# abcam

# Product datasheet

# Anti-Insulin antibody [EPR17359] ab181547



#### ★★★★★ 11 Abreviews 76 References 11 Images

#### Overview

**Product name** Anti-Insulin antibody [EPR17359]

**Description** Rabbit monoclonal [EPR17359] to Insulin

**Host species** Rabbit

Specificity The human recommendation is based on the IHC-P results. We do not guarantee WB for human.

**Tested applications** Suitable for: IHC-FoFr, WB, IHC-P, ICC/IF, mIHC

Species reactivity Reacts with: Mouse, Rat, Human

**Immunogen** Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control IHC-P: Human, mouse and rat pancreas tissue. ICC/IF: BxPC-3 cells. IHC-FoFr: Mouse pancreas

tissue. WB: Mouse & Rat pancreas lysate. mlHC: Human pancreas tissue.

**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® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb 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 Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

**Purity** Protein A purified

Clonality Monoclonal Clone number EPR17359

Isotype lgG

#### **Applications**

#### The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab181547 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
IHC-FoFr		1/1000.
WB	<b>★★★★☆ (1)</b>	Use at an assay dependent concentration. Predicted molecular weight: 12 kDa.
IHC-P	*****( <u>7</u> )	1/64000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		1/200.
mIHC		1/20000.

#### **Target**

#### **Function**

Insulin decreases blood glucose concentration. It increases cell permeability to monosaccharides, amino acids and fatty acids. It accelerates glycolysis, the pentose phosphate cycle, and glycogen synthesis in liver.

#### Involvement in disease

Defects in INS are the cause of familial hyperproinsulinemia (FHPRI) [MIM:176730]. Defects in INS are a cause of diabetes mellitus insulin-dependent type 2 (IDDM2) [MIM:125852]. IDDM2 is a multifactorial disorder of glucose homeostasis that is characterized by susceptibility to ketoacidosis in the absence of insulin therapy. Clinical fetaures are polydipsia, polyphagia and polyuria which result from hyperglycemia-induced osmotic diuresis and secondary thirst. These derangements result in long-term complications that affect the eyes, kidneys, nerves, and blood vessels.

Defects in INS are a cause of diabetes mellitus permanent neonatal (PNDM) [MIM:606176]. PNDM is a rare form of diabetes distinct from childhood-onset autoimmune diabetes mellitus type 1. It is characterized by insulin-requiring hyperglycemia that is diagnosed within the first months of life. Permanent neonatal diabetes requires lifelong therapy.

Defects in INS are a cause of maturity-onset diabetes of the young type 10 (MODY10) [MIM:613370]. MODY10 is a form of diabetes that is characterized by an autosomal dominant mode of inheritance, onset in childhood or early adulthood (usually before 25 years of age), a primary defect in insulin secretion and frequent insulin-independence at the beginning of the disease.

# Sequence similarities

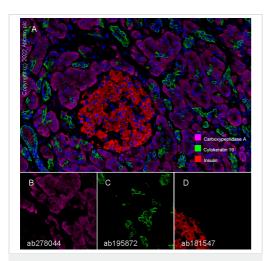
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Belongs to the insulin family.

#### **Cellular localization**

Secreted.

## **Images**



Multiplex immunohistochemistry - Anti-Insulin antibody [EPR17359] (ab181547)

Fluorescence multiplex immunohistochemical analysis of the human pancreas (Formalin/PFA-fixed paraffin-embedded sections).

Panel A: merged staining of anti-Carboxypeptidase A (ab278044, magenta; Opal<sup>™</sup>690), anti-Cytokeratin 19 (ab195872, green; Opal<sup>™</sup>520) and anti-Insulin (ab181547, red; Opal<sup>™</sup>570) on human pancreas. Panel B: anti-Carboxypeptidase A stained on acinar cells. Panel C: anti-Cytokeratin 19 stained on centroacinar cells and ducts. Panel D: anti-Insulin stained on beta cells. Opal Polymer HRP Ms + Rb was used as a secondary antibody.

The section was incubated in three rounds of staining: in the order of <u>ab278044</u> at 1/4000 dilution (0.135  $\mu$ g/ml), <u>ab195872</u> at 1/8000 dilution (0.127  $\mu$ g/ml), and ab181547 at 1/20000 dilution (0.053  $\mu$ g/ml) for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND<sup>®</sup> RX instrument with an Opal<sup>™</sup> 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins. DAPI (blue) was used as a nuclear counter stain.

1 2 3

250 kDa —
150 kDa —
150 kDa —
100 kDa —
75 kDa —
37 kDa —
25 kDa —
20 kDa —
15 kDa —
10 kDa —
10 kDa —
10 kDa —

Western blot - Anti-Insulin antibody [EPR17359] (ab181547)

**All lanes :** Anti-Insulin antibody [EPR17359] (ab181547) at 1/1000 dilution

Lane 1: Human pancreas

Lane 2: Mouse pancreas

Lane 3: Rat pancreas

Lysates/proteins at 20 µg per lane.

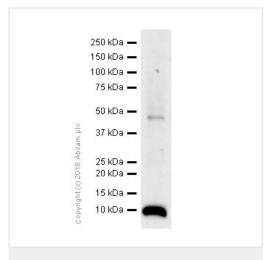
# Secondary

**All lanes :** Goat Anti-Rabbit  $\lg G$  (HRP) with minimal cross-reactivity with human  $\lg G$  at 2000  $\mu g/ml$ 

Predicted band size: 12 kDa

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

Observed MW: 12KDa



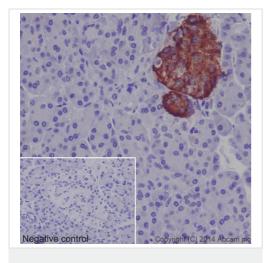
Western blot - Anti-Insulin antibody [EPR17359] (ab181547)

Anti-Insulin antibody [EPR17359] (ab181547) at 1/1000 dilution + Mouse pancreas lysate 20  $\mu g$  at 20  $\mu g$ 

# **Secondary**

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

Predicted band size: 12 kDa Observed band size: 12 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Insulin antibody
[EPR17359] (ab181547)

Immunohistochemical analysis of paraffin-embedded human pancreas tissue labeling Insulin with ab181547 at 1/64000 dilution, followed by Anti-Rabbit HRP (ab97051) at 1/500 dilution.

Cytoplasm staining on islet cells of human pancreas is observed. Counterstained with hematoxylin.

**Negative control:** PBS instead of primary antibody; secondary antibody is Anti-Rabbit HRP (<u>ab97051</u>) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Immunocytochemistry/ Immunofluorescence - Anti-

Immunohistochemical analysis of 4% paraformaldehyde perfusion fixed, frozen section of mouse pancreas tissue labeling Insulin with ab181547 at 1/1000 dilution, followed by Donkey anti-rabbit Alexa Fluor® 594 at 1/1000 dilution. Cytoplasm staining on islet cells of mouse pancreas is observed. Counter stained with DAPI.

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1%

adenocarcinoma cells) cells labeling Insulin with ab181547 at 1/200

Confocal image shows cytoplasmic staining on BxPC-3 cells. The nuclear counterstain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/500 dilution and ab150120 (Alexa Fluor® 594 Goat anti-Mouse secondary) at 1/500 dilution (red).

-ve control 1: - ab181547 at 1/200 dilution followed by ab150120

-ve control 2: - ab7291 (anti-Tubulin mouse mAb) at 1/500 dilution followed by <u>ab150077</u> (Alexa Fluor<sup>®</sup>488 Goat Anti-Rabbit lgG H&L)

(Alexa Fluor® 594 Goat anti-Mouse secondary) at 1/500 dilution.

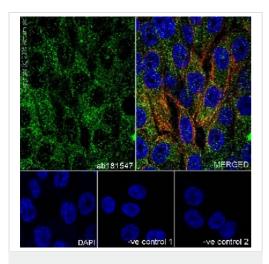
Triton X-100 permeabilized BxPC-3 (Human pancreas

The negative controls are as follows:-

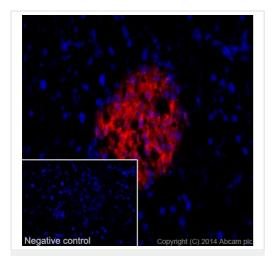
at 1/400 dilution.

dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/400 dilution (green).

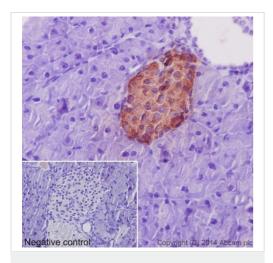
Negative control: PBS instead of primary antibody; secondary antibody is Donkey anti-rabbit Alexa Fluor® 594 at 1/1000 dilution.



Insulin antibody [EPR17359] (ab181547)



Immunohistochemistry (PFA perfusion fixed frozen sections) - Anti-Insulin antibody [EPR17359] (ab181547)



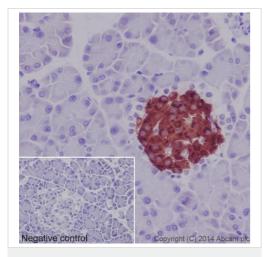
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Insulin antibody
[EPR17359] (ab181547)

Immunohistochemical analysis of paraffin-embedded mouse pancreas tissue labeling Insulin with ab181547 at 1/64000 dilution, followed by Anti-Rabbit HRP (ab97051) at 1/500 dilution.

Cytoplasm staining on islet cells of mouse pancreas is observed. Counterstained with hematoxylin.

**Negative control:** PBS instead of primary antibody; secondary antibody is Anti-Rabbit HRP (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

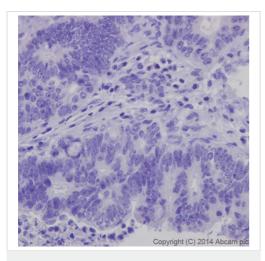


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Insulin antibody
[EPR17359] (ab181547)

Immunohistochemical analysis of paraffin-embedded rat pancreas tissue labeling Insulin with ab181547 at 1/64000 dilution, followed by Anti-Rabbit HRP (<u>ab97051</u>) at 1/500 dilution. Cytoplasm staining on islet cells of rat pancreas is observed. Counterstained with hematoxylin.

**Negative control:** PBS instead of primary antibody; secondary antibody is Anti-Rabbit HRP (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

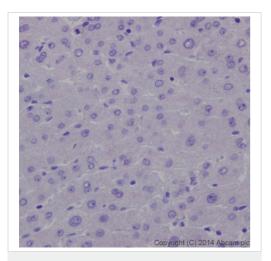


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Insulin antibody [EPR17359] (ab181547)

Immunohistochemical analysis of paraffin-embedded human adenocarcinoma of colon tissue with ab181547 at 1/64000 dilution, followed by Anti-Rabbit HRP (ab97051) at 1/500 dilution.

Negative staining on human colonic adenocarcinoma is observed. Counterstained with hematoxylin.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

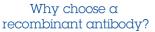


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Insulin antibody [EPR17359] (ab181547)

Immunohistochemical analysis of paraffin-embedded human liver tissue with ab181547 at 1/64000 dilution, followed by Anti-Rabbit HRP (ab97051) at 1/500 dilution.

Negative staining on human liver tissue is observed. Counterstained with hematoxylin.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.





confidence Consistent and reproducible results







production

Anti-Insulin antibody [EPR17359] (ab181547)

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