abcam

Product datasheet

Anti-PGP9.5 antibody [EPR4118] - Neuronal Marker ab108986

敲除验证 重组 RabMAb

★★★★★ 11 Abreviews 69 References 20 图像

概述

产品名称	Anti-PGP9.5抗体[EPR4118] - Neuronal Marker
描述	兔单克隆抗体[EPR4118] to PGP9.5 - Neuronal Marker
宿主	Rabbit
经测试应用	**适用于:** ICC/IF, Flow Cyt (Intra), IHC-Fr, WB, IP, IHC-P
种属反应性	**与反应:** Mouse, Rat, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	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, Neuro2a cells; IHC-Fr: Mouse cerebrum tissue.
常规说明	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**.

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. Stable for 12 months at -20°C.
存储溶液	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.5% BSA
纯度	Protein A purified
克隆	单克隆
克隆编号	EPR4118

同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab108986于以下的经测试应用

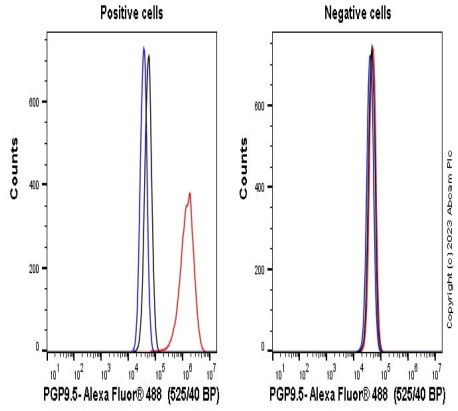
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
ICC/IF	★★★★★ (1)	1/500.
Flow Cyt (Intra)		1/100 - 1/10000. **ab172730** - 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.

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



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

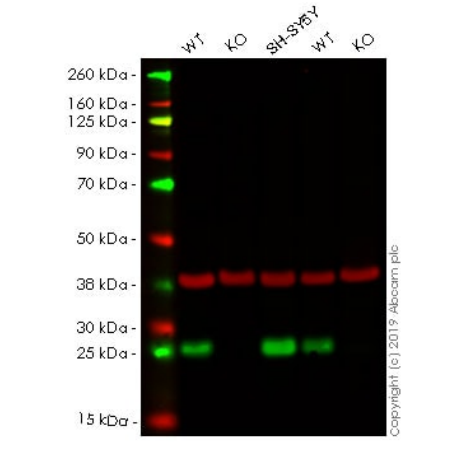
Flow cytometry overlay histogram showing left Neuro2a positive cells and right negative NIH3T3 stained with ab108986 (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilised with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab108986) (1x 10^6 in 100µl at 0.2µg/ml (1/10500)) for 30min at 22°C.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 22°C

Isotype control antibody (black line) was Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.

This antibody gave a positive signal in Neuro2a Fixed with 80% methanol (5 min) / permeabilised with 0.1% PBS-Triton X-100 for 15 min under the same conditions.



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

All lanes : Anti-PGP9.5 antibody [EPR4118] - Neuronal Marker (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.

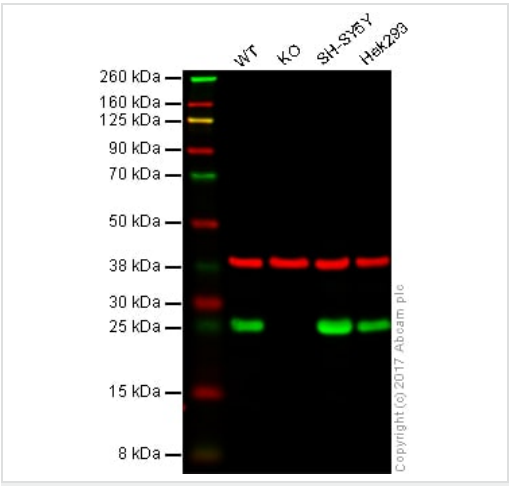
Performed under reducing conditions.

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.



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

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

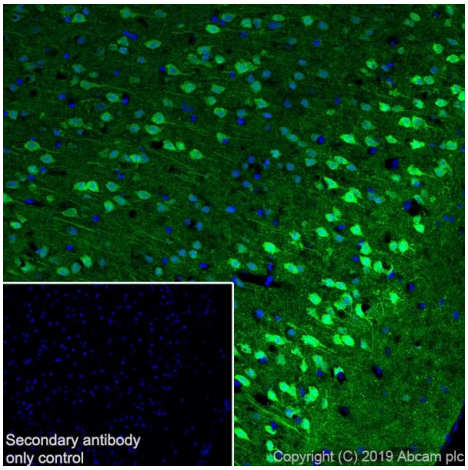
Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : UCHL1 knockout HAP1 whole cell lysate
Lane 3 : SH-SY5Y whole cell lysate
Lane 4 : HEK293 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 24 kDa

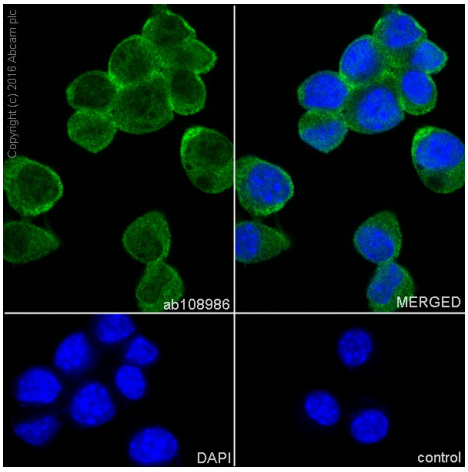
Lanes 1 - 4: Merged signal (red and green). Green - ab108986 observed at 24 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

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] - Neuronal Marker (ab108986)

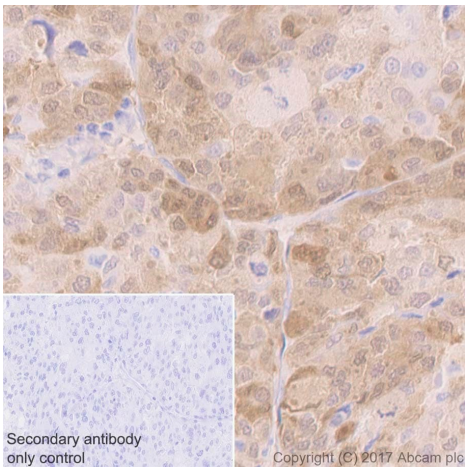
Immunohistochemistry (Frozen sections) analysis of mouse cerebrum tissue sections labeling PGP9.5 with Purified ab108986 at 1/250 (0.5 µg/ml).Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used 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] - Neuronal Marker (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).

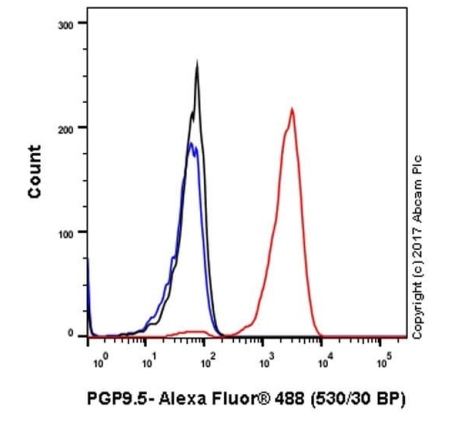
The negative control is PBS only.



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

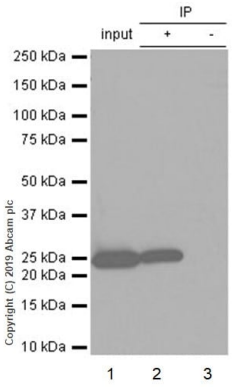
Immunohistochemical analysis of paraffin-embedded human hepatocellular carcinoma tissue labeling PGP9.5 with ab108986, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining on human hepatocellular carcinoma. The section was incubated with **ab229902** for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® 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] - Neuronal Marker (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 Fluorr® 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.



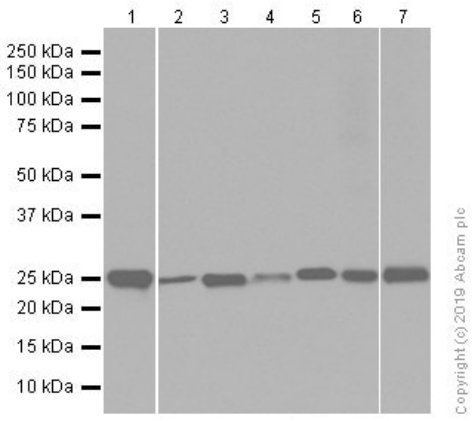
Immunoprecipitation - Anti-PGP9.5 antibody [EPR4118] - Neuronal Marker (ab108986)

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] - Neuronal Marker (ab108986)

All lanes : Anti-PGP9.5 antibody [EPR4118] - Neuronal Marker (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

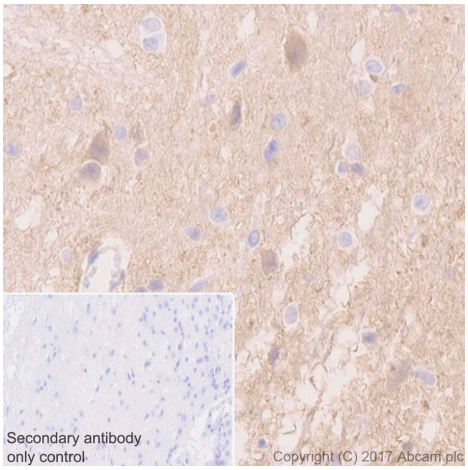
Lysates/proteins at 20 µg per lane.

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

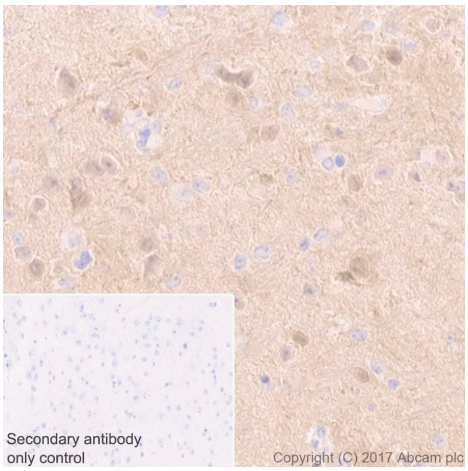
Blocking/Diluting buffer and concentration: 5% NFDM/TBST



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

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® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).

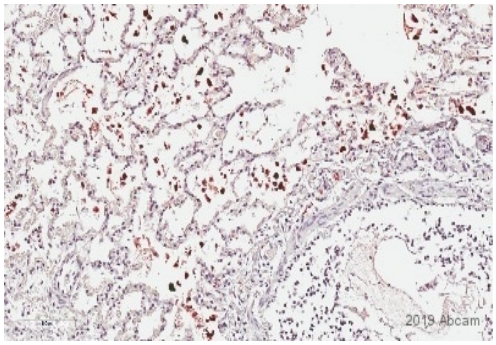
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] - Neuronal Marker (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® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0).

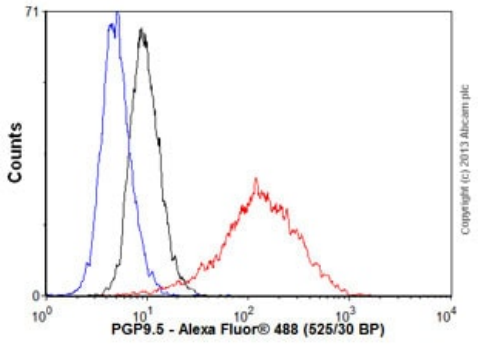
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).



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

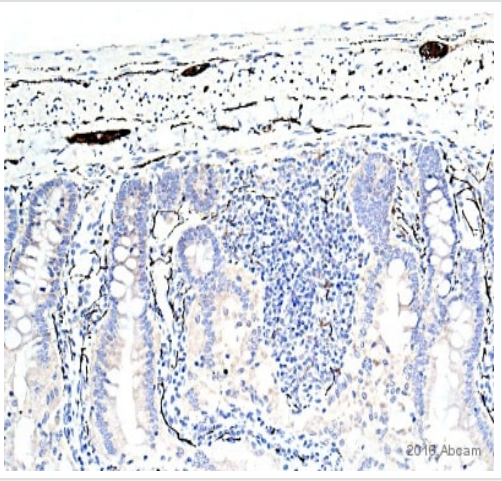
This image is courtesy of an annonymous Abreview

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] - Neuronal Marker (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/1x10^6 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.

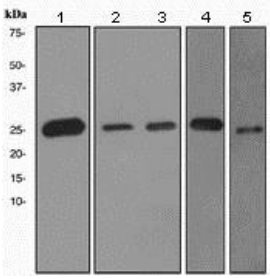


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] - Neuronal Marker (ab108986)

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 dulution 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] - Neuronal Marker (ab108986)

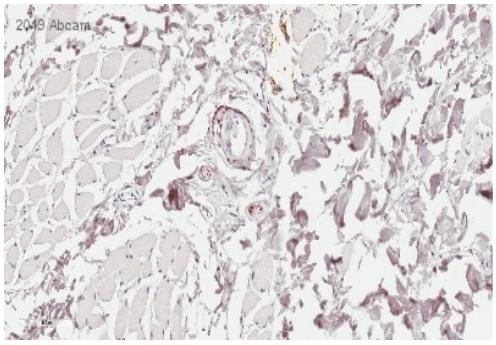
All lanes : Anti-PGP9.5 antibody [EPR4118] - Neuronal Marker (ab108986) at 1/1000 dilution

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

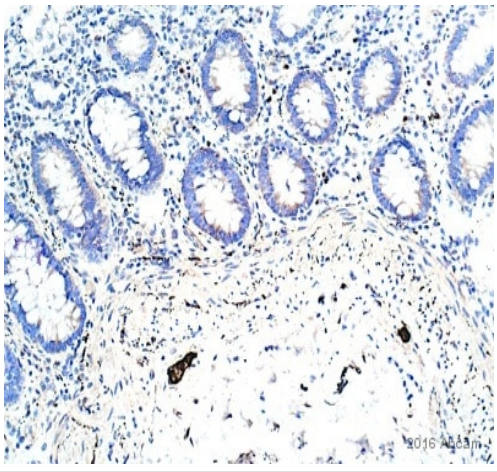


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] - Neuronal Marker (ab108986)

This image was courtesy of an annonymous Abreview

Fomalin-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.

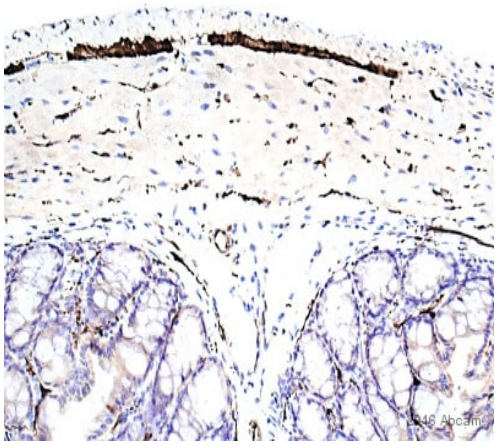
Antigen Retrieval: Heat mediated - Buffer/Enzyme Used: 10 mM citrate, pH6.0



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

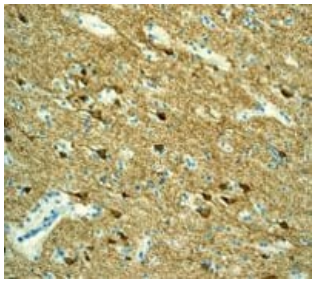


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PGP9.5 antibody [EPR4118] - Neuronal Marker (ab108986)

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 dulution 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] - Neuronal Marker (ab108986)

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

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

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