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Diagnostik & molekulare Diagnostik



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IBRDC2 siRNA (m): sc-44825



The Power to Question

BACKGROUND

p53 is the most commonly mutated gene in human cancer identified to date. Expression of p53 leads to inhibition of cell growth by preventing progression of cells from G₁S phase of the cell cycle. Most importantly, p53 functions to cause arrest of cells in the G₁ phase of the cell cycle following any exposure of cells to DNA-damaging agents. The MDM2 (murine double minute-2) protein was initially identified as an oncogene in a murine transformation system. MDM2 functions to bind p53 and block p53-mediated transactivation of cotransfected reporter constructs. The MDM2 gene is amplified in a high percentage of human sarcomas that retain wildtype p53 and tumor cells that overexpress MDM2 can tolerate high levels of p53 expression. Another p53 target protein is the p53-inducible RING finger protein (p53RFP), an auto-ubiquitinylated protein acting as an E3 ubiquitin ligase. p53RFP, also designated IBRDC2 in mouse and rat, receives ubiquitin from specific E2 ubiquitin-conjugating enzymes and transfers it to substrates that promote their degradation by the proteasome. p53RFP may mediate reentry into the cell cycle.

REFERENCES

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- Fakharzadeh, S.S., et al. 1991. Tumorigenic potential associated with enhanced expression of a gene that is amplified in a mouse tumor cell line. EMBO J. 10: 1565-1569.
- Oliner, J.D., et al. 1993. Oncoprotein MDM2 conceals the activation domain of tumour suppressor p53. Nature 362: 857-860.
- Chen, C.Y., et al. 1994. Interactions between p53 and MDM2 in a mammalian cell cycle checkpoint pathway. Proc. Natl. Acad. Sci. USA 91: 2684-2688.
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CHROMOSOMAL LOCATION

Genetic locus: Rnf144b (mouse) mapping to 13 A5.

PRODUCT

IBRDC2 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 IBRDC2 shRNA Plasmid (m): sc-44825-SH and IBRDC2 shRNA (m) Lentiviral Particles: sc-44825-V as alternate gene silencing products.

For independent verification of IBRDC2 (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-44825A, sc-44825B and sc-44825C.

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

IBRDC2 siRNA (m) is recommended for the inhibition of p53RFP 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 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-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor p53RFP gene expression knockdown using RT-PCR Primer: IBRDC2 (m)-PR: sc-44825-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.

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