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Accumulation of Pesticides in the Organs of Carp, *Cyprinus carpio* L., at 4° and 20°C

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Waste materials originating from intensive agricultural production exert harmful effects when they reach natural waters, since they may become concentrated in the organs of aquatic animals /Salánki et al. 1982/. The degree of accumulation is greater the higher the stage of the given organism in the food chain. Thus, fish are particularly sensitive to environmental contamination of the water and pollutants may significantly damage certain physiological and biochemical processes when they enter the organs of fishes.

Pollutants are not only harmful to adult fish, but may also cause disturbances of development in embryonic stages /Weis and Weis 1977/. Several cases have been reported in which the toxic effect of pollutants may be decreased, or even increased by various water quality factors. These are the pH, temperature, hardness and dissolved oxygen content of the water /Zitko and Carson 1976; Pascoe et al. 1986/. The harmful effects - especially sublethal - retard the development of the surviving individuals and/or are of harmful influence on their normal metabolic processes.

The quantitative distribution of pesticides in aquatic organisms has recently been more intensively studied in Hungary /Salánki et al. 1982/.

In the light of the foregoing, studies were performed on the accumulation of $CuSO_4$ used as fungicide and paraquat: PQ /1,1'-dimethyl-4-4' bipiridilium-dichloride/ used as herbicide, in the various organs of carp at winter and summer /4°C and 20°C/ water temperature, respectively. Our aim was to provide data in relation to the seasonal accumulation of these pesticides, which could increase the possibility of predicting the damaging effects caused by the temperature dependent pesticide-accumulation.

MATERIALS AND METHODS

Carp /Cyprinus carpio L./ weighing 350-400 g obtained from the Fisheries Research Institute at Szarvas were used for our experiment.

After an appropriate adaptation period /3-7 days/ each fish was placed into a 5 l aquarium which was constantly well aerated.

The following compounds were used for the measurements:

14-C-paraquat. Specific activity: 425 /uCi/mg. During the course of the experiment the incorporation of 10 ppm concentration of the labelled paraquat was studied at water temperatures of 4 °C and 20°C, after an exposure time of 2, 8, 24 and 48 hours.

$^{234}CuSO_4$. Specific activity: 2.7 mCi/ml. Copper content: 1,95 mg/ml.
At this experiment, following an exposure time of 2 hours the incorporation of $CuSO_4$ was measured at 4°C and 10 ppm treatment and also at 20°C following 1,0; 10 and 100 ppm treatment. Treatment of longer duration was not performed owing to the fast half-period of the $CuSO_4$ isotope being at our disposal /Half-life: 24 hours/.

The appropriate isotope concentration of the studied compounds was dosed into the water of the aquarium. After a given time the experimental fish were sacrified and the amount of pesticides accumulated in the removed organs was determined on the basis of radioactivity measurements.

About 1 g of the organs removed from the sacrified fish was dissolved in protosol solution /200 mg of organ/ 1 ml of protosol/, then made up to 5 ml with tolulol-coctail solution and the radioactivity measured by means of Packard liquid-scintillation apparatus. Each experiment was repeated 3-5 times. Acetylcholinesterase /AChE/ enzyme activity in fish exposed to different concentrations of $CuSO_4$ was measured with the Ellman reagents.

RESULTS AND DISCUSSION

With the exception of the brain, greater copper accumulation was measurable in every organ at 20°C /Fig. 1/ compared to the values determined at 4°C /Fig 2., Table 1./

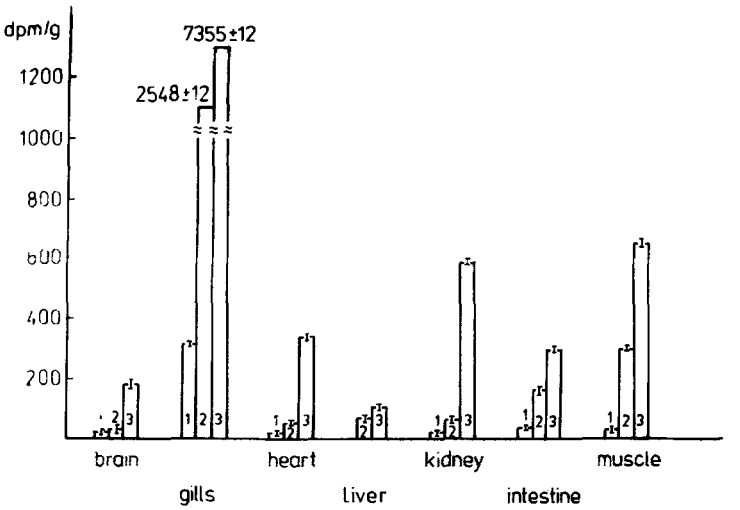

Figure 1. In vivo accumulation of 1,0 /1/; 10,0 /2/ and 100 /3/ ppm $CuSO_4$ into various organs of carp at 20°C following 2 hours treatment. Mean values of samples from 3-5 individuals are given, as dpm/g /± S.E.M./. Specific activity: 2,7 mCi/ml. Copper content 1,95 mg/ml.

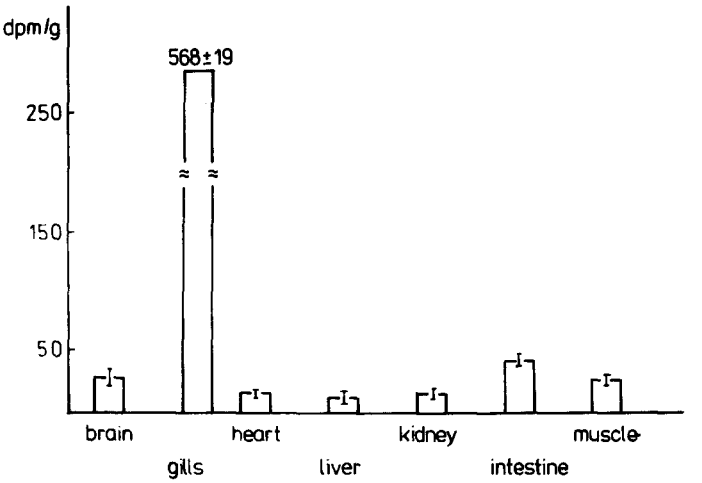

Figure 2. In vivo accumulation of 10 ppm $CuSO_4$ into the various organs of carp at 4°C following 2 hours treatment. Values are the average of the samples measured from 3-5 individuals, expressed in dpm/g /± S.E.M./ Specific activity: 2.7 mCi/ml

The order of the concentrations of copper taken up by the various organs was: skeletal muscle > liver > gills > intestine > kidney > heart > brain /Table 1./

Table 1. In vivo accumulation of 10 ppm $CuSO_4$ into the various organs of carp /Cyprinus carpio L./. The given values mean the percentage of the values measured at 20°C expressed in the percentage of the values measured at 4°C.

Treatment time: 2 hours.	
Brain	75
Gills	448
Intestine	372
Heart	250
Kidney	360
Liver	500
Muscle	1050

The low $CuSO_4$ accumulation detected in the brain may be in correlation with the presence of the blood-brain barrier. The high amount of Cu in the gills and liver is related to the fact that the gills play an important

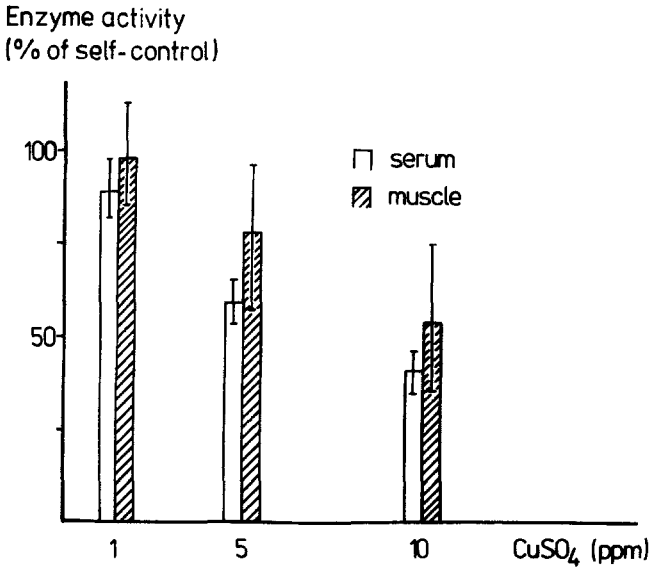

Figure 3. In vivo effect of 1, 5 and 10 ppm $CuSO_4$ on AChE activity in skeletal muscles and serum of carp. Exposure time 2 hours. Values expressed in the percentage of the control are averages of the samples measured from 3-5 individuals.

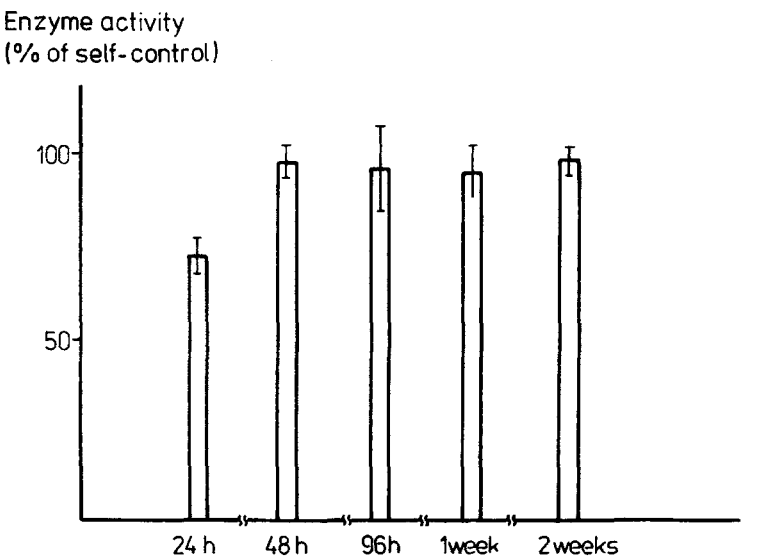

Figure 4. In vivo effect of 5 ppm $CuSO_4$ on AChE activity in serum of carp depending on exposure time. Values expressed in the percentage of the control are averages of the samples measured from 3-5 individuals.

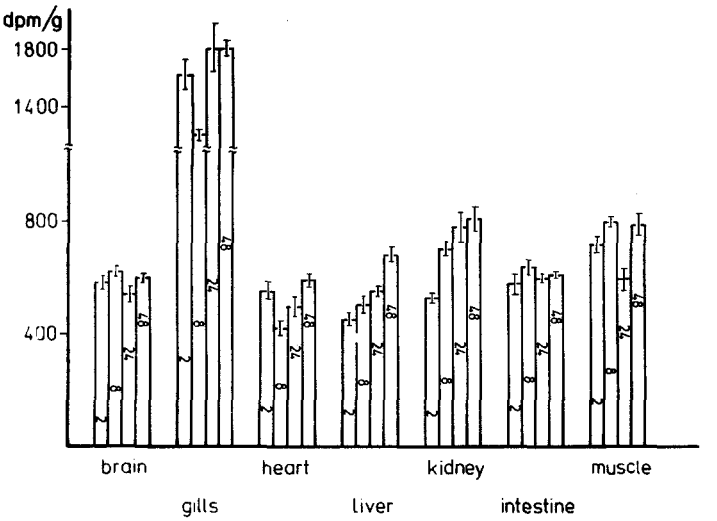

Figure 5. Accumulation of 10 ppm ^{14}C-labelled paraquat into the various organs of carp at 20°C depending on the exposure time. Values are the averages of the samples measured from 3-5 individuals, expressed in dpm/g /± S.E.M./. Specific activity: 425 /uCi/mg.

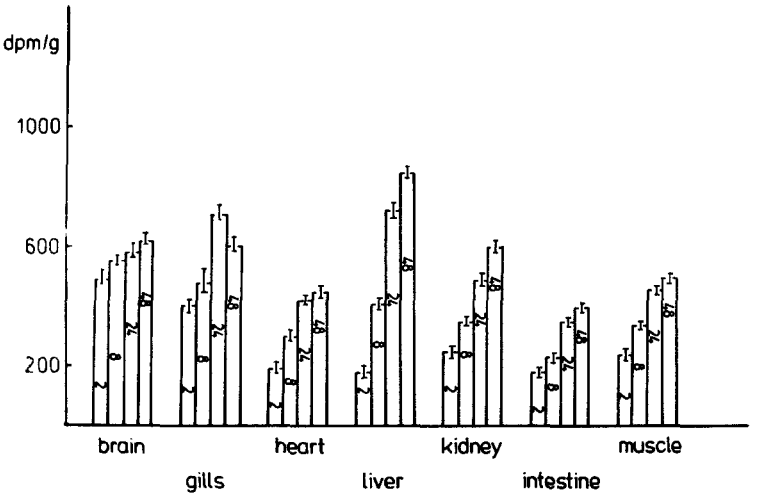

Figure 6. Accumulation of 10 ppm ^{14}C-labelled paraquat into the various organs of carp at 4°C depending on the exposure time. Values are the averages of the samples measured from 3-5 individuals, expressed in dpm/g /± S.E.M./. Specific activity: 425 /uCi/mg.

role in copper uptake, and the liver in its accumulation and detoxication /Reichenbach-Klinke 1972/. The high copper-content of the skeletal muscle - which after 2 hours exposure strongly inhibited the acetylcholinesterase activity in this organ - may be correlated with the greater activity of the fish at 20°C /Fig. 3/. During the course of this movement more copper which has not been transported yet towards other copper storing organs during the treatment period, may enter the organ because of the enhanced blood-supply and accelerated metabolic process of the skeletal muscle.

Treatments of longer duration also confirmed this concept since the considerable acetylcholinesterase inhibition in carp manifested at 5 ppm $CuSO_4$ treatment for two weeks could only be measured in the first 24 hours. Following this the acetylcholinesterase activity returned to normal level as before treatment /Fig.4./. The inhibition of acetylcholinesterase by $CuSO_4$ contamination has also been reported by Olson and Christensen 1980. According other workers copper may also display its damaging effect elsewhere. On the basis of experiments performed on carp, damage to the gills, liver, kidney and nervous system has been recorded at a concentration of 1,5 ppm. At the same concentration, $CuSO_4$ caused adverse alterations in certain blood parameters of the carp: significant increase in haematocrit value, haemoglobin content and protein, as well as in the concentrations of glucose. At the same time, a decrease was observed in the number of leucocytes /Svobodova 1982/.

According to Reichenbach-Klinke /1972/ the major target of copper in the case of fish is the gill epithelium, while Schreck and Lorz /1978/ consider that both the gills and kidney may become damaged by $CuSO_4$.

The electronmicroscopic studies of Rojik et al. /1983/ have verified the foregoing conclusions. During the course of their studies $CuSO_4$ damaged the gills, liver and kidney of carp, silver carp and silure, where mainly the endoplasmic reticulum and mitochondria were damaged.

The effect of increased temperature /from 4°C to 20°C/ was greater accumulation of PQ in every studied organ /Figs. 5, 6, Table 2/. The increase was lower than with $CuSO_4$. According to the time-dependent accumulation of paraquat the degree of accumulation was the highest during the first 2 hours after treatment in practically every organ studied /with the exception of the muscle and brain/.

Table 2. In vivo accumulation of ^{14}C-labelled paraquat into the various organs of carp /Cyprinus carpio L./ depending on the exposure time.
Applied concentration: 10 ppm.
The given values indicate the percentage of the values measured at 20°C expressed in the percentage of the values measured at 4°C.

	2 hours	8 hours	24 hours	48 hours
brain	120	130	93	96
gills	412	250	253	300
intestine	322	280	171	150
heart	289	140	116	130
kidney	212	200	159	130
liver	250	120	76	80
muscle	150	235	130	160

Following this the paraquat concentration in the various organ tended to decline. Even in low concentrations, paraquat could be regarded as one of the most toxic pesticides to living organisms. Its damaging effect is exerted during its metabolism in the organism because of the extremely toxic effect of the formed free radicals /Stancliffe and Pirie 1971/. The developed free radicals may lead to the degradation of the cell membranes, as has successfully been demontrated in fish, with biochemical and electronmicroscopic methods /Rojik et al. 1983/. Paraquat also damages the nervous system of the fish, through the inhibition of acetylcholinesterase /Nemcsók et al. 1984/.

In addition, the pesticides studied have a stress - inducing effect on the fish during the course of long-lasting exposure, as indicated by the increased blood glucose level /Nemcsók and Boross 1982/. In such cases the fish become more susceptible to infectious diseases /Wedemeyer 1970/.

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