

**Table 4**Spirolides concentrations in digestive gland and remaining tissues ( $\mu\text{g SPX eq 13-desMeC kg}^{-1}$ ) during the detoxification period.

Time	Treatments	13,19-didesMeC	13-desMeC	13-desMeD
0	–	412.5 $\pm$ 9.1 (100%)	128.9 $\pm$ 1.2 (100%)	26.1 $\pm$ 1.7 (100%)
1	Without feeding	353.1 $\pm$ 39.6 (85.6%)	88.4 $\pm$ 10.5 (68.6%)	25.8 $\pm$ 1.7 (98.8%)
	With algal food	220.6 $\pm$ 21 (53.5%)	64.3 $\pm$ 10.6 (49.9%)	21.3 $\pm$ 1.2 (81.6%)
4	Without feeding	73.6 $\pm$ 6.1 (17.9%)	27.4 $\pm$ 1.5 (21.2%)	10.7 $\pm$ 0.1 (41%)
	With algal food	56.8 $\pm$ 3.5 (13.8%)	21.4 $\pm$ 1.2 (16.6%)	8.7 $\pm$ 1.3 (33.3%)
7	Without feeding	80.6 $\pm$ 2.9 (19.5%)	25.2 $\pm$ 2.8 (19.8%)	8.0 $\pm$ 0.1 (30.6%)
	With algal food	30.9 $\pm$ 0.7 (7.5%)	10.2 $\pm$ 0.2 (7.9%)	8.0 $\pm$ 0.1 (30.6%)
0	–	76.1 $\pm$ 3 (100%)	29.0 $\pm$ 1.7 (100%)	11.0 $\pm$ 0.4 (100%)
1	Without feeding	37.4 $\pm$ 1.5 (49.1%)	20.3 $\pm$ 1.9 (70%)	9.6 $\pm$ 0.1 (87.27%)
	With algal food	39.7 $\pm$ 1 (52.2%)	21.1 $\pm$ 0.5 (72.7%)	10.0 $\pm$ 0.3 (90.9%)
4	Without feeding	21.7 $\pm$ 0.2 (28.5%)	13.8 $\pm$ 0.6 (47.6%)	9.0 $\pm$ 0.1 (81.8%)
	With algal food	22.1 $\pm$ 0.2 (29%)	14.9 $\pm$ 0.2 (51.4%)	8.9 $\pm$ 0.1 (80.9%)
7	Without feeding	17.5 $\pm$ 0.4 (23%)	12.8 $\pm$ 0.5 (44.1%)	8.7 $\pm$ 0.1 (79.1%)
	With algal food	21.0 $\pm$ 0.7 (27.6%)	13.4 $\pm$ 0.7 (46.2%)	9.2 $\pm$ 0.1 (83.6%)

digestive glands generally contain 80% or more of Paralytic Shellfish Toxin (PST) body concentration (Bricelj and Shumway, 1998) in oysters. Similar results were observed for mussels (Bricelj et al., 1990), and scallops (Cembella et al., 1993; Choi et al., 2003) contaminated with PSP and for clams (Medhioub et al., 2010) contaminated with gymnodimines.

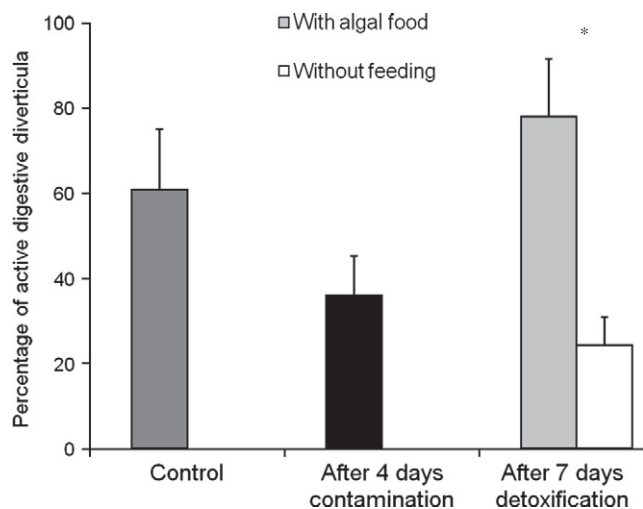
Various physico-chemical treatments including thermal and osmotic stress, electric shocks, pH decrease, ozonised seawater and chlorination have been tested to speed up detoxification of diarrhetic and paralytic toxins accumulated by bivalves. All proposed methods were either too dangerous, too long, too expensive, or modified organoleptic characteristics of the product. Effective ways to detoxify shellfish contaminated by emergent toxins such as spirolides remain to be developed. No attempt has been made so far to detoxify SPX-contaminated shellfish. In this work, food supply was selected as a detoxification process to speed up spirolide elimination. After 7 days detoxification, total spirolide content in digestive gland was found to be lower in oysters fed on *T.Iso* than in unfed oysters. This is in good agreement with previous field and laboratory studies highlighting that availability and quantity of non toxic food was the most important factor regulating PSP and DSP detoxifications (Bauder et al., 1996; Blanco et al., 1999; Guéguen et al., 2008; Haamer et al., 1990; Lassus et al., 1999, 2002, 2005; Marcaillou-Le Baut et al., 1993; Poletti et al., 1996; Sampayo et al., 1990). Similarly, experiments on gymnodimine (another cyclic imine)

detoxification by grooved carpet shell *Ruditapes decussatus* revealed that feeding clams with *T.Iso* accelerated gymnodimine detoxification (Medhioub et al., 2010). In the same way, Guéguen et al. (2008) showed that PST detoxification of Pacific oyster *C. gigas* fed on *Skeletonema costatum* diets optimised oyster detoxification in both digestive gland and remaining flesh. Also, phytoplankton abundance was associated with an increase of detoxification rate after DSP and PSP episodes (Blanco et al., 1997; Sampayo et al., 1990).

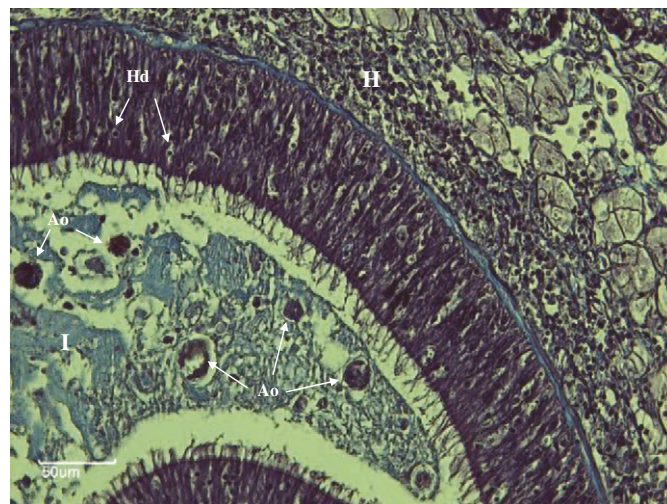
Conversely, detoxification of PST in purple clam, *Hiatala rostrata*, and lipophilic phycotoxin (okadaic acid) in mussels were unaffected by food availability (Chen and Chou, 2001; Svensson, 2003).

In Arcachon Bay, spirolide content in oyster (SPX-A and 13-desMeC) is generally reported as quite low, ranging from 0.6 to 5  $\mu\text{g 13-desMeC eq. kg}^{-1}$  total tissues, and rapidly declining after disappearance of the causative dinoflagellate (Amzil et al., 2007). Similarly, spirolide content rapidly decreased during the detoxification period in our controlled conditions. The final amount of spirolides obtained after detoxification is in the same range as that found in the natural environment (approx. 6  $\mu\text{g 13-desMeC eq. kg}^{-1}$  total tissues).

At the end of the detoxification period, total SPXs content was found in equal amounts in remaining tissues and in digestive gland. Slower detoxifications in remaining tissues (including gill, mantle, and foot) were in agreement with previous studies on other phycotoxins : gymnodimines-contaminated grooved carpet shell, *R. decussatus* (Linné) (Medhioub et al., 2010), PSP in king scallop *Pecten maximus*



**Fig. 3.** Percentage of active digestive gland tubules during the experiment. From left to right: reference before exposure to *A. ostendii*; after contamination with *A. ostendii* and finally after 7 days of detoxification (gray bar referring to fed oyster and white bar referring to unfed oyster). Results are expressed as mean percentages of active digestive gland tubules  $\pm$  confidence limits ( $n=10$ ). \* significant differences between treatment for  $p<0.05$ .



**Fig. 4.** Inflammatory response in the intestine of *A. ostendii*-exposed oysters. Aggregation of hemocytes in the connective tissue surrounding the intestine and hemocytes in diapedesis through the intestine epithelium. Hematoxylin-eosin stained paraffin sections (5  $\mu\text{m}$ ). (Ao) *A. ostendii* in intestine (I) of *C. gigas*, (H) surrounding hemocytes and (Hd) hemocytes in diapedesis.