

## ***Interactive comment on “Picoplankton diversity in the South-East Pacific Ocean from cultures” by F. Le Gall et al.***

### **Anonymous Referee #2**

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This descriptive paper reports on a nice and timely effort to culture the unculturable, here the marine photosynthetic eukaryotic picoplankton (PEP). Marine eukaryotic picoplankton is special in that it is a very heterologous group in which in the last decades novel classes and even a novel division was detected. As with other groups, these findings partially stem from environmental 18S sequencing and the relevant cultures are missing. PEP is more important in oligotrophic waters, so the focus of this study on oligotrophic ocean areas was a wise one. The paper adds new insights into picoplankton species abundance and occurrence.

The major aim of this study, the culturing of novel picoplanktonic groups mostly failed, with one possible exception which unfortunately later died. Was this the fault of the authors? Probably not. They started a large number (1900) of precultures of which

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roughly 10% yielded clean cultures. This number is in good agreement with other microbiological assemblages for which estimates exist of 90-99% to be unculturable. Perhaps it would have been better, to use more than just one medium (K) to culture novel PEP, especially from the very oligotrophic areas, but this is only hypothetical.

The analysis of pure cultures by LM, EM and partial 18S sequencing seems ok to me. I only wonder why not all cultured were sequenced, given that ~ 200 sequencing reactions no longer cost a fortune and the Roscoff Station has modern sequencing facilities. I assume not all could be amplified? Why? I also wonder whether it was always easy, to group the unknown sequences safely into an ARB tree if only partial (500 bp?) of 18S sequence were available. In our hands this sometimes is difficult e.g. because of poor bootstrap support.

Minor things: I could not find the names of the oligos Euk328f and Euk329r in the Moon et al 2000 paper. Were these really the oligos to amplify the total 18S rDNA? I could not read table 2 and 3 and figure 4, unless I use a magnification lense.

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Interactive comment on Biogeosciences Discuss., 4, 2699, 2007.

**BGD**

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