Response to E. Hrustic

We sincerely thank E. Hrustic for the time and effort devoted to the review of the manuscript. Below, we reproduce the E. Hrustic's comments and address their concerns point by point. The reviewer's comments are copied below in regular font with our responses in blue and the revised sections in the new manuscript version in red.

Dear authors,

I have read this manuscript with a great interest since the topic is higly important for the understanding of the complexity of the primary productivity controlling factors and the role of N 2 fixation in nutrient limited oceans.

We would like to greatly thank E. Hrustic for these constructive comments and advices which led us to significantly improve our manuscript. The three hereafter points have been answered in details below, and most of the technical details listed below were took into account in the revised version

General comments:
I find that the manuscript brings interesting combination of field research with modelling. Also, the exploration of nitracline and phosphacline depths variations in oligotrophic and ultra-oligotrophic regions of WTSP, including another analyses, enables a discussion of a high quality. Presented results are clear inspite of the complexity of the conducted research. I find the manuscript excellent, according to my skills to judge an overall scientific contribution. At the same time, I see some possibilites for improvements in Introduction, Methods and Discussion.

Topic – light
Since You mentioned benefitial role of N 2 fixers for the whole plankton community, which is in line with Agawin et al. (2007) and other studies which You cited, it would be interesting to explicitly include a sentence about the interplay between nutrients (dominantly DIN) and light as the common controlling factors in competition between N 2 fixers and non-fixers. It is obvious that Your research went further in more details concerning the link between different types of nutrient limitation and N 2 fixation, but for me as a reader, at least a small part about the light is missing.

I suppose that since the studied area spreads within a short range of geographic latitudes, You did not consider to include the topic of light in this particular manuscript, especially because of small differences in hours of daylight over the year in such an area close to the Equator? Therefore, You focused on the topic of nutrients without mentioning light explicitly, but only indirectly via citations?

It is true that we didn't focus on the role of light in the present manuscript since it has never limited autotrophs' growth, whether or not they are diazotrophs. Intracellular quotas indeed allow us to identify at each moment the most limiting element and, for autotrophs, carbon was never limiting. In most situations, carbon intracellular quotas of autotrophs were at their maximul values, but during the bloom, since cell division accelerated, UCYN and PHYS had sometimes carbon intracellular quotas that were not maximum but phosphorous intracellular quotas were far lower.

Topic – phosphoenzymes
I suggest that You slightly improve the part about the phoshphoenyzmes that are important in usage of DOP during P-limitation (see below my suggestion). In addition, there is a fine opportunity for the inclusion of findings by Dyhrman et al. (2006) reffering to the C-P lyaze enzyme in Trichodesmium, which can enable this organism to get the competitive advantage over other marine phytoplankton that do not use phosphonates that are considered to contribute even up to 25% in DOP (Dyhrman et al. 2006).

We thank E. Hrustic for this interesting suggestion to which we answer in details in the specific comment below (Page 11, Line 26).

Topic – stoichiometric ratios C:N:P for heterotrophs

I did not understand the reason for implementation of Redfield ratios for the heterotrophs since the literature supports molar C:N:P of e.g. 50:10:1. I see that later in the manuscript the rates of N and P mineralization are adjusted in the model, so You did some compensation, which seems to me rational to do. However, I think that many readers would like to see a sentence with explanation for the Redfield ratios for heterotrophs.

As suggested by recent studies, we decided to apply the same Redfield ratio , i.e. 106:16:1 to the C:N:P ratios of the upper and lower ranges of intracellular quotas for all the PFTs, including bacteria and HNF for which the C:N:P ratios of 50:10:1 were used so far. This change also brought a more consistent stoichiometry in ciliates (CIL) which were predating so far on organisms with very different stoichiometries. It is reminded however that PFT's stoichiometry is flexible ($QX \in [QX^{min};QX^{max}]$\$ where $X \in \{C ; N ; P\}$) in the model and that the Redfield ratios are only used to link together the limits of the ranges of C, N and P intracellular quotas, (i.e. $QC^{min} = 106\ QP^{min}$, $QC^{max} = 106\ QP^{max}$,…) thereby allowing a large variety of possible C:N:P ratios in PFTs, including the 50:10:1 ratio. This has been better explained in the revised manuscript (see below).

Technical details

My question marks are there only to provoke Your effort to slightly improve the presentation, rather than expecting the explicit answers during the open discussion, as far as I am concerned.

Page 1:

line 8 - only differing by the presence or absence of diazotrophs

The sentence has been modified as suggested

« [..] only differing by the presence **or absence** of diazotrophs »

line 13 – which seasonal changes? It seems that Abstract would be better if you write precisely here about the main results of your research regarding those seasonal changes.

The sentence has been modified as follow :

« [..] seasonal variations **in primary production and P availability in the upper surface waters** in simWMA [..] »

Line 22 – I feel that the logics of the sentence is inadequate. The word „although“ would suggest opposing statement in the second part of the sentence, but in fact the outcome is pretty logical. The area is oligotrophic and since NH 4 and NO 3 are the main inorganic N species taken up by osmotrophs, those nutrients remain low. So, I suggest this sentence: « Since nitrate (NO 3-) and ammonium (NH 4+) are the two main N sources taken up by autotrophs, their concentrations remain very low in the oligotrophic ocean being frequently growth-limiting factor in most of the open ocean.... »

We are a bit uncertain regarding the suggestion made by E. Hrustic. The word « Since » would suggest that because nitrate and ammonium are the main sources for autotrophs' N needs, their concentrations remain very low in oligotrophic ocean (meaning that this is the consumption of NO3 and NH4 that depletes N in oligotrophic areas). The reasons why oligotrophic areas have low nutrient concentrations are more because of an absence or a low external input of nutrient in the photic zone coming from the bottom of the water column by vertical mixing, by horizontal advection, atmospheric dust or river/land input.

We propose to replace « although » by « while » which could maybe enhance the comprehension of the sentence.
« ~~Although~~ While nitrate (NO$_3^{-}$) and ammonium (NH$_4^{+}$) are the two main N sources taken up by autotrophs [..] »

Page 2:
Line 2 - Some species of prokaryotic organisms....
We thank E. Hrustic for the suggesion that we took into account in the revised manuscript
« Some **species of** prokaryotic organisms (Bacteria, Cyanobacteria, Archaea), commonly called diazotrophs or 'N2-fixers', [..] »

Line 5 – Would it be good to ammend this part with a detail about the nature of dissolved N? I mean on this „diazotrophs release dissolved inorganic and organic N..."?
We agree with E. Hrustic and propose to add the following text to the revised manuscript :
« [..] diazotrophs release **a fraction of the fixed N in the dissolved pool under the form of NH4+ and dissolved organic nitrogen (DON)** in the surface waters [..]
Lines 7-8: I suggest „since it would reduce the N limitation for the phytoplankton and thus enhance primary production in the oligotrophic regions.
Explanation: This is not "characteristic" because some oligotrophic seas are P-limited, therefore "characteristic" seems not to be associated to N exclusively. "in the oligotrophic regions" sounds a bit better at the end of the sentence. Just a suggestion.
We agree with E. Hrustic and took into account this suggestion in the revised manuscript.
« [..] it would reduce the ~~characteristic~~ N limitation for the phytoplakton and thus enhance primary production **in the oligotrophic regions** »

Line 10 - bioavailability of dissolved iron (Fe) and phosphate (P).....
This modification has been made in the revised manuscript as follows :
« [..] availability of **dissolved** iron (**D**Fe) [..] »
Line 13 – It would be nice to have a reference a t the end of this sentence since You write about classical paradigma?
We added the reference of **Zehr and Kudela (2011).**

After this sentence it would be nice to extend the Introduction by Agawin et al. (2007) and Rabouille et al. (2006), which You already cited, but an explicit note about the role of light for N 2 fixation in Your manuscript is lacking. You might briefly add here that N 2 fixation is highly dependent on the circadian clock (Rabouille et al. 2006) and that the success of non-diazotrophs and diazotrophs depend on the interplay between intensity of light and DIN concentration and the competition for those resources.

We thank E. Hrustic for this suggestion that we took into consideration. We therefore added the following sentence :
« [..] thereby calling into question the classical paradigm of the N limitation in the open ocean (Zehr et al., 2011). **Moreover, chemostat experiments have highlightened that N2 fixation activity was highly dependent on the circadian clock and that the success of non-diazotrophs and diazotrophs depend on the interplay between light intensity and DIN concentration, and the competition for those resources (Rabouillet et al,. 2006 and Agawin et al., 2007).** »

Line 22 – DIP availability if the statement is strictly for inorganic P.
Yes it is inorganic P, this has been changed

Page 3:

Line 3 – Is it ecosystem or You explored ecosystems, one with diazotrophy and one without?
We agree that ecosystemS accurates more in the context and modified the sentence as follows :
« [..] to simulate the complex ecosystem**s** observed during the OUTPACE cruise [..] ».
Line 20 – their different biogeochemical characteristics (only a lack of letter „i“ in their)
thank you, this has been corrected

Page 4:
Lines 3,4 – I do not see in the manuscript where did You mention earlier anything about „ten years“?
Yes, indeed, we propose to rephrase the sentence as follow :
« Both simulations were run over ten year~~, as mentioned earlier.~~**, and** since a cyclic steady-state [..] »

Line 14 – „consumers“ could be changed to „grazers“? However, this is less important.
We replaced consumers by grazers as follows :
« [..] three ~~consumers~~ **grazers** (zooplankton) and one decomposer [..] »

Line 25 – after „energy regulator“ it seems appropriate to extend the sentence with „, being itself tightly linked to the daily light cylce (Rabouille et al. 2006). Just an example.
Yes thank you for the suggestion, this has been added to the revised manuscript as follows :
« [..] energy regulator, **being itself tightly linked to the daily light cycle (Rabouille et al. 2006)** »

Page 5:
Line 4 – Hereafter You have numerous dots after some units, these dots should be corrected.
Lines 5,6: Why did You change C:N:P from 50:10:1 for heterotrophs, which is supported by literature (Goldman and Dennett (2000), Fagerbakke et al. (1996), Chan et al. (2012), Alekseenko et al. (2014), for which You cite 50:10:1), to the Redfield ones?
As suggested by recent studies, we decided to apply the same Redfield ratio , i.e. 106:16:1 to the mean intracellular quotas of all the model PFTs, including bacteria and HNF for which the mean ratios 50:10:1 were used so far. This change also brought a more consistent stoichiometry in ciliates (CIL) which were predating so far on organisms with very different stoichiometries. . We propose to insert the following paragraph to justify this choice :

« While several studies have shown that the intracellular C:N:P ratios in heterotrophic bacteria tend to be below Redfield values as they were enriched in N and P (Bratbak, 1985, Goldman and Dennett, 2000, Vrede et al., 2002), more recent studies suggest that these ratios could be higher than 50:10:1 and highly variables in response to physical, chemical and physiological conditions (Cotner et al., 2010, Martiny et al., 2013 ; Zimmerman et al. 2014). This led us to replace the 50:10:1 ratios used so far in the model for bacteria and HNF by the Redfield 106:16:1 ratio as for the other PFTs represented in the model. It is reminded however that the PFT's stoichiometry is flexible (QX Î [QX^{min};QX^{max}]$ where X ∈ {C ; N ; P}) in the model and that these ratios are only used to link together the limits of the ranges of C, N and P intracellular quotas, (i.e. QC^{min} = 106 QP^{min}, QC^{max} = 106 QP^{max},…) thereby allowing a large variety of possible C:N:P ratios in PFTs, including the 50:10:1 ratio.»

Line 9 – Hereafter You write “phosphatase alkaline”. Is it nicer to write alkaline phosphatase, or it has to be phosphatase alkaline?
We thereby changed « phosphatase alkaline » by « alkaline phosphatase » as rightly suggested.

Line 34 – According to which reference You chose the ratios 1000:100:1 ? Can You extend the sentence with a comment about grazing pressure from right to the left in these abundances ratio?
Unfortunately, we are not able to provide a specific reference regarding this ratio. However, it is

reminded that these ratios only concern the initial conditions of zooplankton abundances which have no significant influence on their dynamics thereafter simulated by the model.

Page 7:
Line 21 – "particulate carbon biomass", "particulate organic carbon" and "C biomass" are present in the manuscript. Are some of these names associated to the same variable? Can You simplify this?
« C biomass » in line 28 has been replaced by « POC »
Line 32 – Why not "over ten years" if You are able to perform model for ten years?
As the variations are the same each year, we chose to represent only 3 years to show the seasonal variations. A figure showing the 10-years of simulation has no additional interest.

Page 8:
Line 28 –maybe extending the sentence with "compared to winter mixing".
Yes thank you for this suggestion which has been taken into account.
« [..] followed by a longer stratified period from February to July, with a shallower MLD between 25 and 30 m **compared to the 70 m reached during the winter mixing.** »

Page 9:
Line 26 – exclude one of the words, "new" or "fresh" because they carry the same information.
« fresh » has been removed.

Page 10:
Line 24 – Did you provide an abbreviation explanation for PP before this point in the manuscript?
Yes in section 2.1

Page 11:
Line 7 - However, their development also requires sufficient intensity of light and other nutrients...
Thank you for this suggestion which has been took into account in the revised manuscript.
« However, their development also requires **sufficient intensity of light and** other nutrients [..] »

Line 26 –Why "or"? Is it better to write "e.g." than "or". There are other phosphoenzymes potentially used to get P from DOP, especially by Trichodesmium. I suggest "(alkaline phosphatase, nucleotidase, polyphosphatase, phosphodiesterase)".
We added those other phosphoenzymes as suggested
« [..]phosphenzymes (**alkaline phosphatase, nucleotidase, polyphosphatase, phosphodiesterase**) »
Then You could extend the discussion by "Moreover, C-P lyase found in Trichodesmium (Dyhrman et al. 2006) is another enzyme that enables this organism to use previously considered non-bioavailable fraction of DOP, i.e. phosphonates that represent circa 25% of DOP (Dyhrman et al. 2006). Taking into account this possibility, Trichodesmium thrives in the oligotrophic oceans with a great success".

We really thank E. Hrustic for his suggestion regarding the C-P lyase found in Trichodesmium by Dyhrman et al., 2006. As mentioned in their paper, phosphonates are more difficult to degrade than monophosphoesters, which commonly led to characterize phosphonates as refractory DOP. We understand that the C-P lyase found in Trichodesmium or other Cyanobacteria challenged this point of view since C-P lyase have been found to be able to hydrolyze phosphonates. However, in our model, we only consider the labile pool of DOP (i.e. monosphophoesters), and implicitly took into account the phosphoenzymes activities by decreasing the half-saturation constant for the labile DOP assimilation for all autotrophs, as mentioned in Section 4.2.2. The Dyhrman et al. (2006) results would have been of great interest if we had considered in our model a pool of semi-labile DOP but this could

be one of our future model development since we have already considered this possibility for other reasons than giving an advantage to Trichodesmium...

Page 12:
Lines 17-18 – "around the main thermocline between 100 and 500 m depth". Can You provide a figure for the vertical profiles of temperature?
The following figure represents the vertical profile of temperature during the winter mixing showing that the main thermocline is located between 100 and 700 m depth. We therefore changed the sentence as follows :
« around **the first 400 m of** the main thermocline, between 100 and 500 m depth. »

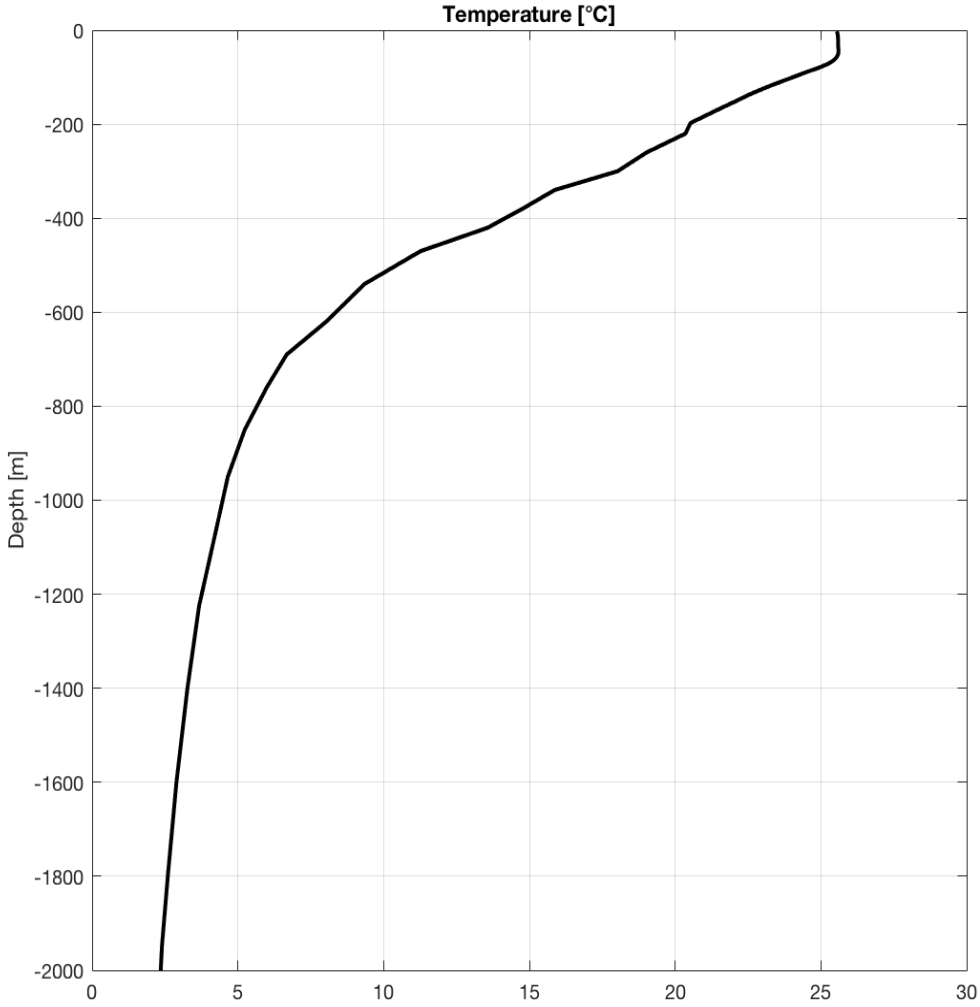

Figure 1 Modeled vertical profile of Temperature during the stratified period (March 2015)

Page 13:
Line 7 – project, Olsen et al. (2016)). Comment: add comma after "project".
Yes thank you, the coma has been added.

Page 14:
Line 6 – nutriclines depths

we replaced « depth of nutriclines » by « nutriclines depths » as suggested in the following sentence :
« [..] (ii) the higher/ lower ~~depth of the nutriclines~~ **nutriclines depths** characteristic of oligotrophic (WMA)/ultra-oligotrophic (WGY) states, [..] »

Lines 8-9 – Is excess DIP ideal for growth of diazotrophs if the light intensity is not suitable? Maybe "Excess DIP in N-limited surface ocean, if supported by sufficient photosynthetically active radiation, would favor the growth of diazotrophs in comparison to non-diazotrophs"?
We propose to add the following section at the end of the sentence :
« [..] intensive N2 fixation, in the WTSP region where light intensity is high enough and not limiting the diazotrophs' growth. »

Page 24:
Figure 4c – Why do You use "Trichos" if You defined the abbreviation in the Methods as TRI?
This is an error, Trichos should be TRI. This has been corrected.

References (*already cited)

*Agawin NSR, Rabouille S, Veldhuis MJW, Servatius L, Harriët M, Van Overzee MJ, Huisman J (2007) Competition and facilitation between unicellular nitrogen-fixing cyanobacteria and non-nitrogen fixing phytoplankton species. Limnology and Oceanography 52(5): 2233–2248.

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Dyhrman TS, Chappell PD, Haley ST, Moffett JW, Orchard ED, Waterbury JB, Webb EA (2006) Phosphonate utilization by the globally important marine diazotroph Trichodesmium. Nature 439(7072): 68–71.

Fagerbakke KM, Heldal M, Norland S (1996) Content of carbon, nitrogen, oxygen, sulphur and phosphorus in native aquatic and cultured bacteria. Aquatic Microbial Ecology (10): 15–27.

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