

Anti-PGP9.5 antibody ab27053



★★★★☆ 6 Abreviews 10 References 8 图像

概述

产品名称	Anti-PGP9.5抗体
描述	兔多克隆抗体to PGP9.5
宿主	Rabbit
特异性	From Jan 2024, QC testing of replenishment batches of this polyclonal changed. All tested and expected application and reactive species combinations are still covered by our Abcam product promise. However, we no longer test all applications. For more information on a specific batch, please contact our Scientific Support who will be happy to help.
经测试应用	适用于: IHC-P, IP, WB, ICC/IF
种属反应性	与反应: Mouse, Rat, Human
免疫原	Synthetic peptide corresponding to Human PGP9.5 aa 150 to the C-terminus conjugated to keyhole limpet haemocyanin.
阳性对照	ICC/IF: PC12 cells and primary rat neurons/glia, DIV14 cells. WB: Wild type HAP1 whole cell lysate. HEK-293 whole cell lysate. Human, rat and mouse brain whole cell lysate. IHC-P: Human pancreas tissue. IP: Mouse brain tissue lysate.
常规说明	<p>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.</p> <p>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</p>

性能

形式	Liquid
存放说明	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.
存储溶液	pH: 7.40 Preservative: 0.02% Sodium azide Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising agent. If you would like information about the formulation of a specific lot, please contact our scientific support team who will be happy to help.

纯度	Immunogen affinity purified
克隆	多克隆
同种型	IgG

应用

The Abpromise guarantee Abpromise™ 承诺保证使用ab27053于以下的经测试应用

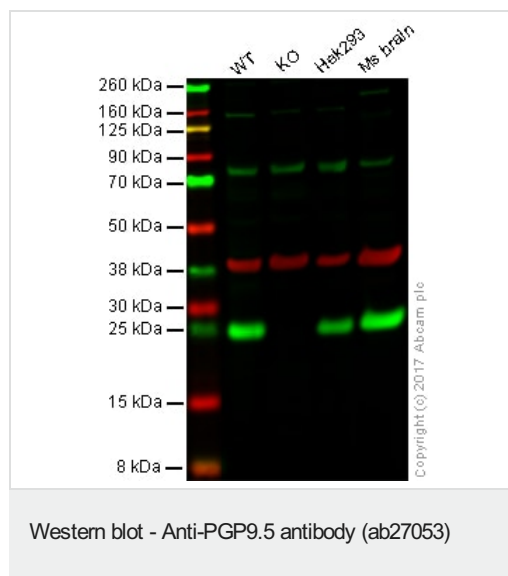
“应用说明”部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
IHC-P	★★★★★ (3)	Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
IP		Use a concentration of 5 µg/ml.
WB	★★★★★ (1)	Use a concentration of 1 µg/ml. Detects a band of approximately 25 kDa (predicted molecular weight: 25 kDa). Abcam recommends using milk as the blocking agent. Abcam welcomes customer feedback and would appreciate any comments regarding this product and the data presented below.
ICC/IF	★★★★★ (1)	Use a concentration of 1 µg/ml.

靶标

功能	Ubiquitin-protein hydrolase involved both in the processing of ubiquitin precursors and of ubiquitinated proteins. This enzyme is a thiol protease that recognizes and hydrolyzes a peptide bond at the C-terminal glycine of ubiquitin. Also binds to free monoubiquitin and may prevent its degradation in lysosomes. The homodimer may have ATP-independent ubiquitin ligase activity.
组织特异性	Found in neuronal cell bodies and processes throughout the neocortex (at protein level). Expressed in neurons and cells of the diffuse neuroendocrine system and their tumors. Weakly expressed in ovary. Down-regulated in brains from Parkinson disease and Alzheimer disease patients.
疾病相关	Parkinson disease 5 Neurodegeneration with optic atrophy, childhood-onset
序列相似性	Belongs to the peptidase C12 family.
翻译后修饰	O-glycosylated.
细胞定位	Cytoplasm. Endoplasmic reticulum membrane. About 30% of total UCHL1 is associated with membranes in brain.

图片



Lane 1: Wild type HAP1 whole cell lysate (20 µg)

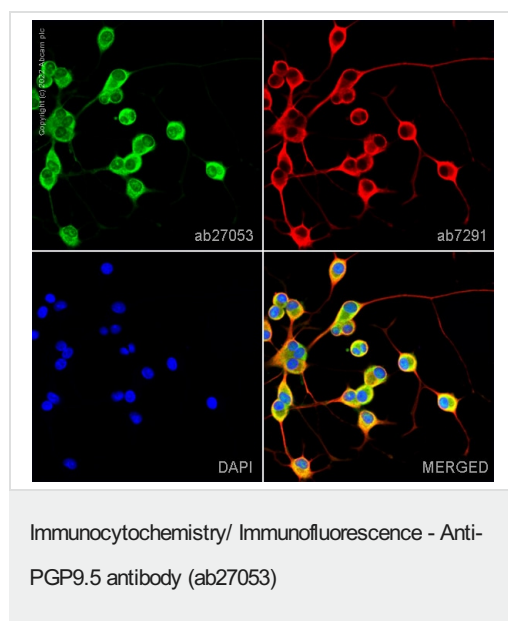
Lane 2: UCHL1 (PGP9.5) knockout HAP1 whole cell lysate (20 µg)

Lane 3: Hek293 whole cell lysate (20 µg)

Lane 4: Ms brain whole cell lysate (20 µg)

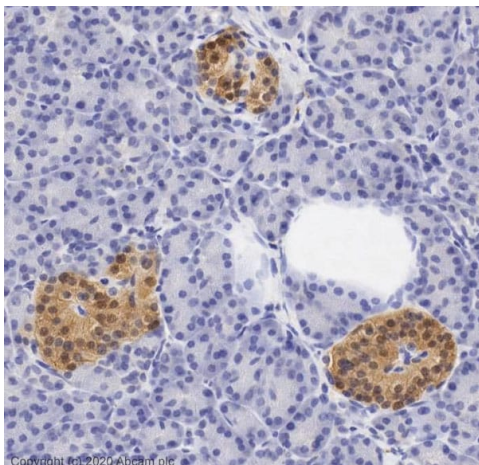
Lanes 1 - 4: Merged signal (red and green). Green - ab27053 observed at 24 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab27053 was shown to recognize UCHL1 (PGP9.5) in wild type cells as signal was lost at the expected MW in UCHL1 (PGP9.5) knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and UCHL1 (PGP9.5) knockout samples were subjected to SDS-PAGE. Ab27053 and **ab8245** (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1 µg/ml 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.



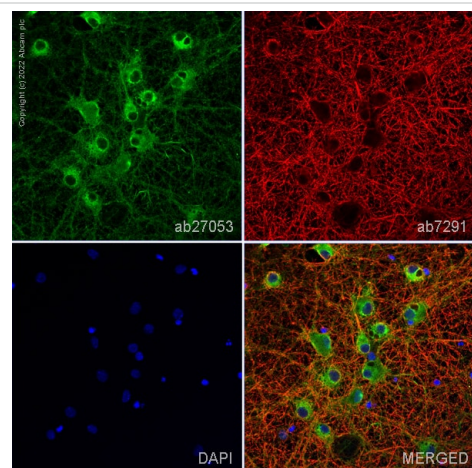
ab27053 staining PGP9.5 in PC12 cells. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% PBS-Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated overnight at 4°C with ab27053 at 1µg/ml and **ab7291**, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with **ab150081**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and **ab150120**, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 594), pre-adsorbed at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody (ab27053)

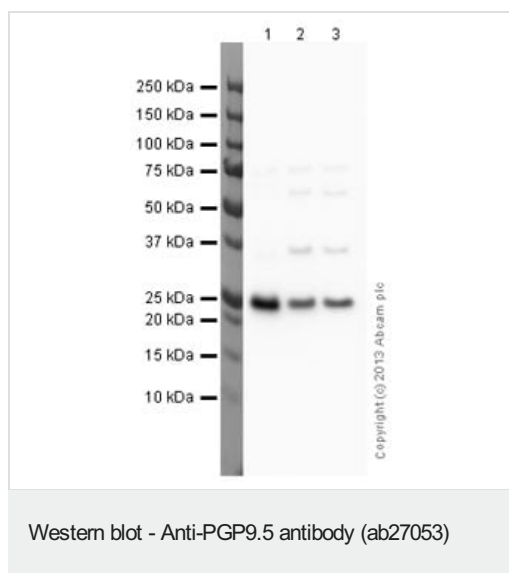
IHC image of Anti-PGP9.5 antibody staining in a section of formalin-fixed paraffin-embedded normal human pancreas performed on a Leica BOND™ system using the standard protocol. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 minutes. The section was then incubated with ab27053, 1 µg/mL, for 15 minutes at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.



Immunocytochemistry/ Immunofluorescence - Anti-PGP9.5 antibody (ab27053)

ab27053 staining PGP9.5 in primary rat neurons/glia, DIV14 (prepared from E18 rat hippocampal brain area, obtained from Transnetix Tissue by BrainBits, LLC, cat.no. SDHEP) cells. The cells were fixed with 4% paraformaldehyde (10 min), permeabilized with 0.1% PBS-Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at 4°C with ab27053 at 5µg/ml and **ab7291**, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with **ab150081**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and **ab150120**, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 594), pre-adsorbed at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.



All lanes : Anti-PGP9.5 antibody (ab27053) at 1 µg/ml

Lane 1 : Human brain tissue lysate - total protein ([ab29466](#))

Lane 2 : Brain (Mouse) Tissue Lysate ([ab27253](#))

Lane 3 : Brain (Rat) Tissue Lysate ([ab7942](#))

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 25 kDa

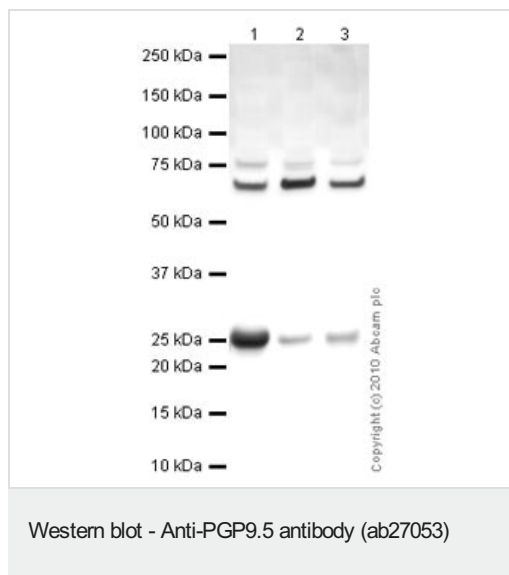
Observed band size: 25 kDa

Additional bands at: 37 kDa (possible non-specific binding), 60 kDa (possible non-specific binding), 75 kDa (possible non-specific binding)

Exposure time: 10 seconds

This blot was produced using a 4-12% Bis-tris gel under the MES buffer system. The gel was run at 200V for 35 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 3% Milk before being incubated with abX overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and visualised using ECL development solution.

Abcam recommends using milk as the blocking agent.



All lanes : Anti-PGP9.5 antibody (ab27053) at 1 µg/ml

Lane 1 : Human brain tissue lysate - total protein ([ab29466](#))

Lane 2 : HEK293 (Human embryonic kidney cell line) Whole Cell Lysate

Lane 3 : Brain (Mouse) Tissue Lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Developed using the ECL technique.

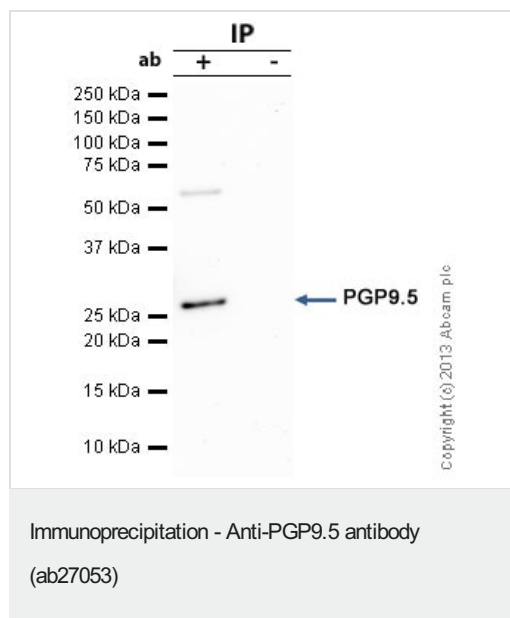
Performed under reducing conditions.

Predicted band size: 25 kDa

Observed band size: 25 kDa

Additional bands at: 65 kDa. We are unsure as to the identity of these extra bands.

Exposure time: 1 minute



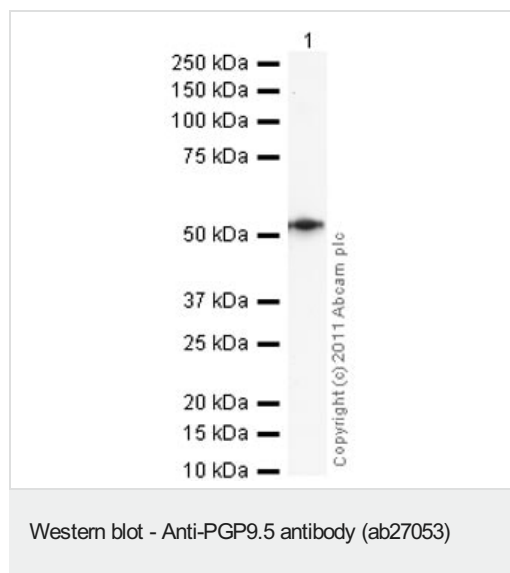
PGP9.5 was immunoprecipitated using 0.5mg Mouse Brain tissue lysate, 5µg of Rabbit polyclonal to PGP9.5 and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).

The antibody was incubated under agitation with Protein G beads for 10min, Mouse Brain tissue lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab27053.

Secondary: Clean-Blot IP Detection Reagent (HRP) at 1/500 dilution.

Band: 26kDa, non specific band - 65kDa: We are unsure as to the identity of this extra band; PGP9.5



Anti-PGP9.5 antibody (ab27053) at 1/250 dilution + Recombinant Human PGP9.5 protein ([ab82628](#)) at 0.01 µg

Secondary

Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) ([ab65484](#)) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 25 kDa

Exposure time: 3 minutes

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