

The effect of hydrodynamic conditions in *Corynebacterium glutamicum* growth

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Corynebacterium glutamicum is a facultative anaerobic, gram-positive bacterium with a GRAS status that grows fast and achieves high cell densities. *C. glutamicum* is commonly used in amino acids production, and is also able to convert sugars in organic acids (OA) and alcohols in specific conditions: anaerobic and limited-oxygen environments. In these conditions, the carbon metabolism is modified, namely the flux shifts from the phosphate pathway to glycolysis and the TCA cycle flux decreases and consequently bacterial growth is strongly affected [1,2].

This work analyses the effect of hydrodynamic conditions in *C. glutamicum* ATCC 13032 growth. Different experiments were conducted in a 5-L stirred-tank bioreactor, with 3.5 L or 2.5 L working volumes. The temperature and pH were automatically controlled at 30 °C and 7, respectively, and dissolved oxygen (DO) was controlled at 30 %, through a cascade control strategy. In the experiments with 2.5 L of working volume, different initial agitation rates were tested: 500, 650 and 800 rpm.

In a first approach, the influence of reducing the working volume on *C. glutamicum* growth was evaluated and it was determined that for the same initial agitation rate. Reducing the working volume from 3.5 to 2.5 L resulted in a 78 % increase on biomass concentration. Considering that the DO control was effective throughout the operation and that the measured DO value was always around 30 %, this result confirms that the oxygen distribution was not homogenous in the 3.5 L experiments, and probably some dead zones were generated, where pO₂ concentration could be lower than the established value. This interpretation was also corroborated by the detection of OA in the fermentation broth of the 3.5 L experiments. The volume reduction eliminates the dead zones, improving the biomass synthesis and reducing OA production.

Further experiments were carried out with a 2.5 L working volume and different initial agitation rates: 500, 650 and 800 rpm. The increase in the agitation rate from 500 to 650 rpm leads to a 1.14-fold increase in growth. Nevertheless, only 1-fold increase was observed in experiments at 800 rpm. The reduction of the working volume and increasing the initial agitation rate to 650 rpm resulted in a 98 % reduction of the OA production.

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[1] Y. Tsuge, T. Tateno, K. Sasaki, T. Hasunuma, T. Tanaka, A. Kondo, *AMB Express* 7, 2013, 1-7.

[2] A. Zahoor, A. Otten, V. Wendisch, *Journal of Biotechnology* 192, 2014, 366-375.