

热动力学的滴定量热法研究2: 单底物酶促反应的热动力学

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摘要 用滴定量热法分别建立了滴定期和停滴反应期单底物酶促反应热动力学的数学模型。根据这两种模型,可由一次实验的滴定量热曲线同时解析出单底物酶促反应的热力学参数(Δ_rH_m)和动力学参数(K_m 和 k_2)。用滴定量热法研究了一个经典的单底物酶促反应---过氧化氢酶催化分解过氧化氢反应的热动力学,由滴定期和停滴反应期热动力学模型解析出在298.15K和pH=7.0时该反应的米氏常数 K_m 分别为 $(5.41\pm 0.24)\times 10^{-3}$ 和 $(5.43\pm 0.21)\times 10^{-3}\text{mol}\cdot\text{L}^{-1}$,酶转换数 k_2 分别为 $(3.58\pm 0.33)\times 10^3$ 和 $(3.60\pm 0.41)\times 10^3\text{s}^{-1}$,摩尔反应焓为 $(-86.75\pm 0.88)\text{kJ}\cdot\text{mol}^{-1}$,实验结果验证了上述热动力学模型的正确性。

关键词 [热动力学](#) [量热法](#) [酶促反应](#) [米氏常数](#) [过氧化氢酶](#) [过氧化氢](#)

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Titration calorimetry applied to the study of thermokinetics 2: Thermokinetics of single-substrate enzyme-catalyzed reactions

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Abstract Titration calorimetry is emerging as an important tool for characterizing interactions of biological macromolecules by virtue of its general applicability, high accuracy and precision. In this paper, two mathematical models for thermokinetics of a single-substrate enzyme-catalyzed reaction in titration period and in the stopped- titration reaction period, respectively, have been developed, by using titration calorimetry. On the basis of the titration calorimetric curve, one can use these two models to calculate not only the thermodynamic data (Δ_rH_m) but also the kinetic data (K_m and k_2) for the reaction. Thermokinetics of a well-studied single-substrate enzymatic reaction, the catalase-catalyzed decomposition of hydrogen peroxide, was thus investigated by titration calorimetry, and the molar enthalpy (Δ_rH_m) was found to be $(-86.75\pm 0.88)\text{kJ}\cdot\text{mol}^{-1}$. The Michaelis constant (K_m) for H_2O_2 and the turn-over number of the enzyme (k_2) were determined by the titration-period thermokinetic model to be $(5.41\pm 0.24)\times 10^{-3}\text{mol}\cdot\text{L}^{-1}$ and $(3.58\pm 0.33)\times 10^3\text{s}^{-1}$, respectively, whereas the corresponding kinetic parameters calculated by the stopped-titration-reaction-period thermokinetic model were $(5.43\pm 0.21)\times 10^{-3}\text{mol}\cdot\text{L}^{-1}$ and $(3.60\pm 0.41)\times 10^3\text{s}^{-1}$, respectively, at 298.15K and pH7.0. Reliability of the above thermokinetic models was verified by the experimental data.

Key words [THERMODYNAMICS](#) [CALORIMETRY](#) [MICHAELIS CONSTANT](#) [CATALASE](#) [HYDROGEN PEROXIDE](#)

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