

# The use of cestode parasites from the largemouth yellowfish, *Labeobarbus kimberleyensis* (Gilchrist and Thompson, 1913) in the Vaal Dam, South Africa as indicators of heavy metal bioaccumulation

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## Abstract

Twenty Largemouth yellowfish *Labeobarbus kimberleyensis* (Gilchrist and Thompson, 1913) were collected in the Vaal Dam in the vicinity of RAU Island (Groot Eiland). Muscle-, spinal cord- and liver tissue were removed from each fish and the intestine opened to obtain *Bothriocephalus acheilognathi*. The tissues were then digested in a microwave digester and the metal concentrations were determined in each tissue with an ICP-MS. The results indicate that the worms have potential use as bioindicators, because in 8 elements (lithium, beryllium, manganese, selenium, mercury, thallium, lead and uranium) out of the 23 elements measured the cestodes have accumulated the highest metal concentrations, and in 7 elements (chromium, iron, zinc, molybdenum, cadmium, tin and barium) out of 23 elements the cestodes had the second highest metal accumulation recorded.

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## 1. Introduction

The term “bioaccumulation indicator” is used to indicate the difference between ecological or pollution indicators, and the presence or unexpected absence of which gives an indication of the habitat or environmental quality. The organisms accumulate polluted substances from their surroundings or food or both, capturing them in their bodies, so that when tissues are analyzed an indirect estimate of prevailing environmental concentrations of these substances may be made (Hellawell, 1986). Indicator organisms that have been used successfully for bioaccumulation studies in South Africa are macrophytes, crabs, and fish (Kotze

et al., 1999; Sanders et al., 1999; Saadi et al., 2002). The tissues, organs or material analyzed are useful because it gives the effect of long-term (chronic) or short-term (acute) exposures to the toxicants. Other sources of metal accumulation were studied to include the water body, invertebrates and the soil (Slabbert, 1996).

Freshwater ecosystem research in South Africa is increasing due to the awareness that water demand is increasing at a pace that in the near future, portable water supply will not be able to meet requirements (Roux et al., 1993). Extensive bioaccumulation studies have been carried out in South Africa on metal accumulation in fishes in the Olifants River (Seymore et al., 1995; Robinson and Avenant-Oldewage, 1997; Nussey et al., 1999) Crocodile River (Heath, 1999) and the Vaal River system (Crafford, 2000; Groenewald, 2000; Kotze, 2003). Studies carried out on the Vaal River (Crafford, 2000; Groenewald, 2000; Avenant-Oldewage, 2001; Kotze,

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2003) have shown that fishes in the Vaal River system accumulate heavy metals discharged into the system by industries.

Cestodes and acanthocephalans accumulate heavy metals even in higher orders of magnitude than their fish host tissues (Galli et al., 1998; Sures and Siddall, 1999; Sures et al., 2003a,b, 2005; Sures, 2004). Sures et al. (2000) reported that the female archiacanthocephalan *Moniliformis moniliformis* from experimentally infected rats absorbed the highest lead concentration followed by the kidneys of the rats. In mammalian parasites (Sures et al., 2003a) the lead concentrations in *Hymenolepis diminuta* adults and *Taenia taeniaeformis* larvae were compared to the rat hosts (*Rattus norvegicus*) and it was observed that the *H. diminuta* also accumulated the highest lead concentration in a polluted site adjacent to the city Cairo, Egypt, when it was compared to the host's intestinal wall, liver, kidney and the larvae of *T. taeniaeformis*.

A study by Koch and Schoonbee (1980) on the Largemouth Yellowfish from the Vaal Dam in South Africa revealed a high prevalence by the Asian Tapeworm (*Bothriocephalus acheilognathi*). Bertasso and Avenant-Oldewage (2005) showed that the prevalence of *B. acheilognathi* in Largemouth Yellowfish collected in this dam ranged between 80% and 90% and that the mean intensities were consistently high and varied from 213.6 in autumn to the lowest value of 68.8 recorded in spring indicating sufficient supply of parasites. The aims of this study were (1) to evaluate whether cestodes accumulate heavy metals or not and (2) to investigate the extent to which cestode accumulate pollutants in comparison to their fish hosts, and the ecological implications thereof.

## 2. Materials and methods

### 2.1. Fieldwork

Twenty Largemouth Yellowfish, *Labeobarbus kimberleyensis* (Gilchrist and Thompson, 1913) specimens with different lengths (ranging from 380 mm to 605 mm) were collected during an autumn survey (April 2005) by means of gill nets in the Vaal Dam in the vicinity of RAU Island (26°52.249'S, 28°10.249'E). Each fish was weighed and the length recorded. They were killed by cutting the spinal cord. The liver, muscle and vertebrae were selected as reference tissues due to the high bioaccumulation shown in previous studies. These tissues were dissected out and placed in 25 ml glass bottles. The intestine was removed from each fish, the length recorded and placed in a plastic Petri dish with saline solution (0.9% NaCl). The intestine was opened with Dumont tweezers exposing the cestodes, and the point of attachment was noted. The worms from each fish were carefully removed and stored in a 25 ml glass bottle before the tissue was frozen until metal analysis. A sediment sample was also taken with an Ekman Grab as well as a grab water sample.

### 2.2. Laboratory work

#### 2.2.1. Microwave digestion

The samples were defrosted and  $\approx 1$  g fish or parasite tissue was weighed off to four decimal spaces with a Sartorius scale. The individual tissue samples were placed in the microwave digestion flasks with 5 ml Suprapur® nitric acid 65% and 1 ml Milli-Q water (18, 2  $\Omega$ ) and digested. Excess NO<sub>2</sub> gas was blown off into an extractor after digestion. The water samples were prepared by filtering the water through a 0.45  $\mu$ m membrane filter and adding 5 ml Suprapur® nitric acid 65%. The sediment samples were divided into two samples. One sample was digested as described above for fish and parasite tissue and the other sample dried in an oven for 3 h.

Pre-cleaned volumetric flasks were used to dilute the digested tissues with Milli-Q water. A calibrated ICP-MS (X Series Thermo Elemental – Thermo Element Corporation) was used to determine the metal concentrations of each tissue. Samples were transferred to Falcon tubes with screw-on lids. Indium internal standard solution was added to each sample. The concentration of arsenic, barium, beryllium, cadmium, chromium, cobalt, tin, tellurium, thallium, uranium, copper, iron, mercury, lithium, manganese, molybdenum, nickel, lead, antimony, selenium, titanium, vanadium and zinc was determined. Graph pad Prism 4.03, Microsoft Excel (Microsoft Office 2003) and SPSS 13.0 for Windows was used to calculate the concentration means to prepare graphs and all the graphs represent results from the wet mass of the sample types. Mauchly's test of sphericity was used to determine the 95% confidence interval. When the sphericity was assured, this *P* value was then used. If the sphericity was not assured the Greenhouse-Geisser test was used at an 80% confidence interval. When the Mauchly test did not show any significance, the paired samples test was used to determine the pairs in the groups that were significant.

## 3. Results

### 3.1. Organic tissue data

In Fig. 1A, it is indicated that the sediment (0.1062  $\mu$ g/ml) had the highest lithium concentration accumulated, followed by the water (0.083  $\mu$ g/ml). When the fish tissue means were compared it was observed that the cestodes had accumulated the highest concentration of lithium after the water and sediment. In the repeated measures test, *P* = 0.141 which indicates that the means are significantly different. In Fig. 1B, it is indicated that the sediment (0.0331  $\mu$ g/ml) had the highest beryllium concentration accumulated. When the fish tissue means were compared it was observed that the cestodes had accumulated the highest beryllium concentration, followed by the liver and the sediment. In the repeated measures test, *P* = 0.283 indicating that the means are significantly different. In Fig. 1C, it is indicated that the mean concentration of titanium was

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