P-17

In vivo effects of cadmium exposure on hematological and detoxification enzymes of the Juvenile Rockfish, *Sebastes schlegeli*

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In fish, Cadmium (Cd) has adverse effects on growth, reproduction, and osmoregulation. Moreover, it affects respiratory functions and the composition of plasma by causing hypocalcemia, hypokalemia and hyperglycemia. The activity of metabolic enzymes in liver, kidney, muscles, and other tissues is disturbed following exposure to Cd (Sastry and Subhadra, 1982). Biotransformation systems are generally regarded as consisting of two subsystems; Phase I and II systems. The phase II matabolizing enzymes have been suggested as a biomaker in hazard assessment of contaminants by a number of authors in laboratory or filed studies (Vander Weiden et al., 1993). The exposure of marine species to metals has been shown to induce oxidative stress through the formation of reactive oxygen and nitrogen species (ROS/RNS) and lipid peroxidation (Viarengo et al., 1990). To protect against oxidative stress, organisms possess antioxidant defenses. For example, glutathione peroxidase (GPx), detoxifies hydrogen peroxide, and glutathione reductase and glutathione S-transferase (GST), conjugates xenobiotics with reduced glutathione (GSH) for excretion.

Although adverse effects of Cd on various physiological functions of fish are well documented, very little research has been done on the capabilities of metals to induce phase II activity. Therefore, the aims of the present study were to estimate haematological varations and to investigate the activity of phase II enzymes; GPx, GST, GSH and GR in juvenile rock fish, after sub-chronic Cd exposure.

Juvenile Rock fish (11.1±0.03 ㎝, 21.6±0.2 g) were obtained from a fish farm in Gyeongnam. After acclimatization, the fish were separated into four experimental lots with 10 animals each. Diets were supplemented with 0, 1, 5 and 10 ㎎ kg^{-1} feed of $Cd(NO_3)_2.4H_2O$ (Aldrich Co.) to investigate the haematological fluctuation

and phase II enzyme activities. After 15 and 30 days exposure, fish were anesthetized and examined. Blood sample was centrifuged at 3000 g for 15min at 4℃. Haematological parameters include the total number of erythrocytes, haemoglobin concentration, haematocrit, chloride, phosphate, magnesium, calcium, tatal protein, glucose, total cholesterol, GOT and GPT. Their assay was perfomed using a diagnostic kit (Asan Pharm, Co., Ltd). Hepatic microsome was prepared by homogenizing in buffer with several passess of a teflon pestle. The hemogenate was centrifuged at 9,000 g for 20 min at 4℃, supernatant was collected, and ultracentrifuged at 100,500 g for 60 min using a Centrikon T-1190 Ultracentrifuge at 4℃ to obtain a microsomal pellet. Glutathione (GSH) was determined according to Sen et al. (1992). The activity of glutathione reductase (GR) was assayed by the method of Carlberg and Mannervik (1985). Glutathione peroxidase (GPx) was assayed by the method of Mohandas et al. (1984). The cytosolic glutathione S-transferase (GST) activity was analyzed by the kinetic method of Habig et al. (1974) with CDNB (1-chloro-2-,4-dinitrobenzene) as the substrate.

It has been observed that higher concentration of Cd causes a decrease in RBC count ($\times 10^4$ mm_3^{-1}), Ht (%) level and Hb (g dL^{-1}) concentration. The predominant haematological finding was a highly significant decrease of the total number of RBC and Hb in fish exposed to 5 mg kg^{-1} group thereby indicating a severe anaemia ($P<0.05$). Also, serum total protein (g dL^{-1}) and cholesterol levels (mg dL^{-1}) showed significant decrease in Cd treatment groups as compared to the control ($P<0.05$). Phosphate (mM), magnesium (mM) and calcium (mM) were found to be significantly increased over the control group. Although plasma total protein level was significantly reduced compared to the control group after 30 days, plasma chloride (mM), glucose (mg $d\ell^{-1}$), GOT and GPT (KU) level were found to be significantly increased over the control group. Exposure to Cd resulted in significant increase in the phese II enzymes activities, such as GST, GR and GPx in 5 and 10 mg kg^{-1}. But, GSH is presented that the acivities were increased at 15 days in 10 mg kg^{-1}. The results of the present study led us to conclude that concentration of Cd 5 mg kg^{-1} of the estuarine could affect the hematological distribution and phase II parameters of the juvenil rockfish.