CHANGES IN LIPID CLASSES, FATTY ACID COMPOSITION AND LIPID PEROXIDATION IN THE GILLS OF THE CLAM *DONAX TRUNCULUS* AFTER PERMETHRIN EXPOSURE

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Abstract
A short term exposure experiment (72 h) to permethrin (PER) of the clam *Donax trunculus* from the Tunisian coast was carried out. Changes in the lipid classes, fatty acid (FA) composition and lipid peroxidation were determined in the gills of adults and juveniles. A significant decrease of total lipid content was followed by a decrease in polar lipids (PL) and polyunsaturated fatty acids (PUFAs) especially n-3 and n-6. As, for the oxidative stress response, a significant elevation in lipid peroxidation (LPO) was found after the exposure of both adults and juveniles.

Keywords: Bivalves, Gulf of Tunis

Introduction
Pesticides cause changes in physiological and metabolic functions of non-target organisms. The permethrin (PER), a synthetic pyrethroid insecticide, can be very toxic to aquatic invertebrate communities. Contaminants like pesticides may induce the formation of reactive oxygen species (ROS) which are responsible of damage to lipids, proteins, carbohydrates and nucleic acids. The aim of the present investigation was to provide new data concerning the changes in the lipid classes and fatty acid composition as well as in the lipid peroxidation in the gills of *Donax trunculus* exposed to PER for 72 h.

Materials and methods
Specimens of *D. trunculus* (adults and juveniles) were collected from the gulf of Tunis (Tunisia) and exposed to 150µg/L PER for 72h. Gills were dissected, immediately frozen in liquid nitrogen and stored at -80°C until analysis. Lipids were extracted according to [1] method. Lipid classes were separated in precoated HPTLC silica gel 60 plates using a double development method. Methylation of fatty acids was performed according to Cecchi et al. [3], and lipid peroxidation was determined using the method of Draper et al. [4] and expressed as nanomoles of malondialdehyde (MDA) per g wet weight of tissues. Significant differences between control and exposed groups was assessed by using one way ANOVA (Statistica 8.0.).

Results and discussion
Exposure of *D. trunculus* adults and juveniles to 150µg/L PER for 72h produced a significant decrease in the total lipid content and polar lipid percentages of the gills Indeed, neutral lipids increased significantly following the elevation of triacylglycerols (TAG) and free fatty acids (FFA) fractions (Table 1). Several studies have demonstrated that TAG and NL increase significantly in aquatic organisms in contaminated sampling sites. Regarding fatty acid composition, a significant decrease ($p<0.05$) in SFA and PUFA of adults and in PUFA of juveniles was found after PER exposure, compared to controls (Table 1). These results may be due to oxidative damage caused by exposure to PER, evidenced by a significant elevation of LPO in the gill of exposed clams (juveniles and adults) (Table 1), potentially due to the antioxidant enzyme system not being able to totally eliminate O^-_2 and induce the production of LPO.

Conclusion The present investigation showed that PER, was able to generate lipid accumulation in the form of TAG and FFA, a decrease in total FA content, especially PUFAs and induce the production of LPO as an oxidative stress response in both adults and juveniles.

Tab. 1. Total lipid content, lipid classes, fatty acid composition and lipid peroxidation in gills of adults and juveniles *D. trunculus* exposed to 150 µg/L PER for 72 hours. Values with the different letters as superscript present significant difference ($p<0.05$), for adults (A, B); for juveniles (a, b).

		Adults		Juveniles	
		Control	Exposure	Control	Exposure
Total lipid contents (g/100g dry weight)		33.6±1.6^A	17.2±2.9^B	27.1±3.7^a	10.1±1.3^b
Lipid classes (% total lipids)	Cholesterol esterified	4.9±1^A	6.5±1.3^A	3.3±0.6^a	5.3±1.5^a
	Triacylglycerol	2.4±1.6^A	9.5±1^B	2.2±1.1^a	11.5±0.9^b
	Free fatty acids	16.1±0.9^A	19.8±1.1^B	13.5±1.4^a	19.7±0.6^b
	Cholesterol	22.3±1.7^A	15.8±2.8^B	28±1.5^a	21.2±1.9^b
	Neutral lipids	45.6±1.3^A	51.5±2.5^B	47±2.2^a	57.7±3.6^b
	Sphingolipids	5.8±1.1^A	3.7±0.3^B	4±0.6^a	2.1±1^b
	Phosphatidylethanolamine	16.2±1.2^A	13.1±0.2^B	16.5±1.1^a	10.1±1^b
	Phosphatidylisontol	10±0.3^A	8.1±0.7^B	9.8±1^a	5.7±1.4^b
	Phosphatidylserine	9.3±1^A	8.4±0.9^A	8.1±0.8^a	6.5±0.6^b
	Phosphatidylcholine	13.2±2.6^A	15.3±3.3^A	14.6±1.3^a	18.9±0.9^b
	Polar lipid	54.4±1.3^A	48.5±2.5^B	53.1±1.8^a	43.3±3.3^b
Fatty acids (g/ 100g dry weight)	SFA	11.5±1.1^A	6.4±1.3^B	4.7±0.5^a	4.1±0.9^a
	MUFA	3.7±0.4^A	2.6±0.3^A	1.5±0.3^a	1.4±0.1^a
	PUFA	8.16±0.7^A	5.9±1.2^B	9.9±3.3^a	2.8±0.4^b
	∑n-3	4.23±0.6^A	2.92±0.6^B	3.4±0.9^a	1.5±0.1^b
	∑n-6	1.74±0.1^A	0.7±0.2^B	3.1±1.1^a	0.7±0.1^b
Lipid peroxidation (Nmol MDA/g ww)		306.7±16.4^A	762.6±50.7^B	438.1±8.9^a	675.4±5.8^b

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