

UBE2O siRNA (h): sc-94199

BACKGROUND

UBE2O (ubiquitin-conjugating enzyme E2O), also known as E2-230K, is a 1,292 amino acid member of the ubiquitin-conjugating enzyme family that is involved in protein modification. Expressed predominately in heart and skeletal muscle, UBE2O functions to catalyze the ATP-dependent covalent attachment of ubiquitin to select proteins, thereby targeting the ubiquitinated proteins for proteasomal degradation. The gene encoding UBE2O maps to human chromosome 17, which comprises over 2.5% of the human genome and encodes over 1,200 genes. Two key tumor suppressor genes are associated with chromosome 17, namely, p53 and BRCA1. Tumor suppressor p53 is necessary for maintenance of cellular genetic integrity by moderating cell fate through DNA repair versus cell death. Malfunction or loss of p53 expression is associated with malignant cell growth and Li-Fraumeni syndrome. Like p53, BRCA1 is directly involved in DNA repair, though specifically it is recognized as a genetic determinant of early onset breast cancer and predisposition to cancers of the ovary, colon, prostate gland and fallopian tubes.

REFERENCES

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2. Yokota, T., et al. 2001. Identification, tissue expression, and chromosomal position of a novel gene encoding human ubiquitin-conjugating enzyme E2-230k. Gene 267: 95-100.
3. Tomsig, J.L., et al. 2003. Identification of targets for calcium signaling through the copine family of proteins. Characterization of a coiled-coil copine-binding motif. J. Biol. Chem. 278: 10048-10054.
4. Nusbaum, R., et al. 2006-2007. Susceptibility to breast cancer: hereditary syndromes and low penetrance genes. Breast Dis. 27: 21-50.
5. Tai, Y.C., et al. 2007. Breast cancer risk among male BRCA1 and BRCA2 mutation carriers. J. Natl. Cancer Inst. 99: 1811-1814.
6. Yan, J., et al. 2007. BLIMP1 regulates cell growth through repression of p53 transcription. Proc. Natl. Acad. Sci. USA 104: 1841-1846.

CHROMOSOMAL LOCATION

Genetic locus: UBE2O (human) mapping to 17q25.1.

PRODUCT

UBE2O 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see UBE2O shRNA Plasmid (h): sc-94199-SH and UBE2O shRNA (h) Lentiviral Particles: sc-94199-V as alternate gene silencing products.

For independent verification of UBE2O (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-94199A, sc-94199B and sc-94199C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

UBE2O siRNA (h) is recommended for the inhibition of UBE2O 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 μ M in 66 μ 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

UBE2O (2C10): sc-293246 is recommended as a control antibody for monitoring of UBE2O 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 κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 UBE2O gene expression knockdown using RT-PCR Primer: UBE2O (h)-PR: sc-94199-PR (20 μ l). 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.