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53BP1 Polyclonal Antibody

Catalog Number **PA5-17578**

Product data sheet

Details	
Size	100 µL
Host/Isotope	Rabbit / IgG
Class	Polyclonal
Type	Antibody
Immunogen	Synthetic peptide corresponding to residues near the center of human 53BP1
Conjugate	Unconjugated
Form	Liquid
Concentration	26 µg/mL
Purification	Antigen affinity chromatography
Storage buffer	0.01M HEPES, pH 7.5, with 0.15M NaCl, 100µg/mL BSA, 50% glycerol
Contains	no preservative
Storage Conditions	-20°C

Species Reactivity	
Species reactivity	Human, Non-human primate
Published species	Human

Tested Applications	Dilution *
Immunohistochemistry (Paraffin) (IHC (P))	1:100
Western Blot (WB)	1:1,000
Immunocytochemistry (ICC/IF)	1:100

Published Applications	
Western Blot (WB)	See 1 publications below
Immunohistochemistry (IHC)	See 1 publications below

* Suggested working dilutions are given as a guide only. It is recommended that the user titrate the product for use in their own experiment using appropriate negative and positive controls.

Product specific information

It is not recommended to aliquot this antibody.

Background/Target Information

53BP1 (P53-binding protein 1) plays a critical role in tumor suppression and is a putative substrate of ATM kinase. Upon DNA damage, 53BP1 is phosphorylated and localizes to the presumptive sites of damage, specifically, double-strand breaks. 53BP1 may have a role in checkpoint signaling during mitosis, enhances TP53-mediated transcriptional activation, and participates in DNA repair by maintaining genomic stability.

For Research Use Only. Not for use in diagnostic procedures. Not for resale without express authorization.

Product Images For 53BP1 Polyclonal Antibody

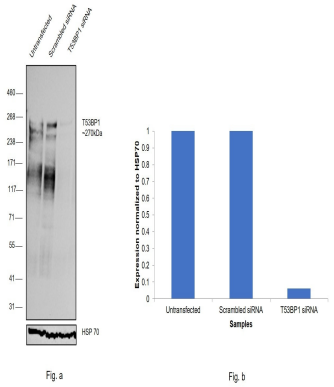
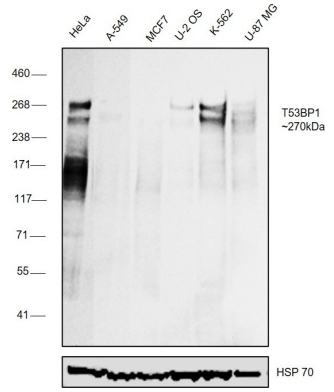


Fig. a Fig. b

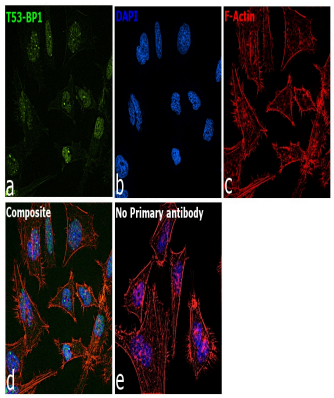
53BP1 Antibody (PA5-17578)

Antibody specificity was demonstrated by siRNA mediated knockdown of target protein. HeLa cells were transfected with TP53-binding protein 1 siRNA and decrease in signal intensity was observed in Western Blot application using Anti-53BP1 Polyclonal Antibody (Product # PA5-17578). {KD}



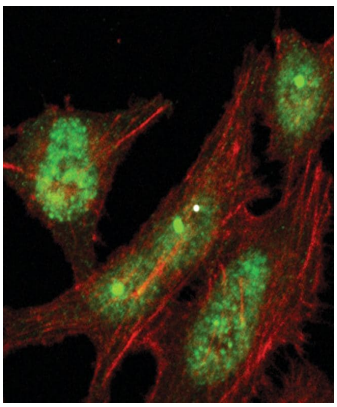
53BP1 Antibody (PA5-17578) in WB

Western blot was performed using Anti-53BP1 Polyclonal Antibody (Product # PA5-17578) and a ~270kDa band corresponding to TP53-binding protein 1 was observed across cell lines tested . Nuclear enriched extracts (30 µg lysate) of HeLa (Lane 1), A549 (Lane 2), MCF7 (Lane 3), U-2 OS (Lane 4), K-562 (Lane 5), U-87 MG (Lane 6) were electrophoresed using NuPAGE™ 3-8% Tris-Acetate Protein Gel (Product # EA0378BOX). Resolved proteins were then transferred onto a Nitrocellulose membrane (Product # LC2002) by iBlot® 2 Dry Blotting System (Product # IB21001). The blot was probed with the primary antibody (1:1000) and detected by chemiluminescence with Goat anti-Rabbit IgG (Heavy Chain) Superclonal™ Recombinant Secondary Antibody, HRP (Product # A27036, 1:4000) using the iBright FL 1000 (Product # A32752). Chemiluminescent detection was performed using Novex® ECL Chemiluminescent Substrate Reagent Kit (Product # WP20005). 53BP1 is reported to pick up as a streak like pattern which is observed in the models tested here.



53BP1 Antibody (PA5-17578) in ICC/IF

Immunofluorescence analysis of TP53-binding protein 1 was performed using 70% confluent log phase HeLa cells. The cells were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton™ X-100 for 15 minutes, and blocked with 2% BSA for 45 minutes at room temperature. The cells were labeled with 53BP1 Polyclonal Antibody (Product # PA5-17578) at 1:100 in 0.1% BSA, incubated at 4 degree celsius overnight and then labeled with Donkey anti-Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor Plus 488 (Product # A32790), (1:2000), for 45 minutes at room temperature (Panel a: Blue). Nuclei (Panel b:Green) were stained with ProLong™ Diamond Antifade Mountant with DAPI (Product # P36962). F-actin (Panel c: Red) was stained with Rhodamine Phalloidin (Product # R415, 1:300). Panel d represents the merged image showing Nuclear localization. Panel e represents control cells with no primary antibody to assess background. The images were captured at 60X magnification.



53BP1 Antibody (PA5-17578) in ICC/IF

Immunofluorescent analysis of 53BP1 in HeLa cells using a 53BP1 polyclonal antibody (Product # PA5-17578) (green). Actin filaments are labeled with a fluorescent red phalloidin.

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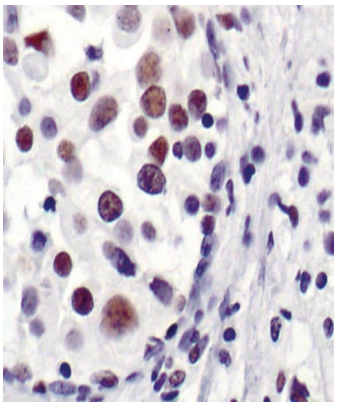


53BP1 Antibody (PA5-17578) in IHC (P)

Immunohistochemical analysis of 53BP1 in paraffin-embedded human breast carcinoma using a 53BP1 polyclonal antibody (Product # PA5-17578) showing nuclear localization.

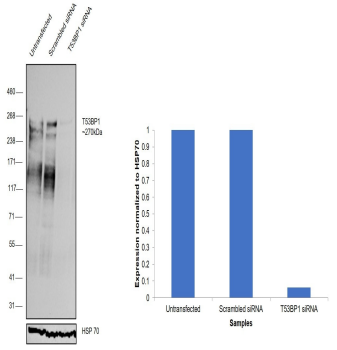
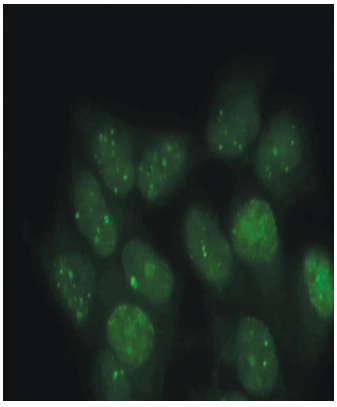


Fig. a Fig. b

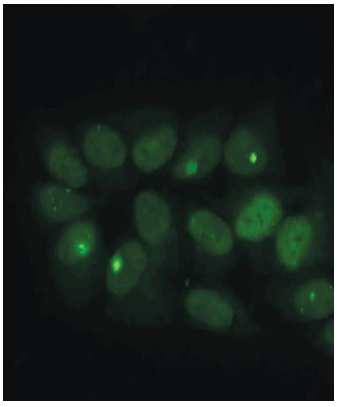
53BP1 Antibody (PA5-17578) in WB

Knockdown of TP53-binding protein 1 was achieved by transfecting HeLa with TP53-binding protein 1 specific siRNAs (Silencer® select Product # s14315, s14314). Western blot analysis (Fig. a) was performed using Nuclear enriched extracts from the TP53-binding protein 1 knockdown cells (lane 3), non-targeting scrambled siRNA transfected cells (lane 2) and untransfected cells (lane 1). The blot was probed with 53BP1 Polyclonal Antibody (Product # PA5-17578, 1:1000) and Goat anti-Rabbit IgG (Heavy Chain) Superclonal™ Recombinant Secondary Antibody, HRP (Product # A27036, 1:4000). Densitometric analysis of this western blot is shown in histogram (Fig. b). Decrease in signal upon siRNA mediated knock down confirms that antibody is specific to TP53-binding protein 1.53BP1 is reported to pick up in a streak like pattern as is seen here.



53BP1 Antibody (PA5-17578) in ICC/IF

Immunofluorescent analysis of 53BP1 using a polyclonal antibody (Product # PA5-17578).



53BP1 Antibody (PA5-17578) in ICC/IF

Immunofluorescent analysis of 53BP1 using a polyclonal antibody (Product # PA5-17578).

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PubMed References For 53BP1 Polyclonal Antibody

1 Western Blot References

Species / Dilution	Summary
	PA5-17578 was used in Western Blotting to investigate snake-venom-induced adaptive response at a relatively noncytotoxic concentration (0.01 µg/ml) in two human fibroblast cell lines of different origin, namely WI-38 fetal lung fibroblasts and BJ foreskin fibroblasts.
Human / 1:100	Journal of cellular physiology (2019; 234: 6147) **"Snake venoms promote stress-induced senescence in human fibroblasts."** Author(s):Lewinska A,Bocian A,Petrilla V,Adamczyk-Grochala J,Szymura K,Hendzel W,Kaleniuk E,Hus KK,Petrillova M, Wnuk M PubMed Article URL:http://dx.doi.org/10.1002/jcp.27382

1 Immunohistochemistry References

Species / Dilution	Summary
	PA5-17578 was used in Immunohistochemistry to find that DNMT2 may take part in the regulation of cell proliferation and longevity in human fibroblasts and speculate that the manipulation of DNMT2 levels that limits cell proliferation may be potentially useful anticancer strategy.
Human / 1:100	Redox biology (2018; 14: 20) **"Reduced levels of methyltransferase DNMT2 sensitize human fibroblasts to oxidative stress and DNA damage that is accompanied by changes in proliferation-related miRNA expression."** Author(s):Lewinska A,Adamczyk-Grochala J,Kwasniewicz E,Deregowska A,Semik E,Zabek T,Wnuk M PubMed Article URL:http://dx.doi.org/10.1016/j.redox.2017.08.012

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