

TH-POK siRNA (m): sc-76650

BACKGROUND

TH-POK (T-helper-inducing POZ/Krüppel-like factor), also known as zinc-finger protein 67 (ZFP67), zinc finger and BTB domain-containing protein 7B or Krüppel-related zinc-finger protein cKrox, is a 539 amino acid protein that contains one BTB (POZ) domain and four C₂H₂-type zinc fingers. Localized to the nucleus, TH-POK functions primarily as a key regulator of lineage commitment of immature T cell precursors. Specifically, the presence of TH-POK directs positively selected thymocytes to the CD4 lineage, whereas its absence causes default development to the CD8 lineage. TH-POK also functions as a transcriptional repressor of various other genes, such as COL1A1, COL1A2 and Fibronectin.

REFERENCES

1. He, X., et al. 2005. The zinc-finger transcription factor TH-POK regulates CD4 versus CD8 T cell lineage commitment. *Nature* 433: 826-833.
2. He, X., et al. 2006. CD4/CD8 lineage commitment: light at the end of the tunnel? *Curr. Opin. Immunol.* 18: 135-142.
3. Kappes, D.J., et al. 2006. Role of the transcription factor TH-POK in CD4:CD8 lineage commitment. *Immunol. Rev.* 209: 237-252.
4. Kimura, H., et al. 2006. Role of DNA methylation for expression of novel stem cell marker CDCP1 in hematopoietic cells. *Leukemia* 20: 1551-1556.
5. He, X., et al. 2008. CD4-CD8 lineage commitment is regulated by a silencer element at the TH-POK transcription-factor locus. *Immunity* 28: 346-358.
6. Bell, J.J., et al. 2008. Putting TH-POK in place. *Nat. Immunol.* 9: 1095-1097.
7. Wang, L., et al. 2008. Distinct functions for the transcription factors GATA-3 and TH-POK during intrathymic differentiation of CD4⁺ T cells. *Nat. Immunol.* 9: 1122-1130.

CHROMOSOMAL LOCATION

Genetic locus: Zbtb7b (mouse) mapping to 3 F1.

PRODUCT

TH-POK siRNA (m) is a pool of 3 target-specific 19-25 nt siRNAs 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 TH-POK shRNA Plasmid (m): sc-76650-SH and TH-POK shRNA (m) Lentiviral Particles: sc-76650-V as alternate gene silencing products.

For independent verification of TH-POK (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-76650A, sc-76650B and sc-76650C.

RESEARCH USE

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

PROTOCOLS

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

TH-POK siRNA (m) is recommended for the inhibition of TH-POK expression in mouse 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.

GENE EXPRESSION MONITORING

TH-POK (A-4): sc-376250 is recommended as a control antibody for monitoring of TH-POK gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor TH-POK gene expression knockdown using RT-PCR Primer: TH-POK (m)-PR: sc-76650-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.