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# Uptake of Heavy Metals by Channa Punctatus from Sewage– Fed Aquaculture Pond of Panethi, Aligarh

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*Abstract* - Investigations on the bioconcentration of heavy metals (Cu, Ni, Fe, Co, Mn, Cr and Zn) were observed in Channa punctatus. The results revealed that heavy metals available in water were in the order Fe > Mn > Zn > Co > Ni > Cu = Cr. The accumulation was also observed in tissues such as gills, liver, kidney, muscle and integument. Their pattern of accumulation in muscle was Fe > Zn > Mn > Cu > Cr > Ni > Co. All the heavy metals showed maximum concentration and persistence in gills with the exception of Cu and Co, which showed maximum accumulation in liver and muscle respectively. Fe was the most abundant metal in the water as well as in the fish tissues. Significant (P < 0.01) relations were observed among the metal accumulations in different organs of the fish. The concentration observed was far exceeding the recommended limits of FAO/ WHO.

Keywords : Bioconcentration, Heavy metals, gills, Channa punctatus.

GJRE-C Classification : FOR Code: 090409

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# Uptake of Heavy Metals by Channa Punctatus from Sewage–Fed Aquaculture Pond of Panethi, Aligarh

Mehjbeen Javed <sup>a</sup> & Nazura Usmani <sup>o</sup>

Abstract - Investigations on the bioconcentration of heavy metals (Cu, Ni, Fe, Co, Mn, Cr and Zn) were observed in *Channa punctatus.* The results revealed that heavy metals available in water were in the order Fe > Mn > Zn > Co > Ni > Cu = Cr. The accumulation was also observed in tissues such as gills, liver, kidney, muscle and integument. Their pattern of accumulation in muscle was Fe > Zn > Mn > Cu > Cr > Ni > Co. All the heavy metals showed maximum concentration and persistence in gills with the exception of Cu and Co, which showed maximum accumulation in liver and muscle respectively. Fe was the most abundant metal in the water as well as in the fish tissues. Significant (P < 0.01) relations were observed among the metal accumulations in different organs of the fish. The concentration observed was far exceeding the recommended limits of FAO/WHO.

*Keywords* : *Bioconcentration, Heavy metals, gills, Channa punctatus.* 

#### I. INTRODUCTION

uman activity has continuously disturbed the natural environment, particularly the aquatic ecosystems. The use of heavy metals in industries has lead to the wide spread environmental contamination. Consequently the waste water from industries and also the sewage water from domestic sources containing heavy metals find their way into the nearby water bodies. The aquatic pollution due to heavy metals is of major concern, due to their persistence and accumulative nature. Aquatic animals live in very intimate contact with their environment thus, absorbed heavy metals from the surrounding contaminated water which ultimately affect their health. Among these animal species, fishes are the inhabitants that cannot escape from the detrimental effects of these pollutants (Olaifa et al., 2004) and are therefore very susceptible to physical and chemical changes which may be reflected in their blood components (Wilson and Taylor, 1993). The studies carried out on various fishes have shown that these metals alter the physiological activities and biochemical parameters both in tissues and blood

(Canli, 1995; Basa and Rani, 2003). The accumulation of trace metals in a fish tissue depends mainly on the concentration of the metal in the water and exposure period. It is therefore necessary to examine their distribution in different fish tissues to understand their physiological, toxicological and hygienic effects. The metal once absorbed is transported by the blood to either a storage point, such as bone or to the liver for transportation. If transported by the liver it may be stored there, excreted in bile, or passed back into the blood for possible excretion by kidney or gills or stored in extra hepatic tissues such as fat. Keeping this in view, a study assess the concentration was conducted to accumulated in different organs of Channa punctatus exposed to potentially toxic chemicals in the wastewater. Fish is also provided as a bioindicator of the deteriorating water quality of sewage fed pond.

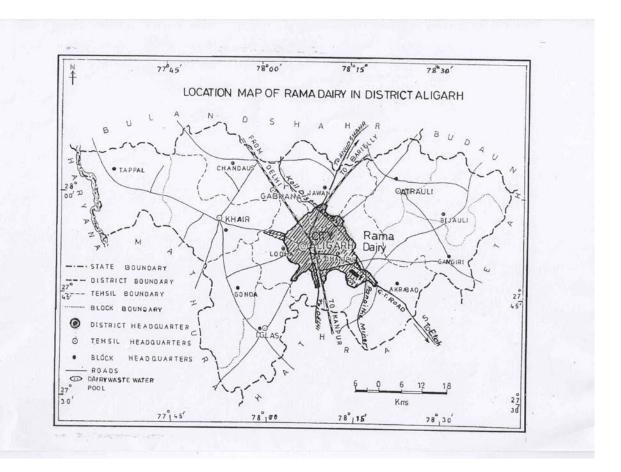
#### a) Description of study area

The study pond is located at Panethi (Latitude 27.88969; Longitude 78.07594), in district Aligarh (Uttar Pradesh), India. This sewage -fed pond is situated at a distance of about 1Km from Dairy products processing factory (Rama Dairy). This factory is now banned for last one year, but in the past the waste water used to reach the pond. Few cold stores are also present nearby. This factory supplied the milk and other processed products in Panethi and around the other regions of Aligarh. The waste water from this factory find its way into the study pond via small streams. This pond also received the domestic waste water of communities living in the area. Fishes thriving in this pond fulfill the need of local peoples living around.

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*Figure 1 :* Location of sewage fed pond (Dairy waste water pool) and Rama Dairy at Panethi, Aligarh.

#### II. MATERIALS AND METHODS

#### a) Collection and analysis of water sample from sewage fed pond

Water was collected in a pre-cleaned and acidified glass bottles. The bottles were immediately brought to the laboratory and acidified with concentrated  $HNO_3$  to pH less than 2.0. Water samples were then analyzed for the presence of heavy metals (Cu, Ni, Fe, Co, Mn, Cr and Zn) according to APHA (2005).

On spot fixation of water was done to measure the dissolved oxygen (D.O). Total solids (T.S), total dissolved solids (T.D.S) and suspended solids (T.S.S) were determined using standard techniques (APHA, 2005).The temperature and pH were recorded at the site using laboratory thermometer (Deluxe, 6) and pH strips (S.D Fine chemicals, 0 - 0.1).

#### b) Collection and processing of fish samples

Fish *Channa punctatus*, (stock = 4, n= 5, Mean length 14.20 cm, Mean weight 35.0 g), were collected from the sewage fed pond. Procured fishes were immediately kept in pre-cleaned polythene bags, sealed and stored in an ice box for further analysis. The present study was conducted to investigate the accumulation of heavy metals (Cu, Ni, Fe, Co, Mn, Cr, and Zn) in various

tissues (gills, liver, kidney, muscle and integument). Tissues were removed and oven dried at 60 °C. The dried samples were ground into fine powder using pestle and mortar, and sieved (0.5 - 1.0 mm). Tissue samples, (0.5g) were digested in 15 ml of solution containing concentrated HNO<sub>3</sub> and HClO<sub>4</sub> (4:1). The digested solution was filtered through Whatmann filter paper (No.42), and washed with distilled water. It was then raised to 50 ml in a volumetric flask (Javed M and Usmani N, 2011).

#### c) Preparation of Blank

Blanks were prepared along with each set of the sample.

#### d) Preparation of standards

Standard solutions for heavy metals were prepared using standard techniques (APHA 2005).

#### e) Instrumentation

The water and fish tissue samples were analyzed for heavy metals Cu, Ni, Fe, Co, Mn, Cr and Zn by Atomic Absorption Spectrometer (Perkin Elmer, AA 800, multiple cathode lamps) with specific cathode lamps for each metal and Nitrous oxide-Acetylene was used as flame. The following analytical conditions of the instrument were used for atomic absorption of these metals.

#### f) Statistical analysis

Samples were taken in triplicates. The values are given as Mean  $\pm$  S.D. The data was subjected to ANOVA. Significant differences among the means was calculated using Duncan's multiple range test (Duncan 1955).

### III. Results and Discussion

The aquatic environment of the sewage fedpond, subjected to many stressful factors, heavy metals are one of the serious pollutants that reach the aquatic habitat and also a matter of concern. For this reason, this work is projected to examine the hazardous effects of heavy metal on one of the most common fish species, *Channa punctatus* in the sewage fed-pond of Panethi.

Table 1 presents the data on physicochemical parameters of sewage-fed pond water. Table 2 and figure 2 shows the mean concentration of metals (mg L<sup>-1</sup>) in water. The heavy metal content in sewage-fed pond water were in the order of Fe > Mn > Zn > Co > Ni > Cu = Cr.

Table 3 revealed concentration of different heavy metals in particular organs of *Channa punctatus.* 

Table 4 and figure 3 present accumulation of particular heavy metals in different organs of *Channa punctatus*.

These results indicate that in general gill was the most affected organ where maximum accumulation of heavy metals takes place followed by muscle, kidney, liver and the integument accumulated the least, and amongst the heavy metals Fe accumulated the most in all tissues.

*Table 1 :* Physicochemical parameters of sewage-fed pond water.

| Parameters             | Water                |
|------------------------|----------------------|
| Temperature            | 30.0°C               |
| рН                     | 7.0                  |
| Dissolved oxygen (D.O) | 5.8mgL <sup>-1</sup> |

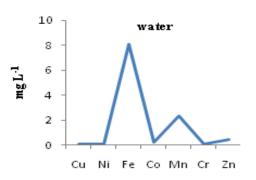
| Total solids (TS)            | 1700mgL <sup>-1</sup>  |
|------------------------------|------------------------|
| Total Dissolved solids (TDS) | 1500 mgL <sup>-1</sup> |
| Total Suspended solids (TSS) | 200 mgL <sup>-1</sup>  |

Values are Mean, (n = 3).

Table 2 : Heavy metal concentrations (mgL-1) in sewage-fed pond water.

| Heavy metals | Water           |
|--------------|-----------------|
| Cu           | 0.07± 0.01      |
| Ni           | $0.08 \pm 0.02$ |
| Fe           | $8.08 \pm 2.88$ |
| Co           | $0.24 \pm 0.02$ |
| Mn           | $2.32 \pm 0.10$ |
| Cr           | $0.07 \pm 0.02$ |
| Zn           | $0.45 \pm 0.03$ |

Values are Mean  $\pm$  S.D, (n= 3)



*Figure 2 :* Heavy metal concentration in sewage-fed pond water, Panethi, Aligarh.

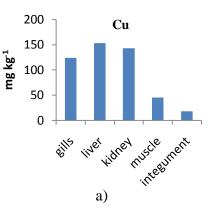
| Heavy metals↓ | Gills                       | Liver                      | Kidney                     | Muscle                     | Integument                |
|---------------|-----------------------------|----------------------------|----------------------------|----------------------------|---------------------------|
| Cu            | 123.80 <sup>d</sup> ±4.12   | 153.33°±7.29               | 143.33°±5.77               | $45.33^{d} \pm 0.57$       | 18.33 <sup>d</sup> ±0.57  |
| Ni            | $30.95^{f} \pm 1.73$        | ND                         | ND                         | $18.33^{f} \pm 0.09$       | 11.33 <sup>e</sup> ±0.57  |
| Fe            | 17609.38 <sup>a</sup> ±4.12 | 14533.13 <sup>a</sup> ±0.5 | $3543.76^{a}\pm0.68$       | 5313.29 <sup>a</sup> ±0.31 | 875.33 <sup>a</sup> ±0.31 |
| Co            | ND                          | ND                         | ND                         | 1.33 <sup>g</sup> ±0.06    | 1.33 <sup>g</sup> ±0.05   |
| Mn            | 1359.51°±0.62               | ND                         | ND                         | 83.28 <sup>c</sup> ±0.06   | 22.31 <sup>c</sup> ±0.57  |
| Cr            | 66.66 <sup>e</sup> ±2.43    | 13.33 <sup>d</sup> ±5.76   | 10.00 <sup>d</sup> ±0.00   | 29.33 <sup>e</sup> ±1.96   | 6.33 <sup>f</sup> ±0.02   |
| Zn            | 1845.22 <sup>b</sup> ±0.57  | 873.31 <sup>b</sup> ±6.06  | 1163.33 <sup>b</sup> ±5.72 | 319.29 <sup>b</sup> ±0.18  | 257.11 <sup>b</sup> ±0.10 |

Values are Mean  $\pm$  S.D, (n= 3), ND= not detected, Values are expressed in mgkg-1.dry weight Means with similar letters in a column are statistically similar at P > 0.01

| Heavy metals↓ | Gills                       | Liver                      | Kidney                     | Muscle                     | Integument                |
|---------------|-----------------------------|----------------------------|----------------------------|----------------------------|---------------------------|
| Cu            | 123.80 <sup>c</sup> ±4.12   | 153.33 <sup>a</sup> ±7.29  | 143.33 <sup>b</sup> ±5.77  | 45.33 <sup>d</sup> ±0.57   | 18.33 <sup>e</sup> ±0.57  |
| Ni            | 30.95 <sup>a</sup> ±1.73    | ND                         | ND                         | 18.33 <sup>b</sup> ±0.09   | 11.33 <sup>c</sup> ±0.57  |
| Fe            | 17609.38 <sup>a</sup> ±4.12 | 14533.13 <sup>b</sup> ±0.5 | 3543.76 <sup>d</sup> ±0.68 | 5313.29 <sup>c</sup> ±0.31 | 875.33 <sup>e</sup> ±0.31 |
| Со            | ND                          | ND                         | ND                         | 1.33 <sup>a</sup> ±0.06    | 1.33 <sup>a</sup> ±0.05   |
| Mn            | 1359.51 <sup>a</sup> ±0.62  | ND                         | ND                         | 83.28 <sup>b</sup> ±0.06   | 22.31 <sup>e</sup> ±0.57  |
| Cr            | 66.66 <sup>a</sup> ±2.43    | 13.33 <sup>c</sup> ±5.76   | $10.00^{cd} \pm 0.00$      | 29.33 <sup>b</sup> ±1.96   | 6.33 <sup>d</sup> ±0.02   |
| Zn            | 1845.22 <sup>a</sup> ±0.57  | 873.31 <sup>e</sup> ±6.06  | 1163.33 <sup>b</sup> ±5.72 | 319.29 <sup>d</sup> ±0.18  | 257.11 <sup>e</sup> ±0.10 |

Table 4: Concentrations of particular heavy metal in different organs of Channa punctatus.

Values are Mean  $\pm$  S.D, (n= 3), ND= not detected, Values are expressed in mgkg-1.dry weight Means with similar letters in a row are statistically similar at P > 0.01



Fe

kidney

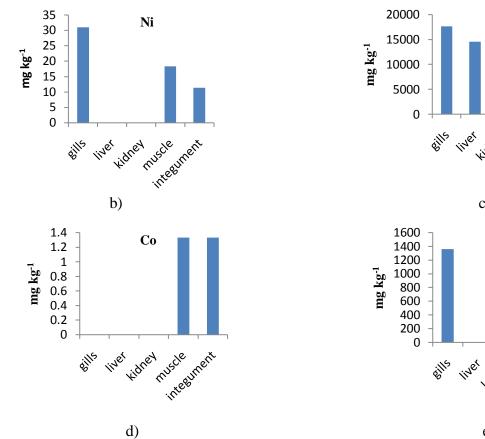
c)

Mn

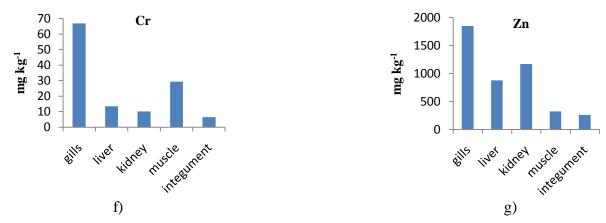
kidney nuscle unent

e)

nuscle interit







*Figure 3 :* a,b,c,d,e,f,g showed mean metal (Cu, Ni, Fe, Co, Mn, Cr and Zn) concentrations (mgkg<sup>-1</sup>.dw) in gills, liver, kidney, muscle and integument of *Channa punctatus.* 

Fe was the most abundant heavy metal in all tissues of *Channa punctatus*, but its highest value was observed in gills followed by liver > muscle > kidney > integument (Table 4) Studies reported in Oreochromis niloticus and Lates niloticus (Mohamed, 2008), Oreochromis mossambicus (Robinson and Avenant-Oldewage, 2006) Liza aurata, Mugil cephalus, Liza ramada (Uysal et al., 2008) also revealed the maximum accumulation of Fe in gills. Highest accumulation in gills indicates that these are the organs which always remain in direct contact with the surrounding water. However in other studies the highest accumulation seen in organs such as liver in the fishes Clarias gariepinus (Osman et al., 2010) and Tinca tinca (Selda Tekin et al., 2005). In the pond water also the Fe concentration was maximum, therefore maximum uptake of this metal takes place by the fish tissues.

Fe accumulation was followed by Zn in the present study where the values were observed to be highest in gills followed by kidney > liver > muscle > and least in integument. Other scientist also reported the highest concentration in gills of Channa punctatus (Vineeta Shukla et al., 2005). These high levels in gill tissue can possibly due to the fact that they are the main sites for Zn uptake, particularly in fresh water fish and due to the large surface area that is in contact with environmental water and the very thin membrane separating the external and internal media of the animal. The large surface area of gills in Channa punctatus (Karuppasamy, 2000) may be favour for metal uptake. Zn content in gills of investigated species was comparable to Labeo dyocheilus (Yousafzai et al., 2010). Other workers, however noticed the highest concentration in organs such as liver of Channa punctatus (Murugan et al., 2008) and Clarias gariepinus (Osman et al., 2010), testes of Oreochromis niloticus and Lates niloticus (Mohamed, 2008) and integument of Labeo dyocheilus and Wallago attu (Yousafzai et al., 2010). In this study, it was also observed that Zn content in liver was higher than muscle. The lower Zn content in muscle may be because the excessive Zn in muscle was transferred to other fish organs when exposed to Zn contaminated system (Madhusudan et al., 2003). This deloading ability of fish has been reported to be advantageous to fish consumers (Murugan et al., 2008). The permissible limits for Zn set by WHO/FAO (1989) is 40 ppm which is much less than the values observed during the study (Table 5).

In the present study, Mn accumulation was significant in tissues and showed accumulation in the order of gills > muscle > integument. It was not observed in liver and kidney. Though, in water its availability was considerably high. Studies reported in Tinca tinca (Selda Tekin et al., 2005), Oreochromis mossambicus (Robinson and Avenant-Oldewage, 2006), Clarias gariepinus (Osman et al., 2010) and Labeo rohita, also revealed the highest concentration of Mn in gills (Javed and Usmani, 2011). However, other workers reported the highest concentration in organs such as kidney and muscle of Channa punctatus and Clarias gariepinus respectively (Javed and Usmani, 2011). However in other studies the Mn content reported in different tissues was much lower than the present study. Mn is an essential micronutrient (Dallas and Day, 1993) and does not occur naturally as a metal in aquatic ecosystems, but is found in form of various minerals and salts. According to the Department of Water Affairs and Forestry (1993), the main route of Mn adsorption occurs through the respiratory and gastrointestinal tracts. The adsorption of Mn in the digestive tract is inversely related to Ca<sup>++</sup> levels in the diet of organism. The permissible limits for Mn set by WHO(1985) is 0.01ppm which is well below the accumulation observed during the study (Table 5).

Copper exhibited highest content in liver and lowest in integument of the investigated species and the pattern observed was liver > kidney > gills > muscle > integument. Other workers also reported the highest accumulation of Cu in liver of fishes *Oreochromis mykiss* and *Cyprinus carpio* (De Boeck et al., 2004), *Tilapia nilotica* (Abdel-Baki et al., 2011), *Oreochromis niloticus* (Mohamed, 2008), *Wallago attu* and *Labeo* 

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dyocheilus (Yousafzai et al., 2010). According to Stokes (1979), liver and kidney have Cu bioaccumulation properties, with the accumulative capacity much greater in liver than in kidney which is evident from the present study (Table 4). Cu accumulation exhibited in liver corroborates to the findings in Cyprinus carpio (De Boeck et al., 2004), Oreochromis mossambicus (Robinson and Avenant-Oldewage, 2006). Cu content in muscle corroborates to the Heterotis niloticus, Clarias gariepinus (Anim et al., 2010), Wallago attu (Yousafzai et al., 2010). According to Stokes (1979) fish muscle has poor accumulative properties, with low concentration of Cu found in the muscles, even systems containing high Cu levels. Present study reports low levels of Cu in water (Table 2). However, other scientists confirmed highest levels in organs such as kidney of Channa punctatus, *Clarias gariepinus* and *Labeo rohita* (Javed and Usmani, 2011), gills of Channa punctatus(Vineeta Shukla et al., 2005) and Lithognathus mormyrus (Uysal et al., 2008). In the present study the content of Cu observed in integument is similar to our earlier findings in Clarias gariepinus and Labeo rohita (Javed and Usmani, 2011). The amount of Cu accumulation observed during the present study is little higher than the permissible limits set for Cu by WHO/FAO (1989) which is 30 ppm (Table 5).

In the present study the concentration of Cr and Cu in water was exactly similar (Table 2), but fish showed different response to these metals (Table 3). It indicates that even in lower amounts Cu has more absorptive and accumulative capacity than Cr under similar natural environment. Cr present in highest amounts in gills followed by muscle and least was in integument. While liver and kidney showed insignificant accumulations. Various studies conducted on Cr also noticed highest concentration in gills of Labeo dyocheilus and Wallago attu (Yousafzai et al., 2010). However, the highest concentration was also reported in other organs such as in kidney of *Clarias gariepinus* and integument of Labeo rohita (Javed and Usmani, 2011), liver of *Clarias gariepinus* (Osman et al., 2010), kidney of Tilapia nilotica (Abdel-Baki et al., 2011), liver of Oreochromis mossambicus (Robinson and Avenant-Oldewage, 2006). The observation that was made for

Cr accumulation in kidney was comparable to *Clarias gariepinus* and *Labeo rohita* (Javed and Usmani, 2011). Concentration in integument corroborates to *Channa punctatus* (Javed and Usmani, 2011). Duffus (1980) and Paasivirta (1991) both regard Cr in its salt form as highly bioaccumulative at high concentrations, and partially dangerous.

Ni occupied the sixth position as far as accumulation was concerned. The order of nickel accumulation observed during the study was gills > muscle > integument. It was not detected in liver and kidney. Other workers also revealed the highest levels of Ni in gills of fishes Cyprinus carpio (Vinodhini and Narayanan, 2007), Catla catla and Heteropneustes fossilis (Abida et al. 2009), Wallago attu (Yousafzai et al. 2010). Ni has a similar chemical behaviour to Fe and Co and commonly substitutes for Fe in ferromagnesian minerals. Gold fish (Carassius auratus) that died during immersion in solutions containing more than 35 mgL<sup>-1</sup> Ni showed elevated concentration in tissues, however, most of nickel was washed off with water, and it is not clear if accumulation occurred after death (Kariya et al.1968). Concentrations of Ni in water from natural occurences are only likely to be of health concern in environments where pH is less than 4.5. The amount of Ni accumulation observed during the present study is within the permissible limits set for Ni (70 - 80 ppm) by USFDA (1993b).

Co was the least accumulated metal in tissues of *Channa punctatus*. Its accumulation was noticed only in muscle and integument while in gills, liver and kidney it was untraceable. Co was also not detected in gills and kidney samples of fishes *Clarias gariepinus, Cyprinus carpio* and *Oreochromis niloticus* (Adeyeye et al., 1996). Ca<sup>2+</sup> competition and dissolved organic matter complexation were the most important factors preventing Co<sup>2+</sup> from binding at the gills in natural water.

To summarize, these results indicate that the fish *Channa punctatus*, as a representative fish species of sewage-fed aquaculture pond, can be a useful vertebrate bio-indicator organisms of heavy metals contamination in water. This species is also a highly sensitive type to heavy metal pollution in the environment.

|              |  | , ,                                    | 0        | <i>,</i> ,                            |
|--------------|--|--|----------|---------------------------------------|
| Heavy metals | Muscle (mgKg <sup>-</sup><br><sup>1</sup> .dw) | Integument<br>(mgKg <sup>-1</sup> .dw) | Average* | Permissible limits<br>(ppm)           |
| Cu           | 45.33  | 18.33                                  | 31.83    | 30 (WHO/FAO 1989)                     |
| Ni           | 18.33  | 11.33                                  | 14.83    | 70-80(USFDA 1993b)                    |
| Fe           | 5313.29  | 875.33                                 | 3094.31  | · · · · · · · · · · · · · · · · · · · |
| Co           | 1.33   | 1.33                                   | 1.33     |                                       |
| Mn           | 83.28  | 22.31                                  | 52.79    | 0.01 (WHO 1985)                       |
| Cr           | 29.33  | 6.33                                   | 17.83    | -                                     |
| Zn           | 319.29   | 257.11                                 | 288.2    | 40(WHO/FAO 1989)                      |

*Table 5*: Permissible limits of heavy metals set by various food agencies in fish and fishery products.

\* At times integument is also consumed with fish muscle therefore, average is taken. Blank cells indicate that no citable information is available.

## IV. Conclusions

This study was carried out to provide information on toxic heavy metal concentrations in *Channa punctatus* from sewage- fed aquaculture pond, India and potential health risk for local population due to their consumption. The majority of heavy metal concentrations in the fish samples analyzed were exceeding the permitted limits set by various authorities and will pose health risks for the local population due to high consumption of fish.

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