

## Produktinformation



Forschungsprodukte & Biochemikalien



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Diagnostik & molekulare Diagnostik



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# caspase-4/5 p10 shRNA (h) Lentiviral Particles: sc-43668-V



The Power to Overtion

#### **BACKGROUND**

Caspases are cysteine proteases which play important roles in the activation of cytokines and in apoptosis. The ICE subfamily of caspases comprises peptides closely related to caspase-1, which promotes maturation of interleukin  $1\beta$  (IL-1 $\beta$ ) and interleukin-18 (IL-18) by proteolytic cleavage of precursor forms to generate biologically active peptides. Both caspase-4 and caspase-5 are members of the caspase-1 subfamily, and are more closely related to each other than to other homologs. Caspase-5 (also designated ICErel-III, TY, ICH-3 and caspase-12 in mouse), can cleave its own precursor, an activity that requires the Cysteine 245 residue. The human caspase-5 gene maps to chromosome 11q22.3 and encodes a protein whose expression is barely detectable in most tissues except brain. Caspase-4 cleaves caspase-1 and has two subunits derived from the precursor sequence by an autocatalytic mechanism or by cleavage by caspase-8. Caspase-4 is widely expressed, with highest levels in spleen and lung, and is not found in the brain.

#### **REFERENCES**

- Lindahl, T., et al. 1995. Post-translational modification of poly (ADP-ribose) polymerase induced by DNA strand breaks. Trends Biochem. Sci. 20: 405-411.
- Duan, H., et al. 1996. ICE-LAP3, a novel mammalian homologue of the Caenorhabditis elegans cell death protein CED-3 is activated during FASand tumor necrosis factor-induced apoptosis. J. Biol. Chem. 271: 1621-1625.
- Fernandes-Alnemri, T.F., et al. 1996. *In vitro* activation of CPP32 and Mch3 by Mch4, a novel human apoptotic cysteine protease containing two FADDlike domains. Proc. Natl. Acad. Sci. USA 93: 7464-7469.
- Duan, H., et al. 1996. ICE-LAP6, a novel member of the ICE/CED-3 gene family, is activated by the cytotoxic T cell protease granzyme B. J. Biol. Chem. 271: 16720-16724.

#### CHROMOSOMAL LOCATION

Genetic locus: CASP4 (human) mapping to 11q22.3.

#### **PRODUCT**

caspase-4/5 p10 shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200  $\mu$ l frozen stock containing 1.0 x 10<sup>6</sup> lentiviral transducing particles per milliliter in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see caspase-4/5 p10 siRNA (h): sc-43668 and caspase-4/5 p10 shRNA Plasmid (h): sc-43668-SH as alternate gene silencing products.

#### **STORAGE**

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

#### **APPLICATIONS**

caspase-4/5 p10 shRNA (h) Lentiviral Particles is recommended for the inhibition of caspase-5 expression in human cells.

#### **SUPPORT REAGENTS**

Control shRNA Lentiviral Particles: sc-108080. Available as 200  $\mu$ l frozen viral stock containing 1.0 x 10<sup>6</sup> lentiviral transducing particles per milliliter; contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

#### **GENE EXPRESSION MONITORING**

caspase-4/5 p10 (H-60): sc-28229 is recommended as a control antibody for monitoring of caspase-5 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

#### **RT-PCR REAGENTS**

Semi-quantitative RT-PCR may be performed to monitor caspase-5 gene expression knockdown using RT-PCR Primer: caspase-4/5 p10 (h)-PR: sc-43668-PR (20  $\mu$ I, 433 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

#### **BIOSAFETY**

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

#### **RESEARCH USE**

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

#### **PROTOCOLS**

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