

## ***Interactive comment on “Functional spatial contextualisation of the effects of multiple stressors in marine bivalves” by Antonio Giacoletti and Gianluca Sarà***

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REVIEWER #1 Reviewer wrote: This paper explores the implications of environmental stress (OA and hypoxia), as determined in lab experiments, on the growth and reproductive potential of mussels in two locations of the Mediterranean with simulations based on a dynamic energy budget model. The study capitalizes on the potential of DEB models to integrate the impacts of multiple environmental drivers on organismal level outcomes, including growth, reproduction, time to maturity, rates of feeding and respiration, and so on. This approach is powerful in potential and the application is new. However, there are some important shortcomings, especially in the way the model is

C1

parameterized. Also, I find the lack of some form of quality assessment problematic. Author's reply: First, we thank the Reviewer #1 for the effort in providing his/her suggestions to the original version of our ms. Author's changes: Apart from all possible not clear parts that we accordingly improved, we followed his/her suggestion in the hope to have increased the quality of this ms.

Reviewer wrote: It is very annoying that all sections consist of a single paragraph. Did something go wrong with the formatting of the manuscript? Author's reply: We appreciated the referee's suggestion about the different paragraphs, but we believe that the current structure is already sufficiently sectioned; to increase the number of paragraphs or sub-paragraphs can only increase the text fragmentation which may limit the logical flow of the text. Reviewer wrote: The author's use of respiration measurements as a proxy for DEB maintenance costs is problematic. In the DEB framework, respiration is emphatically not the same as maintenance, but also include energetic overheads, such that of growth. Respiration is a function of the commitment rate in DEB, of which maintenance could be a minor part, depending on size and nutritional status of the animal. Author's reply: We acknowledge that respiration does not include only maintenance, but also include energetic overheads, such as growth. Nevertheless, there is no way - to our knowledge - to measure the different contribution of every energetic components apart from to experimentally measure oxygen consumption as a proxy for metabolism. Also, the proposed approaches measuring indirectly the  $[\dot{E}]$  values (e.g. van der Veer et al., 2006; Ren and Schiel 2008), are not feasible in the context of the present experimental asset. While this approach is not experimentally feasible when assessing the effect of stressors on the energy budget, the only way to indirectly provide an estimation of the effect of disturbance is through the Jagger et al. (2016) approach which is based on the stress factor "s". Thus, after estimating the effect induced by a treatment on the oxygen consumption, that in the present case study was expressed as a percentage variation, we summed/subtracted the energetic amount due to the effect of a stressor to the species-specific  $[\dot{E}]$  values of *M. galloprovincialis* then we run our models. However, we thank the referee for highlighting this point whose importance

C2

was addressed in the Discussion section of this ms. as we believe that is crucial to increase our understating on how we can mechanistically assess the effect of disturbance on individual performances through the DEB model. All these limitations show how much is important to date to increase the experimental and theoretical research effort in order to unravel this point, which is increasingly crucial to get realistic answers to management questions in a context of environmental change.

Reviewer wrote: In addition, oxygen deprived mussels, and possibly mussels enduring stress of hypercapnia, are able to use anaerobic metabolic pathways to fulfil their maintenance requirements. If stress increases maintenance requirements, one would expect respiration rates to increase with increasing stress intensity. However, we see the opposite happen (see Fig 2). I think this is likely due to the fact that stressed mussels have their shells closed more often than unstressed conspecifics (see Fig. 1), and thus ingest less food. Less food leads to a lower energy reserve buffer and therefore a lower rate at which reserves are committed. Author's reply: As reported in our results and Fig. 2, maintenance requirements in accordance with respiration rates, decreases with stress in agreement with what is reported in the current literature. We are sorry with Reviewer #1 and with all readers as we made a mistake in writing the text commenting the figure 1 (we wrote wrongly "opened" instead of "closed"). Actually, our mussels increased their openness with the increasing stressful conditions. At the present stage, we are not able to provide information about the amount of ingested food under different treatments and then we are not able to infer on the effect of openness degree on energetic performances. Author's changes: Figure was fixed according to both referee's suggestion, and also the text in the paragraph has been rephrased accordingly.

Reviewer wrote: I suggest the authors change the maximum assimilation rate parameter of their model based on their behavioural observations and leave the maintenance rate parameter unchanged. Author's reply: We appreciated the referee's suggestion but we prefer to focus on both the assimilation efficiency and the metabolic effect

C3

(through pM) as i) the main effect of acidification seem to be exerted on metabolism as widely reported in the current literature, and ii) also to show that our mechanistic DEB approach can be really effective in measuring the multiple stressor's effect on LHs. Author's changes: Thus, we enlarged the discussion on these points to include possible shortcomings deriving from the fact that the stressor's effect on maintenance is not still well-experimentally measurable.

Reviewer wrote: The simulations suggest that unstressed mussels only grow to 3 cm in length and do not reproduce in Palermo. This seems implausible. How long do real mussels get in Palermo? Do they reproduce? How sensitive are the simulation results to the particular choices of parameter values? The authors do not reflect at all on the reliability of their assessments, which I find troublesome, especially given the politicized context of the subject matter. Author's reply: Actually, to enlarge the discussion about the magnitude of effects at local level could be not influent for our purposes, although our results are in line with the environmental and trophic conditions reported in section 2.5: "Both sites were chosen as they represent two opposite temperature and food conditions for mussel growth in Italy... etc.". *M. galloprovincialis* in Sicily is observed to be limited by oligo-trophic conditions although it grows in highly trophic-enriched areas such as harbours or under Integrated Multi-Trophic Aquaculture (IMTA) conditions (Sarà et al 2012; 2013b, Giacoletti et al. 2018 in press JEMA) which supports what we gathered in the present ms. through the DEB simulations.

Reviewer specific comment n. 1 Reviewer wrote: Title. Functional spatial contextualization sounds impressive but I've no clue what it could mean. Also, the manuscript deals with only a single species; the title is too general. Author's changes: We agreed with Reviewer's #1 point and changed the title.

Reviewer specific comment n. 2 Reviewer wrote: L27-33 Split up sentence. Author's changes: Sentence was splitted up accordingly.

Reviewer specific comment n. 3 Reviewer wrote: L35 (and elsewhere) Put reference in

C4

the proper place of the sentence Author's changes: All references were checked and put in proper spaces.

Reviewer specific comment n. 4 Reviewer wrote: L40 'lager'? Author's changes: Changed with "larger".

Reviewer specific comment n. 5 Reviewer wrote: L68-70. This is a strong statement and should be substantiated with references. BTW, the only 2 papers using DEB in a OA context I'm aware of are 10.1111/gcb.12547 and 10.1016/j.jembe.2015.09.016 Author's changes: References regarding the effect of OA on functional traits such as feeding and assimilation, and on maintenance costs has been added accordingly.

Reviewer specific comment n. 6 Reviewer wrote: L72 the DEB [p\_M] parameter does not relate to assimilation Author's changes: The sentence was rephrased accordingly.

Reviewer specific comment n. 7 Reviewer wrote: L83 articulated ! consisted of Author's changes: Changed accordingly.

Reviewer specific comment n. 8 Reviewer wrote: Section 2.4 contains material that should go in 2.3 (or combine the sections). Author's reply: Section 2.4 refers to assimilation efficiency, while section 2.3 to oxygen consumption measures, so we consider not easy to combine both sections as we may incur in the risk to reduce the readability of this section.

Reviewer specific comment n. 9 Reviewer wrote: I didn't get how the authors calculate the assimilation efficiency. Author's changes: A detailed explanation on how the assimilation efficiency was estimated, was added with supporting references.

Reviewer specific comment n. 10 Reviewer wrote: Section 3.1 belongs in the Materials and Methods Section. There is no need for a statistical analysis. Delete Table 2. Author's changes: Table 2 was deleted according to both referee's suggestion and details were moved in the Materials and Methods Section.

Reviewer specific comment n. 11 Reviewer wrote: Combine Sections 3.2-4. There

C5

is no need to duplicate in the text what is already presented in the figures. The percentage of closed valves is simply 100 – percentage opened valves, so don't mention the former. I don't understand why the error measures differ so much, though. Author's changes: We agree not to duplicate in text what is already presented in figures, and we worked to avoid this replication. Following Reviewer's #1 suggestion we also expressed the percentage of closed valves 100 – percentage opened valve. Instead, merging the sections can increase the risk of confusion in the reader as section 3.2 is about behavioural observations, while the other two are about physiological measurements.

Reviewer specific comment n. 12 Reviewer wrote: Section 2.7 is incomprehensible for people without DEB modeling background. Include a figure and references to overview texts (e.g. Kooijman's book, Nisbet et al JAE, Sousa et al, and/or most recently Jusup et al Physics of Life Reviews 20:1-39). Author's changes: We agree with Reviewer #1 that section 2.7 is difficult for someone without a DEB modelling background, and in order to made it more clear we added the suggested references and rephrased some parts.

Reviewer specific comment n. 13 Reviewer wrote: L263 addictive ! additive. The way the authors use 'additive' is confusing. Additive refers to impacts that can be summed, like 1+1=2, an unlikely situation with nonlinear models, such as DEB. Author's changes: Rephrased following suggestions to "with a progressive contribution of hypoxia".

Reviewer specific comment n. 14 Reviewer wrote: What are the initial conditions of the simulation runs? Author's reply: Results of simulation performed with unstressed organism were already reported in Table 5, while model parameters have been reported in Table 1.

Reviewer specific comment n. 15 Reviewer wrote: What is the rational for the choices for the frequency of events? Author's reply: While we are aware that hypoxia events are more frequently during summertime, we decided to not apply any timing and frequency

C6

scheme to simulate hypoxia event's occurrence according to many papers published across the recent literature(Crain et al. 2008 Ecology Letters; Miller et al. 2009 PNAS).

Reviewer specific comment n. 16 Reviewer wrote: From Table 5 remove data that are already presented in Figure 3. Round off # of eggs to 6.74e6. Units of frequency should be 1/time Author's changes: Table 5 was corrected accordingly.

Reviewer specific comment n. 17 Reviewer wrote: Figure 3 label y axis 'Change relative to control' Author's changes: Figure was corrected accordingly.

Reviewer specific comment n. 18 Reviewer wrote: L294 delete 'formally' Author's changes: Deleted.

Reviewer specific comment n. 19 Reviewer wrote: L295 delete 'compensatory' and change contrast to compensate Author's changes: Rephrased.

Reviewer specific comment n. 20 Reviewer wrote: L299 suppressed feeding activity Author's changes: Changed.

Reviewer specific comment n. 21 Reviewer wrote: L304 what is crossing effect? Author's changes: Sentence has been rephrased accordingly.

Reviewer specific comment n. 22 Reviewer wrote: L306 on! over. 'that' doesn't refer to anything Author's changes: Rephrased.

Reviewer specific comment n. 23 Reviewer wrote: L333 sustainable and reliable ! practical Author's changes: Changed.

Reviewer specific comment n. 24 Reviewer wrote: L337 write out TW, TRO and TM Author's changes: Written out accordingly.

Reviewer specific comment n. 25 Reviewer wrote: The readability of the manuscript would improve if there were fewer references. Remove unnecessary repetitive references. Author's changes:: All the references has been checked and unnecessary and repetitive ones have been deleted accordingly.

C7

Please also note the supplement to this comment:  
<https://www.biogeosciences-discuss.net/bg-2018-13/bg-2018-13-AC1-supplement.pdf>

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Interactive comment on Biogeosciences Discuss., <https://doi.org/10.5194/bg-2018-13>, 2018.

C8