# abcam

# Product datasheet

# Anti-53BP1 antibody ab21083

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#### Overview

Product name Anti-53BP1 antibody

**Description** Rabbit polyclonal to 53BP1

Host species Rabbit

Tested applications Suitable for: WB, IHC-P

Species reactivity Reacts with: Mouse, Human

**Immunogen** Synthetic peptide corresponding to Human 53BP1 (C terminal). Conjugated to a carrier protein.

Database link: Q12888

General notes

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

#### **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

Storage buffer pH: 7.00

Preservative: 0.025% Proclin 300

Constituents: 79% PBS, 20% Glycerol (glycerin, glycerine)

**Purity** Immunogen affinity purified

**Clonality** Polyclonal

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab21083 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

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Application	Abreviews	Notes
WB	**** <u>(2)</u>	1/500 - 1/3000. Detects a band of approximately 350 kDa (predicted molecular weight: 220 kDa).
IHC-P		1/1000 - 1/4000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

### **Target**

**Function** May have a role in checkpoint signaling during mitosis. Enhances TP53-mediated transcriptional activation. Plays a role in the response to DNA damage.

**Involvement in disease** Note=A chromosomal aberration involving TP53BP1 is found in a form of myeloproliferative

disorder chronic with eosinophilia. Translocation t(5;15)(q33;q22) with PDGFRB creating a TP53BP1-PDGFRB fusion protein.

**Sequence similarities**Contains 2 BRCT domains.

**Post-translational** Asymmetrically dimethylated on Arg residues by PRMT1. Methylation is required for DNA binding. **modifications** Phosphorylated at basal level in the absence of DNA damage. Hyper-phosphorylated in an ATM-

dependent manner in response to DNA damage induced by ionizing radiation. Hyper-phosphorylated in an ATR-dependent manner in response to DNA damage induced by UV

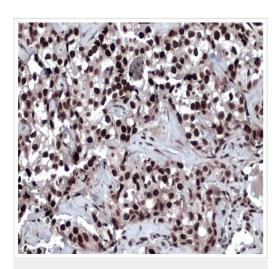
irradiation.

Cellular localization

Nucleus. Chromosome > centromere > kinetochore. Associated with kinetochores. Both nuclear

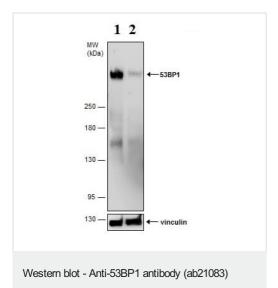
and cytoplasmic in some cells. Recruited to sites of DNA damage, such as double stand breaks. Methylation of histone H4 at 'Lys-20' is required for efficient localization to double strand breaks.

# **Images**



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-53BP1 antibody (ab21083)

Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue labeling 53BP1 with ab21083 at 1/4000 dilution. Nuclear staining is observed. Antigen Retrieval: Citrate buffer, pH 6.0, 15 min.



All lanes: Anti-53BP1 antibody (ab21083) at 1/500 dilution

Lane 1: 53BP1 shRNA non-transfected HeLa whole cell extracts

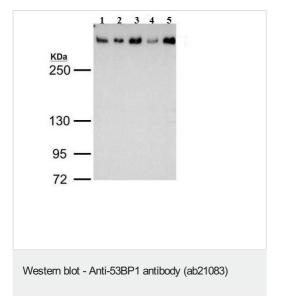
Lane 2: 53BP1 shRNA transfected HeLa whole cell extracts

Lysates/proteins at 50 µg per lane.

## Secondary

All lanes: HRP-conjugated anti-rabbit IgG antibody

Predicted band size: 220 kDa Observed band size: 350 kDa



5% SDS-PAGE

All lanes: Anti-53BP1 antibody (ab21083) at 1/2000 dilution

Lane 1 : 293T whole cell lysate/extract
Lane 2 : A431 whole cell lysate/extract
Lane 3 : HeLa whole cell lysate/extract
Lane 4 : HepG2 whole cell lysate/extract

Lane 5: A375 whole cell lysate/extract

Lysates/proteins at 30 µg per lane.

#### **Secondary**

All lanes: HRP-conjugated anti-rabbit lgG antibody

**Predicted band size:** 220 kDa **Observed band size:** 350 kDa

5% SDS-PAGE.

Running conditions: 80V, 15min; 140V, 40min.

Transfer condition: Semi-dry, 18 V, 60min (Nitrocellulose

membrane).

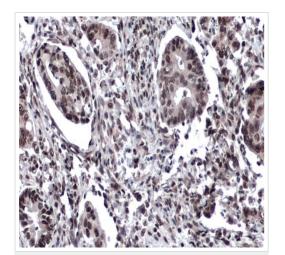
Blocking condition: 5% non-fat milk in TBST, RT, 60min.

Primary antibody incubation: 1/2000, 4?, overnight.

Secondary antibody incubation: Rabbit lgG antibody (HRP), 1/10,000, RT, 1hr.

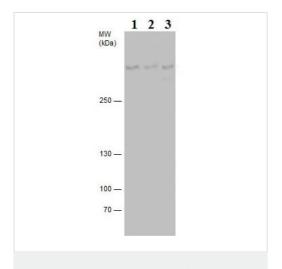
Washing condition: 5 ml TBST, 4 x 5min.

Exposure system: Trident plus Western HRP Substrate.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-53BP1 antibody (ab21083)

Immunohistochemical analysis of paraffin-embedded human colon cancer tissue labeling 53BP1 with ab21083 at 1/4000 dilution. Nuclear staining is observed. Antigen Retrieval: Citrate buffer, pH 6.0, 15 min.



Western blot - Anti-53BP1 antibody (ab21083)

All lanes: Anti-53BP1 antibody (ab21083) at 1/500 dilution

Lane 1 : Neuro2A whole cell extracts
Lane 2 : C8D30 whole cell extracts
Lane 3 : NIH-3T3 whole cell extracts

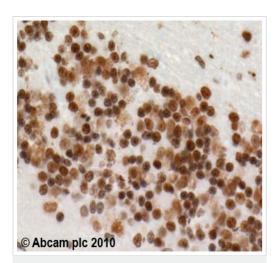
Lysates/proteins at 30 µg per lane.

### Secondary

All lanes: HRP-conjugated anti-rabbit lgG antibody

**Predicted band size:** 220 kDa **Observed band size:** 350 kDa

5% SDS-PAGE



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-53BP1 antibody (ab21083)

ab21083 (2µg/ml) staining 53BP1 in human Brain: Cerebellum using an automated system (DAKO Autostainer Plus). Using this protocol there is ubiquitous nuclear staining throughout.

Sections were rehydrated and antigen retrieved with the Dako 3 in 1 AR buffer EDTA pH 9.0 in a DAKO PT Link. Slides were peroxidase blocked in 3% H2O2 in methanol for 10 mins. They were then blocked with Dako Protein block for 10 minutes (containing casein 0.25% in PBS) then incubated with primary antibody for 20 min and detected with Dako Envision Flex amplification kit for 30 minutes. Colorimetric detection was completed with Diaminobenzidine for 5 minutes. Slides were counterstained with Haematoxylin and coverslipped under DePeX. Please note that, for manual staining, optimization of primary antibody concentration and incubation time is recommended. Signal amplification may be required.

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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