

To the editor,

We hereby present our replies to the Interactive comments by the two reviewers. We have closely followed their recommendations and have proceeded with appropriate revisions throughout the paper. Below, we reply to the reviewers' comments one by one, with relevant reference to modifications in the text, where needed. In the revised version all the changes are highlighted in colour to facilitate location of the new text. We trust that you will find that the revised version fully accounts for the comments and concerns of the reviewers and we look forward to hearing from you soon.

On behalf of all authors

Anna Lagaria

Interactive comment on “The effects of nutrient additions on particulate and dissolved primary production in surface waters of three Mediterranean eddies” by A. Lagaria et al.

Anonymous Referee #1

General

1. RV: Lagaria and co-workers report in this new BOUM manuscript the effect of inorganic nutrient additions on the partitioning of primary production into the particulate and dissolved fractions. It is still quite infrequent to include the latter, potentially important carbon flux, especially in oligotrophic environments such as open Mediterranean waters. I would highlight two interesting results. First, their initial percent extracellular release (PER) values were relatively low (9-18%) and not significantly different between sites that were expected to differ, at least according to their location along the well-described east-west gradient of increasing oligotrophy in the Mediterranean. By the way, this gradient may well be general but it was not evident at all in this study (opposite results in Table 1 are worth of further explanation).

Reply: Firstly, it is important to note that the certain similarities rather than differences among the three selected anticyclonic eddies was one of the prominent findings in other studies too, carried out within the BOUM project. In particular, we refer to the paper by Christaki et al. (2011) where *in situ* stocks and rates were studied within the entire surface layer, and precisely in the 0-150 m depth in the core of the eddies. The results in that paper showed that these three eddies diverted from the W-E gradient by displaying lower stock values compared to adjacent stations outside the eddies and by presenting no significant differences among them in the rates measured, such as PP, BP, GPP, NCP.

The text has been modified and the following is added in the paper (section 4.2):

“The initial conditions of the mixed layer (8 m depth) in the three eddies were oligotrophic and no significant differences between GPP and DCR were observed, indicative of equilibrium between gross production and respiration. Furthermore, *in situ* measurements over the euphotic zone in the three eddies have shown that the west-east gradient was not recognizable in terms of integrated primary and bacterial production rates among the three sites and that gross production roughly balanced respiration (Christaki et al., 2011). These findings were explained by the fact that the centre of established anti-cyclonic eddies are known to be zones of nutrient depletion with low rates of biological activity compared to surrounding areas (e.g. Mouriño-Carballido, 2009).”

Moreover, Lopez-Sandoval et al. (2011) during the same cruise measured *in situ* PPp, PPd over the euphotic zone and did not find significant differences in PER (calculated on integrated data) among the three gyres. The text now reads (section 4.1):

“During the BOUM cruise, PPp and PPd were also determined *in situ*, along vertical profiles. *In situ* PER, in the form of euphotic layer-integrated data, averaged 37% and no significant differences were observed among sites A, B and C (López-Sandoval et al., 2011).”

However, we have to point out that, comparisons of our results to the related papers of the BOUM special issue cannot be straight forward since our data derive from a single depth in surface waters. We believe that experimental results of bioassays (nutrient additions) should rather complementary to the description of *in situ* vertical profiles than being compared to them.

2. RV: Secondly, they found strong evidence against the claimed P-limitation in the Mediterranean (work by Thingstad and colleagues). The addition of phosphorus alone did not enhance primary production or decrease PER values as hypothesized by, among others, Obernosterer and Herndl 1995, MEPS 115: 247-257, please consider this paper in your study). However, in my opinion none of these two findings is sufficiently discussed. This is my first requirement of any subsequent revision.

Reply: The associated paper of Tanaka et al. (2011) fully describes and discusses the lack of P-limitation in these microcosm experiments. The paper by Obernosterer and Herndl (1995) is now considered. We also added the following discussion on this issue, it now reads (section 4.1):

“Theoretically, in the Mediterranean PER should be enhanced under conditions of P-deficiency since depletion of phosphate constrains new cell production inducing the release of dissolved photosynthate compounds by phytoplankton (Baines and Pace, 1991; Nagata, 2008). This was actually confirmed in a study with phytoplankton cultures, where PER was higher under phosphorus-limited conditions of skewed N:P ratios compared to N-limited or N:P balanced conditions (Obernosterer and Herndl, 1995). Interestingly, in our study we did not observe any

decrease in PER with the addition of phosphorus, suggesting a lack of P-limitation. The apparent lack of P-limitation in this experiment is extensively discussed in Tanaka et al. (2011). Moreover, no significant PER variations were observed between the +N and +P treatments (Fig. 1c). It seems that, under ultra-oligotrophic conditions prevailing during the stratified period, limitation by a single nutrient and/or co-limitation are likely in a delicate balance, meaning that addition of one nutrient will quickly push limitation towards the next limiting nutrient (Thingstad et al., 2005; Tanaka et al., 2011). Consequently, the unchanged PER in the +N and +P treatments could be a result of complex initial conditions with perhaps near co-limitation of N and P.”

3. RV: Carbon requirements of heterotrophic prokaryotes were only (very) roughly estimated and this should be clearly stated in the abstract and elsewhere in the text. I may agree that bacterial respiration (BR) should lie between 50% and 100% of total community respiration (CR) but this is certainly too large a range so as to derive sound conclusions. I can envision that statistics are difficult to apply to Table 5 data for the aforementioned reasons, but the authors should then be much more cautious when making statements of the relationships between the (assumed but not demonstrated) degree in oligotrophy and BCD:PP ratios in the +P treatments.

Reply: We modified the text to be more cautious about the estimation of BCD. We also calculated the BGE based on the assumption that BR was 50% or 100% of CR and compared with previous studies. The text now reads (section 4.2):

“Since BR was not directly measured in our study, we estimated BCD assuming that BR is 50% or 100% of DCR, based on the range of values previously reported for the Mediterranean (Lemée et al., 2002; Gonzalez et al., 2003; Navarro et al., 2004). The respective initial BGE would then be on average $15\pm 2\%$ (when BR is 50% DCR) or $8\pm 2\%$ (when BR is 100% DCR) for the three eddies. Generally, in oligotrophic environments BGE is low (<10%-25%, del Giorgio, 1997). Previous studies have reported BGE to be 2-8% in the NW Mediterranean coastal and offshore waters (Gasol et al., 1998; 7% in Almeria-Oran front in Sempéré et al., 2003). It seems, therefore, that BR was likely at least 50% of DCR and even the assumption of 100% still results in a plausible BGE.”

And, in section 4.2, it now reads:

“In the microcosms where net autotrophy was observed ($NCP > 0$, all treatments at site A, +NP at sites B and C, Fig. 2), the carbon-converted GPP and/or PP_{total} , was sufficient to sustain BCD. When the total community was metabolically balanced ($NCP \approx 0$, e.g. in +N), the carbon ratios varied in a relatively narrow range around 1, from 0.5 to 1.4 (Table 5). Finally, whenever the microcosms displayed net heterotrophy ($NCP < 0$, e.g. in +P at sites B and C, Fig. 2), GPP and PP_{total} were not sufficient to supply the BCD, except for PP_{total} at site B with BR assumed as 50 % of the DCR (Table 5).”

4. RV: More importantly, the experimental design to estimate dissolved primary production (PPd) is slightly flawed. Unlike particulate primary production (PPp), PPd was only measured in one of the triplicate microcosms and the authors are surely aware that PPd is usually much more variable than PPp. In my opinion, this methodological constraint importantly affects all subsequent analysis. Also, the fact that control PPd and PER values at station A were below “detection limits” (sic) compromises any comparison between sites when total number of experiments were 3! Similarly, the large errors (standard deviations) associated with O₂ measurements (Table 2) preclude drawing significant conclusions about differences between sites.

Reply: As we have noted in section 2.2.3 “because of the time constraints of sample treatment, dissolved primary production rate (PPd) was measured only in one of the triplicate microcosms of each series”. Furthermore, we could not change the fact that the BOUM cruise focused on only 3 eddies and consequently we have only 3 microcosm experiments. We can only add to our credit the following points: these 3 anticyclonic eddies were situated in 3 distinct Mediterranean basins; they are then representative of the summer situation and the measurements were repeated three times in each experiment (day 0, day 2 and day 4) and finally end up with 24 data points. Finally, we fully recognize the weaknesses of our work -which is mainly due to analytical constraints in oligotrophic waters- but we believe that these data are worthy of publication since (again because of these constraints) data of this kind are almost inexistent in open Mediterranean waters and, while not ideal are of great use for future studies.

5. RV: Stating that 10-20% PER values “closely approximated” 30% (López-Sandoval et al. 2010) is largely missing the point. Please re-write and avoid ambiguous statements such “(PER values in the microcosms): were reasonable”. Also, López-Sandoval results from the same cruise are exactly the opposite to Lagaria and colleagues’ Fig. 3, i.e. PER was constant (mean 37% rather than 30%, see above) along the west-east productivity gradient. The authors should discuss this discrepancy rather than only using supporting references. Since both papers are to appear in the same Biogeosciences special volume, the authors must carefully consider the paper by López-Sandoval et al. and discuss the serious discrepancies accordingly.

Reply: We have eliminated this sentence from the discussion section and have, in turn, developed an extensive discussion of the similarities and discrepancies between the findings of the two papers. It now reads (section 4.1):

“During the BOUM cruise, PPp and PPd were also determined *in situ*, along vertical profiles. *In situ* PER, in the form of euphotic layer-integrated data, averaged 37% and no significant differences were observed among sites A, B and C (López-Sandoval et al., 2011). However, at site A, mean areal PER (30%) was slightly lower than those in sites B and C (35 % and 37%, respectively), a

trend similar to that observed in the initial conditions in our experiments. It should be noted that these areal *in situ* PER values cannot be directly compared to our initial PER values, since our estimates were from a single depth (8 m). A more reasonable comparison is our initial values compared to *in situ* PER values measured at 12.5 m in the core of the eddies at the same day (Figure 3, in López-Sandoval et al., 2011). The same pattern was evident, in the form of site A with minimal values, although our estimates were systematically lower. This can possibly be attributed to different methodologies applied (24-h *in situ* vs 4-h on-board incubations in our study) since longer incubations have been associated with elevated PER (Baines and Pace, 1991). In our enrichment experiments, the incubations were identical in duration and period of day, thus estimates of PER among sites or treatments should be comparable.

Additionally, López-Sandoval et al. (2011) have suggested that when variability of PPd is examined within the same ecosystem, PER tends to remain constant over space and time (Marañón et al., 2004; López-Sandoval et al., 2010) but when contrasting environments are considered, the relative importance of PPd increases under oligotrophic conditions, most probably due to nutrient limitation. Indeed, our experiments showed that under conditions of excess N and P (+NP), chlorophyll *a*, primary production and assimilation efficiencies increased whereas PER tended to decrease. Perhaps due to the extremely low mineral nutrient concentrations present in the surface waters sampled, additions of N-alone or P-alone did not result in large variations of PER. Thus, relieving only one over two co-limiting nutrients did not induce important PER variations.”

6. RV: The afore-mentioned concerns need to be carefully addressed before considering the possibility of resubmission and final publication in the BOUM special volume. In conclusion the paper is not acceptable in its present version.

Reply: We have closely followed the reviewer’s recommendations and have proceeded with appropriate revisions throughout the paper. Below, we reply to the specific comments one by one.

Specific

7. RV: Please include “inorganic” before nitrogen and phosphorus in the abstract.

Reply: We have added this term in abstract and elsewhere in the discussion.

8. RV: The metabolic rates of the osmotrophic community as defined by the authors (phytoplankton plus heterotrophic prokaryotes) were not directly measured. They estimated total respiration, thus including the contribution of other heterotrophs (heterotrophic nanoflagellates, ciliates, larger metazooplankton?).

Reply: The respective sentence in the abstract is modified as follows: “We examined the effects of

nutrient additions on rates of ^{14}C -based particulate and dissolved primary production as well as O_2 -based metabolic rates...”

9. RV: Some indications on the depth of the experiments or whether their analysis of the relationships between PPd and Pp was performed with volumetric or areal units is needed in the abstract.

Reply: The sampling depth is now added in the abstract, it now reads:

“We examined the effects of nutrient additions on rates of ^{14}C -based particulate and dissolved primary production as well as O_2 -based metabolic rates in surface waters (8 m) of three anticyclonic eddies, located in the Western, Central and Eastern Mediterranean.”

We do not think that addition of the term “volumetric” in the abstract is necessary. We believe that with the sampling depth mentioned, it should be clear to the reader that we refer to experiments from a single depth and thus areal values are not applicable.

10. RV: Regarding the latter issue I suggest the authors read carefully the papers by Marañón and co-workers (2004 L&O, 2005 MEPS), and use them in the discussion of their own results. Please see also my next comment of a companion BOUM paper (López-Sandoval et al. 2010 Biogeosciences Discussions).

Reply: We do not think that an extensive discussion about the use of areal or volumetric units is indispensable in this study, since it is clear that measurements were derived from a single depth and thus areal units are not applicable. We have modified the relative discussion part to make this statement more clear. The following has been added (section 4.1):

“In our study measurements were performed with water samples from a single depth and variations of Pp and PPd were principally induced by varying nutrient concentrations. The relation found between Pp and PPd complies with the observation that in excess of both N and P (+NP treatment), PER was minimal while additions of N or P alone resulted in higher PER values (Fig. 1c).”

11. RV: Why do the authors use gross community production (GCP) rather than the more common term gross primary production (GPP)? If there was no other oxygenic phototroph in their water samples rather than phytoplankton I believe the correct term is the latter. In any case, please discuss in Material and Methods your choice.

Reply: We have replaced the term GCP with GPP.

The terms gross community production (GCP) and gross primary production (GPP) are somehow equivalent, both commonly used in the literature and they both account for all inorganic

photosynthetic carbon fixation, whether the organic carbon formed becomes part of the organism or is excreted (or secreted) into the environment as dissolved organic carbon or CO₂. According to the original ecological definition (Riley, 1940), the appropriate term is gross primary production (GPP) and GPP has subsequently been used in the MS.

12. RV: It is not exactly true that planktonic microbes [what do they mean exactly, heterotrophic prokaryotes (bacteria) or microzooplankton?] make up consistently >50% of total respiration. Please check Robinson (2008) chapter in Kirchman's book *Microbial Ecology of the Ocean* (2nd edition) for values below 50%.

Reply: We have modified the respective sentence in the introduction as follows: “Heterotrophic prokaryotes (*Eubacteria* and *Archaea*) are responsible for a significant portion of total respiration in the water column (Robinson, 2008). In the least productive areas, their contribution may even exceed 50% of total respiration (Lemée et al., 2002; Gonzalez et al., 2003; Reinthaler et al., 2006).”

13.RV: The fact that GPP is derived from NCP and CR estimates seriously compromises any consideration about the relative importance of GPP (GCP) or CR in driving NCP values. Unfortunately, this is quite common in ecosystem metabolism (i.e. O₂ fluxes) studies, but the authors should consider it explicitly.

Reply: GPP is derived from NCP and DCR. NCP is the primary measurement. Then assuming that respiration rates in the light and dark bottles do not differ, we can derive GPP rate values. We stated this assumption more clearly in the methods section and have replaced the term CR with the term DCR (dark community respiration) to highlight the embedded hypothesis in our methodology. It now reads (section 2.2.4):

“GPP was calculated as the difference between NCP and the negative DCR, assuming that respiration in the light bottles equals respiration in the dark.”

14. RV: A more exhaustive literature review on the factors that may affect primary production partitioning into PPp and PPd would be appreciated.

Reply: We have further developed the parts concerning the factors that may affect PPp and PPd in the introduction and discussion sections.

1) Added in the introduction: “PER reportedly increases when the phytoplankton are dominated by small-sized cells, most probably because their elevated surface/volume ratio promotes passive diffusion of small metabolites through the cell membrane (Bjørnsen, 1988).”

2) In the discussion –section 4.1 see above the long paragraph related to comment 2

3) and in discussion section 4.1: “PER may also be affected significantly by phytoplankton

community size-structure and species composition (Teira et al., 2001; Wetz and Wheeler, 2007). Unfortunately, taxonomic or size structure analysis was not involved in our experiment but in similar experiments with nutrient additions in nutrient-depleted surface oligotrophic near-shore and offshore waters, increases in autotrophic biomass and production are often associated with community shifts to larger cells and diatoms (Kress et al., 2005; McAndrew et al., 2007). A shift in the composition of the phytoplankton community during our study could be partly responsible for changes in PER. In theory, PER may be expected to be higher when the community is dominated by small-sized organisms compared to larger cells (Bjørnsen, 1988; Teira et al., 2001). However, this is not always observed since there is at least one study where no relationship could be established between PER and phytoplankton taxonomic composition or size structure (López-Sandoval et al., 2010). ”

and

“A potential problem with regard to PER is that measurements are based on the assumption that heterotrophic uptake of dissolved organic carbon produced by phytoplankton is minimized in short time incubations. Heterotrophic prokaryotes can incorporate the phytoplankton-produced labeled dissolved organic carbon and thus transfer it to the particulate pool. This activity would reduce measured PER not only due to underestimated PPd but also due to overestimated Pp, in the form of labelled heterotrophic prokaryotes in the particulate organic matter retained on the 0.2 µm filters. Conversely, labeled DOC may be produced from the particulate pool via trophic-related processes - such as sloppy feeding by grazers. The 4 h incubations used here are supposed to fulfil the assumption that heterotrophic transformations or transfers are minimized. In longer incubations of 5-6 h or more, heterotrophic prokaryotes were found to assimilate ~45% of the excreted carbon (Fernández et al., 1994; Morán and Estrada, 2002). Thus, our estimates of PPd should be considered as net fluxes and PER as a minimum value.”

15. RV: The authors apparently follow the paper by Morán et al. (2002) dealing with phytoplankton-bacterioplankton coupling when considering the role of PPd and Pp in meeting bacterial carbon demand (BCD, please use lower case for the full words). However, their suggestion of comparing total rather than dissolved primary production with BCD differs from what the aforementioned authors use. This should be detailed in the introduction and/or discussion and justified. Do the authors imply that bacteria (and only bacteria) are able to use all primary production concurrently produced in their experiments?

Reply: We have considered the respective reasoning in the aforementioned paper. The authors suggest that comparing the BCD with PPd is a direct way to demonstrate the coupling between phytoplankton and heterotrophic bacteria. We used the rather indirect comparison of total PP with

BCD, as also referred by Morán et al. (2002), and defined how we interpret it in the introduction section. It now reads (introduction):

“Comparing total primary production with BCD does not necessarily imply that all primary production is channeled through the microbial food web, but rather indicates the internal potential of a system to provide carbon sources to heterotrophic prokaryotes, in time and space.”

We also added a brief paragraph in the discussion section explaining the observed coupling in our experiment (section 4.2): “This coupling might be interpreted, in this case, as the common response, i.e., the synchrony of temporal variations of phytoplankton and heterotrophic prokaryotes production in response to forcing factors (e.g nutrient inputs, Fouilland and Mostajir, 2010).”

16. RV: The authors should revise their text for unnecessary verbosity at some parts (e.g. “it is now generally recognized”, “needs further to be investigated”, etc.) as well as the repetition of results in the discussion section.

Reply: We have eliminated and/or rephrased these and other sentences of this type.

17. RV: Why are there 24 data points in the figure? Assuming that control time final at station A was lost I would have expected 26 measurements (11+12+3 initial conditions).

Reply: There are 24 points because there are another two points missing (Cnlt of sites A and B at day 2). The radioactive sub-samples of these two microcosms were not exploitable. Thus, we eliminated these samples from the analysis.

18. RV: Table 2 should include some indication of significant differences between sites.

Reply: Table 2 has been modified and now includes significant differences between sites. The respective results section has been modified accordingly.

19. RV: Tables 3 and 5. Please state that these are mean ratios and provide significant differences if any.

Reply: Table 3 has been modified to include also significant differences between treatments. For this, we have replaced PP_{total} with PPp in calculating the assimilation ratio. Given that PER was generally low in the experiments, the ratios PP_{total}/chl_a and PPp/chl_a do not differ substantially, while microcosm replicates of both PPp and chl_a permit us to apply an accurate statistical test, as recommended. The legend of Table 5 has also been corrected. Given that Table 5 provides ranges of ratios of estimated parameters (BCD), an accurate statistical test is not applicable in this case.

Interactive comment on “The effects of nutrient additions on particulate and dissolved primary production in surface waters of three Mediterranean eddies” by A. Lagaria et al.

Anonymous Referee #2

General Comments:

1. RV: The underlying question, changes in primary production and system metabolism upon nutrient amendment, is a very interesting question and I think it has potentially far-reaching implications (e.g., why some eddies in the open Sargasso Sea are net autotrophic vs. heterotrophic). That said I wonder about the value of the ^{14}C PPd and Pp data. There are the caveats to its interpretation that the authors list but they don't list diel periodicity, filtering artifacts and separating PER from sloppy feeding by grazers (see below) as very important issues when trying to match up Pptot to NCP. In fact, the only meaningful discussion of Pptot is how close it is to NCP. In my opinion the trophic status (oxygen measurements and BCD) is the most compelling part of this data set and the one that should be put forth; the ^{14}C data could easily be left out without any harm to the value of the manuscript. Then the authors could actually discuss why +P drives eddies B & C to net heterotrophy, while +N has no consistent impact on trophic status. +NP I think it is obvious why it stimulates NCP but still worth discussing.

Reply: Our primary interest was to test the effect of nutrient additions on PER. We discuss the difficulties and potential uncertainties related to PPd measurements, but we believe that these data are worth to be included here since such data are very limited, in particular in oligotrophic waters. Moreover, the part in the discussion, regarding the effects of nutrient additions on Pp and PPd, has been extensively rewritten according to the recommendations of the anonymous referee #1. Regarding, diel periodicity and filtering artefacts please see our replies below, in the specific comments section. The ^{14}C incorporation method does not distinguish between DOC physiologically produced and exudated by phytoplankton and phytoplankton DOC released due to sloppy feeding. We have stated this in the method section, it now reads:

“It should also be mentioned that the ^{14}C -incorporation method cannot differentiate the origin of labeled DOC. Therefore, the physiological DOC production by phytoplankton and the release of labeled DOC of trophic-related processes -such as sloppy feeding by grazers -are both included in PPd measurements. However, short-time incubations minimise the contribution of trophic-related processes to DOC production.”

According to the reviewer, we have further developed the discussion regarding the effects of nutrient additions on metabolic balance and the different responses to single additions of site A compared to sites B and C. It now reads (section 4.2):

“As in our experiments, this shows a decoupling of DCR and GPP, with GPP displaying faster

and larger response to **limiting** nutrient additions on a time scale shorter than a week, resulting therefore in positive NCP values and shifting the community balance from net heterotrophy, or balanced, to net autotrophy. It also shows that phytoplankton community was more stimulated by inorganic nutrient additions (+N, +NP) than heterotrophic prokaryotes (Duarte et al., 2000). Addition of P alone had no particular effect on community metabolic balance and responses were similar to those of the unamended controls. The similarity of +P and Control is mainly explained by lack of P-limitation of both phytoplankton and heterotrophic prokaryotes, at all three sites (Tanaka et al., 2011). Meanwhile, since nutrient availability seemed similarly low at all three sites (Table 1, Tanaka et al. 2011), the different character of metabolic balance, in the Control and the +P, at sites B and C (net heterotrophic) compared to site A (rather autotrophic) should indicate differences in food web functioning. This may be attributed to the varying water masses which are important factors determining variability in microbial activity (Martínez, 1997). During the BOUM cruise, physical data indicated that at site A the core of the eddy was formed with Surface Modified Atlantic water, while eddies at sites B and C exhibited deeper cores formed by Levantine Intermediate water (Moutin et al. 2011).”

And (in section 4.2)

“Variability in nutrient availability constitutes an important regulator of plankton metabolism in open ocean waters (Gonzalez et al., 2002; Viviani et al., 2011). Both the O₂-based and C-based rates showed that, when adding limiting nutrients (+NP), rapid shifts in the metabolic balance can occur in favor of net autotrophy, controlled by increases in gross primary production rather than decreases in respiration.”

2. RV: Consistent with this, I find the title to not be representative of the actual work presented. It should, again in my opinion, be something more like “The effects of nutrient additions on trophic status of surface waters within Mediterranean eddies.”

Reply: Following the reviewer’s recommendation we propose the revised title:

“The effects of nutrient additions on particulate and dissolved primary production and metabolic state in surface waters of three Mediterranean eddies”

3. RV: My recommendation is to drop the ¹⁴C, or at least reduce its presence, focus on presenting and discussing the metabolic balance data, and present additional environmental data so it can be evaluated how relevant these surface samples are to the ‘euphotic zone/mixed layer’ of the eddy.

Reply: Regarding usefulness of ¹⁴C-data, we disagree with the reviewer. We believe that the entire ¹⁴C data set is strengthening the paper and that the modified discussion now is more balanced with oxygen metabolic data and justified. The hydrological and biogeochemical properties of the water

column in the three eddies are described in details in the publications by Moutin et al. (2011, in prep.) and Christaki et al. (2011) appearing in the same special issue. The choice of the sampling depth is explained in the companion paper by Tanaka et al. (2011): The following is now added in the Methods section:

“The sampling depth was located at the lower part of the surface mixed layer (13.5 m at site A, 8.5 m at site B, 11.5 m at site C: Moutin et al. 2011).”

Specific comments:

1) P8925, L3 - why only sample at the surface? It seems it could be due to space constraints but surface only data can only be interpreted so far. I think the authors, given that they can't go back and repeat the experiment, need to at least include some CTD profiles to see what the mixed layer depth is, Chla profiles (was 8m the Chla max?), nutrient profiles. This will really help in the interpretation of the metabolic balance data.

Reply: Please see above the previous reply.

2) P8925, L7 – what is the justification for adding those nutrient concentrations and in those ratios, and why is C different from A & B?

Reply: The following is now added in the Methods section:

“Nutrient additions were chosen with the aim to satisfy N or P requirements of heterotrophic prokaryotes and phytoplankton for the duration of the experiment (i.e., 3-4 days), and was based on an approximation of N:P ratio of 16 and 32 of the Western and the Eastern Basins, respectively (Tanaka et al., 2011).”

3) P8926, L11 – what about diel periodicity of photosynthesis. I recognize the authors want to minimize the reincorporation of release DO¹⁴C, but most studies with short incubations that are interested in ‘daily production’ do several incubations over the day? In figure 4 they relate hourly ¹⁴C production to daily O₂ production (converted to C units). Just estimating the slope suggests that there is a factor of 3. Was the incubation duration (4h) 1/3 of the daylength? This all relates back to the value of the ¹⁴C incubation data.

Reply: There are numerous studies where PP measurements are determined through short time incubations, usually around midday and are provided as maximum hourly rates. In order to convert them into daily rates, some studies use simple transformations according to the daily photoperiod length or more developed models (Moutin et al., 1999, *Sciences de la vie*, 322, 651-659). In our case, no model was applicable because of the uncertainties related to the diel periodicity of PPd fraction. Consequently, we had no choice but to work with the hourly rates. The good agreement of

PP_{total} with GPP and of the estimated carbon ratios (Table 5), confirm that ¹⁴C-data are also plausible.

4) P8926, L27 – the authors were obviously concerned about high vacuum pressures giving artificially high PPd values so filtered at <50mm Hg, but then filtered the rest of the sample at 200mmHg to measure Ppp and by using the PER value for the low pressure get a PPd value. This tells me that the Ppp values are all too low, and by calculation the PPd as well. Is it surprising then that the slope of Ppp(5ml) vs. Ppp(160ml) is 1.5? Again, calls to question the value of the Ppp data.

Reply: The difference in the pressure used is relative to the volume of sub-samples. Routinely, samples of ~250 ml are filtered at < 200 mmHg and provide accurate (particulate) primary production measurements. However, this pressure is most probably too high for filtration of only 5 ml sample. For this reason we used a much lower pressure for filtering the 5-ml aliquots. The 1.5-fold difference between the Ppp(5ml) and Ppp(160ml) was rather related to some small bias of the volume-correction factor (first term in eq. 1), rather than filtration artefacts.

5) P8927, L11 – why did the authors use 24,000 for the concentration of DIC? Is this the value calculated from salinity? Is it a measured value? More information is needed.

Reply: We have used the same value used in Moutin and Raibault (2002, Journal of Marine Systems 33– 34, 273– 288) during the MINOS trans-mediterranean cruise, which was determined according to Copin-Montegut (1993, Global Biogeochemical Cycles, 4, 915-925). This value corresponds to 2000 $\mu\text{mol kg}^{-1}$ and approximates the DIC values measured during the BOUM cruise in the surface layer (2220-2280 $\mu\text{mol kg}^{-1}$, Pujo-pay et al., 2011). The reference is now added in the text.

6) P8931, L8 – I don't see that $\text{NCP} = \text{CR}$ except at station C and that's only because of the huge error. Please clarify.

Reply: corrected, it now reads “gross primary production balanced dark community respiration”.

7) P8934, L27 – what large shifts in the properties of the phytoplankton community? Please clarify.

Reply: This sentence has been eliminated in the revised version.

8) P8936, L19 – perhaps I'm missing something but isn't the definition of GCP being sufficient to meet $\text{BCD NCP} > 0$? From here to L24 are just the definitions of metabolic balance. They aren't a discussion. Rather the authors should discuss why +P drives the system to heterotrophy and +N does nothing. I think the paper by Thingstad et al. 2005 might have some relevance here.

Reply: Net community production is defined as the balance between gross primary production and total respiration (in our case dark community respiration, DCR). What is shown here is that the estimated carbon budgets between GPP-BCD followed closely the metabolic state as described by the oxygen fluxes. It now reads (4.2 section):

“The estimated ratios of bacterial carbon demand to autotrophic carbon fixation (Table 5) generally followed the same patterns of metabolic shifts as described by NCP variations.”

9) P8937, L9-10 – Stating that nutrient additions couldn’t relax the competition between heterotrophs and autotrophs is a result, but what would be really interesting is why the different response as from Table 1, it looks like all 3 eddies were equally deplete of nutrients.

Reply: We eliminated this phrase from the conclusion. Please see also our reply to general comment 1.

10) P8937, L13 – again what differences in phytoplankton communities.

Reply: This sentence has been eliminated in the revised version.

Technical Corrections:

1) P8925, L2 – the figure 1 referenced here (I’m guessing a map) is not provided with the manuscript. Please clarify/correct.

Reply: Corrected

2) Page 8924, L1 – missing “in” after P-limited.

Reply: Corrected

3) P8927, L14 – should be “where” not “were”

Reply: Corrected