, 45% of interviewees depend on well water for drinking.

In this study, 33% of the water samples from tankers had higher total coliform than stipulated by the national and international guideline values.<sup>[11,19]</sup> In a previous study in Shebaa area, Southwestern Saudi Arabia, only 2.6% of 39 water samples from tankers was positive for total coliform. The investigators concluded that transportation of desalinated water by water tankers had not significantly contributed to its contamination in their region.<sup>[25]</sup> However, Mihdhir reported that 68.8% and 37.5% of samples from tankers in Makkah Al-Mokarama were positive for total coliform and fecal coliform, respectively.<sup>[15]</sup> Poor microbial quality of community tanker water in our study is likely to be due to the presence of biofilm, contamination of dispensing devices from tankers, contamination of water in the tanker by dust during transportation and the lack of or

**Table 3: Answers to a survey testing the knowledge of the population on various aspects of drinking water in Najran**

Variable	n (%)
Source of drinking water	
Bottled water	546 (45.5)
Municipal water	120 (10)
Well water	534 (44.5)
Use of roof tanks	
Yes	1146 (95.5)
No	54 (4.5)
Types of tanks	
Plastic white	1074 (89.5)
Plastic black	126 (10.5)
Cleaning of water tanks	
Yes	612 (51)
No	588 (49)
Settlements observed	
Yes	696 (58)
No	504 (42)
Connected to network	
Yes	384 (32.5)
No	810 (67.5)
Age of sewage network	
1-3 years	36 (3)
3-5 years	120 (10)
>5 years	204 (17)
Do not know	840 (70)
Sewage flooding	
Summer	528 (44)
Winter	144 (12)
Summer and winter	528 (44)
Do you think drinking water transmit diseases	
Yes	960 (80)
No	240 (20)
Do you think roof tanks water transmit diseases	
Yes	792 (66)
No	408 (34)
Do you think that the water in Najran is suitable for drinking	
Yes	606 (50.5)
No	594 (49.5)
Have you attended educational program on health impact of polluted water	
Yes	300 (25)
No	900 (75)

inadequate water treatment.<sup>[26]</sup> The hygiene of water tankers is also of concern. Communal tankers and their interiors are seldom washed and scrubbed because it is difficult to access their interior. In addition, water is stored in the tanker for long hours thus promoting biofilm formation and development.<sup>[27]</sup>

Bacteriological analysis of water samples from roof tanks in this study revealed higher levels of bacterial indicators in many samples than the national and international guideline



values.<sup>[11,19]</sup> Abu-Zeid *et al.* found that 26.4% of 201 samples from house tanks showed contamination. The investigators suggested that water contamination obviously occurred during storage in house reservoirs, and was possibly implicated, at least partly, in the increased prevalence of diarrhea among residents in the Shebaa area.<sup>[25]</sup> Similarly, our results indicated that the water gets more deteriorated at the point-of-use than at the source. This could be a result of biofilm growth in the household tanks.<sup>[26]</sup> The use of roof tanks for water storage is a common practice in Najran region as stated by most (96%) of interviewees. It is disturbing that 58% of people in this study stated that they had observed sediments in their roof tanks. The lack of cleaning as admitted by 49% of the interviewed people may contribute to water contamination. In many previous studies, diarrhea was strongly associated with the cleaning of water tanks.<sup>[28,29]</sup> A finding of considerable concern in this study is flooding the sewage in winter and summer. This could be the cause of the infiltration of wastewater which in turn may contribute to microbial contamination of water in the wells and house tanks. To maintain the quality of drinking water in roof tanks as received from the source, it would be necessary to implement effective awareness and educational programs. This is especially important for Najran residents. Although most people in this study reported that drinking water and water from roof tanks transmitted diseases, less than one-third of those interviewed had attended any educational program on the effects of polluted water on health. These awareness programs are supposed to show the importance of keeping house tanks closed, hand washing before handling areas close to the nozzle of the hose, cleaning of tanks on a regular basis, and possibly the addition of small amounts of chlorine into the water stored in roof tanks.

The study of the bacteriological quality of the samples of drinking water revealed that *P. aeruginosa*, *E. coli* and *Klebsiella* spp. accounted for 65% of all strains isolated. These genera are pathogenic, and their isolation might be important because of their contribution to water-borne infections.<sup>[29]</sup> Furthermore, the presence of *S. aureus*, *S. epidermidis*, and *Acinetobacter* spp. in water samples from tankers and roof tanks is an indication of hand contamination and inoculation from the human skin.<sup>[23,27]</sup> The contribution of bare hands and fingers to the contamination of drinking water has been emphasized in many previous studies.<sup>[12,14,23,27]</sup>

## CONCLUSIONS

Bacteriological contamination of water samples between the source and point-of-use in Najran region is widespread and highly alarming. The local health authorities should consider a proper regular monitoring program to continuously assess

the quality of well water. Some sustainable means should be found to prevent the deterioration of the quality of well water and eliminate health problems. Communal tankers should be thoroughly washed regularly, and chlorine levels monitored. Safer household water storage and treatment is recommended to prevent postcollection contamination. In addition, this study highlights the importance of the awareness and educational programs for residents on the effect of polluted water on public health.

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