SELECTIVITY OF A BIORECEPTOR ELEMENT OF THE SENSOR BASED ON AN ORGANOSILICON MATERIAL AND MICROORGANISM CELLS

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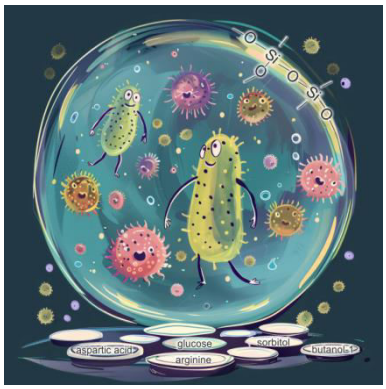
Abstract

The yeast cells from *Ogataea polymorpha*, *Blastobotrys adeninivorans*, and *Debaryomyces hansenii* were immobilized on an organosilicon material by the sol-gel method, resulting in a hybrid biocatalyst. The catalytic activity of the immobilized mixture of microorganisms was evaluated using it as a bioreceptor element in a biosensor. The proposed biohybrid material, consisting of the yeast association immobilized on the organosilicate sol-gel matrix, exhibited broad substrate specificity. Thus, the immobilization of multiple yeast strains provides a heterogeneous catalyst able to biologically oxidize a wide range of organic compounds commonly found in wastewater and surface water.



Key words: sol-gel encapsulation, yeast, association, selectivity.

Introduction

Among the existing technologies, a sol-gel technique stands out as a promising approach for the development of advanced biocatalysts [1]. Sol-gel chemistry, known for its availability of reagents and mild reaction conditions, is widely used in environmental applications [2]. For example, in the development of immobilizing agents for live microbial cells. In environmental monitoring it is necessary to determine the integral characteristics of the degree of wastewater contamination [3], for which microorganisms must be securely anchored without loss of activity and have broad substrate specificity. The immobilization of single microorganisms in organosilicon sol-gel matrices of different compositions has been reported [4-6], but the joint immobilization of several microorganisms will allow for enhancing the substrate specificity of the resulting biocatalyst. This is important for the application of biocatalysts in determining the integral indicators of water pollution.

Results and discussion

The yeast cells *Ogataea polymorpha* VKM Y-2559, *Blastobotrys adeninivorans* VKM Y-2677, and Debaryo*myces hansenii* VKM Y-2482 were immobilized on an organosilicon material by the sol-gel method, resulting in a hybrid biocatalyst. An organosilicon matrix based on tetraethoxysilane and methyltriethoxysilane under basic catalysis conditions was used as the immobilizing agent. Poly(vinyl alcohol) was used to prevent excessive melting of the material. The catalytic activity of the immobilized microbial mixture was evaluated by using it as a bioreceptor element of the biosensor on the surface of the Clark oxygen electrode. The proposed biohybrid material, consisting of the yeast association immobilized on the organosilicate sol-gel matrix methyltriethoxysilane/ tetraethoxysilane 85/15 vol %, showed broad substrate specificity. The encapsulated yeast cells demonstrated the ability to oxidize a wide range of compounds, including alcohols, sugars, amino acids, organic acids, and surfactants, which are commonly found in industrial and domestic wastewater (Fig. 1). This broad substrate specificity is crucial for the development of efficient heterogeneous biocatalysts.

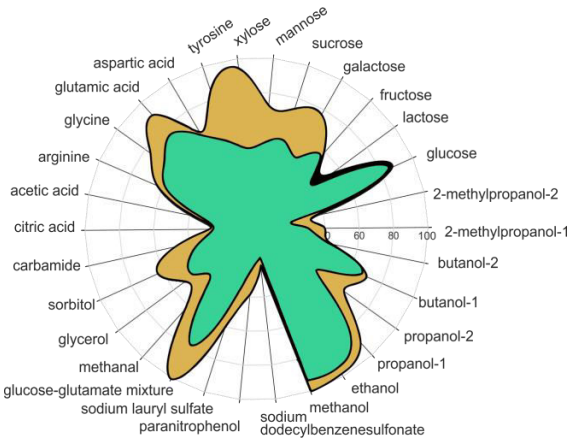


Figure 1. Substrate specificity profile of the artificial yeast association immobilized in the sol-gel matrix MTES/TEOS 85/15 and individual yeast strains.

Compared to the individual microbial cultures, the immobilized yeast association showed the enhanced ability to oxidize a wider range of substrates (Fig. 1). For example, the

yeast association can oxidize disaccharides (*e.g.*, xylose), monohydric alcohols (*e.g.*, 1-propanol, 2-propanol), amino acids, and surfactants. This wide range of the substrates is probably due to the diversity of metabolic pathways present in different yeast strains, allowing the oxidation of intermediate metabolites produced by other strains. Thus, the immobilization of multiple yeast strains allows for the development of a heterogeneous catalyst able to biologically oxidize a wide range of organic compounds, which are commonly found in wastewater and surface water.

Experimental section

Catalytic activity of the cells. An Expert-001 multifunctional analyzer (Ekoniks, Russia) in the thermooximeter mode was used to record the signal. Before and between the measurements, the system was rinsed with a potassium sodium phosphate buffer solution (4 cm^3, 20 $mmol/dm^3$, pH = 6.8). The substrate was introduced into the cuvette where it was oxidized with the cells immobilized on the surface of the oxygen electrode. Simultaneously, the oxygen consumption by the cells increased and the oxygen concentration in the vicinity of the electrode decreased, which was recorded by the electrode. The measured parameter was the maximum rate of change of the dissolved oxygen concentration upon addition of the substrates.

The following compounds were used as the substrates: 2-methylpropan-1-ol, 2-methylpropan-2-ol, glucose, lactose, fructose, galactose, sucrose, mannose, xylose, tyrosine, asparagic acid, glutamic acid, glycine, arginine, citric acid, acetic acid, urea, sorbitol, glycerol, methanol, ethanol, glucose–glutamate mixture, *para*-nitrophenol, sodium lauryl sulfate (SLS), methanal, propan-1-ol, propan-2-ol, butan-1-ol, and butan-2-ol.

Conclusions

To summarize the results presented, the yeast cells from *Ogataea polymorpha* VKM Y-2559, *Blastobotrys adeninivorans* VKM Y-2677, and *Debaryomyces hansenii* VKM Y-2482 were immobilized on the organosilicon material by the sol-gel method, resulting in the hybrid biocatalyst. The encapsulated yeast cells demonstrated the ability to oxidize a wide range of compounds, including alcohols, sugars, amino acids, organic acids, and surfactants, which are commonly found in industrial and domestic wastewater. This broad substrate specificity is crucial for the development of efficient heterogeneous biocatalysts. Hence, the immobilization of multiple yeast strains allows for the development of a heterogeneous catalyst able to biologically oxidize a wide range of organic compounds in wastewater and surface water.

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