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Zn, Cu, and Mn Levels in the Liver of the Dogfish Exposed to Zn

C. Sanpera, M. Vallribera, and S. Crespo*

*Fisiologia Animal, Faculdad de Ciencies, and *Biologia, Faculdad de Veterinária, Universidad Autónoma de Barcelona, Bellaterra, Spain*

Zn contamination has been shown to alter physiological balance in fish. Respiratory processes (SKIDMORE 1970, SELLERS et al. 1975), cardiac respiratory rhythms (HUGHES & ADENEY 1977), O_2 Consumption of the gill tissue (TORT et al. 1982), osmoregulation (LEWIS & LEWIS 1971, WATSON & BEAMISH 1980, CRESPO & KARNAKY 1983) and enzymatic activity (JACKIM 1974) are among the processes that have been proven to be affected.

Heavy metals such as Zn, Cu or Mn are essential elements which exhibit similar atomic structure and therefore compete for the same sites (UNDERWOOD 1977, SANDSTEAD 1981). Interactions among these elements have been described in intertebrates (CUADRAS et al. 1981, SUTHERLAND & MAJOR 1981, ADAMS et al. 1982),fish (SHEARS & FLETCHER 1979, CRESPO et al. 1981) and mammals (BREMNER 1979, SANDSTEAD 1981, SOLOMONS 1981).

To investigate the effects of Zn contamination on the hepatic distribution of these trace elements, Zn, Cu and Mn levels were determined by atomic absorption spectrophotometry in the liver of the dogfish *Scyliorhinus canicula* exposed to 80 and 10 ppm of zinc.

MATERIALS AND METHODS

Experiments were conducted on 25 male dogfish of 150–300 g body weight which were Kept for at least one week before experimentation in an open circulation tank of natural seawater. Eight fish were treated with 80 ppm of Zn ($ZnSO_4$ in natural seawater) for 24h. This concentration corresponds to the LC 50 48h for the dogfish (CRESPO & BALASCH 1980). Eight fish were treated with 10 ppm of Zn for 3 weeks. In order to minimize the reduction of Zn concetration either by precipitation or uptake by the fish and to remove nitrogenous excretions, the Zn solution was replaced every 3 days. Nine fish were used as controls. All fish (experimental and controls) were maintained in 30 litre volume tanks (2 dogfish in each) and were not fed during experimentation (3 weeks). Dogfish were Killed with tricaine methanesulfonate (MS - 222 Sandoz). The liver was taken out from the specimen, carefully washed in de-ionized water, homogenized, oven dried in air at 100°C and weighed. Three samples were taken from each specimen weighing aproximately 0.5 g - 2 g (dry weight). Each sample was digested with 10 ml HNO_3 and 10 ml $HClO_4$. After evaporation to

dryness, samples were diluted to 10 ml with de-ionized water for Cu and Mn determinations. A 10X dilution was prepared for Zn.

Metal levels were determined by atomic absorption spectrophotometry (PYE UNICAM SP 1900).

RESULTS AND DISCUSSION

None of the fish exposed to a sublethal dose (10 ppm-3 weeks) displayed any apparent toxicity symptoms. On the contrary, most of the fish exposed to 80 ppm were overturned in moment of sacrificing.

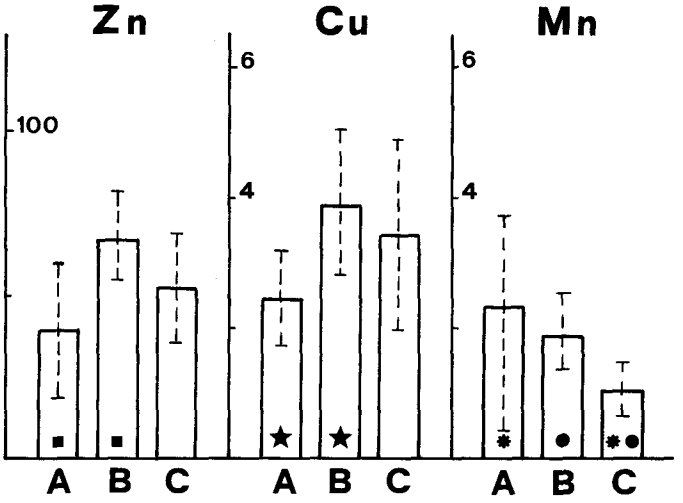

Fig. 1 Zn, Cu and Mn content (μg/g dry weight) in the liver of the dogfish exposed to Zn. Mean ± standard deviation. A = Control animals, B = Experimental animals (10 ppm Zn - 3 weeks), C = Experimental animals (80 ppm Zn - 24h). According to Student t-test: ★ P = 0.01; ■ P = 0.02; ● P = 0.05 According to Mann-Whytney U-test ✱ P = 0.05.

Fig. 1 shows that hepatic Zn levels increased significantly following sublethal Zn treatment. A significant Cu increase was also recorded following 10 ppm - 3 week exposure whereas no differences in Mn were found. After acute Zn treatment, no increase in the hepatic Zn content was detected; however, Mn levels decreased significantly. No differences in Cu were found.

From these data it is apparent that dogfish exposed to 10 ppm Zn for 3 weeks accumulated Zn in the liver. Other internal organs of the dogfish such as kidney, intestine, pancreas and spleen have also been shown to accumulate Zn following sublethal Zn contamination whilst no changes were detected following acute treatment (CRESPO et al. 1981; FLOS et al. 1979).

The mechanisms responsible for the changes in Cu and Mn hepatic levels in the dogfish following Zn contamination are unknown. However, Zn-Cu interactions might be different from Zn-Mn interactions since changes in Cu levels were recorded after

sublethal Zn treatment whereas Mn changes were only found after acute treatment. Taking into account data from SUTHERLAND and MAJOR (1981) showing changes in trace element content in some organs of the blue mussle after Cu treatment, we conclude that heavy metal contamination results in a redistribution of trace elements such as Zn, Cu and Mn within the organism. Trace metal imbalance in cotaminated individuals might be responsible, for The appearance of toxicity symptoms.

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REFERENCES

ADAMS E., K. SIMKISS & M. TAYLOR: Comp. Biochem. Physiol. 72 A, 73 (1982).
BREMNER. I: Proc. Nutr. Soc. 38, 235 (1979).
CRESPO S. & J. BALASCH: Bull. Environm. Contam. Toxicol., 24, 940 (1980).
CRESPO S., E. SORIANO, C. SANPERA & J. BALASCH: Mar. Biol. 65, 117 (1981).
CRESPO S. & K. J. KARNAKY Jr.: J. exp. Biol. 102, 337 (1983).
CUADRAS J., A. GIMENO, R. FLOS & S.CRESPO: Crustaceana 40-1, 79 (1981).
FLOS R, A. CARITAT & J. BALASCH: Comp. Biochem. Physiol, 64, C, 78 (1979).
HUGHES G. M & R.J. ADENEY: Water Res. 11, 1069 (1977).
JACKIM.: In "Pollution and Physiology of Marine organisms". F. J. Vernberg and W,B. Vernberg ed. New York: Academic Press (1974).
LEWIS S.D & W.M. LEWIS: Trans. Am. Fish Soc. 4, 639 (1971).
SANDSTEAD H.H.: J. Lab. Clin. Med. 98-4, 457 (1981).
SELLERS C.M., A.G. HEATH & M.L. BASS: Water Res. 9, 401 (1975)
SHEARS M.A. & G.L. FLETCHER: Comp. Biochem. Physiol. 64 B, 279 (1979).
SKIDMORE J.F.: Q. Rev. Biol. 39-B, 227 (1970).
SOLOMONS, NW: In "Nutrition in the 1980 : Constraints on our Knowledge". Alan R. Liss Inc. ed. New York (1981).
SUTHERLAND J. & C.W. MAJOR: Comp. Biochem. Physiol. 68 C, 63 (1981)
TORT L., S. CRESPO & J. BALASCH: Comp. Biochem. Physiol. 72 C, 145 (1982).
UNDERWOOD E.J.: Trace elements in human and animal nutrition. London: Academic Press (1977).
WATSON T.A & W.H. BEAMISH: Comp. Biochem. Physiol. 66 C, 77 (1980).
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