

AHSP Protein, Human, Recombinant

General Information

Synonyms:	ERAF;alpha hemoglobin stabilizing protein;α hemoglobin stabilizing protein;EDRF
Protein Construction:	A DNA sequence encoding human ERAF (Q9NZD4) (Met1-Ser102) was expressed. Predicted N terminal: Met
Species:	Human
Expression Host:	E. coli
Accession:	Q9NZD4
Molecular Weight:	11.8 kDa (predicted); 12 kDa (reducing conditions)

QC Testing

Biological Activity:	Activity testing is in progress. It is theoretically active, but we cannot guarantee it. If you require protein activity, we recommend choosing the eukaryotic expression version first.
Purity:	> 90 % as determined by SDS-PAGE
Endotoxin:	Please contact us for more information.
Formulation:	Lyophilized from a solution filtered through a 0.22 μm filter, containing PBS, pH 7.4. Typically, a mixture containing 5% to 8% trehalose, mannitol, and 0.01% Tween 80 is incorporated as a protective agent before lyophilization.

Preparation and Storage

Reconstitution:
A Certificate of Analysis (CoA) containing reconstitution instructions is included with the products. Please refer to the CoA for detailed information.

Stability & Storage:
It is recommended to store recombinant proteins at -20°C to -80°C for future use. Lyophilized powders can be stably stored for over 12 months, while liquid products can be stored for 6-12 months at -80°C. For reconstituted protein solutions, the solution can be stored at -20°C to -80°C for at least 3 months. Please avoid multiple freeze-thaw cycles and store products in aliquots.

Actual storage temperature shall be subject to the COA.

Shipping:
In general, lyophilized powders are shipped with blue ice, while solutions are shipped with dry ice.

Protein Background

AHSP, also known as ERAF, is a conserved mammalian erythroid protein which belongs to the AHSP family. It is expressed in blood and bone marrow. AHSP facilitates the production of Hemoglobin A by stabilizing free α-globin. It rapidly binds to ferrous α with association ($k'(AHSP)$) and dissociation ($k(AHSP)$) rate constants of $\approx 1 \mu\text{m}^{-1} \text{s}^{-1}$ and $.2 \text{s}^{-1}$, respectively, at pH 7.4 at 22 °C. A small slow phase was observed when AHSP binds to excess ferrous αCO. This slow phase appears to be due to cis to trans prolyl isomerization of the Asp(29)-Pro(3) peptide

bond in wild-type AHSP because it was absent when α CO was mixed with P3A and P3W AHSP, which are fixed in the trans conformation. This slow phase was also absent when met(Fe(3+))- α reacted with wild-type AHSP, suggesting that met- α is capable of rapidly binding to either Pro(3) conformer. Both wild-type and Pro(3)-substituted AHSPs drive the formation of a met- α hemichrome conformation following binding to either met- or oxy(Fe(2+))- α . The dissociation rate of the met- α ·AHSP complex ($k(\text{AHSP}) \approx .2 \text{ s}^{-1}$) is ~ 1 -fold slower than that for ferrous α ·AHSP complexes, resulting in a much higher affinity of AHSP for met- α . Thus, in vivo, AHSP acts as a molecular chaperone by rapidly binding and stabilizing met- α hemichrome folding intermediates. The low rate of met- α dissociation also allows AHSP to have a quality control function by kinetically trapping ferric α and preventing its incorporation into less stable mixed valence Hemoglobin A tetramers. Reduction of AHSP-bound met- α allows more rapid release to β subunits to form stable fully, reduced hemoglobin dimers and tetramers.

Reference

Miele G., et al., (2001), A novel erythroid-specific marker of transmissible spongiform encephalopathies. *Nat. Med.* 7:361-364.

Zhang Q.-H., et al., (2000), Cloning and functional analysis of cDNAs with open reading frames for 300 previously undefined genes expressed in CD34+ hematopoietic stem/progenitor cells. *Genome Res.* 10:1546-1560.

Kihm A.J., et al., (2002), An abundant erythroid protein that stabilizes free alpha-haemoglobin. *Nature* 417:758-763.

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