

Cloning, Expression, and Characterization of a Novel Putative Aldehyde Dehydrogenase from *Escherichia coli* K-12 Which is Highly Active on 3-Hydroxypropionaldehyde

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Our interest in 3-hydroxypropionic acid (3-HP) production from glycerol, which involves the oxidation of 3-hydroxypropionaldehyde (3-HPA) to 3-HP. Among the ALDHs tested in our laboratory, the putative aldehyde dehydrogenase (AldH) from *Escherichia coli* K-12 exhibited the highest activity on 3-HPA. To assess and characterize its specific function on 3-HPA, the gene *aldH* has been cloned in *E. coli* BL21 under the strong T5 promoter with pQE80L vector. Upon characterizing the purified AldH protein, we observed that this enzyme is highly specific to NAD⁺ over NADP⁺ as hydrogen acceptors. The enzyme activity was significantly high (38.1 U mg⁻¹ protein) with 3-hydroxypropionaldehyde (3-HPA) in pH 8.0 at 37 °C. The low activation energy (E_a) of 20.4 kJ mol⁻¹ for its oxidation of 3-HPA is advantageous in use of this enzyme for commercial applications. The kinetic properties of AldH on 3-HPA and 3-HP indicated that it mostly involved in oxidation reactions and the ratio (%) of oxidation/reduction is 96:4. The enzyme AldH can effectively be utilized for the biological production of 3-HP.