

# reticulocalbin-2 siRNA (m): sc-62248

## BACKGROUND

The CREC family consists of multiple EF-hand, low-affinity  $\text{Ca}^{2+}$ -binding proteins localized to the secretory pathway of mammalian cells. A member of the CREC family, reticulocalbin-2 (Rcn2), also known as TCBP-49 (taipoxin-associated calcium-binding protein 49), is a 320 amino acid protein of the endoplasmic reticulum lumen. Containing 6 EF-hand domains, reticulocalbin-2 is the mouse homolog of human E6BP, a 320 amino acid protein that binds presynaptic snake venom toxin taipoxin and may interact with internalized taipoxin, possibly mediating its activation. E6BP is expressed in rat brain, cerebellum, heart, liver, kidney, skeletal muscle and testis cells. E6BP is not an abundant protein, which suggests that it is not a reticular calcium storage protein. E6BP is believed to share a general E6-binding motif with E6-AP, which binds cancer-related papillomaviral E6 proteins.

## REFERENCES

1. Dodds, D., et al. 1995. Novel reticular calcium binding protein is purified on taipoxin columns. *J. Neurochem.* 64: 2339-2344.
2. Be, X., et al. 2001. Solution structure determination and mutational analysis of the papillomavirus E6 interacting peptide of E6AP. *Biochemistry* 40: 1293-1299.
3. Sherman, L., et al. 2002. Inhibition of serum- and calcium-induced terminal differentiation of human keratinocytes by HPV 16 E6: study of the association with p53 degradation, inhibition of p53 transactivation, and binding to E6BP. *Virology* 292: 309-320.
4. Zanier, K., et al. 2005. Kinetic analysis of the interactions of human papillomavirus E6 oncoproteins with the ubiquitin ligase E6AP using surface plasmon resonance. *J. Mol. Biol.* 349: 401-412.
5. Griffin, H., et al. 2006. Inhibition of papillomavirus protein function in cervical cancer cells by intrabody targeting. *J. Mol. Biol.* 355: 360-378.
6. Lichtig, H., et al. 2006. HPV16 E6 natural variants exhibit different activities in functional assays relevant to the carcinogenic potential of E6. *Virology* 350: 216-227.

## CHROMOSOMAL LOCATION

Genetic locus: Rcn2 (mouse) mapping to 9 B.

## PRODUCT

reticulocalbin-2 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  $\mu\text{M}$  solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see reticulocalbin-2 shRNA Plasmid (m): sc-62248-SH and reticulocalbin-2 shRNA (m) Lentiviral Particles: sc-62248-V as alternate gene silencing products.

For independent verification of reticulocalbin-2 (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-62248A, sc-62248B and sc-62248C.

## STORAGE AND RESUSPENSION

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

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

## APPLICATIONS

reticulocalbin-2 siRNA (m) is recommended for the inhibition of reticulocalbin-2 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  $\mu\text{M}$  in 66  $\mu\text{l}$ . 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

E6BP (30): sc-293069 is recommended as a control antibody for monitoring of reticulocalbin-2 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<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> 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<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor reticulocalbin-2 gene expression knockdown using RT-PCR Primer: reticulocalbin-2 (m)-PR: sc-62248-PR (20  $\mu\text{l}$ ). Annealing temperature for the primers should be  $55-60^{\circ}\text{C}$  and the extension temperature should be  $68-72^{\circ}\text{C}$ .

## 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.