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Assessment of Bacteriological Quality of Drinking Water in some Household Water Filter Systems in Benghazi City

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Abstract: Water is very important to human beings. Although human life can exist for many days without food, the absence of water for only a few days has fatal consequences. A wide variety of commercial water treatment systems are available for application to treat very small quantities, such as for individual homes or taps. These can treat virtually any water quality problem. It is important to have a good understanding of the specific water quality problems before selecting water treatment system. It is also important that products be tested and approved by a qualified independent certification organization to have confidence that the device will indeed perform as the vendor claims. The present study aimed to assess the bacteriological quality of drinking water in some household water filter systems in Benghazi city. The study was carried out on a total of 600 water samples (300 tap water samples and 300 filter water devices samples). All water samples were examined for enumeration of viable heterotrophic bacteria by pour plate method and enumeration of total coliforms (TC) by both the multiple tube fermentation (MTF) and membrane filtration (MF) methods and for thermotolerant coliforms (TTC), *Fecal streptococci* (FS), and *Pseudomonas aeruginosa* (*P. aeruginosa)* by the MF method. According to Libyan guidelines, out of 600 examined drinking water samples 76.8% were acceptable. The highest percentage of acceptable samples was revealed from reverse osmosis (RO) system (90.7%), followed by charcoal filter (83.3%), tap water distribution system (DS) (82.3%), and only 54.7% from tap water tanks. It was concluded from this study that filtered water samples were found to be superior to tap water samples as regards their bacteriological aspects. In addition, *P. aeruginosa* was considered as an excellent indicator for the efficiency of the water filters.

INTRODUCTION:

The key to increase human productivity and long life is good quality water.(1) Drinking water is an important resource all around the globe. (2) It is one of the most important commodities which man has exploited than any other resource for sustenance of his life. (3)

Outbreaks of waterborne infectious diseases via the use of contaminated drinking water still pose a serious health threat worldwide, despite the fact that drinking water is one of the most closely

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monitored and strictly regulated resources. Careful estimates indicate that each year about 350 million people are infected by waterborne pathogens, with 10 to 20 million succumbing due to severe cases of infection.[4] This phenomenon is far from being restricted to developing countries but also threatens developed countries. In the United States (US), almost 430,000 cases were reported in 126 outbreaks of waterborne infectious diseases during years from 2003 to 2005. [5]

The number of different types of pathogens that can be present in water as a result of contamination with human or animal feces is very large and it is not possible to test water samples for each specific pathogen. Therefore, scientists and public health officials typically choose to monitor non pathogenic bacteria that are usually associated with pathogens transmitted by fecal contamination; but are more easily sampled and measured. These associated bacteria are called indicator organisms. [6]

Aeromonas species and *P. aeruginosa* have been advocated as a means of monitoring the hygienic quality of drinking water. They indicate the general cleanliness of the distribution system and are used to assess the quality of filter water devices as their presence suggests non compliance with good manufacturing. [7, 8]

A wide variety of commercial water treatment systems are available for application to treat very small quantities, such as for individual homes or taps. These can treat virtually any water quality problem. A major goal in water treatment is to produce clear, good tasting water. Adsorption has been used for many years to help achieve this goal by removing organics that cause taste, odor and color in drinking water. Recently it is also becoming an important method for protecting public health by removing toxic organic chemicals from drinking water. [9, 10]

There are a number of methods

available to improve or enhance drinking water quality. People work very hard to improve treatments like purification and disinfection and to get perfect product-drink safe for health and necessary for life called simply Household water filters (HHWFs).[11,12]

Water filters have been introduced for many centuries; they are closely related to the history of water. Various filtration methods for reducing microbes in water have been widely known and practiced for decades. [13]

According to the history of water treatment the use of water filters began more than 4000 years ago. As early as 1500 years ago, Egyptians recommended the application of coagulant alum to settle out the suspended solids from water. [14]

Household water management practices have been introduced in approximately 50 developing countries. These range from simple filters made from sari cloth and nylon to commercially produced sachets of flocculant-disinfectants. [15]

Geldreich *et. al*., [16] reported that, installation and use by the public of in-the-home, point of use (POU) water treatment devices have increased during the last several years. This trend has paralleled increased consumer awareness of the potential health risks posed by some community drinking water systems.

Although filtration was recognized for removal of undesirable particles and microbes, several studies showed high bacterial counts in fresh filtered than in tap water after approximately one week of use.[17, 18, 19]

For this reason, the assessment of the biological characteristics of filtered and tap water is of great significance.

Aim of the Work:

The present study aimed to assess the bacteriological quality of drinking water in some household water filter systems in Benghazi city.

Material & Methods:

The study was carried out during 11 months, from the beginning of March 2008 till the end of January 2009, on a total of 600 water samples (300 tap water samples and 300 filter water devices samples). These samples were collected monthly.

Tap water samples*:*

A total of 300 water samples were collected from 21 various areas in Benghazi city (130 samples from DS and 170 samples from tanks).

Filter water devices:

1) Charcoal filter samples

A total of 150 water samples were collected from 14 various areas in Benghazi city.

2) RO filter system samples

A total of 150 water samples were collected from 16 various areas in Benghazi city.

Water samples were aseptically collected in 500 ml sterile bottles. These bottles contained sodium thiosulphate for all water samples.

The water samples were directly transferred to the laboratory and examined within 1-6 hours of collection. Each sample was vigorously shaken and subjected to bacteriological examination.

1) **Enumeration of heterotrophic plate count (HPC) using standard pour plate method**.

It was done using the standard pour plate method. Water samples were subjected to serial 10- fold dilutions using sterile peptone water, and one ml of each sample and / or the appropriate dilutions were dispensed into sterile glass Petri dishes. Duplicate petridishes for appropriate dilutions were inoculated. Then 15 ml volumes of molten sterile plate count agar (PCA) tempered approximately to 45ºC was aseptically poured into each plate. The agar was thoroughly mixed with the sample aliquots by gentle to and fro, clockwise and anticlockwise movements. The agar was allowed to set and solidify. The plates were

then inverted and incubated at 37ºC for 48 hours. The plates showing 30-300 colonies were counted using Quebec colony counter and expressed as cfu /ml of the sample

2) Enumeration of TC by MTF.

i. Presumptive phase

Water sample volumes of 50 and 10 ml were used .The 50 ml volumes were inoculated into bottles containing 50 ml double strength lauryl tryptose broth (LTB) and the 10 ml volumes of water were inoculated into each of five replicate tubes each containing 10ml of double strength LTB.

All the inoculated bottles and tubes were shaken and incubated at 37°C for 24-48 hours. After 24 hours each tube and bottle was shaken gently and was examined for turbidity and/ or gas production. All bottles and tubes showing no gas or turbidity were re-incubated and re-examined at the end of 48 hours.

Production of growth and or gas in tubes or bottles within 48 hours was considered a positive result for presumptive test and was subjected to:

ii . Confirmed Phase:

Two loopfuls from each positive presumptive tube or bottle were inoculated into tubes each containing 5 ml single strength Brilliant Green lactose bile broths (BGLBB). All tubes were shaken on a vortex mixer and incubated at 37°C for 48 hours. Tubes showing turbidity and gas production were considered positive for confirmed phase. The most probable number (MPN) of TC/100 ml of water sample was calculated from MaCradey probability tables.

3) Enumeration of TC, TTC, FS, *P. aeruginosa* by MF.

Membrane filters

Sterile cellulose acetate membrane filters of 0.45um pore size and 47mm diameter were used.

Filtration unit:

The used filter holding assembly consisted of three glass funnels that fasten to three receptacles bearing porous plates to support

the membrane filters. The parts of assembly were sterilized by autoclaving 121° C f or 20 minutes.

Procedures were performed according to the methods described by Eaton *et al.* [20]

Counting:

- Plates showing 20 to 80 coliform colonies not more than 200 colonies of all types per membrane were counted using Quebec colony counter.
- Both typical and atypical coliform colonies were counted.
- Typical TC colonies on m-Endo agar were pink to dark red in color with a green metallic sheen.
- Atypical coliform colonies on m-Endo agar were dark red mucoid, or nucleated colonies without metallic sheen.
- The TC density was calculated by the following equation.

$$TC/100ml = \frac{\text{Coliform colonies counted}}{\text{ml of sample filtered}} \times 100$$

Verification:

Typical colonies and any atypical colonies were verified by picking at least 5 colonies from each plate and incubating them in LTB at 35° C for 48 hours and then in BGLBB at 35° C for 48 hours. Gas formed in LTB and confirmed in BGLBB within 48 hours verified the colony as a coliform.

For verified coliform counts, the initial count was adjusted based upon the positive verification percentage and was reported as "verified coliform counts/ 100ml."

Percentage verified coliforms =

$$\frac{\text{Number of verified colonies}}{\text{Total no. of coliforms colonies subjected to verification}} \times 100$$

3.a. Membrane Filtration Technique for TTC:[20]

It was performed in a similar manner to that employed for enumeration of TC but by using m-FC agar plates. These plates were inverted and incubated at 35-37°C for 2 hours and then incubated at 44.5 °C for 24 hours.

3.b.Membrane Filtration Technique for FS: [20]

It was performed in a similar manner to that employed for enumeration of TC but by using m-KF agar plates. Plates were left for 30 min, then inverted and incubated at 35°C for 48 hours.

3.c. Membrane Filtration Technique for *P. aeruginosa*: [20]

It was performed in a similar manner to that employed for enumeration of TC but by using m-PA agar plates. Plates were left for 30 min, then were inverted and incubated at 35°C for 48 hours.

Results:

According to WHO and Libyan guidelines, out of the 600 examined water samples 461(76.8%) were acceptable and 139(23.2%) were unacceptable.

The results of this study can be summarized as follows:

1) Table (1) showed that the highest percentage of acceptable samples was revealed from RO system (90.7%), followed by charcoal filter (83.3%), tap water DS (82.3%), and only 54.7% from tap water tanks.
2) Table (2) revealed that: a) The highest percentages of TC, TTC were isolated from tap water tanks (18.2% and 4.1%) respectively, followed by tap water DS (9.2% and 3.1%) respectively.

b) No FS were isolated from tap water or filter devices samples.

c) *P. aeruginosa* was isolated from filter water devices samples only (one sample from charcoal filter and three samples from RO filter system).

3) It is clear from tables (3) , (4) & (5) that:

a) The highest percentages of TC, TTC, *P. aeruginosa* and HPC were recorded from filter water devices samples of installment duration above 12 months.

b) No indicator organisms were revealed from water samples of RO filter systems installed for less than 12 months with the exception of only one unaccepted sample for TC.

c) Water samples of charcoal filters installed for 12-18 months revealed indicator organisms as TC 13(17.3%), TTC 5(6.7%) and *P. aeruginosa* one (1.3%).

4) Table (6) showed that the highest mean counts of HPC were revealed from tap water tanks (61.2 cfu/ml), followed by tap water DS (24.6 cfu/ml), charcoal filter (20.4 cfu/ml) and only (18.9 cfu/ml-) from RO system.

5) Table (7) revealed that TC were isolated from RO system and charcoal filter water samples when membranes were not changed according to manufacturer's instructions.

6) Table (8) demonstrated that RO system brand A was the best brand as regards the bacteriological quality and its compliance with the Libyan standards for drinking water showing acceptable samples 98.2%, followed by RO system brands B and C; (93.3 %) and (80.0%) respectively.

7) It is apparent from table (9) that an agreement of 88.2 % was found between both methods MF and MTF in detection of TC from the examined water samples.

Discussion:

The present study revealed that 23.2% of the 600 examined drinking water samples were bacteriologically unsatisfactory and were considered unacceptable as they failed to meet the Libyan guidelines. Nearly a similar percentage of 20% was reported by Brownell *et al*., [21] in Canada. Higher percentages were reported by Copeland *et al*.[22] in Brazil (30.3%) ,Mahjoub *et al*.,[23] in Libya (30.4%) , and Lamka *et al*.[24] in United States (35%). A much lower percentage was reported by Essa [25] in Egypt (3%).

Household water treatment is a practical strategy to prevent waterborne diseases. [26] RO system and charcoal carbon are commonly used in Libya. [27] RO

system in this study showed maximum reduction of microbial contamination and provided the highest percentage of acceptable water samples (90.7%). Nearly a similar percentage was reported by Payment *et al.,* [28] in United States (93%). A lower percentage was reported by Miles *et al*., [18] in United States (73%).

The charcoal filters generally improve the quality of household drinking water by removing objectionable tastes and odors as well as dirt, rust, and sand. [29] In this study, 83.3% of the 150 examined drinking water samples after charcoal filtration were bacteriologically satisfactory and met the Libyan guidelines. Johnston and Burt [30] reported nearly similar results for activated carbon (AC) filters (80.1%).

Taylor *et al* [31] tested four POU carbon filter devices, installed in the cold water line, they found that these devices removed 60-80 % of the influent chlorinated organic compounds when new filter cartridges were first put on line, but after 6-8 weeks, removal was reduced to < 10 % as a result of decreased effectiveness of the filters.

Although coliform organisms may not always be directly related to the presence of fecal contamination or pathogens in drinking water, the coliform test is still useful for monitoring the microbial quality of drinking water. [32]

This study revealed that TC were isolated from 18.2% of examined tank water samples, this could suggest bacterial regrowth, decrease in free chlorine during storage or old tanks. Much higher percentages were reported by Chaidez *et. al*., [33] in United States (32%), and Ojo *et. al*., [34] in Nigeria (40%).

TTC were considered more faecal specific than TC, and their detection was used routinely. [35] This study showed that 4.1% of the examined drinking water samples yielded TTC from tanks. A much higher percentage was reported by Chaidez *et. al*., [36] in United States (20%).

In the present study the frequency of isolation of TC and TTC from RO systems were (6.0%) and (2.0%) respectively. Lechevallier *et. al*., [37] reported isolation rates of (28.7%) and (3.1%) for TC and TTC from granular activated carbon (GAC) filters respectively. This could be explained by the difficulty to maintain free residual chlorine in all parts of the filters.

Some outbreaks of gastrointestinal illness have been attributed to drinking water contaminated with *P. aeruginosa*. This organism is capable of growth in low substrate conditions and in the presence of high chlorine levels. [38] In the present study, *P. aeruginosa* was isolated from filter water devices morning samples (one sample from charcoal filter and three samples from RO filter system). This could be due to the fact that RO systems have plastic storage tanks which might help the colonization of bacteria and their resistance to disinfection: Another factor may be attributed to the membranes being unchanged as recommended by the manufacturers. Nearly a similar result was reported by Vonberg *et al*., [39] where only one water sample yielded *P. aeruginosa* after filtration.

Three different brands of household filter devices designated A, B and C were examined in this study. It was found that RO system brand A was the best brand as regards the bacteriological quality and its compliance with the Libyan standards for drinking water (98.2%). It possessed the highest efficiency in purifying water, followed by RO system brand B and C; (93.3 %) and (80.0%) respectively, this could be due to accuracy, quality and efficiency of manufacturing parts of brand A. This was the same with the studied charcoal filters. In this present study , all the 55 water samples (100%) from brand A RO filter system had HPC <1 cfu/ml. Nearly a similar percentage (95%) was reported by Meckes *et al*., [40] in Canada.

In the present study as regards

installment duration of filter devices and presence of indicator organisms in examined drinking water samples after filtration process, high percentages were recorded from filter water devices samples of installment duration above 12 months. It could be explained by the possibility that with prolonged duration the devices hypothetical durability and efficiency lessens. Another explanation may be that membranes were not changed according to country of origin or manufacturer's instructions time. Reasoner *et al*., [41] reported that microbial colonization of POU devices containing AC occurred after prolonged installation. It is apparent from core analyses that the PAC contained high densities of HPC bacteria after several months of use, suggesting that trapped organic materials within the activated carbon supported growth of heterotrophic bacteria.

MF is a useful technique for the majority of water quality laboratories as it is a relatively simple method to use. Many samples can be processed in a day with limited laboratory equipment by a technician with basic microbiological training. Nevertheless, since this method is not sufficiently specific, a confirmation stage is needed, which could take a further 24 hours after the first incubation period in selective media. [42] This present study showed that out of (600) examined tap water samples, 68(88.2%) were positive for TC detection by MTF and MF methods and only 8 (11.8%) were positive by MTF method and negative by MF method. An agreement of 93 % was found between both methods in detection of TC.

Conclusions:

It can be concluded from this study that:

1. Filtered water samples were found to be superior to tap water samples as regards their bacteriological aspects.
2. The changing of the membranes of the filter devices according to the

manufacturerós instructions resulted in raised efficiency of filter devices where no indicator organisms were isolated from RO systems with changed membranes.

3. *P. aeruginosa* is considered as an excellent indicator for the efficiency of the water filters.

Recommendations:

1- Using water filters improves its quality, with the following measures:

A) To change filters' membranes according to manufacturer's instructions as regards duration and frequency.

B) To select a filter brand of high quality.

2- Guidelines of filtered water should be followed, considering the addition of *P. aeruginosa* as an indicator of filtered water quality.

3- A further research is needed to evaluate the extent and amount of carbon penetration through charcoal filters and RO systems.

Table (1): Results of the examined drinking water samples according to their source.

Water source		Number of examined samples	Acceptable samples		Unacceptable samples		χ^2 (p)
			No.	%	No.	%	
Tap water	DS	130	107	82.3	23	17.7	25.256 (<0.001)
	Tanks	170	93	54.7	77	45.3	
Filter water devices	Charcoal filter	150	125	83.3	25	16.7	73.433 (<0.001)
	RO system	150	136	90.7	14	9.3	
Total		600	461	76.8	139	23.2	

Table (2): Frequency of isolation of the indicator organisms from the examined drinking water samples according to their source.

Water source		Number of examined samples	TC		TTC		FS		P.aeruginosa		χ2(P)
			NO.	%	NO.	%	NO.	%	NO.	%	
Tap water	DS	130	12	9.2	4	3.1	0	0.0	0	0.0	0.723
	Tanks	170	31	18.2	7	4.1	0	0.0	0	0.0	
Filter water devices	Charcoal filter	150	16	10.7	6	4.0	0	0.0	1	0.7	1.000
	RO	150	9	6.0	3	2.0	0	0.0	3	2.0	
Total		600	68	11.3	20	3.3	0	0.0	4	0.7	

Table (3): Frequency of isolation of the indicator organisms from the examined drinking water samples according to the duration of installing the RO filter system devices.

Duration of installing RO devices months	Number of examined samples	Indicator organisms								MCp
		TC		TTC		FS		P. aeruginosa		
		No.	%	No.	%	No.	%	No.	%	
0-	30	0	0.0	0	0.0	0	0.0	0	0.0	1.000
6-	70	1	1.4	0	0.0	0	0.0	0	0.0	
12-18	50	8	16.0	3	6.0	0	0.0	3	6.0	
Total	150	9	6.0	3	2.7	0	0.0	3	2.0	

Table (4): Frequency of isolation of the indicator organisms from the examined drinking water samples according to the duration of installing the charcoal filter devices.

Duration of installing charcoal devices months	Number of examined samples	Indicator organisms								MCp
		TC		TTC		FS		P. aeruginosa		
		No.	%	No.	%	No.	%	No.	%	
0-	25	0	0.0	0	0.0	0	0.0	0	0.0	0.359
6-	50	3	6.0	1	2.0	0	0.0	0	0.0	
12-18	75	13	17.3	5	6.7	0	0.0	1	1.3	
Total	150	16	10.7	6	4.0	0	0	1	0.7	

Table (5): Geometric mean counts of HPC of examined filter water devices samples according to the duration of installing the devices.

Duration of installing the devices (months)	Number of examined samples (300)	HPC (cfu/ ml) charcoal filter	HPC (cfu/ ml) RO
		Mean	Mean
0-	55	1.1	2.04
6-	120	4.2	3.5
12-18	125	15.1	13.4
F (p) ANOVA		1.000	0.567

Table (6): Geometric mean counts of HPC of examined water samples according to water source.

Water source		HPC (cfu ml)
		Mean
Tap water	DS (130)	24.6
	Tanks (170	61.2
Filter water devices	Charcoal filter (150)	20.4
	RO (150)	18.9
F (p)		5.957* (<0.001)

Table (7): Relation between frequency of isolation of indicator organisms from filter water samples and changing of the filter device membranes according to the manufacturers' instructions.

Change of the membrane according to manufacturer's instructions		Indicator organisms								χ^2(P)
		TC		TTC		FS		P. aeruginosa		
		No.	%	No.	%	No.	%	No.	%	
Charcoal filter (150)	Yes*	3	2.0	0	0.0	0	0.0	0	0.0	<0.001
	No**	13	8.6	6	4.0	0	0.0	1	0.7	
RO system (150)	Yes	0	0.0	0	0.0	0	0.0	0	0.0	
	No	9	6.0	3	2.0	0	0.0	3	2.0	
Total 300		25	8.3	9	3.0	0	0.0	4	1.3	

Yes*: change of membrane according to the manufacturer**'s** instructions

No**: no change of membrane

Table (8): Comparison between results of tap water and RO filter system water samples according to the filter brand.

Water source / Result	Tap water (150)		RO water devices (150)								
			A(55)		B(45)		C(50)		Total		
	No.	%	No.	%	No.	%	No.	%	No.	%	
Acceptable samples	98	65.3	54	98.2	42	93.3	40	80.0	136	90.7	27.985 <0.001
Unacceptable samples	52	34.7	1	1.8	3	6.7	10	20.0	14	9.3	
Total	150	100	55	100	45	100	50	100	150	100	

A: Made in United States
B: Made in Thailand
C: Made in China

Table (9): Comparative efficiency of MTF and MF in TC detection in examined tap water samples.

TC by MF / TC by MTF	Positive		Negative		Total		Agreement %
	No.	%	No.	%	No.	%	
Positive	60	88.2	0	0.0	60	88.2	88.2%
Negative	8	11.8	0	0.0	8	11.8	
Total	68	100	0	0.0	68	100	

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