

Development of improved preservation techniques for cyanobacterial and diatom strains in the BCCM collections

Charlotte Crahay¹, Olga Chepurnova², John G. Day³, Annick Wilmotte¹, Wim Vyverman²

1 University of Liège, Centre for Protein Engineering, Allée de la Chimie 13 B6a, 4000 Liège, Belgium, 2 BCCM/DCG Diatoms Collection, Laboratory of Protistology and Aquatic Ecology, Department of Biology, Ghent University, B-9000 Gent, Belgium, 3 Culture Collection for Algae and Protozoa, Scottish Marine Institute, Oban, Argyll PA37 1QA, UK

The implementation of reliable preservation technologies of the biological resources is crucial for the management of the Biological Resources Centers. The BRAIN-be project PRESPHOTO (preservation of photosynthetic micro-algae in the BCCM collections) (www.presphoto.ulg.ac.be) aims to improve the preservation of cyanobacterial and diatoms strains as well as their genetic information in the BCCM/ULC and BCCM/DCG collections, respectively.

We have adapted the traditional two-step cryopreservation technique to photosynthetic microalgae strains. The encapsulation/dehydration technique as alternative to the two-step cryopreservation method was also evaluated. In particular, we have examined the effects of the culture conditions, the cryoprotectants choice. Diatoms were found to be particularly sensitive to post-cryopreservation procedures which greatly affect cell viability. We have also compared storage at -70°C and in liquid nitrogen (-196°C) for cyanobacteria whereas only the latter was used for diatoms. In addition, a vital staining method, allowing the rapid evaluation of post-cryopreservation viability has been assessed for cyanobacteria. The selected cryopreservation protocol was further validated on 26 cyanobacterial strains while for diatoms, best results were obtained by optimization of the generic protocol for individual species. An independent validation of the developed protocols will be performed by the Culture Collection of Algae and Protozoa (UK) (subcontractor) in the final phase of the project.

In addition to the preservation as living strains, we also have focused on the DNA storage improvement. We have compared different DNA extraction, storage and quality control methods. In addition, for filamentous cyanobacteria, different pretreatments were tested to disorganize the polysaccharidic sheaths that hinder DNA extraction.