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Effective bioproduction of natural aromas with coupled bioreactor-pertraction-adsorption hybrid system

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Highlights

- Hybrid system for 2-phenylethanol bioproduction was constructed.
- Biotransformation in hybrid system can be prolonged from 24 hours to 72 hours.
- Low concentrations of 2-phenylethanol avoiding the biomass inhibition were achieved.

1. Introduction

A common characteristic of many biotechnological productions, compared to classical chemical processes, is low reaction rate. Furthermore, bioproduction can suffer from inhibition, which decreases the reaction rate even more. Often, biocatalyst produces a compound causing its own inhibition, which can lead to the production termination. In these cases, a continuous product removal system can be integrated to the reaction system in order to intensify the production. In such integration of a reaction and a separation processes a hybrid system is created which through separation of inhibitory product enhances the production and facilitates further downstream processing. In this study of such hybrid system, a biotechnological production of 2-phenylethanol, which is a higher aromatic alcohol, was chosen. Because of its characteristic rose-like fragrance, 2-phnenylethanol's main application is in cosmetics and food industry where only natural 2-phenylethanol may be used, which can be produced by yeasts *Saccharomyces cerevisiae*. Under growth conditions, yeasts transform the precursor L-phenylalanine to 2-phenylethanol, which has a strong inhibitory effect on the biomass growth [1]. In 24 hours biomass can accumulate in the fermentation medium total growth inhibiting concentration of 4 g/L of 2-phenylethanol [2]. To intensify the bioproduction, low 2-phenylethanol concentration has to be present in the bioreactor. For this purpose, an effective continuous separation system has been designed combining a bioreactor, membrane module and an adsorption column into one operational unit (Fig. 1). Fermentation medium from the bioreactor is continuously fed to the separation part of the hybrid system, where first the pertraction through a supported liquid membrane and subsequently the adsorption in a column takes place. The supported liquid membrane was created by octane in the pores of a membrane module. In the pertraction membrane module, cell free aqueous stripping phase containing 2-phenylethanol can be obtained [3] since the octane is immiscible with both feed fermentation medium and stripping aqueous phase. The cell free aqueous phase is then fed to the adsorption column, where 2-phenylethanol is accumulated on the surface of the adsorbent. Output from the column, free of 2-PEA can be reused in the pertraction membrane module.

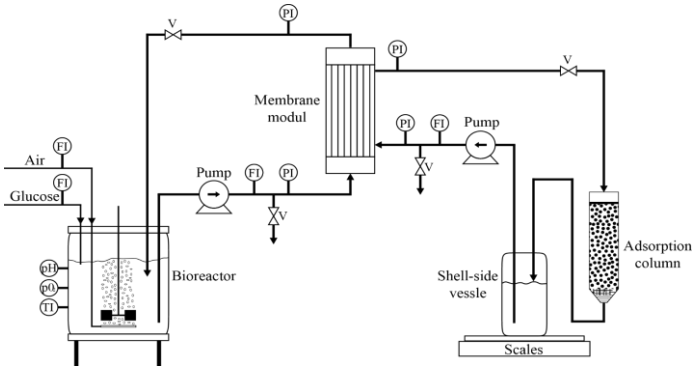

Figure 1. Proposed hybrid system for continuous product removal during bioproduction.

2. Methods

Biotransformation experiment with continuous product removal was carried out in a bioreactor with the working volume of 3 L. Initial concentration of biomass and glucose was 5 g/L and 15 g of L-phenylalanine was added to the bioreactor. Throughout the biotransformation, additional glucose and L-phenylalanine were fed to the bioreactor. After 6 hours of the biotransformation, continuous 2-phenylethanol removal started. Fermentation medium was fed to the membrane module with a 0.75 m^2 of contact area where pertraction occurred. Stripping phase from the membrane module was led to the adsorption column where 107 g of commercial adsorbent Macronet MN270 were placed. Samples for 2-phenylethanol analysis were taken from the bioreactor, from the stripping phase leaving the membrane module and from the output of the adsorption column every three hours and they were analyzed on HPLC.

3. Results and discussion

Membrane module for the pertraction did not allow any biomass from the feed phase to contaminate the stripping phase. Supported liquid membrane created by octane in the membrane pores retained L-phenylalanine in the bioreactor while transporting 2-phenylethanol to the stripping phase, which was further adsorbed onto the adsorbent. The hybrid system was working for 72 hours, during which 54 g of 2-phenylethanol were produced. In the adsorption column, 85% of all produced 2-phenylethanol was adsorbed (Fig. 2-B). During the biotransformation, the 2-phenylethanol concentration did not reach 4 g/L, which would induce total inhibition of the biomass growth, but remained low even in the most intensive production phase, where biomass grew exponentially (Fig. 2-A).

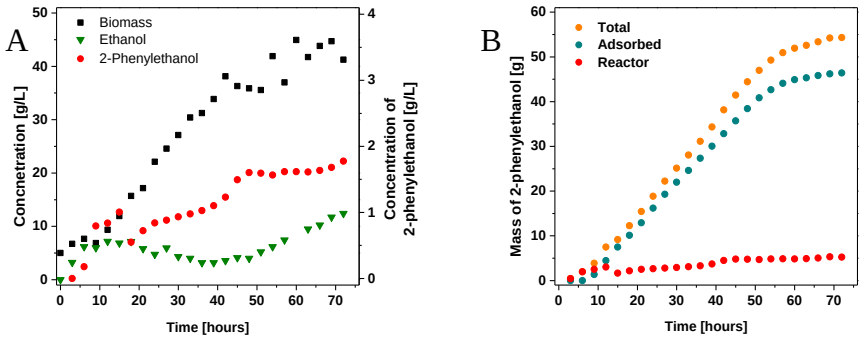

Figure 2. Concentration of biomass, ethanol and 2-phenylethanol during the biotransformation experiment (A) and mass of produced 2-phenylethanol present on the adsorbent and in the reactor (B).

4. Conclusions

The aim of this work was to design an effective hybrid reactive-separation system for 2-phenylethanol bioproduction. For this purpose, one operational unit consisting of a bioreactor interconnected with a membrane module and an adsorption column was constructed. In a 72-hour biotransformation experiment, the concentration of 2-phenylethanol in the bioreactor was below 2 g/L and the average volumetric production rate of 2-phenylethanol was 0.28 g/L/h compared to 0.17 g/L/h in a classical batch fermentation.

Acknowledgements

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References

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Keywords

Product removal, pertraction, adsorption, 2-phenylethanol.