Microbiological Quality Assessment of Drinking Water at Ed-Dueim Town, Sudan.

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Abstract: Water is precious values of human life and should have quality safe and suitable for drinking. The aim of this study was to investigate the microbial quality of drinking water. The bacterial load was determined according to the pour plate standard methods and most probable number techniques for coliform, fecal coliform and fecal streptococci. The results of the viable count of drinking water obtained from Polyvinyl chloride High Density (PVC.HD), Polyvinyl chloride Low Density (PVC.LD), and Asbestos pipes were ranged from 0.3 $\times 10^4$ to 9.3 $\times 10^7$ cfu/ml. Total coliform MPN values were ranged from 0.0 to 11MPN/100 ml. Faecal coliform 0.0 to 7MPN/100 ml, while for faecal streptococci MPN were ranged from 0.0 to 3/100ml. The most predominant bacterial genera found in drinking water were *Bacillus* (44%), *Corynebacterium* (31%), *Micrococcus* (13%), *Staphylococcus* (6%) and *Streptococcus* (6%). In addition, in this study, the physicochemical parameters (e.g. Turbidity, electrical conductivity, pH, temperature, total dissolved solid, chloride, fluoride, calcium, iodine, magnesium and sulfate) were investigated, and the results show all the values except turbidity falls below the maximum limit of Sudanese Standard Metrology Organization (SSMO) and WHO guideline standard. From the results, it may be concluded that the drinking water in Ed-Dueim town has adequate physical and chemical quality and suitable for drinking. However, the bacteriological quality needs more consideration.

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1. Introduction

Water is very important to sustain humanity and life. Good quality of drinking water is one of the most human necessities, and the lack of access to adequate safe water supplies leads to the spread of diseases (Howard and Bartram, 2003; and Bartram and Helmer, 1996). It is unfortunate in many developing countries water-related diseases are a problem's result from contaminated drinking water, so that it will be a high risk to human health, if it contains pathogens (WHO, 2006). It has been reported that the death of most of the children in Africa who die before the age of 5 is caused by inadequate and safe water supplies (Loucks, 1996). In general, terms, the greatest microbial risks are associated with ingestion of water that is contaminated with human or animal faeces (Atiribom, *et al.,* 2007). The contamination of water resources with faecal material, industrial sewage, domestic, agricultural waste still remains a serious problem in the developing countries and became a public health concern. The major problem of drinking water in Sudan and other developing countries is not just a lack of water availability, but in fact, that the people are not concerned with the water quality and pathogenic bacteria which, causes health risk.

Many years in Sudan's peoples did not pay much attention to water pollution problems but today the population is aware of the importance of good water quality and its relation to health. Previous studies in the Sudan have only dealt with the certain aspects of water pollution problems, Dirar (1986) and Abdel Magid *et al.,* (1984) work on pollution of water from the Nile and from the wells in Khartoum. Moreover, contamination of water stored in pots known locally as Zeers was investigated (Hammad and Dirar, 1982a; Hammad and Dirar, 1982b; Abdellah *et al.,* 2012). These studies showed a varied range of microbial contamination of water. Goja (2009) examined the Nile water for faecal contamination and microbial pollution; he indicates that the Nile water was faecally contaminated, and different factors affected the degree of contamination. Chemical pollution in water can also cause a public health risk. This chemical pollutant got into water bodies through different discharge ways, such as domestic, agricultural and industrial waste.

In Ed-Dueim city, the drinking water is supplied by the White Nile River, and the water is very turbid; therefore, it is expected to be contaminated with chemical as well as microorganism, and accordingly; it may cause health problems such as toxicity and some disease, although water was treated and distributed through different pipelines to the consumers. In the present study, the aim was: to investigate the chemical and microbial quality of drinking water samples in Ed-Dueim town and checking for their risk for human health according to the Sudanese Standard Metrology Organization and (SSMO) and WHO guidelines.

2. Materials and Methods

2.1. Sampling

A total number of 192 samples of drinking water were collected from different sites from the network of water distribution in Ed-Dueim city included: Treated and untreated water samples from the treatment unit and water from Taps and Zeros were collected from different pipes (Polyvinyl chloride High Density (PVC.HD), Polyvinyl chloride Low Density (PVC.LD), and Asbestos) after distributed to consumers, during the period from January 2012 to December 2012. Two liters of water from each point were collected especially an in sterile glass bottles (200 ml), the bottles were tightly closed and immediately transported to the laboratory for chemical and for microbiological analysis.

2.2. Microbiological analysis

2.2.1. Total viable count

Serial dilution was prepared by using one ml of water sample and transferred aseptically into the first sterile tube containing 9ml of normal saline (0.85% Nacl) to give 10 dilutions (10^{-1}), then1ml from the first tube was transferred to a second tube containing 9 ml of normal saline to give 10 dilutions (10^{-2}), and then further dilutions were made in a similar manner. The appropriate dilutions of the various samples were inoculated on plate count agar using pour plate method and incubated at 37°C for 24-48 h. The bacterial counts were expressed as colony forming units per ml (cfu/ml) as described in Harrigan (1998).

2.2.2. Enumeration of fecal bacterial indicators

The Most Probable Number (MPN) technique was used for enumeration of Coliforms, fecal Coliforms and fecal streptococci for all samples according to standard methods (APHA, 1995).

2.2.3. Isolation and identification bacteria

The predominant bacterial colonies were isolated from plate count agar; these isolates were purified by streaking twice on nutrient agar and stored in a refrigerator, then further the identification done through biochemical tests (Barrow and Gelthan, 1993).

2.3. Physicochemical Parameters

The physicochemical parameters were determined according to the procedures of Greenberg et al (1998). Turbidity was determined by Nephlometric method, using (HACH 2000) turbidity meter and the results were reported in Nephlometric Turbidity Units (NTU), pH was measured by PH meter (HACH 2000), Electric conductivity (EC) in µs/cm was observed on 25°C using conductivity meter (HACH 2000). Total dissolved solids (TSD) were measured gravimetrically after drying in an oven to a constant weight at 105°C. Chloride, Calcium, Magnesium and Fluoride were determined using titration and colorimetric methods, respectively as described by Greenberg et al (1998). Iodine and Sulphate were measured according to Diethyl-phenylene-Diamine (DPD) and Powder pillows methods using spectrophotometer determination (DR 2000- HACH) at 530 and 450 nm, respectively, as described in Plain (1967).

3. Results

3.1. Biological analysis

The results of the total viable count (TVC), total coliform (TC), faecal coliform (FC) and faecal Streptococci (FS) are shown in Table 1. Total viable count were ranged from 1.7×10^3 to 7.3×10^6 cfu/ml for treating water, for untreated 0.5×10^5 to 0.5×10^9cfu/ml, whiles the ranged for the PVC.HD, PVC.LD and Asbestos pipes were 0.3×10^4 to 6.5×10^6, 0.1×10^4 to 3.3×10^6 and 2.5×10^4 to 9.3×10^7 cfu/ml, respectively. There are no big differences were noticed between the different pipes of network in the different months. However, slight rising in an account of Asbestos pipeline, and the highest count between (5.5×10^7 to 9.3×10^7 cfu/ml) was recorded in Zeer's samples during the summer seasons (March - April) while the lowest count between (0.3×10^4 to 0.7×10^4 cfu/ml) was recorded in Tap samples (PVC. HD pipelines) during winter seasons (October – December) figure 1. Total coliform MPN values were ranged from 0.0 to 75/100 ml, 0.0 to 7/100ml, 0.0 to 3.0/100, 0.0 to 7.0/100ml and between 0.0 to 11MPN/100 ml in untreated, treated water, PVC.HD, PVC.LD and Asbestos pipes, respectively. Faecal coliforms 0.0 to 28MPN/100ml for untreated and 0.0 to 3MPN/100 ml for treating water, while from 0.0 to 2, 0.0 to 4 and 0.0 to 7MPN/100 ml for PVC. HD, PVC.LD and Asbestos pipes, respectively, while for faecal streptococci, MPN were ranged from 0.0 to 20/100, 0.0 to 7/100 ml, 0.0 to 3/100ml, 0.0 to 3MPN/100ml for untreated, treated, PVC.LD and Asbestos, respectively, and no faecal streptococci were found in PVC.HD pipe during the study.

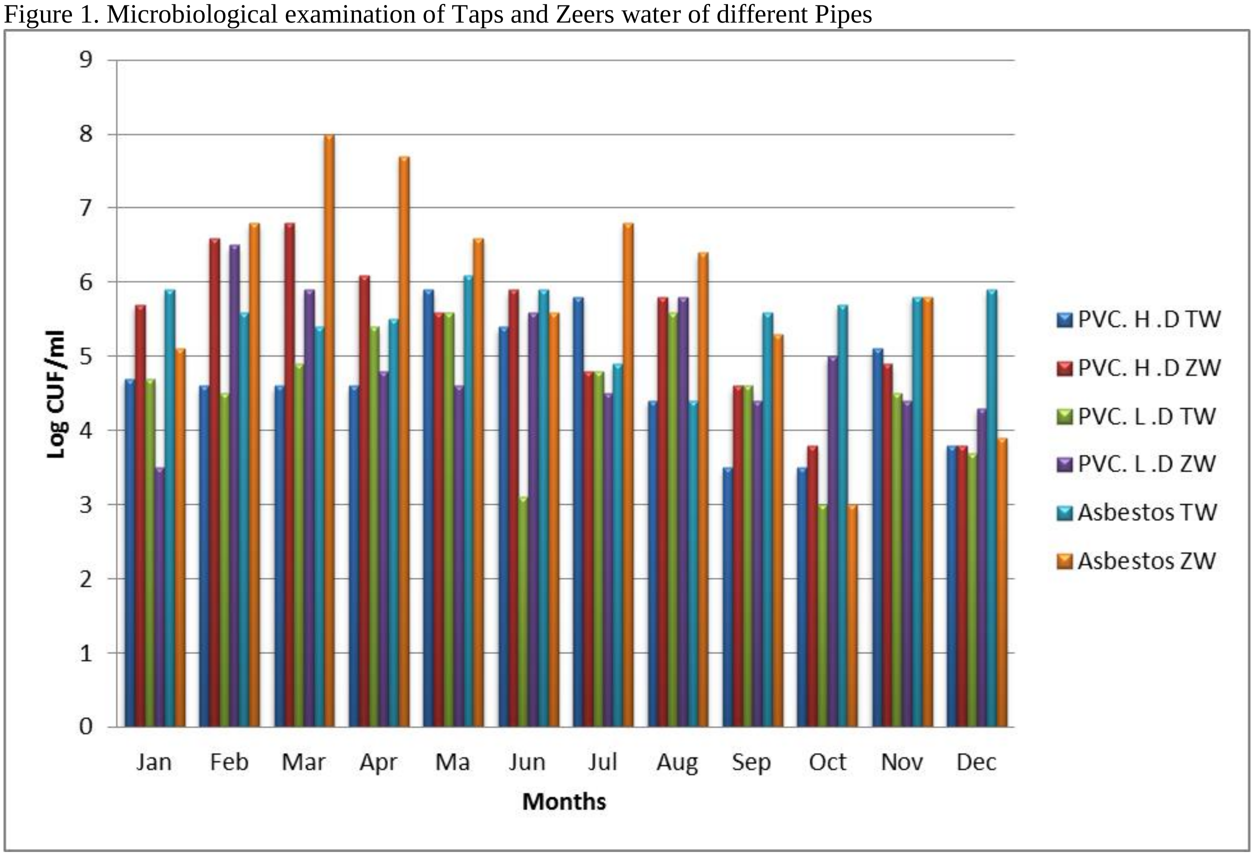
The result also shows that *Bacillus* (44%) is the genus of the greatest proportion ration of isolates, while *Corynebacterium* ranks second (31%) followed by *Micrococcus* (13%) and *Staphylococcus*, *Streptococcus* (6%) counted the smallest proportion in drinking water as shown in Table 2 and figure 2.

Table 1. The microbiological examination of Drinking water in Ed-Dueim Town

Samples	TVC CUF/ml	CC MPN/100ml	FC MPN/100ml	FS MPN/100ml
Untreated	0.5× 105 to 0.5 ×109	0.0 – 75	0.0 - 28	0.0 – 20
Treated	1.7 ×103 to 7.3 ×106	0.0 – 7.0	0.0 – 3.0	0.0 – 7.0
PVC.HD	0.3 ×104 to 6.5 ×106	0.0 - 3.0	0.0 – 2.0	NG
PVC.LD	0.1× 104 to 3.3 ×106	0.0 - 7.0	0.0 – 4.0	0.0 – 3.0
Asbestos	2.5× 104 to 9.3 ×107	0.0 – 11	0.0 – 7.0	0.0 – 3.0
SSMO/WHO (2008)	-/-	10/0MP/100 ml	0 MPN/100 ml	0 MPN/100 ml

TVC ≡ Total viable count; CC ≡ Coliform count; FC ≡ faecal coliform count; FS ≡ faecal streptococci count
NG ≡ No Growth

Figure 1. Microbiological examination of Taps and Zeers water of different Pipes

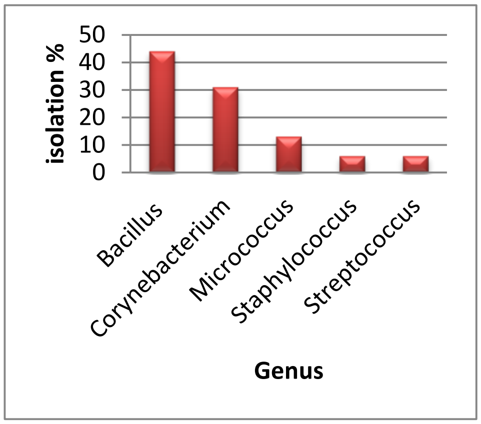


PVC.H.DTW≡Polyvinyl Chloride High Density Tap Water, PVC.H,ZW≡Polyvinyl Chloride High Density Zeer Water, PVC.L.D.TW≡Polyvinyl Chloride Low Density Tap Water, PVC.L.D.ZW≡Polyvinyl Chloride Low Density Zeer Water, Asbestos TW≡ Tape Water, Asbestos ZW≡ Zeer Water

Table 2. Morphological and biochemical identification tests of the bacteria isolated

Isolate	Gram Satin	Shape	Spore Staining	Motility	Growth In air	Cata	Oxi.	Gluc.	O/F	Genus
1	+ve	Rod	+ve	+ve	+ve	+ve	-ve	+ve	F	Bacillus
2	+ve	Rod	-ve	-ve	+ve	+ve	-ve	+ve	F	Corynebacterium
3	+ve	Sphere	-ve	-ve	+ve	+ve	-ve	+ve	O	Micrococcus
4	+ve	Sphere	-ve	-ve	+ve	+ve	-ve	+ve	F	Staphylococcus
5	+ve	Sphere	-ve	-ve	+ve	-ve	-ve	+ve	F	Streptococcus

+ve ≡ Positive; -ve ≡ Negative, Cata. ≡ Catalase test, Oxi. ≡ Oxidase test, Gluc. ≡ Glucose test.



Figuer 2. Prevalence of microorganisms isolates of Ed-Dueim drinking water

3.2. Physicochemical parameters

The physicochemical parameters of drinking water are shown in Tables 3, 4. Turbidity was ranged (2 – 470 NTU), Electrical conductivity's (EC) (130 - 239 µs/cm), pH (6.8 - 7.8). Temperature (11 – 30 ^{o}C) and Total Dissolved Solid (TDS) (59 - 108 mg/L).Chloride 6.0 – 10.5, Fluoride 0.3 – 0.4, Calcium 7.9 – 21.7, Iodine 0.001- 0.13, Magnesium 3.8 – 7.3 and Sulphate 7.9 – 13.3

Table 3. Results of Seasonality physical parameters of drinking water (Jan. – Dec. 2012)

Seasons	Turbidity (NTU)	EC (µs/cm)	TDS (mg/L)	T oC	pH
Summer	2.0 – 402	130 - 211	59 – 78	12 – 30	6.9 – 7.3
Autumn	2.4 – 470	135 - 239	82 – 108	13 – 29	6.8 – 7.6
Winter	2.5 – 450	130 - 228	65 – 90	11 – 27	6.9 – 7.5
SSMO/WHO (2008)	<5	1600	500/1000	NE	6.5 – 8.5

EC ≡ Electrical conductivity; TDS ≡ Total Dissolved Solid; T ≡ Temperature; NE ≡ Not Established

Table 4. Seasonality chemical analysis of minerals (mg/L) of different water pipes (Jan. – Dec. 2012)

Seasons	Mineral	PVC.HD	PVC.LD	Asbestos	SSMO/WHO (2008)
Summer	Cl^{-}	8.40	8.30	6.00	250
	F−	0.39	0.34	0.34	1.3/1.5
	$Ca2^{+}$	13.5	11.2	13.0	100-300/
	I	0.01	0.12	0.12	-/-
	$Mg2^{+}$	6.90	4.20	3.90	50
	$SO4^{-}$	11.0	9.00	7.80	500/400
Autumn	C^{l-}	7.00	9.00	8.00	250
	F−	0.30	0.33	0.30	1.3/1.5
	$Ca2^{+}$	7.90	11.0	10.8	100-300/-
	I	0.06	0.08	0.02	-/-
	$Mg2^{+}$	6.40	3.80	4.10	50
	$SO4^{-}$	12.0	9.70	8.60	500/400
Winter	Cl^{-}	10.5	9.30	8.00	250
	F−	0.40	0.38	0.33	1.3/1.5
	$Ca2^{+}$	15.3	20.0	16.0	100-300/-
	I	0.13	0.10	0.07	-/-
	$Mg2^{+}$	6.60	3.90	5.30	50
	$SO4^{-}$	12.5	13.0	9.00	500/400

PVC.HD ≡ Polyvinyl Chloride High Density, PVC.LD ≡ Polyvinyl Chloride Low Density, SSMO ≡ Sudanese Standard Metrology Organization, WHO ≡ World Health Organization

4. Discussion

4.1. Bacterial load

Data presented in this study Table 1 shows that the TVC of treated water was decreased when compared with untreated water. This is reflecting the chlorine action which, is the most effective water disinfectant and had a major impact on water treatment (Madigan *et al.*, 1997). In addition, chlorine has virtually eliminated most waterborne diseases (USCDC 1997). This finding is lower than the results obtained by Abdo *et al.* (2010) in Ismailia Canal Water. He indicates that the count of viable bacteria ranged from 10.0 x10^6 cfu/ml in summer to 413.0 x10^6 cfu/ml in spring. However, it is higher when compared with the findings obtained by El Bakri (2007) who found that the count of drinking water (surface and ground) were ranged from 3.5×10^2 to 6.8×10^5cfu/ml. This is could be said these older pipes deposits high organic and inorganic matter on the interior walls of pipelines, which serve as a nutrient source for microorganism. Previous studies demonstrated that pipe sediment such as silicon, phosphorous, sulphur, iron, potassium, manganese can serve as the sources of bacterial nutrients (Chowdhury, 2011; Ridgway and Olson 1981).

In search of Taps and Zeer's water analyzed from the pipes (TVC.HD, TVC. LD and Asbstos) Figs (1) shows that the TVC of Zeer samples in all pipes was generally higher than Tap water; this is clear in summer months. That is could be the suitable period for incubation and growth of microorganism, and warm temperature encourages the growth of microorganism. In addition, Zeers usually situated openly in public places and it's used by all those people who want drink water and this is definitely increasing the contamination of Zeer's water as was stated by Goja (2009) who reported that the greater the number of people around a water source, the higher the probability of microbial contamination of that water source and the hence the probability the causation of disease. This finding agrees with Hadi (2006) and Abdellah (2012) they reported that water kept in the series is more contaminated than water kept in another container. Because of the most frequent of usage, lack of cleaning of Zeers, cups, hand of some of the uses (particularly Children) and the fact that Zeers are usually left open without Coverless, which facilitates microorganisms to introduce into water from the surrounding dusts (Abdel Magid 1997; Hammad and Dirar (1982a).

4.2. Bacterial indicators of faecal contamination

It is clear that the total coliform MPN values were decreased substantially after water treated. This reflects the efficiency of the treatment process in the Ed-Dueim treatment unit station. Because of used filtration after coagulation, flocculation and sedimentation by poly aluminium chloride (PAC) which is a very effective compound for high turbidity and rapid coagulants as soon as are added to the water (Madigan *et al.*, 1997). The filtration removed 95% of microbial if these filters are constructed proper and technical operation is good (Madigan *et al.*, 1997). The counts were not exceeded 10/100ml MPN which is the maximum allowable limit for no risk (SSMO, 2008, WHO, 2008), except in tap water (11/100ml MPN) sample of July month in Asbestos pipe. According to quality guidelines for drinking water WHO (1997) and SSMO (2008) standard, the maximum limit for no risk of faecal coliform is 0 cfu/100ml and 10/100ml for the coliform count. The results from the study show that, the treated water was agreeing partially to these standard guidelines due to the recontamination of faecal coliform occurred at some points in the network pipelines. In addition, the concentration of chlorine is not enough to prevent contamination of bacteria, and the injected chlorine is not proportional to the amount of water that passes per unit time. If this inadequately treated water passed via the main network distribution pipelines, it becomes more contaminated when it reaches the consumers. In the Sudan, Hammad and Dirar (1982b) examined sebeel water and suggested faecal streptococci as better indicators than faecal coliforms. The results from this study indicate that faecal streptococci MPN count is lower than the fecal Coliforms count in Ed-Dueim drinking water examined, and therefore, the use of standard indicator bacteria of Faecal streptococci to predict the presence of the pathogens in tropical waters should be interpreted with caution. According to the European Economic community (E.E.C) and WHO standard and guideline's faecal coliform MPN limits for surface water used as raw water to be treated for drinking is 200/100 ml and is less than 1000/100 ml of irrigation water for food crops consumed raw (Tebbut, 1990 and WHO, 1996). In this study, found that during most of the month’s faecal coliform bacteria, count was less than 200/100, so they meet with the limits of raw water treated for drinking.

The finding in this study indicates that the most two dominant bacterial genera isolated from drinking water were *Bacillus,* soil inhabits organism, followed by *Corynebacterium,* which inhabits in a broad variety such as soil, rotting plant material, animal fodder, and in addition it may be found as normal flora in the skin (Green, 2009). This dominancy due to the resistance of spores to disinfection processes Figure 2. It has been reported that *Bacillus* spores as a good indicator of the treatment efficiency Ell-Amin *et al.*, 2012. Similar results were obtained from Goja (2009), Ahmed (2005), Elrofaei (2000), El Tom (1997) and Hassan and Banat (1995); they reported that *Bacillus* the first most common bacterial genera found in water. Oyeleke and Istifanus, (2008) reported that Gram-positive genera, particularly the genus's *Bacillus* where the most abundant bacteria isolated from the Kaduna River in Nigeria.

4.3. Physicochemical parameters

The turbidity of untreated water was recorded higher value in rainy season (Jun, July and August) with the value of (130,700,340 NTU) and (74,400, 350 NTU) for treating water, respectively, while the lowest value (20, 25) and (6.7, 5.5) was recorded in winter season (December and January), respectively (Data not shown). During the rainy season, when a flood occurs, soil was erosion, and water pick up pollutants and wastes of buildings to water bodies. This means increases in organic materials as well increasing in turbidity. Turbidity within the pipes showed big differences between the Taps and Zeers water; the highest values were recorded in Taps water (470.0, 450.0, 402.0 NTU) in autumn, winter and summer, respectivelyTable 3. However, the lowest value was recorded in Zeers (2.0, 2.4, 2.5 NTU) water in summer, autumn and winter, respectively. This could be due to the sediment and precipitation of suspended material of water in Zeer pots. Generally, the turbidity value of Ed-Dueim drinking water exceeds the permissible level of WHO (1993) and SSMO (2008) 5NTU. TDS is indicators present of inorganic salts (e.g. Ca2+, Mg2+ and etc.… in water. TDS ranged from 59 – 108 within the acceptable range (WHO, 2008). Electrical conductivity (EC) which has correlated with TDS, higher value in EC corresponded with high values of TDS Table 3. Other's physicochemical parameters, including Temperature, pH, Ca2+, Mg2+, F, Cl, I, and S were falling below the maximum level of WHO (1993) and SSMO (2008) as shown in Tables 4. Similar findings were reported by many workers (Eltom, 1997, Awad Elgeed, 2006, Siddig, 2007 and Ezzat, 2012) they found a, Mg, F, Cl, I, and S hasn't exceeded the threshold value of the WHO (1993) and SSMO (2008) standards.

Conclusions

Water sources have been influenced by human activities (e.g domestic discharges, industrial's sewage, agricultural waste and…. etc.). Water contaminated with pathogenic microorganism can cause many diseases. Drinking water should be free from pathogenic bacteria and safe for human consumption. From the results of this study, it can be concluded that the drinking water in the Ed Dueim supply network is generally safe for drinking according to the Sudanese Standard Metrology Organization (SSMO, 2008) guidelines. This due to all physical and chemical parameters examined come below the maximum level of thresholds, and the total coliforms counts were recorded less than value 10/100ml MPN. However, the present of fecal coliform and other bacterial genera in some few samples were reflecting the contamination causes by humans or animals, and recontamination of water with bacteria were occurring due to the sediments of organic and inorganic matter inner the old pipes (Asbestos) which, encourage the growth of the organism. Thus, the following recommendations are made: 1. Checking the efficiency of the water-treatment station (filters and tank) frequently to guarantee water quality and should be suitable for drinking. 2. The pipe (Asbestos) in the network distribution must be replaced by PVC. HD or PVC.LD pipes so as to avoid a recontamination resulting from breakdown and degradations. 3. The chlorine treatment must be enough to prevent contamination by microorganisms. 4. Proper intake of water from the White Nile source by using swimming pumps (especially in summer) to decrease the intake of mud.

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