SANTA CRUZ BIOTECHNOLOGY, INC.

ECP siRNA (h): sc-105319

BACKGROUND

ECP (eosinophil cationic protein), also known as RNS3 or RNASE3 (ribonuclease, RNase A family, 3), is a 160 amino acid potent cytotoxic secretory protein that belongs to the pancreatic ribonuclease family. ECP localizes to the granule matrix of the eosinophil and is involved in the immune response system. ECP is a cytotoxin, neurotoxin and helminthotoxin that is secreted by activated human eosinophils. ECP exhibits anti-parasitic, antibacterial and ribonucleolytic activities. It has been suggested that ECP induces the neurotoxic effect known as the Gordon phenomenon, a syndrome manifested by ataxia, muscular rigidity, paralysis and tremor that may lead to death. ECP is considered a marker of eosinophilic inflammation and high levels have been found in cases of active asthma and other allergic diseases.

REFERENCES

1. Fredens, K., et al. 1982. The Gordon phenomenon induced by the eosinophil cationic protein and eosinophil protein X. J. Allergy Clin. Immunol. 70: 361-366.
2. Fredens, K., et al. 1985. Eosinophils and cellular injury: the Gordon phenomenon as a model. N. Engl. Reg. Allergy Proc. 6: 346-351.
3. Newton, D.L., et al. 1994. Toxicity of an anti-tumor ribonuclease to Purkinje neurons. J. Neurosci. 14: 538-544.
4. Rosenberg, H.F. 1995. Recombinant human eosinophil cationic protein. Ribonuclease activity is not essential for cytotoxicity. J. Biol. Chem. 270: 7876-7881.
5. Munthe-Kaas, M.C., et al. 2007. Eosinophil cationic protein (ECP) polymorphisms and association with asthma, s-ECP levels and related phenotypes. Allergy 62: 429-436.
6. Koh, G.C., et al. 2007. Eosinophil cationic protein: is it useful in asthma? A systematic review. Respir. Med. 101: 696-705.
7. Torrent, M., et al. 2008. Eosinophil cationic protein high-affinity binding to bacteria-wall lipopoly-saccharides and peptidoglycans. Biochemistry 47: 3544-3555.

CHROMOSOMAL LOCATION

Genetic locus: RNASE3 (human) mapping to 14q11.2.

PRODUCT

ECP 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 ECP shRNA Plasmid (h): sc-105319-SH and ECP shRNA (h) Lentiviral Particles: sc-105319-V as alternate gene silencing products.

For independent verification of ECP (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-105319A, sc-105319B and sc-105319C.

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 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

ECP siRNA (h) is recommended for the inhibition of ECP 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

ECP (C3): sc-517595 is recommended as a control antibody for monitoring of ECP 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-IgGλ BP-HRP: sc-516132 or m-IgGλ BP-HRP (Cruz Marker): sc-516132-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-IgGλ BP-FITC: sc-516185 or m-IgGλ BP-PE: sc-516186 (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 ECP gene expression knockdown using RT-PCR Primer: ECP (h)-PR: sc-105319-PR (20 µl, 363 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 for detailed protocols and support products.