Investigation of Ionizable Groups that Participate in D-Arginine Dehydrogenase Catalysis and Binding

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Georgia State Undergraduate Research Conference, Atlanta, GA, March 13, 2013

D-Arginine Dehydrogenase (DADH) has been isolated from *Pseudomonas aeruginosa,* a human pathogenic bacteria linked to nosocomial infections in hospitalized patients such as pneumonia, urinary tract infections (UTIs), and bacteremia.[1] DADH catalyzes the deamination of D-arginine in the first stage of the two-enzyme-coupled racemization of D-arginine to L-arginine.[2] The enzyme is active with a number of D-amino acids, displaying the highest activity with residues bearing a positively charged side chain.[3]

Previous studies have established the steady-state kinetic parameters for numerous D-amino acids at pH 8.7, but only for D-leucine a study on the change in the kinetic parameters as a function of pH was carried out.[3] In the present study, the effect of pH on the steady state kinetics has been examined with D-arginine and D-lysine as substrates to determine the contribution of ionizable groups in the reaction mechanism with each substrate. Kinetic rate constants have been derived to classify which steps are relevant to binding and/or catalysis. In addition, L-Arginine, the optical isomer of D-Arginine, has been tested as an inhibitor across a wide pH range to determine an intrinsic pK_a value. Comparison of these pK_a values will offer insight on the nature of how each substrate binds and interacts with the enzyme. Examination of the previously published X-ray crystallographic structure[4], along with the kinetic rate constant equations will lead to the assignment of these pK_a values to ionizable groups on the substrate and/or residues of the enzyme.

References:

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This study was supported in part by NSF MCB-1121695 (G.G.) and a GSU Molecular Basis for Disease Summer Fellowship (J.B.).