



Additional Figure 3. The stability of wild-type and V108A L-iLDH as function of temperature and pH. (A) The effect of temperature on the enzyme stability. The enzyme was incubated at different temperature ranging from 16°C to 58°C for 0.5 h and then assayed. The enzyme activity without treatment (store at 4°C) was defined as 100%. ■, wild-type l-iLDH; ●, V108A l-iLDH. (B) The effect of pH on the enzyme stability. The enzyme was incubated at different pH ranging from 3.0 to 11.0 for 2 h and then assayed. The buffers were: 0.2 M Na₂HPO₄-0.1 M citric acid buffer for pH 3.0-8.0; 50 mM Glycine-NaOH buffer for pH 8.0-12.0. The enzyme activities of wild-type and V108A L-iLDH without pH treatment (stored in 100 mM sodium phosphate buffer, pH 8.0) was defined as 100% severally. MTT was used at a concentration of 0.2 mM instead of 0.0625 mM DCIP as electron acceptor in the assay of pH stability, for the molar extinction coefficient of DCIP changes with different pH. ■, wild-type l-iLDH; ●, V108A l-iLDH. Values are the mean ± SD of 3 separate determinations.