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Microbiological and Chemical Aspects on Some Fresh Water and Industrial Waste Water Samples

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Abstract

Both microbiological and chemical analysis of water and industrial wastewater samples collected from Dakahlia governorate (Egypt) were carried out. The microbiological examination involved the measurement of microbial total count, specific bacterial groups, pathogenic bacteria yeast, and fungi. The isolation and purification of different bacterial groups were also performed from different samples of industrial wastes. Trial to reuse the industrial effluents was also made. The chemical analysis included the measurement of conductivity, alkalinity, hardness, sulphate, PH, total dissolved solids, chloride, as well as dissolved oxygen. Results of the microbiological examination exhibited presence of yeast, fungi and bacteria. Short rods were the most bacterial isolates followed by spore formers and coccoid-shaped bacteria which came last in their count. Data also showed that neither water samples nor industrial wastes contain pathogenic bacteria when using specific cultivation media. Results of the chemical analysis showed that all measured parameters were found within the limitation either national or that of international law. Some samples exhibited higher values than that of permissible limits for some measured parameters. Furthermore, data proved the possibility of using the tested industrial wastes in production of both biomass and microbial enzymes as well. The bacterial treatment of industrial wastewater leads also to heavy metal reduction up to more than 50 per cent in some cases.

Introduction

The water that the employees drink must meet public health standards from both chemical and microbiological point of view. This can achieve when the water source tested by methods recommended in the Standard Methods for the Examination of Water and Wastewater (APHA, 1992). The bacteriological standards for drinking water that coliform bacteria must not be present at levels indicating contamination of the water by sewage. Total plate counts of the water sometimes are made to indicate when trouble may be incipient so that such trouble can be forestalled. More likely to be important is the chemical composition of the water, which must be suited to the use to be made of it. Thus hard water is undesirable in pea canning and brewing. Iron and manganese are also bad in beet canning and in brewing, excessive organic matter may lead to off-flavors etc (Frazier and Westhoff, 1987)

In Dakahlia governorate, industrial wastewater from oil and soap company (Sandoup), fertilizer factory (Talkha) and fodder factory (Sherbin) discharge canals directly attached to the river Nile. Therefore they are considered as serious pollution sources for drinking water. Man and animals are exposed to uptake of heavy metals from air, food and water. Agricultural, industrial and domestic wastes are the major sources of surface water pollution with heavy metals (Polprasert, 1982). The purpose of this investigation is to examine some sources of either drinking or waste water in Dakahlia governorate from both chemical and microbiological viewpoint.

Materials and Methods

Tested samples: Water samples were collected from

different supplies at Dakahlia governorate, from different drinking water, treatment plants, compact units (raw water from the intake of each one and treated water from taps inside the stations), and from river Nile and its canals. Wastewater samples were collected from fodder factory of Sherbin, washing water, (FFSH), fertilizer factory of Talkha, mixed with other wastes, (FFT), oil and soap company of Sandoup (OSCS), sludge from water treatment plant of Sherbin (SWTP) and outlet electrical power plant (EPP). All samples were collected in 5 litre polyethylene bottles for the chemical analysis and one litre sterile glass bottle for the microbiological analysis using the procedure according to the Standard Methods for the Examination of Water and Wastewater (APHA 1992).

Chemical analysis: PH values were measured by HANNA instrument, HI 931401 Microprocessor pH meter. Both conductivity and TDS were measured using OAKLON, TDS/conductivity meter. Chloride, sulfate, alkalinity, hardness and dissolved oxygen were determined as described in the Standard Methods for the Examination of Water and Wastewater (APHA, 1992). The concentration of metals in all tested samples was determined by using 2380-Perkin - Elmer flame atomic absorption spectrophotometer following the recommended standard operating procedure. BDH standard solutions were used and internal standards were applied to check for interference problems. The metals concentration was determined in triplicate and the mean value was recorded. All chemicals used in this investigation were of high grade. This measurement was also done according to the Standard Methods for the Examination of Water and Wastewater (APHA, 1992).

Microbiological Procedures

Cultivation media used: The following general and specific cultivation media were used for different purposes according to the Bacteriological Analytical Manual for Foods (FDA, 1976). Tryptone glucose agar and nutrient agar (TGA & NA); was used for isolation and for total count of the microbial load of the tested water samples. Milk agar (MA); was used for isolation and counting of proteolytic bacteria. Fuchsin lactose agar (Endo C agar) was used to detect the bacterial members of the family *Enterobacteriaceae*. Mannitol sodium chloride phenol red agar (MSPRA), was specifically used for detection of *Staphylococcus sp.* Sabouraud glucose agar medium (SGA) was selectively used for *Enterococcus*. Bactoagar F-medium (BAF) is used for the detection of *Pseudomonas spp.* Nutrient agar containing 1 per cent olive oil (OA) was used for isolation and counting the lipolytic bacteria. The composition of these culture media was as described in Oxoid (1982).

Isolation of microorganisms naturally occurred in tested samples: One ml sample of examined industrial waste was dispersed in appropriate volume (9 ml) of distilled water and different serial dilutions were made with vigorous shaking to give a final dilution of 10^{-3} . The original sample was prepared in case of drinking water. One-ml sample was then taken and plated on different cultivation media and appropriate periods of incubation at 37°C according to the purpose required. The presence of clear zone around the colonies against an opaque white background in Plates of MA indicated that these organisms were proteolytic (Srinivasan *et al.*, 1964). Plates of OA media were flooded with concentrated CuSO_4 solution and bluish green colonies surrounded by precipitates were detected as lipolytic bacteria (Mourey and Kilbertus, 1976). Other bacterial groups were detected and counted on other specific media noted above. The fermentation of Mannitol to acid, which serves as a guideline for pathogens, is indicated by a colour change of the PH indicator, phenol red. Total bacterial count was carried out using TGA and NA media (Oxoid, 1982). Fungi and yeasts were also counted using SGA medium as described in Oxoid (1982).

Purification of obtained bacterial isolates: From the preceding step, well-separated colonies were selected and transferred to the slope agar. In order to make sure the purity of these isolates, each of them was suspended in sterile water and recultivated again for two cycles followed by single colony isolation after streaking onto plates. Slope cultures were considered pure when they showed uniform morphological feature by microscopic examination. Preliminary identification of the three isolates was achieved by Gram reaction, spore formation, motility, capsule formation, presence of water-soluble pigments (Sneath *et al.*, 1986). Five bacterial isolates namely; *Bacillus sp* FFT50; *Pseudomonas sp* OSCS15; *Bacillus sp* FFSH6; *Bacillus sp* SWTP 30 and *pseudomonas sp* EPP22 were

selected and maintained on NA slant at 5°C till use.

Microbiological treatment: The tested wastewater was diluted to 50 per cent by distilled water on which the selected bacterial isolates allow to grow. Each isolate was grew on a waste from which it was isolated for 36 hr incubation period at 30°C. The bacterial growth was then enzymatically detected as follows :

Quantitative measurement of Proteinase activity: The casein digestion method (Lupin *et al.*, 1982) was followed. A unit of proteinase, tyrosine unit (TU/ml cultural filtrate) was defined as that quantity of enzyme which produce TCA-soluble fragments giving blue colour equivalent to μg tyrosine under the assay conditions.

Quantitation of lipase activity: Lipase activity was titrimetrically determined. The reaction mixture contained 5.0 ml of 5 per cent olive oil emulsion in 7 per cent gum acacia, 5.0 ml of 0.2M tris-HCl buffer (7.5), 0.2 M CaCl_2 , 1.0 ml of enzyme solution and 2.0 ml of distilled water. After one-hour incubation, the total amount of liberated fatty acids was titrated against N/100 NaOH. The same procedure was followed with boiled enzyme solution as a control (Chopra and Chander, 1983).

Results and Discussion

Chemical Characteristics

Fresh Water: From the pH values shown in Table 1, it is clear that all raw water (Nile and canals) and drinking water samples have a slight basic effect. This indicated that the pH values of raw water samples did not greatly affect during the treatment processes. All of the values were within the national and international limits (WHO limitation of pH value is 6.5-9.5). Because of the heavy metals in water are stable at the pH values of less than approximately 7.0. The toxicity of the heavy metals can increase with basic pH values. Lead was found to be more toxic at the pH values of 8.5 than pH 6.5. Waters with high dissolved solids generally are inferior palatability and may induce unfavorable physiological reaction in the transient consumer. For this reason, a limit of 500 mg dissolved solids/L is desirable for drinking waters. High-mineralized waters also are unsuitable for many industrial applications. Analyses of dissolved solid are important in the control of biological and physical waste water treatment processes. Data in Table 1 show that TDS of investigated tap water samples did not vary greatly than Nile water. In this respect, World Health Organization (WHO) recommends that total dissolved solid in drinking water must not increase over 500 ppm. This indicates that samples are within limits except sample of oil and soap company Sandoup which exceed the MAC (2000 ppm). Positive relationship should be found between TDS and conductivity since both of them are greatly reflected values to the soluble salts.

The water hardness property is due mainly to the presence of carbonates, bicarbonates, sulphates, or other compounds.

of calcium and magnesium. Calcium compounds are the most common source of hardness in water supplies as they readily dissolved from limestone deposits. All types of samples are within the permissible limits of WHO (guideline value 500 ppm) and Egyptian Standards, ES, (300-400ppm).

Alkalinity of water is its acid-neutralizing capacity, that it is sum of all the titratable bases. Alkalinity is a measure of an aggregate property of water and is significant in many uses and treatments of natural waters and wastewater. Waters with the concentration of sulphate ions more than 250 mg /L may be destructive to concrete structures owing to the formation of gypsum. The presence of chlorides and sulphates in water in appreciable concentrations is indicative of an elevated solute residue which makes the water poorly suitable for feeding of steam boiler plants or as process water in the manufacture of certain polymers, synthetic rubber, some grades of paper, etc. Magnesium and calcium sulphates and chlorides are responsible for noncarbonate hardness of water.

Wastewater: The chemical analysis of tested wastewater including the heavy metals values are illustrated in Table 2. Data in this Table show that all the industrial wastewater samples have the same basic effect of pH but it is also within the national and international limits of pH values. Samples of fertilizer factory of Talkha have high pH value (11.2) exceeds the permissible limit. In case of TDS and conductivity, the sample of OSCS is the highest sample being 2300 ppm, 4600 $\mu\text{s}/\text{cm}$, respectively. The OSCS sample again gave the highest value in case of sulphate and chloride being 322.5ppm and 1349.7ppm, respectively.

Values of heavy metals in water and wastewater: Metallic ions, or positively charged atoms, often accumulate in human organs. Lead and cadmium can displace calcium from bones and cause them to become brittle. Lead, cadmium, mercury and chromium can concentrate in liver and kidneys and cause damage and malfunctioning of these organs. The nervous system is susceptible to concentration of mercury, lead and copper, and the effects vary from brain damage of the peripheral nervous causing uncoordinated muscular control and poor eyesight. Data in Tables 1&2 show that the heavy metals concentrations of investigated water samples are in acceptable range. In addition, the concentration of copper in water is usually attributed to the corrosive action of copper salts as a result of the activity of the aquatic organisms. Manganese is also considered to be one of the essential elements in organic matter, which influence on its occurrence in natural water (El-Sharouny, 1989).

Microbiological View

Sources of fresh waters: Four different microbial groups were examined in four sources of fresh waters as show in Table 3. No pathogenic bacteria were found in all tested

Table 1 : Chemical analysis and heavy metals values of fresh water samples .

Chemical Analysis	Water sources														
	Stations				Compart Units				Canals						
	Sherbin WTP		Bassat WTP		Abu-Ayssa		Fisco		El-Sabai		Mest-Abaret				
	R.W	T.W	R.W	T.W	R.W	T.W1	T.W2	R.W	T.W1	T.W2	R.W	T.W	Mest-Abaret	Mest-Abaret	Mest-Abaret
pH	7.9	7.6	8	7.8	7.9	7.3	6.9	8	7.3	7.6	7.96	8	8	8	8.33
RDS, mg / l	222	224	217	220	169	181	190	176	180	183	319	279	984	984	322
Conductivity, $\mu\text{S}/\text{cm}$	444	448	434	440	338	362	380	352	360	366	638	548	1968	1968	650
Alkaline, mg l	0	0	0	0	0	0	0	0	0	0	0	0	52	52	26
CaCO ₃	0	0	0	0	0	0	0	0	0	0	0	0	104	104	52
Total	192.3	184.6	200	180	173.1	150	115.4	177	153.5	161.5	148	208	624.67	624.67	264
Hardness, mg _{Ca} + Mg ₂₊ / l	46.7	46.7	42	42	37.4	37.4	37.4	43.9	43.9	43.9	38.17	35.12	27.62	27.62	38.1
Total	194.2	194.2	174.8	174.8	155.3	155.3	155.3	190.3	190.3	190.3	140.95	129.52	74.43	74.43	15.28
Sulphate, mg/l	35	50.5	40.1	53.7	32.3	59.4	87	32.7	56.4	32.6	40	40.75	129	129	78.00
Chloride, mg / l	52	60	50	54	40	46	44	40	44	44	41.99	53.98	331.56	331.56	67.98
Dissolved oxygen, mg / l															
Heavy Metals															
Cu	16	35	16	29	13	10	--	13	13	--	19	5	10	10	47
Cd	7.2	10.8	7.1	6.05	5.9	6.5	--	5.1	5.8	--	7.3	7.2	4.1	4.1	6.5
Fe	89	76	77	67	80	78	--	72	74	--	107	78	78	78	97
Mn	99	53	99.4	94.6	60	82	--	73	70	--	103	107	114	114	97

Table 2: Chemical analysis and heavy metals values of industrial wastewater samples.

Chemical Analysis	Fertilizer Factory (Talkha) FFT	Oil & Soap Company Sandoup OSCS	Fodder Factory (Sherbin) FFSH	Sludge of WTP (Sherbin) SWTP	Power plant (outlet) EPP	
pH	11.2	7.5	7.8	8.2	7.8	
TDS, mg/L	283.0	2300.0	11.4	191.0	186.0	
Conductivity; Us	566.0	4600.0	22.7	382.0	372.0	
Alkalinity, mg/L	ph.ph.	184.0	0.0	0.0	0.0	
	OH	48.0	0.0	0.0	0.0	
	CO ₃ ²⁻	272.0	148.0	72.0	188.0	
	Total	320.0	148.0	72.0	188.0	
Hardness, mg/L	Ca ²⁺	66.2	35.8	0.0	37.8	
	Mg ²⁺	0.0	4.0	4.7	16.1	
	Total	165.1	399.9	19.4	155.3	
Sulphate, mg/L	47.6	322.5	0.0	39.5	24.8	
Chloride, mg/L	75.9	1349.7	14.0	39.9	31.2	
Dissolved oxygen, mg/L	8.0	0.0	7.3	8.7	8.0	
Heavy Metals ug/L	Co	16.0	19.0	2.0	41.0	ND
	Cu	6.5	13.4	6.4	7.3	ND
	Fe	68.0	76.0	85.0	75.0	ND
	Mn	94.0	54.0	90.0	76.0	ND

* Each value expresses the mean of three replicates. ND : Not Detected.

Table 3: Microbiological examination of tested water sources.

Source of water	Water	Examined microbial groups (cfu / m1 x 10 ³)							
		Total variable count		Specific group			Fungai and yeast		
		NA	TAG	OA	MA	BAF	SGA		
Water treatment plant:									
Sherbin	R.W.	120.0	145.0	25.8	45.6	130.4	95.4	110.2	
	T.W.	68.5	75.3	15.7	18.4	90.8	30.3	65.8	
Bossatt	R.W.	95.4	110.4	40.6	55.5	156.6	105.7	135.2	
	T.W.	70.7	85.3	30.4	35.2	105.3	65.7	95.3	
Compact unit:									
Abo Arssa	R.W.	185.0	210.0	65.3	73.8	110.3	75.1	30.2	
	Unit 2	T.W.	120.0	165.0	40.2	33.4	85.7	40.3	22.4
	Unit 3	T.W.	108.0	140.0	35.7	22.4	60.4	32.8	25.3
Fasco	R.W.	250.3	285.2	88.6	95.9	125.5	100.2	56.3	
	Unit 2	T.W.	113.4	104.3	39.2	77.3	70.4	66.4	30.0
	Unit 4	T.W.	102.2	98.8	45.6	52.2	45.6	33.7	23.8
Canals									
El-Shahel	R.W.	210.8	255.2	80.4	65.8	230.0	145.0	85.8	
Mahalet Engak	R.W.	240.3	265.4	45.6	25.5	255.0	125.0	65.9	
Ground:									
Meet-Anter	G.W.	80.7	105.8	30.8	13.7	60.8	13.5	25.3	
Meet-EI-Amel	G.W.	95.9	115.6	25.9	18.4	52.5	18.6	30.6	

water samples. This may be ascribed to non-recent sewage effluent entry or present at the time of sampling. In respect to the total viable count, data exhibited that the raw sample of Sherbin (WTP) was more contaminated than the sample of Bossatt (WTP) by about 20.5 per cent when using NA medium in the examination. This ratio was increased to 23.9 per cent in case of using TGY culture

medium indicating the effect of the cultivation media on the appearance of the microbial density. This may be attributed to that the raw water of the treatment plant of Sherbin mixed with El-Sahel canal beside river Nile water. While the water of Bossatt is directly taken only from the river Nile. So, the possibility of contamination by canal water is present in the first case. On the other hand, the sample

Bossatt showed higher population regarding the specific bacterial groups than that one of Sherbin. For example, the different ratios were 36.5, 17.8 and 16.7 per cent for the lipolytic bacteria (OA), proteolytic bacteria (MA) and *Pseudomonas spp.* (BAF), respectively. There were also considerable variations between these two plants (WTP) for both fungi and yeasts when using SGA cultivation medium. faecal coliform in the groundwater of upper Egypt as well as some fungal genera such as *Penicillium sp.*, *Rhizopus sp.*, *Fusarium sp.*, *Trichoderma sp.* and *Aspergillus sp.* were found. The same results were also reported by El-Sharouny (1989).

Efficiency of water purification units: The comparison between the two plants of water treatment (WTP) and compact units (CU) was also considered by means of reduction of the microbial load in raw water. Data exhibited considerable variation between the efficiency of the two types of WTP (Table 4). The calculated percentages of microbial population reduction were 42.9 and 48.1 per cent in Sherbin samples (WTP) when using NA and TGY cultivation media, respectively. These ratios decreased to 25.9 and 22.7 per cent in case of Bossatt WTP. With respect to the CU, data illustrate that unit No. 3 of Abo Arssa was more efficient than the unit No 2. The reduction percentages were 35.1 and 21.4 per cent for unit No 2 While they were 41.6 and 33.3 per cent for unit No. 3

when using NA and TGY media, respectively. The variation is also clear from the higher densities of both fungi and yeasts in case of Bossatt sample by about 9.3 and 18.5 per cent, respectively. *Fusarium sp.*, *Aspergillus sp.* and *Penicillium sp.* are the most common fungal genera found in river Nile stream (El-Sharouny, 1989). Higher microbial population was found in raw water of the Fasco sample than that of Abo Arssa. For the total bacterial count, 26.1 and 26.4 per cent were found in Fasco sample over than AboArssa sample. For the other microbial groups, data proved that Fasco sample increased by 26.3, 23.0, 12.1, 25.1 and 46.4 per cent for lipolytic bacteria (OA), proteolytic bacteria (MA), *Pseudomonas spp.* (BAF), fungi and yeast, respectively. This may be due to that the original source of the raw water of Fasco is more polluted since it have more refuses and organic matter which play main role as energy sources for many of microorganisms. Observable differences were found between the examined two samples of canals, Which may be attributed to the different location. The importance of polluted water of canals that it can act as vehicles for transmitting the contamination microbes. Additionally, the lower contaminated waters were that of ground water either the site of Meet-Anter or the other location of Meet El-Amel (Table 4). *E.coli* and *Streptococcus faecalis* in underground and Nile waters were found (Hosney *et al.*, 1990). For Fasco compact units,

Table 4: Comparison between water treatment plants and compact units by means of the microbial count reduction.

Source of water	Total variable count		Specific group			Fungai and yeast	
	NA	TAG	OA	MA	BAF	SGA	
Water treatment plant:							
Sherbin	42.9	48.1	39.2	59.7	30.4	68.2	40.3
Bossatt	25.9	22.7	25.1	36.6	32.8	37.6	29.5
Compact unit:							
Abo Arssa							
Unit 2	35.1	21.4	38.4	54.7	22.3	46.3	25.8
Unit 3	41.6	33.3	45.3	69.7	45.2	46.3	16.2
Unit 2	54.7	63.4	55.8	19.4	43.9	33.7	46.7
Unit 4	59.2	65.4	48.5	45.6	63.7	66.4	57.7

Table 5: Microbiological examination of tested wastewater

Sources of wastewater	Examined microbial groups (cfu/ml x 10 ³)						
	Total viable count		Specific groups			Fungi and yeasts	
	NA	TGY	OA	MA	BAF	SGA	
Fertilizer factory (Talkha)	90	165	50	85	120	60	45
Oil & soap company (Sandoupe)	180	210	140	60	180	75	15
Fodder factory (Sherbin)	215	260	85	160	210	32	24
Sludge of WTP (Shebin)	205	245	105	180	260	80	90
Electrical Power plant (outlet)	95	115	40	40	95	20	15

Table 6: Activity of some enzymes produced by specific bacterial groups naturally occurred in tested wastewater

Source of wastewater	Bacterial isolated used	After incubation period (hr) of					
		Proteinase activity TU/ml/min.			Lipase activity, 0.01N NaOH/ ml/10 min.		
		12	24	36	12	24	36
Fertilizer factory (Talkha)	<i>Bacillus</i> sp. FFT50	22.0	36.4	65.3	32.8	38.6	49.2
Oil & Soap company (Sandoup)	<i>Pseudomonas</i> sp. OSCS 15	28.4	39.3	58.3	40.0	55.6	72.4
Fodder Factory (Sherbin)	<i>Bacillus</i> sp. FFSH6	82.6	112.4	125.6	38.2	51.8	64.9
Sludge of WTP (Sherbin)	<i>Bacillus</i> sp. SWTP 30	45.0	60.5	99.0	24.8	32.6	45.4
Electrical Power Plant (outlet)	<i>Pseudomonas</i> sp. Pp22	13.2	18.3	36.5	2.8	8.6	15.2

Table 7: Effect of microbial treatment on metal ions concentration in industrial wastewater.

Bacterial	Heavy metal (mg/L)			
	Fe	Mn	Pb	Cd
<i>Bacillus</i> sp. FFT 50				
Before	0.97	0.51	0.49	0.048
After	0.97	0.41	0.37	0.038
Fold	1.00	1.24	1.32	1.260
<i>Pseudomonas</i> sp. OSCS15				
Before	1.09	0.51	0.52	0.042
After	0.92	0.38	0.24	0.037
Fold	118	1.34	2.17	1.140
<i>Nacosis</i> sp. FFSH6				
Before	1.29	0.39	0.45	0.043
After	0.97	0.37	0.35	0.035
Fold	1.33	1.05	1.29	1.230
<i>Bacillus</i> sp. SWTP 30				
Before	1.41	0.41	0.043	0.0045
After	0.62	0.38	0.033	0.0033
Fold	2.27	1.08	1.30	1.360
<i>Pseudomonas</i> sp. EPP 22				
Before	1.07	0.50	0.59	0.044
After	1.07	0.46	0.45	0.320
Fold	1.00	1.09	1.31	1.380

progressive reduction was obtained in bacterial count reached 54.7 and 63.4 per cent in unit No 2 in case of NA and TGY isolation media, respectively. The reduction ratios were 59.2 and 65.4 per cent in unit No 4 within NA and TGY isolation media, respectively. Almost similar trend of these results was obtained in case of both specific bacteria (BAF, OA and MA), fungi and yeasts. Results also proved remarkable differences between the two plants (WTP) and compact units in their efficiencies. Because of the closed system of compact units, the reduction percentages were higher than that of WTP which depends on the open system which may help the proliferation of the microbial load during the time of the process.

Data also demonstrate that, although the efficiency of compact units is higher than that of WTP, the total number of the organisms after treatment is higher in case of compact units than those of raw water in case of the WTP.

This may be demonstrated by the location of compact units that lies on the canals that can increase the chance of the water pollution.

Waste waters: Recorded data in Table 5 showed that wastewater of fodder factory of Sherbin was much higher contaminated than the other wastes regarding the total bacterial count when using either NA or TGY. The sample of oil and soap company of Sandoup showed to be highest in case of lipolytic bacteria (OA). Sludge of WTP (Sherbin) showed to have high values of proteolytic bacteria (MA), *Pseudomonas spp.* (BAF), fungi and yeasts which came last. Both samples of fertilizer factory of Talkha and electrical power plant (outlet) were in moderate area of pollution. The variations of these samples in their microbial contents are ascribed to the differences of their chemical composition, which act as a microbial substrate for their growth and enzyme production.

Biological treatment of wastewater: The possibility to use the tested wastewater as a microbial cultivation media (50% v/v with distilled water) without any addition of nutrients was performed using different bacterial isolates, which their growth can be enzymatically measured. Listed data in Table 6 showed that *Bacillus sp.* FFSH6, gave the highest proteinase activity being 125.6 tyrosine unit per ml cultural filtrate (TU/ml) after 36 hr incubation at 37°C. *Bacillus sp.* SWTP30 obtained 99.0 TU/ml at the same conditions of cultivation. Low units of proteolytic enzyme was obtained by *Bacillus sp.* FFT50 (65.3 TU/ml), *Pseudomonas sp.* OSCS 15 (58.8 TU/ml) whilst 36.5 TU/ml was found in case of *Pseudomonas sp.* EPP22. The measured lipolytic activity was also demonstrated in the same Table. *Pseudomonas sp.* OSCS15 gave the highest lipase activity after 36 hr incubation at 37°C being 72.4 lipase units equal to 0.01 N of NaOH/ml/10 min. *Bacillus sp.* FFSH6 come second giving 64.9 lipase unit using fodder factory waste. The waste of fertilizer factory of Talkha was suitable to give 49.2 lipase unit by *Bacillus sp.* FFT50. Sludge of Sherbin water treatment plant and electrical power plant (outlet) gave 45.4 and 15.2 lipase units by *Bacillus sp.* SWTP30 and *Pseudomonas sp.* EPP22, respectively.

From the results obtained in this investigation, it could be concluded that survival of microorganisms in water supply varied according to the chemical composition, PH, suspended organic matter, and presence of special types of microbial groups. The present study also showed negative results to the presence of *fecal Streptococci*, indicating no recent sewage pollution of such water sources. With respect to the compact unit, this study should attract the attention of the human health authorities to find out urgent procedures to keep these units enough away from the canal, to get rid the high contamination. In view of the fact that polluted waters can act as vehicles for transmitting the microorganisms, so, the authors argue the public health authorities to impose acts regulating the handling of all vegetables which are consumed raw and washed by these waters.

Removal of heavy metals: Table 7 illustrate the effect of bacterial treatment of industrial wastewater by means of heavy metal reduction. Data showed that *Bacillus sp.* SWTP30 was the most efficient in removing Fe, while *Pseudomonas sp.* OCS15 was the most active in reduction of Mn and Pb. The bacterium *Pseudomonas sp.* EPP22 exhibited the higher value in Cd reduction.

Remacle *et al.* (1992) found that the aerobic growth of *Alcaligenes denitrificans* resulted in removal of more than 95 per cent of the dissolved metal. Grappelli *et al.* (1992) reported that many strains of *Arthrobacter spp.* Diffused in the soil and in a condition of environmental stresses, have been proved to be effective in heavy metal capture; Cd, Cr, Pb, Cu, and Zn. Huang and Chiu (1994) studied the removal of trace Cd (II) from aqueous solutions by fungal adsorbents. They found that more compact pellets of *Rhizopus oryzae* were more effective in binding of Cd (II) ions.

References

American Public Health Association, 1992. Standard Methods for the Examination of Water and Waste Water, 18th ed., APHA, Washington, D.C.
Chopra, A.K. and H. Chander, 1983. Factors affecting lipase production in *Syncephalostrum racemosum*. J. Appl. Bacteriol., 54, 163-169.

El-sharouny, H.M.M., 1989. Pollution effects on Fungi in habiting organic debris in the Nile water. Egypt. J. Microbiol., 24: 405-414.
F.D.A., 1976. "Bacteriological Analytical Manual for Foods" 4th ed., Food and Drug Administration, Washington, DC.
Frazier, W.C. and D.C. Westhoff, 1978. Food Microbiology. 3rd ed., Tata Mc Graw-Hill publ. Co. Ltd, New Delhi.
Grappelli, A., L. Campanella, E. Cardarelli, F. Mazzei, M. Cordatore, W. Pictrosanti and M. Suzuki, 1992. Metals removal and recovery by *Arthrobacter spp.* Biomass. Water Sci. Technol., 26: 2149-2152.
Hosny, I., H. El-Zanfaly, M. Fayez and A. Shaban, 1990. Bacteriological evaluation of underground waters in Egypt. Egypt. J. Microbiol., 25: 277-290
Huang, C. and H.H. Chin, 1994. Removal of trace Cd (II) from aqueous solutions by fungal adsorbents: An evaluation of self-immobilization of *Rhizopus oryzae*. Water Sci. Technol., 30: 245-253.
Lupin, I.V., D. Korner, A. Tafel and H. Ruttloff, 1982. Application of automatic protease determination in a fermenter. Enz. Microbiol. Technol., 4; 104-106.
Mourey, A and G. Kilbertus, 1976. Simple media containing stabilized tributyrin for demonstrating lipolytic bacteria in foods and soils. J. Appl. Bacteriol., 40: 47-51.
Oxoid, 1982. "The Oxoid Manual of Culture Media, Ingredients and other Laboratory Services". 5th ed., Oxoid Limited.
Polprasert, C., 1982. Heavy metal pollution in the chaophraya River Estuary. Thailand Water Res., 16: 775 - 784.
Remacle, J., I. Muguruza and M. Fransolet, 1992. Cadmium removal by a strain of *Alcaligenes denitrificans* isolated from a metal polluted pond. Water Res., 26:923-926.
Sneath, P. H.A., N.S. Mair, M. Sharpe and J.G. Halt, 1986. Bergys Manual of Systematic Bacteriology. Vol. 2, Williams & Wilkins Baltimore.
Srinivasan, R.A., M.K.K. Lyengar, I.J. Babbar, S.C. Chakravorty, T.A. Didami and K.K. Lya, 1964. Milk clotting enzyme from microorganisms. Appl. Microbiol., 12: 475-478.
Thompson, L.M. and F.R. Troeh, 1978. Soils and Soil Fertility. 4th ed., McGraw - Hill, Inc., New Delhi.