

## Technical Data Sheet

## Purified Mouse anti-Islet-1

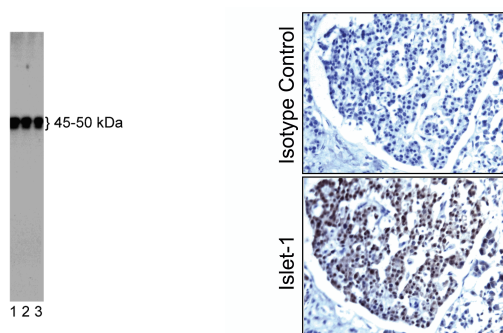
## Product Information

<b>Material Number:</b>	<b>562546</b>
<b>Alternate Name:</b>	ISL-1, ISL1, Islet-2, ISL-2, ISL2
<b>Size:</b>	0.1 mg
<b>Concentration:</b>	0.5 mg/ml
<b>Clone:</b>	Q11-465
<b>Immunogen:</b>	Human Islet-1 Recombinant Protein
<b>Isotype:</b>	Mouse IgG1, $\kappa$
<b>Reactivity:</b>	QC Testing: Mouse Tested in Development: Human
<b>Target MW:</b>	45-50 kDa
<b>Storage Buffer:</b>	Aqueous buffered solution containing $\leq 0.09\%$ sodium azide.

## Description

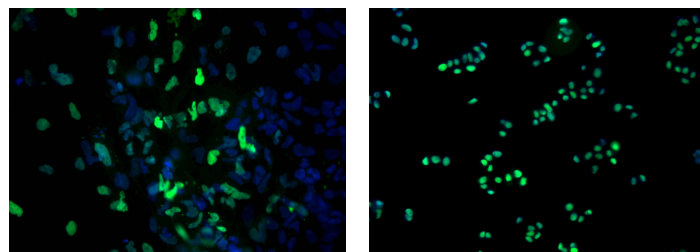
Islet-1 is a LIM-homeodomain transcription factor important for motor neuron differentiation and the formation of islet cells in the pancreas. Various heart cell types, such as cardiac muscle, the conduction system and endothelial cells in multiple heart tissue compartments during cardiogenesis, have been found to originate from Islet-1-positive cardiac precursor cells. Moreover, Islet-1-positive cells from differentiated human embryonic stem cell lines were found to be capable of self-renewal and expansion and could differentiate into the three major cell types of the heart.

Western blot analyses of lysates from transfected cells demonstrate that the Q11-465 monoclonal antibody reacts with human Islet-1 (ISL-1, ISL1) and Islet-2 (ISL-2, ISL2), similar to the 4D5 clone (Tsuchida et al, 1994). Cross-reactivity with mouse Islet is also observed.



**TOP LEFT: Western Blot analysis of Islet-1 in mouse pancreatic tumor (insulinoma).** Lysate from Beta-TC-6 cells (ATCC CRL-11506™) was probed with Purified Mouse anti-Islet-1 monoclonal antibody at titrations of 0.5 (lane 1), 0.25 (lane 2), and 0.125  $\mu\text{g/ml}$  (lane 3). Islet-1 is identified as a band of ~45-50 kDa.

**TOP RIGHT: Immunohistochemical staining of human Islet-1.** Following antigen retrieval with BD Retrieval A buffer (Cat. No. 550524), formalin-fixed paraffin-embedded human pancreas sections were stained with either purified mouse IgG1,  $\kappa$  Isotype control (Cat. No. 550878, top panel) or Purified Mouse Anti-Human Islet-1 (Cat. No. 562546, bottom panel). A three-step staining procedure that employs Biotin Goat Anti-Mouse Ig secondary antibody (Cat. No. 550337), Streptavidin HRP (Cat. No. 550946), and DAB substrate kit (Cat. No. 550880) was used. As shown in the figure, the Islet-1 staining is mainly nuclear in the pancreas islet. Original magnification: 40X.



**BOTTOM ROW: Immunofluorescent staining of Islet-1 in spontaneously differentiated human embryonic stem (ES) cells (left panel) and mouse pancreatic tumor (insulinoma) cells (right panel).** Spontaneously differentiated (Emre et al., 2010) H9 human ES cells (WiCell, Madison, WI) and Beta-T-C6 cells (ATCC, CRL-11506™) were fixed with BD Cytotfix™ Fixation Buffer (Cat. No. 554655), permeabilized with BD Phosflow™ Perm Buffer III (Cat. No. 558050), and stained with Purified Mouse anti-Islet-1 monoclonal antibody (pseudo-colored green) at 1.25  $\mu\text{g/mL}$ . The second-step reagent was Alexa Fluor® 488 goat anti-mouse Ig (Life Technologies). Nuclear staining (pseudo-colored blue) was either with DAPI (left panel) or Hoechst 33342 (Cat. No. 561908, right panel). The images were captured on a BD Pathway™ 435 Cell Analyzer and merged using BD AttoVision™ Software.

## Preparation and Storage

Store undiluted at 4°C.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

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## Application Notes

### Application

Western blot	Routinely Tested
Immunohistochemistry-formalin (antigen retrieval required)	Tested During Development
Bioimaging	Tested During Development
Immunofluorescence	Tested During Development
Intracellular staining (flow cytometry)	Tested During Development

### Recommended Assay Procedure:

For more information please refer to the resources located on the BD Biosciences webpage:

<http://www.bdbiosciences.com/support/resources/index.jsp>

### Suggested Companion Products

Catalog Number	Name	Size	Clone
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)
554655	Fixation Buffer	100 ml	(none)
558050	Perm Buffer III	125 ml	(none)
561908	Hoechst 33342 Solution	1.0 mg	(none)
353219	BD Falcon™ 96-well Imaging Plate		(none)
550524	Retrievagen A (pH 6.0)	1000 ml	(none)
550878	Purified Mouse IgG1 κ Isotype Control	1.0 ml	MOPC-31C
550337	Biotin Goat Anti-Mouse Ig (Multiple Adsorption)	1.0 ml	Polyclonal
550946	Streptavidin HRP	50 ml	(none)
550880	DAB Substrate Kit	500 tests	(none)

### Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to [www.bdbiosciences.com/pharming/protocols](http://www.bdbiosciences.com/pharming/protocols) for technical protocols.
3. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
4. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
5. Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.

### References

Bu L, Jiang X, Martin-Puig S, et al. Human ISL1 heart progenitors generate diverse multipotent cardiovascular cell lineages. *Nature*. 2009; 460(7251):113-117. (Biology)

Ebert AD, Yu J, Rose FF Jr, et al. Induced pluripotent stem cells from a spinal muscular atrophy patient. *Nature*. 2009; 457(7227):277-280. (Biology)

Emre N, Vidal JG, Elia J, et al. The ROCK inhibitor Y-27632 improves recovery of human embryonic stem cells after fluorescence-activated cell sorting with multiple cell surface markers. *PLoS ONE*. 2010; 5(8):e12148. (Methodology: Cell differentiation)

Laugwitz KL, Moretti A, Caron L, Nakano A, Chien KR. Islet1 cardiovascular progenitors: a single source for heart lineages. *Development*. 2008; 135(2):193-205. (Biology)

Osumi N, Hirota A, Ohuchi H, et al. Pax-6 is involved in the specification of hindbrain motor neuron subtype. *Development*. 1997; 124(15):2961-2972. (Biology)

Tsuchida T, Ensini M, Morton SB, et al. Topographic organization of embryonic motor neurons defined by expression of LIM homeobox genes. *Cell*. 1994; 79(6):957-970. (Biology)

Xu C, Police S, Hassanipour M, et al. Efficient generation and cryopreservation of cardiomyocytes derived from human embryonic stem cells. 2011; 6(1):53-66. (Biology)

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