

# Rad53 (A-9): sc-74427

## BACKGROUND

DNA damage results in the arrest of cell cycle progression, allowing the damaged DNA to be repaired prior to replication. Checkpoints exist at several cell cycle phase transitions to maintain this genetic integrity. Rad9, Rad17, Rad24 and Mec3 are involved in activating the G<sub>1</sub> and G<sub>2</sub> checkpoints. Pol2 (also known as Dun2), encoding the catalytic subunit of DNA polymerase  $\epsilon$ , plays a role in activating the S phase checkpoint. The protein kinase Rad53 (also designated Spk1, Mec2 or Sad1) is essential for both G<sub>2</sub> and S phase arrest. Activation of Rad53 is regulated by Mec1 (also known as Esr1 and Sad3), a homolog of the human ATM protein. Pds1 and Mad2 both regulate checkpoints associated with incomplete spindle replication. Dun1, another protein kinase, plays a role in transducing the DNA damage signal.

## REFERENCES

- Li, R., et al. 1993. The mitotic feedback control gene MAD2 encodes the  $\alpha$  subunit of a prenyltransferase. *Nature* 366: 82-84.
- Zhou, Z. and Elledge, S.J. 1993. Dun1 encodes a protein kinase that controls the DNA damage response in yeast. *Cell* 75: 1119-1127.
- Abloussekhra, A., et al. 1996. A novel role for the budding yeast Rad9 checkpoint gene in DNA damage-dependent transcription. *EMBO J.* 15: 3912-3922.
- Siede, W., et al. 1996. Cloning and characterization of Rad17, a gene controlling cell cycle responses to DNA damage in *Saccharomyces cerevisiae*. *Nucleic Acids Res.* 24: 1669-1675.
- Lydall, D., et al. 1996. A meiotic recombination checkpoint controlled by mitotic checkpoint genes. *Nature* 383: 840-843.
- Longhese, M.P., et al. 1996. Yeast pep3/Mec3 mutants fail to delay entry into S phase and to slow DNA replication in response to DNA damage, and they define a functional link between Mec3 and DNA primase. *Mol. Cell Biol.* 16: 3235-3244.
- Sanchez, Y., et al. 1996. Regulation of Rad53 by the ATM-like kinases MEC1 and TEL1 in yeast cell cycle checkpoint pathways. *Science* 271: 357-360.

## SOURCE

Rad53 (A-9) is a mouse monoclonal antibody raised against amino acids 1-300 of Rad53 of *Saccharomyces cerevisiae* origin.

## PRODUCT

Each vial contains 200  $\mu$ g IgG<sub>1</sub> kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Rad53 (A-9) is available conjugated to agarose (sc-74427 AC), 500  $\mu$ g/0.25 ml agarose in 1 ml, for IP; to HRP (sc-74427 HRP), 200  $\mu$ g/ml, for WB, IHC(P) and ELISA; to either phycoerythrin (sc-74427 PE), fluorescein (sc-74427 FITC), Alexa Fluor<sup>®</sup> 488 (sc-74427 AF488), Alexa Fluor<sup>®</sup> 546 (sc-74427 AF546), Alexa Fluor<sup>®</sup> 594 (sc-74427 AF594) or Alexa Fluor<sup>®</sup> 647 (sc-74427 AF647), 200  $\mu$ g/ml, for WB (RGB), IF, IHC(P) and FCM; and to either Alexa Fluor<sup>®</sup> 680 (sc-74427 AF680) or Alexa Fluor<sup>®</sup> 790 (sc-74427 AF790), 200  $\mu$ g/ml, for Near-Infrared (NIR) WB, IF and FCM.

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## APPLICATIONS

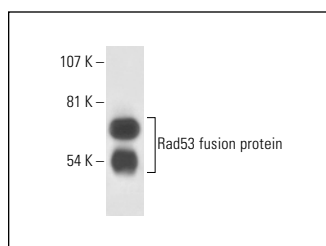
Rad53 (A-9) is recommended for detection of Rad53 of *Saccharomyces cerevisiae* origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2  $\mu$ g per 100-500  $\mu$ g of total protein (1 ml of cell lysate)] and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Molecular Weight of Rad53: 92 kDa.

## RECOMMENDED SUPPORT REAGENTS

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) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml).

## DATA



Rad53 (A-9): sc-74427. Western blot analysis of Yeast recombinant Rad53 fusion protein.

## SELECT PRODUCT CITATIONS

- Chappidi, N., et al. 2019. Replication stress-induced Exo1 phosphorylation is mediated by Rad53/Pph3 and Exo1 nuclear localization is controlled by 14-3-3 proteins. *Cell Div.* 14: 1.
- Gerritelli, S.M., et al. 2020. High density of unrepaired genomic ribonucleotides leads to topoisomerase 1-mediated severe growth defects in absence of ribonucleotide reductase. *Nucleic Acids Res.* 48:4274-4297.

## 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.

## PROTOCOLS

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