

Recombinant *Pyrococcus furiosus* NADP-specific NiFe-type cytoplasmic hydrogenase (4 subunits), 9×His tagged

Cat. No. SHI-61P Lot. No. (See product label)

SPECIFICATION

Product Overview	This recombinant <i>Pyrococcus furiosus</i> NADP-specific NiFe-type cytoplasmic hydrogenase (SHI) is from the hyperthermophilic <i>P. furiosus</i> microorganism that grows optimally at 100 centigrade. The enzyme is active over a broad temperature range, from ambient to 100 centigrade, and is oxygen-sensitive, with a half-life in air of approximately 25 hours at room temperature.
Species	<i>Pyrococcus furiosus</i>
Source	<i>Pyrococcus furiosus</i>
Description	This is an extremely thermostable, but oxygen sensitive enzyme. The [NiFe] hydrogenases contain a minimum of two subunits known as the small (S) and large (L) subunits. The small subunit contains three iron-sulfur clusters while the large subunit contains the active site, a nickel-iron center which is connected to the solvent by a molecular tunnel. To date, periplasmic, cytoplasmic, and membrane-bound hydrogenases have been found. [NiFe] hydrogenases are known to be deactivated by molecular oxygen (O ₂). The [NiFe] hydrogenase of <i>Pyrococcus furiosus</i> is heterotetrameric wherein the additional two subunits allow the enzyme to use NAD(P)(H) as an electron carrier.
Bio-activity	One unit (U) is 1 μmole of H ₂ evolved min ⁻¹ mg ⁻¹ using MV-linked assay.
Molecular Mass	Predicted: 155 kDa, Size Exclusion: 149 kDa +/- 5 kDa

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Purity	> 90%
Unit Definition	1 μ mole of H ₂ evolved min ⁻¹ mg ⁻¹ using MV-linked assay.
Storage	This item is oxygen sensitive. Stable when stored sealed in strictly anaerobic environment (<10 ppm O ₂) at room temperature for up to 6 months. For long-term storage, protein can be flash frozen in nitrogen and stored at -80 centigrade.
Concentration	1 mg/mL (> 100 U/mL)
Storage Buffer	50 mM Tris, 2 mM DT, 300 mM NaCl, pH 8.2
Shipping	Dry ice
References	1. Ma, K. and Adams, M. W. W. (2001) Hydrogenases I and II from <i>Pyrococcus furiosus</i> Meths. Enzymol. 331, 208-216 2. Chandrayan, S. K., McTernan, P. M., Hopkins, R. C., Sun, J., Jenney, F. E., Jr. & Adams, M. W. (2012) Engineering hyperthermophilic archaeon <i>Pyrococcus furiosus</i> to overproduce its cytoplasmic [NiFe]-hydrogenase J. Biol. Chem. 287, 3257-64 3. Hopkins, R.C., Sun, J., Jenney, F.E., Chandrayan, S.K., McTernan, P.M., and Adams, M.W.W., (2011) "Homologous expression of a subcomplex of <i>Pyrococcus furiosus</i> hydrogenase that interacts with pyruvate ferredoxin oxireductase" PLoS ONE 6(1), e26569 4. Patent Pending - University of Georgia research Foundation, Inc. (WO 2009075798)

GENE INFORMATION

Official Symbol	SHI
Synonyms	NADP-specific NiFe-type cytoplasmic hydrogenase; SHI; NADPH-dependent

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hydrogenase/sulfhydrogenase 1 subunit alpha; NADPH-dependent
hydrogenase/sulfhydrogenase 1 subunit beta; NADPH-dependent
hydrogenase/sulfhydrogenase 1 subunit gamma; NADPH-dependent
hydrogenase/sulfhydrogenase 1 subunit delta; Cytoplasmic [NiFe]-Hydrogenase (4 subunits; OE-SHI)

WB

The purified enzyme was analyzed by conventional SDS-PAGE except that the protein was incubated with the SDS-loading buffer for 10 min (lane 1) or for 60 min (lane 3) prior to electrophoresis. Native SHI (treated for 10 min) is shown in lane 2. The arrow indicates the high molecular weight catalytically active protein band seen in lanes 1 and 2. The center lane (M) contains the protein molecular weight ladder (Invitrogen) with corresponding masses as indicated in kDa.

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