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Interactive comment on “CellTracker Green labelling vs. Rose Bengal staining: CTG wins by points in distinguishing living from dead anoxia-impacted copepods and nematodes” by M. Grego et al.

Anonymous Referee #3

Received and published: 15 April 2013

Review “CellTracker Green labelling vs Rose Bengal staining: CTG wins by points in distinguishing living from dead anoxia-impacted copepods and nematodes” by Grego et al

The paper compares the efficiency of the use of two methods of staining to separate dead and living organisms in normoxic and anoxic sediments. The two methods are Rose Bengal and CellTracker Green and the selected organisms are nematodes and copepods. The authors conclude that the CTG appears more efficiency than RB to discriminate dead and living organisms. The results presented here are based on an

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experiment carried out in the Northern Adriatic Sea, using a benthic chamber to create the anoxic conditions. I find the paper very interesting and well organized. However, in my opinion, some points should be revised and re-phrased because they are not clear and supported by the experiment carried out.

My main concern is on the samples treatment. CTG has been incubated for 12h and then samples have been fixed while RB has been added into fixed samples. In my opinion, samples should be treated following the same procedure since the aim of this investigation is to find the best method to discriminate dead and living organisms. So also samples with RB should be incubated for 12h and then fixed.

I find also not correct that the authors used this experiment to “examine fine-scaled and short-term disturbance phenomena” since the experiment is based on a comparison between meiofaunal assemblages from anoxic and normoxic sediments collected at the same time. The experiment does not include different sampling times that describe the evolution of the system from normoxic to anoxic conditions. I think that this point should be clarified in the abstract, in the introduction and conclusion.

Specific comments: Abstract Line 14, delete “Surprisingly”, since It is known that RB stained all material containing proteins, in the case of nematode, the presence of different material on the cuticle can contribute to the overestimate of the living organisms. Lines 15-18, I am not sure that the experimental set up allow to “resolve the course of events”. In the present investigation anoxic and normoxic conditions are compared. There is not an analysis between a T0 and T end of the experiment.

Introduction Pages 3-4, lines: 94-96, At the end of the introduction the aim of this investigation should be better explained. In particular in the present form, the aim seems to be related to “the changes in copepod and nematode community composition can occur in the course of hour”. Again, the experimental set up allow to investigate the differences between anoxic and normoxic sediments but do not allow to discriminate differences during the changes in the environmental conditions.

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Conclusion Page 10, lines: 334-335: please rephrase. I would suggest to remove references from the conclusions. Tables 1 and 2; please check the value of p level. There is a typo in both legends.

Interactive comment on Biogeosciences Discuss., 10, 2857, 2013.

BGD

10, C964–C966, 2013

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