

# CRF-BP siRNA (h): sc-39397

## BACKGROUND

Response to stress in mammals requires an intact hypothalamic-pituitary-adrenal axis. The proximal part of the response is mediated by secretion of corticotropin-releasing hormone (CRH) by the paraventricular nucleus of the hypothalamus. CRH is a 41 amino acid peptide derived by enzymatic cleavage from a 191 amino acid prohormone. CRH is produced not only in the hypothalamus but also in peripheral tissues, such as T lymphocytes; it is highly expressed in human placenta. Glucocorticoids stimulate placental CRH synthesis and secretion in primary cultures of human placenta. This stimulation is in contrast to the glucocorticoid suppression of CRH expression in hypothalamus. The gene which encodes CRH maps to human chromosome 8q13. Human plasma contains a CRH-binding protein, CRH-BP (also designated CRF-BP) which inactivates CRH and which may prevent inappropriate pituitary-adrenal stimulation in pregnancy. The gene which encodes CRF-BP maps to human chromosome 5q13.3.

## REFERENCES

1. Robinson, B.G., et al. 1988. Glucocorticoid stimulates expression of corticotropin-releasing hormone gene in human placenta. *Proc. Natl. Acad. Sci. USA* 85: 5244-5248.
2. Arbiser, J.L., et al. 1988. Human corticotropin releasing hormone gene is located on the long arm of chromosome 8. *Cytogenet. Cell Genet.* 47: 113-116.
3. Potter, E., et al. 1991. Cloning and characterization of the cDNAs for human and rat corticotropin releasing factor-binding proteins. *Nature* 349: 423-426.
4. Vamvakopoulos, N.C., et al. 1995. Mapping the human corticotropin releasing hormone binding protein gene (CRHBP) to the long arm of chromosome 5 (5q11.2-q13.3). *Genomics* 25: 325-327.
5. Behan, D.P., et al. 1995. Corticotropin releasing factor binding protein (CRF-BP) is expressed in neuronal and astrocytic cells. *Brain Res.* 698: 259-264.
6. Behan, D.P., et al. 1996. Urocortin interaction with corticotropin-releasing factor (CRF) binding protein (CRF-BP): a novel mechanism for elevating "free" CRF levels in human brain. *Brain Res.* 725: 263-267.

## CHROMOSOMAL LOCATION

Genetic locus: CRHBP (human) mapping to 5q13.3.

## PRODUCT

CRF-BP 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 CRF-BP shRNA Plasmid (h): sc-39397-SH and CRF-BP shRNA (h) Lentiviral Particles: sc-39397-V as alternate gene silencing products.

For independent verification of CRF-BP (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-39397A, sc-39397B and sc-39397C.

## 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

CRF-BP siRNA (h) is recommended for the inhibition of CRF-BP 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 66  $\mu$ 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

CRF-BP (C-8): sc-365975 is recommended as a control antibody for monitoring of CRF-BP 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 CRF-BP gene expression knockdown using RT-PCR Primer: CRF-BP (h)-PR: sc-39397-PR (20  $\mu$ 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.

## PROTOCOLS

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