

# p21 Waf1/Cip1 siRNA (m2): sc-44271

## BACKGROUND

It is now well established that cyclins play a positive role in promoting cell cycle transitions via their ability to associate with and activate their cognate cyclin-dependent kinases (Cdks). Cdk2 associates with cyclins A, D and E, and has been implicated in the control of the G<sub>1</sub> to S phase transition in mammals. A novel Cdk-interacting protein, designated p21 Waf1/Cip1, Cip1 or WAF1, has been identified in cyclin A, cyclin D1, cyclin E and Cdk2 immunoprecipitates. p21 Waf1/Cip1 is a potent, tight-binding inhibitor of Cdks and can inhibit the phosphorylation of Rb by cyclin A-Cdk 2, cyclin E-Cdk2, cyclin D1-Cdk4 and cyclin D2-Cdk4 complexes. Expression of p21 Waf1/Cip1 is inducible by wildtype, but not mutant, p53. The mouse homolog of p21 Waf1/Cip1 is designated CAP20.

## REFERENCES

1. Sherr, C.J. 1993. Mammalian G<sub>1</sub> cyclins. *Cell* 73: 1059-1065.
2. Harper, J.W., et al. 1993. The p21 Cdk-interacting protein Cip1 is a potent inhibitor of G<sub>1</sub> cyclin-dependent kinases. *Cell* 75: 805-816.
3. El-Deiry, W.S., et al. 1993. WAF1, a potential mediator of p53 tumor suppression. *Cell* 75: 817-825.
4. Hunter, T. 1993. Braking the cycle. *Cell* 75: 839-841.
5. Kato, J., et al. 1993. Direct binding of cyclin D to the retinoblastoma gene product and pRb phosphorylation by the cyclin D-dependent kinase Cdk4. *Genes Dev.* 7: 331-342.
6. Xiong, Y., et al. 1993. p21 is a universal inhibitor of cyclin kinases. *Nature* 366: 701-704.

## CHROMOSOMAL LOCATION

Genetic locus: Cdkn1a (mouse) mapping to 17 A3.3.

## PRODUCT

p21 Waf1/Cip1 siRNA (m2) 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see p21 Waf1/Cip1 shRNA Plasmid (m2): sc-44271-SH and p21 Waf1/Cip1 shRNA (m2) Lentiviral Particles: sc-44271-V as alternate gene silencing products.

For independent verification of p21 Waf1/Cip1 (m2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44271A, sc-44271B and sc-44271C.

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

## APPLICATIONS

p21 Waf1/Cip1 siRNA (m2) is recommended for the inhibition of p21 Waf1/Cip1 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  $\mu$ M in 66  $\mu$ 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

p21 Waf1/Cip1 (F-5): sc-6246 is recommended as a control antibody for monitoring of p21 Waf1/Cip1 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor p21 Waf1/Cip1 gene expression knockdown using RT-PCR Primer: p21 Waf1/Cip1 (m2)-PR: sc-44271-PR (20  $\mu$ l, 600 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Uchida, M., et al. 2016. p27<sup>Kip1</sup> and p21<sup>Cip1</sup>-independent proliferative inhibition of vascular smooth muscle cells cultured in type-I collagen matrix honeycombs. *Microvasc. Res.* 103: 36-40.

## RESEARCH USE

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

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.