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ABSTRACT

This study aims to the evaluation of the microbial quality of the River Nile water at Damietta Branch. Water samples were taken from the branch, during the period from autumn, 2005 till the summer of 2006. Some environmental parameters such as temperature, transparency, depth and pH were monitored. In addition, the bacteriological analyses involved total viable bacterial counts (TVBCs), total coliforms (TC) and estimation of faecal coliforms (FC), faecal streptococci and pathogenic bacteria. The results of physicochemical parameters showed that the temperature values varied from 17°C to 25°C and the transparency from 40cm to 220cm. Whereas, the depths varied from 3m to 25m and pH values from 7.24 to 8.44. The bacteriological analyses showed that TVBCs ranged from 10.8x107 to 150 x107 cfu/ml and from 8.8x107 to152 x107 cfu/ml at 22°C and 37°C, respectively. However, maximum counts were recorded during summer and the minimal were detected in winter. The results of the faecal indicators counts revealed that their densities increased from up- to down stream. The pathogenic bacteria were identified using API 20E strip system (BioMereux). One hundred pathogenic bacterial isolates representing eleven genera were identified to species level. This included, the following suggested names: Esherichia coli (16%), Klebsiella pneumoniae (14%), Pseudomonas aeruginosa (12%), Pseudomonas flourcsence (4%), Salmonella colerasuis (11%), Shigella sp. (9%), Serratia liquefaciens (8%), Proteus vulgaris (8%), Acinetobacter sp.(7%), Brenneria nigrifluens (5%), Flavimonas oryzihabitans (3%) and Chryseomonas lutecla (3%). The results of the present investigation revealed that, the River Nile water at Damietta Branch was subjected to sewage pollution during the study period.

1. INTRODUCTION

One of the most important factors of water pollution is the microbial contamination; especially with pathogenic microorganisms. Enteric pathogens are typically responsible for waterborne sickness (Karaboze *et al.*, 2003). Contamination of water is a serious environmental problem as it adversely affects the human health and the biodiversity in the aquatic ecosystem.

The use of indicator bacteria such as faecal coliforms (FC) and faecal streptococci (FS) for assessment of faecal pollution and possible water quality deterioration in fresh water sources is widely used (APHA, 1995). Youn- Joo *et al.* (2002) monitored coliform bacteria and *E. coli* in Lake Marinas (located on the border of Texas and Oklahoma) to determine the microbial pollution in the lake environment. Currently, coliforms and *E. coli* are of great importance among bacterial indicators used in water quality definition and health risk (Giannoulis *et al.*, 2005).

Pathogens are a serious concern for managers of water resources, because excessive amounts of faecal bacteria in sewage and urban run-off have been known to indicate risk of pathogen-induced illnesses in humans (Fleisher *et al.*, 1998). Several

species of gram-negative bacteria present in municipal wastewater are pathogenic. This pathogenisty is usually associated with certain components of the cell walls, in particular the lipopolysaccharide, also known as LPS or endotoxin, layer (Baron, 1996). Thus, identification of these pathogenic agents in water resources is beneficial for controlling and prevention planning of the infectious diseases.

In Egypt, the River Nile is the main source of drinking water. The Nile, unfortunately, receives heavy loads of industrial, agricultural and domestic wastes. Drinking water must meet specific criteria and standards to ensure that water supplied to the public is safe and free-from pathogenic microorganisms as well as hazardous compounds (WHO report, 1993).

Some studies have been published on the physico-chemical characters of the River Nile. Shaaban-Dessouki *et al.*, 1993; Abdo, 2004 (a) studied the physical and chemical characters of the River Nile at Damietta branch. Elewa and Gohar, (1999) studied the distribution of some heavy metals in the branch. In addition, studies dealing with the microbial quality of the River Nile waters

were recorded by El-Mongy, (1978); Haeikal, (1994); Abu-Shady *et al.* (1996); Rabeh, (2000); El-Fadaly *et al.* (2001); Sabae, (2004) and Sabae *et al.* (2006). The objective of this study was to determine the seasonal variations in microbial pollution indicators throughout Damietta Branch and to identify the pathogenic bacteria.

THE STUDY AREA

The River Nile travels along Egypt for about 940 km behind the High Dam. After passing Cairo, the Nile pursues a north westerly direction for about 23 km and divides at El-Qanater Barrage into two branches, each of which runs separately to the Mediterranean Sea, forming the Delta region between both branches. The western branch is Rossetta branch (239 km in length) and the eastern branch is Demietta Branch. about 242 km long. Damietta Branch receives a large amount of effluents of mostly untreated agricultural domestic and partially treated industrial wastewater (Zyadah, 1996). Damietta Branch is the study area under investigation.



Fig. (1): map showing the sampling locations at Damietta Branch.

2. MATERIAL AND METHODS

Water samples were collected seasonally (from subsurface layer) from seven stations at Damietta Branch between autumn, 2005 and summer, 2006. Sampling stations are shown in the map.

Station 1: located at El-Qanater region; Station 2: located in front of Benha City; Station 3: located in front of Zifta City; Station 4: located in front of Talkha City; Station 5: located at El- Serw region; Station 6: located before Faraskour Barrage; Station 7: located in front of Damietta City.

Water samples were collected using sterile glass bottles and transported in an icebox to the laboratory to be analysed within 8 hours.

1. PHYSICOCHEMICAL CHARACTERS

Some environmental parameters were measured in the field e. g. temperature, transparency, depths and pH.

1.1. BACTERIOLOGICAL EXAMINATIONS

The total viable bacterial counts (TVBCs) at 22°C and 37°C were determined using the spread-plate method (APHA, 1995).

The number of total and faecal coliforms was determined using the most probable number (MPN) method. Statistical tables were used to interpret the results to give the MPN of the bacteria. From each dilution 1ml was added to each of triplicate tubes containing 5ml of MacConkey broth. The tubes were then incubated at 37°C for 48hr for total coliforms and at 44°C (in water bath) for 24 hr for faecal coliforms.

The positive tubes were streaked on the Eosin Methylene Blue (EMB) agar plates using sterile loop and incubated at 37°C for 24 hr. Microscopic examination was carried out to ensure gram-negative, non-spore forming rods (APHA, 1995).

MPN of faecal streptococci was determined using azide dextrose broth at 37°C for 48hr. Positive tube was indicated by dense turbidity and confirmed using ethyl violet azide dextrose broth incubated at 37°C for 24hr. The formation of purple button at the bottom of the tube confirmed the presence of faecal streptococci (APHA, 1995).

1.2. ISOLATION AND PURIFICATION OF GRAM-NEGATIVE BACTERIA

Isolation of gram-negative bacteria in Dameitta Branch water samples were performed using MacConkey agar supplemented with 0.001 g/L crystal violet (Hausler and Koontz, 1970). Three hundred and five isolates were purified, screened and the suspected similar ones were grouped for the purpose of selection and identification processes.

1.3. IDENTIFICATION OF SOME GRAM-NEGATIVE PATHOGENS

One hundred isolates out of three hundred and five from the examined water samples were subjected to identification by biochemical characteristics using API 20E strip system (BioMereux). Each API 20E strip consists of twenty seven wells containing dehydrated media. The isolate to be tested was suspended in sterile saline and added to each well. The inoculated strip was incubated for 16-24h and the colour reactions were noted either positive or negative.

STATISTICAL ANALYSIS

The correlation coefficient (r) between the bacteria and the environmental factors using the computer programme Ms Microsoft Excel (ver., 2003).

3. RESULTS AND DISCUSSION

Table (1) shows that the highest temperature value (29.7°C) was recorded during summer, while the lowest one was (17°C) detected in winter. The results showed a noticeable seasonal trend of temperature. However. the variations in water transparency were almost local, without seasonal trends. The highest value of transparency was detected at Talkha (Station, 4) and the lowest one was recorded at Damietta (station, 7) this agrees with those reported by Abdo, (2004 b), which might be attributed to the effluents discharged from Talkha Electric Power Station.

The results show that the depths of the branch throughout the investigated period ranged between 3 and 25 m. The pH values of the Nile water were on the alkaline side. However, the highest pH value (8.44) was recorded at Damietta (station, 7) and the lowest was detected (7.24) in front of Faraskour Barrage (station, 6). The recorded pH values were favorable for bacterial multiplication.

The results of the total number of bacteria are shown in (Table, 2). The total number of bacteria ranged from 10.8×10^7 to 150×10^7 cfu / ml and from 8.8×10^7 to 152×10^7 cfu/ ml at 22°C and 37°C, respectively. Their highest number was recorded obtained during summer and the lowest one was detected in winter. The data showed that there is gradual increase in the bacterial counts from up-to down stream that is from El-Qanater to Damietta, which might be attributed to domestic, sewage and agricultural effluents discharge into Damietta Branch.

The highest bacterial counts were recorded for both groups (at 22°C and 37°C during summer. This might be due to the high temperature prevailing during this season. This finding was in accordance with El-Fadaly *et al.* (2001) and Sabae (2004).

Statistically, the correlation between the bacterial counts at 22° C and 37° C were

positive (r = 0.82). This agrees with those reported by Sabae, 1999.

The most probable number (MPN) of faecal indicators total coliforms (TC), faecal coliform (FC) and faecal streptococci (FS) are shown in (Table, 3). MPN of TC varied from 240 to $16 \times 10^4 / 100$ ml and from 40 to 7500/100ml water for FC. The counts of FS fluctuated between 4 and 2100/100ml water of Damietta Branch.

The highest bacterial indicators were recorded at Damietta (Station7). This might be explained by the effect of domestic and agricultural wastes discharge from the urbanized surrounding area (Shaaban-Dessouki *et al.*, 1993).

As regards seasonal variations showed that the high counts of bacterial indicators (TC, FC and FS) were detected in the warmer seasons (spring and summer), which might be attributed to high temperature and the discharged waste water during this season (Abu-Shady *et al.*, 1996 and Isobe *et al.*, 2004)

In accordance with (Chao *et al.*, 2003) significant positive correlation (r = 0.74) were recorded between the total number of bacteria and various indicator bacteria in the Nile water.

The ratio FC/FS points to the source of faeces whether it is human (>4) or animal (<0.7) (Geldreich, 1974), accordingly, the FC/FS ratio were in the range of 0.19-11, which means the mixed origin of faecal pollution. On the other hand, Niewolak (1998) reported that the data on absolute values of FC: FS may not reflect the source of contamination in Czarna Hancza River, Poland.

According to the guideline criteria for faecal indicator organisms of (WHO report, 1992) which accept the guide values of the investigated bacteria up to 500/100 ml for total coliforms and 100/100ml for both faecal coliforms and faecal streptococci. The survey of the indicator bacteria along the Damietta Branch waters revealed that the Nile waters at this branch is subjected to sewage pollution.

Seasons		Auti	umu			Win	lter			Spr	ing			Sum	mer	
Stations	°C °C	Trans.	Depth	Hq	v. ccp.	Trans.	Depth	Hq	°C °C	Trans.	Depth	Hq	W. temp. °C	Trans.	Depth	рН
-	18.9	70	8.0	8.30	17.0	60	7.5	8.16	24.3	150	8.0	8.15	27.7	150	8.0	8.14
7	18.8	95	6.0	8.05	17.9	120	8.0	7.87	25.1	60	8.0	7.89	Ð	ΩN	QN	QN
3	19	100	3.5	8.19	18.8	120	8.0	8.11	25.7	150	8.0	7.74	28.7	Q	4.0	7.97
4	23	110	4.0	7.65	19.8	220	3.0	7.58	29.0	70	6.0	7.49	29.1	QN	6.0	8.07
S	18.9	150	16.0	7.58	17.6	130	7.0	7.56	26.9	70	9.0	7.87	29.7	60	0.6	8.04
9	21	200	6.0	7.65	18.1	90	7.0	7.75	27.0	100	7.0	7.24	32.0	80	6.0	7.83
7	19.5	120	19.0	8.22	17.9	40	25.0	8.28	26.1	100	7.0	8.44	32.8	QN	25.0	8.37
ND = Not dete W. temp.= Wat	cted ter tempo	srature			Tra	ins.= Tra	nsparene	h.								

Table (1): Physicochemical characteristics of the River Nile waters at Damietta Branch.

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Seasons	Autu	mn	Wi	nter	Spi	ring	Sum	mer
Stations	22°C	37°C	22°C	37°C	22°C	37°C	22°C	37°C
1	25	28	17.5	8.8	30	20	35	31
2	28	27	25	27	34	27	40	38
3	26	28	19.5	21	39	31.6	80	78
4	30	46	10.8	14.6	44	37.2	84	85
5	43	47	20.7	13	43	41	88	90
6	43	50	19.2	13	64	34.8	108	110
7	46	58	18.7	14	76	77	150	152

Table (2): The total viable bacterial (TVBCs) x 10⁷ cfu/ml water of the River Nile at Damietta branch.

Table (3): The most probable number (MPN) of total coliforms (TC), faecal coliforms(FC) and faecal streptococci (FS) /100ml water of the River Nile.

Seasons		Autumn		,	Winter		<u>r</u>	Spring		s	ummer	
Stations	тс	FC	FS	тс	FC	FS	тс	FC	FS	тс	FC	FS
1	240	21	12	430	40	4	900	400	93	270	70	75
2	440	90	460	460	43	26	2400	930	210	1500	390	400
3	1900	400	150	1100	44	40	11000	1500	1500	7500	400	390
4	2000	700	240	2100	440	40	15000	2100	1600	21000	2000	440
5	9300	2100	240	1500	360	40	15000	2100	430	39000	2300	460
6	11000	700	460	4000	460	150	46000	2400	1500	110000	7000	1100
7	46000	2300	1100	16000	2300	390	110000	6400	1500	160000	7500	2100

In the present study, the isolates of genus 1 were differentiated and confirmed by API 20E as *E. coli* (Table, 4), *E. coli*, the main indicator of faecal pollution, constitute 16% of the identified gram-negative bacteria in the examined water. This also indicated that the water of Dameitta Branch is subjected to sewage pollution.

The genus Pseudomonas is made up of more than 300 species of gram-negative bacteria. Based on morphological and API 20E biochemical reactions (Table, 4) most members of genus 2 are identified as Pseudomonas aeruginosa which is an opportunistic pathogen of humans. In contrast to most enterobaceria, this pathogenic bacterium is the most significant example of bacteria capable of multiplying in water. Thus, P. aeruginosa is common in the tested water of Dameitta Branch of River Nile (12%). On the other hand, P. flourcsence represented 4% of the identified gram negative bacteria from the examined water.

At present, the genus Klebsiella is subdivided into 5 species (Podschun, et al., 2001). The isolates of genus 3 were identified as Klebsiella pneumoniae according to morphological and biochemical characteristics (Table, 4). K. pneumoniae represented 14% of the identified gramnegative bacteria isolated from Dameitta. This pathogenic bacterium has been previously isolated from surface water (McIntosh and Austin, 1990; Podschun et al., 2001). There are 2200 known species of Salmonella which cause intestinal infections such as sallmonellosis. The species of genus 4 was classified as Salmonella cholerasuis because of its biochemical characteristics (Table, 4) such pathogenic bacterium constituted 11% of gram-negative bacteria from the examined water. On the other hand, isolates of genus 5 were identified as Shigella sp and represented 9% of the identified gramnegative bacteria. This pathogenic bacterium is an invasive pathogen which causes shigellasis or Shigella-related diarrhea.

In the present study, isolates of genus 6 were identified as *Serratia liquefaciens* (Table, 4) and represented 8% of gramnegative bacteria isolated from the tested water samples. This bacterium is considered a pathogen of fish (McIntosh and Austin, 1990). On the other hand, genus 7 includes 8 isolates (8%) of gram-negative bacteria which were identified as *Proteus vulgaris* (Table, 4).

Acinetobacter is a member of the family Nisseriaceae that is normally found in soil, water and the moist areas of skin. Members of genus 8 (7 isolates) are oxidase-negative, non-motile, non-spore forming and nonfermentative coccobacillus which were identified as *Acinetobacter* sp. (Table, 4).

Genus (9), *Brenneria*, includes 5 isolates (5%) which were identified as *Brenneria nigrifluens* (Table, 4). This genus, saprophyte or pathogen, is associated with plants and previously isolated from surface water (Geller, 1986).

Three isolates of genus 10 was identified as Flavimonas oryzihabitans, which is gramnegative. vellow pigmented, oxidase negative, and non-fermenting bacillus (Table, 4). This organism has rarely been implicated as human and fish pathogen (Anzai et al., 1997). On the other hand, the three isolates of genus 11 were motile aerobic gram-negative rod with yellow organic pigment, oxidasenegative, non-ferments and identified as Chryseomonas luteola (Table, 4). This bacterium has rarely been reported as a human bacterial pathogen (Chihab et al., 2004). C. luteola can be distinguished from other yellow pigmented non-fermenters by negative-oxidase reaction and from enterobacteria by its strict aerobic growth. The normal habitats of this organism are unclear, but it is frequently found in water soil and other damp environments (Freney et al., 1988).

In conclusion, the water of the River Nile at Damietta Branch is subjected to faecal pollution and monitoring of microbial quality

of water is a must to control the spreading of

pathogens transmitted by contaminated water.

		Dacter	la is	olated	1101	n Da	amietta	a Br	ancn	01	River	mie.
	Genus 1	Ger	nus 2	Genus 3	Genus 4	Genus 5	Genus 6	Genus 7	Genus 8	Genus 9	Genus 10	Genus 11
ONPG	+	-	-	+	-	-	+	-	-	-	-	+
ADH	-	+	+	-	-	-	-	-	-	-	-	+
LDC	-	-	-	-	+	+	-	-	-	-	-	-
ODC	-	-	-	-	+	+	-	-	-	-	-	-
CIT	-	+	+	+	-	-	+	-	-	-	-	+
H2S	-	-	-	-	-	-	-	+	-	-	+	-
URE	-	-	-	+	-	-	-	+	-	-	-	-
TDA	-	-	-	-	-	-	-	+	-	-	-	-
IND	+	-	-	-	-	-	-	+	+	-	- ,	-
VP	-	-	-	+	-	-	+	-	-	+	-	-
GEL	-	-	-	-	-	-	-	-	+	-	-	-
GLU	+	+	-	+	+	+	+	+	+	+	-	+
MAN	+	-	-	+	+	-	+	-	-	+	-	-
INO	-	-	-	+	-	-	+	-	-	-	-	+
SOR	+	-	-	+	+	-	+	+	-	+	-	-
RHA	+	-	-	+	+	-	-	-	-	+	-	-
SAC	-	-	-	+	-	-	+	+	-	+	-	+
MEL	+	-	-	+	-	-	-	-	-	+	-	-
AMY	-	-	-	+	-	-	+	-	-	+	-	+
ARA	+	-	-	+	-	-	-	-	-	+	-	+
ox	-	+	-	-	-	-	-	-	-	-	-	+
NO2	+	-	-	+	+	+	+	+	+	+	•	•
N2	-	-	-	-	-	-	-	-	-	-	-	-
мов	+	+	+	-	+	-	+	+	+	+	+	+
McC	+	+	+	+	+	+	+	+	+	+	+	+
OF-O	+	+	+	+	+	+	+	+	+	+	+	-
OF-F	+	-	-	+	+	+	+	+	-	-	-	-

Table	(4):	API	20E	biochemica	al cha	aracteristics	pathogenic	of	Gram-ne	gative
		bact	eria	isolated	from	Damietta	Branch	of	River	Nile.

Where they confirmed as:

Genus 1: E. coli, Genus 2: P. aeruginosa, P. flourcsence, Genus 3: K. pneumonia, Genus 4: Salmonella cloerasuis, Genus 5: Shigella. sp., Genus 6: Serratia liquefaciens, Genus 7: Proteus vulgaris, Genus 8: Acinetobacter sp., Genus 9: Brenneria nitrifluens, Genus 10: Flavimonas oryzihabitans and Genus 11: Chryseomonas luteola.

REFERENCES

- Abdo, M.H.: 2004 a, Distribution of some chemical elements in the recent sediments of Damietta branch, River Nile, Egypt. *Journal of Egyptian Academic Society for Environmental Development* (D-Environmental Studies) 5(2):125-146.
- Abdo, M.H.: 2004 b, Environmental studies on the River Nile at Damietta Branch region, Egypt. Journal of Egyptian Academic Society for Environmental Development (D-Environmental studies) 5(2): 85-104.
- Abu-Shady, M.R.; El-Moatassem, M.; Heikal, M.T. and Khalafalla. G.M.: 1996, Microbiological quality of the River Nile stretch flows through Cairo. The Second International Conference on Potable Water Management and Water Treatment Technologies, Cairo Aqua-Tech, 96.
- American Public health Association (APHA): 1995, Standard methods for the examination of water and waste water, 19th ed. American Public health Association, Washington, D.C.
- Anzai, Y.; Kudo, Y. and Oyaizu, H.: 1997, The phylogeny of the genera *Chryseomonas, Flavimonas* and *Pseudomonas* supports synonymy of these three genera. *International Journal* of Systematic Bacteriology **25**: 249-251.
- Baron, S.: 1996, *Medical Microbiology*, 4th ed. The University of Texas Medical Branch at Galveston.
- Chao, K.K.; Chao, C.C. and Chao, W.L.: 2003, Suitability of traditional microbial indicators and their enumerating methods in the assessment of fecal pollution of subtropical fresh water environments. *Journal Microbiology Infection* **36** (4): 288-293.
- Chihab, W.; Alaoui, A.S. and Amar, M.:2004, Chryseomonas luteola as the source of serious infections in Moroccan university hospitals. Journal of Clinical Microbiology 42(4): 1837-1839.

- Elewa, A.A. and Goher, M.E.M.: 1999, Environmental factors affecting the precipitation and dissolution of Fe, Mn, Zn, Cu, Pb and Cd in River Nile at Damietta Branch. *Bulletin Faculty of Science, Zagazig University* **21**(2): 114-136.
- El-Fadaly, H.; El-Defrawy, M.; El-Zawawy, F. and Makia, D.: 2001, Chemical and microbiological evaluation of River Nile water in Dakahlia Governorate. *Journal of Environmental Science* **22**: 1-18.
- El-Mongy, M.E.: 1978, Studies on bacterial indices of pollution in Mansoura water resources. M.Sc. Thesis, Fac. Sci. Mansoura University, Egypt.
- Fleisher, J.M.; Kay, D.; Wyer, D. and Godfree, A.F. : 1998, Estimates of the severity of illness associated with bathing in marine recreational waters contaminated with domestic sewage. *International Journal of Epidemiology* 27: 722-726.
- Freney, J.; Hansen, W.; Etienne, J.; Vandenech, F. and Fleurette, J.: 1988, Post operative infant septicemia caused by *Pseudomonas luteola* (CDC group ve-1) and *Pseudomonas oryzihabitants* (CDC group,ve-2). *Journal of Clinical Microbiology* **26**: 1241-1243.
- Geldreich, E.E.: 1974, Buffalo Lake recreational water quality: a study on bacteriological data interpretation. Water Research 6: 913-921.
- Geller, A.: 1986, Comparison of mechanisms enhancing biodegradability of refractory lake water constituents. *Limnology and Oceanography* **31**(4): 755-764.
- Giannoulis, N.; Maipa, V.; Konstantinou, I.; Albanis, T. and Dimoliatis, I.: 2005, Microbiological risk assessment of Agios Georgios source supplies in north western Greece based on faecal coliform determination and sanitary inspection survey. *Chemosphere* **58**: 1269-1276.
- Haeikal, M.T.: 1994, Studies on bio-indicator for water pollution in the River Nile.

M.Sc. Thesis, Institute of Environment Studies and Researches, Ain Shams Univ., Egypt.

- Hausler, W. J. and Koontz, F. P.: 1970, Brucellusis. In: *Diagnostic procedures* for bacterial, mycotic and parasitic infections, 5th ed., APHA, New York.
- Isobe, K.O.; Tarao, M; Chiem, N.H.; Minh, Le. Y.; Takada, H.: 2004, Effect of environmental factors on the relationship between fecal indicator bacteria in tropical (Mekong Delta) and temperature (Tokyo) freshwaters. *Applied and Environmental Microbiology* **79**(2): 814-821.
- Karaboze, I.; Ucar, F.; Eltem, R.; Ozdmir, G. and Ates, M.: 2003, Determination of existence and count of pathogenic microorganisms in Izmir Bay. JES 26:1-18.
- McIntosh, D. and Austin, B.: 1990, Recovery of an extremely proteolytic from *Serratia liquefaciens* as pathogen of Atlantic salmon, *Salmon salar*, in Scotland. *Journal of Fish Biology* **36**: 765-772.
- Niewolak, S.: 1998, Total viable count and concentration of enterio bacteria in bottom sediments fromt eh Czarna hancza River, Northeast Polland. *Polish Journal of Environmental Studies*, **7**(5): 295-306.
- Podschun, P.; Pietsch, S.; Holler, C. and Ulmann, U.: 2001, Incidence of *Klebsiella* species in surface waters and their expression of virulence factors. *Applied and Environmental Microbiology* **67**(7): 3325-3327.
- Rabeh, S.A.: 2000, Thermal and microbial pollution in the River Nile at industrial region of Shoubra El-Kheima, Egypt. Egyptian Academy Society for Environmental Development 1(1): 83-98.

- Sabae, S. Z.: 1999, Bacterial pollution of River Nile waters (Rosstta Branch). Egyptian Journal of Aquatic Biology and Fisheries 3(2): 21-34.
- Sabae, S. Z.: 2004, Monitoring of microbial pollution in the River Nile and the impact of some human activities on its waters. Proceeding 3rd International Conference on Biological Science. Tanta University. 28-29, April, Vol.**3**: 200-214.
- Sabae, S.Z.; Hazaa, M.M.; Aballah, S.A.; Awny, N. and Dabbor, S.M.: 2006, Studies on bacterial indicators of water pollution and bioremediater isolates for Cu²⁺, Fe²⁺ and Zn²⁺ in Rosetta Brach River Nile, Egypt. *Egyptian Journal of Biotechnology* 22: 77-104.
- Shaaban-Dessouki, S.A.; Soliman, A.I. and Deyab, M.A.: 1993, Environmental characteristics and nutrients distribution in Demietta estuary of the River Nile. *Journal of Environmental Science* 6:159-177.
- WHO (World Health Organization): 1992, *Our plants, our health.* Geneva, 133pp.
- WHO (World Health Organization): 1993, *Guideline of Drinking water quality*, 2nd Edition, Vol.1, World Health Organization, Geneva.
- Youn-Joo, A.; Kampbell, D.H. and Breidenbach, G. P.: 2002, *Escherchia coli* and total coliforms in water and sediment at Lake Marinas. *Journal of Environmental Pollution* **120**(3): 771-778.
- Zyadah, M.: 1996. Occurrence of heavy metals in some fish sediment and water samples from River Nile within Damietta Governorate. Proceeding 6th International Conference Environment Protection Is A must, Alex, Egypt: 929-942.