


Product datasheet

Anti-LRRK2 antibody [MJFF2 (c41-2)] ab133474

KO **VALIDATED** Recombinant RabMAB

★★★★★ [2 Abreviews](#) [106 References](#) [5 Images](#)

Overview

Product name	Anti-LRRK2 antibody [MJFF2 (c41-2)]
Description	Rabbit monoclonal [MJFF2 (c41-2)] to LRRK2
Host species	Rabbit
Tested applications	Suitable for: WB, IP Unsuitable for: ICC/IF
Species reactivity	Reacts with: Mouse, Human Predicted to work with: Rat 
Immunogen	Recombinant fragment within Human LRRK2 aa 950 to the C-terminus. The exact sequence is proprietary. Database link: Q5S007
Positive control	WB: A549 and MEF cell lysates; HEK293 cells transfected with LRRK2 cell lysate. IP: Mouse cerebral cortex, A549 .
General notes	<p>Well-characterized antibodies to efficiently detect and purify LRRK2 protein are a critical need in the Parkinson's Disease (PD) research community. To help accelerate LRRK2 research, The Michael J. Fox Foundation (MJFF), working with Abcam, has generated unique and high quality LRRK2 rabbit monoclonal antibodies to be widely available for PD research community.</p> <p>LRRK2 (Leucine-rich repeat kinase 2, dardarin) is a protein kinase belonging to the ROCO family, which is defined by the presence of a ROC (Ras/GTPase of complex proteins) domain and COR (C-terminal of Roc) region. LRRK2 exhibits kinase activity whereby it can undergo autophosphorylation and can phosphorylate generic substrates. In addition, the GTPase domain of LRRK2 can mediate GDP (guanosine-5'-diphosphate)/GTP (guanosine-5'-triphosphate) binding as well as GTP hydrolysis.</p> <p>LRRK2 is mutated in a significant number of Parkinson's disease (PD) patients. Mutations in this gene account for 4% of PD, and are observed in 1% of sporadic PD patients. Clinical symptoms of patients carrying PD-associated mutations of LRRK2 are indistinguishable from typical sporadic PD. The spectra of neuropathological features of PARK8 (type 8), the type corresponding to LRRK2, is broad and appears to encompass those associated with other familial PD cases such as PARK1 (alpha-synuclein) and PARK2 (Parkin). Patients with this gene mutation have typical relatively late onset Parkinsonism with features comparable with idiopathic PD; symptoms include asymmetric rest tremor, bradykinesia, rigidity, and a good response to</p>

3,4-dihydroxy-L-phenylalanine (L-DOPA). The pathology of cases with LRRK2 mutations is pleomorphic.

For more characterization data and protocols using this LRRK2 Antibody, please refer to Davies, et al. 2013. Biochemical J 453(1):101-113 [PMID: 23560750]

Abcam recommended secondaries - Goat Anti-Rabbit HRP ([ab205718](#)) and Goat Anti-Rabbit Alexa Fluor® 488 ([ab150077](#)). Or search our wide range of secondary antibodies for use with your experiment.

This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free production

For more information [see here](#).

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to [RabMAb® patents](#).

This antibody was developed with support from The Michael J. Fox Foundation.



Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 40% Glycerol (glycerin, glycerine), 0.05% BSA, 59% PBS
Purity	Protein A purified
Clonality	Monoclonal
Clone number	MJFF2 (c41-2)
Isotype	IgG

Applications

The Abpromise guarantee Our [Abpromise guarantee](#) covers the use of ab133474 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	★★★★★ (2)	1/10000 - 1/50000. Predicted molecular weight: 286 kDa.
IP		Use at an assay dependent concentration. (2-5 µg)

Application notes Is unsuitable for ICC/IF.

Target

Function

Positively regulates autophagy through a calcium-dependent activation of the CaMKK/AMPK signaling pathway. The process involves activation of nicotinic acid adenine dinucleotide phosphate (NAADP) receptors, increase in lysosomal pH, and calcium release from lysosomes. Together with RAB29, plays a role in the retrograde trafficking pathway for recycling proteins, such as mannose 6 phosphate receptor (M6PR), between lysosomes and the Golgi apparatus in a retromer-dependent manner. Regulates neuronal process morphology in the intact central nervous system (CNS). Plays a role in synaptic vesicle trafficking. Phosphorylates PRDX3. Has GTPase activity. May play a role in the phosphorylation of proteins central to Parkinson disease.

Tissue specificity

Expressed in the brain. Expressed in pyramidal neurons in all cortical laminae of the visual cortex, in neurons of the substantia nigra pars compacta and caudate putamen (at protein level). Expressed throughout the adult brain, but at a lower level than in heart and liver. Also expressed in placenta, lung, skeletal muscle, kidney and pancreas. In the brain, expressed in the cerebellum, cerebral cortex, medulla, spinal cord occipital pole, frontal lobe, temporal lobe and putamen. Expression is particularly high in brain dopaminergic areas.

Involvement in disease

Parkinson disease 8

Sequence similarities

Belongs to the protein kinase superfamily. TKL Ser/Thr protein kinase family.

Contains 12 LRR (leucine-rich) repeats.

Contains 1 protein kinase domain.

Contains 1 Roc domain.

Contains 7 WD repeats.

Domain

The seven-bladed WD repeat region is critical for synaptic vesicle trafficking and mediates interaction with multiple vesicle-associated presynaptic proteins.

The Roc domain mediates homodimerization and regulates kinase activity.

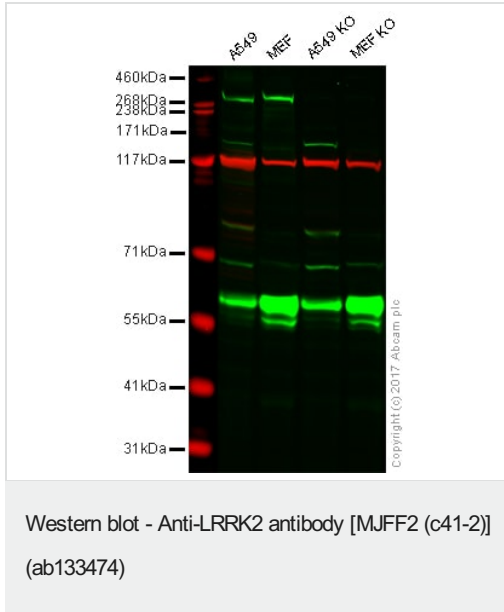
Post-translational modifications

Autophosphorylated.

Cellular localization

Membrane. Cytoplasm. Perikaryon. Mitochondrion. Golgi apparatus. Cell projection, axon. Cell projection, dendrite. Endoplasmic reticulum. Cytoplasmic vesicle, secretory vesicle, synaptic vesicle membrane. Endosome. Lysosome. Mitochondrion outer membrane. Mitochondrion inner membrane. Mitochondrion matrix. Predominantly associated with intracytoplasmic vesicular and membranous structures (By similarity). Localized in the cytoplasm and associated with cellular membrane structures. Predominantly associated with the mitochondrial outer membrane of the mitochondria. Colocalized with RAB29 along tubular structures emerging from Golgi apparatus. Localizes in intracytoplasmic punctate structures of neuronal perikarya and dendritic and axonal processes.

Images



Lane 1: Wild type A549 whole cell lysate (20 µg)

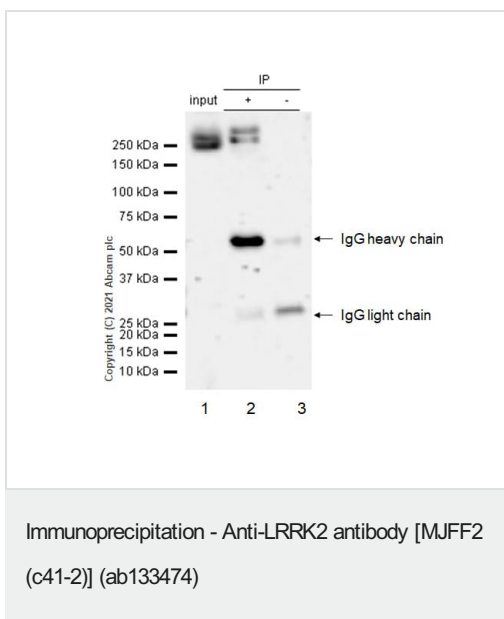
Lane 2: Wild type MEF whole cell lysate (20 µg)

Lane 3: LRRK2 knockout A549 whole cell lysate (20 µg)

Lane 4: LRRK2 knockout MEF whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab133474 observed at 286 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

ab133474 was shown to recognize LRRK2 in wild type A549 and MEF cells along with additional cross reactive bands. Whilst signal was not seen in LRRK2 knockout cells. Wild-type and LRRK2 knockout samples were subjected to SDS-PAGE. Ab133474 and **ab18058** (Mouse anti Vinculin loading control) were incubated overnight at 4°C at 10000 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed **ab216776** secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging. Wild-type and LRRK2 knockout MEF and A549 cells were provided as a generous gift from Professor Dario Alessi, MRC Protein Phosphorylation and Ubiquitination Unit (University of Dundee).



LRRK2 was immunoprecipitated from 0.35 mg A549 (Human lung carcinoma epithelial cell) whole cell lysate 10 µg with 133474 at 1/60 dilution (2µg). VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/5000 dilution.

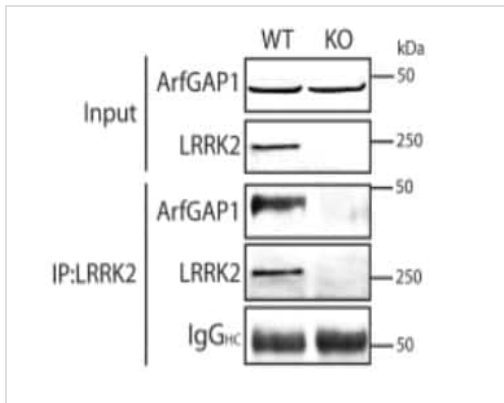
Lane 1: A549 (Human lung carcinoma epithelial cell) whole cell lysate 10 µg

Lane 2: ab133474 IP in A549 whole cell lysate

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab133474 in A549 whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDm/TBST.

Fresh lysate should be used to minimize protein degradation.

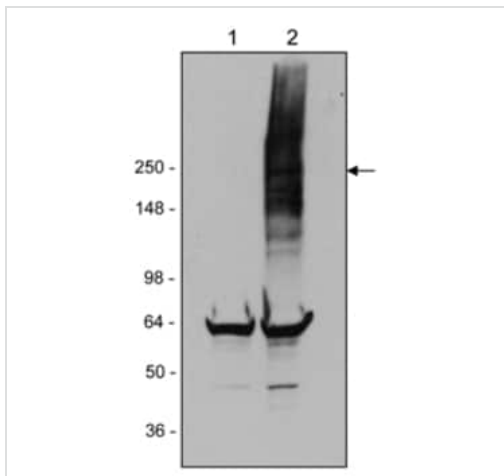


Immunoprecipitation - Anti-LRRK2 antibody [MJFF2 (c41-2)] (ab133474)

Stafa K. et al., PLoS Genet. 2012;8(2):e1002526. Fig 1
doi: 10.1371/journal.pgen.1002526. Reproduced under
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Immunoprecipitation to verify the interaction of LRRK2 and ArfGAP1 *in vivo*. LRRK2 interacts with ArfGAP1 in brain extracts derived from wild-type mice following immunoprecipitation with ab133474, a LRRK2-specific monoclonal antibody (MJFF-2/c41-2), whereas ArfGAP1 is not immunoprecipitated in extracts derived from LRRK2 knockout mice

Protein extracts were prepared from the cerebral cortex of adult wild-type and LRRK2 knockout mice (with targeted deletion of exon 41 of the LRRK2 gene) by homogenization in TNE buffer (10 mM Tris-HCL pH 7.4, 150 mM NaCl, 5 mM EDTA, 0.5% NP-40, 1× phosphatase inhibitor cocktail 1 and 2, 1× Complete Mini protease inhibitor cocktail). Protein concentration was determined by BCA assay. Brain extracts (10 mg protein) were combined with 50 µl Protein G-Dynabeads pre-incubated with rabbit anti-LRRK2 (5 µg; MJFF2/c41-2; Abcam, Inc.), rabbit anti-ArfGAP1 (3 µg) or rabbit IgG (3 µg) antibodies followed by overnight incubation at 4°C. Dynabead complexes were sequentially washed twice with TNE buffer and twice with TBS buffer (10 mM Tris-HCL pH 7.4, 150 mM NaCl). Immunoprecipitates were eluted by heating at 70°C for 10 min, resolved by SDS-PAGE and subjected to Western blot analysis.



Western blot - Anti-LRRK2 antibody [MJFF2 (c41-2)] (ab133474)

This image is courtesy of Zhuohua Zhang Lab
(Sanford-Burnham Medical Research Institute)

All lanes : Anti-LRRK2 antibody [MJFF2 (c41-2)] (ab133474) at 1/10000 dilution

Lane 1 : HEK293 cell lysate transfected with 3*Flag vector

Lane 2 : HEK293 cell lysate transfected with 3*Flag full length wild type LRRK2

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : HRP labelled goat anti-rabbit at 1/2000 dilution

Predicted band size: 286 kDa

Why choose a recombinant antibody?

<p>Research with confidence Consistent and reproducible results</p>	<p>Long-term and scalable supply Recombinant technology</p>
<p>Success from the first experiment Confirmed specificity</p>	<p>Ethical standards compliant Animal-free production</p>

Anti-LRRK2 antibody [MJFF2 (c41-2)] (ab133474)

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