# Hyperthermophilic Carbonic Anhydrase: Application for $CO_2$ Capture System Development

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In order to employ carbonic anhydrases (CAs, potential biocatalysts for CO2 sequestration) under the non-natural process conditions, highly-thermostable CAs should be explored. Here, we expressed the codon-optimized sequence of PMCA, cloned from the thermophile Persephonella marina EX-H1 found in marine vents, and characterized its hyper-thermostable properties. Removal of the PMCA signal peptide (sp) resulted in the production of about 5 times more purified protein, PMCA(sp-), than from the intact gene in an E. coli expression system. PMCA(sp-) has a wide pH tolerance (optimum: pH 7.5), retained 80% of its activity after 15 min incubation at 100°C and almost 50% activity even after 2 h at 100°C. Apparent Km and Vmax for the p-nitrophenylacetate were 5.90 mM and 0.020 μmol min-1. Various metal ions were examined to enhance or inhibit activity. Finally, we demonstrated that in the presence of Ca2+ , PMCA(sp-) readily catalyzed the conversion of CO2 to CaCO3 (calcite form).