

Wee 1 siRNA (m): sc-36836

BACKGROUND

Phosphorylation of Cdc2 on threonine 14 and tyrosine 15 is required to maintain Cdc2 in an inactive state throughout the S and G₂ phases of the cell cycle. The human Wee 1 protein, Wee 1 Hu, encodes a tyrosine-specific protein kinase that phosphorylates Cdc2 on tyrosine 15. Myt 1, a member of the Wee 1 family of protein kinases, has been shown to phosphorylate Cdc2 on both threonine 14 and tyrosine 15 in a cyclin-dependent manner. Activity of both Wee 1 Hu and Myt 1 is regulated during the cell cycle, suggesting that both proteins play a role in mitotic control. Dephosphorylation of Cdc2 on threonine 14 and tyrosine 15 in late G₂ by Cdc25 then activates the Cdc2/cyclin B complex to allow entry into mitosis.

REFERENCES

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2. Krek, W., et al. 1991. Differential phosphorylation of vertebrate Cdc2 p34 kinase at the G₁/S and G₂/M transitions of the cell cycle: identification of major phosphorylation sites. *EMBO J.* 10: 305-316.
3. Igarashi, M., et al. 1991. Wee1⁺-like gene in human cells. *Nature* 353: 80-83.
4. McGowan, C.H., et al. 1995. Human Wee 1 kinase inhibits cell division by phosphorylating Cdc2 p34 exclusively on Tyr15. *EMBO J.* 12: 75-85.
5. Watanabe, N., et al. 1995. Regulation of the human Wee 1 Hu CDK tyrosine 15-kinase during the cell cycle. *EMBO J.* 14: 1878-1891.
6. Liu, F., et al. 1997. The human Myt 1 kinase preferentially phosphorylates Cdc2 on threonine 14 and localizes to the endoplasmic reticulum and Golgi complex. *Mol. Cell. Biol.* 17: 571-583.
7. Squire, C.J., et al. 2005. Structure and inhibition of the human cell cycle checkpoint kinase, Wee 1A kinase: an atypical tyrosine kinase with a key role in CDK1 regulation. *Structure* 13: 541-550.
8. Kiviharju-af Hällström, T.M., et al. 2007. Human prostate epithelium lacks Wee1A-mediated DNA damage-induced checkpoint enforcement. *Proc. Natl. Acad. Sci. USA* 104: 7211-7216.
9. Kamata, M., et al. 2008. Human immunodeficiency virus type 1 Vpr binds to the N lobe of the Wee1 kinase domain and enhances kinase activity for CDC2. *J. Virol.* 82: 5672-5682.

CHROMOSOMAL LOCATION

Genetic locus: Wee1 (mouse) mapping to 7 F1.

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.

PRODUCT

Wee 1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Wee 1 shRNA Plasmid (m): sc-36836-SH and Wee 1 shRNA (m) Lentiviral Particles: sc-36836-V as alternate gene silencing products.

For independent verification of Wee 1 (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-36836A, sc-36836B and sc-36836C.

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

Wee 1 siRNA (m) is recommended for the inhibition of Wee 1 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-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Wee 1 gene expression knockdown using RT-PCR Primer: Wee 1 (m)-PR: sc-36836-PR (20 μ l, 480 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.