

# E-Syt1 siRNA (h): sc-95714

## BACKGROUND

Synaptotagmins are a large gene family that function as regulators of both exocytosis and endocytosis and are involved in neurotransmitter secretion from small secretory vesicles. E-Syt1, E-Syt2 and E-Syt3 are  $\text{Ca}^{2+}$ -regulated intrinsic membrane proteins that belong to the extended synaptotagmins (E-Syts) family. Primary structures and biochemical properties of the E-Syts family are highly conserved from yeast to human and consist of multiple C2 domains, which mediate lipid and calcium binding. Ubiquitously expressed with highest expression in brain, members of the E-Syts family form heteromeric complexes and are thought to play a role in the formation of junctions between endoplasmic reticulum and plasma membrane. E-Syt1 (extended synaptotagmin-like protein 1), also known as MBC2 or FAM62A, is a 1,104 amino acid protein that exists as two alternatively spliced isoforms. The gene encoding E-Syt1 is located on human chromosome 12q13.2.

## REFERENCES

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- Giordano, F., et al. 2013. PI(4,5)P(2)-dependent and  $\text{Ca}^{2+}$ -regulated ER-PM interactions mediated by the extended synaptotagmins. *Cell* 153: 1494-1509.
- Herdman, C., et al. 2014. Loss of Extended Synaptotagmins ESyt2 and ESyt3 does not affect mouse development or viability, but *in vitro* cell migration and survival under stress are affected. *Cell Cycle* 13: 2616-2625.
- Tremblay, M.G., et al. 2015. Extended synaptotagmin interaction with the fibroblast growth factor receptor depends on receptor conformation, not catalytic activity. *J. Biol. Chem.* 290: 16142-16156.
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## CHROMOSOMAL LOCATION

Genetic locus: ESYT1 (human) mapping to 12q13.2.

## PRODUCT

E-Syt1 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  $\mu\text{M}$  solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see E-Syt1 shRNA Plasmid (h): sc-95714-SH and E-Syt1 shRNA (h) Lentiviral Particles: sc-95714-V as alternate gene silencing products.

For independent verification of E-Syt1 (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-95714A, sc-95714B and sc-95714C.

## 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

E-Syt1 siRNA (h) is recommended for the inhibition of E-Syt1 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  $\mu\text{M}$  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

E-Syt1 (C-1): sc-514488 is recommended as a control antibody for monitoring of E-Syt1 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 FAM62A gene expression knockdown using RT-PCR Primer: FAM62A (h)-PR: sc-95714-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.