

p-Chk1 (Ser 345): sc-21866

BACKGROUND

Cell cycle events are regulated by the sequential activation and deactivation of cyclin dependent kinases (Cdks) and by proteolysis of cyclins. Chk1 and Chk2 are involved in these processes as regulators of Cdks. Chk1 and Chk2 both function as essential components in the G₂ DNA damage checkpoint by phosphorylating Cdc25C in response to DNA damage. Phosphorylation inhibits Cdc25C activity, thereby blocking mitosis. Cdc25A, Cdc25B and Cdc25C protein tyrosine phosphatases function as mitotic activators by dephosphorylating Cdc2 p34 on regulatory tyrosine residues. It has also been shown that Chk1 can phosphorylate Wee 1 *in vitro*, providing evidence that the hyperphosphorylated form of Wee 1, seen in cells delayed by Chk1 overexpression, is due to phosphorylation by Chk1. Chk1 is phosphorylated on Serine 345 (S345) in response to UV, IR and hydroxyurea (HU). Chk1 plays an essential role in the mammalian DNA damage checkpoint, embryonic development and tumor suppression.

REFERENCES

1. Gautier, J., Solomon, M.J., Booher, R.N., Bazan, J.F. and Kirschner, M.W. 1991. Cdc25 is a specific tyrosine phosphatase that directly activates p34Cdc2. *Cell* 67: 197-211.
2. Barinaga, M. 1995. A new twist to the cell cycle. *Science* 269: 631-632.
3. Sanchez, Y., Wong, C., Thoma, R.S., Richman, R., Wu, Z., Piwnica-Worms, H. and Elledge, S.J. 1997. Conservation of the Chk1 checkpoint pathway in mammals: linkage of DNA damage to Cdk regulation through Cdc25. *Science* 277: 1497-1501.
4. Peng, C.Y., Graves, P.R., Thoma, R.S., Wu, Z., Shaw, A.S. and Piwnica-Worms, H. 1997. Mitotic and G₂ checkpoint control: regulation of 14-3-3 protein binding by phosphorylation of Cdc25C on Serine 216. *Science* 277: 1501-1505.
5. O'Connell, M.J., Raleigh, J.M., Verkade, H.M. and Nurse, P. 1997. Chk1 is a Wee 1 kinase in the G₂ DNA damage checkpoint inhibiting Cdc2 by Y15 phosphorylation. *EMBO J.* 16: 545-554.

CHROMOSOMAL LOCATION

Genetic locus: CHEK1 (human) mapping to 11q24.2; Chk1 (mouse) mapping to 9 A4.

SOURCE

p-Chk1 (Ser 345) is available as either goat (sc-21866) or rabbit (sc-21866-R) polyclonal affinity purified antibody raised against a short amino acid sequence containing Ser 345 phosphorylated Chk1 of human origin.

PRODUCT

Each vial contains 200 µg IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-21866 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

APPLICATIONS

p-Chk1 (Ser 345) is recommended for detection of Ser 345 phosphorylated Chk1 of mouse, rat and human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

p-Chk1 (Ser 345) is also recommended for detection of correspondingly phosphorylated Chk1 in additional species, including equine, canine, bovine, porcine and avian.

Suitable for use as control antibody for Chk1 siRNA (h): sc-29269, Chk1 siRNA (m): sc-29270, Chk1 shRNA Plasmid (h): sc-29269-SH, Chk1 shRNA Plasmid (m): sc-29270-SH, Chk1 shRNA (h) Lentiviral Particles: sc-29269-V and Chk1 shRNA (m) Lentiviral Particles: sc-29270-V.

Molecular Weight of p-Chk1: 56 kDa.

RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: for goat primary antibody (sc-21866): use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), for rabbit primary antibody sc-21866-R: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Western Blotting Luminol Reagent: sc-2048 and Lambda Phosphatase: sc-200312A. 2) Immunofluorescence: for goat primary antibody (sc-21866): use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941, for rabbit primary antibody (sc-21866-R): use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

SELECT PRODUCT CITATIONS

1. Ullah, Z., Kohn, M.J., Yagi, R., Vassilev, L.T. and DePamphilis, M.L. 2008. Differentiation of trophoblast stem cells into giant cells is triggered by p57/Kip2 inhibition of Cdk1 activity. *Genes Dev.* 22: 3024-3036.
2. Wang, J., Gu, Q., Li, M., Zhang, W., Yang, M., Zou, B., Chan, S., Qiao, L., Jiang, B., Tu, S., Ma, J., Hung, I.F., Lan, H.Y. and Wong, B.C. 2009. Identification of XAF1 as a novel cell cycle regulator through modulating G₂/M checkpoint and interaction with checkpoint kinase 1 in gastrointestinal cancer. *Carcinogenesis* 30: 1507-1516.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

RESEARCH USE

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