

Enzymatic peptide hydrolysis: Measuring kinetics, reaction equilibrium and enzyme activity

Co-solutes have a non-negligible influence on the reaction kinetics and equilibrium position of enzyme-catalyzed reactions. Depending on the kind of reaction and kind of co-solute, this opens the door to design reaction medium of enzyme-catalyzed reactions towards improved rates and yield of the reaction products, while keeping mild reaction conditions and high stereoselectivity.

In order to design a process with enzyme-catalyzed reactions, the stability of the enzyme, the reaction rate and the equilibrium composition have to be known in the whole process window. In this practical course participants will be guided to measure enzyme stability, the Michaelis constant K_M and the equilibrium composition K_x for the hydrolysis reaction of N-succinyl-L-phenylalanine-p-nitroanilide (SPNA) catalyzed by the enzyme α -chymotrypsin in a neat buffer solution and under the influence of the co-solutes urea and dimethyl sulfoxide. Measurements of kinetic and equilibrium data will be performed via UV-Vis measurements after calibration with 4-nitroanilide, which is one of the products of the studied reaction.