MICROBIAL CULTIVATION IN ROCKING SINGLE-USE BIOREACTORS

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The application of single-use bioreactors (SUB) for microbial cultivation, especially of reactor designs beyond the traditional stirred tank, is usually regarded as crucial. Especially the usually low gas mass transfer coefficients are insufficient, however this is not true for 2-dimensionally rocking motion bioreactors like the CELL-tainer®. Volumetric gas mass transfer coefficients (kLa-values) of 600 h^{-1} are achieved (fig. 1), which allow bacterial fed-batch cultivations up to a cell density of 50 gL^{-1} at growth rates of 0.3 h^{-1} w/o any oxygen blending (Junne et al, Chem Eng Technol 2013, 85, p. 57-66). One major asset in this respect are the low maximum shear forces in comparison to stirred tank reactors. This feature might be beneficial when shear sensitive microbes are cultivated, like marine phototrophic and heterotrophic microalgae and filamentous organisms.

Therefore, in this study, the advantages of a rocking single-use bioreactor concept is shown from the mL to the 120 L scale at the example of the marine heterotrophic microalgae *Cryptecodinium cohnii* for the purpose of optimizing the production of the polyunsatured fatty acid docosahexaenoic acid (Hillig et al, Eng Life Sci 2014, 14(3), p. 254-263). Corrosion due to the high chloride concentration in such a media exhibits a major challenge for the cultivation in steel bioreactors. Additionally, the targeted mixotrophic process requires oxygen excess, although the shear forces need to be low in order to exclude any impact of them for a suitable process development.

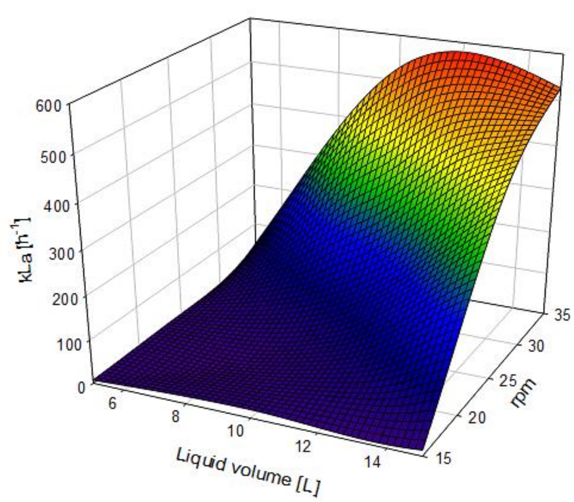

Figure 1 – oxygen transfer in the rocking motion single-use bioreactor CELL-tainer® CT 20 depending on the filling volume and rocking speed

Additionally, the conduction of filamentous cultivations in the CELL-tainer is shown, e.g. for the antibiotics production of *Aspergillus niger* and *Streptomyces clavuligerus*. Both cultivations show tremendously higher productivity while exhibiting different morphologic characteristics than if grown in stirred tank reactors. Pellet formation is avoidable under certain cultivation conditions, thus leading to pure filamentous growth. Since shear forces as low as usually obtained in shake flasks can be achieved in rocking bioreactors, a scale up from the shake flask scale at similar shear forces is performed. When compared to stirred tanks, a study of the impact of shear forces under fully controlled cultivation conditions is conducted. The system, if applied with the so-called expansion channels, can be used for seed train and scale up even w/o changing the reactor, as the liquid volume can vary by two orders of magnitude. Another suitable application of such SUB is a continuous process with cell retention, in which the surface to volume ratio can be kept at high values, if pellet formation is avoided. Thus, a high interfacial area is achieved, in which product secretion can be maintained at considerably high values.

Although rocking systems are not as good described as stirred systems in terms of traditional engineering parameters, the question arises whether it is even useful to consider traditional engineering parameters only. The consideration of physiological and morphological states of cells in order to choose the best-suited system for an individual purpose might circumvent the lack of detailed system descriptions. Such an approach can provide a new process design, in which the bioreactor conditions are optimized for the strain rather than the strain need to be optimized to grow in traditional stirred tank bioreactor designs