# abcam

## Product datasheet

## Anti-CNPase antibody [11-5B] ab6319



★★★★★ 38 Abreviews 135 References 10 图像

概述

产**品名称** Anti-CNPase抗体[11-5B]

描述 小鼠单克隆抗体[11-5B] to CNPase

**宿主** Mouse

pıg

免疫原 Full length native protein (purified) corresponding to Human CNPase.

阳性对照 WB: HeLa and Hap1 cell lysates; Human and Mouse Spinal Cord and Brain tissue lysates; Rat

Brain tissue lysate. IHC-P: FFPE human cerebral cortex tissue sections. ICC: Primary

hippocampal rat neurons/glia, DIV14. cells

常规说明 This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or

conjugation for your experiments, please contact orders@abcam.com.

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.

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

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 Constituents: PBS, 6.97% L-Arginine

1

纯**度** Protein G purified

 克隆
 单克隆

 克隆编号
 11-5B

 同种型
 IgG1

应用

### The Abpromise guarantee Abpromise™承诺保证使用ab6319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	<b>★★★★★ (15)</b>	Use at an assay dependent concentration.
WB	****(8)	Use a concentration of 5 µg/ml. Detects a band of approximately 48 kDa (predicted molecular weight: 48 kDa).
ICC/IF	****(7)	Use at an assay dependent concentration.

#### 靶标

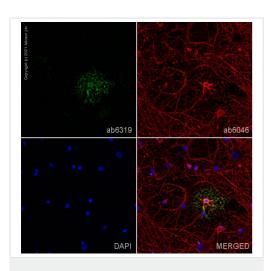
#### 序列相似性

Belongs to the cyclic nucleotide phosphodiesterase family.

细胞定位

Membrane. Melanosome. Firmly bound to membrane structures of brain white matter. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

#### 图片

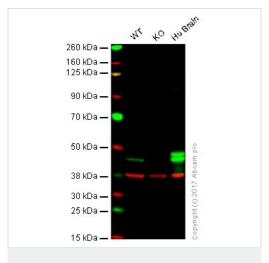


Immunocytochemistry/ Immunofluorescence - Anti-CNPase antibody [11-5B] (ab6319)

ab6319 staining CNPase in primary hippocampal rat neurons/glia, (obtained from Neuromics, cat. no. PC35101), DIV14. 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 ab6319 at 1µg/ml and ab6046, Rabbit polyclonal to beta Tubulin - Loading Control. Cells were then incubated with ab150117, Goat polyclonal Secondary Antibody to Mouse IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (shown in green) and ab150080, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 594) at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Also suitable in cells fixed with 4% paraformaldehyde (10 min).

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



Western blot - Anti-CNPase antibody [11-5B] (ab6319)

All lanes: Anti-CNPase antibody [11-5B] (ab6319) at 5 µg/ml

Lane 1: Wild-type HAP1 whole cell lysate at 40 µg

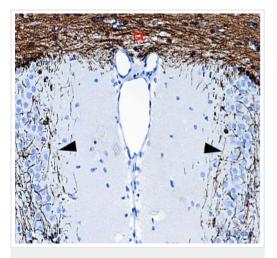
Lane 2 : CNPase knockout HAP1 whole cell lysate at 40  $\mu g$ 

Lane 3: Human Brain whole cell lysate at 20 µg

Predicted band size: 48 kDa

**Lanes 1 - 3:** Merged signal (red and green). Green - ab6319 observed at 48 kDa. Red - loading control, **ab181602**, observed at 37 kDa.

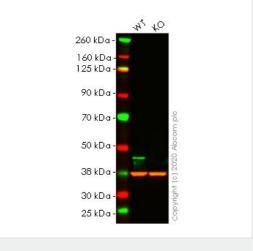
ab6319 was shown to recognize CNPase in wild-type HAP1 cells as signal was lost at the expected MW in CNPase knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and CNPase knockout samples were subjected to SDS-PAGE. Ab6319 and <a href="mailto:ab181602">ab181602</a> (Rabbit anti-GAPDH loading control) were incubated overnight at 4°C at 5 µg/ml and 1/20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed <a href="mailto:ab216772">ab216772</a> and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed <a href="mailto:ab216777">ab216777</a> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CNPase antibody [11-5B] (ab6319)

This image is courtesy of an abreview submitted by Carl Hobbs, King's College London, United Kingdom

IHC-P image of CNPase staining on rat brain sections using ab6319 (1/1600). heat mediated antigen retrieval on paraffin embedded sections was performed using citric acid. The sections were then blocked with 1% BSA for 10 min at 21°C. The primary antibody was incubated for 16 hours at 21°C. The sections were then incubated in Goat anti-mouse (Biotin) at 1:200.



Western blot - Anti-CNPase antibody [11-5B] (ab6319)

All lanes: Anti-CNPase antibody [11-5B] (ab6319) at 5 μg/ml

Lane 1: Wild-type HeLa cell lysate

Lane 2: CNP knockout HeLa cell lysate

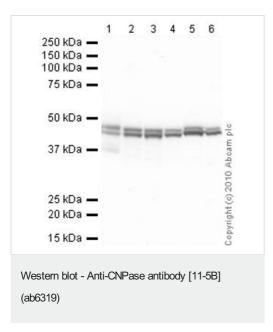
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 48 kDa **Observed band size:** 48 kDa

**Lanes 1-2:** Merged signal (red and green). Green - ab6319 observed at 48 kDa. Red - Anti-GAPDH antibody[EPR16891] - Loading Control (ab181602) observed at 37 kDa.

ab6319 was shown to react with CNPase in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab264949 (knockout cell lysate ab256877) was used. Wild-type HeLa and CNP knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab6319 and Anti-GAPDH antibody[EPR16891] - Loading Control (ab181602) overnight at 4°C at a 5 µg/ml and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Mouse lgG H&L (IRDye®800CW) preadsorbed (ab216772) and Goat Anti-Rabbit lgG H&L (IRDye®680RD) preadsorbed (ab216777) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



**All lanes :** Anti-CNPase antibody [11-5B] (ab6319) at 1/100 dilution

Lane 1: Human spinal cord tissue lysate - total protein (ab29188)

Lane 2 : Human brain tissue lysate - total protein (ab29466)

Lane 3: Spinal Cord (Mouse) Tissue Lysate

Lane 4: Brain (Mouse) Tissue Lysate

Lane 5: Spinal Cord (Rat) Tissue Lysate

Lane 6: Brain (Rat) Tissue Lysate

Lysates/proteins at 20 µg per lane.

#### **Secondary**

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

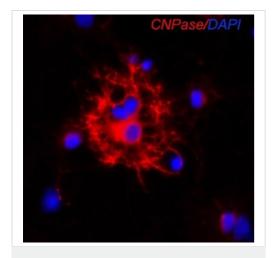
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 48 kDa

Exposure time: 1 minute

This antibody was raised against full length native CNPase and is predicted to recognize both isoforms. The predicted molecular weights of isoforms CNPI and CNPII are 45- and 48-kDa respectively.

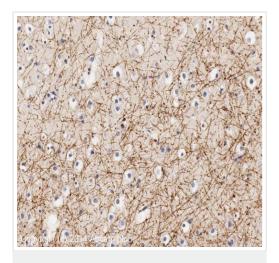


Immunocytochemistry/ Immunofluorescence - Anti-CNPase antibody [11-5B] (ab6319)

This image is courtesy of an anonymous abreview.

ab6319 staining CNPase in rat oligodendrocytes by Immunocytochemistry/ Immunofluorescence.

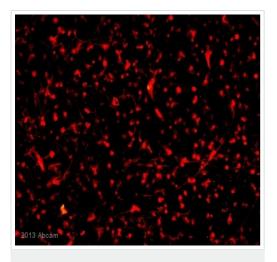
Cells were fixed in paraformaldehyde, blocked using 5% serum for 10 minutes at 25°C, then incubated with ab6319 at a 1/200 dilution for 2 hours at 25°C. The secondary used was a goat anti-mouse Cy3 conjugated polyclonal at a 1/100 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CNPase antibody [11-5B] (ab6319)

IHC image of CNPase staining in human cerebral cortex formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. The section was pretreated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab6319, 5µg/ml, for 15 mins 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.

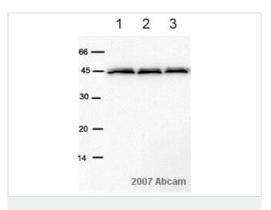
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunocytochemistry/ Immunofluorescence - Anti-CNPase antibody [11-5B] (ab6319)

This image is courtesy of an anonymous Abreview

ab6319 staining CNPase in the rat oligodendrocytes by ICC/IF (Immunoytochemistry/immunofluorescence). Cells were fixed with paraformaldehyde, permeabilized with methanol and blocked with 5% BSA for 1 hour at 37°C. Samples were incubated with primary antibody (1/100 in PBS) for 18 hours at 4°C. An Alexa Fluor<sup>®</sup> 594-conjugated Goat anti-mouse IgG polyclonal (1:200) was used as the secondary antibody.



Western blot - Anti-CNPase antibody [11-5B] (ab6319)

This image is courtesy of an anonymous Abreview

**All lanes :** Anti-CNPase antibody [11-5B] (ab6319) at 1/750 dilution

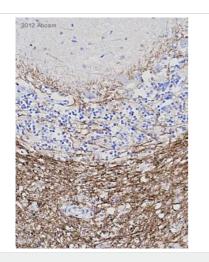
All lanes: Spinal Cord homogenate (whole tissue lysate)

Lysates/proteins at 2 µg per lane.

## Secondary

All lanes: HRP conjugated sheep anti-mouse IgG

**Predicted band size:** 48 kDa **Observed band size:** 45,47 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CNPase antibody [11-5B] (ab6319)

This image is courtesy of an abreview submitted by Carl Hobbs, King's College London, United Kingdom

ab6319 staining CNPase in Dog Cerebellum tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffinembedded sections). Tissue was fixed with formaldehyde and blocked with 1% BSA for 10 minutes at 21°C; antigen retrieval was by heat mediation in a citrate buffer. Samples were incubated with primary antibody (1/1500 in blocking buffer) for 2 hours at 21°C. A Biotin-conjugated Goat anti-mouset IgG polyclonal (1/200) was used as the secondary antibody.

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

#### Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- · Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit <a href="https://www.abcam.cn/abpromise">https://www.abcam.cn/abpromise</a> or contact our technical team.

#### Terms and conditions

· Guarantee only valid for products bought direct from Abcam or one of our authorized distributors