296c In Situ Product Removal by Direct Crystallization: an Attractive Approach for Process Integration of Whole-Cell Biocatalyzed Reactions

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ABSTRACT In biochemical industry, downstream processing is often the cost-limiting factor during the production of pharmaceutical and fine-chemical products because low product concentrations are usually obtained from the reactor, requiring a large train of separation and purification steps. On the other hand, when fermentation or biotransformation is run to reach high product concentrations, volumetric productivity is often limited by either product inhibition or degradation, such that in situ product removal (ISPR) is useful in these cases.

In this work, the synthesis of 6R-dihydro-oxoisophorone (DOIP), also known as levodione, is considered. Levodione is a white crystalline product, with a solubility of about 10 g.L-1 in water, and is an important intermediate in the production of some carotenoids and saffron flavors. The synthesis reaction (Figure 1) involves the asymmetric reduction of 4-oxoisophorone (OIP) using yeast such as Saccharomyces cerevisiae or Saccharomyces rouxii as biocatalyst [1-3]. An enantiomeric excess (e.e.) □ 98% for levodione is obtained in this process. The main product (levodione) is further degraded by the yeast mainly to (4S,6R)-actinol, an unwanted by-product in the process, such that in situ product removal by crystallization is employed to minimize degradation.

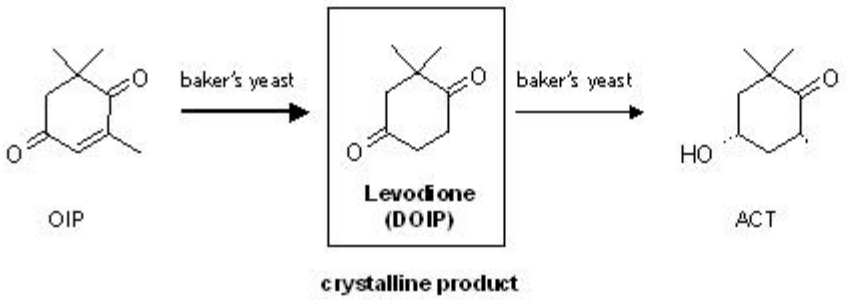
ISPR by crystallization with external configuration has been successfully implemented using resting cells [1] and growing cells [2] of S. cerevisiae resulting in an efficient integrated fermentation-crystallization process. Relative to the conventional batch and fed-batch processes, the integrated fermentation-crystallization process configuration has a better performance in terms of final yield (85%), final selectivity (96-98.7%), and volumetric productivity (0.30-0.92 g.L-1.h-1) [1-2]. In order to further evaluate the process, a conceptual process design is done for the production of levodione implementing ISPR by crystallization, and is compared with the base case, which is a known conventional process equivalent [3].

On the same basis, the comparison indicates that employing ISPR by crystallization has potential advantages over the conventional process (base case) in terms of increased productivity and yield with corresponding decrease in the number of downstream processing steps as well as the quantity of waste streams. This leads to an economically more interesting process alternative.

Figure 1. Synthesis of DOIP from 4-oxoisophorone (OIP) by baker's yeast. By-product formed is actinol (ACT). The desired product (levodione) is recovered from the reactor by in situ crystallization to avoid degradation.

Word count: 370 (maximum 1500)

References [1] Buque-Taboada EM, Straathof AJJ, Heijnen JJ, van der Wielen LAM. 2004. In situ product removal using a crystallization loop in asymmetric reduction of 4-oxoisophorone by Saccharomyces cerevisiae. Biotechnol Bioeng 86, 795-800. [2] Buque-Taboada EM, Straathof AJJ, Heijnen JJ, van der Wielen LAM. 2005. Microbial reduction and in situ product crystallization coupled with biocatalyst cultivation during the synthesis of 6R-dihydro-oxoisophorone. Adv Synth Catal, in press. [3] Fukuoka M, Hiraga K, Sekihara T. 2001. Microbial production of levodione. European Patent (EP) 1074630A2.



crystalline product