Detection of metals and polychlorobiphenyls and their correlation with detoxificant enzymes activity in *Dicentrarchus labrax*

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Abstract

Several pollutants released to the environment, are biotransformed into more soluble molecules, in liver, by several enzymes, as catalase (CAT) and glutathione-S-tranferase (GST), which are fundamental for detoxification and excretion. The aim of this study was to investigate the relationships among xenobiotic levels and CAT and GST enzymatic activities, in reared European sea bass.

Introduction

In the last decades, several pollutants such as metals (Hg, Cd, Cr, and Pb) and polychlorobiphenyls (PCBs) are released into the environment [1]. The consumption of fish and fishery products appears to be one of the most significant way by which humans may be exposed to PCBs [2]. European sea bass (*Dicentrarchus labrax*) is a marine fish species widely distributed in the Mediterranean Sea, one of the species mostly produced in aquaculture in this area [3], chosen in this study as bioindicator species.
Xenobiotics are biotransformed into more soluble molecules in the liver, by several enzymes, as CAT and GST, chosen here as biomarkers of exposure [4].
The purpose of this study was to investigate the relationships among metal and PCB levels and CAT and GST activities, in *D. labrax* under different farming conditions.

Materials and Methods

D. labrax (n=20) were sampled from two different fish farms: off-shore (site 1) and in-shore (site 2). The sampling periods were: summer 2009 and winter 2010. Muscle and liver were used for chemical and biochemical analysis, respectively. To assay the metal concentration a microwave digestion method was used to prepare samples for atomic absorption spectroscopy analysis, according to the prescribed 333/2007/CE; while PCBs levels were assayed by gas-chromatography and high resolution mass spectrometry. CAT was spectrophotometrically assayed at 240 nm, by consumption of H_2O_2. GST activity was analyzed to spectrophotometer, following the formation of s-2,4-dinitro phenyl glutathione, at 340 nm.

Results

Metal concentrations were under the law-limits (Cd not detectable), even though they appeared more elevated in winter than in summer period (tab. 1).

	Site 1		Site 2	
	Summer	Winter	Summer	Winter
Hg (mg/kg)	0.02 ± 0.03	0.03±0.05	0.00±0.00	0.03±0.04
Pb (mg/Kg)	0.02 ± 0.03	0.11±0.04	0.06±0.08	0.14±0.06
Cd (mg/kg)	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
Cr (mg/Kg)	0.05±0.03	0.12±0.09	0.03±0.04	0.05±0.06

Table 1. Metal concentrations in sea bass muscle. Each value is expressed as average ± s. d.

CAT activity was higher in summer than in winter, in both sites. In particular, the CAT levels in summer were: 134.72 ± 22.39 U/mg for site 1 and 155.63 ± 30.36 U/mg for site 2; while in winter were: 89.50 ± 15.89 U/mg for site 1 and 64.14 ± 14.51 U/mg for site 2. Moreover it was observed a significant difference between site 1 and 2, in winter.
The analysis on PCB concentration showed that values were always higher in site 2 than in site 1, both for Dioxin-Like PCBs (DL-PCBs) and for non Dioxin-Like-PCBs (NDL-PCBs) (tab. 2). For DL-PCBs, the concentrations were under the law-limit (8 pg TEQ/g fresh tissue).
GST activity was significantly more elevated in site 2 (168 ± 30.7 mU/mg) than in site 1 (126.8 ± 28.5 mU/mg), in the winter period.

	DL-PCB ng/g fat	
	Site 1	Site 2
PCB-81	0.03± 0.01	0.18± 0.22
PCB-77	0.31± 0.04	1.48± 0.86
PCB-123	0.20± 0.07	1.27± 1.35
PCB-118	15.87± 2.90	50.81± 33.02
PCB-114	0.24± 0.03	1.21± 1.53
PCB-105	4.27± 0.65	14.56± 8.80
PCB-126	0.12± 0.02	0.20± 0.06
PCB 167	1.29± 0.30	4.90± 6.41
PCB 156	1.81± 0.45	3.82± 2.82
PCB 157	0.47± 0.11	1.02± 0.88
PCB 169	0.01± 0.00	0.03± 0.03
PCB-189	0.22± 0.07	0.29± 0.21
	NDL-PCB ng/g fat	
	Site 1	Site 2
PCB 28	4.1± 2.2	29.1± 25
PCB-52	10.3± 3.7	71.9± 52.9
PCB-101	13.9± 4.1	69.8± 51.1
PCB-153	24 ± 8.2	49.5± 39.3
PCB-138	13.1± 3.8	24.5± 16.4
PCB-180	13.1± 3	20.5± 13.1
$\sum_{6\ PCB}$	78.8± 19.2	265.4± 196.4

Table 2. PCB concentrations in sea bass muscle. Each value is expressed as average ± s. d.

Discussion

The chemical analysis demonstrated that metals increased in winter period while, in the same period, the CAT activity decreased, suggesting a no-correlation among CAT activity and metal presence, possibly explained by the low metal levels, under the law-limit and the not detectable Cd concentration. Therefore, the significant difference of CAT activity in the two periods could be due to environmental condition, such as suggested by Radovanovic et al. [5], that has observed a seasonal-specific alteration in Super Oxide Dismutase and CAT activities, due to an increase of ROS production. In the winter period, both PCB concentrations and GST activity increased in site 2 in front of site 1, suggesting a positive correlation between PCB levels and GST activity.

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