The serpulid *Ficopomatus enigmaticus* (Fauvel,1923) as candidate organism for ecotoxicological evaluation of brackish and marine sediments

Matteo Oliva[1,2], Nicola Bigongiari[1], Ludmila Kozinkova[1],Chiara Manzini[1], Elvira Mennillo[2], Carlo Pretti[1,2]

[1] CIBM - Interuniversitary Centre of Marine Biology Livorno, Italy
[2]Department of Veterinary Sciences, University of Pisa, Pisa, Italy

Phone: +39-0586-807287
E-mail: oliva@cibm.it

Introduction: The aim of this study was to investigate the suitability of the marine polychaete serpulid worm *Ficopomatus enigmaticus* (Fauvel, 1927) as model organism in ecotoxicological monitoring of brackish and marine environments. *F. enigmaticus* is a fouling reef-forming species, easy to sample, dioecious with gamete spawning along different seasons in different salinity conditions. Due to its characteristics it could become a good candidate for bioassays. Spermiotoxicity and larval development assays were performed at four different salinity conditions (10-15-30-35 ‰) with the following reference toxicants: copper, cadmium, sodium dodecyl sulphate (SDS) and 4-n-nonylphenol (NP). The same protocols were also used for an environmental sediment elutriate evaluation, together with a canonical bioassay battery.

Methods: *F. enigmaticus* adults were sampled and maintained in aerated filtered (0.45 μm) seawater (T=21 °C). The release of gamete was forced by tube breaking [1]. Spermiotoxicity assay was conducted as reported for *H. elegans* [2]. The sperms were exposed to toxicant at a concentration of $8*10^6$ cells/ml, at selected salinity, for 20'. 1 ml of treated sperm solution was mixed with an egg suspension (250 cell./ml). Fertilization was carried out at S=30‰ and stopped after 90' by adding formaldehyde (4%). 100 eggs per replicate were counted and fertilization rate was calculated. Two or more-cell stages were considered as fertilized. Larval development assay was performed as reported for *G. cespitosa* [3]: about 300 fertilized eggs were exposed to 10 ml of different concentration of reference toxicants at the four different salinities for 48h (21±2°C). On a total number of 100 fertilized eggs per replica, the development rate, as number of normal developed trochophores, was calculated. All experiments ran in triplicate. The methods described above were adopted also for the ecotoxicological evaluation of sediment elutriates. Results were compared with other bioassays conducted on the same samples (*V. fischeri, P. tricornutum* and *P. lividus*)

Results: In Tab. 1 are reported all results obtained with spermiotoxicity and larval development assays with reference toxicants. The ecotoxicity evaluation of sediment elutriates showed a relevant effectiveness in the discrimination of different contamination levels of sediments.

Spermiotoxicity

Reference Toxicant		S=35 ‰	S=30 ‰	S=15 ‰	S=10 ‰
Cd^{2+} (mg/l)	EC_{50}	0.28	0.22	0.21	0.31
	C.L. 95%	(0.20-0.35)	(0.17-0.27)	(0.17-0.24)	(0.15-0.47)
Cu^{2+} (mg/l)	EC_{50}	0.096	0.08	0.08	0.2
	C.L. 95%	(0.07-0.12)	(0.04-0.13)	(0.07-0.10)	(0.14-0.26)
SDS (mg/l)	EC_{50}	2.1	1.76	2.16	1.95
	C.L. 95%	(1.56-2.65)	(1.08-2.44)	(1.26-3.05)	(1.11-2.78)
NP (μg/l)	EC_{50}	1.43	1.38	1.30	1.37
	C.L. 95%	(0.90-1.95)	(0.68-2.08)	(0.87-1.72)	(0.68-2.06)

Larval development

Reference Toxicant		S=35 ‰	S=30 ‰	S=15 ‰	S=10 ‰
Cd^{2+} (mg/l)	EC_{50}	0.79	0.77	1.07	0.5
	C.L. 95%	(0.59-0.99)	(0.59-0.95)	(0.94-1.20)	(0.39-0.61)
Cu^{2+} (mg/l)	EC_{50}	0.2	0.2	0.18	0.03
	C.L. 95%	(0.15-0.24)	(0.14-0.26)	(0.13-0.23)	(0.02-0.04)
SDS (mg/l)	EC_{50}	8.68	8.35	8.76	8.65
	C.L. 95%	(7.17-10.19)	(4.76-11.93)	(6.66-10.86)	(6.67-10.52)
NP (μg/l)	EC_{50}	6.86	6.80	6.03	6.18
	C.L. 95%	(6.56-7.16)	(6.10-7.50)	(5.86-6.19)	(4.60-7.78)

Tab. 1 Results for both spermiotoxicity and larval development assays, all reference toxicant used are reported with EC_{50} values and relative 95% confidence limits.

Discussion: *F. enigmaticus* showed a high degree of tolerance to lower salinities in terms of percent of success in fertilization and larval development. Metal ions showed the same trend of toxicity observed in both assays, copper appeared slightly more toxic then cadmium. NP appeared the compound with the highest toxic effect in both assays while SDS resulted to be the less toxic compound. As expected, the zygote stage exhibited a general higher resistance to all reference toxicants, if compared with spermatozoids. Preliminary assays showed that *F. enigmaticus* spermiotoxicity and larval development assay sensitivities to assessed reference toxicant are comparable with similar assays conducted with other species, some of which are commonly used for marine water and sediment toxicity evaluation [4] [5] [6] [7] [8]. Concluding, results obtained with *F. enigmaticus* assays on sediment elutriates underlined the suitability of this species in the monitoring of marine sediment toxicity.

References: [1]Hadfield et al. (1994) *Recent Development in Biofouling Control*: 65-74; [2]Gopalakrishnan et al. (2008) *Chemosphere* **71**:515-528; [3]Ross and Bidwell (2001) *Arch. Environ. Contam. Toxicol.* **40**:489-496; [4]Arizzi-Novelli et al. (2003) *Environ. Toxicol. Chem.* **22** :1295-1301; [5]Gopalakrishnan et al. (2007) *Arch. Environ. Contam. Toxicol.* **52**:171-178 ; [6] Gopalakrishnan et al. (2008) *Chemosphere* 71:515-528 ; [7]Jorge and Moreira (2005) *Ecotoxicol. Environ. Saf.* **61**:280-285; [8] *Mariani* et al. (2006) *Environ. Toxicol.* **21**:373-379.