

# NPAS4 siRNA (h): sc-96790

## BACKGROUND

The Per-Arnt-Sim (PAS) domain is a 270 amino acid motif that mediates associations among various PAS family transcription factors. The PAS family contains neuronal specific transcription factors known as NPAS1, NPAS2, NPAS3 and NPAS4, which are involved in the development and maintenance of learning and memory pathways. NPAS1 regulates erythropoietin expression in developing brain. NPAS2, also designated PAS 4/MOP-4, associates with MOP-3 to activate transcription. NPAS3, which localizes to the nucleus and is ubiquitously expressed in the adult brain, may be involved in neurogenesis and has been implied to control regulatory pathways relevant to psychotic illness and schizophrenia. NPAS4 (neuronal PAS domain protein 4), also known as NXF, Le-PAS, PASD10 or bHLHe79, is a 802 amino acid nuclear protein that is exclusively expressed in the brain. NPAS4 acts as a transcriptional activator in the presence of Arnt 1, and activates the CME (CNS midline enhancer) element and expression of the Drebrin gene.

## REFERENCES

1. Ooe, N., et al. 2004. Identification of a novel basic helix-loop-helix-PAS factor, NXF, reveals a SIM2 competitive, positive regulatory role in dendritic-cytoskeleton modulator Drebrin gene expression. *Mol. Cell. Biol.* 24: 608-616.
2. Shamloo, M., et al. 2006. NPAS4, a novel helix-loop-helix PAS domain protein, is regulated in response to cerebral ischemia. *Eur. J. Neurosci.* 24: 2705-2720.
3. Ooe, N., et al. 2007. Characterization of *Drosophila* and *Caenorhabditis elegans* NXF-like-factors, putative homologs of mammalian NXF. *Gene* 400: 122-130.
4. Coba, M.P., et al. 2008. Kinase networks integrate profiles of N-methyl-D-aspartate receptor-mediated gene expression in hippocampus. *J. Biol. Chem.* 283: 34101-34107.
5. Ibi, D., et al. 2008. Social isolation rearing-induced impairment of the hippocampal neurogenesis is associated with deficits in spatial memory and emotion-related behaviors in juvenile mice. *J. Neurochem.* 105: 921-932.

## CHROMOSOMAL LOCATION

Genetic locus: NPAS4 (human) mapping to 11q13.1.

## PRODUCT

NPAS4 siRNA (h) 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see NPAS4 shRNA Plasmid (h): sc-96790-SH and NPAS4 shRNA (h) Lentiviral Particles: sc-96790-V as alternate gene silencing products.

For independent verification of NPAS4 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-96790A, sc-96790B and sc-96790C.

## RESEARCH USE

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

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

## APPLICATIONS

NPAS4 siRNA (h) is recommended for the inhibition of NPAS4 expression in human 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  $\mu$ M in 60  $\mu$ 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

NPAS4 (4E4): sc-293239 is recommended as a control antibody for monitoring of NPAS4 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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 $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 NPAS4 gene expression knockdown using RT-PCR Primer: NPAS4 (h)-PR: sc-96790-PR (20  $\mu$ l, 582 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.