Planctomycetes diversity in lakes with differing trophic status: tests of specific primer sets and comparative analysis of their composition

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Planctomycete diversity in lakes with differing trophic status: test of specific primer sets and comparative analysis of their composition

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Context of the study

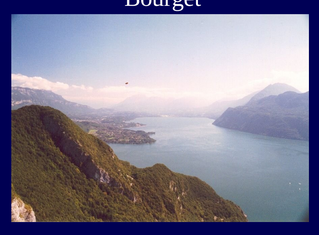
Knowledge on bacterioplankton diversity is crucial for understanding their dynamics and function in aquatic ecosystems. Over the last 20 years, most of the studies on this topic have dealt with the most abundant bacterial groups (e.g. *Actinobacteria, Proteobacteria, Flavobacteria*). Recent works suggested that other bacteria, considered less abundant, may also play a key role in aquatic system functioning. *Planctomycetes* are one of these groups, and are ubiquitous. They might play key roles in nitrogen cycling and in the degradation of dissolved organic matter[1,2]. However, little is known about these bacteria in lakes.

Objectives of PhD research*: Study the dynamics and diversity of *Planctomycetes* and their links with nitrogen cycle in lakes with differing trophic status.
First step of the study: Test the suitability of the existing primer sets and, if necessary, develop our own primer set.

Study sites

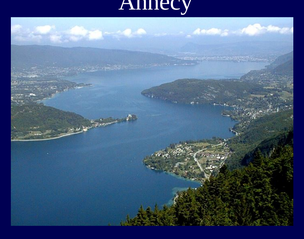
Subalpine lakes (France) | tropical lake (Burkina Faso)

Bourget



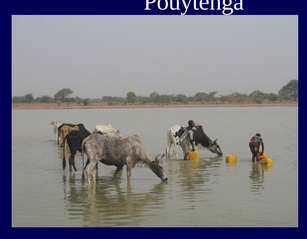
- Mesotrophic
- 145m
- Cyano blooms

Annecy



- Oligotrophic
- 65m

Pouytenga



- Eutrophic
- 0.7m

Methods

- 250 mL filtered through 2 and then 0.2µm membrane filters.
- Phenol-Chloroform extraction
- DNA amplification with 3 different primer sets described as *Planctomycete* specific
  - Pla46F/Pla886R
  - Pla40F/P530R
  - Pla352F/Pla920R
- Amplification products cloned and positive transformants sequenced
- Sequences alignment using Genedoc
- Sequences identification using RDPII (chimeric sequences excluded from alignment)

Results and discussion

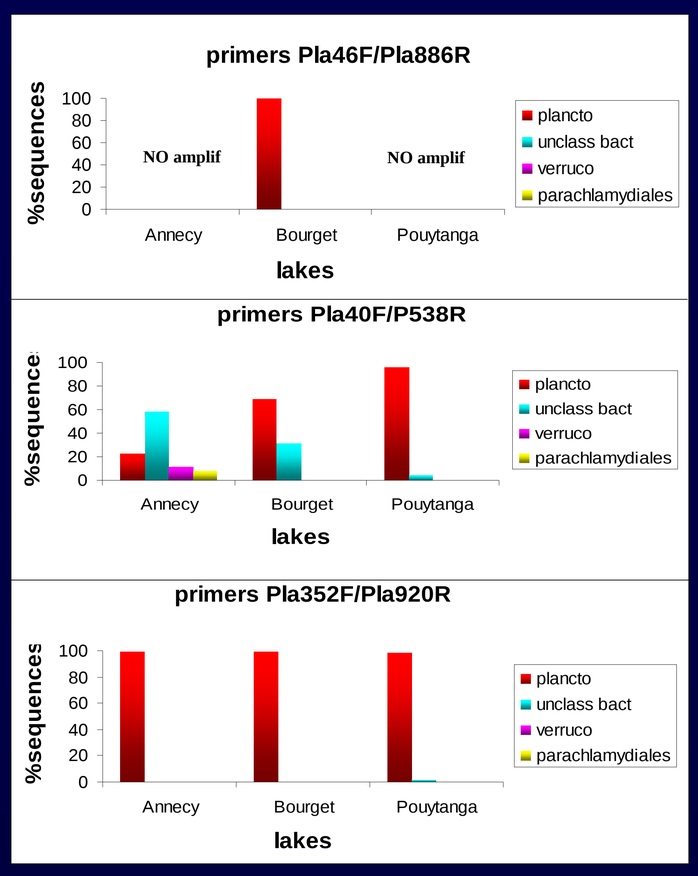


Fig 1: Percentage of sequences detected

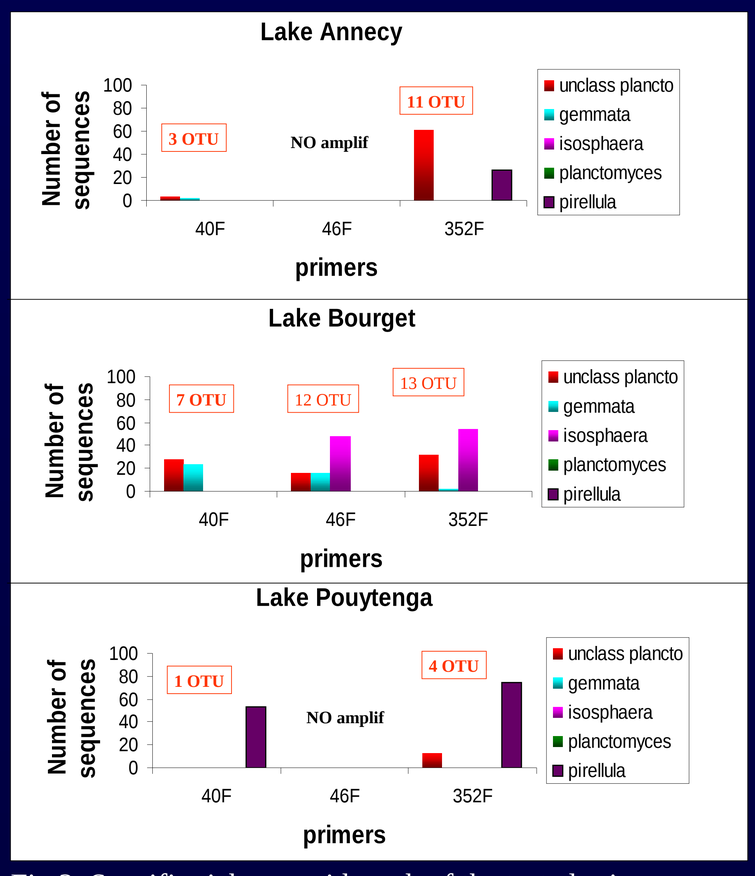


Fig 2: Specific richness with each of the tested primer set

No amplification with 46F
Number of OTUs: 352F >> 40F
≠ genera according to the primer

Number of OTUs: 352F and 46F>> 40F
No *Isosphaera* from 40F

No amplification with 46F
Number of OTUs: 352F >> 40F
≠ genera according to the primer

352F/920R detected a higher number of OTUs
However, need to increase the number of samples

- 46F/886R: good specificity BUT no amplification in Annecy and Pouytenga
- 40F/530R: poor specificity BUT positive amplifications from all lakes
- 352F/920R: good specificity AND positive amplifications from all lakes

352F/920R: best results for specificity

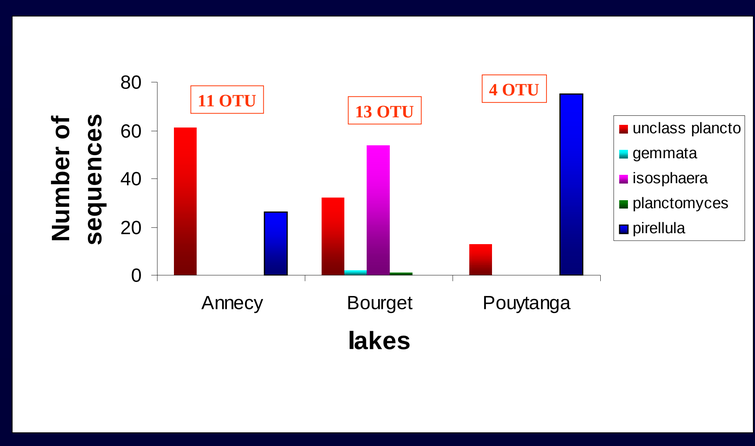


Fig 3: *Planctomycete* composition in the 3 studied lakes

- Higher specific richness and diversity in the subalpines lakes than in the African lake
- Similar specific richness in subalpine lakes but strong differences in the *Planctomycete* composition

Strong differences in the *Planctomycete* composition of the 3 lakes BUT results are preliminary and based on a small number of samples

Conclusion

The primer Pla352F/Pla920R was the best compromise to detect *Planctomycetes* in the studied lakes
- even though it seemed to underestimate some groups (e.g. *Gemmata* in Lake Bourget, Fig. 2),
- it was *Planctomycete* specific, detected them in all samples and detected higher number of OTUs.

Next steps: spatial and temporal distribution, experiments to test the effects of nutrients and links with nitrogen functional genes.

References: 1 – Strous et al 1999: Nature. 400: 446-449; 2 – Tadonléké, 2007: FEMS Microb. Ecol. 59:543-555

Acknowlegments

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