Notes

s8a.m1.o5 **Crystal Structure and Enzyme Mechanism of ?^{5}-3 Ketosteroid Isomerase**. B.-H. Oh, H.-S. Cho, G. Choi, K. Y. Choi, *Department of Life Science, Division of Molecular and Life Science, Pohang University of Science and Technology, Pohang, Kyungbuk, 790-784, Korea*
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?^{5}-3-ketosteroid isomerase has been intensively studied as a prototype with which to build a model of enzyme-catalyzed allylic isomerization. It catalyzes the isomerization of ?^{5}- to ?^{4}-3-ketosteroid at a diffusion controlled reaction rate. Asp38 (p*K*a ~4.7) has been identified as the general base abstracting the steroid C4? -proton (p*K*a ~12.7) to form a dienolate intermediate. A key issue regarding catalysis of this proton abstraction is the question of how the energy required for the unfavorable proton transfer can be provided at the active site of the enzyme, and/or how the thermodynamic barrier can be drastically reduced. The crystal structure and the enzyme in complex with an reaction intermediate analogue and proton nuclear magnetic resonance analyses of several mutant enzymes indicate that the Tyr14 OH forms a low-barrier hydrogen bond with the dienolic oxyanion of the intermediate while the Asp99 COOH forms a normal hydrogen bond with the same atom The catalytic power of the enzyme is likely to originate from a perturbation of the proton affinities of Asp38 and the steroid C4-proton at the active site owing to Tyr14 and Asp99 that are predisposed to form hydrogen bonds to the steroid oxyanion. [Supported by the Brain Korea 21 Project]