Docosahexaenoic acid (DHA) production in the heterotrophic marine microalgae Cryptothecodinium cohnii

Friederike Hillig, Agnieszka Niedziółka, Stefan Jahns, Stefan Junne, Peter Neubauer

Technische Universität Berlin, Department of Biotechnology, Chair of Bioprocess Engineering, Ackerstraße 71-76, ACK 24, 13355 Berlin, Germany. Tel.: +49-30-314-72576, e-mail: f.hillig@tu-berlin.de, http://www.bioprocess.tu-berlin.de

Motivation

Docosahexaenoic acid (DHA) is a polyunsaturated fatty acid (PUFA) with a beneficial effect on human health. The main source of DHA for the human diet is fatty sea fish. To maintain a high amount of DHA in fish grown in aquaculture, fishes are fed with fish oil. Fish oil is also used as an additive in the food industry. The utilization of this fish oil has several drawbacks, as it contributes strongly to the ocean’s overfishing and contains environmental pollutions [1].

C. cohnii is a heterotrophic marine dinoflagellate which can be used for biotechnological DHA production [1]. The process is divided in two phases: in a growth phase and a shorter production phase, in which the amount of DHA per cell can be doubled. In the project a screening system in deep well plates with the application of oxygen vectors was developed. With this screening system a DHA content of 26 % per cell dry weight was reached at the end of the growth phase. The possibility to obtain this results in larger scale has to be demonstrated.

Challenges in process development:

• High oxygen demand during growth and synthesis of fatty acids
• Aigal cells are sensitive to high shear forces → hamper the application of directly aerated systems [2].
• High chloride concentration of the media → corrosion of steel reactors.
→ To overcome these limitations the application of polymer based bioreactors as alternative are evaluated.

Results

CELL-tainer® – wave - mixed system (CELLution Biotech)

Cultivation:
• Start volume: 8 L
• Obtained cell dry weight (CDW): 53 g/L
• DHA content per CDW: 29 %
→ The results are similar to those obtained in a 5 L stirred tank bioreactor (data not shown)
• The cell dry weight and the volumetric DHA content was higher than in the Kühner reactor due to sophisticated process control and a higher oxygen transfer rate
• Application of the celltainer for a fed-batch cultivation with Cryptothecodinium cohnii was feasible
→ For the purpose of industrial application, a scale up of this system has to be performed

SB 200x orbital shaking system (Kühner)

Cultivation:
• Start volume: 100 L
• CDW: 37 g/L
• DHA content per CDW: 28 %
→ Cells could withstand oxygen limitation better than at other reactor systems
→ The cell dry weight and the volumetric DHA content was smaller than in the celltainer
→ In future application of improved process control will allow for the achievement of higher amounts of biomass

Outlook

• Both system designs seem to be adequate for further development for the application in marine bioprocesses.
• A process control using the exhaust gas analysis will improve process robustness.
• Applying our developed plate screening system, the influence of different hydrophobic and hydrophilic additives on the DHA production is investigated to enhance the volumetric DHA content in the production phase.

Acknowledgements

The authors would like to thank Mare Nutrica GmbH (Niendorf, D) for financial support, CELLution Biotech BV (Assen, NL) and Kühner AG (Birsfelden, CH) for providing the bioreactors.