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Biogeosciences Discussions

Interactive comment on "The significance of nitrogen regeneration for new production within a filament of the Mauritanian upwelling system" *by* D. R. Clark et al.

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Response to general comments. We thank the reviewer for this detailed review. We aim to address all points raised by adding clarification, expanding the discussion and providing an alternative representation for Figure 1. The reviewer requests that we separate results and discussion sections; on this point our preference is to retain the format as presented. 'Biogeosciences' welcomes both formats (combined or separate results and discussion sections). We do not feel that the argument for separating these sections (to 'help expert readers identify patterns and compare rates') should in any way be hindered by the choice of format. We are open to guidance from the Editor on

this issue and will accommodate accordingly. We provide detailed responses below.

'The reference to previous studies and interpretation of the data in the light of the planktonic community responsible for the fluxes measured is insufficient. Despite the authors only have larger phytoplankton counts and no flow cytometry data, their fluxes could be better discuss in the light of previous works in the NW African upwelling.'

We are willing to expand the discussion and citations related to the NW African region and thank the reviewer for this information. We would however suggest that the factors and topics critical to the discussion of our results have been included. Although we did not include it, we have analytical flow cytometry (AFC) data. However, this information was not especially insightful, partly due to the limited duration of the study. Using all parameter sets available to us, we undertook statistical analysis of this data in an attempt to understand links between physical, biological and chemical characteristics of the water column. Unfortunately, limitations (e.g. the duration of the filament study) precluded any statistically significant interpretation.

'Given the existing literature on this subject published in the last 20 years, and the different approaches taken to measure new and regenerated production, it is perhaps the time to produce a review paper comparing the data obtained in different systems, homogenize results, and even provide the community with a consensus formulation for new and regenerated production (concepts which need to be revisited, as the authors discuss "it is likely that NO3 based exportable production from such systems has been over-estimated historically").'

This would be a valuable exercise, time permitting. We have gathered an extensive data set of N-cycle parameters across a range of oceanographic provinces; the process of synthesising this data into a review structure has started and we are very much interested in taking this further. We would welcome input from this reviewer should they wish to contribute.

ABSTRACT 'The abstract contains many details on the data, but none on their inter-

pretation.'

The abstract provides an overview of the study objectives, the location, the method and the data obtained. The last sentence provides an insight into the implications of the study. We believe that this adequately fore fills the function of an abstract and do not propose to change it.

INTRODUCTION 'The introduction could highlight better how considering regenerated production may change our view of upwelling ecosystems and global carbon and nitrogen cycling in marine systems.'

'Page 17785, line 5: please also mention all other nitrogen fluxes that may influence f-ratio calculations (DON release, N2 fixation. . .), including appropriate references.'

We will improve the consideration of regenerated nitrogen in the introduction and include N-cycle processes in the introduction of f-ratio calculations. We would not wish to cover this extensively in the introduction as it is covered further in the discussion, and elsewhere in the literature.

MATERIALS AND METHODS 'Page 17785, line 20: please provide details on the remote sense data (satellite used etc), how much time passed between detecting the filament and starting sampling it'

We have added this detail.

'Page 17785, line 25: "GC" is not described elsewhere in the text. More details on the SF6/3He detection method should be provided (i.e. are these compounds detected in a continuous way, like with seawater pump continuously through the GC? Or sampled discretely?'

These details have been added.

'Page 17786, line 1: when is that? Provide detection ranges, limits.'

This information has been added

'Page 17786, lines 5-6: it would be clearer to list which stations were sampled at which depth levels and which were not.'

Stations sampled at 1% sPAR are identified in Fig. 6; this only relates to NO2- oxidation rate data at 6 stations. Our view is that an additional table is not justified, but this information will be clarified in the text.

'Page 17786, line 12: do the authors know what's the temperature range within their flushing boxes along incubations, and/or how does it compare to in situ temperature?'

Incubation boxes were flushed with seawater using the ships surface seawater supply, consistent with standard practice. The ships seawater supply (to both labs and decks) was collected from a depth of approximately 5m. We did not measure the temperature of individual incubation boxes. However, very high flow rates were used for this flushing process and we would not anticipate large deviations between surface seawater temperature and that of incubation boxes.

'Page 17786, line 25: the authors mention chemicals and where they were acquired from before actually saying what they used them for. I would recommend describing how 15N additions were made and then stating in parentheses where the chemicals were obtained from.'

This is essentially a point about writing style. Rather than be extremely repetitive (as many chemicals were sourced from 'Sigma-Aldrich' for example) we chose to gather this information together for collective presentation. We do not propose to change this.

'Page 17786, line 27: why was the sample blacked-out? Was it only maintained in the dark until being dispensed in different incubation bottles and amended with 15N? Why regeneration fluxes were measured in the dark and assimilation ones in the light? Do the authors have any evidence that nitrogen regeneration is not performed by photoheterotrophic bacteria for example?'

The collection of seawater and 15-N amendments were conducted with blacked out

containers to avoid light shock (some samples were collected from low light environments). However, the text is quite clear that all incubations were performed in deck incubators using simulated light and temperature according to JGOFS protocols. At no point is the use of 'dark' incubations introduced.

'Page 17787, line 9: so one bottle or set of bottles was used to measure NH4 regeneration in the light and another one in the dark? It is unclear. Bottles were incubated in monoplicates but then split in triplicate subsamples for analysis? Are the error estimates given in Figure 5 analytical standard deviation? It is not clear from the methods.'

The text in line 9 (P17787) states; 'Amended seawater was used to fill a 2.2 L incubation bottle, which was placed in a deck incubator at simulated light and temperature for approximately 8 h.' This seems quite clear – a single 2.2L bottle is filled with 15-N amended seawater and placed in an incubator under simulated light and temperature. We are open to suggestions as to how this can be made any clearer. The remaining amended seawater (i.e. 4L minus the 2.2L used for incubation) is filtered and triplicate 100mL volumes used for pre-incubation analysis. Similarly, the text states that post-incubation, the 2.2L bottle contents are filtered and split into triplicate samples for post-incubation analysis. Again, we are open to suggestions as to how improve clarity here. We can improve clarity in the figures by stating what error bars represent; one standard deviation of triplicate rate estimations for Fig 5, or one standard deviation for triplicate concentration measurements for Fig 2 for example). We will add this clarification. However, to re-iterate, at no point do we discuss the use of 'dark' incubations and it is unclear how the reviewer has reached this interpretation.

'Page 17787, line 18 and elsewhere: the range of %15N enrichment caused by 15Nlabeled substrate addition should be mentioned somewhere in the methods.'

Indeed. This was an oversight and will be stated in the methods section.

'Pages 17788-17789: consider reducing methods and referring to previous publications where possible (see general comments above).'

We find that this is a difficult balance – some readers (and reviewers) prefer to have full method details while others prefer very concise descriptions. We have made reference to our previous work for method details which are not provided here.

'Page 17790, lines 4 and following: 660 mL for 15NO3 and 15NH4 incubations seems a really small volume. What was the range of PON concentrations measured? What is linearity limit of the IRMS used? Can 15N values be given with enough accuracy which such low PON concentrations (if they were low)?'

The volume and duration of incubations for N assimilation measurements was absolutely appropriate for high productivity systems. The range of PON concentrations was presented in Fig 5 (panel b) and reported as 0.6-3.0μM-N. A calibration for our IRMS is presented in the attachment (IRMS graph), in addition to variance in atom% 15-N for this standard range. The correlation provides an r2 of 0.999 with a variance in atom%15-N of these standards of 0.16%. This is normal for such systems. Sample sizes <0.5μM-N are not used. Enrichments achieved during this study were as high as 1.0 atom%15N, and generally >0.5atom%15N. Consequently, analytical performance was not an issue during this study.

'Page 17790, line 9: assimilation incubations were performed for 6 h, while regeneration ones were 8 h. The use of short incubations for the measurement of regeneration fluxes is common, but the assimilation incubation times seem short to me. The authors should state why these timings were chosen, as well as discuss how this might have affected their rates and compare to other studies using different incubation times.'

We disagree. Quite the reverse in fact. Assimilation studies are often kept short; historically the rationale for this was that it minimised both the influence of isotope dilution (in the dissolved fraction) due to N-regeneration and the risk of substrate depletion. In high productivity systems, incubations as short as 3 hours have been reported. This constrains atom%15N enrichment of PON thus avoiding analytical complications with excessive enrichment. Conversely, analytical (and method) limitations have often re-

quired long incubations for N regeneration studies, as these rates are typically (though not always) lower than assimilation rates. The incubation durations used in our study were appropriate to the study location and are consistent with previous studies. We do not believe that justification is necessary; comparisons with other studies on this point would detract from the manuscripts focus and would be of little value.

'Page 17790, lines 10 and following: it is not clear how many replicates were done.'

The text states on line 6 (p17790); 'Triplicate 660mL volumes of seawater were separately amended. . .. . .' i.e. there were 3 replicates.

'Page 17791, line 9: this notation (T-1-T7) has not been explained before in the methods section.'

Correct – this notation was 'introduced' later on page 17793. We will address this sequence.

'Page 17791, lines 15-20: a table including the variability of RNO3 values along the study would be helpful. So, in summary, the proportion of RNO3 with respect to total NO3 increases with distance to the recently upwelled water near the coast? Please add a conclusive sentence at the end of this paragraph.'

The reviewers text is confusing here. 'RNO3 increases with distance to the recently upwelled water near the coast'. From where then? The term 'RNO3' represents the fraction of the total NO3- pool represented by 'new' NO3-. Upwelled water must, by convention at least, represent new NO3- alone. This fraction diminished as photic zone nitrification diluted the 'new' NO3- pool with 'regenerated' NO3-. That is the concept we are introducing here and representing by Fig. 7. RNO3 values are presented in Fig 7 and an additional table is not justified.

'Page 17792, line 10: It is a pity that flow cytometry data are not available.'

It is available. However the data adds little to the interpretation of results presented and so was not included in the manuscript.

RESULTS AND DISCUSSION 'At the time of their sampling the authors found that the filament contained mainly NACW, which has a lower nutrient content than SACW. Can the authors discuss, in the light of water mass depiction in this upwelling system (previous publications) how different water mass proportions and seasonality may affect microbial nitrogen fluxes on a yearly basis?'

Unfortunately, the simple answer is no. We attempted to draw out some statistically meaningful interpretations between nitrogen fluxes and water column characteristics but were unable to do so due to data limitations, both spatially and temporally. While other studies can add to the data set, this type of analysis is beyond the scope of the study.

'Despite the authors do not have prokaryote abundance data, it is worth discussing their role in regeneration processes referring to previous works. Also, the use of GF/F filters and the related potential underestimation of nitrogen fluxes should be discussed (Morán et al., 1999).'

We do have AFC data but it adds little value. We will expand the discussion of the role of prokaryotes in N-regeneration processes. The paper referred to by the reviewer relates to the estimation of primary production using 14C and its overestimation using GF/F filters due to DO14C adsorption. This is a completely separate issue to the use of GF/F for PON retention and 15N analysis. The only potential issue for N flux (i.e. N assimilation) rate estimations would be the underestimation of PON due to non-quantitative retention of all cells capable of utilising enriched inorganic nitrogen (i.e. the bacteria). This is unlikely to be a significant issue given that productivity in this system was dominated by large cells (> 2μm, Fig 4).

'Page 17793, lines 26-27: the description of T-1 etc should be included in the methods section.'

This issue has been addressed.

'Page 17793, lines 18-20: it is unclear how the mapping was done. Regular (multiple) CTD casts? Moving vessel profilers, SeaSoar? It should be better explained in the methods.'

Extensive details of the mapping exercise are provided in a highly technical form by Meunier et al. (2012), and referred to in the manuscript. While some additional details can be provided here, this is a complex area and interested readers should refer to the associated paper from this research program.

'Page 17793, lines 25-27: so the Lagrangian study was successful from days 0 to 7, but not beyond (lines 5-7), or is it not fully reliable from days 0 to 7?'

This is a confusing interpretation of the manuscripts text. We will add text to clarify the constraints of the study duration; these details have already been requested by the reviewer in a previous comment.

'Page 17794, line 17: Even if N2 fixation was not targeted in this study (and is likely minimal in this upwelling system) it should be mentioned and discussed. The low N:P ratios found could promote diazotrophic activity. It is worth discussing works like Raimbault and Garcia (2008), Sohm et al. (2011) and others. The subject is briefly mentioned in Page 17795, line 6, but could be further developed.'

We have attempted to focus the text on topics directly relevant (and informed) by the data. Deviation in N:P is directly relevant for subsequent N fixation as the water mass leaves the study area and advects offshore. We discuss this. Noting that we are not offering a review, without further data we question the value of discussing N fixation in any depth as we are unable to offer anything other than speculation.

'Page 17794, line 27: you mean denitrification?'

No. We mean nitrification as stated. Denitrification is a strictly anaerobic process, most often associated with sediments. We are referring to pelagic nitrification, which is well documented in oxygen minimum zones (OMZ). A by-product of the nitrification process

is N2O. Consequently, pelagic OMZ's are a source of N2O.

'Page 17795, line 2: the authors mention here P* without having explained what it is, how it is calculated etc. '

We have included this detail.

'Page 17795, line 2: It is probably out of the scope of this paper, but can the authors use offshore transversal velocities to estimate how much phosphate (and other nutrients) are exported offshore (export rates) by the filament and compare with other upwelling systems? It would be interesting to compare with data from other filaments like those in California, off the Iberian Peninsula and Cape Ghir, for example. Presumably physical data can be easily obtained from Meunier et al. 2012 (?). This could be included at the end of the discussion (Page 17802).'

This is an interesting point and we had considered how to arrive at an estimation of P-export (specifically) as this would be most relevant to the basin scale P* debate. However, the complexity of this issue justifies a separate manuscript and is beyond the scope of the present contribution.

'Page 17795, line 8: very vague, please be more precise.'

Is the reviewer referring to the statement '. . ..biological processes. . .' in relation to the drawdown of inorganic nutrients? We could add that the processes relate to primary production, but this really is a statement of the obvious. Perhaps the reviewer is referring to the second part of this sentence; '. . .horizontal and vertical mixing' but again, in the context of the preceding sentence the meaning should be clear? We do not propose to modify this sentence as we do not agree that its meaning is vague.

'Page 17795, line 12: add references to figures where needed.'

Additional figure references will be added as needed.

'Page 17795, lines 13-21: and why is this? Different nutrient regime? Try to compare

with other upwelling systems (reasons behind different primary production rates).'

There are far too many possible reasons behind this to make meaningful comparisons within the constraints of available space. Such information has been reviewed and links to nutrient regimes and seasonality have been made. For individual studies, productivity could also depend upon the 'age' of upwelled water (i.e. since it reached the photic zone and supported photosynthetic processes) which cannot necessarily be known. We do not propose to develop this point as it would be highly speculative.

'Page 17795, lines 22-23: state ranges, refer to figure.'

This information has been included.

'Page 17796, lines 9-10: state ranges of phytoplankton abundance (here and in the following lines).'

This information has been included.

'Page 17796, line 11: at least state % of carbon provided by diatoms. '

This information has been included.

'Page 17796, line 15: the high proportion of flagellates is important and explains the predominance of regeneration fluxes. Discuss further, cite other works where the protagonism of flagellates in upwelling systems has been highlighted (e.g. Anabalón et al., 2014; Böttjer and Morales, 2007).'

There is a coincidence between peak NH4+ regeneration rates and peak flagellate abundance but one does not necessarily explain the other. We discuss the routes of NH4+ regeneration (17798 line 14-20) and support this with numerous citations. Within the constraints of the data we believe this is adequate (we will be including more information about regeneration processes in the introduction).

'Page 17796, line 17: POC would be better if available.'

It isn't.

'Page 17796, lines 20-24: See Benavides et al. (2013).'

We will include appropriate information from this publication.

'Page 17798, line 17: or active release.'

Indeed. Will will provide any appropriate support to this statement.

'Page 17798, line 20: and probably negligible in your samples due to the small volume used.'

We suspect that the reviewer is referring to lines 18-19 (not line 20) which describes as minor the contribution from zooplankton activity to NH4+ regeneration. The sample volume will have been likely to exclude zooplankton and so their contribution to NH4+ regeneration would have been negligible. However, we believe that the text already makes this point adequately.

'Page 17800, lines 4-24: before speculating on particle-attached nitrifying organisms, I suggest discussing the much more oxygenated character of this upwelling system in comparison with Peru for example and how this affects nitrification/denitrification processes.'

Firstly, there is evidence for particle attached nitrifying organisms presented in the citation (Ward 2008). Secondly, denitrification is not taking place in the incubation bottles used here and cannot be considered as a mechanism to explain these observations. Yes, denitrification takes place in systems like the Peruvian upwelling, but how does this help to explain the observations made here, in aerobic water samples taken from the photic zone? The discussion suggested by the reviewer would be entirely speculative. By contrast, the proposed mechanism aligns with the observations (e.g. DIN budgets) and provides a rationale for related observations (such as the challenge of linking pelagic nitrification rates with environmental drivers). Finally, we have direct (but yet unpublished) observational data from NERC's (UK) Shelf Seas Biogeochem-

istry (http://www.uk-ssb.org/) program that supports the association between very high rates of N regeneration and marine particles; N regeneration rates vary in relation to particle composition, depth and season.

'Page 17800, lines 25 and following: here the authors could refer to the RNO3 table proposed above.' The RNO3 data is already presented in a figure and does not need to be replicated in a table.

'Page 17802: Have the authors tried converting new production to carbon using Redfield (or C:N ratios from their own samples) and comparing those rates to 14C-based primary production data?'

The rationale for this exercise is not clear.

'Page 17802, conclusions: Can the authors estimate how much new production rates in upwelling systems are overestimated by not discerning between 'new' and 'regenerated' NO3?'

We do this! P17802 line 5-10. We present annual new production estimated by the classical f-ratio and then present how much this decreases by with each 'correction' strategy.

'Figure 1: This figure needs to be improved. The longitude is not aligned between panels. The path of the labeled water mass (SF6 distribution) should be superimposed on the map. Satellite images of Chl and/or temperature would be helpful for the reader to see the structure of the upwelling filament.'

We will develop a new figure using a combination of satellite data and regional maps.

'Figure 3: This figure is barely discussed in the text. The differences in N:P ratios between MLD and below MLD are interesting and merit discussion. Also, MLD is not written in full at first use in the text (Page 17798, line 4).'

We will define MLD and expand the discussion of this figure. The data demonstrates

the concept of P export (i.e. P*) by this filament. As suggested by the reviewer, this point could be expanded upon to arrive at a useable number for this export flux. We will also speculate (to a limited extent) upon the difference between values between depths.

TECHNICAL CORRECTIONS 'Lagrangian and Eulerian should be capitalized throughout the text. Page 17786, line 17: CTD is fully written here, but in fact was mentioned before in the text (line 3 of the same page).'

We will address these points.

'Page 17794, line 28: here the authors use 'N' instead of 'nitrogen', please be consistent throughout the text. Same in Page 17796, line 1 and elsewhere.'

We will correct this for consistency.

'Page 17795, line 24: 'northwest African', also written as 'North West', and 'NW' elsewhere in the text. Please be consistent.'

We will correct this for consistency.

'Page 17800, line 3: typo "μmnol" '

Well spotted. We will address this point.

Interactive comment on Biogeosciences Discuss., 12, 17781, 2015.

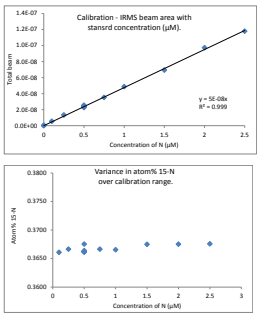

Fig. 1. IRMS graph