SANTA CRUZ BIOTECHNOLOGY, INC.

MAST205 siRNA (m): sc-62603

BACKGROUND

Syntrophin is an adapter protein that functions to bind certain signaling molecules to the dystrophin-associated protein complex. This complex connects the extracellular matrix to the intracellular cytoskeleton for construction and maintenance of the postsynaptic structures in the neuromuscular junction and the central nervous system. Microtubule-associated serine/threonine-protein kinase 2 (MAST205) is a testis-specific, cytoplasmic protein that functions in a multi-protein complex in the maturation of spermatids. MAST205 is involved in linking the dystrophin/utrophin network with microtubule filaments via syntrophin. By forming a complex with TRAF6, MAST205 regulates lipopolysaccharide-induced IL-12 synthesis in macrophages. This leads to the inhibition of TRAF6 NFκB activation. Two isoforms exist for MAST205 due to alternative splicing. Isoform 1 represents the full length protein, while isoform 2 lacks the residues 327-396 and 1091-1113. The N-terminus of MAST205 must be phosphorylated in order for ubiquitination to occur at the same site. This ubiquitination leads to the degradation of MAST205 via proteasome-mediated proteolysis.

REFERENCES

1. Walden, P.D., et al. 1994. A novel 205-kilodalton testis-specific serine/threonine protein kinase associated with microtubules of the spermatid manchette. Mol. Cell. Biol. 13: 7625-7635.
2. Walden, P.D., et al. 1997. Increased activity associated with the MAST205 protein kinase complex during mammalian spermiogenesis. Biol. Reprod. 55: 1039-1044.
3. Lumeng, C., et al. 1999. Interactions between β2-syntrophin and a family of microtubule-associated serine/threonine kinases. Nat. Neurosci. 2: 611-617.
4. Xiong, H., et al. 2004. Interaction of TRAF6 with MAST205 regulates NFκB activation and MAST205 stability. J. Biol. Chem. 279: 43675-43683.
5. Zhou, H., et al. 2004. Microtubule-associated serine/threonine kinase-205 kDa and Fc γ receptor control IL-12 p40 synthesis and NFκB activation. J. Immunol. 172: 2559-2568.
6. Valiente, M., et al. 2005. Binding of PTEN to specific PDZ domains contributes to PTEN protein stability and phosphorylation by microtubule-associated serine/threonine kinases. J. Biol. Chem. 280: 28936-28943.

CHROMOSOMAL LOCATION

Genetic locus: Mast2 (mouse) mapping to 4 D1.

PRODUCT

MAST205 siRNA (m) 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 MAST205 shRNA Plasmid (m): sc-62603-SH and MAST205 shRNA (m) Lentiviral Particles: sc-62603-V as alternate gene silencing products.

For independent verification of MAST205 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-62603A, sc-62603B and sc-62603C.

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

MAST205 siRNA (m) is recommended for the inhibition of MAST205 expression in mouse 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

MAST205 (A-7): sc-377198 is recommended as a control antibody for monitoring of MAST205 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 MAST205 gene expression knockdown using RT-PCR Primer: MAST205 (m)-PR: sc-62603-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.