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Assessment of Microbial and Chemical Contamination in Re-Usable 18 Liter Bottled water Marketed in Tripoli City and its Suburbs

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تقدير التلوث الكيميائي والميكروبي في عبوات المياه، المعبأة سعة 18 لتر المتداولة في مدينة طرابلس وضواحيها

نظرا لتزايد السكان ومحدودية الموارد المائية الصالحة للشرب قد عزز من ضرورة استخدام المياه المعبأة في زجاجات وانتقال الأمراض المنقولة عن طريق المياه هو مصدر قلق كبير للصحة العامة وأنه من المهم أن نعرف التلوث الكيميائي والميكروبي في المياه المعبأة في زجاجات التي يعاد استخدامها. وكانت هذه الدراسة للتحقيق في التلوث الميكروبي والكيميائي في الزجاجات المياه المعبأة ذات سعة 18 لتر المستهلكة في مدينة طرابلس وضواحيها. تم جمع 40 عينة من المياه المعبأة في زجاجات عشوائيا من عشر ماركات مختلفة. جميع العينات تتوافق مع اللوائح القياسية لمنظمة الصحة العالمية. وكانت متوسط قيم النترات ، والنتريت ،

التعكر ، Na^+، K^+، درجة الحموضة (pH) ، 7.10مليجرام/لتر،0.013 مليجرام /لتر،0.36 NTU ، 8.6 مليجرام /لتر ، 0.32 مليجرام /لتر ، 7.64علي التولي. وأظهرت النتائج من المتوسط والانحراف المعياري لمجموع البكتيريا ا،لقولونية تم الكشف عنها في 14.4,2 ٪ من زجاجات المياه المعبأة. ارتفاع مستوي التلوث بمؤشرات التلوث الميكروبي في عبوات المياه ذات سعة 18 لتر المعروضة للبيع في السوق المحلي يشير إلي احتمال وجود إحياء دقيقة وطفيليات ممرضة في هذه المياه وهذا قد يشكل تهديدا علي الصحة العامة . ونظرا لان العبوات قبل إعادة تعبئتها واستخدام مياه من مصادر غير أمنة وعدم كفاءة عملية تطهير المياه من العوامل التي ربما تكون قد ساهمت في ارتفاع المحتوى الميكروبي للمياه في بعض العبوات الممثلة للأصناف التي شملتها الدراسة.

الكلمات الدالة: التلوث الميكروبي والكيميائي- المياه المعبأة- مدينة طرابلس-ليبيا

ABSTRACT

The increasing population and limited drinking water, resources have enhanced the necessity of using bottled water. Transmission of the waterborne disease is a major concern of public health, and it is important to know chemical and microbial contamination of bottled water. This study was to investigate the microbial and chemical contamination in 18liters refilled water samples consumed inTripoli city and its suburbs. 40 samples of bottled water were collected randomly from ten different brands. All the samples meet standard of the WHO guidelines. The mean values of nitrate, nitrite, turbidity, Na^+, K^+ and pH parameters were 7.10 mg/l, 0.013 mg/l , 0.36 NTU, 8.60 mg/l, 0.32 mg/l and 7.64, respectively. Results showed that the mean and standard devation of total coliforms

bacteria were detected in 14.42% bottles water. The high levels of microbial faecal indicators recorded in this study indicate the possibility that pathogenic microorganisms and parasites might be present in the 18liters refilled bottles currently offered for sale at the local market. Such a situation might pose a threat to public health. Since the bottles are of the re-usable type, thus, the reasons behind the high levels of contamination of most brands of bottled water included in this study could be due to inefficient cleaning and sanitizing of the bottles before refilling, utilization of water from unsafe sources and inefficient sanitizing of water.

Key words: Microbial and chemical contamination - Bottle water-Tripoli City-Libya

INTRODUCTION

About 97 % of earth's water is salty water, and only 3 % is fresh water. Although fresh water is a renewable source, most of the world 's fresh water supply is ground water. This precious element is needed for the basic survival of all living organisms (Crittenden et al., 2012). Good quality is of basic importance for human physiology, and human lives depend basically on availability. Average human body weight is ranging from 53 kg - 63 kg, requires about 3 liters of water in the liquids and food daily to keep healthy (Wardlow et al., 2004). Many human diseases are related to lack hygienic water. Today, it is estimated that about 450million people in 29 countries suffer from water shortages (Guler, 2007). In recent years, bottled water gained popularity in several

countries including Libya. Bottled water is a beverage that is rapidly gaining in popularity and according to the latest World's Water report the average annual consumption is over 100 liters per person in 15 countries from the world (Diduch et al. 2013). The increasing demand for access to safe drinking water, the global population, has turned to the use of bottled water. It is stated that over 89 billion liters of bottle water are annually used worldwide (Guler, 2007; Salvato, 2003). This may be due to increasing water salinity (Morton et al.,2011). The supply and demand theory, in this case, bottled water, has necessitated a growing need for various water sources to manufacturing this product (Leivadara et al., 2008; Zazouli et al., 2013). People are thus choosing bottled water over tap water because it is perceived to be safer and of higher quality than tap water (Eric and Isaac, 2013). Water should be free from any organisms. However, unfortunately, water is not always found pure. The contamination of natural water with faecal material, domestic and industrial sewage may result in an increased risk of disease transmission to humans (Gleson and Gray, 1997; Hunter, 1997; OECD, 2003). Today fecal coliforms found in water tables have been identified as the main pollutants (Williams, 2001). Several studies have documented the detection of coliform and heterotrophic bacteria in bottled water (Bharath et al.,2003; Liguori et al., 2010). Consumption of contaminated drinking water was associated with 80 per cent of disease and one-third of death in developing countries (Mellor et al.,2013; Echoru et al.,2015). This study aimed to investigate the chemical and microbial quality in 18 liter refilled water samples consumed in Tripoli city and its suburbs.

Materials and methods

The study was carried out on ten brands of bottled water produced in the Tripoli city and its suburbs. In total, four samples from each brand (44 samples) were randomly collected during the period from Jun 2016 to November 2016. The collected samples were transferred to the advanced center for chemical analysis laboratory Tripoli and refrigerated at standard conditions (at 4°C). The nitrate anion and nitrite anion were measured by a spectrophotometer (Hach Long/USA). The pH level and Total dissolved solids (TDS) were determined by the Milwaukee – SM-801. The measurement of sodium and potassium cations was accomplished by Ion Chromatography device (Metrohm/Switzerland). The turbidity level was measured by the Hach turbidity meter device (USA). To ensure sample testing accuracy in each measurement according to the mentioned parameters, each parameter was carried out in triplicate, and the mean of each parameter was calculated. The values of each parameter were compared to set by the World Health Organization,2003 (WHO) guidelines for drinking water.

Determination of total coliforms:

Coliforms bacteria were determined by MPN (Most Probable Number) method (5-tubes test) through incubation of sample into tubes of lactose broth at 35 ^{o}C for 48h. Then tubes were examined for the appearance of gas bubbles within the Durham tubes in Brilliant Green Bile Broth 2% at 37 ^{o}C for 24hr and the tubes were selected for determination of fecal

coliforms which have produced gas. Fecal coliforms are bacteria that are associated with human or animal waste.

Detection of the fecal coliforms:

It is indicative of fecal pollution and the possible presence of enteric pathogens. A loop of a positive tube was inoculated to EC broth tube and peptone water. Tubes incubated for 24 to 48 hr at 44.5 ^{o}C. Cultures that produced gas in EC broth tubes and followed by subsequent positive reaction in Indole test (Hitchins et al., 1998). The total coliforms and fecal coliforms counts were performed based on international Standard No.9308(2000).

Statistical Analysis

The data were analyzed by SPSS software, V.16.0 (Mean ± SD). A probability level of ($P<0.05$) was considered as significant.

Results and Discussion

According to (Table 1)., among examined brands, the highest and lowest mean value of nitrate were related to brands of G (22.78 mg/l) and J (0.85 mg/l), respectively; and were B (0.02 mg/l) and C (0.007 mg/l), respectively for nitrite . Also, a significant difference was observed ($p<0.05$) in nitrate and nitrite levels among different brands, but they were ranging into an acceptable level. The results of this study were in agreement with the study by Jahed -Khaniki et al. (2008) and Godini et al. (2012). The mean value of the sodium cation in the J brand was the highest with 19 mg/ l, whereas F, C and D brands had the lowest values (2 mg/l). Also, among the different measured brands, the I brand (0.7

mg/l), and C and D brands had the lowest values (0.1 mg/l) level of potassium cation. The high amount of these minerals is of matter of health concern. Because of the elevated potassium level in bottled water can be a cause of renal disorders and sodium plays a role in developing high blood pressure (EPA, 2006). Statistical analysis showed a significant difference ($p<0.05$) between the mean values of these two cations and all of them were in accordance to the standard level. The level of total dissolved solids (TDS) and turbidity parameters were below the acceptable limit (Table 1). The values of pH for all samples ranged between 6.9–8.0 and the values of TDS ranged between 118–339 mg/l which falls within limits allowed by the WHO Standard of Drinking Water. Shown in Table 1. In this study, pH and TDS levels in the samples were in optimal standard limits. There is a study on bottled water in Kuwait indicated to about 44% of the samples had pH values higher than 8 and 8% of them were slightly acidic (Alfraji et al., 1999). The prevalence of total coliforms and fecal coliforms are shown in Table 2. Coliform bacteria were detected in 5 out of 40 of bottles of water. None of the bottle samples was positive for fecal coliforms. The findings showed that 14.42 % of the bottled water sampled contains coliform organisms and they failed to meet the World Health Organization standard for coliforms in bottled water (WHO,2003). Similar findings were found by El-Abagy et al. (2000) in Greece. There are studies conducted in Bangladesh showed that most of the bottled water in Dakar the capital of Bangladesh were contaminated by microorganisms (Ahmed et al., 2013). 18-liter refilled water samples were more exposed to pollution because of their content of microorganisms (Alabdu and Khan,

1995). This may be due to using for purposes other, and therefore be a source of water pollution when filling bottles again (Isam and Bahaa,2010). Therefore, compounding the suspicion of most consumers that bottled water may not, after all, be the clean and safe alternative they were looking for this reason (Lisa et al., 2015). The presence of coliforms in bottled water suggests the potential presence of pathogenic enteric microorganisms, and it requires an improved surveillance system for bottled water. The total coliform of bottled water according to WHO is zero (WHO,2003). Total coliforms in 100 ml water were also zero in the A, C, D, E, G and I brands. It was more than zero among the brands of B, F, H and J are shown in (Table 2). These results did not agree with the study conducted by Alabdula and Khan (1995). There was the meaningful difference ($P<0.05$) between total mean values of coliforms/100 ml in a different sample and it shows that the quality of these bottled waters is somewhat unhealthy for public consumption. Coliforms bacteria accepted as microbial contamination indicators by WHO. The presence of coliform bacteria can indicate diseases causing pathogens in water. Therefore, the consumption of contaminated bottled waters should be limited (Hammer and Hammer, 2007). In this study, the B, F, H and J brands are positive from coliforms bacteria, and they are not suitable for consumption.

There are a study conducted in South Africa showed that eight local and two brands of bottled water were free of total and fecal coliforms of bacteria (Ehlers et al.,2004). Abed and Alwakeel (2007) indicated to there is contamination with some bacteria in two of the 30 (6.7%) bottled samples. It is important to identify contaminated bottled water and to prevent the consumption of these samples.

Table 1: Physico-chemical parameters of bottled water brands in the Tripoli city and its suburbs

Brands Samples	pH	K^+ (mg/l)	Na^+ (mg/l)	Turbidity NTU	Nitrite (mg/l)	Nitrate (mg/l)	TDS (mg/l)
A	7.52±0.2	0.2±0.003	13±0.3	0.30±0.01	0.010±0.001	2.52±0.22	213.2±0.4
B	7.49±0.15	0.2±0.002	14±0.1	0.35±0.02	0.020±0.002	6.24±0.35	254.5±0.4
C	8.0±0.1	0.1±0.003	2±0.3	0.50±0.02	0.007±0.001	5.45±0.54	118±0.3
D	7.89±0.15	0.1±0.002	2±0.1	0.40±0.01	0.014±0.002	9.19±0.34	180±0.7
E	7.82±0.1	0.3±0.002	7±0.2	0.39±0.01	0.018±0.001	10.0±0.4	169.1±0.6
F	7.79±0.2	0.4±0.003	2±0.1	0.30±0.02	0.014±0.001	2.81±0.55	146.2±0.5
G	7.98±0.15	0.2±0.003	3±0.1	0.36±0.01	0.014±0.002	22.78±0.35	165±0.30
H	7.31±0.1	0.6±0.004	9±0.3	0.33±0.02	0.010±0.001	6.45±0.50	124±0.4
I	7.72±0.2	0.7±0.003	15±0.2	0.34±0.02	0.013±0.002	4.61±0.60	259±0.4
J	6.90±0.1	0.5±0.002	19±0.4	0.33±0.01	0.012±0.002	0.85±0.10	339±0.5
WHO*	***6.6-8.5***	***20***	***200***	***5***	***1.0***	***45***	***1000***

* The World Health Organization,2003, guidelines for drinking water.

Table 2: Prevalence of total coliforms in sample bottled water according to brands of purchase.

Brands	No. Samples tested	No. (%) of sample positive	
		Total coliforms	Fecal coliforms
A	4	0(0.0)	0(0.0)
B	4	2(40)	0(0.0)
C	4	0(0.0)	0(0.0)
D	4	0(0.0)	0(0.0)
E	4	0(0.0)	0(0.0)
F	4	1(20)	0(0.0)
G	4	0(0.0)	0(0.0)
H	4	1(200)	0(0.0)
I	4	0(0.0)	0(0.0)
J	4	1(20)	0(0.0)
Overall	40	5(14.42)	0(0.0)

CONCLUSION

Based on the recommended zero tolerance for microbial faecal in drinking water by WHO. The high levels of microbial indicators recorded in this study indicate the possibility that pathogenic microorganisms and parasites might be present in the 18 liter refilled bottles currently offered for sale at the local market. Such a situation might pose a threat to public health. Since the bottles are of the re-usable type, thus, the reasons behind the high levels.

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