Revision 1

Store at -20C

#3428

Phospho-53BP1 (Thr543) Antibody

Cell Signaling TECHNOLOGY®

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For Research Use Only. Not for Use in Diagnostic Procedures.

Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
W	H	Endogenous	450	Rabbit	#Q12888	7158

Product Usage Information

Application	Dilution
Western Blotting	1:1000

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA and 50% glycerol. Store at –20°C. Do not aliquot the antibody.

Specificity/Sensitivity

Phospho-53BP1 (Thr543) Antibody detects endogenous levels of 53BP1 protein only when phosphorylated at Thr543.

Species predicted to react based on 100% sequence homology

Monkey

Source / Purification

Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surounding Thr543 of human 53BP1. Antibodies are purified using protein A and peptide affinity chromatography.

Background

p53-binding protein 1 (53BP1) was originally identified as a p53 binding partner that could enhance the transcriptional activity of p53 (1,2). 53BP1 consists of two BRCA1 carboxy terminal (BRCT) domains that allow for binding to p53 and a separate domain responsible for binding to phosphorylated histone H2A.X (3). 53BP1 rapidly translocates to nuclear foci following treatment of cells with ionizing radiation (IR) or radiomimetic agents that cause DNA double strand breaks (DSBs) (4,5). Because of this localization to DSBs and homology to the yeast protein Rad9, a role for 53BP1 in DSB repair has been proposed. Recruitment of 53BP1 to sites of DNA damage has been demonstrated to be independent of ATM, NBS1, and DNA-PK (4) and retention of 53BP1 at DNA breaks requires phosphorylated H2A.X (6). In cells lacking 53BP1, phosphorylation of ATM substrates is reduced, suggesting that 53BP1 is upstream of ATM (7). In response to IR, phosphorylation of 53BP1 at serines 6, 25, 29, and 784 by ATM has been demonstrated, but phosphorylation at these sites is not required for localization of 53BP1 to sites of DSBs (6). Phosphorylation of 53BP1 at Ser1618 has been reported to be enriched in human cells arrested in mitosis (8).
Threonine 543 of 53BP1 has been shown to be phosphorylated in an ATM/ATR-dependent manner in response to DNA damage (8,9).
Phospho-53BP1 (Thr543) Antibody is directed at a site that was identified at Cell Signaling Technology (CST) using PhosphoScan®, CST's LC-MS/MS platform for modification site discovery. Phosphorylation at Thr543 was discovered using an ATM/ATR substrate antibody and was shown to be induced by UV treatment. Please visit PhosphoSitePlus®, CST's modification site knowledgebase, at www.phosphosite.org for more information.

Background References

1. Iwabuchi, K. et al. (1994) *Proc. Natl. Acad. Sci. USA* 91, 6098-102.
2. Iwabuchi, K. et al. (1998) *J. Biol. Chem.* 273, 26061-8.
3. Mochan, T.A. et al. (2004) *DNA Repair (Amst)* 3, 945-52.
4. Schultz, L.B. et al. (2000) *J. Cell Biol.* 151, 1381-90.
5. Anderson, L. et al. (2001) *Mol. Cell. Biol.* 21, 1719-29.
6. Ward, I.M. et al. (2003) *J. Biol. Chem.* 278, 19579-82.
7. DiTullio, R.A. et al. (2002) *Nat. Cell Biol.* 4, 998-1002.
8. Dephoure, N. et al. (2008) *Proc Natl Acad Sci U S A* 105, 10762-7.
9. Stokes, M.P. et al. (2007) *Proc. Natl. Acad. Sci. USA* 104, 19855-19860.
10. Matsuoka, S. et al. (2007) *Science* 316, 1160-1166.

Species Reactivity

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

Western Blot Buffer

IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

Applications Key

W: Western Blotting

Cross-Reactivity Key

H: Human

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