**Interactive comment on** “N-limited or N and P co-limited indications in the surface waters of three Mediterranean basins” *by T. Tanaka et al.*

**Anonymous Referee #2**

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Ambient concentrations of both dissolved inorganic N and P are exceptionally low in the Mediterranean and the availability of either one or both of these nutrients is likely to limit plankton growth. While there are evidence that the East Mediterranean is probably P-limited (Krom et al 1991, Zohary and Robarts, 1998) and that the West Mediterranean is probably N-limited (Raimbault and Cost 1990, Thingstad and Rassulzadegan 1995), several studies showed that limitation shifts from N to P and vice versa depending on the period of the year (Fiala et al 1976, Dolan et al 1995) or the area considered (Woodward and Owens 1989). The authors have conducted 3 experiments over 4 days each to explore the possibility of concurrent P and N limitations on phytoplankton and bacterial production in 3 basins located in the Mediterranean Sea. Despite the fact that the observation of N and P co-limitation in the Mediterranean is not original, this study is very interesting as it presents the response of a broad range of variable to N, P or
N+P additions in three basins from the Mediterranean. It would have been remarkable to replicate these experiments as spatial variability within the 3 basins would be interesting to study. Nevertheless, oceanographic cruises are always too short to conduct such extensive experiments.

The presentation and interpretation of the data seems straightforward, but I request that the authors consider a number of points before the manuscript is accepted for publication:

I suggest modifying the title “N-limited or N and P co-limited indications in the surface waters of three Mediterranean basins”. Here it says that you have only weak conclusions (indications) about the nutrient status of the Mediterranean. It is therefore not very attractive to your potential readers. Moreover, you should indicate what is limited: here, grammatically, the indications are limited or co-limited.

The English should be improved by a native speaker. Some sentences are very long and/or confusing and make the manuscript hard to follow here and there. Numerous abbreviations are not given in full letter at first use.

Abstract: L2-5: this sentence needs to be rewritten for English. Why using “respectively” at the end of this sentence?

L9 “…were set up for the carboys”. Since you introduce the experiment as being manipulated in microcosm, I would keep this term instead of carboys. Moreover, this sentence should be rewritten as well.

L13: I don’t think that “purely” is a well chosen term here. I suggest changing all over the manuscript.

L16: I think “the” is missing between “at” and “three study sites”

L16-17: See Figure 4: the stimulation of PP is much greater in +NP suggesting co limitation rather than N limitation. Please rectify. This is true for most of the parameters presented in this study and that should be mentioned.
L19-20: I don’t understand this sentence. What do you mean?

Introduction: P8185 L24: I don’t think that “ultra-oligotrophic” is necessary here.
P8185 L24 to P8185 L1: Can you provide references?
P8185 L1: I think “the” is missing between ”understand” and “biogeochemical”. Carbon should be written in all letters before introducing C.
P8185 L7: Particulate Organic Carbon (POC)

L12: I would remove the brackets for “relatively”

L12-13: “The deep waters have…” – How deep?

L16: “...the biogeochemical evidences...” - I suggest: “...these biogeochemical parameters suggest that...”

L19: “in the lower part of the plankton food web,” – un-necessary, I would remove.

P8147 L6: “P” - do you mean phosphate or phosphorus?

P8148 L12: objectives... were...

P8147 L15: One of the main objectives was to test “if the bypass and tunneling mechanisms for P exist”. Nevertheless, the authors did not answer this objective and just mention in the discussion “Hence it was impossible to test in this study if the bypass and tunneling mechanisms for P exist in different sites of the Mediterranean Sea.” I would thus consider removing this objective from the introduction and just discuss this point. Moreover, even if you introduced these concepts in the previous paragraph, you did not mention the terms of bypass and tunneling. If the reader has not read Thingstad et al (2005), they will not follow. Please define the terms of bypass and tunneling earlier.

Materials and methods

P8149 L1: Can you explain why did you choose this depth?
L3: delete “volume”
L3-4: delete “Four different treatments were set up to examine the limiting nutrient for the plankton community.” But add “four” between “The” and “treatments” L5.
L7: Please explain why did you add twice as much N at Stn C?
L10: Please explain why did you add NH4 only, while NO3 is undetectable at StnA and lower than NH4 at StnB in the initial conditions?
L12: “During the incubation, samples were taken...” – Please indicate the sampling frequency.
P8150L9: “For each sample, samples (500 ml) were...” – I suggest: “For each sample, 500 ml were...”
L19: remove “respectively”
P8151L9: Do you mean that you first pre-concentrated by gravity and then concentrated again in Utermohl chamber? This is unclear.
L16: What was the final TCA concentration?
L20: What were your background levels?
P8152L18-22: Why using all these conversion factors to get P-biomass from C while you measured POP? This is certainly a large source of error.
P8153L21: Do you mean the supernatant was aspirated and the pellet was washed 3 times (washed with what?).

Results
P8154L12: What is DIN?
L13-17: Please indicate that you are providing ranges; it is unclear until we read the last sentence.
L17: Please remove “That is”

P8155L1-7: Please provide numbers such as factors of decrease in NH4 and PO4 concentrations between Ti and Tf. PO4 at Stn A treatment +N: how can you measure significant decrease since PO4 was under the detection limit at the beginning of the experiment (<10 nM, Table 1).

L8-9: “No significant change of NO3+2 concentration was detected between the start and the end of the incubation in 8 out of 12 cases (t-test, P >0.05, data not shown).” Can you specify in which treatments it changed and discuss this interesting result in the discussion?

P8156L1-2: Please remove; this is part of the Method section.

L5-7: I do not understand this sentence

L21: Please replace “smaller” by “lower”

L16-17: “PP was significantly higher in +N than the Control and +P, and highest in +NP at all stations (Tukey HSD test, P < 0.05).” Even though PP was significantly higher in +N, it was largely higher in +NP (Fig 4) and this result should be emphasized since the authors provide misleading conclusions in the abstract (i.e. “Primary production was consistently limited by N”).

P8157L5: Please replace “cases” by “occasions”

Discussion

L1-15: I would remove this sentence as it is not really relevant. This study might be the only one to study the effect of nutrient additions on these parameters all together but it is far from being the first study to look at the limiting nutrients in the Mediterranean.

L15-17: "are the first that"

Guerzoni et al 1999 Progress in Oceanography p169-70 or Handbook of environmental
chemistry vol 5 - C. Migon p250: "Despite early observations of P limitation (Fiala et al 1976, Berland et al 1973, 1980), recent works suggest that the Mediterranean surface waters are N-limited (Andersen and Nival 1988, Owens et al 1980). In fact, there is growing evidence that the east Mediterranean is P-limited (Krom et al 1991) and that the West Mediterranean is probably N-limited (Raimbault and Cost 1990, Thingstad and Rassulzadegan 1995), or that limitation shifts from N to P and vice versa depending on the period of the year (Fiala et al 1976, Dolan et al 1995) or the area considered (Woodward and Owens 1989)."

Again, this study is not a first.

See also in your discussion "phytoplankton were N and P co-limited in surface waters in May 2002 in the same area (Thingstad et al., 2005; Zohary et al., 2005)"

Please remove "are the first that".

P8158L10: which depths?

L23-27: Again, why not using POP data?

P8159L29: What was different? Microcosm size (bottle effect?), final concentration of the amended nutrients? Other? Please describe. Please provide some hypothesis to explain these differences in plankton response to P additions.

P8160L28: there is no "on one hand" so there should not be any "on the other hand". Please modify.

L29: PP and Chla were not measured at the trophic level

P8161L8: “...during the winter overturn, and annual phytoplankton...” Here we understand that the N and P inputs happen during the overturn AND the bloom. I think that you meant that the input during mixing event results in a bloom which exhausts PO4 and NO3 to residual levels?

L12: Please remove “That is”
P8161L25-30: This sentence is very long and hard to understand. I would suggest to write 2 sentences and to improve the English.

P8162L2-6: same comment

L8-9: Since you are not the first one to raise this question, I would remove this sentence or rewrite it.

L10-11: “the skewed PON:POP ratio but the microbial food web being N-limitation or N and P co-limitation” – Please rewrite

L19-20: This sentence is irrelevant here. I don’t understand the link with the previous and following ideas.

L20-21: Please explain how.

Table 1: â€˜I suggest the following title: “Initial conditions at the three sampling sites.” â€˜Values are shown as mean ± SD (n = 3) except for water temperature, nutrient stoichiometry, and heterotrophic nanoflagellates.” Do you mean that n=1 for the exceptions? â€˜ATP turnover time at Stn B is very high compared to Stn A and C. Are you confident that it is representative of the initial conditions at Stn B? Indeed it influences the results on Figure 2 where ATP turnover decrease while they were pretty much constant at Stn A and C. â€˜How can you calculate DIN:PO4 when NO3 and PO4 are under the detection limit?

Figures: I suggest adding A, B, C... letters to the different panels in order to help the reader in the result section.

Figure 1: How do you explain the increase in PO4 in the control and +N treatment at Stn B and C? Please comment in the discussion

Figure 3: It is too small

Figure 4: the stimulation of PP is much greater in +NP suggesting co limitation rather than N limitation as the authors mention in the abstract.
Interactive comment on Biogeosciences Discuss., 7, 8143, 2010.