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Datasheet for ABIN968860 anti-TP53BP1 antibody (AA 149-259)

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Overview

Quantity:	50 µg
Target:	TP53BP1
Binding Specificity:	AA 149-259
Reactivity:	Human
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This TP53BP1 antibody is un-conjugated
Application:	Western Blotting (WB), BioImaging (BI)

Product Details

Immunogen:	Human 53BP1 aa. 149-259
Clone:	19-53BP1
lsotype:	lgG2b
Characteristics:	1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
	2. This antibody has been developed and certified for the bioimaging application. However, a
	routine bioimaging test is not performed on every lot. Researchers are encouraged to titrate the
	reagent for optimal performance.
	3. Triton is a trademark of the Dow Chemical Company.
	4. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
	5. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide
	compounds in running water before discarding to avoid accumulation of potentially explosive

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Product Details	
	deposits in plumbing. 6. Please refer to us for technical protocols.
Purification:	The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Target Details	
Target:	TP53BP1
Alternative Name:	53BP1 (TP53BP1 Products)
Background:	The p53 protein is critical to regulation of normal cell growth and is a suppressor of tumor cell

The p53 protein is critical to regulation of normal cell growth and is a suppressor of tumor cell proliferation. Inactivation of p53 by a number of mechanisms, such as missense mutations or interaction with oncogenic viral or cellular proteins, can result in tumor progression. In addition, Bcl2 and p53 are involved in apoptosis in an antagonistic fashion such that overexpressed Bcl2 inhibits p53-induced apoptosis. 53BP1 and 53BP2 were identified in a yeast two-hybrid screen of proteins that bind p53. Both 53BP1 and 53BP2 bind wild type p53, but not mutant p53 found in tumor cells. p53BP1 is localized to the cytoplasm and nucleus, while p53BP2 is found only in the cytoplasm. 53BP1 has BRCT motifs found in proteins involved in cell cycle control and DNA repair. DNA damage leads to 53BP1 hyperphosphorylation, which may be mediated by ATM. 53BP2 has four ankyrin repeats and a SH3 domain that are required for interactions with Bcl2 and p53. Overexpression of 53BP2 in 293 cells inhibits progression of the cell cycle in G2/M phase, while co-transfection of 53BP2 with p53 in H358 cells enhances p53-mediated transcriptional activation. The interaction between 53BP2 and p53 may be regulated by Bcl2, since competition experiments demonstrate that Bcl2 prevents p53 binding to 53BP2. In addition, 53BP2 can also bind the apoptotic-related p65 subunit of NFkappaB and this subunit can inhibit 53BP2-induced cell death.

Molecular Weight:

Pathways:

DNA Damage Repair

345 kDa

Application Details

Application Notes:	Bioimaging
	1. Seed the cells in appropriate culture medium at \sim 10,000 cells per well in an 96-well Imaging
	Plate and culture overnight.
	2. Remove the culture medium from the wells, and fix the cells by adding 100 myl of Fixation
	Buffer to each well. Incubate for 10 minutes at room temperature (RT).

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	3. Remove the fixative from the wells, and permeabilize the cells using either 90% methanol, or
	Triton™ X-100: a. Add 100 myl of -20°C 90% methanol to each well and incubate for 5 minutes
	at RT. OR b. Add 100 myl of 0.1% Triton™ X-100 to each well and incubate for 5 minutes at RT.
	4. Remove the permeabilization buffer, and wash the wells twice with 100 myl of $1 \times$ PBS.
	5. Remove the PBS, and block the cells by adding 100 myl of to each well. Incubate for 30
	minutes at RT.
	6. Remove the blocking buffer and add 50 myl of the optimally titrated primary antibody (diluted
	in Stain Buffer) to each well, and incubate for 1 hour at RT.
	7. Remove the primary antibody, and wash the wells three times with 100 myl of $1 \times$ PBS.
	8. Remove the PBS, and add the second step reagent at its optimally titrated concentration in
	50 myl to each well, and incubate in the dark for 1 hour at RT.
	9. Remove the second step reagent, and wash the wells three times with 100 myl of $1 \times$ PBS.
	10. Remove the PBS, and counter-stain the nuclei by adding 200 myl per well of 2 myg/ml
	Hoechst 33342 in 1× PBS to each well at least 15 minutes before imaging.
	11. View and analyze the cells on an appropriate imaging instrument.
Comment:	Related Products: ABIN968535, ABIN967389
Restrictions:	For Research Use only

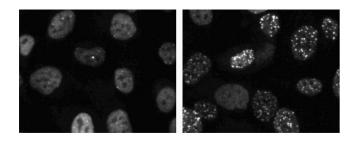
Handling

Format:	Liquid
Concentration:	250 μg/mL
Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤ 0.09 % sodium azide.
Preservative:	Sodium azide
Precaution of Use:	This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.
Storage:	-20 °C
Storage Comment:	Store undiluted at -20°C.
Publications	
Product cited in:	Rappold, Iwabuchi, Date, Chen: "Tumor suppressor p53 binding protein 1 (53BP1) is involved in DNA damage-signaling pathways." in: The Journal of cell biology , Vol. 153, Issue 3, pp. 613-20, (2001) (PubMed).

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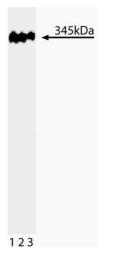
Iwabuchi, Bartel, Li, Marraccino, Fields: "Two cellular proteins that bind to wild-type but not mutant p53." in: **Proceedings of the National Academy of Sciences of the United States of America**, Vol. 91, Issue 13, pp. 6098-102, (1994) (PubMed).

Images



Immunofluorescence

Image 1. Immunofluorescent staining of HT1080 cells (ATCC CCL-121). Cells were seeded in a 96 well imaging plate at ~10,000 cells per well. After overnight incubation, cells were either mock treated (PBS, left) or exposed to hydrogen peroxide (400uM, right) for 30 minutes and allowed to recover in media for 30 minutes. After treatment cells were stained using the alcohol perm protocol and the anti-53BP1 antibody. The second step reagent was Alexa Fluor® 488 goat anti-mouse IgG (Invitrogen). The image is a confocal collapsed stack, taken on a BD Pathway[™] 855 bioimaging system with a 40x objective. This antibody also stains A549 (ATCC CCL-158), HeLa (ATCC CCL-2) and U-2 OS (ATCC HTB-96) cells and can be used with either fix/perm protocol.



Western Blotting

Image 2. Western blot analysis of 53BP1 on a HeLa lysate. Lane 1: 1:1000, lane 2: 1:2000, lane 3: 1:4000 dilution of the anti-53BP1 antibody.

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