SANTA CRUZ BIOTECHNOLOGY, INC.

4E-BP1 siRNA (h): sc-29594

BACKGROUND

The translation of proteins from eukaryotic mRNA is initiated by the multisubunit complex eIF-4F, which associates with the mRNA 5' cap structure. eIF-4E, a component of eIF-4F, is responsible for binding to the 5' cap structure and for the assembly of the eIF-4F complex. The regulatory protein 4E-BP1, also referred to as PHAS-I, inhibits eIF-4E function. Phosphorylation of 4E-BP1 by S6 kinase p70, MAP kinases or PKCs causes the disassociation of 4E-BP1 from eIF-4E, promoting translation. A protein that is functionally related to 4E-BP1, designated 4E-BP2, also associates with eIF-4E.

CHROMOSOMAL LOCATION

Genetic locus: EIF4EBP1 (human) mapping to 8p11.23.

PRODUCT

4E-BP1 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 4E-BP1 shRNA Plasmid (h): sc-29594-SH and 4E-BP1 shRNA (h) Lentiviral Particles: sc-29594-V as alternate gene silencing products.

For independent verification of 4E-BP1 (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-29594A, sc-29594B and sc-29594C.

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

4E-BP1 siRNA (h) is recommended for the inhibition of 4E-BP1 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.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

GENE EXPRESSION MONITORING

4E-BP1 (P-1): sc-9977 is recommended as a control antibody for monitoring of 4E-BP1 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor 4E-BP1 gene expression knockdown using RT-PCR Primer: 4E-BP1 (h)-PR: sc-29594-PR (20 µl, 424 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

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3. Lin, Z., et al. 2014. Epstein-Barr virus-encoded latent membrane protein 2A promotes the epithelial-mesenchymal transition in nasopharyngeal carcinoma via metastatic tumor antigen 1 and mechanistic target of rapamycin signaling induction. J. Virol. 88: 11872-11885.
4. Lee, Y.C., et al. 2018. ABT-263-induced MCL1 upregulation depends on autophagy-mediated 4EBP1 downregulation in human leukemia cells. Cancer Lett. 432: 191-204.
5. Gao, Q., et al. 2019. Distinct role of 4E-BP1 and S6K1 in regulating autophagy and hepatitis B virus (HBV) replication. Life Sci. 220: 1-7.
6. Plaisance-Bonstaff, K., et al. 2019. Isolation, transfection, and culture of primary human monocytes. J. Vis. Exp. 10.3791/59967.
7. Kruse, T., et al. 2020. Mechanisms of site-specific dephosphorylation and kinase opposition imposed by PP2A regulatory subunits. EMBO J. 39: e103695.
8. Chaturvedi, N.K., et al. 2020. A novel combination approach targeting an enhanced protein synthesis pathway in Myc-driven (group 3) medulloblastoma. Mol. Cancer Ther. 19: 1351-1362.
9. Vittori, C., et al. 2022. Mechanisms of miR-3189-3p-mediated inhibition of c-Myc translation in triple negative breast cancer. Cancer Cell Int. 22: 204.
10. Yamamoto, V., et al. 2024. GRP78 inhibitor YUM70 upregulates 4E-BP1 and suppresses c-MYC expression and viability of oncogenic c-MYC tumors. Neoplasia 55: 101020.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.