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The *Candida rugosa* lipase adsorbed onto titania as nano biocatalyst with improved thermostability and reuse potential in aqueous and organic media

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Abstract
The immobilization of *Candida rugosa* lipase by adsorption was performed onto commercial titania powder (Degussa P25). The change of titania particles surface was diagnosed by means of FTIR and FESEM analysis, as well as by shift of zeta potential value towards that of lipase. A detailed study of the effect of immobilization on enzyme kinetic, temperature stability, as well as on potential for its reuse in aqueous organic media was undertaken. Immobilization of lipase altered enzyme affinity toward substrates with different length of carbon chain in hydrolytic reaction. The Vmax value decreased 2-8-fold, where major constraint was registered for the ester containing the longest carbon chain. Thermostability of lipase improved more than 7-fold at 60 ^{o}C. Significant potential for reuse in water solutions was also found after immobilization. In cyclohexane immobilized lipase catalyzed synthesis of amyl octanoate by ping-pong bi-bi mechanism with inhibition by amyl alcohol. Obtained kinetic constants were Vmax=26.4 μmol/min, K_{Ac}=0.52 mol/l, K_{Al}=0.2 mol/l and $K_{i,Al}$=0.644 mol/l. Esterification activity remained 60 % after 5 reuse cycles in cyclohexane indicating moderate reuse stability.

Keywords
Candida rugosa lipase, Degussa P25 titania, Immobilization, Kinetic parameters, Cyclohexane