

Population analyses of microfauna at a Water treatment Plant at Shebeen Alkoom, Minufeya, Egypt.

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ABSTRACT

Safe Drinking water is a priority for human health all over the world. Accordingly, the microfauna of raw, filtered, treated, and tap water were investigated in this study. The results proved the efficiency of the Water Plant, but slight occurrence of non-pathogenic bacteria in tap water, indicated the necessity to renovate the distribution system.

Most of the physico-chemical characters are within the standards for drinking water apart from phosphate, nitrate, iron and manganese. Raw water samples only were positive for *E. coli*, T. & F. coliform, *Salmonella* and *Shigella*. This could be referred to the illegal discharge of sewage and other organic pollutants into the water source. Sixty one protozoan genera were identified only in raw water which can be attributed to the influence of chlorination during water treatment. Ciliated protozoan densities proved seasonal variation with highest values during Autumn. Protozoan monthly densities showed significant correlations with certain physical and chemical parameters. Therefore ciliates are considered as bio-indicators for pollution in various aquatic habitats. Simultaneously, rotifers can be used to indicate the performance level of certain stages of water treatment process as they showed significant correlations with different ecological factors.

Keywords: Fecal bacteria, Protozoa, Water-treatment.

INTRODUCTION

Micro-organisms play an important role in nutrient recycling in nature with particular reference in aquatic ecosystems. Bacteria oxidize the highly complicated organic materials into simpler inorganic salts which are useful to biotic and abiotic components of the aquatic ecosystems. Protozoan organisms help directly and indirectly in both nutrient recycling and removal of different types of pollutants. This was carried out through the predation of bacteria and consequently keeping bacteria in permanent biological activity. On the other hand, different protozoa feed upon pathogenic and harmful bacteria like fecal type.

MATERIAL AND METHODS

Study Area

The study included the different stages of water treatment at Shebeen Alkoom Water Treatment Plant. It is located south of the city and feeds the western side of the city.

Sample Determinations and Collection Procedures

Water samples for microbiological analysis were collected aseptically in 500ml glass bottles containing 1ml of 3% freshly prepared sodium thiosulphate solution ($\text{Na}_2\text{S}_2\text{O}_3$) to neutralize the residual chlorine. The samples were preserved in ice box during transportation (APHA, 1998), brought to the laboratory three times monthly

during a period from November 2008 to November 2009 and examined within 6 hours of collection. For physico-chemical analysis the samples were collected in 1 liter glass bottles.

Physio-chemical Characteristics

Turbidity

Turbidity is an expression of the amount of light scattered and absorbed by the particles in a sample and is measured as "Nephelometric turbidity units" (NTU) by using turbidity meter (PCH019054, Germany).

Chemical Characteristics

The chemical analyses were carried out in the laboratory at faculty of science-Minofia University. The study of the chemical properties of the water included.

Hydrogen Ion Concentration (pH)

The pH values of the collected field samples are determined by the use of bench-top pH/Ion Meter (Model 6500, China).

Determination of Hardness (Calcium and Magnesium)

The hardness of water is a measure of the concentration of calcium and magnesium salts dissolved in water. Calcium and magnesium in water samples were determined using EDTA (Ethylene diamine tetra acetate) titrimetric method according to APHA (1998).

Dissolved Oxygen (DO)

The oxygen content of the water samples was measured by SB70D DO Bench top meter S/NDO 0800, U.S.A. and expressed as mg/L.

Ammonia (NH₃), nitrite (NO₂), nitrate (NO₃), phosphate (Po₄), silica (Si), iron (Fe), manganese (Mn) and fluoride (Fl) contents were determined using assay colorimetric kits (HANNA instrument, C200 Multiparameter Ion Specific Meter, Hungary) according to the manufacture's protocols. The results were expressed as mg/l.

Biological Examination

Zooplankton

Samples for analysis of zooplankton were collected from the studied four stages (raw water- filtered water – treated water – tap water) into glass bottles of 500 ml capacity. In the laboratory, water sample in each bottle was mixed well by inverting the bottle at least ten times smoothly, then for each stage, three replicates each of 10 ml were taken, centrifuged by using a cooling centrifuge (Hermle, Z252 MK, Germany) for three minutes at 7 °C and at 1500 rpm. The volume of each replicate was concentrated to three ml by decanting the supernatant and the residual was transferred to a cavity slide drop by drop and then the different zooplankton organisms were counted by using a Carl Zeiss transmitted light inverted Microscope, Germany. The identification was performed according to Patterson and Hedley (1996). The results were expressed as organism x10³/l.

Bacteria

Enumeration of Indicator Organisms

Water samples were analyzed for total coliform (TC), fecal coliform (FC) and *E. coli* using Most Propable Number (MPN/100 ml according to Boothman *et al.*, 2002 and Michiels and Moyson; 2000). This method was given in Standard Methods for Examination of Water and Wastewater (AHPA, 1998).

For TC counts a series of five fermentation tubes containing 10 ml double strength lauryl tryptose broth (LTB) (Merck) were inoculated with 10 ml volumes of water samples (treated water, tap water). In the examination of the non-potable water (raw water) 1.0, 0.1, 0.01 ml portion of the sample was used as inoculum for a series

of five fermentation tubes containing 5 ml single strength lauryl tryptose broth. The tubes were incubated at 37 °C for 48 h. All gas positive (yellow) LTB tubes were subcultured to tubes of brilliant green lactose bile broth (BGLB, Merck), and incubated at 37 °C for 48 h. Gas-positive BGLB tubes were considered positive for the presence of TCs. Gas-positive LTB tubes were subjected to further analysis with *E. coli* broth (Merck). The Ec tubes were incubated at 44.5 °C for 24 h. *E. coli* positive tubes were confirmed for the presence of *E. coli* by performing Gram stain. All cultures appeared as Gram-negative, short rods re-inoculated back into LTB to confirm gas production.

Water samples were analyzed for bacterial indicators, *Salmonella* and *Shigella* by using Deoxycholate citrate agar. It is a modification of deoxycholate agar formulated by Leifson (1935) and modified by Hynes (1942). The results are expressed as CFU/ml (APHA, 1998).

Principles of the Procedure

Meat extract and meat peptone provides the nitrogen and vitamin sources in Deoxycholate Citrate Agar. Lactose is the fermentable carbohydrate. Sodium Deoxycholate and Sodium Citrate inhibit growth of Gram-positive bacteria, coliforms and *Proteus* spp. Ferric Citrate aids in the detection of H₂S producing bacteria. Neutral Red is a pH indicator. Agar is the solidifying agent.

In the presence of Neutral Red, bacteria that ferment lactose produce acid and form red colonies. Bacteria that do not ferment lactose form colorless colonies. If bacteria produce H₂S, colonies will have black centers. The majority of normal intestinal bacteria ferment lactose and do not produce H₂S (red colonies without black centers). *Salmonella* spp. and *Shigella* spp. do not ferment lactose, but *Salmonella* may produce H₂S (colorless colonies with or without black centers). Lactose-fermenting colonies may have a zone of precipitation around them caused by the precipitation of deoxycholate in the presence of acid.

Sodium desoxycholate and sodium citrate present in the medium inhibit the growth of the Gram positive bacteria. Lactose fermenting bacteria cultivate with pink colonies surrounded by a zone of precipitated bile salts. Lactose non fermenting bacteria cultivate with colorless colonies, with or without a black center caused by the production of hydrogen sulphide, as illustrated in Table 1.

Table 1: Culture Response on Desoxycholate Citrate Agar at 35 °C after 48 Hours Incubation*.

Organisms (ATCC)	Growth	Color of colony
<i>Salmonella typhi</i> (6539)	+++	colorless to tan
<i>Salmonella typhimurium</i> (14028)	+++	colorless to tan
<i>Salmonella enteritidis</i> (13076)	+++	black centered
<i>Shigella sonnei</i> (25931)	++	colorless to pink
<i>Shigella flexneri</i> (12022)	++	colorless to pink
<i>Escherichia coli</i> (25922)	±	Pink with zone of precipitation
<i>Enterococcus faecalis</i> (29212)	-	-

*After, Leifson (1935), Hynes (1942) and MacFaddin (1985)

Statistical Analysis

Statistical Analysis was carried out at 1% and 5% level of significance. The correlation and regression coefficients were carried out using Microsoft Office Excel and Statistical Package for the Social Sciences (SPSS) version 15.

RESULTS

Zooplankton Distribution

Zooplankton groups occurred only in raw water samples and were completely absent in filtered, treated and tap water samples. This might be attributed to the effect of pre-chlorination during water treatment process in the plant.

Abundant Zooplankton ($\times 10^3$)

In the present investigation as illustrated in Table 2, seasonal variation of the most abundant zooplankton groups attained a high number of 1722 in Autumn, and the second peak of 1611 in Spring, while the minimum value of 1048 was observed in Winter.

Table 2: Seasonal Variation of the Most Abundant Zooplankton Group ($\times 10^3/L$)

Season	Sum of Sarcodines	Sum of Flagellates	Sum of Ciliates	Total Protozoa	Rotifers	Protozoa & Rotifera as main Zooplankton Groups
Winter	255	159	570	984	64	1048
Spring	285	234	905	1424	187	1611
Summer	377	77	877	1331	196	1527
Autumn	289	174	1143	1606	116	1722
Total	1206 (23%)	644 (12%)	3495 (65%)	5345 (90.5%)	563 (9.5%)	5908

Zooplankton populations are differentiated into many groups. The most abundant are protozoa and rotifera. Their percentage frequencies are 90.5 % and 9.5%, respectively.

Total Protozoa

Protozoa persisted all over the year in the collected water samples. A total of 61 genera were identified, 42 genera of ciliates, 12 genera of phytoflagellates and 7 genera of sarcodines, in raw water samples of the studied area, as shown in Table 3.

The monthly variations of total density of protozoa in raw water are given in Table 4. It was found that population density of protozoa in raw water attained a climax of 730 organism $\times 10^3/l$ in September, the second peak 719 organism $\times 10^3/l$ was observed in April, while the minimal value (266 organism $\times 10^3/l$) was detected in February.

Table 3: Identified Genera of Protozoa.

	Sarcodina	<i>Amoeba</i> <i>Arcella</i>	<i>Centropyxis</i> <i>Diplophrys</i>	<i>Actinosphaerim</i> <i>Hedriocystis</i>	<i>Nuclearia</i>
	Phyto-Mastigophora	<i>Dinoflagellate</i> <i>Peranema</i> <i>Euglena</i>	<i>Anthophysa</i> <i>Ceratuim</i> <i>Eudorina</i>	<i>Entosiphon</i> <i>Anisonema</i> <i>Cryptomonas</i>	<i>Astasia</i> <i>Urceolus</i> <i>Cartenia</i>
Protozoa	Ciliophora	<i>Dinoflagellate</i> <i>Peranema</i> <i>Euglena</i> <i>Dinoflagellate</i> <i>Peranema</i> <i>Euglena</i> <i>Dinoflagellate</i> <i>Peranema</i> <i>Euglena</i> <i>Dinoflagellate</i>	<i>Anthophysa</i> <i>Ceratuim</i> <i>Eudorina</i> <i>Anthophysa</i> <i>Ceratuim</i> <i>Eudorina</i> <i>Anthophysa</i> <i>Ceratuim</i> <i>Eudorina</i> <i>Anthophysa</i>	<i>Entosiphon</i> <i>Anisonema</i> <i>Cryptomonas</i> <i>Entosiphon</i> <i>Anisonema</i> <i>Cryptomonas</i> <i>Entosiphon</i> <i>Anisonema</i> <i>Cryptomonas</i> <i>Entosiphon</i>	<i>Astasia</i> <i>Urceolus</i> <i>Cartenia</i> <i>Astasia</i> <i>Urceolus</i> <i>Cartenia</i> <i>Astasia</i> <i>Urceolus</i> <i>Cartenia</i> <i>Astasia</i>

Common, Frequent and Rare Protozoa (number of organisms x 10³).

The common genera of protozoa in raw water samples, were represented by thirteen genera. These genera were *Amaeba* sp., *Arcella* sp., *Actinosphaerium* sp., *Centropyxis* sp., *Peranema* sp., *Anisonema* sp., *Cryptomonas* sp., *Vorticella* sp., *Paramecium* sp., *Cinetochilum* sp., *Aspidisca* sp., *Tachysoma* sp., and *Cyclidium* sp. They contributed different percentages among the total protozoan numerical density (5345 organism x10³/l).

The results also revealed that, *Amaeba* reached a maximum of 89 in July and a minimum of 13 in February. The maximum population density of *Arcella* sp. was 78, recorded in June, while it was not easily detected in May. The population density of *Actinosphaerium* sp. ranged between a maximal value of 77 in Jan and minimal value of 3 in August. The maximum population density of *Centropyxis* was 81 on August, while it couldn't be detected in January, February, March, April, May and July. *Peranema* sp. reached its maximal count of 54 during April and it was more or less absent during August.

Anisonema sp. reached a maximum value of 70 in September and it was missing in December, May and November. *Cryptomonas* sp. reached a climax of 57 in January, while it could not be easily detected in December, May, July, August, September, October and November. *Vorticella* sp. reached its maximal count of 83 in September and minimal count of 3 in December. *Paramecium* reached a climax of 39 in November, while the minimal value of 1 detected in September. *Cinetochilum* sp. attained a maximal population density of 190 in September, while it could not be detected in December, January, February and August.

Aspidisca sp. population density ranged between a maximal value of 127 in September and minimal value of 6 in February. *Tachysoma* sp. and *Cyclidium* sp. reached a maximal value of 151 in April and 184 in June respectively. *Tachysoma* sp. could not be detected in May, June and August, while *Cyclidium* sp. was absent during August.

Genera of frequent Protozoa

These were represented by eight genera: *Dinoflagellate*, *Euglena*, *Chilodonella* sp., *Halteria* sp., *Lembadion* sp., *Litonotus* sp., *Strombilidium* sp. and *mesodinium* sp. They contributed various percentages of the total protozoa (5345 organism x10³/l).

Dinoflagellate sp. reached a maximum value of 13 in September and completely missed in May. *Euglena* peaked with a value of 17 in May, but was absent during March and August.

Halteria sp. reached a climax of 41 in March, while it was not found in December, January, April, June, October and November. *Lembadion* sp. reached a maximum value of 36 in March and missed during April, June, July, August and September. *Litonotus* sp. reached 36 in April and September, while it could not be detected in May, July, August, and October.

Chilodonella sp. reached a climax of 37 in August and difficult to be sampled in December, January, February, March, April, May, July and October. *Strombilidium* sp. reached about 31 in January, while it could not be detected in December, April, May, June and July. *Mesodinium* sp. population density reached 32 in July, while it could not be easily detected during December, February, March, and May.

Rare Genera of Protozoa

These were represented by 40 genera having varying percentages among the total protozoa (5345 organism x10³/l).

Herdriocystis sp., *Diplophrys* sp., *Nuclearia* sp., *Entosiphon* sp., *Anthophysa* sp., *Ceratuim* sp., *Eudorina* sp., *Astasia* sp., *Urceolus* sp., *Cartenia* sp., *Stentor* sp.,

Strombidium sp., *Euplotes* sp., *Cothurnia* sp., *Urocentrum* sp., *Coleps* sp., *Didinium* sp., *Dileptus* sp., *Stylonychia* sp., *Tintinnidium* sp., *Blepharisma* sp., *Parurolptus* sp., *Pleuronema* sp., *Loxodes* sp., *Lacrymaria* sp., *Vaginicola* sp., *Holotrichia* sp., *Podophyra* sp., *Chlamydodon* sp., *Loxophyllum* sp., *Epistylis* sp., *Spirostomum* sp., *Telotrochidium* sp., *Chaenea* sp., *Strichotricha* sp., *Spathidium* sp., *Acineta* sp., *Colnilembus* sp., *Colpidium* sp. and *Trachelophyllum* sp.

Herdriocystis sp. was found to be scattered throughout the year with irregular mode of occurrence, with a maximum count of 5 in July and a minimum count of 1 in April, June and September. *Diplophrys* sp. appeared in June only, where its density was 5. *Nuclearia* sp. appeared also in September only with a count of 1.

Entosiphon sp. was found during January (13). *Ceratuim* sp. existed during January and early Spring (March–April), where its population densities ranged between a minimum of 1 in January and April, and maximum of 4 in March. *Anthophysa* sp. was observed in January, February, April, July, August, September and November with a maximum value of 18 in July. *Eudorina* sp. existed during May, June, August and September, where its population densities ranged between a minimum of 5 in June and a maximum of 12 in August.

Astasia sp. existed during two months (September and October) and ranged between four in September to one in October. *Urcealus* sp. appeared in October only where its density was only one. *Cartenea* sp. appeared also in October only with a count of 1. *Stentor* sp. was found to be scattered throughout of the year with irregular mode of occurrence with a maximum count of 6 in February and a minimum count of one. in August. *Strombidium* sp. existed during October only with a value of 4.

Euplotes sp. was observed in June, August and March with densities of 17, 6 and 4, respectively. *Cothurnia* sp. appeared in five months (February, March, April, September and October) with a maximum value of 15 in September and a minimum value of one in February and March.

Urocentrum sp. appeared in January only, where its density was two. *Coleps* sp. was found during four months (December, June, August and November) with a maximum count of 18, but was only one in August. *Didinium* sp. appeared in June only, where its density was two. *Dileptus* sp. was found during February, March, April, May, June and August with a maximum value of 5 in May and a minimum value of one in February, March, June and August. *Stylonychia* sp. appeared only in March where its density was 1. *Tintinnidium* sp. appeared in January only where its density was 6. *Blepharisma* sp. appeared in June only where its density reached 16. *Parurolptus* sp. appeared in three months of the year (December, January, April) with a maximum count of 9 in April and a minimum count of one in January. *Pleuronema* sp. appeared only in four months (January, February, March and November) with the numbers one, three, seven, and one respectively. *Loxodes* existed during early Spring and Summer (April, May, June and July), where its density ranged between a maximum of 16 in April and a minimum of two in July. *Lacrymaria* sp. reached its maximum value (3) in July and September, while the minimum value (1) was noticed in April and June. *Vaginicola* sp. appeared only during early Spring (March and April) by 6 and 3, respectively. *Holotrichia* sp. was found during March, April, June and November with a maximum count of three in June and a minimum count of one in March and April. *Podophyra* sp. appeared during three months (December, April and August) with a maximum count of five in April and a minimum count of one organism in December. *Chlamydodon* sp. appeared only during April and July, where its density was 1 and 2. *Loxophyllum* sp. existed during August and September, where

its densities were one and two, respectively. *Epistylis* sp. appeared only in September with a density of two.

Spirostomum sp. appeared in four months (March, July, September and October) with maximum count of 2 in October and a minimal count of 1, in the first three months. *Telotrochidium* sp., *Chaenea* sp., *Colnilembus* sp., *Colpidium* sp., *Strichotricha* sp., *Trachelaphyllum* sp. appeared only in one month of a year (November, for the first two genera; January *Colnilembus* sp., June for *Colpidium* sp. and *Strichotricha* sp. and September for *Trachelaphyllum* sp.) with a count of 1. *Spathidium* sp. appeared in December and June (2 and 1). *Acineta* sp. appeared in December and February, with the number of 3 and 2, respectively.

Rotifera

It was found that the population densities of Rotifera in raw water samples attained a climax of 86, during April. The second peak of 76 was observed in July. The minimal counts of 14 and 17 were detected in November and December respectively.

Bacterial Investigations

Total coliform (Tc), fecal coliform (Fc) and *Escherichia coli*. (*E. coli*) were counted using Most Propable Number (MPN) method, while *Salmonella* and *Shigella* were counted using Spread Plate Method, according to APHA (1998).

Raw water samples showed that MPN of Tc varied from 109 to 390/100 ml in September and July respectively, while those of Fc increased from 2 to 18/100 ml water for Fc observed in November and May respectively. The counts of *E. coli* fluctuated between 2 and 18/100 ml water with the highest average value observed in May and the lowest average value observed in November, as shown in Table 5. Count for *Salmonella* and *Shigella* showed that the highest average numbers were found in September and November, being 153 and 152 CFU/ml, respectively for *Salmonella*, while the highest average numbers for *Shigella* were observed in August, June and July (340, 338, and 335 CFU/ml respectively).

Table 5: Monthly occurrence of Bacteria (Total coliform, Tc; fecal coliform, Fc; and *E. coli*) existed in raw water (R), filtered (F), and tap water at Shebeen Alkoom.

	R	R	R	R	R	F	F	Tap
Month	Tc	Fc	<i>E. coli</i>	<i>Salmonella</i>	<i>Shigella</i>	Tc	Fc	Tc
December 2008	260	12	12	89	185	1	1	1
January 2009	195	12	12	3	82	1	1	1
February 2009	195	11	11	51	258	1	1	1
March 2009	285	5	5	68	219	1	1	1
April 2009	265	7	7	42	193	4	1	4
May 2009	236	18	18	66	157	4	2	2
June 2009	307	8	8	68	338	2	1	1
July 2009	390	8	8	0	335	2	1	3
August 2009	123	5	5	50	340	2	1	2
September 2009	109	5	5	153	266	4	1	4
October 2009	180	4	4	81	220	1	1	1
November 2009	205	2	2	152	147	2	2	1

The lowest average numbers for *Salmonella* in raw water samples were zero, and 3 CFU /100ml in July and January respectively. For *Shigella*, the lowest densities

were 82, 147 CFU /ml in January and November respectively. In Filtered water samples, the highest average numbers were observed in April, May and September being 4 MPN/100ml for total coliform. Those of Fecal coliform were observed in May and November, being 2 MNP/100 ml, while *E. coli*, *Salmonella* and *Shigella* were completely absent.

Regarding seasons the highest counts of bacterial indicator Tc in raw water samples were detected in the warmer seasons (Spring and Summer) as shown in Table 6.

Table 6: Seasonal Variations of Total Coliform, Fecal Coliform and *Escherichia coli*, *Salmonella* and *Shigella* recorded in Raw water.

Seasons	Total Coliform MPN/100ml	Fecal Coliform MPN/100ml	E. Coli MPN /100ml	Salmonella CFU/ml	Shigella CFU/ml
Winter	217	11	11	48	175
Spring	262	10	10	59	190
Summer	273	7	7	39	337
Autumn	165	3	3	128	211

In treated water, all samples from outlet water treatment were free from total coliform, fecal coliform, *E. coli*, *Salmonella* and *Shigella* groups. These results indicate the safety of treated water according to the Egyptian Standard, (2007) for drinking water. Tap water samples, showed that number of total coliform ranged between 1 and 4 MPN/100 ml, but were free from fecal coliform, *E. coli*, *Salmonella* and *Shigella* groups.

Overall correlations between the studied species and specific variables, are presented in Table 7. From that Table, it is clear that the increase of manganese, ammonia, dissolved oxygen will be accompanied by an increase of protozoan abundance, while Ca and turbidity play a reducing effect on its prevalence. However, a negative relationship between protozoa and rotifers was obtained. Ciliates as a group of protozoa were found to increase by rising in turbidity and/or manganese.

On the other hand, either water temperature or turbidity increases the abundance of rotifers, while alkalinity or total hardness negatively affects their abundance.

Table 7: Correlations coefficient (r) of total protozoa (P), rotifers, ciliates, and certain physico-chemical parameters.

Parameters	Correlation, r	Type of relation
P & Mn	0.55	+ve
P & Ca	0.60	-ve
P & Turbidity	0.55	-ve
P & Ammonia	0.58	+ve
P & DO	0.57	+ve
P & Rotifera	0.44	-ve
P & Total Coliform	0.43	-ve
Ciliates & Turbidity	0.56	+ve
Ciliates & Mn	0.60	+ve
Rotifera & Alkalinity	0.64	-ve
Rotifera & Total hardness	0.55	-ve

DISCUSSION

Main Zooplankton (Protozoa and Rotifera)

The maximum population density of main zooplankton in this study was observed during Autumn and Spring with a peak in April (805 organism $\times 10^3/l$) and

September (792 organism $\times 10^3/l$). This might be attributed to the relatively higher water temperature (24.3 and 22.8 °C, respectively). The rise of water temperature was favored by most plankton organisms as indicated by Forsyth and McCallam, (1980), and Benzie (1984).

Further more, those mentioned periods were characterized by abundant protozoa, which perform major food for zooplankton. That was even emphasized by a significant correlation ($r = 0.67$) between the mentioned communities.

Although physical factors (Benzie, 1984), and chemical conditions (Lind, 1974) were reported to control the abundance of zooplankton, no significant relationships was obtained between these parameters in the present study.

Protozoa

On considering water pollution, protozoa seem to be an excellent tool to assess both toxicity and pollution: they are regarded as biological indicators of pollution when their presence or absence can be related to particular environmental conditions, and they are considered test organisms when a species or population is used to evaluate the toxicity of relevant toxic compounds (Nicolau *et al.*, 2001). Certain genera of ciliated protists representing healthy environmental conditions, could be employed as biological indicators for system performance process, because there is a strong positive correlation between the abundance of some protozoa and chemical parameters (Scholz and Martin, 1998; Jiang and Shen, 2007).

In the present study, Protozoa persisted all over the year, and they contributed 90.5 % of the total population density of the examined zooplankton in raw water samples. During the present study, the total of 61 genera of protozoa was identified in raw water samples and completely missing in filtered, treated and tap water samples. This may be attributed to the added primary chlorine at the beginning of water treatment, indicating its effectiveness.

Generally, the maximum density of Protozoa was observed during Autumn and Spring with a peak in September and April, which agreed with El-Bassat (1995). Monthly results showed a significant correlation between total protozoa and: manganese, Calcium, Ammonia plus dissolved oxygen. On the other hand, a negative significant correlation was found to occur between population density of protozoa and both rotifers, and total coliform bacteria, which agreed with Sibille *et al.* (1998); Stevik (1998); Bomo *et al.* (2004); Chabauda *et al.* (2006); and Papadimitriou *et al.* (2010).

Ciliates are considered as important creatures in nutrient recycling (Senler and Yildiz, 2004). Moreover, ciliates respond more quickly to environmental contamination than other organisms because of their high reproduction rates, sensitivity, and variety of occupied niches (bacterivores, algivores, carnivores, omnivores). Changes in species diversity and structure are reliable and generally useful means for assessing the biological effects of pollution (Velho *et al.*, 2005; Madoni and Zangrossi, 2005). Recent studies demonstrated that many periphytic forms play a positive role in improving and maintaining water quality (Zhang and Song, 2000; Zhang *et al.*, 2001a,b; Xu *et al.*, 2004). This could be observed in the present study where monthly results of ciliates showed significant positive and negative correlations with physicochemical parameters. On examining those relationships in further detail, only three ciliate protozoans (*Cinetochilum*, *Lembadion* and *Lacrymaria* spp.) showed significant correlation with ambient temperature. In addition, eight ciliated protozoans (*Euplotes*, *Cyclidium*, *Didinium*, *Dileptus*, *Blepharisma*, *Holoticha*, *Strichotricha*, *Colpidium* spp.) showed significant correlation with ammonia. These results agreed with Xu *et al.* (2004a and 2004b) who

found that ciliates are likely to play a positive role in maintaining and improving water quality in aquatic environments with high-level ammonium, such as sewage treatment systems.

Rotifera

Rotifers are sensitive to water quality changes (Marneffe *et al.*, 1996). They are good indicators of saprobity (Authman, 1998). They can be used to indicate the performance of certain types of water treatment process (Spellman, 2003). They have positive correlation with water temperature, turbidity; and a negative one with alkalinity. Previous works indicated that temperature control birth and growth rates of rotifers (Galkovskaga, 1987). Turbidity (Menzel and Roth, 1972; Abd El- Mageed, 1995; and Authman, 1998) and alkalinity (Authman;1998) affects negatively on growth and distribution of rotifers.

Bacterial Investigation

In raw water samples, the presence of total coliform, fecal coliform, and *E. coli* indicates that the water was contaminated by fecal material from municipal waste water discharge, or septic leachate (An *et al.*, 2002). Total coliform and *E. coli* coexist with pathogenic organisms (*Salmonella* and *Shigella*). Thus, their presence may reflect the presence of enteric pathogens (USEPA, 1999; An *et al.*, 2002). In a survey by Seyfried and Harris (1990), over 94% of the thermo tolerant coliforms isolated from human faeces were identified as *E. coli*. Accordingly, the observed high counts of bacterial indicator (TC), here, were detected in the warmer seasons, (Spring and Summer).

In filtered water samples, total coliform and fecal coliform occurred very scarcely, which may be attributed to contamination of sand filter system by wastes of warm-blooded animal such as birds.

In treated water samples, the absence of total coliform, fecal coliform, *Salmonella* and *Shigella* groups could be attributed to the effectiveness of disinfection process. These results indicate the safety of treated water according to the WHO (1998) and Egyptian Standards (2007).

In tap water samples, Total coliforms occurred in very few numbers, though should be absent immediately after disinfection, indicating inadequate treatment (WHO, 2008). A number of research studies have shown that coliform bacteria can grow within drinking water distribution systems and can be a significant contributor to biofilm populations (LeChevallier, 1990; Chowdhury, 2012). It has been shown that the presence of significant densities of coliforms within distribution systems represent a health risk to water consumers (Edberg *et al.*, 1994), but can reveal contamination through ingress of foreign material (WHO, 2008). Marciano-Cabral *et al.* (2010) reported that despite effective treatment of drinking water, microbes can enter water utility distribution systems and hence the plumbing within building premises. Additionally, biofilm formation may add account for the persistence of microbes in the distributing system. These findings indicate that further treatment of drinking water before consumption is essential for avoiding potential health hazards.

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ARABIC SUMMARY

دراسة عشائر الكائنات الدقيقة في محطة معالجة مياه الشرب بشبين الكوم-المنوفية - مصر

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اجريت هذه الدراسة بمحطة شبين الكوم الجديدة لمعالجة مياه الشرب، ولقد درست الكائنات الدقيقة بمراحل مختلفة من المعالجة. ولقد اثبتت النتائج كفاءة عملية المعالجة إلا انه وجدت بعض البكتريا فى صنابير المياه بالمنازل مما يتطلب تجديد شبكة المياه. ولقد اثبتت الدراسة الحالية ان اغلب القياسات الفيزيائية والكيميائية تقع داخل الحدود المسموح بها فى القوانين. كما ان البكتريا القولونية والبرازية متواجدة فى المياه قبل دخولها محطة المعالجة وكذلك الكائنات الدقيقة وذلك نتيجة تلوث تلك المياه بملوثات مختلفة. كما اثبتت هذه الدراسة أيضاً وجود ارتباط احصائى بين تلك الاولييات خاصة الهدبية منها وبين بعض الملوثات العضوية والمعدنية ولذلك توصى الدراسة باستخدام هذه الكائنات ككواشف حيوية لأنواع التلوث المختلفة. كما ان العجليات ثبتت امكانية استخدامها للدلالة على كفاءة بعض مراحل المعالجة.