Anti-Parkin Antibody [JF82-09]

ET1702-60

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP, FC
Molecular Wt:	52 kDa
Clone number:	JF82-09

Description: Parkin is a zinc-finger protein that is related to ubiquitin at the amino terminus. The wild type Parkin gene, which maps to human chromosome 6q25.2-27, encodes a 465 amino acid full-length protein that is expressed as multiple isoforms. Mutations in the Parkin gene are responsible for autosomal recessive juvenile Parkinson's disease and commonly involve deletions of exons 3-5. In humans, Parkin is expressed in a subset of cells of the basal ganglia, midbrain, cerebellum and cerebral cortex, and is subject to alternative splicing in different tissues. Parkin expression is also high in the brainstem of mice, with the majority of immunopositive cells being neurons. The Parkin gene has been identified in a diverse group of organisms including mammals, birds, frog and fruit flies, suggesting that analogous functional roles of the Parkin protein may have been highly conserved during the course of evolution.

Immunogen: Synthetic peptide within N-terminal human Parkin.

Positive control: 293T cell lysates, SH-SY5Y, N2A, PC-3M, rat brain tissue, mouse testis tissue,Jurkat cell lysate,293 cell lysate.

Subcellular location: Mitochondrion, mitochondrion outer membrane, endoplasmic reticulum, cytosol, Nucleus, neuron projection, postsynaptic density, presynapse.

Database links: SwissProt: O60260 Human | Q9WVS6 Mouse | Q9JK66 Rat

Recommended Dilutions:

WB	1:500-1:1,000
IF-Cell	1:50-1:200
IF-Tissue	1:50-1:200
IHC-P	1:50-1:200
FC	1:500-1:1,000
IP	Use at an assay dependent concentration.

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

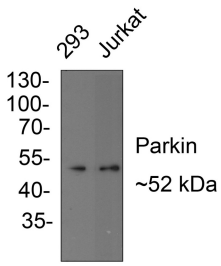

Fig1: Western blot analysis of Parkin on different lysates with Rabbit anti-Parkin antibody (ET1702-60) at 1/1,000 dilution.

Lane 1: 293 cell lysate
Lane 2: Jurkat cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 52 kDa
Observed band size: 52 kDa

Exposure time: 1 minutes;
10% 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 (ET1702-60) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.

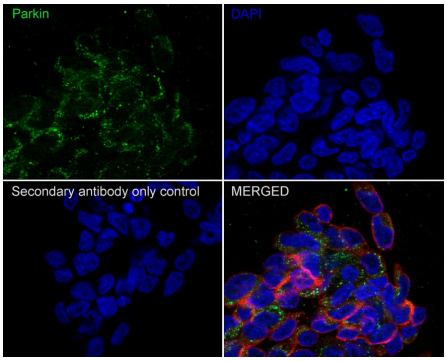

Fig2: Immunocytochemistry analysis of SH-SY5Y cells labeling Parkin with Rabbit anti-Parkin antibody (ET1702-60) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 ℃, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-Parkin antibody (ET1702-60) at 1/100 dilution in 2% negative goat serum overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/200 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

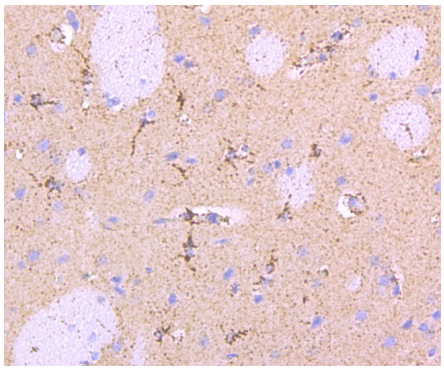

Fig3: Immunohistochemical analysis of paraffin-embedded rat brain tissue using anti-Parkin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1702-60, 1/50) for 30 minutes 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.

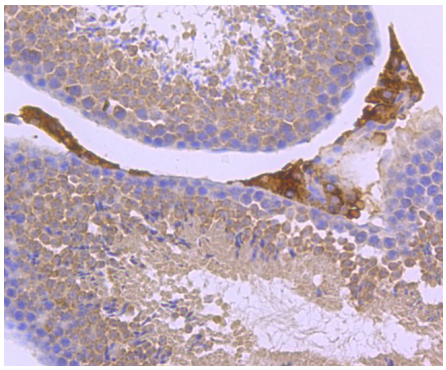

Fig4: Immunohistochemical analysis of paraffin-embedded mouse testis tissue using anti-Parkin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1702-60, 1/50) for 30 minutes 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.

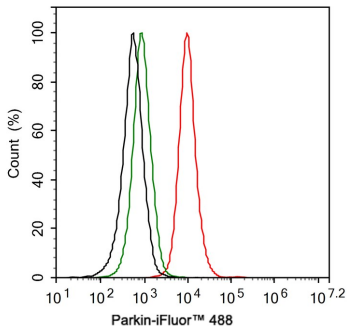

Fig5: Flow cytometric analysis of SH-SY5Y cells labeling Parkin.

Cells were fixed and permeabilized. Then stained with the primary antibody (ET1702-60, 1ug/ml) (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).

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

Background References

1. Lu Y et al. Beneficial effects of astragaloside IV against angiotensin II-induced mitochondrial dysfunction in rat vascular smooth muscle cells. Int J Mol Med 36:1223-32 (2015).
2. Seillier M et al. Defects in mitophagy promote redox-driven metabolic syndrome in the absence of TP53INP1. EMBO Mol Med 7:802-18 (2015).