EFFECT OF HEAVY METAL CONCENTRATIONS ON SOME FISHES IN KAFR EL-ZAYAT AREA, ROSETTA BRANCH, RIVER NILE, EGYPT.

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Introduction:
In Egypt, the effect of heavy metals on fish is of great importance since fish is becoming a necessity as relatively cheap source of animal protein and may compensate for the present deficiency of other expensive protein (Zaghloul et al., 2001). Heavy metals exert a wide range of effects on fish, from metabolic and physiological to behavioral and ecological aspects (El-Naggar et al., 1998).

The level of metals bioaccumulation in fish tissues is influenced by abiotic and biotic factor, such as fish biological habitat, chemical form of metals in the water, water temperature, pH value, dissolved oxygen, water transparency as well as by fish age, gender, body mass and physiological conditions (Has-Schon et al., 2006). In addition, there are significant differences in the concentrations of elements across fish species related to organisms mobility, food preference or behavior with respect to environment (Demirezen and Uruc, 2006).

The aim of the present study is to study the effect of different pollutants such as industrial and sewages on water quality of River Nile in Egypt and some species of Nile fishes; the Nile tilapia or bulti, bayad or silver catfish and the African catfish or catfish, which were collected from different Nile sites in Kafr El Zayat area.

Materials:
Study sites:
This study was conducted at Kafr El- Zayat town. It located on Rosetta branch of the River Nile. Five sites in the study area were selecte:
1. Tala drainage: drain sewage water directly in Nile water.
2. The second site is located by about 200 m away from the first site.
3. This site is in-between sites 2 and 3 in middle of Rosetta branch water.
4. The area of industrial companies which drain their waste water into Nile water. These companies change their drain outlet from this area into Ganag drain which drains directly into Borolos Lake in the north.

5. This site after site 4 by about 150-200m.

Water samples:-
were taken with a water sampler from each site seasonally. Duplicates of water samples were taken from each of the five studied sites, between 10:00 and 12:00 a.m. at a depth of 30cm below the water surface and stored at 4˚c in clean1000 ml sampling glass bottles according to Boyd (1990).

Fish species:-
The present comparative study was carried out on three different species of fish in the River Nile in Egypt; the Nile tilapia or bulti, Oreochromis niloticus (Perciformes; Cichlidae); bayad or silver catfish Bagrus bayad (Siluriformes; Bagridae); and the African catfish or catfish Clarias gariepinus (Siluriformes; Clariidae). The three fish species were collected seasonally during (2015-2016) from the studied sites. The fishes were immediately dissected to extract muscle samples.

Methods:-
Water heavy metals concentration (Fe, Mn, Cu, Cd, Ni) were determined by a Perkin-Elmer 2380 atomic absorption spectrophotometer (AAS) according to APHA(2005).

The collected fishes were washed with de-ionized water, put in cleaned plastic bags and stored frozen until analysis was carried out. One gram of muscle tissue sample (wet weight) was subjected to digestion by adding 10 ml of freshly prepared 1:1 concentrated HNO$_3$-HClO$_3$ in beaker, covered with a watch glass till initial reaction subsided in about 1 hour and gently heated at 160ºC in a sand bath on a hot plate till reduction of volume to 2-5 ml. The digested samples were allowed to cool and transferred into 25 ml volumetric flasks and made up to mark with de-ionized water (Olaifa et al., 2004). Heavy metal concentrations (Fe, Mn, Cu, Cd, Ni) were determined using flame atomic absorption spectrophotometer (Thermo Scientific ICE 3300, UK) with double beam and deuterium background corrector according to (APHA, 2005).

Results:-
The concentration of heavy metals (Fe, Mn, Cu, Cd and Ni) in water samples expressed in unit of mg/l were show in Table ( 1 ). The concentrations of the heavy metals tested in this study were significantly different (p ≤ 0.05) in either sites or seasons of the present study, except for that of Fe, which show no significant differences in sites as well as seasons. However, generally speaking, the concentrations of iron in each of sites and seasons were higher than that of the other heavy metals tested.

Regarding the concentration of these heavy metals recorded in the different seasons, it is noticed that; the concentration of each of Fe, Cu, and Cd were higher in spring season. For the concentrations of each of Mn and Ni, were higher in summer and autumn seasons, respectively. Lower values of each of Fe and Cd were recorded in summer season, and for Cu and Ni in winter season, whereas Mn in autumn season. Site 1 has higher concentrations of each of Fe, Cu, and Ni; while site 5 has high values of each of Mn and Cd. On the other hand, site 2 has low values of Fe and Mn and site 5 has low values of Cd and Ni whereas, site 3 has low value of Cu.

Table 1:- Seasonal variation of heavy metal concentrations in water samples in different sites.

| Metal | Site | Summer2015 | Autumn | Winter2016 | Spring |
|-------|------|------------|--------|------------|--------|
| Fe    | 1    | 2.331$^a$  | 5.000$^a$ | 4.953$^a$  | 5.463$^a$ |
|       | 2    | 1.040$^a$  | 4.140$^a$ | 4.077$^a$  | 5.030$^a$ |
|       | 3    | 3.416$^a$  | 4.750$^a$ | 3.647$^a$  | 4.637$^a$ |
|       | 4    | 2.334$^a$  | 5.010$^a$ | 4.332$^a$  | 5.003$^a$ |
|       | 5    | 1.111$^a$  | 4.620$^a$ | 3.667$^a$  | 4.253$^a$ |
| Mn    | 1    | 1.341$^a$  | 0.170$^c$ | 0.994$^{bc}$ | 1.028$^{bc}$ |
|       | 2    | 0.943$^{bc}$ | **0.120**$^g$ | 0.471$^{bc}$ | 0.868$^{cd}$ |
|       | 3    | 0.874$^{cd}$ | 0.160$^g$ | 0.166$^{c}$ | 0.504$^{c}$ |
|       | 4    | 1.200$^{cd}$ | 0.140$^g$ | 0.242$^{c}$ | 0.701$^{bc}$ |
|       | 5    | **1.356**$^a$ | 0.200$^g$ | 0.131$^{c}$ | 0.139$^{c}$ |
and the muscles of each of bulti and catfish, respectively. On the other hand, there were significant negative correlation at \((p \leq 0.05)\). There are significant positive correlation at \((p \leq 0.05)\). The correlation coefficient and regression analysis between concentrations of heavy metals in water are shown in Table (2).

### Table 2: Seasonal variation of heavy metals in muscles of three species of Fish

| Metal | Fish | Sum 2015 | Aut 2015 | Win 2016 | Spr 2016 |
|-------|------|----------|----------|----------|----------|
| Fe    | bulti | 2.146h   | 3.000e   | 2.872f   | 2.907i   |
|       | bayad | 1.432i   | 1.23j    | 1.177k   | 0.893l   |
|       | catfish | 3.752a   | 3.365b   | 3.670c   | 3.420d   |
|       | bulti | 0.134a   | 0.111b   | 0.152c   | 0.043f   |
| Mn    | bayad | 0.088c   | 0.003d   | 0.087e   | 0.052h   |
|       | catfish | 0.188e   | 0.152f   | 0.178g   | 0.163h   |
|       | bulti | 0.017abcda | 0.012bcd | 0.018bc | 0.014cde |
| Cu    | bayad | 0.004cde | 0.001de  | 0.004cde | 0.000f   |
|       | catfish | 0.032a   | 0.022ab  | 0.032a   | 0.021ab  |
| Cd    | bulti | 0.017abcda | 0.014cde | 0.018ab  | 0.015cde |
|       | bayad | 0.003cde | 0.000d   | 0.003cde | 0.000d   |
|       | catfish | 0.026a   | 0.019ab  | 0.022a   | 0.021a   |
| Ni    | bulti | 0.109c   | 0.131d   | 0.151c   | 0.138cd  |
|       | bayad | 0.072f   | 0.030g   | 0.071f   | 0.004b   |
|       | catfish | 0.190e   | 0.163b   | 0.188a   | 0.185s   |

Means with different superscript letters for each metal are statistically significant at \(p \leq 0.05\)

The concentration of heavy metals (Fe, Mn, Cu, Cd and Ni) in muscles of three species; *Oreochromis niloticus* (bulti), *Bagrus bayad* (bayad) and *Clarias gariepinus* (catfish) are expressed in unit of \(\mu g/g\) wet wt and were show in Table (2).

The concentration of each of Fe, Mn, Cu, Cd, and Ni expressed in \(\mu g/g\) wet wt. in the muscle tissues of each of bulti, *O. niloticus*; bayad, *B. bayad*; and the catfish, *C. gariepinus*, were significantly different during all seasons, except that of Cd-ion in the case of *C. gariepinus* (Table 2). It is noticed that, the highest values concentrations of each of Fe, Mn, Cu, Cd, and Ni were recorded in summer season in both of the fish species bayad, *B. bayad*; and the catfish, *C. gariepinus*. However, the highest concentrations of these metals in the muscle tissue of bulti, *O. niloticus*, were recorded during winter season except that of Fe metal, which recorded in autumn season. On the other hand, the lowest values of the five heavy metals concentrations were, in general, recorded during autumn season with some values which recorded during either spring or summer seasons.

The correlation coefficient and regression analysis between concentrations of heavy metals in muscles of each of the studied fishes; bulti, bayad, and catfish and the concentrations of these heavy metals in water are shown in Table (3). There are significant positive correlation at \((p \leq 0.05)\) between the concentrations of each of Fe and Mn in water and the muscles of each of bulti and catfish, respectively. On the other hand, there were significant negative
correlation at \((p \leq 0.05)\) between the concentrations of each of Cu and Cd in water and the concentration of these metals in the muscles of each of bulti and catfish.

Table 3:- The correlation coefficient \((r)\) and regression analysis between heavy metal concentrations in water and muscles of the tested fishes.

| Metal | Fish   | \(r\)     | Regression |
|-------|--------|-----------|------------|
|       |        |           | \(a\)  | \(B\) |
| Fe    | bulti  | 0.978*    | 1.54    | 0.30 |
|       | bayad  | -0.807    | 1.86    | -0.17|
|       | catfish| -0.733    | 4.2     | -0.16|
| Mn    | bulti  | 0.562     | -0.02   | 0.22 |
|       | bayad  | 0.730     | 0.003   | 0.127|
|       | catfish| 0.823*    | 0.15    | 0.051|
| Cu    | bulti  | -0.968*   | 0.028   | -0.072|
|       | bayad  | -0.774    | 0.011   | -0.054|
|       | catfish| -0.843*   | 0.045   | -0.158|
| Cd    | bulti  | -0.34     | 0.19    | -0.045|
|       | bayad  | -0.567    | 0.005   | -0.043|
|       | catfish| -0.883*   | 0.028   | -0.073|
| Ni    | bulti  | 0.533     | 0.091   | 0.171|
|       | bayad  | 0.707     | -0.033  | 0.323|
|       | catfish| 0.464     | 0.152   | 0.121|

*significant at \(p \leq 0.05\)

Discussion:-

The Nile River has been subjected to different sources of pollution and contamination through several and complicated routes. Industrial effluents constitute a real threat to The Nile River. Recently, the risk of water pollution with toxic chemicals not limited to the public health and veterinary public health only but extended and jointed as toxic chemicals causing zoonotic diseases as reported by (El-Tras et al., 2011). The Nile River at Kafr El-Zayat, is impacted by several industrial outfalls from oil and soap, fertilizers, pesticides and sulfur industry (El-Malh and Soda, El-Malyia and El-Mobidat factories). There were other sources of pollution and contamination as agricultural drains, laying carcasses and sewage which discharged from several villages distributed along the two banks of the Rosetta Branch. In general, the distribution of these metals at the area under investigations are strongly affected by the industrial effluents produced from El-Mobidat, El-Malyia and Salt and Soda companies which directly discharge industrial effluents at this area without any treatment (Issa et al. 1996).

Different species of fish may be used to determine the mechanisms of action of pollutants on specific physiological functions (Gul et al., 2004). Because fish respond with great sensitivity to changes in aquatic environment, they are one of the most indicative factors in aquatic environment for estimation of trace metals pollution and risk potential of human consumption (Authman et al., 2013). Hence, it is important to determine the concentration of heavy metals in commercial fish in order to evaluate the possible risk of fish consumption (Yilmaz et al., 2007). Bioaccumulation of heavy metals in tissues of aquatic organisms has been identified as an indirect measure of the abundance and availability of metals in the aquatic environment (Kucuksegin et al., 2006). For this reason, monitoring fish tissue contamination represent an important function as an early warning indicator of related water quality problems and enables ou to take appropriate action to protect public health and the environment (Murtala et al., 2012). Fish accumulate metals may from metal complex with the structural proteins, enzymes and nucleic acids which interrupt their functions and leading to morphological, histological and biochemical alteration in the tissues which may critically influence fish health (Ismail and Saleh, 2012).

Generally metal concentration may differ greatly between one organism and another and between different organs of the same organism (Watanabe et al., 2003; Massoud et al., 2007). The distribution pattern of the heavy metals Fe, Mn, Cd, Cu and Ni, had different concentration in muscles of the three species of fish. According to Sorensen (1991) bioaccumulation of heavy metals depend on the interaction between metals and the target organ. The obtained results of all toxic trace metals in muscles in different species of fish indicate the presence of agricultural
and domestic wastes as environmental pollutants among the studied sites and their accumulation edible and non-edible fish tissues.

In conclusion, the results demonstrate that the Nile River at Kafr El-Zayat industrial area is heavily polluted and consequently harmful effects to the aquatic environment and to the quality of the water are established. So, pretreatment of different wastes through chemical and microbiological unit before discharging to the River Nile stream is recommended.

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