

p-Claspin (Ser 945): sc-31718

BACKGROUND

Claspin is a large acidic protein that undergoes phosphorylation and binds to Chk1 in the presence of checkpoint-inducing DNA templates in *Xenopus* egg extracts. Serine 864, Serine 895 and Threonine 906 undergo phosphorylation during the DNA replication checkpoint response. Phosphorylation of Threonine 906 creates a docking site for Plx1, the *Xenopus* Polo-like kinase. This interaction promotes the phosphorylation of Claspin on a nearby Serine 934 by Plx1. Phosphorylation of Claspin at Threonine 916 and Serine 945 is required for the DNA-dependent binding of Claspin to Chk1. A mutant of Claspin containing substitutions at positions 864 and 895 cannot bind to Chk1 and is unable to mediate its activation. Claspin, ATR and Rad17 may collaborate in checkpoint regulation by detecting different aspects of a DNA replication fork. Downregulation of Claspin augments the premature chromatin condensation induced by hydroxyurea, inhibits the UV-induced reduction of DNA synthesis and decreases cell survival. ATR can regulate Claspin phosphorylation in response to DNA damage and replication stress, resulting in recruitment and phosphorylation of BRCA1.

REFERENCES

1. Kumagai, A., et al. 2000. Claspin, a novel protein required for the activation of Chk1 during a DNA replication checkpoint response in *Xenopus* egg extracts. *Mol. Cell* 6: 839-849.
2. Kumagai, A., et al. 2003. Repeated phosphopeptide motifs in Claspin mediate the regulated binding of Chk1. *Nat. Cell Biol.* 5: 161-165.
3. Lee, J., et al. 2003. Claspin, a Chk1-regulatory protein, monitors DNA replication on chromatin independently of RPA, ATR, and Rad17. *Mol. Cell* 11: 329-340.
4. Chini, C.C., et al. 2003. Human Claspin is required for replication checkpoint control. *J. Biol. Chem.* 278: 30057-30062.
5. Jeong, S.Y., et al. 2003. Phosphorylated Claspin interacts with a phosphate-binding site in the kinase domain of Chk1 during ATR-mediated activation. *J. Biol. Chem.* 278: 46782-46788.

CHROMOSOMAL LOCATION

Genetic locus: CLSPN (human) mapping to 1p34.3; Clspn (mouse) mapping to 4 D2.2.

SOURCE

p-Claspin (Ser 945) is a rabbit polyclonal antibody raised against a short amino acid sequence containing Ser 945 phosphorylated Claspin of human origin.

STORAGE

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

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.

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-31718 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

APPLICATIONS

p-Claspin (Ser 945) is recommended for detection of Ser 945 phosphorylated Claspin 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), immunohistochemistry (including paraffin-embedded sections) (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

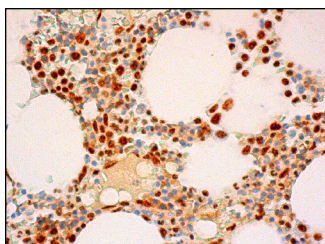
p-Claspin (Ser 945) is also recommended for detection of correspondingly phosphorylated Claspin in additional species, including equine, canine, bovine, porcine and avian.

Suitable for use as control antibody for Claspin siRNA (h): sc-45412, Claspin siRNA (m): sc-45413, Claspin shRNA Plasmid (h): sc-45412-SH, Claspin shRNA Plasmid (m): sc-45413-SH, Claspin shRNA (h) Lentiviral Particles: sc-45412-V and Claspin shRNA (m) Lentiviral Particles: sc-45413-V.

RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: 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 A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: 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. 3) Immunohistochemistry: use ImmunoCruz™: sc-2051 or ABC: sc-2018 rabbit IgG Staining Systems.

DATA



p-Claspin (Ser 945): sc-31718. Immunoperoxidase staining of formalin fixed, paraffin-embedded human bone marrow tissue showing nuclear staining of a subset of hematopoietic cells.

RESEARCH USE

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