# abcam

# Product datasheet

# Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407] ab189891



# 1 References 8 Images

#### Overview

Product name Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407]

**Description** Rabbit monoclonal [EPR18407] to ENO1 + ENO2 + ENO3

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), ICC/IF, WB

Species reactivity Reacts with: Mouse, Rat, Human, Recombinant fragment

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Human ENO1, ENO2 and ENO3 full length recombinant proteins; Human fetal heart, fetal

kidney and fetal spleen lysates; HeLa, Jurkat, MCF7, A431, C6, RAW 264.7 and NIH/3T3 whole cell lysates; Mouse brain and heart lysates; Rat brain and heart lysates. ICC/IF: HeLa and NIH/3T3

cells. Flow Cyt (intra): NIH/3T3 cells.

**General notes**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<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

#### **Properties**

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

1

ClonalityMonoclonalClone numberEPR18407

**Isotype** IgG

#### **Applications**

#### The Abpromise guarantee

Our Abpromise guarantee covers the use of ab189891 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
Flow Cyt (Intra)		1/70.
ICC/IF		1/500.
WB		1/1000. Detects a band of approximately 47 kDa (predicted molecular weight: 47 kDa).

#### **Target**

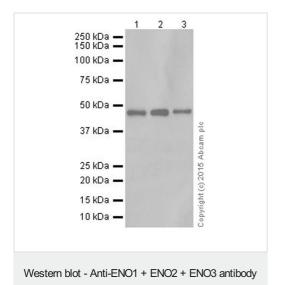
#### Relevance

Enolase 1 is a multifunctional enzyme that, as well as its role in glycolysis, plays a part in various processes such as growth control, hypoxia tolerance and allergic responses. May also function in the intravascular and pericellular fibrinolytic system due to its ability to serve as a receptor and activator of plasminogen on the cell surface of several cell-types such as leukocytes and neurons. Stimulates immunoglobulin production. MBP1 binds to the myc promoter and acts as a transcriptional repressor. May be a tumor suppressor. Enolase 2 has neurotrophic and neuroprotective properties on a broad spectrum of central nervous system (CNS) neurons. Binds, in a calcium-dependent manner, to cultured neocortical neurons and promotes cell survival. Enolase 3 appears to have a function in striated muscle development and regeneration.

#### **Cellular localization**

ENO1: Cytoplasm. Cell membrane. Cytoplasm, myofibril, sarcomere, M-band. Note: Can translocate to the plasma membrane in either the homodimeric (alpha/alpha) or heterodimeric (alpha/gamma) form. ENO1 is localized to the M-band. ENO2: Cytoplasm. Cell membrane. Note: Can translocate to the plasma membrane in either the homodimeric (alpha/alpha) or heterodimeric (alpha/gamma) form ENO3: Cytoplasm. Note: Localized to the Z line. Some colocalization with CKM at M-band.

#### **Images**



[EPR18407] (ab189891)

**All lanes :** Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407] (ab189891) at 1/1000 dilution

 $\textbf{Lane 1:} \ \textbf{Human ENO1 full length recombinant protein}$ 

Lane 2: Human ENO2 full length recombinant protein

Lane 3: Human ENO3 full length recombinant protein

Lysates/proteins at 0.02 µg per lane.

### **Secondary**

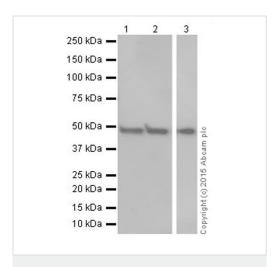
**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

**Predicted band size:** 47 kDa **Observed band size:** 47 kDa

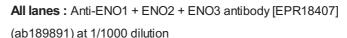
Exposure time: 5 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

Human ENO1 full length recombinant protein (Cat#:<u>ab89248</u>) contains aa1-434. Human ENO2 full length recombinant protein contains aa1-434 with a His-Tag®. Human ENO3 full length recombinant protein(Cat#:<u>ab113127</u>) contains aa1-434 with a His-Tag®. Human ENO2 full length recombinant protein was made inhouse.



Western blot - Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407] (ab189891)



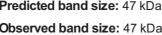
Lane 1: Human fetal heart lysate Lane 2: Human fetal kidney lysate Lane 3: Human fetal spleen lysate

Lysates/proteins at 10 µg per lane.

## Secondary

All lanes: Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 47 kDa Observed band size: 47 kDa



Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1 and 2: 5 seconds; Lane 3: 15 seconds.

All lanes: Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407] 3 250 kDa -(ab189891) at 1/5000 dilution 150 kDa -

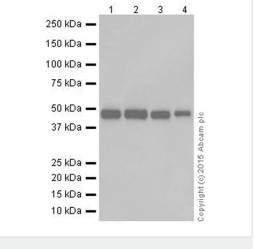
> Lane 1: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 2: Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

Lane 3: MCF7 (Human breast adenocarcinoma cell line) whole cell lysate

Lane 4: A431 (Human epidermoid carcinoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.



Western blot - Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407] (ab189891)

# **Secondary**

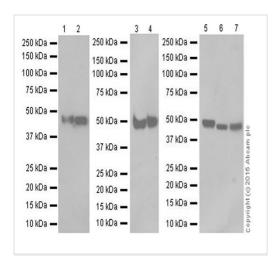
All lanes: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at

1/100000 dilution

**Predicted band size:** 47 kDa **Observed band size:** 47 kDa

Exposure time: 3 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407] (ab189891) Lanes 1-4: Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407]

(ab189891) at 1/1000 dilution

Lanes 5-7: Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407]

(ab189891) at 1/5000 dilution

Lane 1: Mouse brain tissue lysate

Lane 2: Mouse heart tissue lysate

Lane 3: Rat brain tissue lysate

Lane 4: Rat heart tissue lysate

Lane 5: C6 (Rat glial tumor cell line) whole cell lysate

Lane 6: RAW 264.7 (Mouse macrophage cell line transformed

with Abelson murine leukemia virus) whole cell lysate

Lane 7: NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell

lysate

Lysates/proteins at 10 µg per lane.

#### Secondary

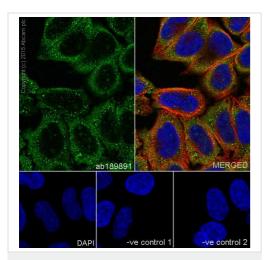
All lanes: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/10000

dilution

**Predicted band size:** 47 kDa **Observed band size:** 47 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

**Exposure time:** Lane 1-4: 5 seconds; Lane 5-7: 1 second.



Immunocytochemistry/ Immunofluorescence - Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407] (ab189891)

ab189891

DAPI

-ve control 1

-ve control 2

Immunocytochemistry/ Immunofluorescence - Anti-ENO1 + ENO2 + ENO3 antibody [EPR18407] (ab189891)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling ENO1 + ENO2 + ENO3 with ab189891 at 1/500 dilution, followed by Goat Anti-Rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing mostly cytoplasmic staining on Hela cells.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin - Loading Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse lgG (AlexaFluor®594) preadsorbed (ab150120) at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab189891 at 1/500 dilution followed by **ab150120** at 1/1000 dilution.

-ve control 2:  $\underline{ab7291}$  at 1/1000 dilution followed by  $\underline{ab150077}$  at 1/1000 dilution.

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling ENO1 + ENO2 + ENO3 with ab189891 at 1/500 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing mostly cytoplasmic staining on NIH/3T3 cells.

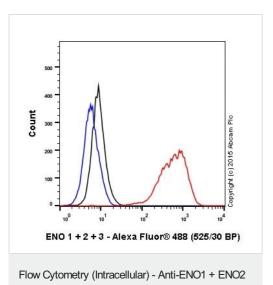
The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin antibody -Loading Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse IgG (AlexaFluor®594) preadsorbed (ab150120) at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab189891 at 1/500 dilution followed by  $\underline{ab150120}$  at 1/1000 dilution.

-ve control 2:  $\underline{ab7291}$  at 1/1000 dilution followed by  $\underline{ab150077}$  at 1/1000 dilution.



+ ENO3 antibody [EPR18407] (ab189891)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed NIH/3T3 (Mouse embryonic fibroblast cell line) labeling ENO1 + ENO2 + ENO3 with ab189891 at 1/70 dilution (red) compared with a Rabbit  $\lg G$ ,monoclonal- lsotype control (ab172730) (black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti Rabbit  $\lg G$  (Alexa Fluor 488) at 1/500 dilution was used as the secondary antibody.



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