REVERSIBILITY OF METAL INDUCED MALFORMATIONS IN SEA URCHIN EMBRYOS

Lorenzo Morroni [1]*, David Pellegrini [2] and Francesco Regoli [1]
[1] Department of Earth, Environment and Life Sciences, Polytechnic University of Marche, via Ranieri Monte D'Ago, 60100 Ancona, Itay - l.morroni@univpm.it
[2] ISPRA, Institute for Environmental Protection and Research, Piazzale dei Marmi 12, 57123 Livorno, Italy

Abstract
Trace metals are widespread pollutants in marine sediments. Exposure to these elements can induce well documented malformations during the early embryogenesis of sea urchin, but the reversibility of such effects is largely unknown. Our results showed the highest effects at pluteus stage, but the damage observed at 48 h after fertilization was mostly reversible. This study suggests the need to perform the sea urchin embryo test at different times of incubation, in order to discriminate between delay and block of embryogenesis, thus obtaining a more realistic interpretation on embryotoxicity potential of marine sediments.

Keywords: Ecotoxicology, Mediterranean Sea, Metals, Sediments, Echinodermata

Introduction
Trace metals are accumulated in marine sediment, constituting an important risk for living organisms. The sea urchin embryo test is one of the most sensitive and widespread bioassays to assess the ecotoxicological effects of marine sediments [1]. Although the sensitivity of sea urchin embryos to trace metals has been largely reported, no studies have assessed the possible reversibility of these effects during the early life stage. The aim of this study was to characterize and quantify the main anomalies induced by metals during embryo development of *Paracentrotus lividus*, investigating whether the damage is reversible.

Materials and methods
P. lividus embryo test was conducted with standardized methodology [2]. Moreover, different aliquots of the same pool of embryos were incubated with metals for 24 h and then allowed to continue developing in filtered control seawater for up to 72 h. Analysis of developmental defects was performed at 24 h, 48 h (standard time of incubation) and 72 h after fertilization both for washed and non-washed embryos, in order to understanding whether embryos exhibit block or delay in embryogenesis. The percentage of well-developed embryos (n=100) was recorded along with: 1) stage of development 2) occurrence of malformation.

Results and discussion
All trace metals to which the embryos were exposed showed a minimum of toxicity at 24 h, with an increase of the toxic effects at 48 h and a partial recover at 72 h (Fig. 1A). Pluteus stage appeared to be more sensitive than embryos at the end of gastrulation. The majority of embryos analysed at 48 h presented reversible anomalies, reaching the correct developmental stage at 72 h. This trend was particularly evident in embryos transferred to clean seawater at 24 h; despite no significant differences were observed at 48 with non-washed embryos, at 72 h washed embryos exhibited higher values of normally developed plutei (Fig 1B). In addition, specific malformations, time of occurrence and magnitude of effects was influenced by the considered metal.

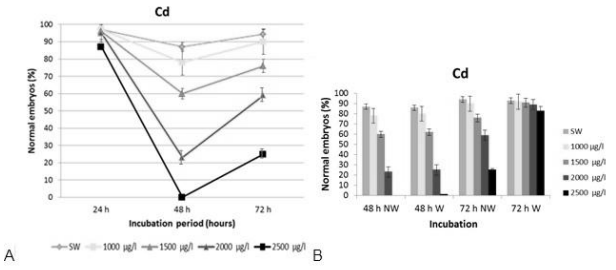

Fig. 1. A: Non-washed embryos: percentage of normal embryos (ordinate) at different times of incubation (abscissa) for each Cd concentration (coloured lines). SW: control. B: Histograms refer to non-washed embryos (NW) and to washed embryos (W) 48 h after fertilization and 72 h after fertilization.

Conclusions
In conclusion, results suggest the need to perform the sea urchin embryo test not only at the standard time of 48 hours, considering also the effects on pre-pluteus stage and the temporal evolution of these effects, allowing a more realistic estimation of the ecotoxicity of environmental matrices.

References
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