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BACTERIOLOGICAL PARAMETERS FOR EVALUATING THE UNDERGROUND WATER QUALITY

Underground water samples were examined for total bacterial counts, total and fecal coliforms, fecal streptococci, total anaerobic bacteria, anaerobic sporeformers and total clostridia. The samples represented well water, which had been used directly or after mixing with chlorinated treated water from the River Nile. The Nile River is a source of drinking water in 3 districts of Cairo.

Total bacterial counts ranged between 10^1 and 10^4 CFU/cm^3. Different levels of bacteria indicators were detected in the samples of well water. Of the samples tested, the ones which gave positive results for the presence of total coliform, fecal coliform, fecal streptococci and total clostridia amounted to 92, 55, 90 and 45%, respectively. On the other hand, all of the samples gave positive results for the presence of anaerobic bacteria and anaerobic sporeformers.

1. INTRODUCTION

Groundwater withdrawal from wells at Greater Cairo waterworks represents 11–17% of the total production of the municipal water. The management of groundwater quality is as important as its quality.

The sanitary quality of water is currently judged on the basis of bacteria indicators. Total bacterial counts have been used to monitor water pollution [11], [19], [29], [30]. Previous studies showed great variations in bacterial densities in different well waters examined, which ranged from 0–10^4/cm^3 in Israel [17] to 10^4–10^7/cm^3 in Minnesota, USA [2].

It is the total coliform group that is considered the primary indicator of bacteriological quality of potable water [15]. Bacteria of this group are taken as an indicator of pollution since they are found in large numbers in fecal wastes. In general, potable water is said to be of good bacteriological quality if it contains less than one coliform per 100 cm^3 of water sample [9]. In a study given by TODD [28], in which water samples were taken from 95 wells representing three types (dug, driven

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and drilled), the following positive results for total coliforms were obtained: 100% for dug wells, 43% for driven wells and 24% for drilled wells.

It has been suggested by many investigators to use fecal coliform subgroup of the coliform bacteria for providing evidence of fecal pollution [4], [5], [8]. A correlation between fecal coliform density and the presence of *Salmonella* was reported by many investigations [2], [3], [32]. Significant levels of *E. coli* were observed in groundwaters of urban areas in the USA [25].

The use of supplementary bacterial indicators of pollution such as streptococci has been suggested by several investigators [7], [21], [26]. Finding the ratio between fecal coliform and fecal streptococci could be helpful in defining the possible sources of fecal pollution [6]. Atypical strains of enterococci were isolated from well water samples of good sanitary quality [24].

The particular significance of *Clostridium perfringens* in water tested lies in the fact that it is a spore-forming bacterium of fecal origin which survives in water for a much longer time than the other bacteria indicators. It is particularly useful in evaluating the quality of water coming from well or bore hole supplies, since the water drawn from ground may have travelled far or spent much time travelling from the source of fecal contamination [12]. The results obtained from five treatment plants indicate greater reliability of *Clostridium* spore tests, compared to the traditional procedure of estimating fecal coliform density [22].

This investigation was undertaken to provide detailed information about sanitary conditions of wells used as a source of drinking water and to show suitability of different bacteriological parameters for evaluation of water quality.

2. MATERIALS AND METHODS

Water samples were collected from 15 wells at three water treatment plants which supply drinking water to three districts in Greater Cairo. At Mostrod waterworks, the pumped drinking water was a mixture of pumped well water and treated surface water (prechlorination; coagulation and precipitation; filtration and postchlorination) taken from Ismailia Canal. Six wells were examined. They differed in depth which ranged from 75 to 95 m and in the flow rate which ranged from 100 to 200 m^3/h. There were four other wells under investigation at El-Maadi, 55–65 m deep, with the flow rate of 100–200 m^3/h. The pumped water was a mixture of untreated groundwater and treated surface water from the River Nile. The third group of wells (five altogether) was located at El-Marg and ranged between 45 and 65 m in depth, with the flow rate of 100–200 m^3/h. Untreated groundwater was the source of drinking water in that region.

In the samples collected during 1985–1986 the following bacteriological parameters were examined:

a) total viable bacterial counts at 22°C and 37°C, using standard agar plate count and applying poured plate method,

b) MPN (most probable number) of total coliforms, using MacConkey broth at 37°C,

c) MPN of fecal coliform, using EC medium at 44.5°C,

d) MPN of fecal streptococci, using azide dextrose broth and ethyl violet azide broth as control medium,

e) MPN of total anaerobic bacteria, using fluid thioglycolate medium,

f) MPN of anaerobic sporeformers, using fluid thioglycolate medium and heating the sample to 80°C for 10 min,

g) MPN of total *Clostridia*, using sulfadiazine polymyxine-B-sulfite agar (SPS).

3. RESULTS AND DISCUSSION

It has been observed how the total viable bacterial densities varied in particular water samples taken from the wells located at the same and/or at different places. The calculated averages ranged from 6.3×10 to 3.3×10^3 (at Mostrod), from1.7×10 to 1.3×10^2 (at El-Maadi), and from 6.2×10 to $1.6 \times 10^2/cm^3$ (at El-Marg). Generally, the count was less numerous at 37°C than that obtained at 22°C. According to GRABOW [13], natural water bacteria grow freely at temperatures between 20 and 30°C, whereas parasitic bacteria and bacteria of soil and sewage origin tend to grow best at 37°C. Bacterial growth in water supplies has been well documented and counts as large as $2.5 \times 10^5/cm^3$ have been reported [10]. In the recent years, an interest has grown in the use of heterotrophic plate count for enumeration of bacteria in potable water. A standard value, for the total bacterial count in drinking water has been established at 100 bacteria/cm^3 in Germany [20], and it is suggested that this standard should not exceed the value of 500 bacteria/cm^3 in distributed water [10]. According to the American standard, all the wells examined except Nos. 7, 40, 43, 46 at Mostrod and No. 2 at El-Maadi are acceptable as sources of drinking water (table 1). Sizeable number of bacteria obtained as a result of standard plate count procedure may include genera which are hazardous to pubilic health [16] or can become serious secondary pathogenic invaders in post-operative infections [33]. Some bacteria create taste, odour or spoilage problems in food and drug industries [1]. There are some limitations in using the total viable bacterial counts as an indicator of water pollution: 1) the agar plate count selects the bacteria that may comprise only a small percentage of the bacterial population actually present, 2) the limited sample volume used in poured plate method restricted its application to some extent [10].

The results showed that 14.4 and 11.1% of the well water samples from Mostrod and El-Maadi, respectively, were negative for coliform test. On the other hand, all the wells at El-Marg were positive for the same test (table 2). The range of coliform densities varied from about 1 to 11.7 MPN/100 cm^3. The lowest average values were obtained for samples of Mostrod wells (table 1). MCFETERS et al. [18] suggested that

Table 1

Average values for the bacteriological indices in well water samples from different locations

Locations	Well No.	Bacteriological indices*							
		Total bacterial counts		Coliforms		Fecal streptococci	Anaerobic bacteria		Clostridia
		at 22°C	at 37°C	Total	Fecal		Total	Spores	
Mostrod	3	63	65	0.8	0.2	3.8	556	22**	0**
	7	2138	1313	3.3	1.2	3.3	703	49***	7***
	24	72	57	1.8	0.3	2.0	332	41***	0***
	40	2390	1490	1.2	1.0	2.7	285	39**	5**
	43	3310	2390	1.7	1.2	4.0	542	54**	7**
	46	1420	1600	1.7	0.7	5.2	463	48**	4**
El-Maadi	1	443	245	1.1	0.6	3.4	5754	58	3
	2	1298	1009	7.8	0.3	4.3	6269	53	1
	3	223	172	11.7	3.9	6.4	4718	47	5
	4	549	429	3.9	0.8	6.3	5298	63	2
El-Marg	1	62	122	2.8	0.7	5.5	3143	34	3.3
	2	120	77	2.2	0.8	2.8	2158	39	4.5
	10	164	92	6.7	1.5	1.3	308	13	2.0
	13	115	101	4.3	1.8	2.5	1655	47	6.6
	20	72	65	2.2	0.7	2.8	497	59	5.3

* All bacteriological indices were determined as MPN/100 cm^3 except total bacterial counts which were determined as CFU/cm^3.

** Only 4 samples were examined.

*** Only 5 samples were examined.

the lack of demonstrable coliform levels in groundwater, which is partly caused by microbial interferences and limitations of test media, does not preclude the occurrence of pathogens. The American bacteriological standards limit the coliform densities to less than one MPN per 100 cm^3 [31]. Accordingly, only well No. 3 at Mostrod can meet this standard. The Egyptian standards recommended a level of not more than 5 MPN/100 cm^3 as acceptable level of total coliforms in groundwater. According to our local standards, all wells are acceptable as a source of drinking water except wells Nos. 2 and 3 at El-Maadi and No. 10 at El-Marg. The coliform group includes organisms, that differ in their natural habitats, which limits their suitability as the indicators of pollution. In addition, excessive densities of non-coliform organisms in water may desensitize assay procedure for total coliforms [14].

The percentages of positive samples for fecal coliforms varied from well to well and ranged between 17 and 83%. In this respect, the three locations under investiga-

Table 2

Number and percentage of well water samples the testing of which gave positive results for different bacteriological indices

Location	Well No.	Bacteriological indices											
		Total coliforms		Fecal coliforms		Fecal streptococci		Total anaerobes		Anaerobic spores		Clostridia	
		+	%	+	%	+	%	+	%	+	%	+	%
Mostrod	3	3	50	1	17	5	83	6	100	4	100	0	0
	7	5	83	4	67	6	100	6	100	5	100	4	80
	24	5	83	2	33	6	100	6	100	5	100	0	0
	40	5	83	5	83	6	100	6	100	4	100	2	50
	43	5	83	3	50	6	100	6	100	4	100	4	100
	46	6	100	4	67	6	100	6	100	4	100	2	50
El-Maadi	1	9	64	7	50	14	100	14	100	14	100	4	28
	2	8	100	2	25	8	100	8	100	8	100	2	25
	3	14	100	11	78	14	100	14	100	14	100	10	7
	4	9	100	5	55	9	100	9	100	9	100	4	44
El-Marg	1	6	100	3	50	5	83	6	100	6	100	1	16
	2	6	100	5	83	6	100	6	100	6	100	6	100
	10	6	100	3	50	4	66	6	100	6	100	1	16
	13	6	100	3	50	6	100	6	100	6	100	5	83
	20	6	100	3	50	6	100	6	100	6	100	6	100

tion can be arranged in the following ascending order: Mostrod (52.7% positive samples for fecal coliforms), El-Maadi (55.5%), and El-Marg (56.6%). The average for MPN index ranged between 0.2 and 3.9 of coliforms/100 cm^3. Only well No. 3 at Mostrod gave more than 90% samples free from fecal coliforms. All standards rejected the presence of the fecal coliforms in drinking water.

The use of more persistent bacterial indicator is necessary, specially under adverse environmental conditions such as in the case of underground water. 96.3% of the well water samples showed various levels of contamination with fecal streptococci. Negative results gave the samples collected at Mostrod (well No. 3) and El-Marg (wells No. 1 and 10). Percentage of fecal streptococci positive samples exceeded the percentage of fecal coliform positive samples (54.9). This finding was confirmed by the results of ROSENBERG et al. [23]. Fourty-seven well water samples were negative for fecal coliforms, being positive for fecal streptococci (table 3). These samples were distributed as follows: 16 from Mostrod, 20 from El-Maadi and 11 from El-Marg. Only one sample from Mostrod (well No. 3) and two samples from El-Marg (wells Nos. 1 and 10) were negative for both fecal coliforms and fecal streptococci. Generally, densities of fecal streptococci were higher than those of fecal coliforms, however they never exceeded 10 MPN/100 cm^3 (table 1). According to the results of fecal coliforms to fecal streptococci ratio, it is possible to conclude that animal wastes are the main source of pollution as mentioned by GELDREICH et al. [6].

Total anaerobic bacteria counts in the wells located at Mostrod, El-Maadi and El-Marg were averaged as follows: 10^2–10^3, 10^3–10^4 and 10^2–10^4 MPN/100 cm^3, respectively (table 1). At high level of anaerobic bacteria, the sporulated cells do not exceed an average of 59 MPN/100 cm^3 (table 1). It is worth mentioning that all the samples were positive for both anaerobic and spore-forming bacteria (table 2). *Clostridium* as a natural inhabitant of intestinal tract was detected in all wells under investigation except Nos. 3 and 24 at Mostrod (table 2). Its density did not exceed the average value of 10 MPN/100 cm^3 (table 1). Over 30% well water samples taken at Mostrod were positive for clostridia, while at El-Maadi, these amounted to more than 40%, and to 60% at El-Marg. Comparing the results of clostridia testing with those of other bacteria testing, it was observed that out of the total samples examined, fecal coliform, fecal streptococci and clostridia were present in 25% at Mostrod, 44.4% at El-Maadi and 43.3% at El-Marg. Only 8.3, 17.7 and 20% of the samples taken at Mostrod, El-Maadi and El-Marg were positive for both fecal streptococci and clostridia. One sample from Mostrod and one from El-Marg wells were negative for the three indicators. The isolation of other indicators in the absence of coliform was considered a feature which would be important in detecting unusual water pollution. So, the presence of *Clostridium* in the absence of coliform organisms indicates on of the two possibilities: 1) that contamination occurred too long ago, or 2) that water had been subjected to conditions lethal to coliform bacteria [13].

Table 3

Presence or absence of different bacteriological indices in well water samples collected from three locations

No. of samples examined	T.c.+	F.c.+	F.s.+	Cl.+	F.c.+ F.s.+	F.c.+ F.s.−	F.c.− F.s.−	F.c.− F.s.+	F.c.− Cl.+	F.c.+ Cl.+	F.s.+ Cl.+	F.s.− Cl.+	F.s.− Cl.−	F.c.+ F.s.+ Cl.+	F.c.+ F.s.− Cl.+	F.c.− F.s.− Cl.+	F.c.− F.s.+ Cl.+
							Mostrod wells										
36	29	19	35	12	19	0	1	16	3	9	12	0	1	9	0	0	3
							El-Maadi										
45	40	25	45	20	25	0	0	20	8	12	20	0	0	20	0	0	8
							El-Marg										
30	30	17	27	19	16	1	2	11	8	11	18	1	2	13	0	1	7

T.c.: Total coliforms, F.c.: Fecal coliforms, F.s.: Fecal Streptococci, Cl.: Clostridium

The assumption that groundwater from deep aquifers is generally of good bacteriological quality because percolation of water through soil results in the removal of microbial pollution should not be taken for granted. Unless otherwise demonstrated, it is to be assumed that groundwater also contains microbiological contaminants and needs to be treated. Specifically, all treatment plants for household supplies should provide disinfection or at least have disinfection capability. Continuous bacteriological monitoring of the suitable bacterial indicator is required to demonstrate continuous integrity of the system.

4. CONCLUSIONS

The results obtained during the testing of the samples are as follows:

1. 54% of the samples gave positive results for the presence of both fecal coliforms and fecal streptococci.
2. 0.9% of the samples gave positive results for fecal coliforms and negative results for fecal streptococci.
3. 42.3% of the samples were negative for fecal coliforms, being however, positive for fecal streptococci.
4. Both fecal coliforms and fecal streptococci were absent in 27% of the samples.
5. 15.3% of the samples were negative for fecal coliforms and positive for clostridia. On the other hand, 28.8% of the samples gave positive results for both fecal coliforms and clostridia.
6. 0.9% of the well water samples gave negative results for fecal streptococci and positive results for clostridia, but 45% of the samples were positive for both fecal streptococci and clostridia.

The isolation of other indicators in the absence of coliforms (traditional indicator) was considered a feature which would be important in detecting the underground water pollution, when coliform bacteria were absent or not differentiated during water analysis. Much more accurate results could be obtained by the examination of more than one indicator of pollution, especially fecal streptococci and clostridia.

The assumption that groundwater from deep aquifers is generally of good bacteriological quality should not be taken for granted. Disinfection of underground water is the only way of overcoming possible public health hazards.

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WSKAŹNIKI BAKTERIOLOGICZNE W OCENIE JAKOŚCI WÓD PODZIEMNYCH

W próbkach wód podziemnych pochodzących ze studni określono wskaźniki bakteriologiczne w odniesieniu do wszystkich bakterii, bacterii z grupy *coli*, fekalnych bakterii z grupy *coli*, bakterii z grupy *Streptococcus faecalis*, wszystkich bakterii anaerobowych, anaerobowych form przetrwalnikowych i wszystkich bakterii z grupy *Clostridium*. Badana woda była wykorzystywana jako źródło wody pitnej albo bezpośrednio, albo po zmieszaniu z uzdatnioną i chlorowaną wodą z Nilu.

Wskaźnik bakteriologiczny w badanych próbach wahał się od 10 do $10^4/cm^3$. Stwierdzono znaczne wahania w wartościach pozostałych wskaźników bakteriologicznych w poszczególnych próbkach wody studziennej. Obecność bakterii z grupy *coli*, fekalnych bakterii z grupy *coli*, bakterii z grupy *Streptococcus faecalis* i bakterii z grupy *Clostridium* stwierdzono odpowiednio w 92, 55, 90 i 45% badanych prób. Z drugiej strony, we wszystkich badanych próbach stwierdzono obecność anaerobowych bakterii i anaerobowych form przetrwalnikowych.

БАКТЕРИОЛОГИЧЕСКИЕ ПОКАЗАТЕЛИ В ОЦЕНКЕ КАЧЕСТВА ПОДЗЕМНЫХ ВОД

В пробах подземных вод, происходящих из колодцев, определены бактериологические показатели по отношению ко всем бактериям, бактериям из группы *coli*, фекальным бактериям из группы *coli*, бактериям из группы *Streptococcus faecalis*, всем анаэробным бактериям, анаэробным склероциевым формам и всем бактериям из группы *Clostridium*. Исследуемую воду использовали или в качестве питьевой воды, или непосредственно после смешения с подготовленной и хлорированной водой из Нила.

Бактериологический показатель в исследуемых пробах колебался от 10 до $10^4/см^3$. Были установлены значительные колебания значений остальных бактериологических показателей в отдельных пробах воды из колодцев. Наличие бактерий из группы *coli* фекальных бактерий из группы *coli*, бактерий из группы *Streptococcus faecalis* и бактерий из группы *Clostridium* было установлено соответственно в 92, 55, 90 и 45% исследуемых проб. С другой стороны, во всех исследуемых пробах было установлено наличие анаэробных бактерий и анаэробных склероциевых форм.