Bacteriological and Physicochemical Assessment of Drinking Water Collected from the Private Selling Sites of Urban Areas of Khamis Mushait City-KSA

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Abstract: This study was conducted in the urban area of Khamis Mushait City, Aseer Region, and Saudi Arabia. A total of 50 samples of drinking water were collected from private selling sites of unbolted water in the urban area of Khamis Mushait city. Physicochemical parameters such as pH, total dissolved solids (TDS), and electrical conductivity (EC) of water samples were measured using standard methods to assess the quality of drinking water. The pH values of water samples ranged from 6.56±0.1 to 8.5±0.49, TSD changed from 104.7±0.28 to 202.4±1.7 mg/liter and EC from 163.6±0.45 to 316.3±2.61 µS/cm. The previous results indicated that all physicochemical parameters are within the permissible limits of WHO (2011) for drinking water quality. The bacteriological examination of water samples included the most probable number (MPN/100 ml) of coliforms, *E.coli* and *Pseudomonas aeruginosa*. The results showed that *E. coli* bacterium was not detected in all samples, while total coliform group was found in 7 samples out of 50 samples analyzed (14%) and *Pseudomonas aeruginosa* was found in 24 samples out of a total of 50 samples (48%). Furthermore, this study has revealed that about 54% of the water samples collected from private selling sites is incompatible to the World Health Organization (WHO) standards for drinking water quality.

Keywords: Khamis Mushait City, Aseer Region, coliforms, E. coli, Pseudomonas aeruginosa, (TDS), pH

INTRODUCTION

Clean-safe drinking water is required for the sustenance of life and it is a fundamental human need. Drinking water must be safe to drink or to use for food preparation. Water is essential to life; each person on earth requires at least 20 to 50 liters of clean safe water a day for drinking, cooking, and cleaning. But, normal healthy person needs to drink about 8 glasses (2 liters) of water per day, depending upon several factors, including gender, age, level of activity and environment. World Health Organization (WHO, 1996), has developed guidelines for drinking water quality, provides the basis for the development of national standards that, if properly implemented, will ensure the safety and quality of drinking water, as well as preparation of international standards on water quality and human health in a form of guidelines of regulations and standards that serve as the basis for organization and standard setting around the world.

Saudi Arabia is the world's largest producer of desalinated water which covers 70% of the total water demand (Ahmad and Bajahlan, 2009). The private selling sites in the urban areas of Khamis Mushait city provide the drinking water by tankers from the main source of government water desalination and sold it in a small twenty-liter reusable container to the consumers. Contaminate water, poor hygiene, and bad sanitation cause over 80% of diseases in developing countries (WHO, 1998). Many researchers such as Zacheus *et al.* (2001), Leguori (2010) and Rifaat *et al.* (2007) reported bacterial contamination of drinking water in developing and some developed countries. Detection of *E. coli* or enterococci is recommended for monitoring fresh water, whereas enterococci are the preferred indicator bacteria for marine waters because of their salt tolerance (USEPA, 2004).

In a previous study about the bacteriological assessment of urban water sources in Khamis Mushait City, Fecal coliform was detected in desalinated, surface water and well water with percentages of 3.23, 60.0, and 87.88, respectively (AlOtaibi E. L. Sh., 2009).

In general, organisms which are potential disease-producers are five types, bacteria, protozoa, worms, viruses, and fungi. The presence of certain organisms of these various types can lead to such infectious diseases as typhoid fever, dysentery, cholera, jaundice, hepatitis, guardians, undulant fever, and tularemia, as well as other diseases which spread through unfit drinking water. In addition to the presence of contaminants, other factors assessed when determining portability include taste, odor, and turbidity, or cloudiness. Some of these issues can be resolved as the water goes through processes such as settling, filtering, and disinfecting. Drinking water should be clear, not saline and free from compounds that can change color, taste, and odor. Presence of Coliform bacteria in drinking water indicates that disease-causing organisms (pathogens) could be in the water system. Most pathogens that can contaminate water supply come from the feces of humans or animals.

Pseudomonas aeruginosa is a common bacterium, Gram-negative opportunistic pathogen capable of infecting humans with compromised natural defenses and causing severe pulmonary diseases, occurs widely in the environment such water, soil, sewage, animal feces, and on vegetation, also occurs to many foodstuffs and may often be present in the digestive tract of humans without causing any signs of illness. *Pseudomonas aeruginosa* is a waterborne opportunistic pathogen which may have impacts on human health, especially in immune-compromised populations (Wang *et al.,* 2012).

pH value is an important factor in maintaining carbonate and bicarbonate levels in water whereas, total dissolved solids (TDS) is used to describe the

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inorganic salts and small amounts of organic material present in water (WHO, 1996).

The Main purposes of the present study are to investigate the bacteriological and physicochemical quality of drinking water sold in the urban area of Khamis Mushait City, which provide us good information about the safety of this water. Furthermore, the study expected to provide important and valuable information to Khamis Mushait Municipality about the extent of pollution that facing the private selling sites of drinking water. In addition, can also help devise long-term strategies to improve water quality.

MATERIALS AND METHODS

Location of Study area:

Khamis Mushait is one of an important city found in Aseer Region, Saudi Arabia. It is located 18.30 latitude and 42.73 longitudes and it is situated at elevation 1998 meters above sea level. Khamis Mushait has a population of 387,553 making it the biggest city in Aseer region.

Water samples were collected from an urban area of Khamis Mushait city. The urban area was divided into five geographic regions (Central, Eastern, Western, Southern, and Northern regions) in terms of the distribution of private water selling points (Figure-1).

Collection of water samples:

The study was conducted from September to October 2018. The Samples of the drinking water were collected from the urban area of Khamis Mushait city covering, Northern region (9 samples), Western region (7 samples), Southern region (11 samples), Eastern region (13 samples) and Central Region (10 samples) Table (1)

Table (1): The number of drinking water samples taken from sites in each region

Geographic Region	Number of drinking water sites
Northern Region	9
Southern Region	11
Western Region	7
Northern Region	9
Eastern Region	13
Total samples	50

A total of 50 drinking water samples were collected and analyzed for bacteriological (Total coliform, *E. coli*, and *Pseudomonas aeruginosa*) and physicochemical parameters (pH, TDS, EC), following standard procedures to avoid any contamination. The samples were collected in clean sterile glass bottles with screw caps under aseptic conditions (Bottles for bacteriological analysis contain sodium thiosulfate to neutralize any residual disinfectant). After collection, the water samples immediately kept on the ice-box, and transported to the Food Safety and Environment laboratory of Khamis Mushait Municipality, Saudi Arabia, and preserved at 4°C until analysis within 3 to 6 hours.

Bacteriological Analysis:

Enumeration of Total Coliforms and *E. coli*:

The most probable number (MPN) technique was followed as described in the Standard Methods for the Examination of Water and Wastewater (APHA, 2000) by using the Colilert 18 medium (IDEXX Laboratories, Westbrook, ME, USA-2015) with appropriate dilutions. 100 ml of water sample was mixed with one snap of colilert 18, the mixture poured into the Colilert tray - Quanti-Tray/2000, and sealed within an IDEXX Quanti-Tray Sealer. The sealed Quanti-tray was incubated at 35°C±0.5 for 18-24 hours. A yellow color after incubation was considered as a positive total coliform while the wells with yellow color gave fluorescence under UV illumination (366 nm) was considered as *E. coli* positive. The number of positive wells in each Quanti-Tray/2000 was counted and the corresponding most probable (MPN) was obtained from the MPN table provided with the Quanti Tray/2000. Results of each sample were calculated and reported as MPN/100ml (IDEXX Laboratories, 2013).

Enumeration of *Pseudomonas aeruginosa*:

One hundred milliliters of water sample was added to a sterile 120 mL vessel containing an antifoam reagent (ISO 16266-2:2018). One snaps pack of Pseudalert® reagent was added, the vessel capped and the sample was shaken to dissolve the reagent before being left to stand for any foam to settle. The sample was then poured into a Quanti-Tray/2000 (IDEXX Laboratories, USA-2013), sealed and incubated at 38±0.5°C for 24–28 hours. After incubation, the Quanti-Trays® were examined under UV irradiance (365 nm), and all wells demonstrating blue fluorescence compared to a negative blank sample were counted as positive for *Pseudomonas aeruginosa*. Results were reported as MPN/100ml using the table provided with Quanti-Tray/2000, (IDEXX Laboratories, USA-2013).

Physicochemical Analysis:

Physicochemical parameters such as pH, total dissolved (TDS), and electrical conductivity (EC) of the drinking water samples were measured using standard methods as described in the APHA (1998).

Determination of pH:

The pH values of the water samples was determined by using a digital pH meter AB15j Fisher Scientific, 100 ml of each sample was poured into a sterile beaker and the electrode of the pH meter was dipped into it and readings were obtained when it was stable.

Determination of TDS and EC:

The Electrical Conductivity (EC) was determined in the laboratory by a conductivity meter (SesIon 7, Hach Company, USA). The electrode was dipped into 100 ml of the water sample, and the reading was recorded when it was stable. The TDS can be calculated by multiplying the EC by a predetermined factor. The factor determined gravimetrically ranges between 0.55 and 0.9, in this study the factor value used was 0.64.

Statistical Analysis:

Each sample was analyzed in triplicate and the figures were then averaged. The statistical analysis was performed with SAS program (SAS.1990) using of variance (ANOVA) and means were separated by Duncan's multiple range tests with a probability P≤ 0.05 (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

Physicochemical parameters:

The results of the physicochemical parameters of drinking water (50 samples) collected from all regions were summarized in Table (2) (a, b, c, d, and e).

The results of 10 samples collected from the central region showed that the pH values ranged from 7.09 to 7.82 with an average of 7.42, TDS from 133.6 to 154.9 mg/liter with an average of 148.8, and EC values varied from 208.7 to 242.0 µS/cm with an average 232.5 (Table 2a). The results of 11 samples from the southern region; revealed that pH, TDS, and EC were ranged from 7.22 to 8.20 with an average value7.59, from 149.2 to 161.9 with an average value 155 mg/l, from 233.1 to 252.9 with an average value 242.2 µS/cm respectively, (Table 2b).

Table (2a): Bacteriological and physicochemical parameters of drinking water samples collected from Central Region of City

Physicochemical parameters	Values determine of central region			Allowed limits by WHO
	Minimum	Maximum	Mean	
pH	7.09	7.82	7.42	6.5-8.5
TDS mg/l	133.6	154.9	148.8	1000
E.C µS/cm	208.7	242.0	232.5	1500
Enumeration of bacteriological analysis	**Values determine of central region**			**Allowed limits by WHO**
	Minimum	**Maximum**	**Mean**	
Coliform MPN/100ml	0.0	3.1	0.31	0
***E.coli* MPN/100ml**	0.0	0.0	0.0	0
***Pseudomonas aeruginosa* MPN/100ml**	0.0	8.6	2.3	0

- Total number of samples = 10 Drinking water
- Each sample was done in duplicate

Table (2b): Bacteriological and physicochemical parameters of drinking water samples collected from Southern Region of City

Physicochemical parameters Mean	Values determine of Southern region			Allowed limits by WHO
	Minimum	Maximum	Mean	
pH	7.22	8.20	7.59	6.5-8.5
TDS mg/l	149.2	161.9	155.0	1000
E.C µS/cm	233.1	252.9	242.2	1500
Enumeration of bacteriological analysis Mean	**Values determine of Southern region**			**Allowed limits by WHO**
	Minimum	**Maximum**	**Mean**	
Coliform MPN/100 ml	0.0	0.0	0.0	0
***E.coli* MPN/100 ml**	0.0	0.0	0.0	0
***Pseudomonas aeruginosa* MPN/100 ml**	0.0	9.8	4.9	0

- Total number of samples = 11 Drinking water site
- Each sample was done in duplicate

Table (2c): Physicochemical and bacteriological parameters of drinking water samples collected from Western Region of City

Physicochemical parameters	Values determine of Western region			Allowed limits by WHO
	Minimum	Maximum	Mean	
pH	6.56	7.65	7.36	6.5-8.5
TDS mg/l	148.4	202.4	162.2	1000
E.C µS/cm	231.9	316.3	253.5	1500
Enumeration of bacteriological analysis	**Values determine of Western region**			**Allowed limits by WHO**
	Minimum	**Maximum**	**Mean**	
Coliform MPN/100ml	0.0	17.2	3.5	0
***E.coli* MPN/100ml**	0.0	0.0	0.0	0
***Pseudomonas aeruginosa* MPN/100ml**	0.0	2149.6	392.7	0

- Total number of samples = 7 Drinking water site
- Each sample was done in duplicate

Values of 7 samples from Western region varied from 6.56 to 7.65 with a mean of 7.36, from 148.4 to 202.4 with a mean of 162.2 and from 231.9 to 316.3 with a mean253.5 for pH, TDS, and E.C, respectively (Table 2c).for pH, TDS, and EC, respectively (Table 2c). On the other hand Table (2d) Showed that the values of nine samples of the northern region were found to be ranged from 6.56 to 8.50 for pH with an average 7.41, from 124.9 to 192.3 with an average 154.9for TDS and from 195.2 to 300.5 with an average 242.0 for EC. Data of samples from Eastern Region recorded in (Table 2e) indicated that pH, TDS, EC values ranged from 6.85 to 8.33, with an average 7.68, from 104.7 to 188.2 with an average 141.8, and from163.6 to 294.1with an average 221.5, respectively.

The pH of water is controlled by the carbon dioxide-bicarbonate. The increase of carbon dioxide will lower the pH concentration, whereas the decrease in it will raise it, as well as the lower than 7 are considered acidic and, that with the pH more than 7 considered basic. Control of pH is important in drinking water to minimize the corrosion of water mains and pipes and maintain the taste, odor and appearance (WHO, 1996). Environmental Protection Agency (EPA) regulation doesn't include pH in drinking water quality, because it's considered an aesthetic quality of water. However, the agency recommends that municipal drinking water suppliers keep their water supply at a pH of 6.5 to 8.5. Total dissolved solids (TDS) are used to describe the inorganic salts and small amounts of organic material present in water. TDS in water is directly related to conductivity and effect on the taste of water.

The present study concluded that the selected physicochemical parameters of the drinking water fifty samples collected from the private selling sites of the urban area of Khamis Mushait City are varied from 6.56 to 8.50, 104.70 to 202.40, and 163.60 to 316.30 for pH, TDS, and EC, respectively, these values are falling within the range of the WHO (2011) standards guidelines of drinking water. Therefore, all drinking water samples were found to be fit for human consumption.

Table (2d): Bacteriological and physicochemical parameters of drinking water samples collected from Northern Region of City

Physicochemical parameters	Values determine of Northern region			Allowed limits by WHO
	Minimum	Maximum	Mean	
pH	6.56	8.50	7.41	6.5-8.5
TDS mg/l	124.9	192.3	154.9	1000
E.C µS/cm	195.2	300.5	242.0	1500
Enumeration of bacteriological analysis	**Values determine of Northern region**			**Allowed limits by WHO**
	Minimum	**Maximum**	**Mean**	
Coliform MPN/100 ml	0.0	8.6	1.0	0
***E.coli* MPN/100 ml**	0.0	0.0	0.0	0
***Pseudomonas aeruginosa* MPN/100 ml**	0	343.6	76.8	0

- Total number of samples = 9 Drinking water site.
- Each sample was done in duplicate.

Table (2e): Physicochemical and bacteriological parameters of drinking water samples collected from Eastern Region of City

Physicochemical parameter	Values determine of Eastern region			Allowed limits by WHO
	Minimum	Maximum	Mean	
pH	6.85	8.33	7.68	6.5-8.5
TDS mg/l	104.7	188.2	141.8	1000
E.C µS/cm	163.6	294.1	221.5	1500
Enumeration of bacteriological analysis	Values determine of Eastern region			Allowed limits by WHO
	Minimum	Maximum	Mean	
Coliform MPN/100ml	0.0	20.5	2.2	0
***E.coli* MPN/100ml**	0.0	0.0	0.0	0
***Pseudomonas aeruginosa* MPN/100ml**	0	178.6	4.3	0

Total number of samples = 13 Drinking water site
Each sample was done in duplicate

Bacteriological analysis:

The bacteriological results are summarized in tables (2a, 2b, 2c, 2d, 2e) and compare them to WHO (2011), guidelines of drinking water quality.

The results showed that total coliforms were found in drinking water collected from, Central, Western, Northern and Eastern with a value varied from 0.0 to 3.1 with an average 0.31, from 0.0 to 17.2 with an average 3.5, from 0.0 to 8.6 with an average 1.0 and from 0.0 to 20.5 with an average 2.2 MPN/100ml, respectively. However, no coliforms found in the southern region. The drinking water should be free of coliform according to WHO (2011) guidelines for drinking water; their presence in treated drinking water may be due to the ineffectiveness of treatment or post contamination after treatment or poor hygiene. Also, its presence in drinking water indicates that disease-causing organisms (pathogens) could be in the water system. The *E. coli* count is not detected in all drinking water samples taken from different geographic regions, therefore no fecal contaminant. The *E. coli* bacteria are one the coliform group that indicate fecal pollution and it is also strictly of fecal origin.

The total count of *Pseudomonas aeruginosa* of Central region was varied from 0.0 to 8.6 with mean 2.3, Southern region from 0.0 to 9.8 with mean 4.9, the Western region from 0.0 to 2149.6 with mean 392.7, Northern region, from 0.0 to 343.6 with mean 76.8.7, and Eastern region from 0.0 to 178.6 with mean 4.3 MPN/100ml. Majority of *Pseudomonas* species are not harmful to humans but *Pseudomonas aeruginosa* can cause infections in immune-suppressed patients.

The contamination percentage of total coliforms was found (14%), *Pseudomonas aeruginosa* (48%), and *E. coli* (0%). This study also indicated that about 54% of the water samples are incompatible with WHO, (2011) standards (Fig. 3), so the study suggests that these private water selling sites need urgent action to control the source of contamination. The cause of high bacteriological contamination may be due to ineffective disinfectant in the system, contamination during transportation by trucks, water storage tanks in the plants, pipelines, poor hygiene in the plants and post contamination after treatment. Moreover, plastic containers of water may be a source of contamination when reused many times in order to save money.

Most of the drinking water has been collected from private water sites (54%) are not fit for drinking. Therefore, we recommend the need to periodically intensify quality control programs with the addition of an appropriate method of disinfection in the system.

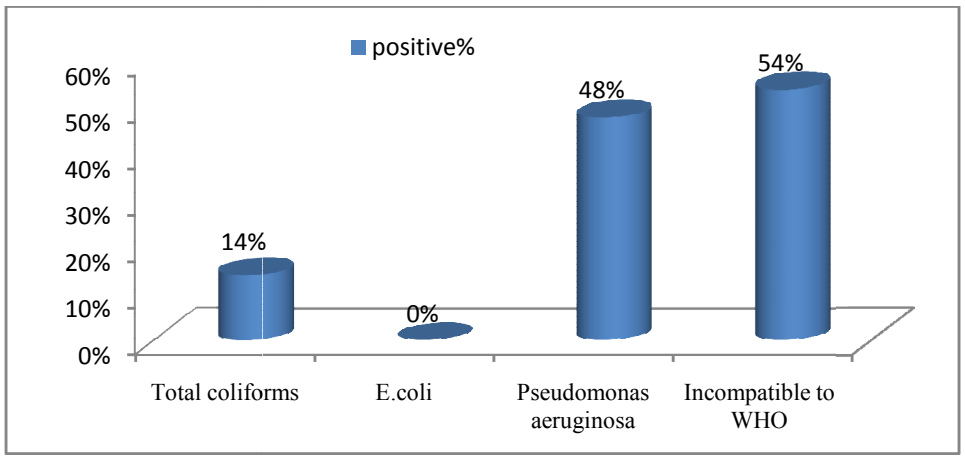

Fig. (3): Contamination percentage of total coliforms, *E. coli,* and *pseudomonas aeruginosa* and total incompatible percentage samples to WHO (2011) standards

CONCLUSIONS

The present study was concluded that the results of physicochemical parameters (pH, TDS, and EC) of the drinking water samples collected from the private selling sites of the urban area of Khamis Mushait city were within the permissible limits to WHO (2011) guidelines of drinking water. Bacteriological results indicated that 14% of the water samples were positive for coliforms contamination, but, *Escherichia coli* bacterium was not found in all samples. About 48% of the samples were contaminated with *Pseudomonas aeruginosa*. Furthermore, our results indicated that 27 samples out of 50 water samples collected from private selling sites (54%) were incompatible to WHO (2011) standards for drinking water.

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التقييم البكتيريولوجي والفيزيوكيميائي لمياه الشرب المجمعة من مواقع البيع الخاصة في المناطق الحضرية بمدينة خميس مشيط ـ المملكة العربية السعودية

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أجريت هذه الدراسة في المنطقة الحضرية في مدينة خميس مشيط، منطقة عسير، المملكة العربية السعودية. حيث تم جمع عدد ٥٠ عينة مياه الشرب من مواقع البيع الخاصة للمياه غير المعبأة في المنطقة الحضرية في مدينة خميس مشيط. تم قياس بعض الصفات الفيزيوكيميائية مثل الرقم الهيدروجيني (pH) ، المواد الصلبة الذائبة الكلية (TDS) والتوصيل الكهربائي (EC) باستخدام الطرق القياسية لتقدير جودة مياه الشرب. تراوح الرقم الهيدروجيني لعينات المياه من ٦.٥٦±٠.١ إلى ٨.٥±٠.٤٩، المواد الصلبة الذائبة الكلية من ١٠٤.٧±٠.٢٨ إلى ٢٠٢.٤±١.٧ ملغم/لتر وحساب قيمة التوصيل الكهربي من ١٦٣.٦±٠.٤٥ إلى ٣١٦.٣±٢.٦١ ميكروسيمنز/سم. أشارت النتائج السابقة إلى أن جميع المعايير الفيزيوكيميائية التي تم قياسها هي ضمن الحدود المسموح بها لمنظمة الصحة العالمية (٢٠١١م) فيما يتعلق بجودة مياه الشرب. شمل الفحص البكتيريولوجي لعينات المياه العدد الأكثر إحتمالا (MPN/100ml) لمجموعة القولون الكلية، بكتيريا الايشريشيا كولاي (*E. coli*) وبكتيريا الزائفة الزنجارية (*Pseudomonas aeruginosa*). وأظهرت النتائج أن بكتيريا الإيشريشيا كولاي لم يتم اكتشافه في كل العينات، بينما تم العثور على مجموعة القولون الكلية في ٧ عينات من أصل ٥٠ عينة تم تحليلها بنسبة تعادل (١٤٪) وتم العثور على الزائفة الزنجارية في ٢٤ عينة من إجمالي ٥٠عينة تم تحليلها بنسبة (٤٨٪). علاوة على ذلك، كشفت هذه الدراسة أن حوالي ٥٤٪ من عينات مياه الشرب التي تم جمعها من مواقع البيع الخاصة لا تتوافق مع معايير منظمة الصحة العالمية بجودة مياه الشرب.