abcam

Product datasheet

Anti-LRRK2 antibody [MJFF3 (c69-6)] - BSA and Azide free ab183216

KO VALIDATED Recombinant RabMAb

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Overview

Product name	Anti-LRRK2 antibody [MJFF3 (c69-6)] - BSA and Azide free
Description	Rabbit monoclonal [MJFF3 (c69-6)] to LRRK2 - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** IHC-P, WB
Species reactivity	**Reacts with:** Mouse, Human **Predicted to work with:** Rat
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	IHC-P: Human brain hippocampus tissue WB: Jurkat cells transfected with LRRK2
General notes	ab183216 is the carrier-free version of **ab133475**. 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 Epitomics, Inc., has generated unique and high quality LRRK2 rabbit monoclonal antibodies to be widely available for PD research community. 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. 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 3,4-dihyroxy-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]

Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

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.

THE MICHAEL J. FOX FOUNDATION
FOR PARKINSON'S RESEARCH

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	MJFF3 (c69-6)
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab183216 in the following tested applications.

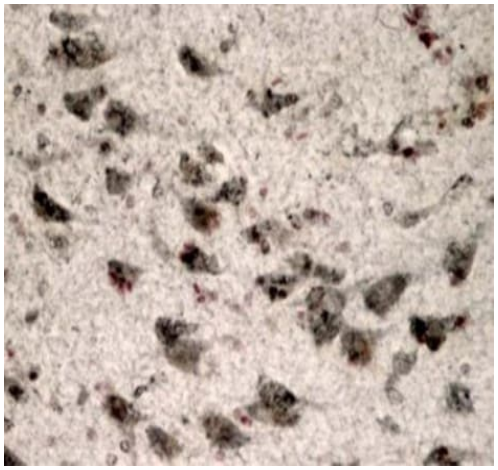
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB		Use at an assay dependent concentration. Predicted molecular weight: 286 kDa. Please check the parent abID, **ab133475**, for a recommended dilution.

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 dopaminoceptive 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

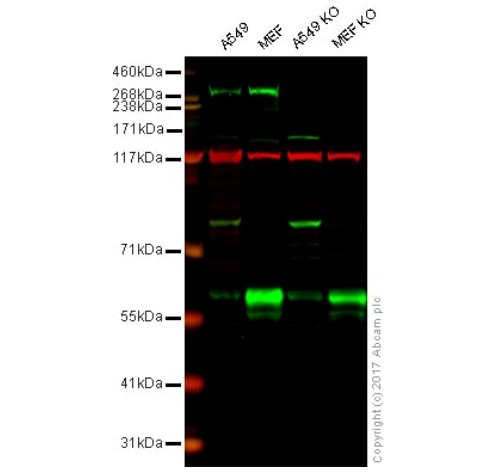


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-LRRK2 antibody [MJFF3 (c69-6)] - BSA and Azide free (ab183216)

This IHC data was generated using the same anti-LRRK2 antibody clone, MJFF3 (c69-6), in a different buffer formulation (cat# **ab133475**).

ab133475, at 1/200 dilution, staining LRRK2 in paraffin embedded Human hippocampus tissue using immunohistochemical analysis.

Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-LRRK2 antibody [MJFF3 (c69-6)] - BSA and Azide free (ab183216)

This WB data was generated using the same anti-LRRK2 antibody clone, MJFF3 (c69-6), in a different buffer formulation (cat# **ab133475**).

Lane 1: A549 whole cell lysate (20 µg)
Lane 2: MEF whole cell lysate (20 µg)
Lane 3: A549 (KO) whole cell lysate (20 µg)
Lane 4: MEF (KO) whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - **ab133475** observed at 286 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

ab133475 was shown to specifically react with Wild type A549 and MEF cells whilst no band was observed when knockout samples were used. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and knockout samples were subjected to SDS-PAGE. Ab133475 and **ab18058** (Mouse anti Vinculin loading control) were incubated overnight at 4°C at 1000 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.

Western blot - Anti-LRRK2 antibody [MJFF3 (c69-6)] - BSA and Azide free (ab183216)

Anti-LRRK2 antibody [MJFF3 (c69-6)] - BSA and Azide free (ab183216) + NIH/3T3 (mouse embryo) whole cell lysate at 10 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**)

Predicted band size: 286 kDa

Exposure time: 10 seconds

Blocking buffer and concentration: 5% NFDM/TBST

Diluting buffer and concentration: 5% NFDM/TBST

Anti-LRRK2 antibody [MJFF3 (c69-6)] - BSA and Azide free (ab183216)

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