SANTA CRUZ BIOTECHNOLOGY, INC.

BOULE siRNA (m): sc-60281

BACKGROUND

Spermatogenesis represents the intricate developmental process of mitotic and meiotic cell divisions that ultimately leads to the production of haploid spermatozoa. BOULE, a member of the human deleted in azoospermia (DAZ) family, functions as a key conserved switch that regulates the progression of germ cells through meiosis in man. BOULE is an RNA-binding protein that regulates the expression of twine, a Cdc25 phosphatase, which promotes progression through meiosis. BOULE is expressed not only in the testis, but also in the nervous system, where it may play a role in neural communication. Mutations in the BOULE gene are be associated with male infertility, and the relative proportions of the three BOULE isoforms (B1, B2 and B3) may function as predictive markers for meiotic efficiency.

REFERENCES

1. Xu, E.Y., et al. 2002. Human BOULE gene rescues meiotic defects in infertile flies. Hum. Mol. Genet. 12: 169-175.
2. Joiner, M.L., et al. 2004. Nervous system function for the testis RNA-binding protein BOULE in *Drosophila*. J. Neurogenet. 18: 341-363.
3. Luetjens, C.M., et al. 2004. Association of meiotic arrest with lack of BOULE protein expression in infertile men. J. Clin. Endocrinol. Metab. 89: 1926-1933.
4. Ezeh, U.I., et al. 2005. Human embryonic stem cell genes Oct-4, Nanog, STELLAR, and GDF-3 are expressed in both seminoma and breast carcinoma. Cancer 104: 2255-2265.
5. Lin, Y.M., et al. 2005. Messenger RNA transcripts of the meiotic regulator BOULE in the testis of azoospermic men and their application in predicting the success of sperm retrieval. Hum. Reprod. 20: 782-788.
6. Urano, J., et al. 2005. Interaction of the conserved meiotic regulators, BOULE (BOL) and Pumilio 2 (PUM2). Mol. Reprod. Dev. 71: 290-298.
7. Westerveld, G.H., et al. 2005. Mutations in the human BOULE gene are not a major cause of impaired spermatogenesis. Fertil. Steril. 83: 513-515.
8. Tung, J.Y., et al. 2006. Evolutionary comparison of the reproductive genes, DAZL and BOULE, in primates with and without DAZ. Dev. Genes Evol. 216: 158-168.

CHROMOSOMAL LOCATION

Genetic locus: Boll (mouse) mapping to 1 C1.2.

PRODUCT

BOULE 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 BOULE shRNA Plasmid (m): sc-60281-SH and BOULE shRNA (m) Lentiviral Particles: sc-60281-V as alternate gene silencing products.

For independent verification of BOULE (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-60281A, sc-60281B and sc-60281C.

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

BOULE siRNA (m) is recommended for the inhibition of BOULE 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

BOULE (B-2): sc-166660 is recommended as a control antibody for monitoring of BOULE 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 BOULE gene expression knockdown using RT-PCR Primer: BOULE (m)-PR: sc-60281-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.