

Hemoglobin ϵ siRNA (r): sc-270013

BACKGROUND

Hemoglobin (Hgb) is coupled to four iron-binding, methene-linked tetrapyrrole rings (heme). The α and β globin loci determine the basic Hemoglobin structure. The globin portion of Hemoglobin consists of two α chains and two β chains arranged in pairs forming a tetramer. Each of the four globin chains covalently associates with a heme group. The bonds between α and β chains are weaker than between similar globin chains, thereby forming a cleavage plane that is important for oxygen binding and release. High affinity for oxygen occurs upon relaxation of the $\alpha 1$ - $\beta 2$ cleavage plane. When the two $\alpha 1$ - $\beta 2$ interfaces are closely bound, Hemoglobin has a low affinity for oxygen. Hemoglobin A, which contains two α chains plus two β chains, comprises 97% of total circulating Hemoglobin. The remaining 3% of total circulating Hemoglobin is comprised of Hemoglobin A-2, which consists of two α chains plus two δ chains, and fetal Hemoglobin (Hb F), which consists of two α chains together with two γ chains. Hemoglobin ϵ is a 147 amino acid β -type Hemoglobin chain that exists as a tetrameric complex with two Hemoglobin α chains and is a component of early embryonic Hemoglobin.

REFERENCES

1. Liebhaber, S.A., Goossens, M. and Kan, Y.W. 1981. Homology and concerted evolution at the $\alpha 1$ and $\alpha 2$ loci of human α -globin. *Nature* 290: 26-29.
2. Goodbourn, S.E., Higgs, D.R., Clegg, J.B. and Weatherall, D.J. 1983. Molecular basis of length polymorphism in the human ζ -globin gene complex. *Proc. Natl. Acad. Sci. USA* 80: 5022-5026.
3. Giardina, B., Messana, I., Scatena, R. and Castagnola, M. 1995. The multiple functions of hemoglobin. *Crit. Rev. Biochem. Mol. Biol.* 30: 165-196.
4. Adachi, K., Zhao, Y. and Surrey, S. 2002. Assembly of human hemoglobin (Hb) β - and γ -globin chains expressed in a cell-free system with α -globin chains to form Hb A and Hb F. *J. Biol. Chem.* 277: 13415-13420.
5. Sudha, R., Anantharaman, L., Sivaram, M.V., Mirsamadi, N., Choudhury, D., Lohiya, N.K., Gupta, R.B. and Roy, R.P. 2004. Linkage of interactions in sickle hemoglobin fiber assembly: inhibitory effect emanating from mutations in the AB region of the α chain is annulled by a mutation at its EF corner. *J. Biol. Chem.* 279: 20018-20027.
6. Feng, L., Gell, D.A., Zhou, S., Gu, L., Kong, Y., Li, J., Hu, M., Yan, N., Lee, C., Rich, A.M., Armstrong, R.S., Lay, P.A., Gow, A.J., Weiss, M.J., Mackay, J.P. and Shi, Y. 2004. Molecular mechanism of AHSP-mediated stabilization of α -hemoglobin. *Cell* 119: 629-640.
7. Baudin-Creux, V., Vasseur-Godbillon, C., Pato, C., Prehu, C., Wajcman, H. and Marden, M.C. 2004. Transfer of human α - to β -hemoglobin via its chaperone protein: evidence for a new state. *J. Biol. Chem.* 279: 36530-36533.

CHROMOSOMAL LOCATION

Genetic locus: Hbe1 (rat) mapping to 1q32.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

PRODUCT

Hemoglobin ϵ siRNA (r) is a target-specific 19-25 nt siRNA designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Hemoglobin ϵ shRNA Plasmid (r): sc-270013-SH and Hemoglobin ϵ shRNA (r) Lentiviral Particles: sc-270013-V as alternate gene silencing products.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Hemoglobin ϵ siRNA (r) is recommended for the inhibition of Hemoglobin ϵ expression in rat cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Hemoglobin ϵ gene expression knockdown using RT-PCR Primer: Hemoglobin ϵ (r)-PR: sc-270013-PR (20 μ l). Annealing temperature for the primers should be 55 - 60° C and the extension temperature should be 68 - 72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.