Abstract

Understanding Hysteresis in Human UDP-Glucose Dehydrogenase

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Human UDP-glucose dehydrogenase (hUGDH) catalyzes the NAD^+-dependent production of UDP-glucuronic acid, an essential substrate in drug metabolism. Some cancers have been shown to co-opt glucuronidation as an effective drug resistance mechanism. Thus, controlling hUGDH activity is a promising strategy for sensitizing some drug resistant tumors to existing chemotherapeutics. The progress curves of the hUGDH reaction reveal a significant lag in enzyme activity. Such hysteresis occurs when an enzyme undergoes a slow transition from an inactive (E*) to an active (E) conformation when the reaction is initiated. Understanding the nature of the E* state may illuminate new strategies for regulating hUGDH. The E* state in hysteretic enzymes can be caused by: i) the substrate-induced association of inactive subunits to form an active complex; ii) the dissociation of an inactive aggregate; or iii) the substrate-induced isomerization of an enzyme from an inactive state. Here we use transient state analysis, sedimentation velocity and a new crystal structure to characterize the E* state of hUGDH. Combined with steady state analysis and binding studies, we show that the hysteretic transition results produces an asymmetric complex that displays negative cooperativity. This work identifies the E* state as a possible target to controlling the activity of hUGDH. For more information, see the accompanying poster presented by Mr. Nathaniel R. Beattie that tests this model (abstract no.171).