Cryopreservation of microbial consortia for industrial biotechnology

The Department of Plant Sciences, University of Cambridge has three strategic targets currently - global food security, various types of synthetic biology and biotechnology, and climate science and ecosystem conservation and the results of their work can relate to those targets or be in pure science applicable in diverse areas including medicine. One aspect being investigated is how algal-bacterial relationships influence algal productivity.

Microalgae are found in almost every aquatic ecosystem as well as many terrestrial ecosystems. This diversity of lifestyles has resulted in an extremely large group of organisms with divergent metabolic and functional characteristics. As a result, they possess considerable biotechnological potential, particularly in the bioenergy, cosmeceutical and nutraceutical markets, but much is still unknown on their biology and exploitability.

Chris Ridley from Prof Alison Smith's lab in Cambridge has been working at the Scottish Association for Marine Science/University of the Highlands and Islands to explore the possibility of conserving an algal-bacterial consortium using cryopreservation. By culturing algae with symbiotic bacteria, they have managed to reduce nutrient inputs and prevent contamination by pathogenic bacteria – a major problem in commercial processes.

Chris Ridley of the Department of Plant Sciences, University of Cambridge

To develop commercial algal biotechnology, consistent and functional inocula are required. Cryopreservation is widely regarded as the preferred method for the long-term, genetically and phenotypically stable, storage of stock cultures. They utilised a model symbiotic interaction between the green alga *Lobomonas rostrata* and the soil bacterium *Mesorhizobium loti*, in which the alga receives cobalamin (vitamin B_{12}) from the bacterium in exchange for photosynthate. To help in this they needed to develop a 'co-cryopreservation' method. Their

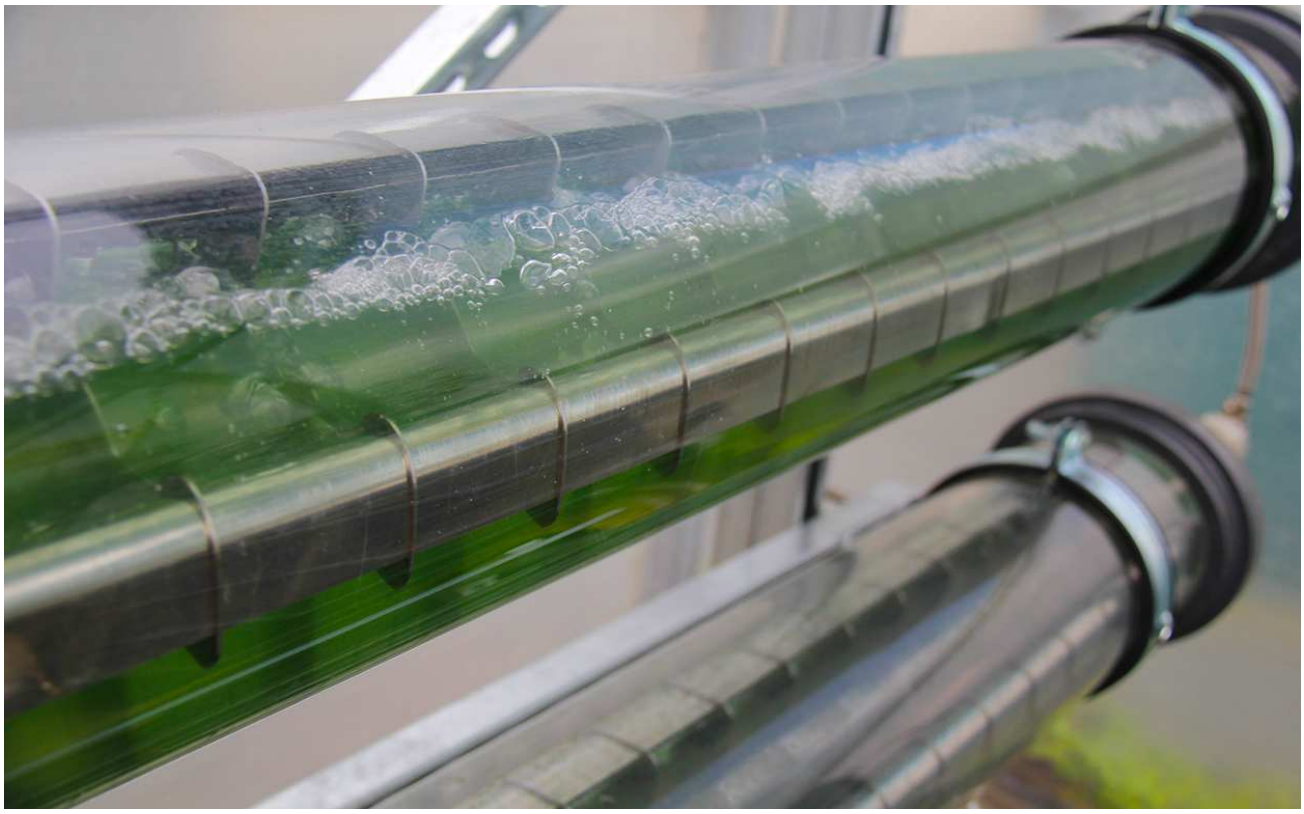
resulting technique ensures a maximum post-thaw viability (PTV) of both algae and bacteria, and preserves the symbiotic interaction.

The work was achieved utilising a Planer Kryo controlled rate cryogenic freezer, taking advantage of the ability to cool at a defined rate. The protocol was optimised to ensure maximum post-thaw viability (PTV) of both algae and bacteria, and so to maintain the symbiotic interaction. Over 90% PTV was achieved after testing several colligative cryoprotectants, including different concentrations of DMSO and MeOH, as well as varying the conditions of storage immediately post-thawing.

Controlled rate freezers (CRF), such as Planer's Kryo 560, are widely used when freezing biological material that cannot well withstand the uncontrolled freezing rates of mechanical or liquid nitrogen immersion. Samples, usually treated with cryoprotectants, are placed in the CRF in vials, bags or 'straws' and vaporised liquid nitrogen is fanned through the chamber. This can be very accurately controlled and so programmes governing the rate of freezing, and any holds needed to equilibrate the chamber, can be written and programmed into the CRF's controller. Apart from the advantages of repeatable, consistent results using a CRF, help with improved post thaw cell viability by handling latent heat at freezing points and then taking samples below the glass transition point to -180C

Chris worked under Prof John Day at the Scottish Association for Marine Science. John is head of the culture collection of algae and protozoa at there and focusses on algae and protozoa especially when these relate to algal biofuels and biotechnology, cryopreservation and cryoinjury and the use of molecular method in culture collections

This work by John Day and Chris Ridley is the first demonstration of the potential for long-term storage of microbial consortia for industrial biotechnology.



Green alga Chlamydomonas reinhardtii growing in a photobioreactor at the Algal Innovation Centre, University of Cambridge

Planer controlled rate freezer

Prof John Day

See more at
http://www.sams.ac.uk/john-day
http://www.plantsci.cam.ac.uk/directory/smith-alison
http://planer.com/