

SUPPLEMENTARY MATERIAL

Highly stable cold-active aldehyde dehydrogenase from the marine Antarctic *Flavobacterium* sp. PL002

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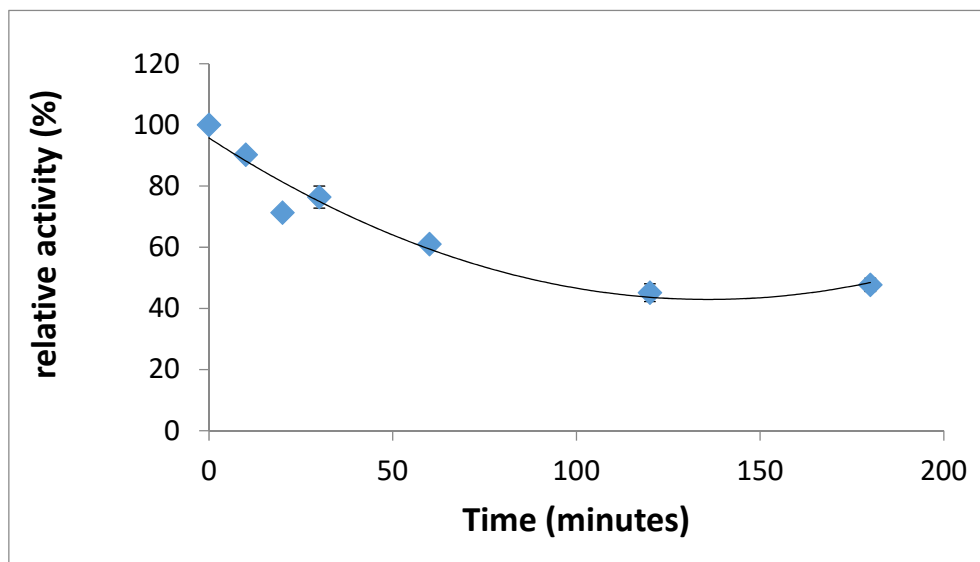


Figure S1. Thermal stability of F-ALDH at 50°C. The residual activity of the NAD⁺-dependent reaction was measured at 25°C as described in Methods after enzyme incubation at 50°C for various times up to 3 hours. The relative activity was calculated considering 100% the activity measured in the absence of high temperature incubation.

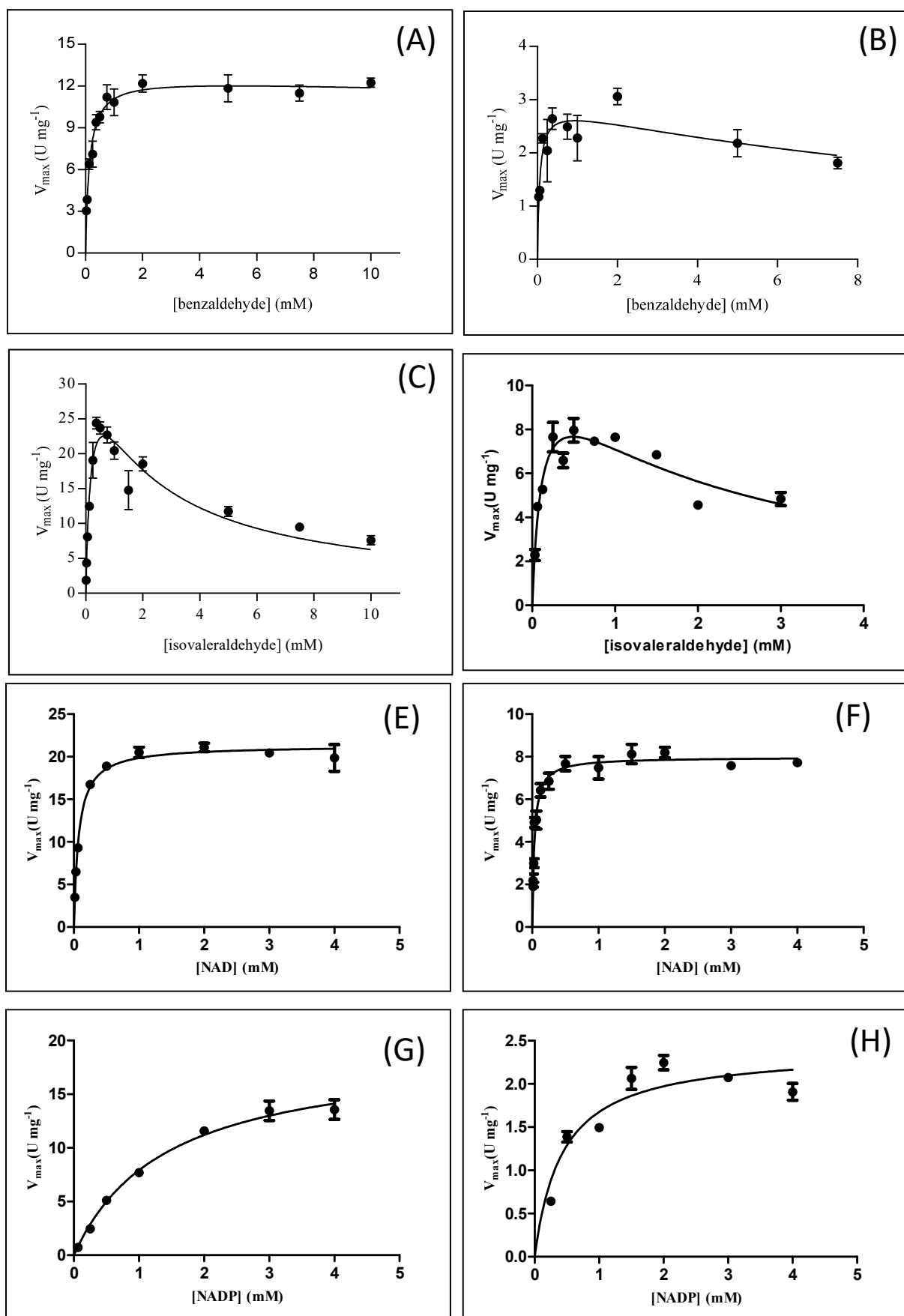


Figure S2. Substrate saturation curves of F-ALDH. The ALDH activity was measured at 30°C in the presence of increasing concentrations of (A) isovaleraldehyde and 1 mM NAD⁺; (B) isovaleraldehyde and 1 mM NAD⁺; (C) benzaldehyde and 1 mM NAD⁺; (D) benzaldehyde and 1 mM NADP⁺, as described in Methods.

Table S1. Catalytic parameters of bacterial ALDHs

Organism / Enzyme	Substrate / Cofactor	K_M (mM)	k_{cat} (s ⁻¹)	k_{cat} / K_M (s ⁻¹ mM ⁻¹)	Reference
<i>Flavobacterium</i> PL002 / F-ALDH	Isovaleraldehyde / NAD ⁺	0.197	100.04	507.6	Current study
	Isovaleraldehyde / NADP ⁺	0.070	34.11	487.2	
	Benzaldehyde / NAD ⁺	0.136	34.74	255.4	
	Benzaldehyde / NADP ⁺	0.057	10.72	188	
<i>Flavobacterium frigidimaris</i> / AldDH	Benzaldehyde / NAD ⁺	82.8	-	-	Yamanaka, 2002
<i>Pseudomonas putida</i> / BADH*	Benzaldehyde / NAD ⁺	63.4	137	2160	McLeish
	Benzaldehyde / NADP ⁺	39.9	42.5	1,065.16	
<i>Klebsiella pneumoniae</i> / ALDH	Isovaleraldehyde / NAD ⁺	0.21	25.47	121.9	Raj
<i>Escherichia coli</i> K-12 / AldDH	Isovaleraldehyde / NAD ⁺	0.68	27.31	40.16	Mohan (Jo)
	Benzaldehyde / NADP ⁺	5.37	17.37	3.23	
<i>Homo sapiens</i> / ALDH1B1	Benzaldehyde / NAD ⁺	0.05	-	-	Stagos (20616185)
<i>Homo sapiens</i> / ALDH2	Benzaldehyde / NAD ⁺	0.000018	5.83	323,888.8	Klyosov
<i>Homo sapiens</i> / ALDH3A1	Benzaldehyde / NAD ⁺	0.16	-	-	Giebułtowiec
<i>Euglena gracilis</i> / ALDH	Benzaldehyde / NAD ⁺	0.15	-	-	Rodriguez – Zavala 2006
<i>Ratus norvegicus</i> / ALDH1	Benzaldehyde / NADP ⁺	1.6	-	-	Lindahl, 1984

*(BADH) benzaldehyde dehydrogenase