P-22

Effect of dietary di-butyll phthalate and di-ethylhexyl phthalate on glutathione level and glutathione related enzyme activities of bagrid catfish, *Pseudobagrus fulvidraco*

Yoo-Hwa Keum, Jung-Hoon Jee, Ju-Chan Kang
Department of Aquatic Life Medicine, Pukyong National University

INTRODUCTION

Dibutyl phthalate (DBP) and di ethylhexyl phthalate (DEHP) are widely used as a plasticizer. However they cause a peroxisome proliferation and have been implicated as having estrogenic properties. Especially, the toxicity of DEHP has been extensively studied, but DBP have not been thoroughly studied. Effects of dietary DBP and DEHP on glutathione level and glutathione related enzymes of bagrid catfish (*Pseudobagrus fulvidraco*), were assessed for 8 weeks.

MATERIALS AND METHODS

Bagrid catfish (*Pseudobagrus fulvidraco*) were obtained from an Inland Fisheries Research Institute in Chung Cheong, Korea. After acclimatization, fish (mean length, 17.9±2.1cm, body weight 54.3±1.7g) were selected for the experiments. Fish were administered four concentrations of DBP and DEHP (0, 100, 500, 1000 mg DBP or DEHP/kg diet) for 4 or 8 weeks. At the end of each period (at 4, 8 weeks) fish were anesthetized and weighed. Livers, gill and kidney were isolated from the experimental fish and homogenized ice cold buffer using teflon pestle. The homogenates were centrifuged at 10,000g for 20 min at 4 ℃, and the supernatant obtained was stored -70℃ before analysis. Glutathione level were measured by Baker *et al.* (1990) method and glutathione peroxidase activity were used after (Bell *et al.*, 1985), modified for microplate assay, using H_2O_2 as the substrate and sodium azide as a catalase inhibitor. Glutathione reductase activity were measured by the increase in absorbance caused by the reduction of DTNB [5,5'-dithiobis(2-nitrobenzoic acid)] at 412nm. Protein concentration was determined by the method of bradford (1976) with bovine serum albumin as a standard.

Significant differences between groups were determined using one-way ANOVAs and Duncan's test for multiple comparions.

RESULT

For DBP fed fish, glutathione content in liver and gill showed a significant increase after 4 or 8 weeks of exposure in fish fed 500 and 1000 mg/kg diet concentration, whereas for DEHP fed fish, only liver glutathione content was increased at the same DBP concentrations. Fish fed th highest concentration of DBP and DEHP had significant higher renal glutathione content compared with control. Highest concentration of DBP (1000 mg/kg diet) induced a significant increse in the activity of glutathione peroxidase in all organs at time intervals of 4 and 8 weeks. Liver and gill glutathione reductase activity in fish showed a significant increase in its activity in the all DBP exposed fish when compared to control at 8th weeks. However, renal glutathione reductase activity was significantly increased in fish fed 1000 mg DBP/kg diet. Liver glutathione reductase activity in fish fed DEHP were investigated same pattern like DBP, whereas gill and kidney glutathione reductase activities showed significant increase in highest concentration of DEHP after 8 weeks exposure.

REFERENCE

Bell, J.G., C.B. Cowey, J.W. Adron and A.M. Shanks. 1985. Some effects of vitamin E and selenium deprivation on tissue enzyme levels and indices of tissue peroxidation in rainbow trout (*Salmo gairdneri*). Br. J. Nutr., 53, 149-157.

Baker, M.A., G.J. Cerniglia and A. Zaman. 1990. Microtiter plate assay for the measurement of glutathione and glutathione disulfide in large numbers of biological samples. Anal. Biochem., 190, 360-365.

Bradford, M.M., 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal. biochem., 72, 248-254.