

Interactive comment on “Effects of increased $p\text{CO}_2$ and temperature on trace element (Ag, Cd and Zn) bioaccumulation in the eggs of the common cuttlefish, *Sepia officinalis*” by T. Lacoue-Labarthe et al.

Anonymous Referee #2

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General comment: This study addresses problems potentially arising from global warming and reduced pH (simulating CO_2 driven ocean acidification), which modulate trace metal uptake into the eggs and embryos of cuttlefish *Sepia officinalis*. Reprotoxicity in organisms which play a key role in the marine foodnet such as cuttlefish strongly influence sustainability of populations and communities. A mechanistic understanding of effects of increasing temperature and low pH on the bioavailability of trace metals as well as their bioaccumulative compartmentation inside of developing eggs and embryos is the basis for further risks assessment studies of metal toxicity in chang-

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ing oceans. This pioneer approach is a good starting point to expand the research towards ecotoxicological aspects in collaboration with experts in this field. Cell biological and toxicological aspects should be addressed in future such as metal uptake via membranes, metal binding affinity to metallothioneins and the role of specific MRP transporters for conjugated metals during embryogenesis of marine invertebrates.

Specific comments.

Abstract and Introduction: Please, explain, why you choose exactly Ag, Zn and Cd? Are these metals especially relevant in the marine environment, in general, or in the geographic area of your interest or in the habitats of cuttlefish? Please, comment.

You mention egg shell hardening due to pH-induced seawater polymerisation. Throughout the genera from plants to humans eggshell hardening occurs due to the acrosom reaction of the sperm in order to avoid lethal polyspermy. Please, add more recent references addressing the processes of membrane hardening. That the eggshell protects the embryo against mechanic and chemical injury is a secondary and certainly true aspect.

M&M: Under item 1 you mention the use of Mediterranean seawater without any further information on 1. source of water 2. chemical analysis of the water with respect to metal contamination and other chemicals. Please, explain why you used water from the field and no artificial seawater in order to exclude contaminating factors.

The exact design of the experiment is not evident for the reader (number of replicates?). Furthermore, please explain, how many eggs/individuals you used. In the text it is written that you exposed a number of 300 eggs randomly assigned to six bottles with seawater. Looking at the data presented in Figure 3 it appears that you analysed 703 individuals (13 time points, each 3 eggs, 3 pHs, 3 metals, 2 temp.).

With respect to the concentrations of metals it would be interesting to receive information on which basis these concentrations have been chosen. Do they correspond to

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natural levels found in seawater or in biota?

Discussion: Please, explain what you mean when you write about anti-stress peptides produced by the embryo ?

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