MICROBIOLOGICAL EVALUATION OF RESERVOIRS DRINKING WATER IN HURGHADA CITY
PART1: STUDIES ON SOME PHYSICAL, CHEMICAL PROPERTIES AND ISOLATION OF WATER BORNE PATHOGENIC BACTERIAL STRAINS FROM HURGHADA PLANTS AND RESERVOIRS DISTRIBUTED IN THE CITY.

By

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ABSTRACT

This study deals with the some physical, chemical and microbiological parameters of drinking water samples collected from Hurghada city which included five plants and twenty eight different types of reservoirs from different localities involved entire city where the total collected samples are 33.The isolation of water–borne pathogenic bacteria was conducted using 5 general and specific nutrient agar media for isolating Salmonella & Shigella, Gram negative bacteria, fecal coliform bacteria, *Vibrio cholera* and other Vibrio species. Ihe physical and chemical properties and the microbial analysis for water samples from water treatment plants of drinking water are in a compliance with the Egyptian standards (2007) for drinking water quality which recommended that the drinking water sample should be free from bacterial indicators and pathogens that is refer to success of chlorination processes in the plants. The samples collected from all reservoirs give high bacterial count on different nutrient media. The obtained results revealed that the microbial analysis of water samples from water reservoirs are not in a compliance with the Egyptian standards (2007) for drinking water quality which refer to storage condition for the drinking water are not suitable .

INTRODUCTION

Water is essential for life and plays a vital role in the proper functioning of the Earth's ecosystems. The pollution of water has a serious impact on all living creatures, and can negatively affect the use of water for drinking, household needs, recreation, fishing, transportation and commerce. The last decades, parallel with rapidly developing technology, increasing populations we have been witnessing alarming phenomena all over the world, the lack and bad quality of the drinking water (Chhatwal *et al*. 1993).

According to the World Health Organization, a third of the world population suffers from diseases derived from contaminated drinking water. It is widely acknowledged that the major threat to public health from drinking water is from microbial contamination (bacteria, virus, fungi, parasite and algae) coming from human, and to a lesser degree, animal feces. Disease causing organism are still in our environment and especially that part of the environment that is water would be foolhardy and deadly (Spellman, 1997). Water is not a medium for the growth of pathogenic microorganisms, but is a mean of transmission of the pathogenic microbes to the place where an individual inadvertently consumes it, and the outbreak begins (Spellman, 1997; Spellman and Drinan, 2000).

To ensure the safety of drinking-water supplies within the building system, pumbing practices must prevent the introduction of hazards to health by ensuring water storage systems are intact and not subject to intrusion of microbial and chemical contaminants (WHO, 2008).

Storage of samples for microbiological analysis:The time between sample collection and analysis should not exceed 6 hours, and 24 hours is considered the absolute maximum. The samples are

immediately kept in a lightproof insulated box containing melting ice or ice-packs with water to ensure rapid cooling. If ice is not available, the transportation time must not exceed 2 hours .It is imperative that samples are kept in the dark and that cooling is rapid. When water that contains or may contain traces of chlorine is sampled, the chlorine must be inactivated. If it is not, microbes may be killed during transit . The bottles in which the samples are collected should contain sodium hyposulfite to neutralize any chlorine present; the box used to carry samples should be cleaned and disinfected after each use to avoid contaminating the surface of the bottles and the sampler's hands (WHO, 1997).

Microbial hazards associated with drinking: Infectious diseases caused by pathogenic bacteria, viruses and parasites are the most common and widespread health risk associated with drinking-water. The public health burden is determined by the severity of the illness(es) associated with pathogens, their infectivity and the population exposed.Breakdown in water supply safety may lead to large-scale contamination and potentially to detectable disease outbreaks. Other breakdowns and low-level, potentially repeated contamination may lead to significant sporadic disease, but is unlikely to be associated with the drinking-water source by public health surveillance. Quantified risk assessment can assist in understanding and managing risks, as those associated with sporadic disease (WHO, 2008).

Water characteristics:

A- Physical characteristics:

1) Taste and odor: Taste and odor problems in surface waters are usually caused by algae, actinomycetes and microorganism, taste is used to determine the acceptability of drinking water from a judgment based on sensory evaluation. Odor of water is created by organic chemicals- or natural process of decomposition of vegetable matter or microorganisms activity (Wiley & Sons, 1997).

2)pH:Raw water examined for use as drinking water has an expected pH value between4-9but,more than likely,encountered value will be between 5.5-8.6(Wiley & Sons, 1997).

3)–Turbidity: Caused by the presence of suspended matter as clay, silt, and fine particles of organic and inorganic matter, plankton, and other microscopic organisms. The major problem associated with turbidity is it`s effect on disinfection, because high levels protect microorganisms from the action of disinfection (WHO, 1997).

4) Temperature: An ideal water should have constant temperature or with minimum variation, between 8-14 ºC.Temperature has an influence on the treatment of water supplies and above 27 ºC is unsuitable and above 32 ºC is unfit for public use.(Wiley& Sons, 1997).

5) Color: Color when noticed in drinking water by the consumer, is an objectionable characteristic that would make the water supply psychologically un acceptable (Wiley& Sons, 1997)

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Table (1): Guide value of physical character of water according to Egyptian standards (2007).

Physical character	Unit	Guide Value
Color	)Co/Pt(	Colorless
Temperature	°C	8-14^{O}c
pH	-log H^{+} concentration	6.5 - 8.5
Turbidity	NTU	0-1

Nephelometric turbidity units (NTU).

B-Chemical characteristics:

1) Hardness: Hardness may be considered a physical or chemical parameter of water. It represents the total concentration of calcium and magnesium ions, reported as calcium carbonate. Other metallic ions may be present in specific cases in such a considerable amount as to require inclusion in hardness reporting quantity. Therefore, it can also be defined as the sum of polyvalent cations present in the analyzed

water. Originally, hardness was examined and evaluated in raw water sampling as an indicator of water quality in terms of precipitating soap (Wiley & Sons, 1997). The taste threshold for the calcium ion is in the range of 100–300 mg/liter, depending on the associated anion, and the taste threshold for magnesium is probably lower than that for calcium. In some instances, consumers tolerate water hardness in excess of 500 mg/liter (WHO, 1997).

2)-Residual chlorine: Most individuals are able to taste or smell chlorine in drinking-water at concentrations well below 5mg/litre, and some at levels as low as 0.3 mg/liter. At a residual free chlorine concentration of between 0.6 and 1.0 mg/liter, there is an increasing likelihood that some consumers may object to the taste (WHO, 2003).

Table(2):Guide value for some chemicals in drinking water according to Egyptian standard by ministry of health 2007.

Parameter	unit	Guide Value
Residual Chlorine	mg / l	5
Ammonia	mg / l	0.5
Nitrates	mg / l	45
Nitrites	mg / l	0.2
Total Hardness	mg / l	500

3)-Ammonia:The threshold odour concentration of ammonia at alkaline pH is about1.5 mg/l., and a taste threshold of 35 mg/l. has been proposed for the ammonium action.Ammonia has a toxic effect on healthy humans if the intake becomes higher than the capacity to detoxification (WHO, 1997).

4)-Nitrate and nitrite: Nitrates could be detected in soil and consequently are widespread in the environment from food to atmosphere and water. Higher concentrations are expected where fertilizers are used, in decayed animal and vegetable matter, in leakages from sludge and refuse disposal, and in industrial discharges. Nitrates are always higher in concentration than nitrites (Wiley & Sons, 1997).

AIMS OF THE STUDY.

A-Evaluate microbial pollution in drinking water reservoirs in hurghada city by isolate water-borne pathogenic bacteria on different types of media.

B-Identify these isolated water-borne pathogenic bacteria.

C-Study some physiological characteristics for these isolated bacteria.

D-Study the effect of some antibiotics on growth and virulence of the isolates with reference to the following:- 1- The city located at remote provinces, which does not contain any branch of the River Nile, but the population depends mainly on water storage.2 - The city has a great economic importance & it is very famous for tourism in the world and therefore contains a large number of workers in the tourism sector. 3 - The city is the capital of Red Sea province with a population of the largest cities if compared with the rest cities in this province. 4– Most of complaints of citizens in the city on the contamination of drinking water.

Sources of drinking water in Hurghada city: The people in Red Sea Governorate depend on the storage of drinking water in reservoirs where the River Nile is far from this area. Hurghada was taken as the one city of this Governorate as an example to measure the microbial pollution in drinking water. There are the line of pipes extend from Giza to feed Hurghada city with drinking water through many water plants which feed the people by water. There are different type 5 of reservoirs in Hurghada city such as (metal, fiber plast ,and in the new building in Hurghada such as moubarak building 5 have ground reservoirs), the majority types f reservoirs found in Hurghada is metal because it is cheap, most

of people don't clean their reservoirs for along period of time.

This study will be carried out during three parts:

1-(Part 1): Study some physical, chemical properties and isolation of water borne pathogenic bacteria from Hurghada plants (5 plants) and reservoirs (28 reservoirs) distributed to entire city.

2-(Part2): Characterization of 110 water-borne pathogenic bacteria from drinking water reservoirs.

3- (Part 3): Study some physiological properties of twenty two (22) drinking water-borne pathogenic bacteria and the effect of different antibiotics on these isolates to control it`s growth and virulence.

MATERIAL AND METHOD

The present study was carried out to evaluate and determine the bacterial pollution of drinking water in Hurghada city by monitoring some bacterial indicators and pathogens. The total numbers of samples are (33) from water plants and reservoirs during a period from March 2007 to February 2008.

Plan of the study:1-Collecting drinking water samples from the five plants in hurghada city.

2-Collecting drinking water samples from water reservoirs in Hurghada city which include ten different type reservoirs.3-Study of some physical parameter as pH, turbidity, conductivity, temperature, and chemical parameter as total hardness, residual chlorine, ammonia, nitrate and nitrite for the samples.

4-Isolation of different bacteria by membrane filtration method on different types of media.

1-Sampling: Dechlorinated treated drinking water samples collected from the outlet of five treated plants (feeding from the River Nile) located in Hurghada city and twenty eight samples from different types of reservoirs distributed in Hurghada areas in dark glass bottles were covered by aluminum foil, and autoclaved at 121°C for 20 minutes. Some samples were taken directly from reservoirs and others were taken from the tap where the tap opened for some time and the samples were collected in a clean and sterile bottles. In case of chlorinated water samples the bottles were charged with sodium thiosulphate (0.1 ml of 3% solution) to neutralize residual chlorine. The time between sample collection and analysis not exceed 6 hours according to Guideline for drinking water quality (WHO ,1997).

II-Isolation media

A) Nutrient agar (NA) medium (APHA, 1998): contained the following (g/l*):* Beef extract, 3;peptone, 5; sodium chloride, 5; and agar, 15.All ingredients were dissolved in distilled water and completed up to 1000 ml. pH of the medium was adjusted at 6.8 and sterilized at 12 1°C for 15 min.

B)Salmonella *&* Shigella (SS) agar medium (Barrow and Feltham,1993). The Salmonella *&* Shigella-agar medium consists of the following (g/l): Proteose peptone, 5; yeast extract, 4; lactose, 10; sodium thiosulphate, 5.75; sodium citrate, 7.75; ferric citrate, 1.0; bile salt, 8.5; neutral red, 0.025; brilliant green, 0.00033; and agar, 15.

Salmonella *&* Shigella agar ready made medium (Biolife, Milane, Italy) was prepared by adding54 g to l000ml distilled water, dissolved, stirred frequently, and steamed for 2 min; avoid any over heating and adjusted pH at 7.0. The medium was cooled to 45-50°*C* and distributed into sterile Petri-plates under aseptic conditions.

C) MacConkey agar (MA) medium: (APHA, 1998): contained the following (g/l)*:* Peptone,20; sodium chloride,5.0;sodium taurocholate,5.0; distilled water was completed to 1000ml.All ingredients were dissolved in water, and then heated. pH was adjusted at 8.0, the ingredients were then boiled for 20 min, cooled, and filtered. Agar, 20; lactose, 10; neutral red (1% aqueous solution), 10 ml. after filtration of the ingredients which were mixed well, pH was adjusted at 7.4 and then sterilized at 115°C for 20 min. This medium was distributed into sterile Petri-plates under aseptic conditions.

D) Thiosulphate citrate bile salt sucrose medium(TCBS).(Corry,*et al*,2003):This medium contained the following (g/l):

peptone,10.0;Yeast extract,5.0;Oxbile,8.0; Sodium thiosulphate,10.0; Sodium chloride,10.0; Sodium citrate,10.0; Sucrose,20.0;Ferric citrate,1.0; Bromothymol blue ,0.04 ; agar,14.0; distilled water was completed to 1000 ml.All ingredients were dissolved in water, pH was adjusted at 7.4 The medium was distributed in sterile Petri-plates under aseptic conditions.

E)Endo agar media (APHA, 1998):consists of (g/l):
yeast extract,1.2.csitone,3.7;thiophene,3.7; lactose,9.4;tryptose.7.5;Dipotassium hydrogen phosphate , 3.3 ; potassium dihydrogen phosphate,1.0; sodium chloride,3.7;sodium desoxycholate,0.1;sodium laurryl sulfate,0.05; sodium sulfite,1.6;basic fuchsin,0.8; agar,15; distilled water up to 1000 ml .All ingredients were dissolved in water, and20 ml of 95 % ethanol were added, mixed well, heat with frequent agitation and boil for 1 minute to dissolve the ingredients, pH was adjusted at 6.6 and was distributed into sterile Petri-plates under aseptic conditions.

III:- Membrane filter test (MF):The MF test allows scientist to conform the presence and estimate the number of pathogenic bacteria in the sample, it is easier to perform than the MPN test (Most probable number) because of less handling. In this technique, a measured amount of water (usually 100 ml for drinking water is pass through a membrane filter (pore size 0.45 μm) that traps bacteria on its surface. then membrane is then placed on a thin absorbent pad that has been saturated with a specific medium designed to permit growth and differentiation of the organisms being sought the success of the method depends on using effective differential or selective media that can facilitate identification of the bacterial colonies grown on the membrane filter surface(APHA 1998).

IV .Purification of water pathogenic bacteria: The Purification procedure was carried out by the agar steak plate method .All colonies of different forms and color showing separate growth on the different agar media were picked up and restreaked following the agar streak method onto the agar surface of plate. At the end of incubation period, only the growth which appeared as a single separate colony of distinct shape and color was picked up and restreaked again for several consecutive times onto the surface of agar plate of isolation media to insure its purity. Purity was checked up microscopically and morphologically using Gram stain. Pure isolates were sub cultured on slants of its specific isolation medium and kept for further investigation. The purified colonies were prepared to be used for a complete identification process and other studies.

IIV. Physical and chemical analysis: Physical parameters were measured by using laboratory equipment such as pH meter, Conductivity meter, and thermometer, and Chemical parameter such as Residual Chlorine , Hardness, Ammonia, Nitrate and Nitrite were measured according (APHA. 1992)standard methods for the examination of water and waste water 18 th edition

RESULTS

Isolation of bacteria: Thirty three samples were collected from hurghada city include five samples from five plants in hurghada city and twenty eight samples collected from different locations &different type or reservoirs and the bacterial isolates was conducted using five general and specific agar media,.

1-Nutrient Agar(NA).general media.2-Salmonella & Shigella Agar (SS), for isolating Salmonella species & Shigella species 3- MacConkey Agar (MA) for isolating gram negative bacteria.4- Endo Agar(EA)for isolating total coli form at 35 °C (TC) and fecal coliform at 44.5 °C (FC).5-Thiosulphate citrate bile salt sucrose (TCBS) for isolating *Vibrio cholera* and *Vibrio* species.

The collections of these samples are divided into four groups.

Group (A): Involves five plants found in hurghada city, these plants are listed in table (3).

Group (B): Involves ten reservoirs from different locations are listed in table (6).
Group(C): Involves nine reservoirs from different locations are listed in table (8).
Group (D): Involves nine reservoirs from different locations are listed in table (٩1).

Group (A) as shown in table (3) was collected from the plants in Hurghada and there are five plants in Hurghada as follow:-
1- EL- kilo 59(M1)The main plant which receives the water from krimat source (Giza pipe line, river Nile water) then it distributes this quantity of water to other plants. , 2-El - Dahar plant (M2).3- EL – Mina Plant (M3). 4- EL- kilo 56 Plant (M4). 5- EL –Yoser Plant (M5) this produce water from sea water by treatment process via (R.O) reverse osmosis method and feed this water to people after mixing by fresh drinking water.
Note: all plants make chlorination to disinfect water before pump the water to the people reservoirs.

Table (3): Names of five plants found in hurghada (Group A).

Sample number	Sample Code	Sample site
1	M1	EL- kilo 59 Plant
2	M 2	El - Dahar Plant
3	M 3	EL - Mina Plant
4	M 4	EL- kilo 56 Plant
5	M 5	EL - Yoser Plant

Bacterial Count for samples which were collected from Hurghada (group A): It was found that there are no detected bacterial count in group A on the media used which may be due to chlorination processes in the plants (Table5) in which the physico-chemical parameters reveal that the residual chlorine values are identical to the Egyptian standards (2007).

Table(4):Physico-chemical parameters of samples collected from Hurghada Plants (group A).

Sample / Physical and chemical parameter	Plants M1	M2	M3	M4	M5
Turbidity (NTU)	0.2	0.4	0.2	0.7	0.4
pH	8.04	8.5	7.9	7.9	8.5
Temperature (TC°)	26.5	25.5	27.2	27	25.5
Residual chlorine(mg/l)	1	0.8	0.9	0.8	1.2
Conductivity (ms/m)	37.9	49	39.5	45.5	124.7
Total Hardness (mg/l)	130	140	120	110	240
No2 (mg/l)	0.01	0.01	0.05	0.01	0.02
No3 (mg/l)	0.9	0.4	0.9	0.7	0.6
NH3 (mg/l)	0.01	N.D	0.03	0.03	N.D

Note: (N.D) means not detected.

Group (B) as shown in table (6) it involved ten reservoirs found in these sites as the follows:-
1- EL –Nasre area (Plastic reservoir)., 2- Building 144 area (Metal reservoir).3- Building 144 area (Plastic reservoir)., 4- Government Building area (Metal reservoir).5- Megahed area (Metal reservoir)., 6- Mubarak Five building area (Ground reservoir).7- Zahran area (Metal reservoir)., 8- Stadium area (Metal reservoir)., 9- Bus station area. (Metal reservoir)., 10- El – Dahar Square area (Metal reservoir).

Table(5):Reveal sample sites, sample code & types of reservoirs for group B.

Sample no.	Sample site	Sample Code	Type of reservoirs
1	EL –Naser area	Hp1	Plastic
2	building 144area	Hm2	Metal
3	building 144area	Hp3	Plastic
4	Government Building area	Hm4	Metal
5	Megahed area	Hm5	Metal
6	Mubark Five building area	HG6	Ground
7	Zahran area	Hm7	Metal
8	Stadium area	Hm8	Metal
9	Bus station area	Hm9	Metal
10	El – Dahar Square area	Hm10	Metal

Table(6):Bacterial Count for samples collected from Hurghada houses reservoirs (group B).

Media / Sample	Isolation media					
	NA CFU/100 ml	TCBs CFU/100 ml	TC CFU/100 ml	F C CFU/100 ml	M A CFU/100 ml	SS CFU/100 ml
Hp1	40	7	NIL	NIL	1	NIL
Hm2	2040	448	644	250	238	106
Hp3	5950	168	440	396	272	420
Hm4	1300	550	472	160	720	272
Hm5	8200	180	144	130	890	167
HG6	7600	600	122	4	500	170
Hm7	1770	912	380	116	1100	125
Hm8	5520	396	252	4	980	134
Hm9	2400	400	396	8	640	142
Hm10	5400	4	NIL	NIL	460	108

Note: (NIL) means no colony Appear

Group (C): as shown in table (9) it involved nine reservoirs found in these sites as follows:-

1- School area (Metal reservoir) 2- El-Sakala-behind Policeman building area (Plastic reservoir).

3- Sheraton area (Metal reservoir). 4- Fishermen port area (Plastic reservoir).5- Abu Nawas area (Metal reservoir).6- Al – Zahra area (Metal reservoir).7- Hafre El-Baten area (Metal reservoir).8- Traffic area (Metal reservoir).9- Hafre -El Baten area (Metal reservoir).

Table(7): Physico-chemical parameters of samples collected from Hurghada houses reservoirs (group B).

Sample / Physical & chemical parameter	Samples									
	Hp1	Hm2	Hp3	Hm4	Hm5	HG6	Hm7	Hm8	Hm9	Hm10
Turbidity (NTU)	0.9	0.7	0.6	0.7	0.8	0.3	0.9	0.7	0.7	0.5
pH	8	8.5	8.4	7.5	8.5	7.9	8.1	8	7.5	7.9
Temperature (TC°)	28	27.2	27.8	26.7	27.6	25	26.5	26.5	27.1	26.7
Conductivity (ms/m)	50	40.5	45.6	52.7	52.8	46.5	52.5	96.5	44.2	42.1
Total Hardness (mg/l)	140	220	120	140	142	200	142	134	140	140
NO_2 (mg/l)	N.D	N.D	0.04	N.D	N.D	0.05	N.D	0.01	0.01	N.D
NO_3(mg/l)	1.1	0.96	1.02	0.8	1.3	1.04	1.1	0.75	1.07	1.02
NH_3 (mg/l)	0.05	0.35	0.07	0.08	N.D	0.11	0.06	0.11	0.10	0.10

Note: (N.D) means not detected.

Table(8): Reveal sample sites ,sample code & types of reservoirs for group C.

Sample no.	Sample site	Sample Code	Type of reservoirs
1	School area	Hm11	Metal(cool water)
2	El- Sakala behind Policeman building area	Hp12	Plastic
3	Sheraton street area	Hm13	Metal
4	Fishermen port area	Hp14	Plastic
5	Abu Nawas area	Hm15	Metal
6	Al-Zahra area	Hm16	Metal
7	Hafre El baten area	Hm17	Metal
8	Traffic area	Hm18	Metal
9	Hafre al baten area	Hm19	Metal

Table (9): Bacterial Count of samples collected from Hurghada houses reservoirs (group C).

Media / Sample	Isolation media					
	NA CFU/100ml	TCBs CFU/100ml	TC CFU/100ml	F C CFU/100ml	M A CFU/100ml	SS CFU/100ml
Hm11	130	50	NIL	NIL	NIL	NIL
Hp12	1950	850	420	90	1520	760
Hm13	1440	600	NIL	NIL	1280	900
Hp14	45	NIL	NIL	NIL	NIL	NIL
Hm15	1600	100	6	NIL	NIL	NIL
Hm16	7500	NIL	14	NIL	50	NIL
Hm17	350	30	8	NIL	NIL	10
Hm18	380	NIL	10	NIL	NIL	60
Hm19	420	30	12	NIL	NIL	10

Note: (NIL) means no colony Appear.

Table(10): Physico-chemical parameters of samples collected from Hurghada houses reservoirs (group C).

Physical and chemical parameter / Sample	Samples									
	Hm11	Hp12	Hm13	Hp14	Hm15	Hm19	Hm16	Hm17	Hm18	Hm19
Turbidity (NTU)	0.5	0.9	0.8	1	0.2	0.8	0. 9	0.2	0.6	0.8
pH	8.1	8	8.2	8.1	7.8	8.2	8.2	7.5	8.1	8.2
Temperature (TC°)	16.5	27.2	27.6	26.9	27	27	26.5	25.5	25	27
Conductivity (ms/m)	42	44.8	62.4	43.6	53	42.6	53.8	42.6	65.4	42.6
Total Hardness (mg/l)	150	140	180	142	210	210	142	150	142	210
NO_2 (mg/l)	N.D	0.01	0.04	0.01	N.D	0.01	N.D	N.D	0.01	0.01
NO_3 (mg/l)	1.8	1.3	1.3	1.2	1.06	1.04	1.2	1.03	1.6	1.04
NH_3 (mg/l)	0.06	0.05	0.05	0.09	0.04	0.45	0.03	0.08	0.56	0.45

Note: (N.D) means not detected.

Group (D):- as shown in table (12) it involved nine reservoirs found in these sites as follows:-

1- Al Nagda area (Plastic reservoir). 2- EL Slam area (Plastic reservoir). 3- EL Slam area (Metal reservoir) 4- Mubark Five building area (Ground reservoir). 5- AL Amal area (Metal reservoir).

6- El malaha area (Metal reservoir). 7- Abu Ashara area (Metal reservoir). 8- Arab area (Metal reservoir). 9- Al- ahyaa area(Metal reservoir).

Table (11): Reveal sample sites, sample code & types of reservoirs for group D.

Sample number	Sample site	Sample Code	Type of reservoirs
1	Al Nagda area	Hp20	Plastic
2	EL Slam area	Hp21	Plastic
3	EL Slam area	Hm22	Metal
4	Mubarak Five building area	HG23	Ground
5	AL Amal area	Hm24	Metal
6	El malaha area	Hm25	Metal
7	Abu Ashara area	Hm26	Metal
8	Arab area	Hm27	Metal
9	Al- ahyaa area	Hm28	Metal

Table (12): Bacterial Count of samples collected from Hurghada houses reservoirs (group D).

Sample	Isolation media					
	NA	TCBs	TC	F C	M A	SS
Hp20	3800	30	88	2	40	230
Hp21	2600	510	144	80	720	950
Hm22	1520	950	152	14	140	160
HG23	1540	336	340	4	40	130
Hm24	3500	2400	230	171	1520	190
Hm25	1520	360	45	11	1140	360
Hm26	1052	150	350	150	600	580
Hm27	1440	105	200	190	135	NIL
Hm28	420	90	60	12	648	370

Note: (NIL) means no colony Appear.

Table(13): Physico-chemical parameters of samples from Hurghada houses reservoirs (group D).

Sample / Physical and chemical parameter	Samples									
	Hp20	Hp21	Hm22	HG23	Hm24	Hm28	Hm25	Hm26	Hm27	Hm28
Turbidity (NTU)	0.9	1	0.6	0.3	0.5	0.8	0.7	0.6	0.4	0.8
pH	7.9	7.9	8	8.1	8	7.5	7.5	7.6	8	7.5
Temperature (TC°)	26.2	27	26.4	25.6	26	26	26	25.5	27	26
Conductivity	56.3	59.6	68.5	58.9	42.5	42.5	40.5	43.6	43.3	42.5
Total Hardness	134	186	250	130	140	210	142	160	210	210
NO_2 (mg/l)	0.01	0.04	N.D	N.D	N.D	N.D	N.D	0.01	N.D	N.D
NO_3 (mg/l)	1.07	1.03	1.02	1.03	1.1	1.1	1.2	1.08	1.1	1.1
NH_3 (mg/l)	0.587	0.065	0.045	0.053	0.040	0.03	0.03	0.03	0.03	0.03

Note: (N.D) means not detected.

After finishing the count of each plate by using colony counter equipment, the negative plates were excluded where positive plates were taking to purified microorganism.

After Purification for these isolated bacteria. One bacterial isolate from each plate were taken for investigation of the morphological and physiological characteristics (biochemical tests)to identify it (110 bacterial Isolates) by using the keys of Cowen and steel's Manual for the Identification of medical bacteria (Barrow and Feltham, 1993) Bergey's Manual of determinative Bacteriology 9th edition (Hensyl, 1994). Bergey's Manual of systematic Bacteriology. 2nd edition. Volume 2 part B (George M. Garrity. *et al* (2005).

DISCUSSION

The greatest risk of waterborne diseases are infants and young children, people who are

debilitated or living under unsanitary conditions. Safe drinking-water is suitable for all usual domestic purposes, including personal hygiene (WHO, 2008). In this study the physical and chemical properties, the pH, turbidity,total hardness, nitrites , nitrates and ammonia of the samples of drinking water are in a compliance with the Egyptian standards (2007) for drinking water quality which recommended that the drinking water sample should be free from bacterial indicators(eg TC,FC) and pathogens (eg. *Salmonella*).

There are three different groups of microorganisms that can be transmitted via drinking water (Viruses, bacteria and Protozoa) (Mason,1996) Bacteria are the most important group in terms of frequency of isolation in drinking water and reported outbreaks of diseases (Wiley & Sons, 1994).

In this study the membrane filter technique was used for detecting bacteria in water. Because of the MF technique significant advantage over the MPN (The most probable number) method, the examination of larger volumes of water is feasible, which leads to greater sensitivity and reliability. MF also offers a quantitative enumeration comparatively to the semi- quantitative information given by the MPN method.

In the present study no bacterial growth could be detected in five plants which feed the people by drinking water in Hurghada city .this results may be refer to the successes of chlorination processes in these plants by added the suitable proportion of chorine compound where the value of Residual chlorine(mg/l) range from 0.8 mg/l to 1.2 mg/l.

For purposes of disinfection of municipal supplies, chlorine is applied primarily in two forms: as a gaseous element, or as a solid or liquid chlorine-containing hypochlorite compound. Gaseous chlorine is generally considered the least costly form of chlorine that can be used in large facilities. Chlorine is shipped in cylinders, tank cars, tank trucks, and barges as a liquefied gas under pressure. Chlorine confined in a container may exist as a gas, as a liquid, or as a mixture of both. Thus, any consideration of liquid chlorine includes consideration of gaseous chlorine (Troyan & Hansen, 1989).

Hypochlorite forms (principally calcium or sodium) have been used primarily in small systems (less than 5,000 persons) or in large systems where safety concerns related to handling the gaseous form outweigh economic concerns. Present day commercial, high-test calcium hypochlorite products contain at least 70 percent available chlorine and are usually shipped in tablet or granular forms. Sodium hypochlorite is provided in solution form .containing 12%or15% available chlorine (Troyan & Hansen, 1989).

Generally, disinfection is accomplished through the addition of an oxidant. Chlorine is, by far, the most common disinfectant used to treat drinking water; but other oxidants, such as chloramines, chlorine dioxide, and even ozone, are also used (Craun, 1993; Pepper, *et al*., 1996). Payment*et al*. (1985). found that most water treatment plants

continuously produce water that is free of detectable indicator bacteria. Where the samples collected from different types of reservoirs gave bacterial growth. which refer to the contamination of drinking water occurred during the storage process. This have been mentioned in the Guideline for drinking water quality by World Health Organization, 2008 as the follow (Further hazards and hazardous situations that can have an impact on storage reservoirs and intakes and that should be taken into consideration as part of a hazard assessment).

The media used in the present investigation are five media, one of which nutrient agar medium as common medium (not selective medium) and the others are selective media which are:-

(1) MacConkey agar medium, a selective medium used to isolate gram negative bacteria.

(2)Salmonella & Shigella -agar medium a selective medium used to isolate Salmonella & Shigella species.

(3) Thiosulphate citrate bile salt sucrose medium, a selective medium used to isolate Vibrio species.

(4)Endo agar medium, a selective medium used to isolate total coliform and fecal coliform bacteria

The obtained results revealed that the microbial analysis of water samples from water reservoirs are not in a compliance with the Egyptian standards (2007) for drinking water quality.

Surveillance should cover the whole of the drinking-water system, including sources and activities in the catchments, transmission infrastructure, treatment plants, storage reservoirs and distribution systems (WHO, 2008). According to World Health Organization the microbial contamination can be introduced to the distribution system through open treated- water reservoirs, for example water borne outbreaks have occurred in communities where birds contaminated the water either because the reservoir was uncovered or because they gained access to the reservoir through unscreened roof vents . Uncovered reservoirs can also permit the growth of toxin –forming cyanobacteria.

Mazari-Hiriart *et al* (2003). detected in 23% of drinking water samples, with significant differences between total coliforms, fecal coliforms, and fecal streptococci before and after chlorination. Freshwater bacteria are a very diverse assemblage of prokaryote organisms, varying in their morphology, physiology, and ecological preferences (Wiley& Sons, 2005).

Finally: the characterization of the purified bacterial isolates using different types of nutrient media will be prepared in the next article of this study.

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