

## ***Interactive comment on “Differential response of planktonic primary, bacterial, and dimethylsulfide production rates to vertically-moving and static incubations in upper mixed-layer summer sea waters” by M. Galí et al.***

**Anonymous Referee #1**

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### GENERAL COMMENTS

This paper explores, through an experimental approach (four experiments performed in oligotrophic Mediterranean coastal and open stratified waters), the influence of a static (three fixed-depths) and dynamic (experimental vertical movement) gradient of solar radiation exposure within the upper mixed layer (UML) on two groups of response variables: one acting as phyto- and bacterioplankton physiological indicators (e.g. algal pigments, Fv/Fm, proportion of bacteria with intact membrane integrity) and the other group with biogeochemical relevance (e.g., primary and bacterial production,

C3495

gross and net biological DMS production and photolysis). The response variables were measured by means of properly applied and described methods. Authors posed two interesting questions falling within the scope of BG, one photobiological and the other biogeochemical-methodological, which were quite well elucidated through a nice integration (data analysis, presentation, and discussion) of the experimental results obtained from the four experiments.

I find that paper is well written and contributes novel information on how dynamic solar exposure within ULM slightly disrupted the photoinhibition and photoacclimation processes associated to vertical gradient of ultraviolet radiation in marine UML. Nevertheless, I have found some deficiencies, indicated in the detailed comments below, which can be easily remedied to strengthen the final version of the paper.

### SPECIFIC COMMENTS

Title:

I propose (optionally) the following title because, as acknowledged by authors in conclusions, “the irradiance dose-response in mixing bottles was distinct (though subtle) in each of the processes measured. . .”; besides, it may reinforce the idea of static vs. dynamic light field: “Subtle differential response of planktonic primary, bacterial, and dimethylsulfide production rates to static vs. dynamic incubations in upper mixed-layer summer sea waters”

Abstract:

OK, it provides a concise and complete summary. . . But, in order to improve consistency throughout the paper, introduce the word “subtle” before “disruption” in pg. 8853, line 15.

Introduction:

Good review of the scientific background, and interesting questions posed. However, I do not feel entirely comfortable with the statements in pg. 8855 lines 18 to 21. I think

C3496

that mixing treatment resembles more realistic conditions than fixed-depth incubations within UML (because water and organisms indeed experience vertical movement and dynamic light exposure in real UML), even though the experimental mixing times were faster than current mixing times, according to calculations and statements in pg. 8861, lines 10 to 11. Hence, I would include this point of view in the Introduction (e.g., in pg. 8855, after lines 18 to 21).

#### Methods:

Good description of suitable materials and methods. Some caveats concerning descriptions are indicated below:

- Pg. 8856, line 4: Was temperature of samples controlled during their transport to lab and the pier? Make a statement about it.
- Pg. 8856, line 10: Describe here the incubation bottles (were the 2.3L Teflon bottles?) and number of replicates per treatment.
- Pg. 8856, lines 12-16: Mixing times were distinct between coastal (C1, C2) and oceanic experiments (O1, O2). This introduced a different fluctuating light regime between these environments, even though in C1 and C2 the bottles were incubated at shallower depths to approximate the equivalent in situ optical depths. This could make the two types of environments less comparable between them, and perhaps making less appropriate the pooling of results from all experiments. . . This would deserve some discussion, particularly regarding to the apparent different behavior shown by C1 for most of variables (even different to C2).
- Pg. 8857, lines 24 to 26. Describe better how this calculation was made. . . e.g. were expressed as rates per hour or per (incubation) period? How was integrated the 2 h of incubation under dark in the presence of tracer with the prior incubation-time under light?
- Pg. 8859, line 22: Replace "...a photolysis rate constant (...) was used at each

C3497

experimental location to correct. . ." with "...a distinct photolysis rate constant (...) for each type of experimental location (i.e. coastal or oceanic) was used to correct. . ."

#### Statistical analysis:

- Pg. 8860, line 2: How the integration was calculated? Please, detail further.
- Pg. 8860, line 4: Please, report n or degrees of freedom.
- Pg. 8860, lines 5-6. It would be worth to use modern robust statistical methods instead or complementarily to classical non-parametric statistic to corroborate differences among treatments when assumptions for parametric tests are not met (e.g. ANOVA based on percentile bootstrap method; see Erceg-Hurn & Mirosevich 2008, Rose et al. 2009).

#### Results and discussion:

Good description of results and discussion, conclusions, and the arrangement of the sections. Nevertheless, I miss a discussion about broader ecological implications of the results. I feel the valuable responses to solar radiation found, particularly of variables with biogeochemical relevance (e.g. primary and bacterial production, DMS production. . .) through the depth-gradient (fixed incubations) and the subtle effects of mixing, deserve a more extensive discussion focused on their implications in the context of global warming, and within theoretical frameworks of (controversial) CLAW hypothesis, summer DMS paradox, and Earth-system theory (after Vallina & Simó 2007; Quinn & Bates 2011, Lana et al. 2011, 2012, Galí et al. 2013). Thus, as an example, the results found at surface and middle static incubations jointly may mimic the scenario of expected prolonged shallower stratification due to global warming, confining plankton long within hypothetical photoactive UVR damage layer (Fig. 1). In this way, (i) the maximum PPp found at middle depth, offset by mixing (resembling values from surface depth, i.e. subjected to inhibition), (ii) the absence of significant variation with depth (and mixing) of LIR measured under complete light exposure in presence

C3498

of tracer (Fig. 5B) that may be judged as very realistic measurement of bacterial activity, and (iii) the sharp vertical gradient of gross and net biological DMS production (increasing with irradiance, but largely compensated by DMS photolysis), with a neutral or slight reduction due to mixing, are results that, in overall, give room to discuss in the context of shallower stratification (global warming) and the CLAW hypothesis, particularly after controversies introduced by Quinn & Bates (2011). In this line, not only DMS but also organic matter (dissolved and particulate, of biological origin) or even (volatile and non volatile) photodegradation products of DMS (also of biological origin) can affect the formation of cloud condensation nuclei (by bubble bursting at the ocean surface) at large scale. The underlying idea of this claim is that the integrated operation of biotic and abiotic variables can reinforce regulation of Earth-system (after Cresser et al. 2008, Kleidon 2010, 2012).

I strongly encourage authors to include some of these aspects in discussion to reinforce the implications of their results.

#### References:

Good bibliographic review in quantity, quality and actuality.

#### Tables and Figures:

- Table 1: Include between parentheses absolute or percentage values of biomass of the “dominant phytoplankton (biomass)” within the field “Initial sample characteristics”.
- Fig. 3: Display axe title and units; also report R-squared and significance (p-value) of regression.
- Figs. 4 and 5: Why ANOVA results (p-values) and multiple comparisons are not included in Figs. 4E to I, and 5B to C)? If all variables (except DMS production rates) were measured in duplicate incubation bottles, as stated in pg. 8859 lines 24-25, there are enough variability to perform the statistics... In figure captions (Fig. 4) state that different letters indicate significant differences ( $p < 0.05$ ) between treatments from post-

C3499

hoc analysis.

- Fig. 7: Report values of R-squared, slope and significance of regression, for fixed-depth and also for mixing (either alone or fixed-depth + mixing) incubations, in order to show which extent mixing treatment disrupted photoacclimating and photodamage processes.

#### TECHNICAL CORRECTIONS

- Pg. 8854, lines 22-23: Replace citation Helbling & Villafañe 2013 with Helbling et al. 2013.
- Pg. 8859, line 6: Is it “cell-permeant” instead of “cell-permanent”?
- Pg. 8859, line 6: In “. . . SybrGreen I (Molecular Probes, Eugene, OR). . .” include the acronym SGI within those parentheses.
- Pg. 8867, line 25: What is the difference between DMSPt and DMSP (DMSPt was not defined).

#### \*\*References cited in the review:

Cresser MS, Aitkenhead MJ, Mian IA (2008) A reappraisal of the terrestrial nitrogen cycle: What can we learn by extracting concepts from Gaia theory? *Science of the Total Environment* 400: 344-355. DOI:10.1016/j.scitotenv.2008.06.047

Erceg-Hurn DM, Mirosevich VM (2008) Modern robust statistical methods. An easy way to maximize the accuracy and power of your research. *American Psychologist* 63: 591-601. DOI:10.1037/0003-066X.63.7.591

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C3500

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Lana A, Bell TG, Simó R, Vallina SM et al. (2011) An updated climatology of surface dimethylsulfide concentrations and emission fluxes in the global ocean. *Global Biogeochemical Cycles* 25: GB1004 DOI:10.1029/2010GB003850

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Rose JM, Feng Y, DiTullio GR, Dunbar RB et al. (2009) Synergistic effects of iron and temperature on Antarctic phytoplankton and microzooplankton assemblages. *Biogeosciences* 6: 3131-3147.

Vallina SM, Simó R (2007) Strong relationship between DMS and the solar radiation dose over the global surface ocean. *Science* 315: 506-508. DOI:10.1126/science.1133680

End of review —

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