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Chromium and Potassium Accumulation Influenced by Body Weight in Goldfish (*Carassius auratus*)

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Chromium has long been recognized as toxic, and even if it is not as toxic as other trace metals, such as Hg and Cd, the threshold limit value for some chromium compounds is being reduced as its toxic effects become better known (LANGARD 1980). Cr (VI) is considered more toxic than Cr (III), but these forms are readily interconvertible under natural conditions (SCHROEDER & LEE 1975). As most wastes from industrial uses that contain Cr are released into continental waters, effects on aquatic life have grown in interest. As with other trace metals, chromium levels in fish from unpolluted waters have been shown to be correlated with fish weight for some species, but not in others. When fish are placed in Cr polluted waters, they accumulate chromium; unpublished results obtained in our laboratory suggest that the size of the animal had an influence on the rate of accumulation. One purpose of this paper is to publish these data of chromium levels when fish samples are taken from polluted waters. We investigated the influence of body weight and chromium levels in several organs of goldfish (Carassius auratus) subjected to a subacute potassium dichromate treatment. In most studies on toxic effects of Cr (VI), where potassium dichromate is employed, only chromium levels are usually recorded and interest is focused on this metal. Low levels of potassium are usually not toxic, but high levels can be so. Thus, knowing potassium levels as well as chromium levels is therefore useful and can allow more understanding of potassium dichromate toxicity mechanisms. We have measured potassium levels as well.

MATERIALS AND METHODS

Experiments were performed in goldfish (Carassius auratus) of weights between 2 and 12 g. Fish were kept and acclimatized in laboratory conditions at least 2 weeks before their use in any test (SPRAGUE 1973). Water conditions: pH 7.3; free chlorine 0; combined chloride 0; hardness 155.6 mg/L. Dissolved oxygen: at saturation level by passing air constantly through the aquarium water. Temperature: 22 °C. Test were

conducted by adding $K_2Cr_2O_7$. Having determined that TL 50 25 ppm Cr (VI) for 82 days, a concentration of 20 ppm of Cr (VI) as potassium dichromate was used;it was high enough to be considered subacute but low enough to allow the analysis of live treated fish. Moribund fish not can be used in metal levels studies (RIVA et al. 1981). Specimens of different sizes were kept as controls, and the others were subjected to 20 ppm Cr (VI). Water was changed weekly, and animals were fed only once a week. Samples were taken after 4,6, 8, 11, 13, 15,18, 20, 22, 27, 29, 32 and for small specimens 42 days after treatment. The group of large fish was the only one that demonstrated mortality during treatment. Live fish were processed separately in two groups according to size each day. Levels of chromium and potassium were measured by spectrophotometric methods as described in ROUSSELET (1971). Results were analyzed using the "t" Student test for independent samples and correlation test (BAILEY 1976). In all cases 0.05 was adopted as the level for which significance was accepted.

RESULTS

Chromium levels (µg/g dry weight) in gills, liver, bile, and muscle are shown in Table 1 for control group, treated large fish group, and treated small fish group. Levels of chromium were analyzed in pools where specimens had aproximately the same weight (range from 2 to 12 g). Studying the relation between levels of chromium in gills, liver, bile, and muscle and the weight mean, it was shown that levels of the metal in the organs of control animals were not correlated with fish weight in this range and conditions.

A time influence on chromium levels in treated fish was only clear for liver where one peak before the 11th day of treatment was found both for large and small fish treatment groups. So we analyzed results without taking time into account except for liver. Chromium levels in gills of large and small treated fish were significantly higher than control fish, with small specimens significantly higher than those in large specimens. Levels in liver increase abruptely in both small and large fish. Levels of 773 µg/g dry weight for 8 days of treatment in large fish group and 1315 µg/g dry weight for 11 days of treatment in small fish group. After these initial changes, levels decreased but remain significantly higher than control ones, both for large and small fish groups, but differences between means of both treated groups were not significant. The mean of chromium levels in the bile in small fish group was significantly higher than in large and control fish. Chromium levels in muscle

Table 1. Chromium levels (μg/g dry weight) in gills, liver, bile, and muscle in control and treated fish. Mean ± standard deviation. a,b,...q: differences are significant. Student "t' test (0.05).

Fish	N°of fish in pool	Gill	Liver	Bile	Muscle
Control (2-12g)	22	42±15 (a,b)	55±26 (d,e)	143±83 (n)	11±4 (p,q)
Treated (6-12g)	36 (24 for liver)(1)	93±22 (a,c)	143±68 (d)	192±94 (m)	40±14 (p)
Treated (2-4g)	39 (27 for liver)(1)	163±75 (b,c)	214±99 (e)	345±152 (m,n)	47±24 (q)

(1) values for treated fish after the 11th day of treatment. See text for comment.

Table 2. Potassium levels (mg/g dry weight)in gills, liver,bile, and muscle in control and treated fish. Mean ± standard deviation a, b, c,: differences are significant. Student "t" test (0.05).

Fish	N°of fish in pool	Gill	Liver	Bile	Muscle
Control (2-12g)	22	6.5±1.6	15.3±2.7	8.9±3.3	10.4±1.6 (b,c)
Treated (6-12g)	36	5.2±1.3 (a)	13.4±4.0	6.2±1.1	14.2±2.2 (b)
Treated (2-4g)	39	6.9±1.1 (a)	14.4±4.9	10.1±7.1	15.7±3.5 (c)

of both small and large fish groups are significantly higher than in control group, but there was not a significant difference between both treated groups.

Potassium levels (mg/g dry weight) in gills, liver, bile, and muscle are shown in Table 2 for control group, treated large fish group, and small fish treated group. Levels of potassium in these organs of control animals have not shown any correlation with body weight in this range of weights and conditions. In treated groups, a time influence in potassium levels from 4 to 42 days of treatment does not appear. All pools are considered together separating large and small specimens. Means of potassium levels in gills of both big and small fish group are not significantly different from mean for control group, but the difference between large and small treated fish is significant. In liver and bile there are no significant differences between means of potassium levels from any group. Levels of potassium in muscle significantly increase in both treated groups (small and large fish) compared with control values, but there are no differences between them.

Relation between chromium and potassium levels. Levels of chromium and potassium in these organs have been analyzed to show correlation between them. In controls no correlation has been found. In treated fish, chromium and potassium levels are positively correlated in gills and bile.

DISCUSSION

Our results show no correlation between chromium levels in gills, liver, muscle, bile, and body weight in Carassius auratus in a weight range of 2-12 g even if body weight is one of the factors that can influence several metal levels in fish. Results from chromium studies do not show a consistent relationship between chromium concentrations and size of fish. A positive correlation, an inverse relationship or no correlation at all have been reported in several species between body size and chromium levels in body and/or organs (TONG et al. 1974; GIESY & WIENER 1977; MEARS & EISLER 1977; ELWOOD et al. 1980). These discrepances could be related, in part, to both small sample size and small weight range used in several of these studies as well as ours. Similar facts can be reported for potassium in fish where a consistent relationship has not been established (GOLDBERG 1962; EISLER & LAROCHE 1972).

Over the period of potassium dichromate treatment, all organs (except bile in large specimens) showed

accumulation of chromium. During these 32-42 days only Cr levels in liver showed the clear influence of time having a peak before the 11th day of treatment both for big and small treated fish group, and being lower but still higher than controls afterwards. Except for this high initial levels in liver, accumulation in the organs was constant. In rainbow trout exposed to 0.05-0.15 mg/L or maintained on a diet containing chromium but in low concentration, fish accumulated chromium but did not reach equilibrium quickly (FROMM & STOKES 1962; SINGH & FERNS 1978). But this was reached within aproximately 1 day when trout were subjected to 2.5 mg/L Cr VI (BUHLER et al. 1977). So our results appear to corroborate that high levels of chromium in circulating waters are needed to reach equilibrium quickly and this could involve an increased rate of loss rather than a reduced rate of uptake (SINGH & FERNS 1978). Metal excretion could be related to body levels exceeding certain limits, levels that would have been reached quickly when Cr concentrations in water were higher.

Our results are suggestive too that regulatory ability for K does differ considerably from ability for Cr. Potassium levels in muscle significantly increased both in small and large fish groups. Gills, liver, and bile failed to show any significant difference between controls and treated fish. In tench (Tinca tinca L) exposed to 8.5 mg/L K^+ (Potassium nitrate), plasma potassium increased after 3 days but returned to normal levels afterwards, and liver tissue accumulated relatively large quantities of K^+ during the first week, but levels returned to normal slowly (DEMAEL et al.1980). A similar pattern of accumulation is not followed by the two metals, but in treated fish, chromium and potassium levels are positively correlated in gills and bile, two organs related with uptake (gills) and excretion (gills and bile) of chromium.

Body weight affected chromium levels in treated fish only for gills and bile, small specimens having higher levels than big fish. It is noteworthy that in the case of bile, only small specimens showed levels significantly higher than controls. In excretory organs, accumulation has been thought to be correlated with this function (KNOLL & FROMM 1960). This, together with the fact that differences in accumulation between large and small fish have been registered in gills and bile, could reflect that these organs play a different role in chromium excretion in small and large specimens with the biliary pathway more important in small than in big fish. As chromium and potassium levels are correlated in organs, some relation between their excretion could exist. In gills and liver of

trout (BUHLER et al. 1977) (bile was not reported), chromium was shown to accumulate preferentially in the cell cytosol providing tentative evidence for the presence of high concentrations of the soluble Cr VI anion in those tissues, a possible rapid turnover pool of chromium, that could be important in the uptake and excretion of the metal. In slow turnover pools, chromium could be bound to tissue proteins. Perhaps the relative capacity of this two pools could change with age.

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