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Biogeosciences Discussions

Interactive comment on "Diversity of cultivated and metabolically active aerobic anoxygenic phototrophic bacteria along an oligotrophic gradient in the Mediterranean Sea" *by* C. Jeanthon et al.

Anonymous Referee #2

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Review of Journal: BG

Title: Diversity of cultivated and metabolically active aerobic anoxygenic phototrophic bacteria along an oligotrophic gradient in the Mediterranean Sea

Author(s): C. Jeanthon et al. MS No.: bg-2011-146 MS Type: Research Article Special Issue: Interactions between planktonic organisms and the biogeochemical cycles of biogenic elements in the Mediterranean Sea during intense summer stratification: the BOUM experiment

The authors present another chapter in the story of Aerobic Anoxygenic Phototrophic (AAP) bacteria dealing with their diversity of different oligotrophic water masses sampled along a transect of the Mediterranean Sea. This study focuses on (i) the cultivation, (ii) the molecular biological analyses of the pufM mRNA of metabolic active AAP bacteria and (iii) the phylogenetic analyses of both, AAP isolates and pufM mRNA reverse transcribed. The authors isolated about 50 AAP strains (all affiliated to the Alphaproteobacteria) by plating on oligotrophic media and using infra red fluorescence for detection. However, clone libraries of pufM gene transcripts revealed that the large majority of the phylotypes affiliated to the Gammaproteobacteria. Diversity analyses of pufM transcripts revealed highest diversity in the ultra-oligotrophic region of the Mediterranean Sea and that most environmental sequences were not affiliated to any cultivated bacteria. Due to their findings the authors emphasize the divergence of culture-dependent and culture-independent methods.

General comments:

The manuscript is a companion investigation to Lamy et al. (2011) reporting about the BChl-a and AAP bacterial abundance and linking to other (a-)biotic parameters, also published in BG. This study is a well addition to present knowledge regarding the in situ metabolically active AAP bacteria and completes the assemblage of several studies dealing with the same sample set within the BOUM experiment. Therefore, it is worth to be published in the special issue. Moreover, this study shows once again the gap of cultivation and molecular analyses, but nonetheless the use of a more degenerated primer recovering a wider diversity would have been preferable and may avoid the strengthening of discrepancy of both culture-dependent and culture-independent methods (see specific comments).

Specific comments:

Abstract, chapter 3.2, Tab 1+2: Did you mention the right number of isolates? Abstract 52, chapter 3.2: 54 minus the one strain not recovered = 53, 54 in Tab 1 and 53 in Tab

2.

Page 4423 line 7-9: Cell counting of AAP bacteria by the IREM method need to be interpreted with caution (Schwalbach and Fuhrman, 2005). That is the reason why abundance and significance of AAP bacteria were overestimated in the studies from Kolber et al. (2000, 2001). Data of Schwalbach and Fuhrman (2005) raise the question about where AA Photosynthesis is advantageous in marine ecosystems.

Page 4426 line 1: In which depths did the DCM occur? Give values or at least refer to section 3.2.

Page 4426 line 25: Illumination by which device? Give name, manufacturer, etc. of the used lamp for reproducibility regarding the spectrum of wavelengths.

Page 4426 line 28: Did you screen all plates and all media, respectively, for AAP bacteria? Refer media (MAD, MiA, MA?).

Page 4428 section 2.4: Not clear from which samples you generated RT pufM libraries.

Pages 4428 line 21: Why did you choose the nucleotide based primer pufMF of Béjà et al. (2002) for cDNA amplification of your diversity study? In 2005 Yutin and colleagues published fully degenerate pufM primers recovering a wider diversity of marine AAnPs than the nucleotide-based original pufM primers from Achenbach et al. (2001) and Béjà et al. (2002) and were used for e.g diversity analyses by DGGE (Yutin et al. 2008). Also other studies, e.g. Hu et al. (2006), used pufMF as forward primer and reported of abundant presence of AAP clones affiliated to the Gammaproteobacteria in oxic oceanic surface waters. Furthermore, primer pufMF discriminates several Roseobacter clones and strains. Maybe the choice of a more degenerated forward primer would have revealed another diversity pattern of the pufM transcripts and weaken the strong gap between culture-dependent and culture-independent methods.

Tab 3: Add a column with the accession numbers of the representative clones or refer it in the caption. May add a column of the phylogenetic affiliation or give abbreviations

in upper case.

Fig 1: What is the meaning of “SeaWiFS”? What is the meaning of “both”? Delete it and replace by e.g. additionally, etc.

Technical comments:

Abstract page 4422 line 18: . . .affiliated to. . .

Pages 4431 line 1-2: Syntax . . . from 21.4°C (station 5) to 26.9°C in the western basin.

Page 4432 line 7: . . .synthesize. . .

Page 4436 line 22: dimethylsulphoniopropionate

Page 4437 line 16: BChl-a synthesis

Page 4438 line 29: . . .abundance. Moreover,. . .

Tab 2: Roseovarius halotolerans

Fig 2: Description of the phylogenetic groups is different. Choose second one: Alpha-4 Proteobacteria, Alpha-1-Proteobacteria

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Interactive comment on Biogeosciences Discuss., 8, 4421, 2011.