

Anti-PARP1 Antibody [SU03-68] - BSA and Azide free

HA750154



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC
Molecular Wt:	Predicted band size: 113 kDa
Clone number:	SU03-68

Description: The main role of PARP (found in the cell nucleus) is to detect and initiate an immediate cellular response to metabolic, chemical, or radiation-induced single-strand DNA breaks (SSB) by signaling the enzymatic machinery involved in the SSB repair. Once PARP detects a SSB, it binds to the DNA, undergoes a structural change, and begins the synthesis of a polymeric adenosine diphosphate ribose (poly (ADP-ribose) or PAR) chain, which acts as a signal for the other DNA-repairing enzymes. Target enzymes include DNA ligase III (LigIII), DNA polymerase beta (pol β), and scaffolding proteins such as X-ray cross-complementing gene 1 (XRCC1). After repairing, the PAR chains are degraded via Poly(ADP-ribose) glycohydrolase (PARG). PARP enzymes are essential in a number of cellular functions, including expression of inflammatory genes: PARP1 is required for the induction of ICAM-1 gene expression by cardiac myocytes and smooth muscle cells, in response to TNF.

Immunogen: Synthetic peptide within N-terminal human PARP1.

Positive control: HeLa cell lysate, Jurkat cell lysate, NIH/3T3 cell lysate, C2C12 cell lysate, C6 cell lysate, PC-12 cell lysate, HeLa whole cell lysate, HeLa treated with 1 μ M staurosporine for 3 hours whole cell lysate, HeLa, human breast cancer tissue, human colon tissue, mouse colon tissue, rat colon tissue.

Subcellular location: Nucleus, nucleolus, chromosome.

Database links: SwissProt: P09874 Human | P11103 Mouse | P27008 Rat

Recommended Dilutions:

WB	1:2,000
IF-Cell	1:500
IF-Tissue	1:1,000
IHC-P	1:4,000
FC	1:1,000

Storage Buffer: PBS (pH7.4).

Storage Instruction: Store at +4 $^{\circ}$ C after thawing. Aliquot store at -20 $^{\circ}$ C or -80 $^{\circ}$ C. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

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Orders:0086-571-88062880

Technical:0086-571-89986345

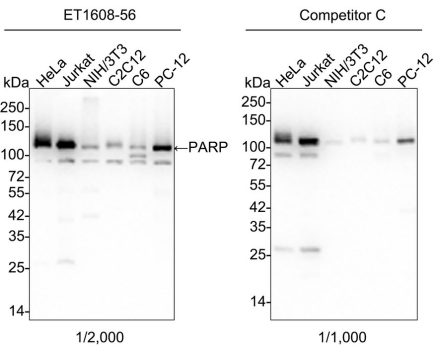
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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

Fig1: Western blot analysis of PARP1 on different lysates with Rabbit anti-PARP1 antibody (HA750154) at 1/2,000 dilution and competitor's antibody at 1/1,000 dilution.



Lane 1: HeLa cell lysate (15 µg/Lane)
Lane 2: Jurkat cell lysate (15 µg/Lane)
Lane 3: NIH/3T3 cell lysate (15 µg/Lane)
Lane 4: C2C12 cell lysate (15 µg/Lane)
Lane 5: C6 cell lysate (15 µg/Lane)
Lane 6: PC-12 cell lysate (15 µg/Lane)

Predicted band size: 113 kDa
Observed band size: 113 kDa

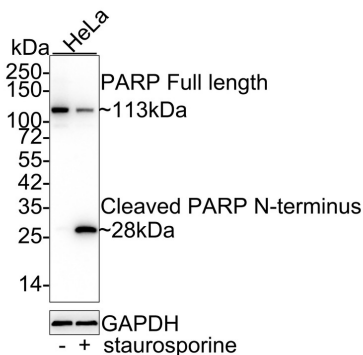
Exposure time: 6 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA750154) at 1/2,000 dilution and competitor's antibody at 1/1,000 dilution were used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

Fig2: Western blot analysis of PARP1 on different lysates with Rabbit anti-PARP1 antibody (HA750154) at 1/2,000 dilution.

Lane 1: HeLa whole cell lysate
Lane 2: HeLa treated with 1µM staurosporine for 3 hours whole cell lysate



Lysates/proteins at 20 µg/Lane.

Predicted band size: 113 kDa
Observed band size: 113/28 kDa

Exposure time: 1 minute 9 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA750154) at 1/2,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

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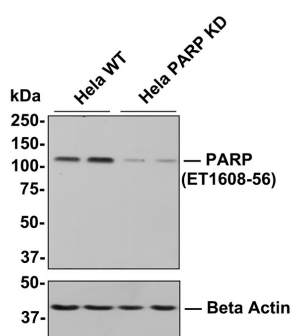


Fig3: All lanes: Western blot analysis of PARP with anti-PARP1 antibody[SU03-68] (HA750154) at 1:500 dilution.

Lane 1/2: Wild-type HeLa whole cell lysate (10 μ g).

Lane 3/4: PARP knockdown HeLa whole cell lysate (10 μ g).

HA750154 was shown to specifically react with PARP in wild-type HeLa cells. Weakened bands were observed when PARP knockdown samples were tested. Wild-type and PARP knockdown samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (HA750154, 1/500) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG-HRP Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.

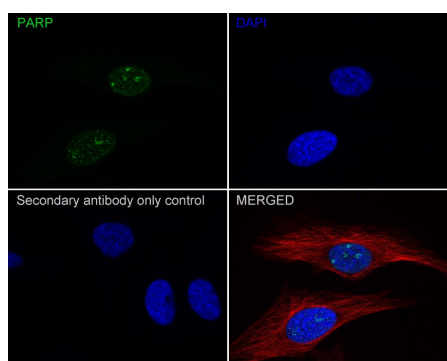


Fig4: Immunocytochemistry analysis of HeLa cells labeling PARP1 with Rabbit anti-PARP1 antibody (HA750154) at 1/500 dilution.

Cells were fixed in 100% methanol for 5 minutes, blocked with 2% normal goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-PARP1 antibody (HA750154) at 1/500 dilution in 2% normal goat serum overnight at 4 $^{\circ}$ C. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, green) was stained at 1/200 dilution overnight at +4 $^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluorTM 488, HA1125) was used as the secondary antibody at 1/800 dilution.

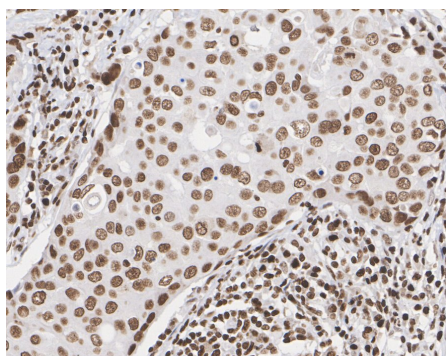


Fig5: Immunohistochemical analysis of paraffin-embedded human breast cancer tissue with Rabbit anti-PARP1 antibody (HA750154) at 1/4,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750154) at 1/4,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

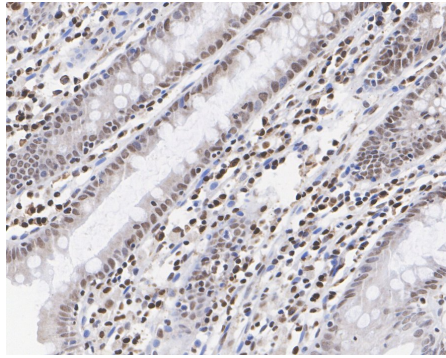


Fig6: Immunohistochemical analysis of paraffin-embedded human colon tissue with Rabbit anti-PARP1 antibody (HA750154) at 1/4,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750154) at 1/4,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

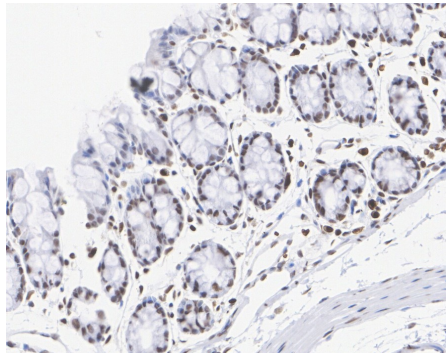


Fig7: Immunohistochemical analysis of paraffin-embedded mouse colon tissue with Rabbit anti-PARP1 antibody (HA750154) at 1/4,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750154) at 1/4,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

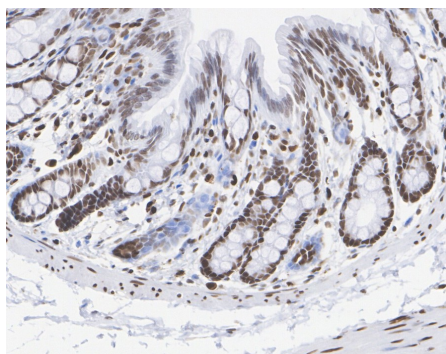


Fig8: Immunohistochemical analysis of paraffin-embedded rat colon tissue with Rabbit anti-PARP1 antibody (HA750154) at 1/4,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750154) at 1/4,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

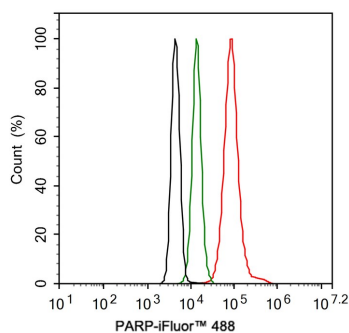


Fig9: Flow cytometric analysis of HeLa cells labeling PARP1.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA750154, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

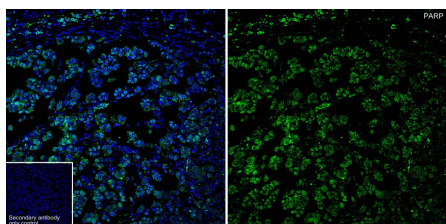


Fig10: Application: IF-Tissue

Species: Human

Site: breast cancer

Sample: Paraffin-embedded section

Antibody concentration: 1/1,000

Note: All products are “FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE”.

Background References

1. Cao C et al. The long intergenic noncoding RNA UFC1, a target of MicroRNA 34a, interacts with the mRNA stabilizing protein HuR to increase levels of -catenin in HCC cells. *Gastroenterology* 148:415-26.e18 (2015).
2. Gao S et al. Ischemia-reperfusion injury of the retina is linked to necroptosis via the ERK1/2-RIP3 pathway. *Mol Vis* 20:1374-87 (2014).

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