

# Investigation and Enhancement of Protein Stability – Bommarius

*Determination of long-term process stability of protein biocatalysts through short-term experiments*

The deactivation of protein biocatalysts even at relatively low temperatures is one of the principal drawbacks to their use. We have derived an equation for both time- and temperature-dependent activity of the biocatalyst based on known concepts such as transition state theory and the Lumry-Eyring model. We then derived an analytical solution for the total turnover number (ttn), under isothermal operation, as a function of the catalytic constant  $k_{cat}$ , the unfolding equilibrium constant  $K$ , and the intrinsic first-order deactivation rate constant(s)  $k_{d,i}$ . Employing an immobilized glucose isomerase biocatalyst in a CSTR and utilizing a linear temperature ramp beyond the  $T_m$  of the enzyme, we demonstrate an accelerated method for extracting the thermodynamic and kinetic constants describing the biocatalyst system. In addition, we demonstrate that the predicted biocatalyst behavior at different temperatures and reaction times is consistent with the experimental observations.

## Stabilization of enzyme catalysts



$$ttn = \frac{k_{cat}}{Kk_u}$$

