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The Distribution of Certain Heavy Metals Between Intestinal Parasites and their Fish Hosts in the River Nile at Assuit Province, Egypt.

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Abstract

The present investigation was carried out on two species of freshwater fishes ***Oreochromis niloticus niloticus*** and ***Clarias gariepinus*** collected from EL-Ibrahimia and EL-Malah canals at Assuit province. Water samples were also collected for determination of some water quality parameters and the concentration of certain heavy metals including Cu, Cd and Pb. The results of water quality analysis indicated significant differences between the two selected sites. The examination of the intestinal parasites in both investigated fish species indicated that *O. niloticus niloticus* harbors one species of acanthocephalans, ***Acanthosentis tilapiae*** at the two investigated sites while, ***Clarias gariepinus*** was found harboring one species of nematodes, ***Paracamallanus cyathopharynx*** at site(1) and one species of trematodes, ***Orientocreadium lazeri*** at the two sites. The results also indicated that the mean concentration of the three heavy metals in water at site (1) are ranked as Cu > Cd > Pb, while at site (2) they are ranked as Cu > Pb > Cd with a highly significant difference ($P<0.01$) between the two investigated sites. The data also showed a differences between the fishes and their parasites from heavy metal ranking point of view, where it is ranked as Cu > Pb > Cd in both infected and uninfected fishes. In case of parasites heavy metals concentration at site (1) the investigated heavy metals are ranked as Pb > Cu > Cd in both ***Acanthosentis tilapiae*** and *Orientocreadium lazeri*, while they are ranked as Pb > Cd > Cu at site (2) in ***Acanthosentis tilapiae*** and Pb > Cu > Cd in ***Orientocreadium lazeri***. Also, a significantly higher concentration of heavy metals was recorded in the parasites compared to their host fishes.

Key words: Heavy metals, Water quality, *Oreochromis*, *Clarias*, bioaccumulation, helminth parasites.

Introduction

Heavy metal pollution in the aquatic ecosystem has attracted serious concern during the recent years. The development of human activities; the increase of industrialization and the discharge of wastes to the environment might be the main sources of contamination especially in developing countries that might threatened biolife **(McGlashan & Hughies, 2001; Bishop, 2002; ECDG, 2002; Santos et al., 2005 and Saeed & Shaker, 2008).**
The River Nile is the principal freshwater resource for Egypt, meeting nearly all demands for drinking water, irrigation, and industry **(Mohamed *et al*., 1998)**. It receives many pollutants, including heavy metals with levels, in water and sediments of some parts of the River Nile are higher than the tolerance levels or limits set by the Egyptian General Authority for standards and Quality control **(Anwar, 2003).** Modern industrial and agricultural activities have introduced several polluting substances such as organic matter, chemical fertilizer, insecticides, etc. into the River Nile and drainage systems at Assiut Province. Several methods were used for detecting and assessment of pollutants in water environments, from these, the use of fauna inhabiting these habitats.
In the recent years, there has been increasing interest in the interrelationship between parasitism and pollution, especially in aquatic habitat and the role of parasites as bioindicator of heavy metals pollution **(Huspeni & Lafferty, 2004; Sures, 2006 and Vidal-Martinez, 2007).** This relationship is not simple and in essence involves a double edged phenomenon, in which parasitization may increase host susceptibility to toxic pollutants or in which pollutants may result in increase (or in some decrease) in the prevalence of certain parasites.

Kakacheva–Avramova (1975) studied the influence of pollution on the occurrence of helminthiasis in the Bulgarian section of River Danube. He noticed that the pollution has reduced the number of intermediate hosts of digeneans, cestodes, acanthocephalans and nematodes resulting in decrease of the helminth fauna in the river.

Poulin (1992) reported that the toxic substances can negatively affect the parasites, either directly, by harming the free- living stages, or indirectly, by reducing the invertebrate intermediate hosts. **Riggs *et al*. (1987)** reported that, if the intermediate hosts (Crustacea and snails) of the Acanthocephala and Digenea suffered more from pollution than did their fish host, many parasites could eventually disappear from fish population in the polluted areas.

Khan and Thulin (1993) mentioned that the chronic fish exposure to pollutants over a period of time causes biochemical, physiological and behavioral changes of the host that ultimately can influence the prevalence and intensity of parasitism by impairing the host's immune response or favouring the survival and reproduction of the intermediated hosts.

Invertebrate animals especially (helminth parasites) can accumulate heavy metals in their tissues with concentration in many cases higher than surrounding water **(Eisler, 1981; Rainbow, 1990; Phillips and Rainbow, 1993, AbdAllah and Moustfa, 2002, Sures, 2003, AbdAllah, 2004, Thielen, et al., 2004 and AbdAllah, 2006).**

Sures *et al.* (1994) studied lead accumulation in the adult acanthocephalan, *Pomphorhynchus laevis* isolated from the intestine of chub, *Leuciscus cephaleus*. They found that the mean level of lead in parasite samples were 284 times more than that in the host intestine, 771 times more than that in host liver and 2700 times more than that in host muscles.

Sures and Taraschewski (1995) investigated the cadmium concentration in the acanthocephalan, *Acanthocephalus lucii* parasitizing the intestine of perch, *Perca fluviatilis* and found that cadmium was significantly higher in isolated acanthocephalan parasite than in any organ (muscles, intestine and liver) of the infected perch. Moreover**, Sures *et al.* (1999)** reported that the potential value of fish parasites in monitoring metal contamination is more convenient than those of mammalian parasites.

In the present study, two common species of freshwater fishes; ***Oreochromis niloticus niloticus*** and ***Clarias gariepinus*** from two selected sites in the River Nile at Assiut Province, were investigated for identifying the infecting helminth parasites and determining their prevalence, intensity and abundance. Also, the present study aims to record the major physical and chemical water quality parameters, heavy metals concentrations in water and to calculate the bioaccumulation factors (BAF) of heavy metals for helminth parasites in the two investigated areas so as to examine their efficiency as sentinel organisms for heavy metal pollution.

Materials and methods

Study areas:

Site 1 (El-Ibrahimia canal);

El-Ibrahimia canal is graduated from the Nile to the Assiut barrage and ends at the Ashimnt status of Wasta, Beni Suef governorate. The samples were collected from the beginning of canal. The selected site is away from the industrial effluents and domestic sewage disposal and the position is 27^0 **11**' 18" N and 31^0 **11**' 21" E (Fig.1).

Site 2 (El-Malah canal):

EL-Malah canal is located at about 5 KM North Western from the Assiut city. It is exposed to domestic sewage disposal and industrial effluent **(Gabr *et al*., 2008)** and its position is 27^0 **10'** 19" N and 310 **0**9' 44" E (Fig.1)

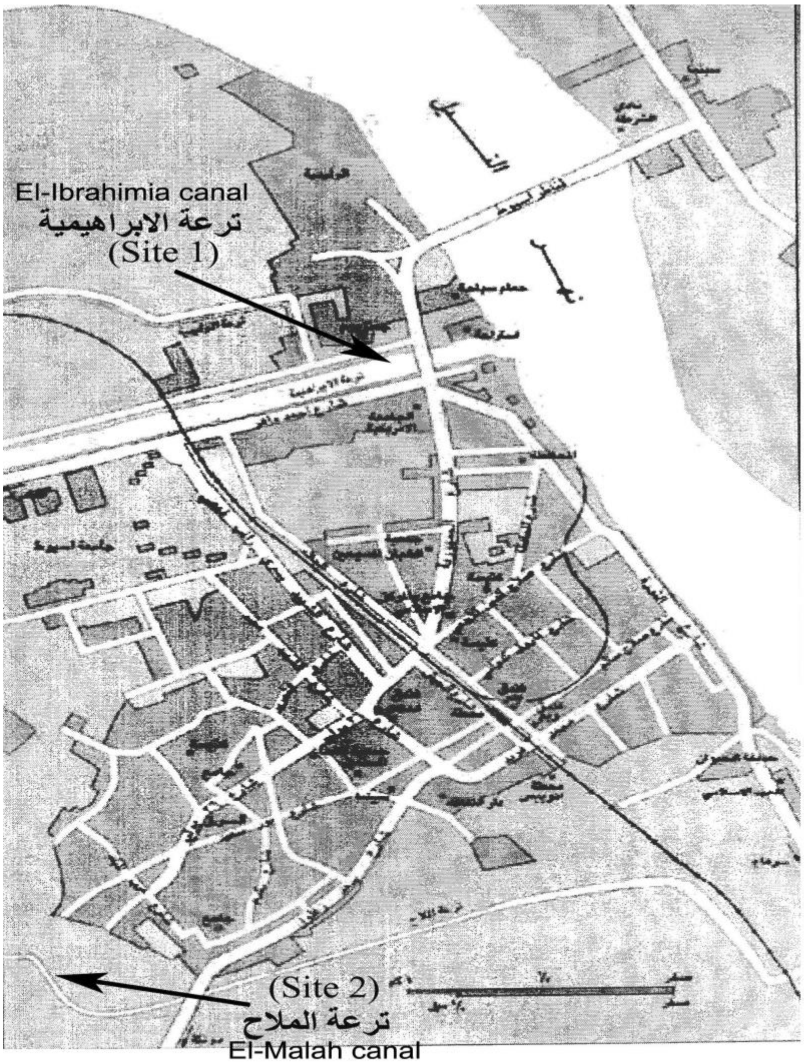

Fig. (1): A map of Assiut governorate showing the two investigated sites from the River Nile.The first site; El-Ibrahimia canal and the second site; El-Malah canal.

<u>Samples collection:</u>
1025 fish of *Oreochromis niloticus niloticus* and *Clarias gariepinus* were collected during the period from November 2005 to December 2006. The Specimens were collected as follow: 181 of *Oreochromis niloticus niloticus* and 293 of *Clarias gariepinus* from EL-Ibrahimia canal (site1) and 251 of *Oreochromis niloticus niloticus* and 300 of *Clarias gariepinu*s from EL-Malah canal (site2). Specimens were transported alive to the laboratory for examination. Six water samples, from each site, were collected and transported to the laboratory for water analysis.

<u>Parasites survey and identification:</u>
Fish were dissected in the laboratory. Gills, intestine, liver and kidney were removed, placed in Petri dish, opened in 0.7 physiological saline solutions and examined for helminth parasites. The collected worms were washed several times in saline solution to be freed from the mucous or any other debris and examined alive using a binocular dissecting microscope. All co-specific helminth species were counted and recorded for each fish.

The collected parasites were identified after **El-Naffar and Saoud, 1974; ; Imam, 1971; Wannas, 1977; Sahlab, 1982; Negm El- Din, 1987;Abu El-Ezz, 1988; Khattab, 1990; Imam and El-Askalany, 1990; El-Gehaeny,1995; Abd El-Monem, 1998; Mohammed, 2002 and Thabit, 2004 .**

Prevalence, abundance and mean intensity of infection were calculated according to **Margolis *et al.* (1982).**

$$\textbf{Parasite prevalence} = \frac{\text{Number of infected fish}}{\text{Total number of examined fish}} \times 100$$

$$\textbf{Parasite abundance} = \frac{\text{Number of parasites}}{\text{Total number of examined fish}}$$

$$\textbf{Mean intensity} = \frac{\text{Number of parasites}}{\text{Total number of infected host}}$$

Measuring of physicochemical parameters:

The collected water samples from the two investigated sites were analyzed for major physical and chemical water quality parameters such as; water temperatures, pH**,** electrical conductivity (EC), dissolved oxygen and salinity. These parameters were measured by using water checker U-10 Horiba Ltd. Another parameters like; total alkalinity, nitrite, ammonia and total hardness were measured by traditional manual methods according to **(USEPA , 1983 and Eaton *et al*., 1995).**

Determination of heavy metals:

The collected water samples from the two investigated sites were analyzed to determine the concentration of heavy metals (Cu, Cd and Pb) as process of **Clescerl *et al.*(1999).** The fish tissue (intestine) and intestinal parasites samples were dried as the methods of **Sures *et al.* (1995b)** and analyzed for heavy metals by using a Perkin–Elmer Analyst 100 Atomic Absorption Spectrophotometer (**Kruse, 1980, AbdAllah and Moustafa, 2002 and AbdAllah, 2006**).

Statistical analysis:

SPSS 13 for Windows software was used for the statistical analysis of the three heavy metal contents in fish intestine, their helminths and water samples at the two investigated sites by using multivariate ANOVA (95% significance level).

The Bioaccumulation factors (BAF) were calculated according to **Neuhauser *et al* (1995) and AbdAllah and Moustafa (2002)** using the following formula:

$$\textbf{BAF} = \frac{\text{Metal concentration in fish tissue (mg/kg)}}{\text{Metal concentration in water (mg/l)}}$$

Results

Mean ± SD of water quality parameters at the two investigated sites were tabulated in Table (1) and the Permissible level was recommended by Egyptian Organization for Standardization (1993). Statistical analysis of variance (one way ANOVA) has revealed highly significant differences in all parameters at the two sites during the period of investigation ($P<0.01$), except temperature where it was significant ($P<0.05$).

Table (1): Water quality parameters measured (Means ± SD) at the two sites:

Water Quality Parameter	Permissible limit	Localities	
		Site 1	Site 2
Temperature (c°)	Over 5 c°	22.33 ± 0.58	25.0 ±1.00
pH	7-8.5	7.23 ± 0.25	8.0± 0.10
Conductivity (ms/cm)	-	0.34 ± 0.01	0.39 ± 0.01
Salinity (g/l)	-	0.02 ± 0.00	0.033 ± 0.002
dissolved oxygen (mg/l)	Not less than 5	8.01 ± 0.01	5.8 ± 0.66
Nitrite (mg/l)	45	0.002 ± 0.001	0.037 ± 0.001
Ammonia (mg/l)	0.5	0.02 ± 0.01	3.23 ± 0.21
Total alkalinity (mg/l)	20 – 150	123.00 ± 1.00	176.00 ± 2.00
Total hardness (mg/l)	-	143.00 ± 1.0	206.33 ± 1.53

Table (2) showing the helminthes fauna of the studied fishes at the two sites, ***Oreochromis niloticus niloticus*** was found harboring one species of acanthocephalans**,** ***Acanthosentis tilapiae*** recovered from the intestine at the two investigated sites and one species of trematodes, ***Clinostomium phalacrocoracis*** **was** recovered from the gills at site(1).

Clarias gariepinus **was** found harboring one species of nematodes, ***Paracamallanus cyathopharynx*** recovered from the intestine at site (1) and one species of trematodes, ***Orientocreadium lazeri*** recovered from the intestine at the two sites. One way ANOVA showing highly significance (P < 0.01) of *Acanthosentis* ***tilapiae*** and **Clinostomum** ***phalacrocoracis*** in Oreochromis ***niloticus niloticus*** at the two sites. In ***Clarias gariepinus*,** the ***Paracamallanus cyathopharynx*** was highly significant (P < 0.01) while, ***Orientocreadium lazeri*** was significant (P< 0.05) at the two investigated sites.

Mean ± SD of heavy metals concentrations in water at the two sites were tabulated in Table (3).The mean concentration of the three metals in site (1) are ranked as Cu > Cd > Pb, while in site (2) the three metals are ranked as Cu > Pb > Cd. Applying one way ANOVA, showed highly significant difference (p<0.01) between the two investigated sites for all heavy metals concentration.

The mean concentrations of the three heavy metals in the tissues of ***Oreochromis niloticus niloticus*** and ***Clarias gariepinus*** and their helminthes parasites in the two investigated sites are ranked as Cu > Pb > Cd in both infected and uninfected fishes (Table 4). The mean concentrations of heavy metals in ***Acanthosentis tilapiae*** (Fig. 2) and ***Orientocreadium lazeri*** (Fig. 3) are ranked as Pb > Cu > Cd in site (1), while in site (2) they are ranked as Pb > Cd > Cu in ***Acanthosentis tilapiae*** and Pb > Cu > Cd in ***Orientocreadium lazeri*** (Table 4 and Fig. 2).

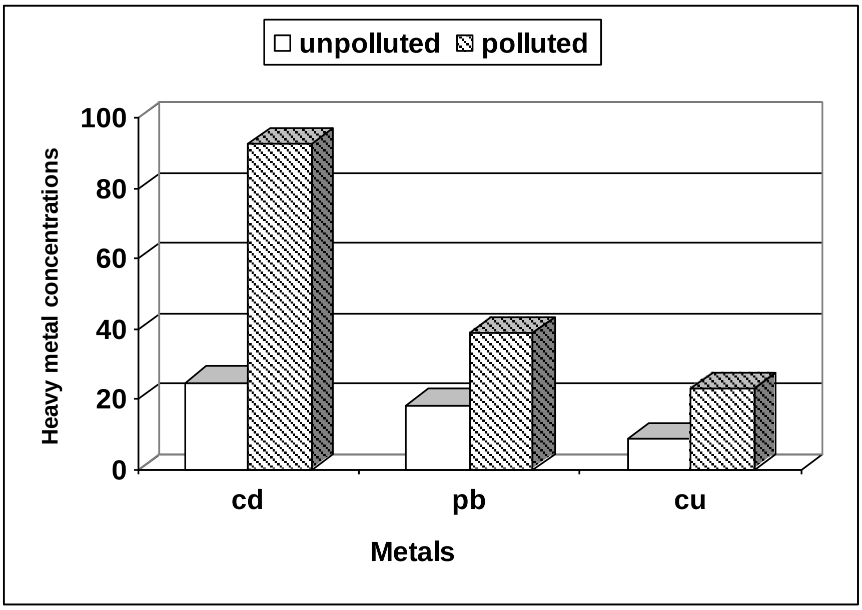

Fig (2) Cadmium, lead and copper in the acanthocephalan Acanthosentis tilapiae in Ibrahimia canal (unpolluted) and ElMalah canal (polluted).

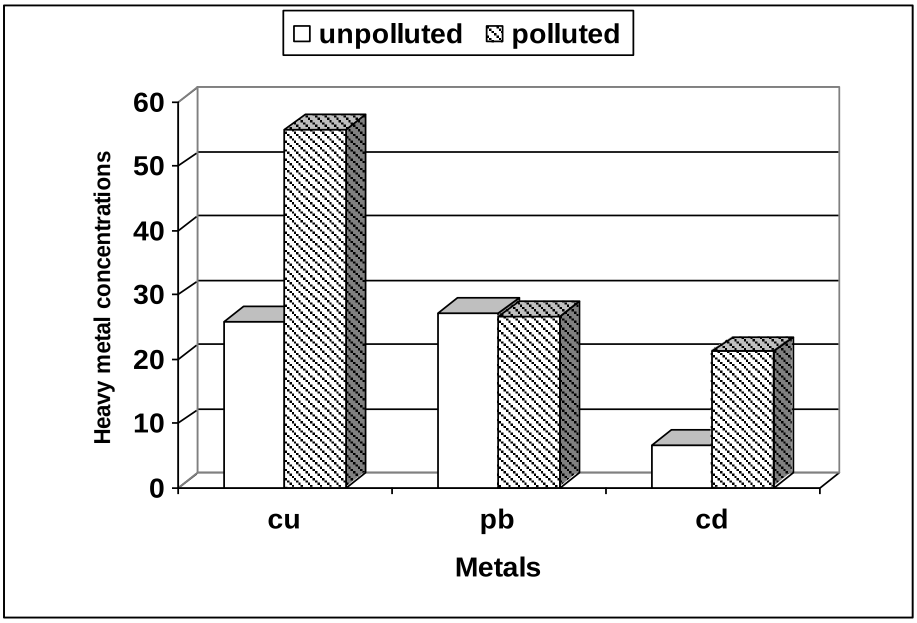

Fig (3) Heavy metal concentration in the trematode Orientocreadium lazeri in ElMalah canal (polluted) and Ibrahimia canal (unpolluted).

Table (2): Composition of parasite species and their prevalence, abundance and intensity at the two sites.

Localities	Host of fish	Species of parasite	No. of examined fish	No. of infected fish	Total no. of parasite	Prevalence	Intensity	Abundance
EL-Ibrahimia canal (unpolluted area)	*O. niloticus*	***Acanthosentis tilapiae***	181	96	1864	53.04	19.4 ± 0.55	10.30
		Clinostomium phalaacrocorsis	181	31	241	17.13	7.8 ± 0.45	1.33
	C. gariepinus	***Orientocreadium lazeri***	293	151	2657	51.54	17.60 ± 0.55	9.07
		Paracamallanus cyathopharynx	293	68	216	22.21	9.00 ± 000	2.09
EL-Malaha canal (Polluted area)	*O. niloticus*	***Acanthosentis tilapiae***	251	143	3185	56.79	22.27 ± 0..45	12.70
		Clinostomium phalaacrocorsis	251	0	0	0	00.00 ± 0.00	00.00
	C. gariepinus	***Orientocreadium lazeri***	300	140	2352	46.67	16.8 ± 0.45	07.48
		Paracamallanus cyathopharynx	300	0	0	0	00.00 ± 0.00	00.00

Table (3): Mean ±SD of heavy metals concentrations (µg / l) in water at the two investigated sites

Metal	Site (1)	Site (2)	Permissible limit In water
	Mean ±SD	Mean ±SD	
Cu	0.40±0.02	0.70±0.04	1.00
Cd	0.04±0.002	0.07±0.004	0.01
Pb	0.024±0.001	0.08±0.004	0.05

One way ANOVA of the two investigated sites showing significance ($p < 0.05$) of all means of heavy metals concentrations in infected ***Oreochromis niloticus niloticus,*** but in the uninfected ones, it was highly significant ($P < 0.01$) in case of copper and lead and non significant in case of cadmium (Table 4). In ***Clarias gariepinus***, one way ANOVA of the two sites showing significance ($P < 0.05$) of cadmium and lead mean concentrations and non significance of copper in the infected fishes, while in the uninfected ones, all means of heavy metals concentrations were non significant except in the case of lead, it was significant ($P<0.05$).

Table (4): Mean ±SD of Cu, Cd and Pb concentrations in the hosts intestines and intestinal parasites (µg / g) of the infected and uninfected fishes at the two sites.

			Intestinal parasites		Fish intestine	
			A. tilapiae in *O. niloticus*	O. *lazeri* in *C. gariepinus*	*Oreochromis niloticus niloticus*	*Clarias gariepinus*
Cu	**Site 1**	**uninfected fish**	-	-	3.41 ± 0.75	4.86 ±1.06
		Infected fish	32.08 ± 6.85	25.95± 1.22	3.02 ± 0.66	4.01 ± 0.88
	Site 2	**uninfected fish**	-	-	13.98 ± 3.06	6.55 ± 1.43
		Infected fish	92.60 ± 1.89	55.69 ± 2.62	7.57 ± 1.98	4.90 ± 1.07
Cd	**Site 1**	**uninfected fish**	--	--	0.584 ± 0.13	0.987± 0.39
		Infected fish	28.82 ± 0.41	6.61± 0.31	0.402 ± 0.088	0.457± 0.099
	Site 2	**uninfected fish**	-	--	0.867 ± 0.23	1.202± 0.26
		Infected fish	188.37± 17.80	21.21 ± 0.99	0.862± 0.22	1.102 ± 0.29
Pb	**Site 1**	**uninfected fish**	--	--	1.26 ± 0.33	2.03 ± 0.52
		Infected fish	118.38 ± 0.86	27.25 ± 1.28	0.76 ± 0.17	1.38 ± 0.30
	Site 2	**uninfected fish**	--	--	5.64 ± 1.23	3.56 ± 0.79
		Infected fish	253.44 ± 15.23	158.38 ±23.70	1.71± 0.37	2.75 ± 0.60

Table (5): Mean values of bioaccumulation factor for different heavy metals in the tissues of infected and uninfected *Clarias gariepinu*s and *Oreochromis niloticus niloticus* at the two sites during the period of study.

Fish species		Heavy metals	Site (1)	Site (2)	Mean ±SD
Oreochromis niloticus niloticus	**Infected**	**Cu**	7.15 ± 1.57	10.67 ± 2.79	8.91 ± 2.79
		Cd	10.08 ± 2.24	12.31 ± 3.17	11.20 ± 2.74
		Pb	31.81 ± 6.99	21.30 ± 4.66	26.56 ± 7.83
	Uninfected	**Cu**	8.06 ± 1.77	19.69 ± 4.31	13.88 ± 6.99
		Cd	14.67 ± 3.19	12.39 ± 3.24	13.53 ± 3.13
		Pb	52.64 ± 3.78	70.56 ± 15.24	61.60 ± 16.33
Clarias gariepinus	**Infected**	**Cu**	9.48± 2.07	6.90 ± 1.51	8.19 ± 2.16
		Cd	11.42 ± 2.53	15.80 ± 4.29	13.61± 3.96
		Pb	59.86 ±15.37	34.31 ± 7.52	47.09 ± 15.65
	Uninfected	**Cu**	11.48 ± 2.52	9.22 ± 2.02	10.35 ± 2.41
		Cd	24.75 ± 9.78	17.17 ± 3.76	20.96 ± 7.81
		Pb	84.58 ± 21.70	44.81 ± 9.82	64.70 ± 26.49

Three factors ANOVA showing highly significant ($P<0.01$) effect of metal types and sites, separately, on the concentration of heavy metals, while fish species showing non significant effect. Two factors interaction revealed highly significance ($P<0.01$) in the case of (heavy metal*site), significance ($P<0.05$) in the case of (Fish*heavy metal) and non significance in the case of (Fish*site) while, three factors interaction (fish*site*heavy metal) showing highly significance ($P<0.01$) on the concentration of heavy metals in the infected fishes at the two sites**.** In the Parasite tissue (***Acanthosentis tilapiae*** and **Orientocreadium lazeri**), one way ANOVA showing highly significance (P<0.01) for all heavy metals concentration at the two investigated sites (Table 6).

Table (6): Mean values of bioaccumulation factor for different heavy metals in the tissues of *Clarias gariepinus* and *Oreochromis niloticus niloticus* parasites at the two sites.

Parasite species	Heavy metals	Site (1)	Site (2)	Mean ±SD
Acanthosentis tilapiae	**Cu**	4932.64 ± 36.00	3167.96 ± 190.30	4050.30 ± 974.28
	Cd	720.500 ± 10.45	2690.95 ± 254.28	1705.73 ± 091.20
	Pb	76.38 ± 16.29	130.42 ± 2.66	103.40 ± 31.39
Orientocreadium lazeri	**Cu**	1135.42 ± 53.25	1979.71 ± 296.25	1557.57 ± 500.09
	Cd	165.25 ± 7.75	303.00 ± 14.23	234.13 ± 76.14
	Pb	61.49 ± 2.65	78.44 ± 3.68	69.97± 9.71

Discussion

a. Water quality parameters:

This study shed light on some of the environmental factors that might have an impact directly or indirectly on the concentration ability of heavy metals into the bodies of the investigated hosts and parasites.

For instance, the mean value of dissolved oxygen at the two investigated sites showed highly significant (P<0.01) decrease toward the site 2 (polluted area). This decrease may be referred to the consumption of oxygen in decomposing the organic matter and the oxidation of chemical effluents. This interpretation agrees with the finding of **Haggag *et al.* (1999); Abd El-Monem (2001) and Ait Alla *et al*. (2006**).

The highly significant (P<0.01) difference between the salinity at the two sites may be attributed to two important factors; the high levels of dissolved salts in agricultural drainage water which discharged directly into site 2 and this interpretation is parallel with the finding of **(Ueda *et al*., 2000 and** Emara **& Belal, 2004),** and the extensive evaporation of the water from such closed ecosystem increases concentration of heavy metals, pesticides and other pollutants (**Ali, *et al.* 2008 and Mohamed, 2009**).

There is also a highly significant (P<0.01) difference between the two sites, regarding pH value and nitrite - amonia contents. The slight increase in pH values of site 2 may be attributed to a number of factors such as industrial effluents, agricultural drainage and waste municipal and/or the changes in the total hardness and total alkalinity that involves the uptake of free carbon dioxide from water and precipitation of calcium carbonate. This opinion is in accordance with **(Boyed, 1990; Saeed, 1999 and Abdo, 2005).**

The increase in Nitrite content at site 2 may be due to the increase in microbial activity and nitrification process. While, the increase in Ammonia may be pointed to the industrial effluents and agricultural drainage water which discharged directly into site 2 and this agrees with the finding of **(Svobodova *et al*., 1993 and Anzfcc& Armcanz, 2000).**

The water temperature at site (1) was (22.33 ± 0.58 C^o), while at site (2) was (25 ±1 C^o) and this difference is significant at (p<0.05). The variation in water temperature depends mainly on the climatic conditions, sampling times, the number of sunshine hours and also affected by specific characteristics of water environment such as turbidity, wind force, plant cover and humidity**.** This view is parallel with the finding reported by **Mahmoud (2002) and Tayel (2002)**.

The recorded conductivity at site (1) was (0.34 ±0.01 ms /cm) while, at site (2) was (0.39 ±0.01 ms /cm). The difference between the two sites is highly significant (P<0.01) and the present authors accept the interpretation of Abdo **(2005)** and **Abdo *et al.* (2010)**, who mentioned that the increase in EC values is related to the increase in total dissolved solids and water temperature.

The total alkalinity at site (1) was (123±1 Mg/l) while, at site (2) was (176± 2 Mg/l). The difference between the two sites is highly significant ($P<0.01$) and the increase in total alkalinity at site 2 may be attributed to the increase in bicarbonate concentration which produced from the decomposition of organic matter by bacteria, where the HCO3 is the final product of this decomposition. This finding is in agreement with **(Abdo, 2002).**

Also, there is highly significant difference ($P<0.01$) were shown in the total hardness at the two investigated sites, this increase difference at site 2 may be produced as a result of the interaction between domestic sewage disposal and/or industrial effluent, which increases the dissolved divalent metallic ions (calcium and magnesium) in water.

b. Parasitic fauna of the investigated fishes at the two sites:

The examination of ***Clarias gariepinus*** and ***Oreochromis niloticus niloticus*** revealed that they harboured many species of helminths. Comparing the data and characters of these helminths with those of **Amin (1975a&b); El-Naffar *et al.* (1983); Abu El-Ezz (1988); Ebraheem (1992); El-Ganiny (1995); Bayoumy (1996); Abd El-Monem (1998) and Thabit (2004),** revealed that ***Oreochromis niloticus niloticus*** harboured the acanthocephalan, ***Acanthosentis tilapiae*** in the alimentary canal and the digenetic trematode metacercaria, ***Clinostomium phalacrocoracis*** in the gills. While, ***Clarias gariepinus*** harboured the digenetic trematode, ***Orientocreadium lazeri*** and the nematode, ***paracamallanus cyathopharynx*** in the intestine.

The present work showed that the prevalence, intensity and abundance of infection with ***Orientocreadium lazeri*** and ***paracamallanus cyathopharynx*** in ***Clarias gariepinus*** was higher in site 1 than site 2. The same was reported in case of ***Clinostomium phalacrocoracis*** and ***Oreochromis niloticus niloticus*** (**Table 2**). This result is parallel with **Sinderman (1990); Kuperman (1992) and Dusek *et al.* (1998).** The mentioned authors above attributed the decrease in the infection rate of fishes lived in highly polluted areas to the fact that, effluents including heavy metals could alter the availability or reducing the number of invertebrate intermediate hosts necessary for life cycle of these parasites.

On other hand, the prevalence, intensity and abundance of ***Acanthosentis tilapiae*** in ***Oreochromis niloticus niloticus*** were higher in site 2 than site 1 **(Table 2)**. Consequently, the pollution increases the prevalence, intensity and abundance of ***Acanthosentis tilapiae*** and this increase might be attributed to the municipal and industrial effluents. This result corresponds with **Billiard and Khan (2003)** who reported an increase in the prevalence and intensity of the acanthocephalans which infected the fish, *Tautogolabrus adspersus* in the area contaminated with municipal and industrial effluents.

c- Heavy metals pollution at the two investigated sites:

The contamination of freshwater with a wide range of heavy metals has become a matter of great concern over the last few decades **(Yilmaz *et al.*, 2007)** and a lot of studies have been published on the heavy metals at all levels of aquatic ecosystems **(Wagner and Bomam, 2003; Dugong *et al.*, 2006 and Jayakumar and Paul, 2006).** Many authors associated the heavy metal pollution in water with industrial and municipal discharges. These heavy metals may be taken up by living organisms, deposited in the sediments or remain for some period in the water itself **(Haggag *et al.*, 1999; and Zaghloul, 2000).**

In the present study, the highest concentrations for all studied heavy metals were recorded at site (2) when compared with site (1). The mean value of lead (Pb) was highly significant increase ($p<0.01$) at site 2. This result is parallel with that recorded by **Ibrahim (2007)** while, **Abd El-Monem, (2001) and Gabr *et al.* (2008)** recorded higher concentrations of lead however, **Abu El-Fadl (2008)** recorded lower levels. Also, the difference between mean values of Cadmium (Cd) at the two investigated sites (1&2) is highly significant ($p<0.01$). This result is parallel with the data recorded by **Abd El-Monem (2001),** relatively higher than the finding showed by **Abu El-Fadl (2008)** and less than those recorded by **Ibrahim (2007).** In addition to, the mean value of Copper (Cu) at site (1) was (0.4± 0.02 ug/l), while at site (2) it was (0.7± 0.04 ug/l). The difference between the two sites is highly significant ($p<0.01$) and parallel with the result of **AbuEl-Fadl (2008)**, but relatively higher than the finding recorded by **Abd El-Monem (2001).**

The differences in heavy metals concentration between site 1 and site 2 might be attributed to the highly discharge of mixture of industrial, municipal and agricultural drains into site 2, this opinion agrees with **Nagdi and Shaker (1998); Haggag *et al.* (1999) and Zaghloul (2000**)

Several authors reported that the variation in heavy metals concentration in water might be attributed to the contaminated sediment; these sediments reflect the quality of water current and form the major repository of heavy metals in aquatic systems. They added that the rate of accumulation depends, mainly, on the environmental parameters. Therefore, sediments can be used to detect the presence of contamination that does not remain soluble after the discharge into water **(Awadallah *et al*., 1996 and Chapman & Wang, 2001)**.

d- Fish parasites as bioindicator of heavy metals at the two investigated sites:

The data of the present work revealed that the two intestinal parasites, ***Acanthosentis tilapiae*** (Acanthocephala) and ***Orientocreadium lazeri*** (Trematoda) accumulate higher values of Pb, Cd and Cu than the infected organ (intestine) of their host fish species, ***Oreochromis niloticus niloticus*** and ***Clarias gariepinus***, with highly significant differences between the two sites. These results are in agreement with **Tenora et al.(2000)**; **Lohan *et al*. (2001)**; **Abd El-Monem (2001)** ; **Sures and Reimann (2003); Thielen et al. (2004), Sures (2008b)** , **Eira et al., (2009) and Jankovska et al. (2010)**.

Also, the accumulation of lead, cadmium and copper in the ***Orientocreadium lazeri*** was lower than the ***Acanthosentis tilapiae*** at the two sites. Moreover, this study revealed that the accumulation values of both ***Acanthosentis tilapiae*** and ***Orientocreadium lazeri*** were greatly different at the two sites, where, the accumulation values at site 2 were higher than site 1. This finding corresponds with **Abd El-Monem (2001)** and explains the great capability of both acanthocephalans and trematodes to accumulate heavy metals.

Sures (2003 and 2004) reported significantly higher quantity of heavy metals in the tissues of fish endoparasites than their hosts.

The heavy metals concentration in tissues reflects, post exposure via water and/or food **(Canli and Kalay, 1998 and Velcheva, 2006).**

Fish parasites, particularly intestinal acanthocephalans and cestodes, can accumulate heavy metals at concentrations significantly higher than those in host tissues or the environment **(Sures, 2001 & 2003; Schludermann *et al.*, 2003; Thielen *et al*., 2004 and Tekin-Ozan & Kir, 2005).**

Siddall and Sures (1998) considered the acanthocephalans useful and promising sentinels in environmental monitoring of metal pollution in aquatic habitats due to their remarkable capacity for metal accumulation. They added that the ability to bioconcentrate metals far above the ambient levels is restricted to the adults in the intestine of their definitive hosts, whereas larvae in the haemocoel of the intermediate hosts failed to show elevated metal levels**. Sures and Reimann (2003)** reported that the adult acanthocephalans are superior in accumulating metals than their definitive hosts not only in freshwater ecosystems but also in the marine environment.

The study of heavy metals accumulation in parasites can be subdivided into two main approaches; the first is the study of interrelationship between the heavy metals concentration in parasites and the tissues of their hosts and the second concerning with the relationship between the heavy metals concentration in parasite and the surrounding environment.

The bioaccumulation factor (BAF) calculated for the tissues of ***Orientocreadium lazeri*** and ***Acanthosentis tilapiae*** at site 2 were higher than those of the corresponding elements at site 1 for all metals and the difference was significant at ($p<0.01$) between the two sites (Tables 6). Also, the calculated bioaccumulation factor for the tissues of infected and uninfected ***Clarias gariepinus***, revealed insignificant difference between site1 and site2 for Cd and Cu but significant ($p<0.05$) for Pb in uninfected ***Clarias gariepinus*** only (Table 5). The same data were shown in ***Oreochromis niloticus niloticus***, but highly significant ($p<0.01$) for Cu in the uninfected ***Oreochromis niloticus niloticus*** (Table 5). In all cases above the concentration of heavy metals was ranked as: Lead > cadmium > Copper. The present study also, revealed the high bioaccumulation factor of both

Acanthosentis tilapiae and *Orientocreadium lazeri* in one hand and the increase difference of bioaccumulation factor between these parasites and their hosts on the other hand with a wide range of variation between the two sites (Table 6).

The heavy metals accumulation inside the tissues of different parasites and their hosts affected by many external and internal factors (**Retief *et al*., 2009)**. **Also, Chen *et al.* (2000)** mentioned that the concentration of metals in whole body tissues is often correlated with ambient metal levels in the contaminated habitat. Nearly, the same was recorded by **Ravera *et al.* (2007)** who mentioned that the abundance of available element forms in water and/or food at different sites in addition to some water characteristics can affect metal intake and accumulation**. Philips (1980**) suggested that both temperature and salinity could affect the rate of metal accumulation.

Many authors reported that the variability in the rate of accumulation may be attributed to the proximity of tissues to toxic medium, physiological state of the tissues, structural and functional of organs and the presence of ligands in tissues of organs having an affinity to the heavy metal (**Jayakumar and Paul, 2006; Dugo *et al*., 2006 and Sures, 2008a)**.

Bergey *et al.* (2002) considered the location of parasite within the host may be an important factor in its amount of accumulation, they added that the developmental stage of the parasite and the amount of time which the parasite is living in a particular host, are other factors that can influence metal accumulation. In the same context, they recorded that behavioral and physiological changes, resulting from parasitism, could alter a host's feeding and metabolism and consequently the accumulation of heavy metals could be affected.

Szefer *et al.* (1998) suggested that the bioaccumulation of helminthes may reflect the higher ability of host to clear heavy metals. **Sures and Siddall (1999), Taraschewski (2000) and Malek *et al*. (2007)** considered the parasites beneficial and might act as a heavy metal sanitizer for the host**.**

Several authors have discussed the passage and entry of heavy metals into aquatic animal tissues. They mentioned that heavy metals can enter the animal tissue via different routes such as the uptake from surrounding water or intake from food **(Ravera, 2001).** Soluble metal ions are the most available for uptake by aquatic organisms **(Rainbow, 2002 and AbdAllah, et al., 2003)**. Bioaccumulation of heavy metals not only depends on the structure of the organ, but also on the interaction between metals and the target organs (**Sorenen, *et al*., 1980 and AbdAllah, 2006)**. The strategy of metal interaction depends mainly on the metal ion speciation and the comparative concentrations of each metal **(Parrott & Sprague, 1993 and Wang *et al*., 1995).**

The results of the current study, revealed certain correlation between the heavy metals under study. This correlation may be present between some or all heavy metals or may not be. The positive correlation between metals indicating to the synergistic effect of both metals on their uptake and bioconcentration by fish tissues and their parasites, but the negative significance revealing the antagonistic effect of those metals on their bioconcentration within the examined tissues **(Otitoloju, 2002 and AbdAllah, 2006)**. The correlation between heavy metals may be attributed to many internal and/ or external factors such as physiological and behavioral conditions concerning the parasites and their hosts and the physicochemical parameters**. Ravera *et al.* (2007**) reported that the positive correlation between some elements pairs could be the result of their metabolic analogy and/or the seasonal variations of the available forms in the environment, whereas the negative correlation may be due to the competitive inhibition of the metabolic sites**.**

Further studies are still needed to investigate the effect of heavy metal contamination on the biochemical parameters, light and fine structure of helminth parasites to evaluate their potentiality as sentinel organisms to heavy metal contamination in aquatic habitats.

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توزيع بعض العناصر الثقيلة بين الطفيليات المعوية وعوائلها من أسماك نهر النيل في محافظة أسيوط

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فى هذه الدراسة تم تجميع عدد 1025 سمكة من الأسماك النيلية الشائعة هما، البلطى النيلى والقرموط، من منطقتين مختلفتين بمحافظة أسيوط ، منطقة ترعة الابراهيمية (المنطقة الاولى) و هى منطقة غير ملوثة ومنطقة ترعة الملاح (المنطقة الثانية) وهى منطقة ملوثة حيث يتم صرف مخلفات الصرف الصحى و الزراعى و بعض الورش و المصانع اليها مباشرة. وقد تضمنت هذة الدراسة عدة موضوعات هى:

1- دراسة العوامل التى تؤثر على جودة المياة فى كلا المنطقتين موضوع الدراسة.
2- التعرف على الديدان الطفيلية وتوزيعها بين منطقتى الدراسة مع دراسة تاثير العناصر الثقيلة على بعض الجوانب البيئية للطفيليات.
3- قياس تركيز العناصر الثقيلة (الكادميوم , الرصاص و النحاس) فى عينات الماء، الطفيليات المعوية و الامعاء (العضو محل الاصابة) لنوعى الاسماك موضع الدراسة وقياس عامل التركيز Bioaccumulation factor لتلك المعادن فى الطفيليات المعوية و الامعاء.

وقد اظهرت الدراسة:

1- وجود فروق معنوية فى متوسطات العوامل البيئية التى تؤثر على جودة المياة بين منطقتى الدراسة،فكلا من العسر الكلى للماء , القلوية الكلية , الملوحة , الامونيا ,التوصيل الكهربى, النيتريت ، الاس الهيدروجينى (PH) و الاكسجين الذائب (O_2) كانت ($p < 0.01$).بينما درجة حرارة المياة كانت.($p< 0.05$).
2- أن تركيزات العناصر الثلاثة ؛ الكادميوم(Cd)،النحاس(Cu) و الرصاص(Pb) كانت اعلى فى المنطقة الثانية مقارنة بالمنطقة الاولى واوضح التحليل الاحصائى للتباين وجود فروق معنوية ($p<0.01$)
3- **معدل الاصابة,الكثافة و الوفرة للطفيليات :**
اوضحت الدراسة الحالية و جود علاقة بين كلا من التلوث والتطفل ، فقد اظهرت النتائج ان اسماك القرموط المجمعة من المناطق الملوثة و غير الملوثة مصابة بطفيل الاورينتوكريديم لازيرى , بينما الاصابة بطفيل الباراكاملينيس سيا ثوفارينكس كانت مقتصرة على الاسماك المجمعة من المنطقة الغير ملوثة فقط (ترعة الابراهيمية) . على الجانب الاخر وجد ان اسماك البلطى المجمعة من كلتا المنطقتين مصابة بالاكنثوسينتس تلابى اما الكلينوستوميم فالكركورسيس فقد كانت الاصابة به مقصورة على الاسماك المجمعة من المنطقة الغير ملوثة فقط (ترعة الابراهيميه).
4- **قياس تركيز العناصر الثقيلة:** بقياس تركيز العناصر الثقيلة فى انسجة العائل والطفيل فى المنطقتين وجد:
أ- فى حالة اسماك القرموط الغير مصاب ،اظهرت التحاليل الاحصائية وجود فروق معنوية بين منطقتى الدراسة فى تركيز الرصاص(Pb) ($p<0.05$)
ب- أما فى حالة اسماك القرموط المصاب ،فقد اظهرت التحاليل الاحصائية وجود فروق معنوية بين منطقتى الدراسة فى تركيز الرصاص (Pb) وتركيز الكادميوم (Cd).($p<0.05$)
ت- فى حالة اسماك البلطى الغير مصاب ،اظهرت التحاليل الاحصائية وجود فروق معنوية بين منطقتى الدراسة فى تركيز الرصاص (Pb) وتركيز النحاس (Cu) .($p<0.01$)
ث- أما فى حالة اسماك البلطى المصاب ، فقد اظهرت التحاليل الاحصائية وجود فروق معنوية بين منطقتى الدراسة فى تركيز جميع العناصر($p<0.05$) .
ج - فى حالة طفيل الاكانثوسينتس تيلابى وطفيل الاورينتوكريدم لازيرى ،اظهرت التحاليل الاحصائية وجود فروق معنوية بين منطقتى الدراسة واظهرتحليل المتغيرات وجود فروق معنوية .($p<0.01$) فى تركيز جميع العناصر.

ومما سبق يمكن استنتاج مايلى:-

اولا: دور الطفيليات كمؤشرات بيولوجية للتلوث بالعناصر الثقيلة و كذا مقدرتها على تجميع الجزء الاكبر من هذة العناصر المتراكمة فى العضو المصاب بهذة الطفيليات.
ثانيا:مقدرة الطفيليات على ترسيب العناصر الثقيلة تختلف باختلاف الانواع حيث وجد ان مقدرة الاكنسوسيفلاعلى الترسيب اعلى من التريماتودا سواء عند مقارنتها بانسجة العائل او البيئة المحيطة.
ثالثا: وجود علاقة بين كلا من التلوث والتطفل حيث وجد ان التطفل يلعب دور كبير فى انتشار الطفيليات فقد تختفى بعض الانواع او تقل او تزيد بوجودة.