abcam

# ab315816

# Anti-P5CS antibody [EPR28827-56]

Recombinant | RabMAb | KO Validated | 20ul selling size

## Key facts

| | |
|---|---|
| **Isotype** | IgG |
| **Host species** | Rabbit |
| **Storage buffer** | pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA |
| **Form** | Liquid |
| **Clonality** | Monoclonal |
| **Immunogen** | The exact immunogen used to generate this antibody is proprietary information. |
| **Clone number** | EPR28827-56 |
| **Purification technique** | Affinity purification Protein A |
| **Specificity** | Unsuitable for mouse and rat IHC. |
| **Concentration** | 0.498 mg/mL The concentration of this product may be batch-dependent<br>Batch concentration finder → |

## Reactivity data

## WB

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/1000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | 1/1000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | 1/1000 |
| **Notes** | - |

## IP

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/30 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | 1/30 |
| **Notes** | - |

### Expected

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

## IHC-P

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/100 |
| **Notes** | Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |

### Not recommended

| | |
|---|---|
| **Species** | Mouse, Rat |
| **Dilution info** | - |
| **Notes** | - |

## Flow Cyt (Intra)

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/500 |
| **Notes** | - |

### Expected

| | |
|---|---|
| **Species** | Mouse, Rat |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

## ICC/IF

### Not recommended

| | |
|---|---|
| **Species** | Human, Mouse, Rat |
| **Dilution info** | - |
| **Notes** | - |

# Target data

See full target information ALDH18A1

| | |
|---|---|
| **Function** | Bifunctional enzyme that converts glutamate to glutamate 5-semialdehyde, an intermediate in the biosynthesis of proline, ornithine and arginine. |

# Storage

| | |
|---|---|
| **Shipped at conditions** | Conditional Ambient |

| | |
|---|---|
| **Appropriate short-term storage duration** | 1-2 weeks |
| **Appropriate short-term storage conditions** | +4°C |
| **Appropriate long-term storage conditions** | -20°C |
| **Aliquoting information** | Upon delivery aliquot |
| **Storage information** | Avoid freeze / thaw cycle |

## Notes

**Patented technology**
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

**What are the advantages of a recombinant monoclonal antibody?**
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 batch production

For more information, read more on recombinant antibodies.

## Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

| | |
|---|---|
| **Activity summary** | Pyrroline-5-carboxylate synthetase (P5CS) also known as ALDH18A1 plays an important role in proline biosynthesis. This enzyme catalyzes the conversion of glutamate to pyrroline-5-carboxylate. The P5CS protein has a molecular mass of approximately 87 kDa and is mainly expressed in tissues with high rates of protein synthesis such as the liver and kidney. It has significant expression in the mitochondria reinforcing its function in energy metabolism and amino acid biosynthesis. |
| **Biological function summary** | P5CS influences cellular responses to oxidative stress due to its role in proline metabolism and redox balance. As part of the proline synthesis pathway P5CS contributes to the production of proline a critical amino acid for protein synthesis. Proline acts as an osmoprotectant and plays a role in cellular structure. P5CS operates within a complex pathway regulating amino acids acting cooperatively with other proteins involved in synthesis processes. |
| **Pathways** | P5CS is integral to the proline biosynthetic pathway and is linked to the glutamate pathway. Its activity closely coordinates with the functioning of enzymes like |

| | |
|---|---|
| | pyrroline-5-carboxylate reductase which converts pyrroline-5-carboxylate to proline. This relationship ensures the continuous supply of proline vital for collagen synthesis and also intersects with pathways involving protein metabolism and cellular stress responses. |
| **Associated diseases and disorders** | Altered P5CS function associates with conditions like cutis laxa and hyperprolinemia. Mutations in the P5CS gene can lead to dysfunction in proline metabolism resulting in elastic skin joint and bone issues and mental retardation in some individuals. The enzyme's interaction with proteins such as the collagen metabolism enzymes highlights its relevance in dermatological and connective tissue disorders. |

## Product promise

**Tested**

We have tested this species and application combination and it works. It is covered by our product promise.

**Expected**

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

**Predicted**

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

**Not recommended**

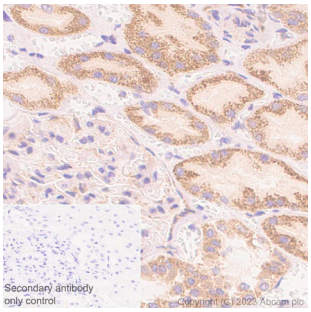
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

## 9 product images




## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-P5CS antibody [EPR28827-56] (ab315816)

Immunohistochemical analysis of paraffin-embedded Human kidney tissue labeling P5CS with ab315816 at 1/100 (4.98 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

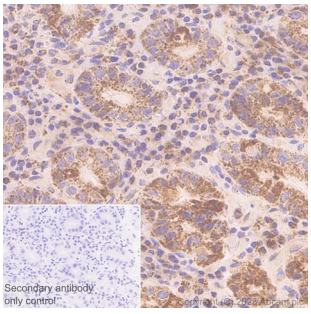
Positive staining on human kidney. The section was incubated with ab315816 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.

---




## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-P5CS antibody [EPR28827-56] (ab315816)

Immunohistochemical analysis of paraffin-embedded Human stomach tissue labeling P5CS with ab315816 at 1/100 (4.98 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

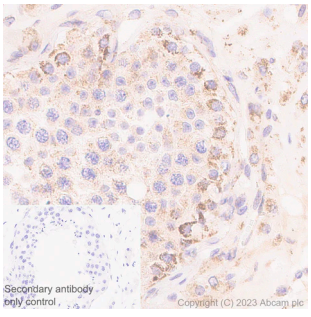
Positive staining on human stomach. The section was incubated with ab315816 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.

---




## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-P5CS antibody [EPR28827-56] (ab315816)

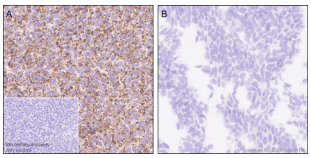
Immunohistochemical analysis of paraffin-embedded Human testis tissue labeling P5CS with ab315816 at 1/100 (4.98 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Positive staining on human testis. The section was incubated with ab315816 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-P5CS antibody [EPR28827-56] (ab315816)

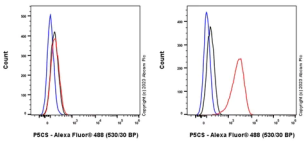
Immunohistochemical analysis of paraffin-embedded (A) Wild-type HEK293T (human epithelial cell line from embryonic kidney) cell pellet (B) ALDH18A1 knockout HEK293T cell pellet tissue labeling P5CS with ab315816 at 1/100 (4.98 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Positive staining on (A) Wild-type HEK293T cell pellet, negative staining on (B) ALDH18A1 knockout HEK293T cell pellet. The section was incubated with ab315816 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

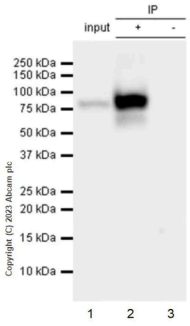
Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



## Flow Cytometry (Intracellular) - Anti-P5CS antibody [EPR28827-56] (ab315816)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized ALDH18A1 KO HEK293T (human ALDH18A1 knockout embryonic kidney epithelial cell, Left) / Parental HEK293T (Right) cells labelling P5CS with ab315816 at 1/500 dilution (0.1 ug)/Red compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue).

Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody.




## Immunoprecipitation - Anti-P5CS antibody [EPR28827-56] (ab315816)

P5CS was immunoprecipitated from 0.35 mg HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysate with ab315816 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab315816 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution.

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

All lanes:
Immunoprecipitation - Anti-P5CS antibody [EPR28827-56] (ab315816) at 1/1000 dilution

Lane 1:
HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysate

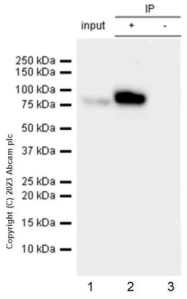
Lane 2:
ab315816 at 1/30 IP in HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysate

Lane 3:
Rabbit monoclonal IgG (ab172730) instead of ab315816 in HepG2 whole cell lysate

Secondary

All lanes:
Immunoprecipitation - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/5000 dilution

Exposure time: 3s




## Immunoprecipitation - Anti-P5CS antibody [EPR28827-56] (ab315816)

P5CS was immunoprecipitated from 0.35 mg NIH/3T3 (mouse embryonic fibroblast) whole cell lysate with ab315816 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab315816 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP) (ab131366) was used at 1/5000 dilution.

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

All lanes:
Immunoprecipitation - Anti-P5CS antibody [EPR28827-56] (ab315816) at 1/1000 dilution

Lane 1:
NIH/3T3 (mouse embryonic fibroblast) whole cell lysate

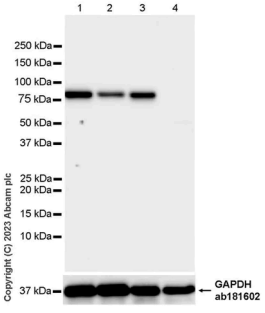
Lane 2:
ab315816 at 1/30 IP in NIH/3T3 (mouse embryonic fibroblast) whole cell lysate

Lane 3:
Rabbit monoclonal IgG (ab172730) instead of ab315816 in NIH/3T3 whole cell lysate

Secondary

All lanes:
Immunoprecipitation - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/5000 dilution

Exposure time: 6s




## Western blot - Anti-P5CS antibody [EPR28827-56] (ab315816)

P5CS Western blot staining using rabbit Anti-P5CS antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

In Western blot, ab315816 was shown to bind specifically to P5CS.

Target of interest was observed at 90 KD in wild-type HEK293T cell lysates (lane 3, wild-type cell line ab255449) with no signal observed at this size in ALDH18A1(P5CS) knockout cell line (lane 4, knockout cell line ab266378/ knockout cell lysate ab258300).

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/200000 dilution.

All lanes:
Western blot - Anti-P5CS antibody [EPR28827-56] (ab315816) at 1/1000 dilution

Lane 1:
HepG2 (human hepatocellular carcinoma epithelial cell) transfected with scrambled siRNA control whole cell lysate at 10 µg

Lane 2:
HepG2 transfected with siRNA specifically targeting P5CS whole cell lysate at 10 µg

Lane 3:
Wild-type HEK293T (human embryonic kidney epithelial cell) whole cell lysate at 10 µg

Lane 4:
ALDH18A1(P5CS) knockout HEK293T whole cell lysate at 10 µg

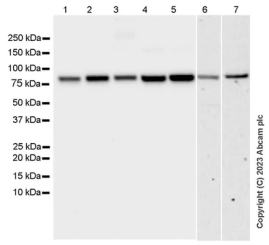
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Performed under reducing conditions.

Observed band size: 90 kDa, 36 kDa

Exposure time: 26s




## Western blot - Anti-P5CS antibody [EPR28827-56] (ab315816)

P5CS Western blot staining using rabbit Anti-P5CS antibody

Exposure time: Lane1-5: 26 seconds, lane 6-7: 180 seconds

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

All lanes:
Western blot - Anti-P5CS antibody [EPR28827-56] (ab315816) at 1/1000 dilution

Lane 1:
MCF7 (human breast adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 2:
A431 (human epidermoid carcinoma epithelial cell) whole cell lysate at 20 µg

Lane 3:
A549 (human lung carcinoma epithelial cell) whole cell lysate at 20 µg

Lane 4:
NIH/3T3 (mouse embryonic fibroblast) whole cell lysate at 20 µg

Lane 5:
4T1 (mouse mammary gland carcinoma epithelial cell) whole cell lysate at 20 µg

Lane 6:
Mouse brain tissue lysate at 20 µg

Lane 7:
Rat testis tissue lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 90 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.