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Research Paper

Neoechinorhynchus rutili (Acanthocephala) from *Capoeta trutta* as a Bioindicator for Heavy Metal Pollution in the Keban Dam Lake

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ABSTRACT: *In this study, some heavy metal (Cd, Cu, Cr, Fe, Pb, Zn and Mn) were at higher levels in muscle and organ tissues (liver, intestine and gill) of Capoeta trutta and its parasite, Neoechinorhynchus rutili (Acanthocephala), from a location of Keban Dam Lake were analyzed. The sampling station (Kockale) were chosen based on ecological conditions and human activities in the area. Twenty-four fish (11 non-parasitized and 13 parasitized) were collected in May to September 2015. There were differences between the heavy metal levels in fish tissues and parasite, N. rutili. The highest level of Cd and Cr, was recorded in the N. rutili compared to fish tissues.*

KEYWORDS: *Keban Dam; Capoeta trutta; Neoechinorhynchus rutili; Metal concentration; Accummulation*

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I. INTRODUCTION

Heavy metals pollution of water sources is an environmental problem [1]. Monitoring of heavy metals in water is, therefore, important for control of environmental pollution. Also, many of these heavy metals can occur as ecological toxins in aquatic ecosystems and may pose a risk to human health [2, 3, 4].

Parasite species and intensity directly affect fish health. Heavy metals can cause a disruption of metabolism. Once these metals are accumulated in parasites, metal tolerance appears to be high in accumulating species. This situation may pose a threat to other aquatic animals within the food chain. Fish parasites are highly sensitive because of their physiological response to aquatic contaminants. Also, their characteristically accumulate metals [5]. Therefore, fish parasites may serve as the perfect bioindicator.

Fish parasites are an important part of the aquatic ecosystem in terms of species diversity and abundance Their use as biomonitor and bioindicator species in aquatic ecosystems has attracted ecologists' interest in recent years and has resulted in an increase of their importance in biomonitoring. Because they quickly respond to environmental changes. Fish parasites are also used as biological indicators of the phylogenetic position of hosts, population biology, stock removal and migration and feeding. As accumulation indicators, they can accumulate higher concentrations of heavy metals than the aquatic environment. They are also useful as ecosystem effect indicators due to possible changes in biomass, community structure, species diversity, richness, and abundance of fish parasites in environmental pollution surveys [6].

Previous studies show higher concentrations of heavy metals in parasites compared to that of host tissue. Tenora et al. [7] studied heavy metals in fish and demonstrated higher concentrations of lead, chromium and cadmium in *Philometra ovata* (Nematoda) than in its hosts. Brazova et al. [8] reported that Acanthocephalans as well as Cestodes seem to be good indicators of environmental conditions. Therefore, this study focused on *N. rutili* (Acanthocephala) and its host (*C. trutta)* to determine levels of heavy metal concentrations in Keban Dam (Elazig, Turkey).

II. MATERIALS AND METHODS

The research protocol (Protocol number: 2016/03) for all animal studies was approved by Kahramanmaraş Sutcu Imam University, Faculty of Agriculture Ethics Committee (KSUZİRHADYEK, Kahramanmaraş, Turkey).

A total of 24 *C. trutta* were obtained from Kockale location of Keban (Elazig, Turkey) between May 2015 and September 2015 (Figure 1). Length and weight of fish were recorded and sex was determined. After the fish were brought to the laboratory they were necropsied and *N. rutili* were removed from the intestines. Parasites recovered were first rinsed in a 0.9% saline solution, placed in cold (4^{o}C) distilled water to force osmotic evagination of the proboscis. Thereafter some were fixed in alcohol/formalin/acetic acid (AFA) fixative and preserved in 70% ethanol until processed for identification. The *N. rutilii* were identified using parasite identification keys [9, 10] and the number of *N. rutilii* per fish were counted. Prevalence, abundance and mean intensity of infection were calculated according to Bush et al. [11]. Tissue samples (gill, liver and intestine) and parasites were frozen at -20 ^{0}C (Siemens, GS33VVW30N, Germany) until further analysis.

Prevalence =Number of parasitised fishes / Total number of fishes x 100 Average density = Total number of parasites / Number of parasitised fish Average abundance = Total number of parasites / Total number of fish

A part of tissue was cut away at the gastrointestinal junction to ensure only intestinal tissue was analyzed.

The samples were digested with concentrated nitric acid and perchloric acid (3:1) until a clear and transparent solution was obtained. The digested samples were diluted to make the volume by using distilled water. After wet digestion, samples were analyzed for Cd, Cr, Cu, Fe, Mn, Pn and Zn concentrations according to American Public Health Association (APHA) [12] through Atomic Absorption Spectrometry (Perkin Elmer, As 800) , and the instrument was calibrated with standard solutions prepared from commercially available chemicals Merck, Germany [13].

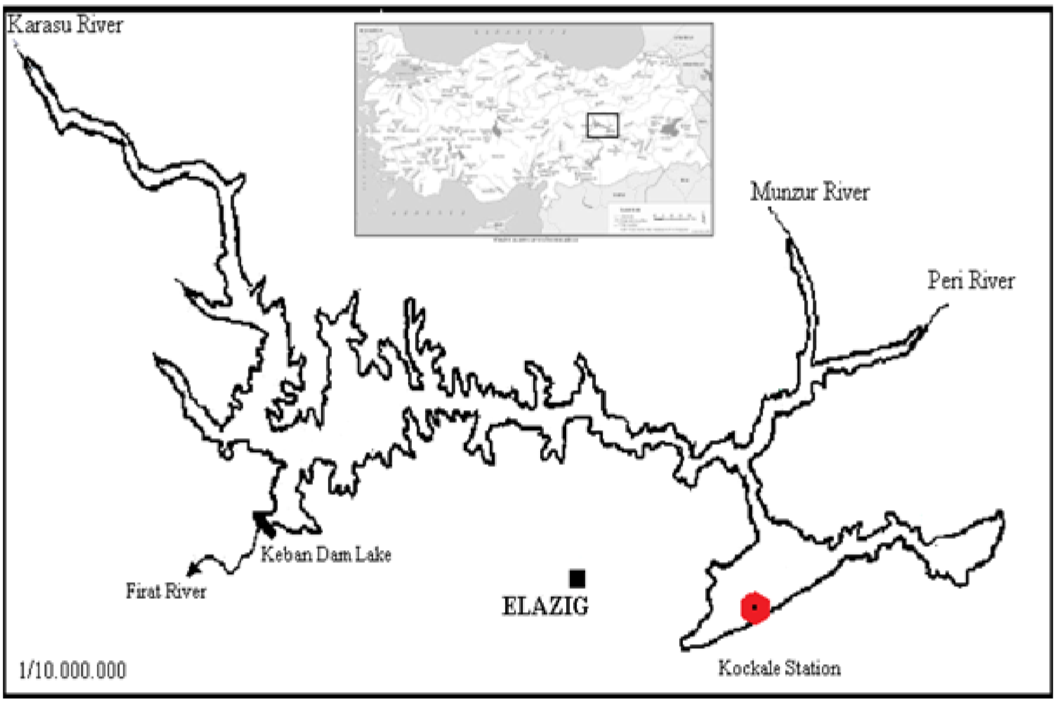

Figure 1: Keban Dam Lake map showing the site of sampling

Means and standard errors were calculated for each experimental group. Statistical analysis of data was carried out with SPSS statistical package.

III. RESULTS AND DISCUSSION

The mean weight and length of *C. trutta* collected from Keban Dam were 213 ± 25 g and 24.4 ± 32 cm, respectively. A total of 13 (54.16%) of the 24 *C. trutta* examined were infected with *N. rutilii* and a total of 371 parasite were collected from the infected fish. Mean abundance and mean intensity of parasite is shown in Table 1.

Table 1: Distribution of *Neoechinorhynchus rutili* in *C. trutta* from Keban Dam Lake

Parameters	
Number of fish examined	24
Number of infected fish	13
Total number of parasites	371
Mean abundance	15.46
Mean intensity	28.54
Prevalence of infection (%)	54%
Minimum number of parasites	9
Maximum number of parasites	61

The concentrations of heavy metals in un-parasitized and parasitized fish tissues are given in Tables 2 and 3. There were no significant differences between heavy metals con- centrations in tissues of unparasitized *C. trutta*. The bioaccumulation data showed that the analysed is this all metals were accumulated in the following order: Parasites > Muscle > Liver > Intestine. Heavy metal concentrations in host-parasite systems including *N. rutili* and their fish host are shown in Table 4. Concentrations of the detected heavy metals (Cd and Cr) accumulated in the biomass of *N. rutili* were higher than those in the muscles of *C. trutta*.

Table 2: Mean Cd, Cu, Cr, Fe, Pb, Zn and Mn contents (mg Kg^{-1} dry weight) in non-infected *C. trutta* of Kockale on Keban Dam (Elazig, Turkey)

Metals	**Liver**	**Gills**	**Intestine**	**Muscle**
Cd	0.99±0.04	1.81±0.03	0.76±0.04	1.08±0.03
Cr	3.87±1.09	10.23±0.63	8.44±0.88	0.51±0.18
Cu	8.18±1.21	7.11±0.78	6.78±1.54	3.09±0.57
Fe	1102.91±96.04	987.48±78.0	789.21±52.4	189.21±38.3
Mn	20.13±8.09	50.77±7.80	48.22±6.98	8.13±4.01
Pb	1.23±0.41	1.01±0.43	0.92±0.78	0.82±0.18
Zn	26.48±4.91	61.82±6.14	72.56±7.15	30.08±3.04

Table 3: Mean Cd, Cu, Cr, Fe, Pb, Zn and Mn contents (mg Kg^{-1} dry weight) in infected *C. trutta* of Kockale on Keban Dam (Elazig, Turkey)

Metals	**Liver**	**Gills**	**Intestine**	**Muscle**
Cd	0.87±0.01	1.64±0.07	0.82±0.03	0.38±0.01
Cr	0.97±0.06	18.09±0.71	11.14±0.62	0.107±0.08
Cu	7.44±1.80	11.94±0.84	7.96±1.01	2.92±0.41
Fe	1003.63±81.3	1393.64±59.72	972.44±47.21	340.53±21.2
Mn	16.09±0.61	34.77±4.53	32.12±4.28	5.87±0.72
Pb	0.88±0.52	0.95±0.68	0.83±0.61	0.64±0.69
Zn	23.49±12.83	62.18±5.49	49.24±5.18	28.37±2.92

Table 4: Mean Cd, Cu, Cr, Fe, Pb, Zn and Mn contents of *N. rutili* in *C. trutta* from Kockale on Keban Dam Lake (Elazig, Turkey)

Metals	**Parasite (*N. rutili*)**
Cd	2.61±1.49
Cr	9.72±0.43
Cu	11.75±0.52
Fe	521.56±28.19
Mn	13.98±0.22
Pb	6.11±0.68
Zn	27.48±13.39

Some heavy metal levels (Cd, Cr, Cu, Fe, Mn, Pb and Zn) in muscle and organ tis- sues (gill, liver and intestine) of *C. trutta* and it's a parasite, *N. rutili* (Acanthocephala), from a location of Keban Dam Lake were analyzed. Twenty-four fish (13 parasitized and 11 non-parasitized) were collected in May to September 2015. A higher concentration of Cd and Cr, was recorded in the *N. rutili* compared to fish tissues.

In their study on *N. rutili* in *C. trutta*, Sağlam and Sarıeyyüpoğlu [14] reported that out of the 37 fish from the Koçkale region of Keban Dam, where the sewage water of Elazığ was discharged, 14 were infested; and they found that the prevalence was 2.70%, concentration was 14 and abundance was 38%. In Keban Dam Lake, Dörücü et al. [15] found that 5 out of 7 *C. trutta* were infested, the prevalence was 71.43%, the was 36.8 and the abundance was 26.3. In the present study, it was found that 13 out of 44 *C. trutta* were infested, the total number of parasites was 371, mean intensity was 28.54, prevalence was 0.54%, and mean abundance was calculated to be 15.46.

Fish are at a higher trophic level organism, such as humans, they accumulate heavy metals such as Pb, Cd, Cr, Hg, Zn and Fe. In addition, the chemical properties of water and biological factors such as organic compounds, habitat selection, diet and growth rate also affect the concentration of heavy metals in fish [16, 17, 18, 19, 20, 21]. Furthermore, the physiological status, length and age of the fish plays a role and the relationships between different elements affect the accumulation and toxicity of heavy metals [22]. During heavy metal analysis in tissues and organs of carp and tench in Beyşehir Lake, Fe and Zn were detected, in all tissues and in every season [23]. Similarly, in the present study, heavy metal analysis of muscles, gills, liver and intestines of *C. trutta*, in Keban Dam Lake showed that the concentration of Fe and Zn were the highest in all organs and tissues.

Other studies in Turkey, Canlı et al. [24] found that the liver and gills of the *C. carpio* (gill: 7.46±2.3, liver: 10.21±7.6, muscle: 3.78±1.4), *B. capito* Gill: 7.52±2.63, liver: 15.09±11.39, muscle: 4.45±2.92) and *C. regium* gill: 8.14±9.07, liver : 26.61±21.66, muscle: 3.48±1.65) Seyhan River accumulated higher concentrations of metal when compared to the muscle tissue; Canlı and Atli [25], found that the metal concentration in the liver of some fish species living in the Black Sea had higher metal concentrations when compared to their muscle tissues; Küçükbay and Örün [26] found that Cu (0.07±0.01) and Zn (0.44±0.16) accumulate in high concentrations in the liver of *C. carpio* living in the Karakaya Dam Lake; Canpolat and Çalta [27] found that the Cu, Fe, Mn and Zn in the skin and muscle tissues of *C. capoeta umbla* living in Keban Dam was lower than in the liver and gill; Karadede et al. [28] found that the Co, Cu, Fe, Mn, Mo, Ni and Zn in the liver and gills of *L. abu* and *S. triostegus* in Atatürk Dam were higher than in the muscle tissue. In this study, it was observed that the metals accumulated mostly in liver and then highest in the liver in (166.25±15.97) followed by the gill (160.03±13.4), intestines (132.41±9.96) and muscle (33.27±6,61) tissue. It was determined that the lowest concentration of accumulated metal in the liver was Cd (0.99±0.04). Fe (1102.91±96.04 and Zn (26.48±4.91) accumulated in the liver at high levels. The high accumulation of metals in the liver was due to the fact that it is a metabolically active organ. Metal accumulation was observed to be lower in the gill (160.03±13.4) than in the liver (166.25±15.97). It was determined that the metal that accumulated least in the gill was Pb (1.01±0.43) and that the metal that accumulated most was Fe (987.48±78.0). The high concentration of metal in the gill was due to the fact that the metals absorbed from water dissolved into the mucus on the gill and stayed between the lamellae [29]. It was seen that the metal accumulation in the intestinal tissue (132.41±9.96) was lower than that in the gills (160.03±13.4). The metal that accumulated the least in the gill was Pb (1.01±0.43) and the metal that accumulated the most was Fe (987.48±78.0). Metal accumulation in muscle tissue (33.27±6.61) was low. The metal that accumulated least in the muscle was Cr (0.51±0.18) and most was Fe (189.21±38.3). The low accumulation of metal in muscular tissue can be explained by the activity of Metallothioneins. Because, this protein which have the ability to bind heavy metals are absent from muscle, and thus allow the tissue to accumulate their metals at a high concentration.

In Turkey, Tekin Özan [22] evaluated the concentrations of the heavy metals in the plerocercoid of *Ligula intestinalis* in terms of seasons (Spring, Summer, Autumn and Winter). It was reported that Cu and Mn concentration increased in summer and the concentration of Fe and Zn increased in spring. When metal concentration in the plerocercoid of *L. intestinalis* was compared to the concentration in tissues it was observed that the Fe in muscle tissue, Cu and Fe in the liver and the Fe and Mn in the gill were higher when compared to the that in the plerocercoid of *L. intestinalis.* In this study, when the metal in *N. rutili* were compared to the metal in the tissues and organs of the host, the Zn (28.37±2.92) in muscle tissue, Mn (16.09±0.61) and Fe (1003.63±81.3) in the liver, Fe (1393.64±59.72), Mn (34.77±4.53), Zn (62.18±5.49) and Cr (18.09±0.71) in the gill, Cr (11.14±0.62), Fe (972.44±47.21), Mn (32.12±4.28) and Zn (49.24±5.18) in the intestines were higher than in the *N. rutili* (Zn: 27.48±13.39; Mn: 13.98±0.22; Fe: 521.56±28.19; Cr: 9.72±0.43). The fact that the metals in the tissues and organs of the host, were higher than the concentration in *N. rutili* was due to the position of the parasite in the host body and the age of the parasite [30]. Whereas it was determined that the Cd in the *N. rutili* was 3 times higher than the Cd in the liver, 1.59 times higher than the Cd in the gill, 3.18 times higher than the Cd amount in the intestines, 6.86 times higher than the Cd in the muscle tissue; its Cr was found to be 10.02 times higher than the Cr in the liver, 90.84 times higher than the Cr in the muscle tissue; its Cu was 1.57 times higher than the Cu in the liver t, 1.47 times higher than the Cu in the intestines, 4.02 times higher than the Cu in the muscle tissue; its Fe was 1.53 times higher than the Fe in the muscle tissue; its Mn was 2.38 times higher than the Mn in muscle tissue; its Pd was 6.94 times higher than the Pd in the liver, 6.43 times higher than the Pd in the gill, 7.36 times higher than the Pd in the intestines, 9.54 times higher than the Pd in the muscle tissue; and, its Zn was 1.16 times higher than the Zn in the liver. Since *N. rutili* occur in the intestines of the host, its potential to accumulate and exposure the metals were higher than that of endoparasites living in the abdominal cavity. Hence, this type of endoparasites directly absorbed the metals passing through the digestive system through food. Therefore, the fact that the *C. trutta N. rutili* of certain metals was measured in high amounts in the study may depend on the parasite living in the intestine of the host.

IV. CONCLUSION

Parasites and heavy metals seem to influence the fish population. The present study shows that is important to monitor bioaccumulation of heavy metals in fish and their intestinal parasites. Therefore, the Acanthocephalan parasite have potential as bioindicators to monitor heavy metal concentrations in because of its high bioaccumulation capacity.

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