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Microbial Evaluation of Some Groundwater Resources for Human Domestic Consumption in the Western Area of the River Nile Basin of Assuit Governorate, Egypt

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Abstract:A microbiological water analysis is mainly based on the concept of fecal indicator bacteria. This study was conducted to evaluate some private groundwater wells and groundwater well based-plants in the Assuit Governorate during the summer and autumn of 2013. With the assessment of the presence of thermotolerant fecal coliform and thermotolerant fecal streptococci bacteria as potent indicators for fecal contamination, we used multiple-tube fermentation or membrane filters for enumerating the themotolerant fecal coliform and thermotolerant fecal streptococci using the most-probable-number (MPN) index. In this study, we used 178 water samples. The results showed that there were 9 water wells not suitable to be consumed as drinking water for humans with an additional 5 other wells suddenly contaminated in the autumn so as to need a continuous sterilization system; 2 water well plants and 2 water networks needed replacement and renovation. In addition, 4 other water well plants and 4 water networks suddenly contaminated in the autumn had to be treated and disinfected before usage for drinking or human consumption. The physicochemical parameters of the water samples of the most studied locations were in permissible limits of the Egyptian standards for drinking water.

Keywords: Groundwater, Themotolerant fecal Coliform, Thermotolerant fecal Streptococci, and physicochemical parameters.

1.Introduction

The two sources of potable water supply in Egypt are groundwater and surface water, either from the River Nile or from the main irrigation canals. In 2008, the total drinking water production in Egypt was about 7.5 billion m3/year, the contributions from the Nile and groundwater being about 60% and 40%, respectively [14].

[18]Studied the evaluation of some private groundwater wells in the El-Rhawy (10 wells) and ManshiatRadwan (7 wells) regions, Giza governorate, Egypt. The total viable bacterial counts, total coliforms (TC), fecal coliforms (FC) and fecal streptococci (FS), as bacterial indicators, were examined. Ammonia, nitrates, sulfate, iron, total dissolved solids (TDS), chlorides, total hardness (CaCO3), biological oxygen demand (BOD), chemical oxygen demand (COD), pH, temperature, electric conductivity (EC) and turbidity were measured as physicochemical parameters of these wells. And, they showed that 11 wells were not suitable for drinking since they showed high total viable bacterial counts (>50 CFU mL^{-1}) and the presence of TC, FC and FS. In addition, some wells showed high concentrations of ammonia (n=16), iron (n=15), and turbidity (n=11) that exceeded the permissible limits of the Egyptian standards for drinking water, 2007.

The use of bacteria as indicators of the sanitary quality of water probably dates back to 1880 when Von Fritsch described *Klebsiellapneumoniae* and *K. rhinoscleromatis*as micro-organisms characteristically found in human feces [16]. In 1885, Percy and Grace Frankland started the first routine bacteriological examination of water in London, using Robert Koch's solid gelatine media to count bacteria [22]. Also in 1885, Escherich described *Bacillus coli* [31],renamed *Escherichia coli* by [3] from the feces of breast-fed infants (cited in [25]. Enterococci may be considered an essential part of the autochthonous microflora of humans and animals. Because of its wide distribution, Enterococci can also occur in different food commodities, especially those of animal origin [29, 23]. *Escherichia coli*is the predominant member of the facultative anaerobic portion of the human normal flora. *E. coli* is a member of the fecal coliform group and is a more specific indicator of fecal pollution than other fecal coliforms [27]. The presence of *E. coli* in drinking water is still considered to indicate that faecal contamination of water has occurred. *E. coli* monitoring of drinking water as a verification measure is a useful tool within a risk management approach to water quality. For more than 100 years, the microbial safety of drinking water has primarily been determined by testing for bacterial 'indicators' of fecal pollution, mainly *Escherichia coli* or alternatively thermotolerant fecal coliforms and total coliforms. These indicators are used to assess the potential public health risk of drinking water, and their presence or absence are key elements of most drinking water quality guidelines, water supply operating licenses and agreements between bulk water suppliers and retail water companies

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There are a number of other useful indicators, both microbial and physical, which can be used to monitor both drinking water system operation and performance, and which provide better support for system management than total coliforms [30]. Two key factors have led to the trend towards the use of *E. coli* as the preferred indicator for the detection of fecal contamination in drinking water [23].

[15]Stated that a faecal coliform and faecal streptococci of a ratio of four or greater may indicate human pollution; whereas, ratios of two or less may indicate animal pollution.

2. Materials and methods

2.1 Sampling

One hundred and seventy eight samples were collected in clean and sterile polypropylene plastic bottles from groundwater wells, expulsion of groundwater well plants, and normal tap water (Table 1); these bottles were covered with aluminum foil, and sterilized in an autoclave at 121ºC for 20 minutes. Sodium thiosulfate was used as a satisfactory dechlorinating agent that neutralized any residual halogen and prevented continuation of the bactericidal action during the sample transit. The time between sampling and analysis was not more than 6 hours [9].

Table (1): Different sampling locations:

Code	Location	Code	Location
1	Well No. 1, water well plant, Khalaf Rashid village	20	The expulsion of water well plant, Durunkha village
2	Well No. 2, water well plant, Khalaf Rashid village	21	Drinking water Network, Workshop Samir Carpentry, Durunkha village
3	Expulsion of water well plant, Khalaf-Rashid village	22	Well No. 4, water well plant, Alnamaysa village
4	Tap water from Mosque Emam Ali ibnAbi- Talib, Khalaf-Rashid village	23	Well No. 5, water well plant, Alnamaysa village water well plant
5	Well No. 2, water well plant, Al-Burah village	24	Well No. 7, water well plant, Alnamaysa village
6	Well No. 4, water well plant, Al-Burah village	25	Expulsion of Alnamaysa village tank
7	Tank expulsion of water well plant, Al-Burah village	26	Tap water, Drinking water network, Alnamaysa village
8	Tap water from water network of water well plant , Al-Burah village	27	Well No. 4, Seed water well plant,Seed
9	Well No. 2, water well plant, Musha village	28	Expulsion of water well plant, Farm of Assuiy University.
10	Well No. 3, water well plant, Musha Village	29	Well No. 1, water well plant, Mir
11	The expulsion of water well plant, Musha village tank	30	Well No. 2, water well plant, Mir
12	Drinking water of network of Musha village, Pharmacy Mahmoud Hassan	31	Well No. 5, water well plant, Mir
13	Well No. 5, water well plant, Shotp village	32	Well No. 6, water well plant Mir
14	Well No. 6, water well plant, Shotp village	33	Expulsion of water well plant , Mir
15	The expulsion of the Shotp village tank	34	Well No. 3, water well plant, Awlaad-Elias village
16	Drinking Water Network of Shotp village, unit ambulance	35	Well No. 6, water well plant, Awlaad-Elias village
17	Well No. 1, water well plant, village Durunkha	36	Expulsion of water well plant, Awlaad-Elias village
18	Well No. 6, water well plant, Durunkha village	37	Tap water from Dr. Heba-Farmacy, Awlaad-Elias, water network
19	Monastery Durunkha Village tank		

2.2. Laboratory Examination (Microbial Analysis)

a. Estimation of Coliform Group by the Multiple Tube Fermentation Technique (MPN)

i- Presumptive Phase:

Lauryl Tryptose Broth, abbreviated as LTB, was used in the presumptive phase of the Standard Total Coliform Fermentation Technique in the examination of water [4].

ii- Confirmer Phase:

The Brilliant green bile broth, 2%, was formulated according to AOAC and APHA [17] specifications for use in the confirmation of the presumptive tests for coliforms. The Brilliant green bile broth, gL^{-1}, for Total Coliform contained: Peptone 10.0, Lactose 10.0, Oxgall 20.0, and Brilliant green 0.0133Reagent-grade water 1 L. Thedehydrated ingredients were added to the water, mixed thoroughly, and heated to dissolve. The pH had to be 7.2 ± 0.2 after sterilization.

iii- Complete phase:

The EC Medium [1] developed by [26]was used for the detection of the coliform group and *E. coli*. This medium consisted of a buffered lactose broth with the addition of a 0.15% bile salt mixture. The growth of spore-forming bacteria was inhibited by the bile salts. The formation of gas in the Durham tube of the Brilliant green tubes, at any time within 48 ± 3 h, constituted a confirmed positive result. The formation of gas in the Durham tube of the EC tubes, at any time within 24 ± 2 h, constituted a confirmed positive result.

The MPN value of the number of positive Brilliant green lactose bile tubes and the EC tubes was calculate from the MPN index. In the case of inoculating one bottle with 100 ml of the sample portion, the report resulted as present or absent. The MPN values were for a variety of positive and negative tube combinations. The sample volumes indicated in the indexes illustrate the MPN values for the concentrations of Positive and Negative results when Five 20-ml or Ten 10-ml portions were used. This was a detailed procedure for the detection and enumeration of the Fecal Streptococcus group (FS) and Enterococcus group by using the Multiple Tube Technique in water samples in 48 hours or less on the basis of the reduction of TriphenylTetrazolium Chloride (TTC). Eosine Methylene Blue agar (EMB) was used for the isolation of *E. coli* [24].

In this method, *E. coli* was defined as coliform bacteria that possessed the enzyme β-glucuronidase and were capable of cleaving the fluorogenic substrate 4-methylumbelliferyl-β-D-glucuronide (MUG) with the corresponding release of the fluorogen when grown in EC-MUG medium at 44.5°C within 24 ± 2 h or less. The procedure was used as a confirmatory test after the prior enrichment in a presumptive medium for the total coliform bacteria [8].

iv-Physiological and biochemical examination of *E. coli*

Four to five suspected colonies from each bacterial plate were picked, cultured and then identified by the various biochemical tests.Biochemical tests were performed to confirm *E. coli* using Gram staining, Catalase test, Indole, Methyl red, Voges-Proskauer test, Nitrate reduction, Urease production, Simon citrate agar [6], and various sugar fermentation tests.

b. Isolation and identification of Themotolerant Fecal streptococci

i. Azide dextrose broth was used for the enumeration of fecal streptococci [35].

The medium was heated to boiling with agitation and the pH was adjusted at 7.2 before autoclaving at 121°C for 15 hours; then cooled to 45°C.

A positive test was indicated by turbidity (cloudiness) in the broth. A negative test remained clear. Azide Dextrose Broth tubes showing turbidity after 24 – 48 hours incubation had to be subjected to the Confirmed Test Procedure. Consult appropriate references for details of the Confirmed Test Procedure [4] and further identification of Enterococcus.

ii. Pfizer selective Enterococcus:

Pfizer Selective Enterococcus Agar was used for the selective isolation and cultivation of Enterococci. This medium was formulated as per Isenberg [19] by reducing the concentration of bile salts and sodium azide from the original formulation. The importance of esculin hydrolysis in differentiating Enterococci and streptococci was first reported by Rochaix as streptococci do not exhibit esculin hydrolysis [5].

iii. Presumptive Test Procedure:

A series of tubes of azide dextrose broth were inoculate with the appropriate graduated quantities of the sample; samples of 10 mL portions or less and double-strength broth for 10-mL inoculum were used. The portions usedvaried in size and number with the sample character. Only decimal multiples of 1 ml was used. Inoculated tubes were incubated at 35 ± 0.5°C. Each tube was examined for turbidity at the end of 24 ± 2. If no definite turbidity was present, it was reinsulated and read again at the end of 48 ±3 h.

iv. Confirmed Test Procedure:

All azide dextrose broth tubes showing turbidity after 24- or 48-h incubation were subjected to the confirmed test. A portion of the growth from each positive azide dextrose broth tube was streaked on the PSE agar. The inverted dish was incubated at 35 ± 0.5°C for 24 ± 2 h. Brownish-black colonies with brown halos confirmed the presence of fecal streptococci.

PSE agar (gL^{-1}):

Peptone C 17.0, Peptone B 3.0, Yeast extract 5.0, Bacteriological bile 10.0, Sodium chloride, NaCl 5.0, Sodium citrate 1.0, Esculin 1.0, Ferric ammonium citrate 0.5, Sodium azide, NaN3 0.25, Agar 15.0, and reagent-grade water 1 L.

The pH had to be 7.1 ± 0.2 after sterilization. The medium was held for not more than 4 h at 45 to 50°C before plates were poured.

Colonies showing esculin hydrolysis were analyzed for catalase activity. At least two catalase negative colonies from each plate were characterized by cultural and biochemical tests: Gram-staining reaction, growth in 6·5% NaCl broth, at 45 °C for 48 h and at 60 °C for 30 min, haemolysis on 5% blood agar, acid production from dextrose, mannitol, trehalose, and arabinose.

2.3. Physicochemical analysis:

Table (2): Physicochemical parameters of groundwater and analytical methods:

	Parameter	Method
1	pH	Digital pH meter.
2	Turbidity	Turbidimeter [10b]
3	Ca,	Ethylene diaminetetraacetic acid (EDTA) titrimetric method
4	Sulfate	Turbidimetric method (UV/Visible spectrophotometer, at wave length of 420 nm [9]
5	Chlorides	Silver nitrate titrimetric method [7]
6	Ammonia & Nitrate	Technicon Auto Analyzer.
7	Total dissolved solids (TDS)	Titrimetric method (Spectrophotometer at 600 nm)
8	Total hardness	EDTA Titrimetric Method [9]
9	Iron	The phenanthroline method
10	Manganese	The persulfate method

3. Result and discussion

3.1 Microbial analysis

Table 3reveals that there was contamination with thermotoleranr fecal coliform especially *E. coli* in the following locations: wells no.1, 2, expulsion of water well plant of Khalaf-Rashid village and tap water of its water network (MPN-Index/100ml= 2.6-4.6, code no. 1-4); tap water from water network of water well plant in Al-Burah village (MPN-Index/100ml= 8 for fecal coliform & 4.6 for *E. coli*, code 8); wells no. 5, 6 of water well plant, Shotp village (MPN-Index/100ml= 2.6, 4.6 *E. coli*, code no. 13, 14); well no. 7, water well plant Alnamaysa village tap water of its water network (MPN-Index/100ml= 2.6, 4.6 for fecal coliform, code no. 24, 26); well no. 4, water well plant, Seed (MPN-Index/100ml= 2.6 fecal coliform, code no. 27); wells no. 2, 6 Mir water well plant (MPN-Index/100ml= 4.6, 2.6 *E. coli*, code no. 30,32); and well no. 3, expulsion; tap water of water network of Awlaad-Elias village. There was no presence of fecal streptococci bacteria recorded in all tested samples.

There was no thermotolerant fecal streptococ*cus* spp. detected in any samples with one exception of the Monastery Durunkha Village tank (MPN-Index/100ml= 1.1, code 19).

We found thermotolerant fecal coliform bacteria in 15 locations of water samples (9 wells, 2 expulsions of water well plants, and 4 tap water samples) that indicated the fecal contamination of these locations. So, there were 9 water wells not suitable for consumption as drinking water for humans. There were 22 locations of water samples corresponding to the standard international specifications and Egyptian standards.

Table (3): Average of total viable indicator bacterial count in water samples (MPN-Index/100ml) of different locations in the summer of 2013:

Code	Indicator bacteria (MPN-Index/100ml)				Code	Indicator bacteria (MPN-Index/100ml)			
	T-Coliform	Fecal Coliform	*E. coli*	Fecal *Streptococci*		T-Coliform	Fecal Coliform	*E. coli*	Fecal *Streptococci*
1	4.6	4.6	4.6	_	**20**	<1.1	<1.1	<1.1	_
2	4.6	4.6	4.6	_	**21**	<1.1	<1.1	<1.1	_
3	2.6	2.6	2.6	_	**22**	1.1	1.1	1.1	_
4	2.6	2.6	2.6	_	**23**	<1.1	<1.1	<1.1	_
5	<1.1	<1.1	<1.1	_	**24**	2.6	2.6	1.1	_
6	<1.1	<1.1	<1.1	_	**25**	1.1	1.1	1.1	_
7	<1.1	<1.1	<1.1	_	**26**	4.6	1.1	1.1	_
8	8	8	4.6	_	**27**	2.6	2.6	2.6	_
9	1.1	1.1	1.1	_	**28**	<1.1	<1.1	<1.1	_
10	<1.1	<1.1	<1.1	_	**29**	<1.1	<1.1	<1.1	_
11	<1.1	<1.1	<1.1	_	**30**	4.6	4.6	4.6	_
12	<1.1	<1.1	<1.1	_	**31**	1.1	1.1	1.1	_
13	2.6	2.6	2.6	_	**32**	2.6	2.6	1.1	_
14	4.6	4.6	4.6	_	**33**	<1.1	<1.1	<1.1	_
15	1.1	1.1	1.1	_	**34**	4.6	4.6	4.6	_
16	<1.1	<1.1	<1.1	_	**35**	1.1	1.1	1.1	_
17	<1.1	<1.1	<1.1	_	**36**	2.6	2.6	1.1	_
18	<1.1	<1.1	<1.1	_	**37**	2.6	2.6	2.6	_
19	<1.1	<1.1	<1.1	<1.1					

The results in Table 4 reveal that in the autumn of 2013 there was fecal contamination for most samples with thermotolerant fecal Coliform and thermotolerant fecal streptococci as the following: wells no. 1, 2, expulsion, tap water of water well plant of Khalaf-Rashid village. (MPN-index/100ml= >8 *E. coli*, code no. 1-4); wells no. 2, 4 water well plant in Al-Burah village (MPN-index/100ml= 2.6 coliform & 1.1 *E. coli*, code no. 5, 6); wells no. 3, 4, expulsion, tap water of Musha water well plant (MPN-index/100ml= 4.6-2.6 coliform & 2.6-1.1 *E. coli* , code no. 9-12); well no. 5, expulsion, tap water of water well plant in Shotp (MPN-index/100ml= 2.6, 4.6, 4.6 coliform & 1.1, 2.6, 2.6 *E. coli*, code no. 13, 15, 16); wells 1, 6, monastery tank, expulsion of water well plant, tap water in Durunkha village (MPN-index/100ml= >8 *E. coli*, code no. 17-21); well no. 4, expulsion Alnamaysa tank, tap water in Alnamaysa village (MPN-index/100ml= 2.6 E. coli, code no. 22, 25, 26); wells 2, 5 water well plant, Mir (MPN-index/100ml= 2.6 coliform & 1.1, 2.6 *E. coli*, code no. 30, 31); and wells 3, 4, expulsion of water well plant, tap water in Awlaad-Elias village.

We found thermotolerant fecal coliform bacteria in the samples of 27 locations (14 wells, 1 tank, 6 expulsions of water well plants, and 6 tap water samples).

Thermotolerant fecal streptococci were found in: well no. 4, water well plant, Al-Burah village (MPN-index/100ml= 2.6, code no. 6); well no. 6, monastery tank, expulsion of water well plant, water network in Duronkha village (MPN-index/100ml= 8, 4.6, 2.6, 2.6, code no. 18-21); well no. 1, water well plant, Mir (MPN-index/100ml= 4.6, code no. 29); and wells no. 3, 4, expulsion, water well plant, water network in Awlaad Elias village (MPN-index/100ml= 4.6, 2.6, 2.6, 2.6, code no. 34-37).

Table (4): Average of the total viable indicator bacterial count in the water samples (MPN-Index/100ml) of different locations in autumn, 2013:

Code	Indicator bacteria (MPN-Index/100ml)				Code	Indicator bacteria (MPN-Index/100ml)			
	T-Coliform	Fecal Coliform	*E. coli*	Fecal *Streptococci*		T-Coliform	Fecal Coliform	*E. coli*	Fecal *Streptococci*
1	>8	>8	>8	1.1	20	>8	>8	>8	2.6
2	>8	>8	>8	1.1	21	>8	>8	>8	2.6
3	>8	>8	>8	1.1	22	2.6	2.6	2.6	1.1
4	>8	>8	>8	1.1	23	<1.1	<1.1	<1.1	<1.1
5	2.6	2.6	1.1	1.1	24	<1.1	<1.1	<1.1	1.1
6	2.6	2.6	1.1	2.6	25	2.6	2.6	2.6	1.1
7	1.1	1.1	1.1	1.1	26	2.6	2.6	2.6	1.1
8	1.1	1.1	1.1	1.1	27	1.1	1.1	1.1	<1.1
9	4.6	2.6	2.6	1.1	28	<1.1	<1.1	<1.1	<1.1
10	4.6	2.6	2.6	1.1	29	1.1	4.6	4.6	4.6
11	2.6	1.1	1.1	1.1	30	2.6	<1.1	<1.1	<1.1
12	2.6	1.1	1.1	1.1	31	2.6	2.6	2.6	1.1
13	2.6	1.1	1.1	1.1	32	<1.1	<1.1	<1.1	1.1
14	1.1	1.1	1.1	1.1	33	<1.1	<1.1	<1.1	1.1
15	4.6	2.6	2.6	1.1	34	>8	>8	>8	4.6
16	4.6	2.6	2.6	1.1	35	>8	4.6	4.6	2.6
17	>8	>8	>8	1.1	36	>8	>8	>8	4.6
18	>8	>8	>8	8	37	>8	>8	>8	4.6
19	>8	>8	>8	4.6					

There were 10 locations contaminated with thermotolarant fecal streptococci (5 wells, 1 tank, 2 expulsions, and 2 water networks). There were 10 locations of water samples corresponding to the standard international specifications and Egyptian standards.

Note: the total number of allowable MPN-Index/100ml of 95% samples = 2 colonies of coliform bacteria on condition that this number is not recurrent more than one time in the same samples from the same location is zero CFU of *Streptococcus* spp.(Egyptian standard for the quality of drinking water).

Fecal contamination of groundwater wells results from animal contamination or wells deep in domestic sewage in some houses near the location of groundwater wells. As for the contaminant expulsion of some water well plants, there is no sterilization with chlorine (chlorine system) and filtration of water for water wells that are very bad.

The indicator bacteria recorded in the study were thermotolerant coliforms and fecal streptococci. In temperate climates, it has been reported that 95% or more of

thermotolerant coliforms are *E. coli*, which are the preferred fecal indicator bacteria [39]. Fecal streptococci have been proposed as possible alternative indicator bacteria to *E. coli*. Previous literature has suggested that they have greater persistence in water and do not multiply in polluted environments. As a result, their use has been recommended for determining whether groundwater has received contaminated discharge (The logistic regression models for the presence of fecal streptococci (i.e., over 0 cfu 100 ml^{-1}) and numbers exceeding 10 cfu 100 ml^{-1} contain four and three sanitary risk factors, respectively). Isolation of fecal streptococci colonies were primarily related to the erosion of the backfill area, lack of fences, surface water uphill and rainfall occurring within the previous 48 h. The number of risk factors in the model for the number of fecal streptococci exceeding10 cfu 100 ml^{-1} were fewer and only eroded backfill, surface water uphill and rainfall within the previous 48 h were included. Population density was not included in the model for either water quality targets, which implies that the control on the presence of fecal streptococci was primarily related to pathway factors allowing the direct ingress of poorly disposed of fecal matter and not on sub-surface microbial loading [20].According to the WHO, the mortality of water associated diseases exceeds 5 million people per year. From these, more that 50% are microbial intestinal infections, with cholera standing out in the first place. In general terms, the greatest microbial risks are associated with ingestion of water that is contaminated with human or animal feces. Wastewater discharges in fresh waters and costal seawaters are the major source of fecal microorganisms, including pathogens [38]. Fecal streptococci also belong to the traditional indicators of fecal pollution. Fecal streptococci are Gram-positive, catalase-negative, non-sporeformingcocci that grow at 35°C in a medium containing bile salts and sodium azide. Cells hydrolyze esculin[36].

3.2 Isolation and identification of Escherichia coli and Streptococcus spp.:

A total of 120 isolates from environmental isolates were identified as *E. coli*, as confirmed by conventional laboratory tests using Gram staining, Catalase test, Indole, Methyl red, VogesProskauer test, Nitrate reduction, Urease production, Simon citrate agar, and various sugar fermentation tests. *Escherichia*, a member of *Enterobacteriaceae*, are oxidase-negative catalase-positive straight rods that ferment lactose. The cells were positive in the Methyl-Red test but negative in the Voges-Proskauer assay. The cells did not use citrate, did not produce H2S or lipase, and did not hydrolyze urea [12]. Fecal coliforms (or thermotolerant coliforms) are traditionally defined as coliforms that fermentlactose at 44.5°C in a medium with bile salts [28]. The detection of β-D-glucuronidase activity (at 44.5°C) is, generally, a good marker for fecal coliforms in environmentally polluted waters and very specific for *E. coli* [33].

A total of 62 isolates from environmental isolates were identified as fecal streptococci, as confirmed by conventional laboratory tests. About 90% of the isolates from the PSE agar were positive, characterized by cultural and biochemical tests: Gram-staining reaction, growth in 6·5% NaCl broth, at 45°C for 48 h and at 60°C for 30 min; haemolysis on 5% blood agar acid production from dextrose, mannitol, trehalose, arabinose, sucrose, and melezitose; arginine decarboxylation; reduction of tellurite, pyrrolidonylarylamidase, and phosphatase; susceptibility to optochin b-D-glucuronidase and b-D-glucoside a- D-galactoside; resistance to bacitracin and novobiocin. Some tests were carried out using an automatic system [10].

Physicochemical analysis (Table 5)

Hydrogen Ion Concentration (pH): ThepH values in the present study showed a slightly basic range of about 7.5 in well no. 2, Khalaf-Rashid and 8.67 in the expulsion of the water well plant, Durunkha.

Turbidity: Nephelometric Turbidity Unit (NTU) at all groundwater samples ranged from 0.2 well no. 6, Awlaad-Elias, water well plant and its tap to 9.64 well no. 1, Mir water well plant.

Sulfate: The sulfate values were found to be less in all groundwater samples. The minimum value of 3.6 mgL^{-1} was observed at the expulsion of the farm water well plant, Assuit University; while the maximum value of 11.92 mgL^{-1} was observed at well no. 4, water well plant, Al-Burah village. All sulfate concentrations in the groundwater samples were within the permissible limits (< 12 mgL^{-1}).

Chlorides: The minimum value of 15 mgL^{-1} of chloride was observed at well no.1, water well plant, Alnamaysa village; whereas, the maximum value of 140 mgL^{-1} was noted at well no. 7, water well plant, Durunkhavillage. The values were still within the permissible limits at both regions (< 250 mgL-1).

Total Dissolved Solids (TDS): The TDS levels ranged from 211 to 1163 mgL^{-1}. The minimum level was observed at well no. 4, Alnamaysa village; while the maximum level was observed at well no. 3, Musha village.

All TDS results were within the permissible limits (< 1000 mgL-1) with the exception of well no. 3, expulsion and tap water in Musha village.

Total Hardness (CaCO3): The total hardness values ranged from 221 to 466.4mgL^{-1}. The maximum value was observed at well no. 6, water well plant, Mir; while the minimum value was observed at the water network, Shotp village. The total hardness concentrations in all groundwater samples were within the permissible limits (< 500 mgL^{-1}).

Ammonia: The ammonia content of the samples was not detected in some samples; and in other samples it ranged from 0.07 in the tap water in Alnamaysa village to the maximum value was 1.95mgL^{-1} in well no. 4 in water well

plant, Seed. The results of the ammonia in most of the examined water samples were at the permissible limits (< 0.5 mgL^{-1}).

Iron: The minimum value was 0.05 mgL^{-1} recorded at 10 locations. Whereas, the maximum value was 0.4 in two locations with the exception of well no. 1, water well plant, Mir, which was 2mgL^{-1}, mgL^{-1}.. However, it was not detected in the expulsion of the Shotp tank and tap water in Alnamysa village. Most samples were at the permissible limits (< 0.3 mgL^{-1}).

Manganese: The minimum value was 0.05mgL^{-1} recorded at well no. 5, water well plant, Mir locations. Whereas, the maximum value was 0.6 in well no.4, water well plant, Awlaad-Elias. It was not detected in 11 locations and most samples were at the permissible limits (< 0.1 mgL^{-1}).

An appropriate assessment of the suitability of groundwater requires the concentrations of some important parameters like pH, electrical conductivity (EC), TDS, Ca2+, Mg2+, K+, Na+, Cl−, HCO3 -, SO4 2-, F−, NO3 -, PO4 3-, and a comparison with the guideline values set for potable water [39]. Hardness has no adverse effect on human health; however, water above a hardness of 200mgL^{-1} may cause scale deposition in the water distribution system and more soap consumption. Soft water below the hardness less than 100mgL^{-1} is more corrosive for water pipes [37].

[11]Suggested that the high nitrates were indicative of high pollution load. The increase of nitrate levels was caused by intrusion of sewage and industrial effluents into the natural water [13]. High levels of nitrates in water may cause serious illness and sometimes death. Nitrates have the potential to cause shortness of breath, “blue babies” syndrome in infant diuresis, an increase in starchy deposits and hemorrhaging at the spleen [34]. Iron is biologically an important element. It is essential to all organisms and present in the hemoglobin system. A stringent taste is detectable by some persons at levels above 1 mgL^{-1}[21]. In the present study, the iron contents were slightly higher than the permissible limits. The high concentrations may have been due to the dumping of wastes around the bore wells. TDS represents the amount of inorganic substances (salts and minerals). High TDS is commonly objectionable or offensive to the taste. A higher concentration of TDS usually serves as no health threat to humans until the values exceed 10mgL^{-1}[2].

Table 5: Physicochemical parameters of the water samples:

Code	Parameters										
	P.H	Turb (NTU)	Sulfate (mg/l)	Chloride (mg/l)	T-D-S (mg/l)	T-H (mg/l)	Ca-H (mg/l)	Mg-H (mg/l)	Ammonia (mg/l)	T-Iron (mg/l)	T-Manganese (mg/l)
1	7.93	1.34	6.84	75	703	288.2	170	118.2	0.24	0.35	0.2
2	7.5	3.93	9.41	85	821	400	218.8	181.2	0.24	0.4	0.3
3	7.81	2.8	8.91	73	801	367.6	204.4	163.2	N.D.	0.3	0.25
4	8	1.27	9.37	71	798	363.8	199.6	164.2	N.D.	0.15	0.2
5	7.97	2.01	8.55	75	852	261	151	110	0.12	0.1	0.1
6	7.9	1.48	11.92	77	836	310	187	123	0.24	0.35	0.15
7	8.23	0.36	9	78	832	138.6	78	60.6	N.D.	0.05	N.D.
8	8.11	0.3	10.27	79	875	296.8	163.6	133.2	N.D.	0.05	N.D.
9	7.75	1.68	5.89	103	961	333	213.4	119.6	0.4	0.4	0.2
10	7.69	3.54	6.69	128	1163	393.6	246.6	147	0.35	0.05	0.4
11	7.96	0.58	7.98	115	1080	370.2	227.8	142.4	0.05	0.05	N.D.
12	7.98	0.41	8.25	112	1072	370	251.2	118.8	N.D.	0.05	N.D.
13	7.83	1.56	5.43	50	689	244.4	125.6	118.8	0.35	0.05	0.15
14	7.99	1.02	4.1	40	630	226.6	133.4	93.2	0.55	0.1	0.2
15	8.1	1.1	4.78	50	633	248.6	138.2	110.4	0.1	N.D.	N.D.

16	**8.07**	**0.77**	**4.54**	**45**	**646**	**221**	**112.5**	**108.5**	**N.D.**	**0.05**	**N.D.**
17	**7.7**	**0.2**	**20.12**	**140**	**892**	**375.6**	**208.8**	**166.8**	**0.24**	**0.1**	**0.2**
18	**7.72**	**0.62**	**23.26**	**94**	**742**	**369.2**	**206.4**	**162.8**	**0.44**	**0.15**	**0.2**
19	**8.03**	**0.15**	**24.33**	**95**	**696**	**323.2**	**184.2**	**134**	**0.37**	**0.1**	**N.D.**
20	**8.08**	**0.07**	**22.66**	**94**	**678**	**333.2**	**165.4**	**167.8**	**N.D.**	**N.D.**	**N.D.**
21	**8.2**	**0.38**	**22.6**	**128**	**813**	**357.4**	**179.2**	**178.2**	**0.19**	**N.D.**	**N.D.**
22	**8.67**	**3.19**	**3.29**	**20**	**211**	**280**	**80**	**200**	**0.48**	**0.3**	**0.2**
23	**7.97**	**5.39**	**3.36**	**18**	**235**	**162.8**	**82.8**	**80**	**0.56**	**0.35**	**0.25**
24	**7.72**	**3.19**	**3.41**	**15**	**251.5**	**186.4**	**91.6**	**94.8**	**0.24**	**0.3**	**0.25**
25	**7.68**	**2.7**	**3.42**	**22**	**263.5**	**148.8**	**96.2**	**52.6**	**0.12**	**0.05**	**0.05**
26	**7.61**	**2.64**	**3.35**	**17**	**242**	**163.6**	**89**	**74.6**	**0.07**	**N.D.**	**0.15**
27	**7.92**	**2.78**	**3.6**	**134**	**689**	**360**	**186**	**174**	**1.95**	**0.5**	**0.1**
28	**8.02**	**0.51**	**9.6**	**130**	**705**	**352**	**179**	**173**	**N.D.**	**0.05**	**N.D.**
29	**7.68**	**0.97**	**10.28**	**96**	**749**	**466.4**	**308**	**158**	**0.5**	**2**	**0.2**
30	**7.7**	**2.39**	**3.6**	**30**	**408**	**228.4**	**148**	**80**	**0.45**	**0.5**	**0.05**
31	**7.66**	**9.46**	**6.8**	**61**	**642**	**419.4**	**243**	**176**	**0.3**	**2**	**0.3**
32	**8.18**	**19.1**	**6.8**	**42**	**442**	**249.6**	**166**	**83**	**0.4**	**0.2**	**0.5**
33	**8.26**	**0.47**	**3.8**	**30**	**433**	**284.8**	**144**	**140**	**0.15**	**0.1**	**N.D.**
34	**7.8**	**0.73**	**6.19**	**94**	**684**	**364**	**231**	**133**	**0.63**	**0.4**	**0.5**
35	**7.72**	**0.51**	**6.76**	**105**	**753**	**382**	**212**	**170**	**1.46**	**0.15**	**0.6**
36	**7.78**	**0.49**	**6.19**	**90**	**700**	**326**	**215**	**111**	**0.36**	**0.2**	**0.5**
37	**8.13**	**0.2**	**6.32**	**95**	**663**	**331**	**221**	**110**	**0.89**	**0.1**	**0.5**

N.D. = not detectable

4. Conclusion

Microbial analysis of the water samples from the groundwater, water well plants and tap water showed that:Thermotolerant fecal coliform bacteria were found in the water samples of 15 locations (9 wells, 2 expulsions of water well plants, and 4 water networks); that indicated fecal contamination of these locations. There were 22 locations of water samples corresponding to the standard international specifications and Egyptian standards in the summer.So, we found 2 water well plants that needed a sterilization system and filtration system and 2 water networks that had to be regenerated. But in the autumn, thermotolerant fecal coliform bacteria were found in the water samples of 27 locations (14 wells, 1 tank, 6 expulsions of water well plants, and 6 water networks)**.** There were 10 locations contaminated with thermotolarant fecal streptococci (5 wells, 1 tank, 2 expulsions, and 2 water networks). The microbial contamination in the autumn was more than in the summer which led to the sudden fecal contamination with external factors such as animals or the absence of a chlorine system at that time and not for the physical environmental factors such as temperature. The physicochemical parameters of most of the samples were at the permissible limits.

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