Human/Mouse Proinsulin Biotinylated Antibody

Monoclonal Mouse IgG_{2A} Clone # 253627

Catalog Number: BAM13361

DESCRIPTION

Species Reactivity	Human/Mouse
Specificity	Detects human and mouse Proinsulin in direct ELISAs. In direct ELISAs, no cross-reactivity with mature Insulin is observed.
Source	Monoclonal Mouse IgG_{2A} Clone # 253627
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	*E. coli*-derived recombinant human Proinsulin Phe25-Asn110 Accession # P01308
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with BSA as a carrier protein. See Certificate of Analysis for details.

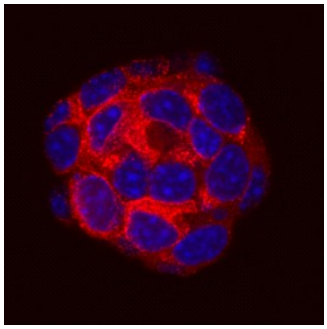
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Immunocytochemistry	8-25 µg/mL	See Below
Intracellular Staining by Flow Cytometry	2.5 µg/10^6 cells	βTC-6 mouse beta cell insulinoma cell line fixed with paraformaldehyde and permeabilized with saponin

DATA

Immunocytochemistry



Proinsulin in βTC-6 Mouse Cell Line. Proinsulin was detected in immersion fixed βTC-6 mouse beta cell insulinoma cell line using Mouse Anti-Human/Mouse Proinsulin Biotinylated Monoclonal Antibody (Catalog # BAM13361) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Streptavidin (red; Catalog # NL999) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

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BACKGROUND

Proinsulin is synthesized as a single chain, 110 amino acid (aa) preproprecursor that contains a 24 aa signal sequence and an 86 aa proinsulin propeptide. Following removal of the signal peptide, the proinsulin peptide undergoes further proteolysis to generate mature insulin, a 51 aa disulfide-linked dimer that consists of a 30 aa B chain (aa 25-54) bound to a 21 aa A chain (aa 90-110). The 34 aa intervening peptide (aa 55-89) that connects the B and A chains is termed the C-peptide. Human proinsulin shares 84% and 80% aa sequence identity with rat and bovine proinsulin, respectively. Most of the sequence variation between species occurs in the region of the C-peptide (1). This peptide generates a structural conformation that allows for the correct formation of the intrachain disulphide bonds (1). Insulin is a molecule that facilitates the cellular uptake of glucose. This is accomplished by regulating the appearance of membrane glucose transporters. Low insulin levels or lack of insulin are associated with type 2 and type 1 diabetes mellitus, respectively. These conditions are associated with an increased risk for microvascular complications such as retinopathy, nephropathy, and peripheral neuropathy (3). Proinsulin also circulates, but its physiologic role is less well understood. It does possess about 25% of the activity of mature insulin, but it would seem unlikely to be a natural substitute for insulin (4). In type 2 diabetes, an elevated proinsulin to insulin ratio in the circulation is a well-known abnormality (5-9). Perhaps this abnormality represents either compromised proteolytic processing or a general inability to process increased levels of insulin precursor (5). In any event, proinsulin will stimulate amylin secretion by β-cells, and amyloid formation in pancreatic islets that promotes decreased β cell function (10). Studies also suggest that fasting serum proinsulin may be a better predictor of future type 2 diabetes than fasting insulin levels in obese children (11).

References:

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