# SANTA CRUZ BIOTECHNOLOGY, INC.

# Rad9 (B-8): sc-74464



## BACKGROUND

DNA damage or incomplete replication of DNA results in the inhibition of cell cycle progression at the  $G_1/S$  or  $G_2/M$  phase checkpoints by conserved regulatory mechanisms. Chk1, Rad9 and Hus1 are involved in the signal transduction cascade that regulates cell cycle arrest at the  $G_2$  checkpoint. Chk1 functions as an essential component in the  $G_2$  phase DNA damage checkpoint, as it phosphorylates Cdc25C in response to DNA damage and thereby inhibits mitosis. Two related mammalian proteins, Hus1 and Rad9, share conserved sequence identity and function to the yeast homologs of the same names. *In vivo*, Rad9 is highly phosphorylated and directly associates with two other checkpoint control proteins, Rad1 and Hus1. Additionally, Rad9 associates with anti-apoptotic Bcl-2 family proteins Bcl-2 and Bcl- $x_L$ , but not with the pro-apoptotic Bax and Bad proteins. Overexpression of Rad9 induces apoptosis and indicates that Rad9 may have an additional role in regulating apoptosis after DNA damage.

# **CHROMOSOMAL LOCATION**

Genetic locus: RAD9A (human) mapping to 11q13.2; Rad9 (mouse) mapping to 19 A.

# SOURCE

Rad9 (B-8) is a mouse monoclonal antibody raised against amino acids 1-389 representing full length Rad9 of mouse origin.

#### PRODUCT

Each vial contains 200  $\mu$ g IgG<sub>2a</sub> kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Rad9 (B-8) is available conjugated to agarose (sc-74464 AC), 500 µg/0.25 ml agarose in 1 ml, for IP; to HRP (sc-74464 HRP), 200 µg/ml, for WB, IHC(P) and ELISA; to either phycoerythrin (sc-74464 PE), fluorescein (sc-74464 FITC), Alexa Fluor® 488 (sc-74464 AF488), Alexa Fluor® 546 (sc-74464 AF546), Alexa Fluor® 594 (sc-74464 AF594) or Alexa Fluor® 647 (sc-74464 AF647), 200 µg/ml, for WB (RGB), IF, IHC(P) and FCM; and to either Alexa Fluor® 680 (sc-74464 AF680) or Alexa Fluor® 790 (sc-74464 AF790), 200 µg/ml, for Near-Infrared (NIR) WB, IF and FCM.

# **APPLICATIONS**

Rad9 (B-8) is recommended for detection of Rad9 of mouse, rat and human 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)], 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).

Suitable for use as control antibody for Rad9 siRNA (h): sc-36364, Rad9 siRNA (m): sc-36365, Rad9 shRNA Plasmid (h): sc-36364-SH, Rad9 shRNA Plasmid (m): sc-36365-SH, Rad9 shRNA (h) Lentiviral Particles: sc-36364-V and Rad9 shRNA (m) Lentiviral Particles: sc-36365-V.

Molecular Weight of Rad9: 65 kDa.

Positive Controls: HeLa nuclear extract: sc-2120, NIH/3T3 whole cell lysate: sc-2210 or KNRK nuclear extract: sc-2141.

#### **RECOMMENDED SUPPORT REAGENTS**

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG K BP-HRP: sc-516102 or m-IgG K BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® 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). 3) Immunofluorescence: use m-IgG K BP-FITC: sc-516140 or m-IgG K BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

# DATA





expression in HeLa (A), KNRK (B), NIH/3T3 (C), A-431 (D), PC-3 (E) and CCRF-CEM (F) nuclear extracts Rad9 (B-8): sc-74464. Western blot analysis of Rad9 expression in HeLa (A) and NIH/3T3 (B) whole cell lysates and HeLa (C) and KNRK (D) neclear extracts.

#### SELECT PRODUCT CITATIONS

- Zhang, L., et al. 2010. Proteolysis of Rad17 by Cdh1/APC regulates checkpoint termination and recovery from genotoxic stress. EMBO J. 29: 1726-1737.
- Ackerson, S.M., et al. 2020. Human CTC1 promotes TopBP1 stability and CHK1 phosphorylation in response to telomere dysfunction and global replication stress. Cell Cycle 19: 3491-3507.
- Ho, K., et al. 2021. Critical role of SMG7 in activation of the ATR-CHK1 axis in response to genotoxic stress. Sci. Rep. 11: 7502.
- Xin, D., et al. 2023. Pre-rRNA facilitates TopBP1-mediated DNA doublestrand break response. Adv. Sci. 10: e2206931.

#### **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 for detailed protocols and support products.

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