SANTA CRUZ BIOTECHNOLOGY, INC.

VACVase siRNA (h): sc-106687

BACKGROUND

Vase, also known as valacyclovir hydrolase, BPHL (biphenyl hydrolase-like) or MCNAA, is a 291 amino acid member of the AB hydrolase superfamily. Highly expressed in liver and kidney and weakly expressed in heart, intestine and skeletal muscle, VACVase is a serine hydrolase that functions to catalyze the hydrolytic activation of amino acid ester prodrugs and may play a role in chemical detoxification. VACVase exists as a monomer and contains a serine residue at its active site, allowing it to enzymatically hydrolyze and activate compounds such as valacyclovir (VACV), an antitherapeutic drug. VACVase is expressed in several carcinoma cell lines and, due to its enzymatic specificity, may be a potential activation target for anticancer and antiviral prodrugs. VACVase exists as two alternatively spliced isoforms designated α and β.

REFERENCES

1. Puente, X.S. and López-Otín, C. 1995. Cloning and expression analysis of a novel human serine hydrolase with sequence similarity to prokaryotic enzymes involved in the degradation of aromatic compounds. J. Biol. Chem. 270: 12926-12932.
2. Puente, X.S., Pendás, A.M. and López-otín, C. 1998. Structural characterization and chromosomal localization of the gene encoding human biphenyl hydrolase-related protein (BPHL). Genomics 51: 459-462.
3. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 603156. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
4. Kim, I., Chu, X.Y., Kim, S., Provoda, C.J., Lee, K.D. and Amidon, G.L. 2003. Identification of a human valacyclovirase: biphenyl hydrolase-like protein as valacyclovir hydrolase. J. Biol. Chem. 278: 25348-25356.
5. Kim, I., Song, X., Vig, B.S., Mittal, S., Shin, H.C., Lorenzi, P.J. and Amidon, G.L. 2004. A novel nucleoside prodrug-activating enzyme: substrate specificity of biphenyl hydrolase-like protein. Mol. Pharm. 1: 117-127.
6. Kim, I., Crippen, G.M. and Amidon, G.L. 2004. Structure and specificity of a human valacyclovir activating enzyme: a homology model of BPHL. Mol. Pharm. 1: 434-446.

CHROMOSOMAL LOCATION

Genetic locus: BPHL (human) mapping to 6p25.2.

PRODUCT

VACVase 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see VACVase shRNA Plasmid (h): sc-106687-SH and VACVase shRNA (h) Lentiviral Particles: sc-106687-V as alternate gene silencing products.

For independent verification of VACVase (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-106687A, sc-106687B and sc-106687C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

VACVase siRNA (h) is recommended for the inhibition of VACVase 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 µM in 66 µl. 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

VACVase (E-3): sc-514189 is recommended as a control antibody for monitoring of VACVase 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 VACVase gene expression knockdown using RT-PCR Primer: VACVase (h)-PR: sc-106687-PR (20 µl). 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 for detailed protocols and support products.