

Datasheet for 600-401-346

Rad9 S1129 Antibody

Overview

Description:	Anti-Rad9 S1129 Yeast (pan reactive) (RABBIT) Antibody - 600-401-346
Item No.:	600-401-346
Size:	100 µg
Applications:	ELISA, WB
Reactivity:	<i>S. cerevisiae</i>
Host Species:	Rabbit

Product Details

Background:

Rad9 is required for the MEC1/TEL1-dependent activation of *Saccharomyces cerevisiae* DNA damage checkpoint pathways mediated by Rad53 and Chk1. DNA damage induces Rad9 phosphorylation, and Rad53 specifically associates with phosphorylated Rad9. Cells have evolved multiple strategies for tolerating genomic damage. The most important of these are numerous repair systems that remove or bypass potentially mutagenic DNA lesions. Another cellular strategy is to delay cell-cycle transitions at multiple points. The genetic control of these delays, termed 'checkpoints', was first established in budding yeast where it was shown that the RAD9 gene functions in G₂/M arrest after irradiation with X-rays. Subsequently, it has become clear that Rad9 also functions at the G₁/S, intra-S and mid-anaphase checkpoints. Defects in checkpoint regulation can lead to genome instability and, in higher eukaryotes, neoplastic transformation. Rad9 also controls the transcriptional induction of a DNA damage regulon (DDR). Rad9 may also have a pro-apoptotic function. This is suggested in that Rad9 from *Schizosaccharomyces pombe* (SpRad9) contains a group of amino acids with similarity to the Bcl-2 homology 3 death domain, which is required for SpRad9 interaction with human Bcl-2 and apoptosis induction in human cells. Overexpression of Bcl-2 in *S. pombe* inhibits cell growth independently of rad9, but enhances resistance of rad9-null cells to methyl methanesulfonate, ultraviolet and ionizing radiation. Rad9 conveys the checkpoint signal by activating Rad53p and Chk1p; is hyperphosphorylated by Mec1p and Tel1p; and is a potential Cdc28p substrate. Mature yeast Rad9 is reported to have an apparent molecular weight of ~148kDa. The human homolog is reported at 48.5 kDa.

Synonyms:	Rabbit anti-Rad9 antibody, Rabbit anti-Rad9 S1129 antibody, Rad 9, Rad-9, Cell cycle checkpoint control protein antibody, Cell cycle checkpoint control protein RAD9A antibody, DNA repair exonuclease rad9 homolog A antibody, DNA repair protein RAD9, YDR217C, YD9934.02C
Host Species:	Rabbit

Clonality: Polyclonal

Format: IgG

Target Details

Gene Name: RAD9

Reactivity: *S. cerevisiae*

Immunogen Type: Conjugated Peptide

Immunogen: This affinity purified antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to an internal region near aa 1120-1145 from the aa1309 yeast Rad9 protein conjugated to KLH.

Purity/Specificity: This affinity purified antibody is directed against an internal sequence of yeast Rad9 at the S1129 residue. The product was affinity purified from monospecific antiserum by immunoaffinity purification. Antiserum was purified against the immunizing peptide. This pan reactive polyclonal antibody reacts equally with both phosphorylated and non-phosphorylated yeast Rad9 at S1129. No reactivity is expected against human and mouse homologs. Reactivity to Rad9 from others sources is unknown.

Relevant Links:

- [GenelD - 851803](#)
- [UniProtKB - P14737](#)
- [NCBI - NP_010503.1](#)

Application Details

Tested Applications: ELISA, WB

Application Note: This pan reactive polyclonal antibody was tested by immunoblotting and ELISA. Data from both immunoblotting and ELISA indicate the antibody is pan reactive with both the phosphorylated and non-phosphorylated forms of the peptide and protein. Immunoblotting detects yeast Rad9 protein. No reactivity is expected against human and mouse homologs. Reactivity to Rad9 from others sources is unknown. Although not tested, this antibody is likely functional by immunohistochemistry and immunoprecipitation. This product has been assayed against 0.1 µg of immunizing peptide (S1129) in a standard capture ELISA using TMB (3,3',5,5'-Tetramethylbenzidine) code # TMBE-100 as a substrate for 30 minutes at room temperature. A working dilution of 1:5,000 is suggested for this product. Reactivity was detected against both the phosphorylated and non-phosphorylated form (S1129 and pS1129) of the immunizing peptide. This antibody appears to be pan reactive for both forms of the protein. Dilute the antibody 1:100 to 1:500 for immunoblotting. Researchers should determine optimal titers for other applications.

Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:5,000
WB:	1:100-1:500

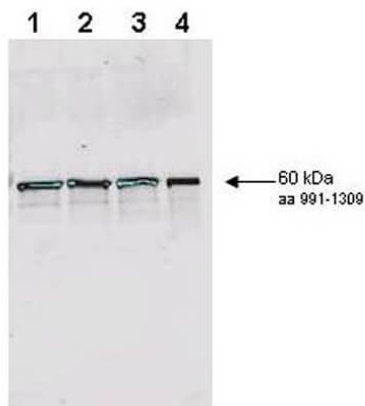
Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	1.0 mg/mL by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	None

Shipping & Handling

Shipping Condition:	Dry Ice
Storage Condition:	Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.
Expiration:	Expiration date is one (1) year from date of receipt.

Images



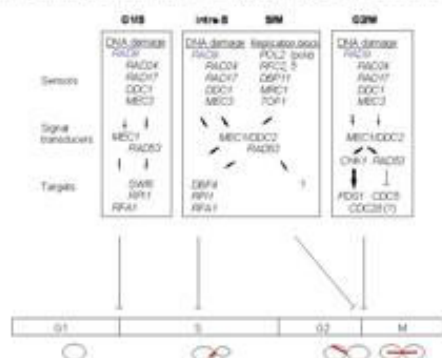
Western Blot

Affinity purified antibody to yeast Rad9 (1125-1139 pan reactive) was used at a 1:200 dilution incubated 8 h at room temperature to detect Rad9 by Western blot. Lanes were loaded with 50 ng each of recombinant GST fusion protein containing *S. cerevisiae* Rad9 (aa 991-1309 ~60 kDa) on a 4-20% Criterion gel for SDS-PAGE as follows: Lane 1 - non-phosphorylated wild type yeast Rad9, Lane 2 - in vitro phosphorylated wild type yeast Rad9, Lane 3 - non-phosphorylated S1129A/S1260A double mutant Rad9, Lane 4 - in vitro phosphorylated S1129A/S1260A double mutant. Phosphorylation of Rad9 was by treatment with ATP and Rad53 kinase. Detection occurred using a 1:5,000 dilution of IRDye™800 conjugated Donkey anti-Rabbit IgG (code # 611-732-127) for 1h at room temperature. LICOR's Odyssey® Infrared Imaging System was used to scan and process the image. Other detection systems will yield similar results.

Pathway

Checkpoints are mechanisms that impose delays in the cell cycle in response to DNA damage or defects in DNA replication, to ensure that mitotic transmission is error-free. Failure to delay the cell cycle in the presence of damage converts an easily repairable DNA lesion into one far more deleterious, provoking genomic instability or cell death. This figure shows a summary of our current knowledge about the DNA damage checkpoints in yeast. Genetic analysis of the pathway has allowed classification of its components into "Sensors", which detect different sorts of damage, "Signal transducers" which are signal-integrating kinases, and "Targets" which carry out the essential functions of suppressing progress through the cell cycle (i.e. inducing repair genes and preventing late origin firing or sister chromatid segregation. Contributed by C. Frei and K. Shimada, laboratory of S. Gasser, U. Geneva.

Schematic summary of the DNA replication and DNA damage checkpoints in *S. cerevisiae*.



Disclaimer

This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information. All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation. All properties listed are typical characteristics and are not specifications. All suggestions and data are offered in good faith but without guarantee as conditions and methods of use of our products are beyond our control. All claims must be made within 30 days following the date of delivery. The prospective user must determine the suitability of our materials before adopting them on a commercial scale. Suggested uses of our products are not recommendations to use our products in violation of any patent or as a license under any patent of Rockland Immunochemicals, Inc. If you require a commercial license to use this material and do not have one, then return this material, unopened to: Rockland Inc., P.O. BOX 5199, Limerick, Pennsylvania, USA.