Anti-Islet 1 Antibody [SC05-64] - BSA and Azide free HA750211

Product Type: Recombinant Rabbit monoclonal IgG, primary antibodies Species reactivity: Human, Mouse, Rat, Cynomolgus monkey, Pig Applications: WB, IF-Cell, IHC-P, FC, IP, IHC-Fr Molecular Wt: Predicted band size: 39 kDa Clone number: SC05-64 Description: Islel-1 (ISL1 transcription factor, LIM/homeodomain) and Islel-2 (ISL2 transcription factor, LIM/homeodomain) contain amino-terminal LIM domains and a carboxy-terminal homeodomain and both influence developmental events. Islel-1 influences embryogenesis of the pancreatic islets of Langerhans and neural tube motor neuron differentiation. In developing mouse teeth, Islel-1 mediate patterning of dentition as an activator of Bmp4 expression in incisor (distal) areas of the stomatodeal epithelium. Islet-1 expression defines cardiac progenitor cell populations and is required for normal cardiac development and asymmetry. Isle-2 activity in newly generated motor neurons parmits the diversification of visceral and somatic motor neuron subtypes in the developing spinal cord. Murine Islet-2 specifies retinal ganglion cell (RGC) laterality by repressing an ipsilateral pathfinding program unique to the ventral-temporal crescent (VTC) of RGCs in a Zic2- and EphB1-dependent manner. Immunogen: Synthetic peptide within Human Islet 1 aa 262-205 / 349. Positive control: SH-SYSY cell lysate, Neuro-2a cell lysate, PC-12 cell lysate, SH-SYSY, Neuro-2a, human pancreas tissue. Subcellular location: Nucleus. Database links: SwissProt: P61371 Human P61372 Mouse P61374 Rat<		
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Purity: Protein A affinity purified.	-	Store at +4 $^\circ\!\!\!{\rm C}$ after thawing. Aliquot store at -20 $^\circ\!\!\!{\rm C}$ or -80 $^\circ\!\!\!{\rm C}$. Avoid repeated freeze / thaw
	Purity:	Protein A affinity purified.

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Images

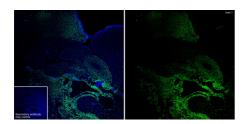


Fig1: Application: IHC-Fr

Species: Mouse

Site: E14.5 embryo

Sample: Frozen section

Antibody concentration: 1:500

Antigen retrieval: The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven.

Fig2: Application: IHC-Fr

Species: Mouse

Site: E14.5 embryo

Sample: Frozen section

Antibody concentration: 1:500

Antigen retrieval: The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven.

Fig3: Application: IHC-Fr

Species: Mouse

Site: E14.5 embryo

Sample: Frozen section

Antibody concentration: 1:500

Antigen retrieval: The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven.

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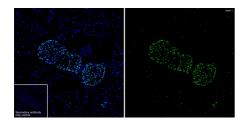


Fig4: Application: IHC-Fr

Species: Mouse

Site: Pancreas

Sample: Frozen section

Antibody concentration: 1:500

Antigen retrieval: The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven.

Fig5: Western blot analysis of Islet 1 on different lysates with Rabbit anti-Islet 1 antibody (HA750211) at 1/2,000 dilution.

Lane 1: SH-SY5Y cell lysate (5 µg/Lane) Lane 2: Neuro-2a cell lysate (30 µg/Lane) Lane 3: PC-12 cell lysate (30 µg/Lane)

Predicted band size: 39 kDa Observed band size: 45 kDa

Exposure time: Lane 1: 10 seconds; Lane 2-3: 1 minute; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA750211) at 1/2,000 dilution was used in 5% NFDM/TBST at 4° C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

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Islet 1

- - GAPDH

45kDa

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250

150-100-75-

55

45

35-25-

14

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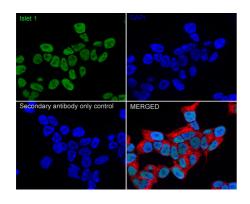


Fig6: Immunocytochemistry analysis of SH-SY5Y cells labeling Islet 1 with Rabbit anti-Islet 1 antibody (HA750211) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Islet 1 antibody (HA750211) at 1/100 dilution in 1% BSA in PBST overnight at 4 $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluorTM 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4 °C. Goat Anti-Mouse IgG H&L (iFluor ™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

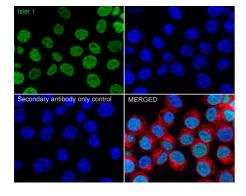


Fig7: Immunocytochemistry analysis of Neuro-2a cells labeling Islet 1 with Rabbit anti-Islet 1 antibody (HA750211) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Islet 1 antibody (HA750211) at 1/100 dilution in 1% BSA in PBST overnight at 4 $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluorTM 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

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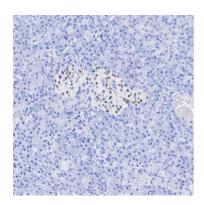


Fig8: Immunohistochemical analysis of paraffin-embedded human pancreas tissue with Rabbit anti-Islet 1 antibody (HA750211) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750211) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

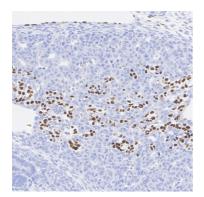


Fig9: Immunohistochemical analysis of paraffin-embedded mouse embryo tissue with Rabbit anti-Islet 1 antibody (HA750211) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750211) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

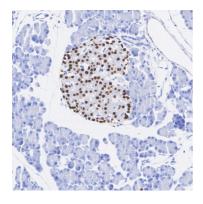


Fig10: Immunohistochemical analysis of paraffin-embedded rat pancreas tissue with Rabbit anti-Islet 1 antibody (HA750211) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750211) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

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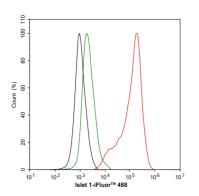


Fig11: Flow cytometric analysis of SH-SY5Y cells labeling Islet 1.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA750211, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4°C for an hour, the cells were stained with a iFluorTM 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

- 1. Beaudet MJ et al. High yield extraction of pure spinal motor neurons, astrocytes and microglia from single embryo and adult mouse spinal cord. Sci Rep 5:16763 (2015).
- 2. Wrighton PJ et al. Signals from the surface modulate differentiation of human pluripotent stem cells through glycosaminoglycans and integrins. Proc Natl Acad Sci U S A 111:18126-31 (2014).

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Celt=Immunofluorescence (Celt) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

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