

## ECM29 siRNA (h): sc-92599

### BACKGROUND

HEAT repeats are tandemly repeated, 37-47 amino acid long modules that occur in a variety of cytoplasmic proteins. HEAT repeats form a rod-like helical structure and likely operate as protein-protein interaction surfaces. ECM29 (proteasome-associated protein ECM29 homolog), also known as KIAA0368, is a 1,845 amino acid protein containing 27 HEAT repeats. Localizing primarily to endoplasmic reticulum, ECM29 acts as an adapter or scaffolding protein, binding to the 26S proteasome, motor proteins and other compartment specific proteins. The N-terminus of ECM29 binds endocytic proteins, including Rabaptin, Rab11-FIP4 and VPS11, while the C-terminus binds myosins and kinesins. ECM29 may also play a role in endoplasmic reticulum (ER)-associated protein degradation (ERAD) in addition to other enhanced proteolysis.

### REFERENCES

1. Leggett, D.S., et al. 2002. Multiple associated proteins regulate proteasome structure and function. *Mol. Cell* 10: 495-507.
2. Gorbea, C., et al. 2004. Characterization of mammalian ECM29, a 26S proteasome-associated protein that localizes to the nucleus and membrane vesicles. *J. Biol. Chem.* 279: 54849-54861.
3. Kajava, A.V., et al. 2004. New HEAT-like repeat motifs in proteins regulating proteasome structure and function. *J. Struct. Biol.* 146: 425-430.
4. Humphray, S.J., et al. 2004. DNA sequence and analysis of human chromosome 9. *Nature* 429: 369-374.
5. Gorbea, C., et al. 2010. A protein interaction network for ECM29 links the 26S proteasome to molecular motors and endosomal components. *J. Biol. Chem.* 285: 31616-31633.
6. Lehmann, A., et al. 2010. ECM29 fulfills quality control functions in proteasome assembly. *Mol. Cell* 38: 879-888.
7. Wang, X., et al. 2010. Regulation of the 26S proteasome complex during oxidative stress. *Sci. Signal.* 3: ra88.

### CHROMOSOMAL LOCATION

Genetic locus: KIAA0368 (human) mapping to 9q31.3.

### PRODUCT

ECM29 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 ECM29 shRNA Plasmid (h): sc-92599-SH and ECM29 shRNA (h) Lentiviral Particles: sc-92599-V as alternate gene silencing products.

For independent verification of ECM29 (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-92599A, sc-92599B and sc-92599C.

### 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  $\mu$ 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

ECM29 siRNA (h) is recommended for the inhibition of ECM29 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.

### RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ECM29 gene expression knockdown using RT-PCR Primer: ECM29 (h)-PR: sc-92599-PR (20  $\mu$ l). 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](http://www.scbt.com) for detailed protocols and support products.