Interactive comment on “Hidden biosphere in an oxygen-deficient Atlantic open ocean eddy: future implications of ocean deoxygenation on primary production in the eastern tropical North Atlantic” by C. R. Löscher et al.

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Response to Referee #1 I would like to thank Referee 1 for considering this manuscript being adequate for publication in Biogeosciences. The comments and thoughts provided by Referee 1 were particularly helpful to improve this manuscript. Questions and comments were addressed as described in the following; modifications are highlighted in yellow in the main text:

General comments:

Referee comment (RC)1: I am fairly surprised to not see organisms of the SUP05 clade being present/abundant here as they are normally found in OMZs. Was the amplicon dataset screened for them?

Author comment (AC)1: SUP05 was indeed present in the dataset; we presented the diversity on the phylum level, SUP05 would only show up on the family level and clusters within the phylum of Proteobacteria, as shown in figure 4. In order to provide a deeper insight into the distribution of SUP05 and other organism groups, we now included a table and a bar plot including all taxa on the family level to the supplement and added the information of SUP05 distribution to the text.

RC2: If the figures are already the size they later should appear in the manuscript, then I would suggest increasing it for the sake of readability. They are really nice, so no need to make them so small.

AC2: Thanks for this nice remark- we will increase the figures in the final manuscript.

RC3: While the manuscript is generally well written, sometimes complicated sentence structure with multiple sub-sentences makes reading bothersome. Streamlining this a little could enhance the reading flow significantly.

AC3: The manuscript has now been checked by a native speaker in order to increase the reading flow.

Specific comments:

RC4: p. 14178, l. 10 ff: “…at depth around 40 to 100 m…” sounds weird. Is “…at depths of around 40…” meant?

AC4: Changed as suggested.

RC5: p. 14180, l. 6-13: Why state the aim and then the ultimate aim? Wouldn’t it make more sense to clearly define what the aim was? To identify differences in microbial community structure between eddy and surrounding waters was just a step towards
In this study, we investigated differences in microbial community structure in an O2-depleted eddy, surrounding ETNA open waters, and upwelled waters on the Mauritanian shelf. This was achieved using a combined high-throughput 16S rDNA amplicon sequencing/qPCR approach along with carbon uptake rate measurements and hydrochemical observations. This study aimed to understand the microbial community response to O2 depleted conditions with regard to primary production and remineralization in these poorly described anomalies to further understand the sensitivity of the ETNA biogeochemistry to future ocean deoxygenation.

It would be nice to cite the original papers here.

Sure, as a result of our sampling strategy both options are possible; however, it would not matter with regard to the difference in community structure between samples inside and outside the eddy. Ganesh et al. (2015 in ISME j) show in the supplement of their study that a certain proportion of euryarchaeota are present in the filter fraction of 0.2-1.6 \( \mu \text{m} \). The authors consider this fraction as free-living organisms. They, however, do not show more details on the phylogeny of this group so that it remains unclear whether these are methanogens or not.

To confirm if the upper chlorophyll maximum is caused by eukaryotic phototrophs, it might be an idea to look into the sequences classified as cyanobacterial 16S rRNA and see if 16S genes from plastids were detected and are hidden in there.

Actually, the idea of recovering information on eukaryotic phototrophs from the plastids is interesting (although a characterization of eukaryotes remains impossible from plastid sequences) - still the extraction protocol for nucleic acids may not be as efficient for eukaryotes as it is for microbes. This has to be considered. In addition, we tested the applied primer set in silico on the SILVA database and determined a relatively low coverage of only 38% for chloroplast sequences. Therefore, the information obtained from chloroplast sequence analysis can only be taken indicative for the presence and abundance of eukaryotic phototrophs; however, using it as a quantitative tool is only possible to a limited extent. Particularly, it can’t be quantitatively compared to the abundance of cyanobacteria because of the different coverage as described above.

Judging from the phylogenetic information on the metabolisms of the organisms is not always possible, so the statement is fairly strong here. While I think the authors are right I still would word it a little bit more careful.

The close proximity of increased abundances of the HNLC-PCC maximum to the O2 minimum in the eddy may thus point towards a beneficial relation between the HNLC-PCC and the eddy core water microbial community that largely consists of heterotrophic organisms.

original studies are missing in the references (Morris 2002, Rappe 2002)

We added the suggested references.
terial 16S rRNA gene copy numbers? What part of the population has the capability for
denitrification? Because of differences in cell numbers between eddy and the outside
the actual difference might not be as high as suggested.

AC11: The part of the community containing nirS is very minor in all samples, which
does not mean that the corresponding process is not active or important. (For compar-
ison, N2-fixers are considered to account for only 2% in ocean surface metagenomes,
however, they contribute up to 80% of new nitrogen to these waters). The absolute
number of nirS sequences measured against a standard dilution series cannot be
compared to 16S rRNA copy numbers from the amplicon sequencing, as this is would
mean comparing two different methods. However, we see a highly pronounced effect
denitrification on nitrous oxide (N2O) production within the eddy core waters based
on molecular and isotopic data (Grundle, Löscher et al.’ Extreme N2O activity in an
oxygenated ocean’, under review). This effect is not present outside the eddy, where
N2O is supposed to be exclusively produced by nitrification (see, e.g., Löscher et al.,
2012, Ryabenko et al., 2012). Therefore, we are convinced that our nirS dataset mir-
rors the presence/activity of the denitrification process, which is the key finding, here.
We added the reference (Grundle et al.) to the text.

RC12: p.14195, l. 12: With similar reasoning as in the comment above, it would make
sense to compare the transcripts of the nirS gene to a housekeeping gene.

AC12: I do not agree on that. The potential for nitrite reduction is not depending on the
overall abundance of bacteria or microbes or the biomass. It is more a question of the
presence of certain organisms in an environment that enables the respective process.
Absolute gene quantification is also from a methodological perspective preferable as it
mitigates the risk of including a second PCR bias.

RC13: p. 14197, l. 22: space missing
AC13: Changed.

Please also note the supplement to this comment:
http://www.biogeosciences-discuss.net/12/C7625/2015/bgd-12-C7625-2015-
supplement.pdf

Interactive comment on Biogeosciences Discuss., 12, 14175, 2015.