# Human health aspects of certain metals in tissue of the African sharptooth catfish, *Clarias gariepinus*, kept in treated sewage effluent and the Krugersdrift Dam: Chromium and mercury

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## Abstract

Chromium and mercury concentrations in water, sediment and fish tissue were studied to assess the health risks concerned when such fish are consumed. Metal concentrations were studied in the liver, kidney and muscle tissues of the African sharptooth catfish, *Clarias gariepinus*, kept in treated sewage effluent from a biofilter treatment plant and in the Krugersdrift Dam, Bloemfontein, South Africa as well as in the water and sediment of the mentioned localities.

Chromium concentrations were noticeably higher in the livers and kidneys than in the muscle tissue. In contrast, Hg concentrations were higher in the muscle tissue than in the livers and kidneys. However, no set seasonal patterns could be established regarding the incidence of these elements in both habitats. The occurrence of Hg in the water of both habitats was very low and could therefore not be considered harmful or toxic to aquaculture. Chromium concentrations showed a fluctuating occurrence and exceeded international limit values during some months. The Hg and Cr concentrations found in the muscles of *C. gariepinus*, kept in treated domestic effluent can be a health hazard to consumers if fish is consumed in excess. Only gutted fish with the gills removed would be recommended for intake due to the Hg and Cr concentrations found in the kidney and liver tissue.

## Introduction

Loading of aquatic habitats with non-degradable, non-nutritious, cumulative pollutants such as Hg, Pb and As can result in undeniably complex alterations of numerous trophic levels. These effects can last for centuries and studies spanning the globe, document the ubiquity of such contamination (Sorenson, 1991). Industrial and/or sewage effluents are major sources of metal poisoning in fish in Africa (Greichus et al., 1978).

The utilisation of human and animal waste in fish dams has already been known for decades (Feachem et al., 1978), but little information regarding the human health aspects of fish living in treated domestic effluent is currently available. The prevalence and accumulation of a series of metals in the tissue of local fish fauna as well as the accompanying health risks for the consumer thereof, were investigated by Van den Heever and Frey (1994). The primary goal of the study was to determine whether treated sewage effluent could be suitable for the culture of fish for safe human consumption. These usually abundant and nutritious waters are normally discharged into natural water sources without further use. In South Africa especially, it must be endeavoured to improve the quality of available water and to make use of treated effluents. A secondary goal of the study was to compare the pollution status of the Krugersdrift Dam, as a natural water source, and treated sewage effluent discharged from a biofilter treatment plant.

This paper specifically deals with the occurrence of Cr and Hg in treated sewage effluent and natural dam water and the extent of bio-accumulation of these metals in the tissue of the African sharptooth catfish (*Clarias gariepinus*). This fish was chosen as experimental species, as research conducted by Prinsloo et al. (1989) on *C. gariepinus* in maturation ponds in Lebowa, indicated that this fish thrives on the high organic content of ponds.

## Materials and methods

### Water sampling, preparation and analysis

Water samples were collected monthly from January to December 1991 according to the method of Watling (1981). Sampling bottles were washed and left to stand in a 5% Decon 75 non-metallic detergent solution for 48 h. After rinsing with distilled water, the bottles were soaked in 50% HCl for 24 h. Finally, the items were washed with distilled water and stored in 1% $HNO_3$ prior to their use.

Pre-cleaned high density polyethylene bottles with a 2*ℓ* capacity were used for sampling. Samples were acidified on site with 10 m*ℓ* concentrated nitric acid (SAARCHEM) (AR) per litre sample and the bottles sealed with high density polyethylene screw-tops. Samples were transported to the laboratory in a sample case at 15°C and stored in high density polyethylene bottles until they were analysed. The collected samples were divided into three subsamples and then filtered through a 0.45 µm membrane filter into a 335 m*ℓ* high density polyethylene bottle with a polyethylene top. The first 20 m*ℓ* of the filtered samples were discarded. A separate subsample of the 0.45 µm filtered water sample was collected in a test tube for analysis. The samples were analysed according to the method of Kempster (1986).

The Hg concentrations of water samples collected from each locality were determined by means of a Varian Spectra 30 atomic absorption spectrometer (Watling, 1981). Chromium concentrations were determined by means of an ARL inductively coupled plasma (ICP) emission spectrometer (Model 3410). Various physical properties of the water of the two habitats such as pH, hardness, conductivity, redox potential and temperature variations were also determined monthly with a HANNA water test meter in order to

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give possible explanations for unexpectedly high or low concentrations of metals in solution.

### Collection, preparation and analysis of sediment samples

For the collection of uncompacted sediment samples, a polyvinyl chloride pipe of 50 mm dia. was manually pushed into the sediment to a depth of 300 mm whereafter the top end was covered with a thick plastic bag. The pipe was then removed from the water and the rod-shaped samples placed into plastic bags. Each sample was divided into 3 lengths of 100 mm, sealed in polyethylene bags and transported to the laboratory in cool-bags filled with ice (Watling, 1981). Before drying, the samples were exposed to a temperature of -20°C for 24 h (Bruwer et al., 1985) and thereafter passed through a 1 mm nylon sieve. Finally they were placed on a filter-disk between sheets of filter-paper and left to dry at room temperature for 7 d (Watling, 1981).

Sample dissolution for both metals was carried out according to the method of Watling (1981). Samples were chemically analysed by means of AA spectrophotometry for Hg (Watling, 1981) and ICP spectrometry (Kempster, 1986) for Cr.

### Collecting and keeping of fish

Healthy adult catfish were netted from a natural pond in the Brandfort district (Free State) near Bloemfontein, South Africa, in December 1990. The fish were kept in a polyethylene pond with a capacity of 2 150 $\ell$ for 2 weeks to rid the tissue of any foreign substances such as bacteria. The pond system had a domestic tap-water flow-through of approximately 20$\ell\cdot h^{-1}$. During this phase of the study the fish were fed commercial breakfast food (Weet-bix) with high protein content (0.116 $g\cdot g^{-1}$) every second day. Fish were fed in the ratio of 0.6 $g\cdot kg^{-1}$ body mass.

After the above cleansing process, 30 fishes were transported to the first maturation pond of the Bloemspruit Sewage Works and the inlet of the Krugersdrift Dam respectively (see Fig. 1 in Van den Heever and Frey, 1996). Here they were placed in cube-shaped 10 x 50 mm galvanised mesh cages, each with a capacity of 8 $m^3$. The cages were firmly positioned on the bottom in such a way that the water could flow through freely. Approximately 20 cm of each cage protruded above the water allowing the fish to surface for air if necessary. The fish were not fed in the cages as sufficient food was found in the stomachs of individual specimens after periodic examination. A random sample of 2 fishes per month showed stomach fullness of 50%. The number of fish that died in captivity was negligible (3%).

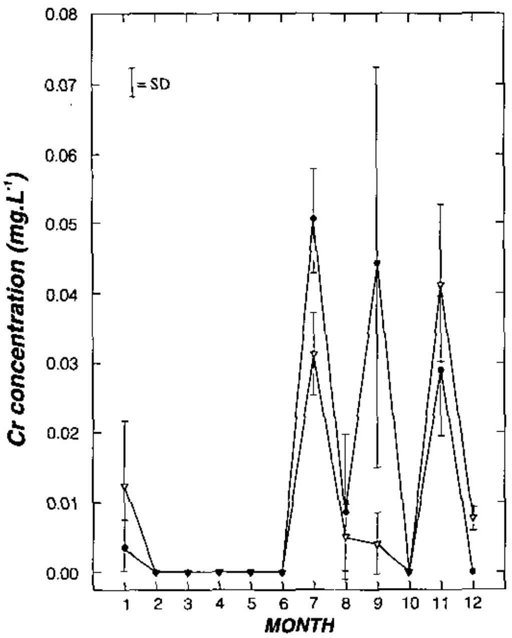

**Figure 1**
*Concentrations of Cr and standard deviations from the mean of treated water from the Bloemspruit Sewage Works (•—•) and water from the intake of the Krugersdrift Dam (▽—▽) measured from January 1991 (month 1) to December 1991 (month 12)*

### Tissue sampling, preparation and analysis

Tissue sampling, preparation and analysis for Cr were carried out as described by Van den Heever and Frey (1994). A Varian Spectra 30 atomic absorption spectrophotometer was used to determine the Hg concentration in tissue samples (Watling, 1981).

A preliminary study was carried out at the start of the experiment in order to establish to what extent uniform concentrations of metals were present in tissues of the group. Insignificant low concentrations were found and are therefore not mentioned.

## Results

### Water analyses

The mean concentrations of Cr with the standard deviations from the mean of treated effluent from the Bloemspruit Sewage Works and water from the intake of the Krugersdrift Dam measured from January 1991 to December 1991, are shown in Fig. 1. Means and standard deviations were calculated for 9 samples collected at each locality during each month.

Concentrations of Cr in the water of these two localities were insignificantly low during the first six months of 1991. However, during the latter half of 1991 large monthly variations were found (Fig. 1). In spite of this phenomenon no significant differences were found between the average Cr concentrations (12.4$\mu g\cdot\ell^{-1}$) for treated sewage effluent and (9.24$\mu g\cdot\ell^{-1}$) for natural dam water. The mentioned values were also lower than those of Watling and Watling (1982a-f) determined for 12 Cape rivers.

Concentrations of Hg in treated sewage water and in natural dam water were lower throughout than the detection limit of 0.001 $mg\cdot\ell^{-1}$. Because of insignificantly low values and to avoid an unnecessary increase in the cost of the project, the actual concentrations were not determined.

### Sediment analyses

The average Cr concentration for sediment samples of the Krugersdrift Dam was higher than that of treated sewage effluent. In contrast, the average Hg concentrations in the sediment of treated sewage effluent and natural dam water were found to be very similar (Table 1).

### Chromium and mercury concentrations in fish tissue

Chromium concentrations found in the different tissue types of *Clarias gariepinus* are shown in Fig. 2. During December the

**TABLE 1**

**CONCENTRATIONS OF Cr ($mg \cdot g^{-1}$ DRY MASS) AND Hg ($\mu g \cdot g^{-1}$ DRY MASS) IN SEDIMENT SAMPLES COLLECTED MONTHLY OVER A 12 MONTH PERIOD DURING 1991 FROM THE FIRST MATURATION POND OF THE BLOEMSPRUIT SEWAGE WORKS AND THE KRUGERSDRIFT DAM**

| | **Maturation pond** | | | **Krugersdrift Dam** | | |
|---|---|---|---|---|---|---|
| **Element** | **Range** | **$\bar{x}$** | **SD** | **Range** | **$\bar{x}$** | **SD** |
| Cr | 3.40 - 8.60 | 4.31 | 0.85 | 2.02 - 4.83 | 3.61 | 0.84 |
| Hg | 0.052 - 0.096 | 0.08 | 0.005 | 0.041 - 0.051 | 0.047 | 0.003 |

$\bar{x}$: Mean concentration
SD: Standard deviation from the mean value

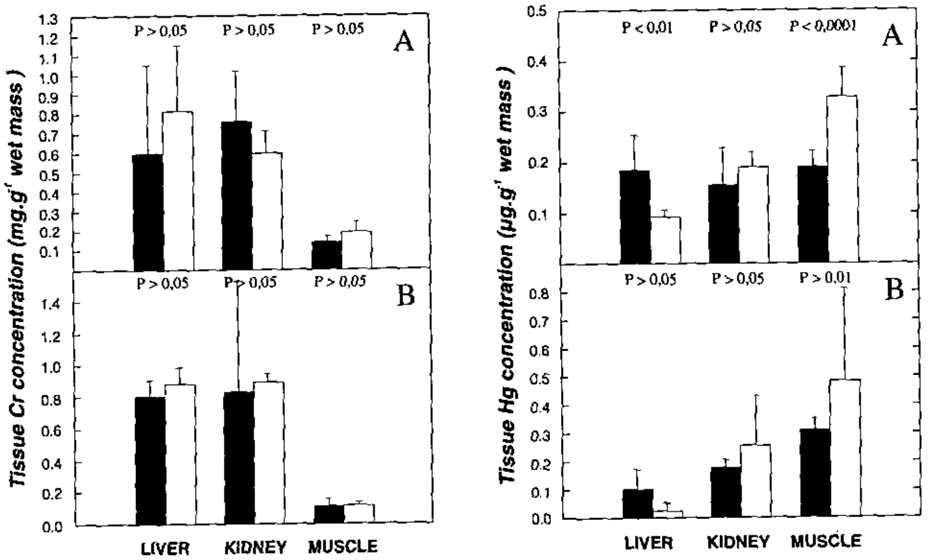

***Figure 2***

*Concentrations of Mn and Fe measured in tissue samples of* Clarias gariepinus *kept in treated sewage water (■) and natural dam water (□)*

*A: Fish kept from January to June 1991 whereupon tissue analysis was carried out*

*B: Fish kept from January to December 1991 whereupon tissue analysis was carried out*

highest concentrations of Cr were measured in the liver (0.888 $mg \cdot g^{-1}$ wet mass) and in the kidneys (0.894 $mg \cdot g^{-1}$ wet mass) of the fish. The lowest values of Cr were found in the muscle tissue with the June concentration for fish in natural dam water slightly higher than that for December. The average June-December concentrations of Cr in muscle tissue were very similar, i.e. 0.125 $mg \cdot g^{-1}$ (wet mass) for fish in treated sewage water and 0.151 $mg \cdot g^{-1}$ (wet mass) for fish in natural dam water.

The average Hg concentration in the muscle tissue of the fish in the Krugersdrift Dam was significantly higher in June as well as in December than that measured in treated sewage effluent (Fig. 2). In June a noticeably bigger difference was found between the concentrations of this metal in the muscle tissue. No significant differences were found in the concentrations of Hg in the kidneys of fish sampled from the respective habitats.

In contrast with the kidney and muscle tissues, the concentration of Hg in the liver during June was significantly higher in treated sewage effluent than in natural dam water. Concentrations in the livers of fish of both habitats also seem to be higher in winter than during the summer months.

The values in Fig. 2 compared favourably with concentrations of Hg measured in the muscle tissue of other fish species in the literature. Potter et al. (1975) measured 0.25 $\mu g \cdot g^{-1}$ (wet mass) for *Cyprinus carpio* in a man-made reservoir. Catfish caught in the Mississippi River showed an Hg concentration of 0.281 $\mu g \cdot g^{-1}$ (wet mass) after analysis (Hartung, 1974).

## Discussion

The average Cr concentrations of treated sewage effluent from the Bloemspruit treatment plant and the water of the Krugersdrift Dam calculated for the 12 months of 1991, were 12.4 and 9.24 $\mu g \cdot \ell^{-1}$ respectively. These are considerably higher than values reported by Watling and Watling (1983a; b) for Cr in various South African

**TABLE 2**
**A COMPARISON BETWEEN CONCENTRATIONS (μg·$\ell^{-1}$) OF Cr AND Hg MEASURED FOR SOME SOUTH AFRICAN RIVERS AND FOR WATER OF THE KRUGERSDRIFT DAM AND TREATED SEWAGE EFFLUENT OF THE BLOEMSPRUIT SEWAGE WORKS**

| **River** | Vaal River | | 12 Cape rivers | Transkei rivers | Maturation dam | Natural dam |
|---|---|---|---|---|---|---|
| **Period** | Oct 1981 | Sept 1982 | May 1975-Aug 1981 | Sept 1980-Jan 1982 | Jan 1991-Dec 1991 | Jan 1991-Dec 1991 |
| **Source** | Rand Water Board (1982) | | Watling (1982a-f) | Du Preez (1985) | | |
| **Hg** | 1.00 | 1.00 | 0.03 | 26.0 | <1.00 | <1.00 |
| **Cr** | 60.0 | 60.0 | 0.80 | 5.00 | 12.40 | 9.24 |

rivers. However, the average values obtained for Cr in this study are again notably lower than the recommended standard of 50 μg·$\ell^{-1}$ (USEPA, 1976; WHO, 1984) for total Cr. The latter standard was set to provide for Cr in the highly toxic hexavalent form (WHO, 1984). Despite the relatively low average concentrations obtained for Cr in the water of the habitats investigated in this study, individual concentrations measured in both localities for some months during the second half of 1991, were in the order of 40 to 50μg·$\ell^{-1}$. The lower pH and total hardness values of the water measured during the second half of 1991, especially in the Krugersdrift Dam (pH of 7.4 and total hardness of 119.2 mg·$\ell^{-1}$ respectively), apparently contributed to the higher concentrations of Cr in the water to near unacceptable levels. During the first half of the year, the corresponding values were 7.9 for pH and 105.7 mg·$\ell^{-1}$ for total hardness. In addition the World Health Organisation (WHO, 1984) claims that in the case of aerated and chlorinated water with a total Cr content of 100 μg·$\ell^{-1}$, as much as 40 to 66% of the total Cr released by sediment can be in the form of Cr(VI). It can therefore be deduced that shallow dam water with a high total Cr and oxygen content, as in the case of the Krugersdrift Dam, could pose an extremely high toxic state in terms of Cr(VI). Further possible reasons for the incidence of higher concentrations of dissolved Cr in the Krugersdrift Dam and in treated sewage effluent during the second half of 1991 are dumping of waste by developing communities along the upper reaches of the Modder River, drainage of fertiliser from farm-lands on the banks of the river and Cr-containing effluent from electroplating plants in the catchment area. Illegal dumping of damaging substances into the river cannot be overlooked.

Snyder et al. (1975) estimate that the food intake of a person with a mass of 70 kg should contain an average of approximately 150 μg Cr per day. The average Cr concentration of 125.8 μg·g$^{-1}$ (wet mass) in the muscle tissue of *C. gariepinus* in treated sewage effluent and 151.9 μg·g$^{-1}$ (wet mass) in natural dam water can in view of the above-mentioned intake pose a health risk especially due to the Cr(VI) content of muscle tissue being an unknown factor. However, the higher Cr concentrations measured in the water of both localities during the second half of 1991, were in no way reflected in the tissue of *C. gariepinus* during December 1991. It would appear that *C. gariepinus* accumulated this metal in the muscle tissue up to specified concentration levels only, regardless of the concentration in the water.

Symptoms relating to the excessive intake of Cr by man are not yet known (Carson et al., 1991). According to the National Academy of Sciences (NAS, 1980) an intake of 1 μg Cr(III) per day is necessary to maintain a normal glucose metabolism in the body. It is therefore considered safe to take small quantities of total Cr daily. Davids and Lieber (1951) also reported on a family of four individuals drinking water with a concentration of 450 μg·$\ell^{-1}$ over a period of three years without showing any symptoms of Cr poisoning. Studies conducted by MacKenzie (1958) on rats also showed that an intake of water with a Cr concentration of 450 to 2 500 μg·$\ell^{-1}$ had no toxic effects even after a year.

The concentration of Cr(VI) in the water of the Krugersdrift Dam and the treated sewage effluent of the Bloemspruit Sewage Works is still an unknown factor. Therefore the danger of Cr pollution in these localities cannot be deemed insignificant. At least it can be stated that treated sewage effluent appeared not to pose a bigger health risk than the water of the Krugersdrift Dam. The more stable physical parameters of treated sewage effluent, e.g. pH and hardness, could ensure that increases of Cr up to unacceptable levels will not occur. It is, however, suggested that the authorities concerned with health and environmental conservation should urgently pay attention to the prevention of Cr pollution in the natural waters of the Free State region. The total Cr content in freshwater fish should be regulated by means of legislation.

The Hg concentrations in water samples taken from both localities were lower than 1 μg·$\ell^{-1}$ which is much less than the recommended level of 5 μg·$\ell^{-1}$ for drinking water as suggested by the South African Bureau of Standards (SABS, 1984). The WHO (1984), however, recommends that only 1 μg·$\ell^{-1}$ Hg should be allowed in drinking water seeing that an intake of 2$\ell$ of water a day at the mentioned concentration would result in approximately 10% of the Hg tolerance of man, based on the Hg being in the methylated form. In contrast, the USEPA (1976) recommends a maximum concentration of 2 μg·$\ell^{-1}$ for domestic drinking water and 0.05 μg·$\ell^{-1}$ for freshwater sources supporting aquatic life. The concentrations of both the above-mentioned organisations are much lower than the 20 μg·$\ell^{-1}$ allowed for treated sewage effluent by South African legislation (South Africa, 1984).

Although the water of the Krugersdrift Dam and that of the maturation pond of the Bloemspruit Sewage Works both met the criteria set by the above-mentioned organisations regarding Hg concentrations, Hannerz had already proved in 1968 that algae and water plants accumulated Hg by means of surface absorption where concentrations as low as 100 μg·$\ell^{-1}$ were found in the water. McKim (1976) also found that fish accumulated more than

$0.5 \mu g \cdot g^{-1}$ Hg in their tissues over a period of 20 to 48 weeks in water with a methyl mercury concentration which varied between 0.08 and 0.0030 $\mu g \cdot \ell^{-1}$. Fish which were kept in water with 0.8 to 0.41 $\mu g \cdot \ell^{-1}$ Hg, died within three months after exposure (Mount, 1974).

From the results obtained during the course of this study, it can be deduced that the low Hg levels in the water of both the sampling localities were not dangerous to either fish or man. However, if the Hg concentrations in the Krugersdrift Dam increase, aquatic life could be affected adversely. The incidence of low Hg levels in water can be very misleading in view of the fact that this highly toxic metal can accumulate to high levels in tissue. This fact can be supported by the relatively high Hg concentrations found in the muscle tissue of fish in comparison with that of liver and kidney tissue (Fig. 2). The lowest concentrations of methyl mercury in blood and hair associated with toxic symptoms were, according to epidemiological data in *Methyl Mercury in Fish* (Anonymous, 1971), calculated to be 0.2 $\mu g \cdot g^{-1}$ and 60 $\mu g \cdot g^{-1}$ respectively. These values correlate with the chronic exposure of approximately 300 $\mu g \cdot 70$ kg body $mass^{-1} \cdot d^{-1}$. To ultimately assure the safe intake of Hg, the latter level was decreased with factor 10 to 30 $\mu g \cdot person^{-1} \cdot d^{-1}$ by the above-mentioned source. This source also emphasised that if all other food sources as well as water intake was excluded and only fish eaten, the maximum daily consumption of fish containing 0.5 $mg \cdot kg^{-1}$ Hg should not exceed 60 g (420 $g \cdot week^{-1}$). The latter level is also suggested by the Food and Drug Administration (FDA, 1974) for all living organisms utilised by man as food source. The USEPA (1976) also considers fish fit for human consumption when the water in which the fish are kept does not exceed the Hg level of 0.05 $\mu g \cdot \ell^{-1}$.

Although the average Hg concentrations in the muscle tissue of fish in treated sewage effluent as well as in natural dam water were lower than the limit of 0.5 $mg \cdot kg^{-1}$ set by the FDA (1974), the average value of 0.406 $\mu g \cdot g^{-1}$ measured in the muscle tissue of fish in the Krugersdrift Dam was alarming.

## Conclusions

No set seasonal patterns could be established regarding the incidence of Cr and Hg in the tissue of fish kept in treated effluent water and natural dam water. The Hg concentrations in the kidneys were higher for fish in the Krugersdrift Dam than in the treated sewage effluent. Higher concentrations of Hg were measured in the livers of fish kept in treated sewage effluent.

Hg and Cr concentrations in the muscle tissue of *Clarias gariepinus* were inclined to reach the maximum permissible level, especially in the Krugersdrift Dam. The relatively high Hg concentrations in the muscle tissue of catfish in the Krugersdrift Dam, as well as those of Cr in fish of both localities, posed a possible health risk to man if the fillets of the fish were eaten. The relatively high Cr concentrations could possibly be attributed to electroplating plant effluent which had not been completely purified and not been discharged into the sewage works of the Bloemfontein City Council, but had reached the dam via other routes. This study revealed that the total Cr content of water and muscle tissue of fish from both habitats contained unknown quantities of Cr(VI) which could pose a health risk. Consumption of fish from both habitats (treated sewage effluent and natural dam water) was equally risky. Concentrations of trace elements found in the livers and kidneys of *C. gariepinus* in both habitats may well pose a health risk when eaten. Human consumption of these organs is not recommended.

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