Activity and stability of β-galactosidase in SC CO_2

Mateja Primožič, Katja Vasić, Željko Knez, Maja Leitgeb*
University of Maribor, Faculty of Chemistry and Chemical Engineering, Laboratory for separation processes and product design, Smetanova 17, SI-2000 Maribor, Slovenia
*e-mail: maja.leitgeb@um.si

Modern immobilization methods allow enzyme immobilization with or without a carrier in the form of micro- or nano-particles. Their applications can be found in various fields, such as biomedicine, biosensors or bioreactors for different industries or environmental protection.
Production of cross-linked enzyme aggregates (CLEAs) is a simple and cheap method of enzyme immobilization where no carrier is used. There is also no need of enzyme purification prior the CLEAs formation.
The enzyme β-galactosidase was immobilized by two different methods: as cross-linked enzyme aggregates (CLEAs) and as magnetic cross-linked enzyme aggregates (mCLEAs).
Prepared immobilized enzyme was exposed in the supercritical carbon dioxide (SC CO_2) at different temperatures (50 °C and 80 °C) and at different pressures (100 bar, 200 bar and 300 bar). Activity of the immobilized enzyme was measured after each exposure to SC CO_2. Activity of immobilized enzyme (CLEAs and mCLEAs) after lyophilization was determined as well. Also, stability of pure enzyme in SC CO_2 was determined. Activity assay for β-galactosidase was performed using o-Nitrophenyl β-D-Galactoside as a substrate. Later on, the activity of immobilized and pure enzyme was defined spectrophotometrically with measuring absorbance at 405 nm. Regarding to the immobilized enzymes after lyophilisation, in comparison to CLEAs, the mCLEAs shows better stability at storage conditions.

The highest residual activity for CLEA and mCLEAs was obtained when applying the pressure of 300 bar at 80 °C.