

## Product datasheet

# Anti-PGP9.5 antibody [EPR4118] ab108986

**KO VALIDATED** Recombinant RabMAb

★★★★★ 10 Abreviews 38 References 19 Images

### Overview

<b>Product name</b>	Anti-PGP9.5 antibody [EPR4118]
<b>Description</b>	Rabbit monoclonal [EPR4118] to PGP9.5
<b>Host species</b>	Rabbit
<b>Tested applications</b>	<b>Suitable for:</b> ICC/IF, Flow Cyt (Intra), IHC-Fr, WB, IP, IHC-P
<b>Species reactivity</b>	<b>Reacts with:</b> Mouse, Rat, Human
<b>Immunogen</b>	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
<b>Positive control</b>	WB: Fetal brain, Y79, U87-MG, SH-SY5Y, HAP1, HeLa and HEK-293T cell lysates; IHC-P: Human glioma, colon, and hepatocellular carcinoma tissue, mouse colon, mouse cerebral cortex tissue, rat Jejunum and cerebral cortex tissue; ICC/IF: Neuro-2a cells; IP: Human fetal brain lysate; Flow Cyt (intra): SH-SY5Y and Y79 cells; IHC-Fr: Mouse cerebrum tissue.
<b>General notes</b>	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> <li>- High batch-to-batch consistency and reproducibility</li> <li>- Improved sensitivity and specificity</li> <li>- Long-term security of supply</li> <li>- Animal-free production</li> </ul> <p>For more information <a href="#">see here</a>.</p> <p>Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <a href="#">RabMAb<sup>®</sup> patents</a>.</p> <p><b>We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.</b></p>

### Properties

<b>Form</b>	Liquid
<b>Storage instructions</b>	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Stable for 12 months at -20°C.
<b>Storage buffer</b>	pH: 7.20

Preservative: 0.01% Sodium azide  
Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.5% BSA

**Purity** Protein A purified  
**Clonality** Monoclonal  
**Clone number** EPR4118  
**Isotype** IgG

## Applications

**The Abpromise guarantee** Our [Abpromise guarantee](#) covers the use of ab108986 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
ICC/IF		1/500.
Flow Cyt (Intra)		1/100 - 1/10000. <a href="#">ab172730</a> - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IHC-Fr		1/250. Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20)
WB		1/1000 - 1/10000. Detects a band of approximately 25 kDa (predicted molecular weight: 24 kDa).
IP		1/10 - 1/100.
IHC-P	★★★★★ (10)	1/250 - 1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

## Target

**Function** 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.

**Tissue specificity** 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.

**Involvement in disease** Parkinson disease 5  
Neurodegeneration with optic atrophy, childhood-onset

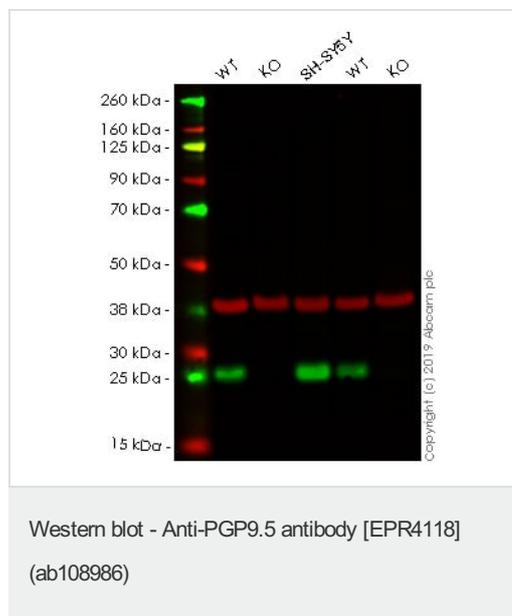
**Sequence similarities** Belongs to the peptidase C12 family.

**Post-translational modifications** O-glycosylated.

## Cellular localization

Cytoplasm. Endoplasmic reticulum membrane. About 30% of total UCHL1 is associated with membranes in brain.

## Images



**All lanes** : Anti-PGP9.5 antibody [EPR4118] (ab108986) at 1/1000 dilution

**Lane 1** : Wild-type Hap1 cell lysate

**Lane 2** : UCHL1 knockout Hap1 cell lysate

**Lane 3** : SH-SY5Y cell lysate

**Lane 4** : Wild-type HEK-293T cell lysate

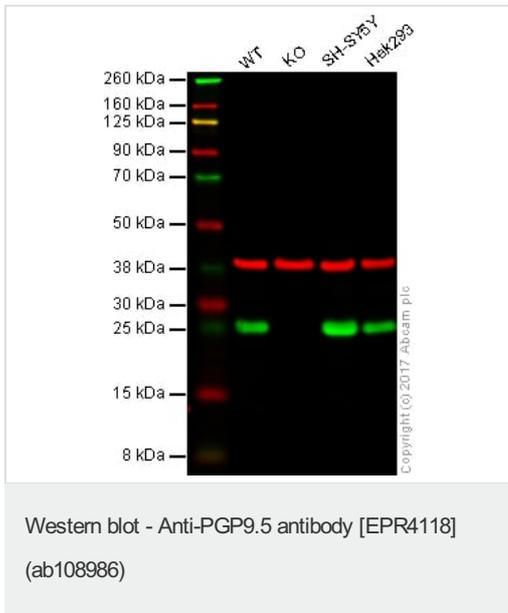
**Lane 5** : UCHL1 knockout HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

**Predicted band size:** 24 kDa

**Lanes 1 - 5:** Merged signal (red and green). Green - ab108986 observed at 25 kDa. Red - loading control, ab8245 observed at 37 kDa.

ab108986 was shown to react with PGP9.5 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line ab255443 (knockout cell lysate ab263773) was used. Wild-type and PGP9.5 knockout samples were subjected to SDS-PAGE. ab108986 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4 °C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



**All lanes** : Anti-PGP9.5 antibody [EPR4118] (ab108986) at 1/1000 dilution

**Lane 1** : Wild-type HAP1 whole cell lysate

**Lane 2** : UCHL1 knockout HAP1 whole cell lysate

**Lane 3** : SH-SY5Y whole cell lysate

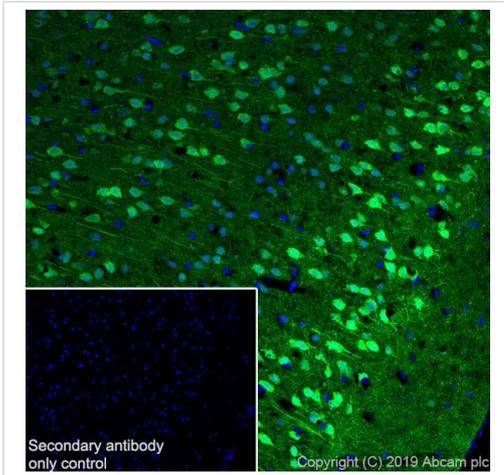
**Lane 4** : HEK293 whole cell lysate

Lysates/proteins at 20 µg per lane.

**Predicted band size:** 24 kDa

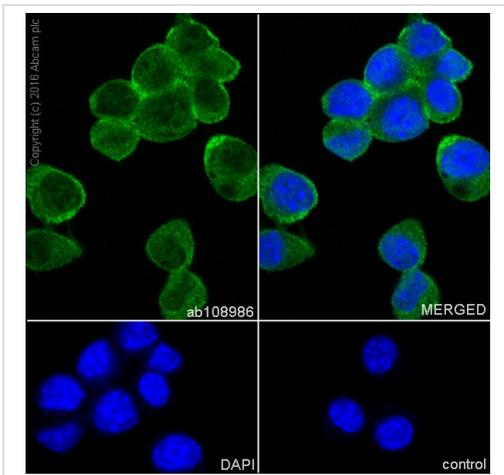
**Lanes 1 - 4:** Merged signal (red and green). Green - ab108986 observed at 24 kDa. Red - loading control, [ab8245](#), observed at 37 kDa.

Ab108986 was shown to specifically react with UCHL1 in wild-type cells as signal was lost in UCHL1 knockout HAP1 cells. Wild-type and UCHL1 knockout samples were subjected to SDS-PAGE. Ab108986 and [ab8245](#) (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 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.



Immunohistochemistry (Frozen sections) - Anti-PGP9.5 antibody [EPR4118] (ab108986)

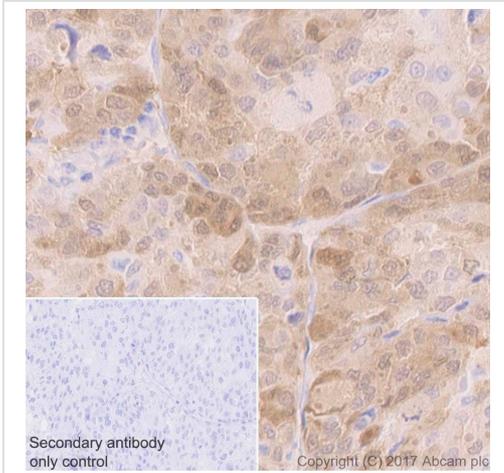
Immunohistochemistry (Frozen sections) analysis of mouse cerebrum tissue sections labeling PGP9.5 with Purified ab108986 at 1/250 (0.5 µg/ml). Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. DAPI was used as a counterstain.



Immunocytochemistry/ Immunofluorescence - Anti-PGP9.5 antibody [EPR4118] (ab108986)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Neuro-2a (Mouse neuroblastoma cell line) cells labeling PGP9.5 with ab108986 at 1/500 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining on Neuro-2a cell line. The nuclear counter stain is DAPI (blue).

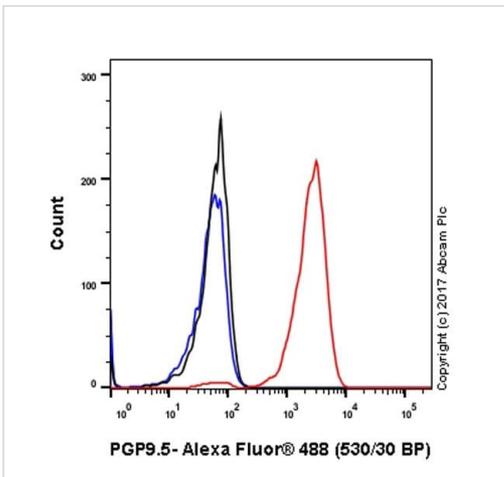
The negative control is PBS only.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] (ab108986)

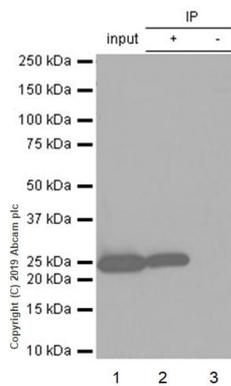
Immunohistochemical analysis of paraffin-embedded human hepatocellular carcinoma tissue labeling PGP9.5 with ab108986, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining on human hepatocellular carcinoma. The section was incubated with ab229902 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND<sup>®</sup> RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).



Flow Cytometry (Intracellular) - Anti-PGP9.5 antibody [EPR4118] (ab108986)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized Y79 (Human retinoblastoma retinoblastoma) cells labelling PGP9.5 with ab108986 at 1/20 dilution (Red) compared with a Rabbit monoclonal IgG (ab172730) isotype control (black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor<sup>®</sup> 488, ab150077) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-PGP9.5 antibody [EPR4118] (ab108986)

PGP9.5 was immunoprecipitated from 0.35 mg Human fetal brain lysate with ab108986 at 1/20 dilution (0.5µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab108986 1/500 dilution (0.17 µg/ml). VeriBlot for IP Detection Reagent (HRP) (ab131366) was used as the secondary antibody at 1/1000 dilution.

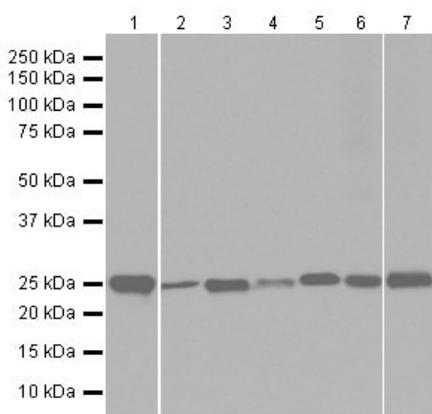
**Lane 1:** Human fetal brain lysate 10µg

**Lane 2:** ab108986 IP in Human fetal brain lysate

**Lane 3:** Rabbit monoclonal IgG (ab172730) instead of ab108986 in Human fetal brain lysate.

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

**Exposure time:** 1 second.



Western blot - Anti-PGP9.5 antibody [EPR4118] (ab108986)

**All lanes :** Anti-PGP9.5 antibody [EPR4118] (ab108986) at 1/5000 dilution

**Lane 1 :** HEK-293 (Human embryonic kidney epithelial cell) whole cell lysates

**Lane 2 :** SH-SY5Y (Human neuroblastoma epithelial cell) whole cell lysates

**Lane 3 :** C6 (Rat glial tumor glial cell) whole cell lysates

**Lanes 4 & 6 :** PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysates

**Lane 5 :** Neuro-2a (Mouse neuroblastoma neuroblast) whole cell lysates

**Lane 7 :** Mouse brain lysates

Lysates/proteins at 20 µg per lane.

**Secondary**

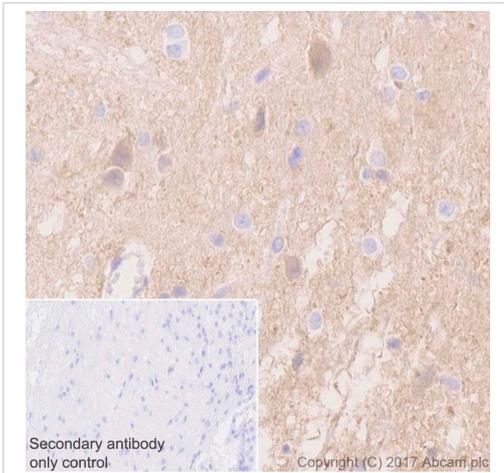
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

**Predicted band size:** 24 kDa

**Observed band size:** 25 kDa

**Exposure time:** 10 seconds

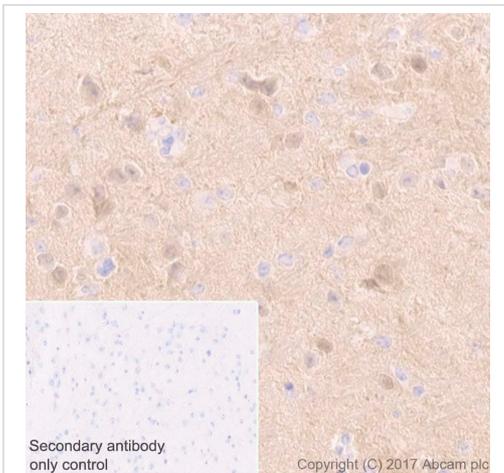
Blocking/Diluting buffer and concentration: 5% NFDN/TBST



Immunohistochemical analysis of paraffin-embedded rat cerebral cortex tissue labeling PGP9.5 with ab108986, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining on rat cerebral cortex. The section was incubated with [ab229902](#) for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND<sup>®</sup> RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval using [ab93684](#) (Tris/EDTA buffer, pH 9.0).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

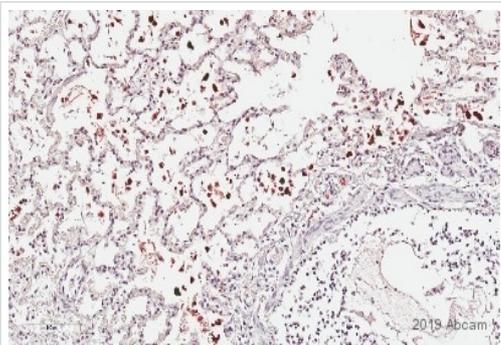
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] (ab108986)



Immunohistochemical analysis of paraffin-embedded mouse cerebral cortex tissue labeling PGP9.5 with ab108986, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining on mouse cerebral cortex. The section was incubated with [ab229902](#) for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND<sup>®</sup> RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval using [ab93684](#) (Tris/EDTA buffer, pH 9.0).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

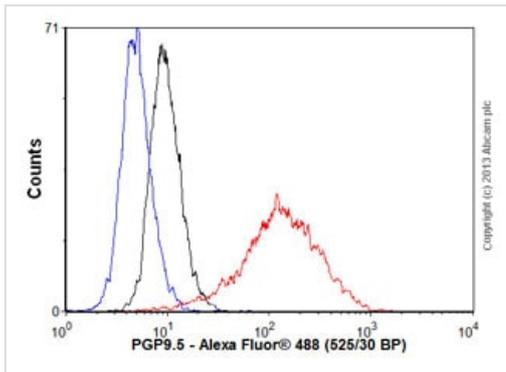
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] (ab108986)



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] (ab108986)

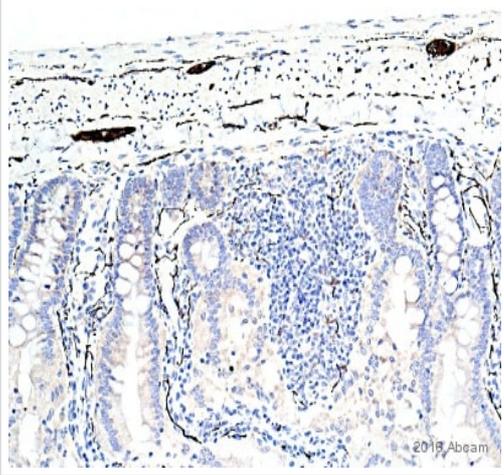
This image is courtesy of an anonymous Abreview

Formalin-fixed, paraffin-embedded Cat lung tissue stained for PGP9.5 using ab108986 at 1/250 dilution in immunohistochemical analysis. The secondary antibody was ImmPRESS™ HRP Universal Antibody (Anti-Mouse IgG/A). Antigen retrieval: Heat mediated - Buffer/Enzyme Used: 10 mM citrate, pH6.0.



Flow Cytometry (Intracellular) - Anti-PGP9.5 antibody [EPR4118] (ab108986)

Overlay histogram showing SH-SY5Y cells stained with ab108986 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab108986, 1/10000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (0.1µg/1x10<sup>6</sup> cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in SH-SY5Y cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

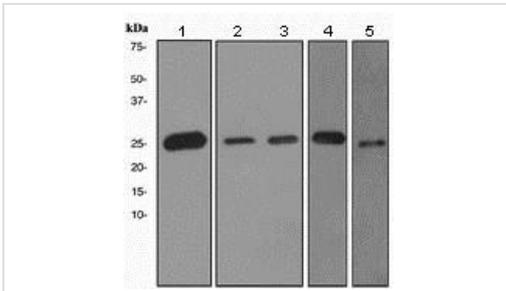


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] (ab108986)

This image is courtesy of an Abreview submitted by Carl Hobbs

Immunohistochemical analysis of rat Jejunum tissue sections labeling PGP9.5 with ab108986 at a dilution of 1/1000. Sections were fixed with Formaldehyde. A Biotin conjugated Goat Anti-Rabbit IgG at 1/300 was used as the secondary antibody. Antigen retrieval was heat mediated using citric acid.

All nerve components of enteric plexuses appear to be very well demonstrated, particularly the fine fibres of the lamina propria and the muscularis mucosa.



Western blot - Anti-PGP9.5 antibody [EPR4118] (ab108986)

**All lanes** : Anti-PGP9.5 antibody [EPR4118] (ab108986) at 1/1000 dilution

**Lane 1** : Fetal brain cell lysate

**Lane 2** : Y79 cell lysate

**Lane 3** : U87-MG cell lysate

**Lane 4** : SH-SY5Y cell lysate

**Lane 5** : 293T cell lysate

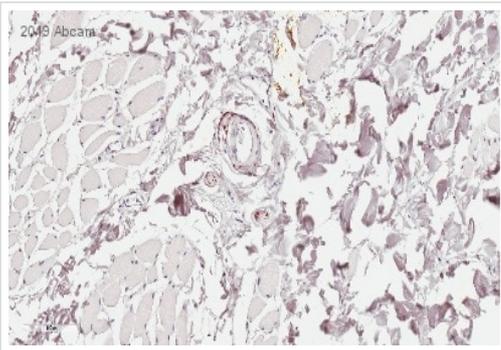
Lysates/proteins at 10 µg per lane.

### Secondary

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

**Predicted band size:** 24 kDa

**Observed band size:** 25 kDa

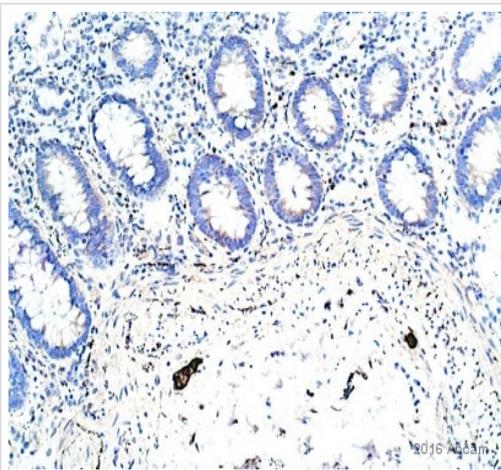


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] (ab108986)

This image was courtesy of an anonymous Abreview

Formalin-fixed, paraffin-embedded Dog skin tissue stained for PGP9.5 using ab108986 at 1/500 dilution in immunohistochemical analysis. ImmPRESS™ Anti-Rabbit IgG Polymer Detection Kit was used as the secondary antibody.

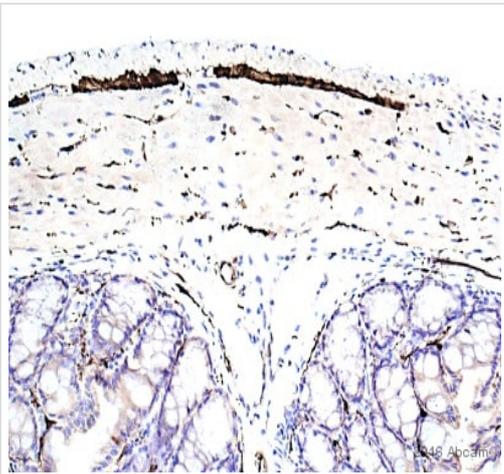
Antigen Retrieval: Heat mediated - Buffer/Enzyme Used: 10 mM citrate, pH6.0



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] (ab108986)

This image is courtesy of an Abreview submitted by Carl Hobbs.

ab108986 staining PGP9.5 in human colon tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with formaldehyde and blocked with 2% BSA for 10 minutes at 21°C; antigen retrieval was by heat mediation in citric acid. Samples were incubated with the primary antibody (1/500 in TBS/BSA/azide) for 16 hours at 21°C. A Biotin-conjugated goat anti-rabbit IgG polyclonal (1/300) was used as the secondary antibody.



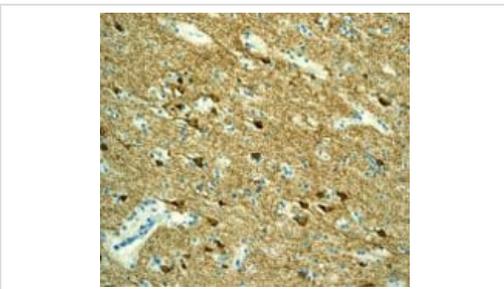
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody

[EPR4118] (ab108986)

This image is courtesy of an Abreview submitted by Carl Hobbs

Immunohistochemical analysis of mouse colon tissue sections labeling PGP9.5 with ab108986 at a dilution of 1/1500. Sections were fixed with Formaldehyde. A Biotin conjugated Goat Anti-Rabbit IgG at 1/300 was used as the secondary antibody. Antigen retrieval was heat mediated using citric acid.

All nerve cell/fibre components of enteric plexuses are demonstrated very well.



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

[EPR4118] (ab108986)

Immunohistochemical staining of PGP9.5 in paraffin embedded Human glioma tissue, using ab108986 at a 1/250 dilution.

**Why choose a recombinant antibody?**

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

Anti-PGP9.5 antibody [EPR4118] (ab108986)

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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