

**Fig. S6. Determination of Michaelis-Menten kinetic parameters for** *Sa***M1PDH oxidoreductase activity.** (**A**) Determination of  $K_m$  (substrate concentration that yields a half-maximal velocity) and  $V_{max}$  (maximal velocity) values for *Sa*M1PDH mannitol-1-phosphate (M1P) oxidase activity. (**B**) Determination of  $K_m$  and  $V_{max}$  values for *Sa*M1PDH fructose-6-phosphate (F6P) reductase activity. In a typical reaction, purified *Sa*M1PDH was incubated with cofactor (M1P oxidation, NAD<sup>+</sup>; F6P reduction, NADH) and various concentrations of substrate (oxidation reaction, 0–250 µM M1P; reduction reaction, 0–450 µM F6P). At each substrate concentration, initial enzymatic velocity was determined after 1min reaction at 30 °C by measuring change in absorbance of NADH at 340 nm in reaction with substrate compared to that in control reaction without substrate and expressed as absorbance change per second (A/sec). Graphs were constructed by plotting initial enzymatic velocities against substrate concentrations of the means. Solid lines represent the best-fit curves determined by nonlinear regression analyses using GraphPad Prism 7 for Mac (http://www.graphpad.com) to calculate  $K_m$  and  $V_{max}$  values.