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Datasheet for 200-301-A33

GAPDH Antibody

Overview

Description:	Anti-Glyceraldehyde-3-Phosphate Dehydrogenase (GAPDH) (MOUSE) Monoclonal Antibody - 200-301-A33
Item No.:	200-301-A33
Size:	100 µg
Applications:	ELISA, Multiplex, WB
Reactivity:	Human, Mouse, Rabbit
Host Species:	Mouse

Product Details

Background:	GAPDH Loading Control Monoclonal Antibody detects Glyceraldehyde-3-Phosphate Dehydrogenase. GAPDH is a metabolic enzyme responsible for catalyzing one step in the glycolytic pathway, the reversible oxidative phosphorylation of glyceraldehyde 3-phosphate. Because GAPDH is a protein expressed in large amounts and which is required at all times for important "house keeping" functions, levels of GAPDH mRNA are often measured and used as standards in studies of mRNA expression. Increasingly, scientists are making use of specific antibodies to GAPDH in comparable studies of levels of protein expression. Apart from a role in glycolysis, GAPDH may have other roles such as in the activation of transcription. GAPDH is reported to bind to a variety of other proteins, including the amyloid precursor protein, mutations in which cause some forms of Alzheimer's disease, and the polyglutamine tracts of Huntingtin, the protein product aberrant forms of which are causative of Huntington's disease. Associations with actin and tubulin have also been reported. The protein may also have a role in the regulation of apoptosis, and interestingly migrates from the cytoplasm into the nucleus when cells become apoptotic. Anti-GAPDH antibody is widely used as a loading control for western blotting experiments, allowing comparison between the level of this protein and others in a cell or tissue and is ideal for Neuroscience, Cell Signaling and Cancer Research.
Synonyms:	mouse anti-GAPDH antibody, Loading Control Antibody, G3PDH, GAPD, Glyceraldehyde-3-phosphate dehydrogenase, Peptidyl-cysteine S-nitrosylase GAPDH
Host Species:	Mouse
Clonality:	Monoclonal
Clone ID:	33F2.C8.G6.E3
Format:	IgM

Target Details

Gene Name:	GAPDH
Reactivity:	Human, Mouse, Rabbit
Immunogen Type:	Native Protein
Immunogen:	Anti-GAPDH Monoclonal Antibody was produced by repeated immunizations in mice with rabbit GAPDH protein.
Purity/Specificity:	Anti-GAPDH Monoclonal Antibody is directed against rabbit Glyceraldehyde-3-Phosphate Dehydrogenase. The antibody is a total IgM fraction prepared from ascites fluid by selective precipitation and tandem molecular sieve chromatography followed by extensive dialysis against the buffer stated above. Anti-GAPDH antibodies are expected to react with the following species based on 100% sequence homology: rabbit, human, and mouse. Reactivity against other sources is not known.
Relevant Links:	• UniProtKB - P46406 • NCBI - NP_001075722.1 • GeneID - 100009074

Application Details

Tested Applications:	ELISA, Multiplex, WB
Application Note:	Anti-GAPDH (Mouse) has been tested in ELISA and western blot. This product is suitable in IHC and IF. Specific conditions should be optimized by the end user. Expect a band of ~38kDa in size corresponding to Glyceraldehyde 3-Phosphate Dehydrogenase protein by Western blot in the appropriate cell lysate or extract.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	User Optimized
IF:	1:100
IHC:	User Optimized
IP:	User Optimized
WB:	1:1,000

Formulation

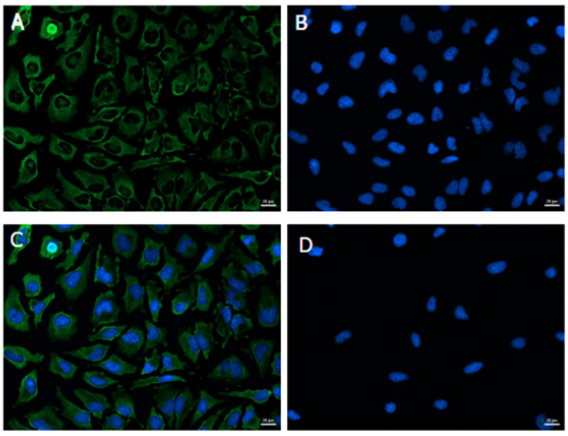
Physical State:	Liquid (sterile filtered)

Concentration:	1.0 mg/ml by UV absorbance at 280 nm
Buffer:	0.1 M Tris Chloride, 0.5 M Sodium Chloride, pH 8.0
Preservative:	0.1% (w/v) Sodium Azide
Stabilizer:	None

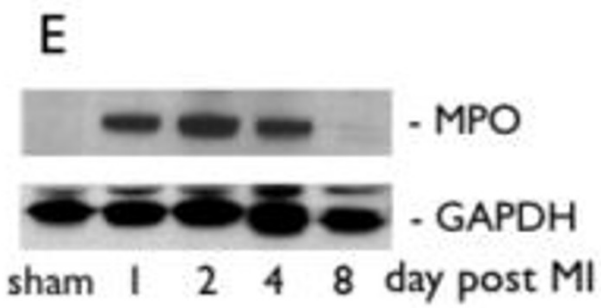
Shipping & Handling

Shipping Condition:	Dry Ice
Storage Condition:	Store vial at -20° C prior to opening. This product is stable at 4° C as an undiluted liquid. For extended storage, aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing. Dilute only prior to immediate use.
Expiration:	Expiration date is one (1) year from date of receipt.

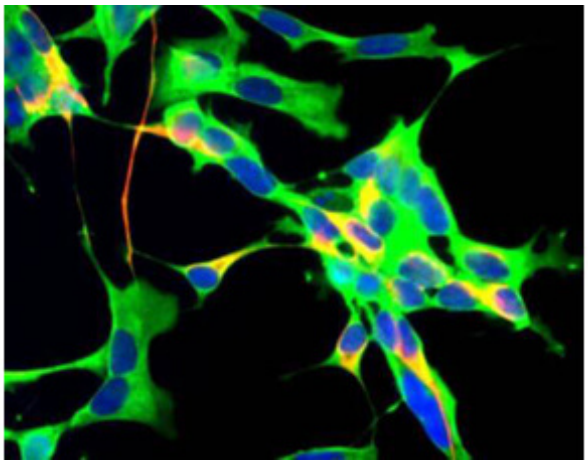
Images



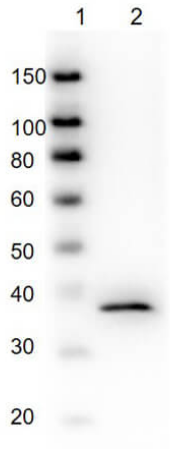
Immunofluorescence Microscopy
Immunofluorescence Microscopy of Mouse Anti-Glyceraldehyde 3-Phosphate Dehydrogenase Antibody.
Cells: HeLa cells.
Fixation: Ice Cold Methanol.
Permeabilization: Ice Cold Methanol.
Primary antibody: GAPDH antibody at 4µg/mL (1:250) overnight at 4°C.
Secondary antibody: Goat anti-Mouse DyLight™488 (p/n 610 -141-121) at 1:1,000 for 1hr at RT.
Localization: GAPDH is cytoplasmic.
Staining: (A) Anti-GAPDH (green), (B) Hoechst Nuclear stain, (C) merge, (D) secondary only and Hoechst Nuclear stain.
Magnification Objective: 20x.



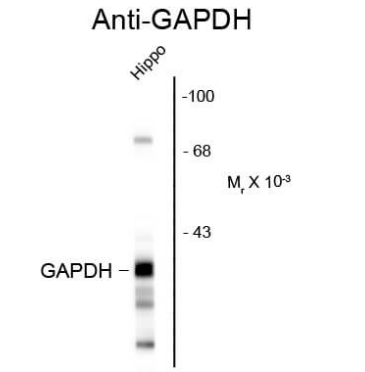
Western Blot
Western blots show no MPO in the hearts of sham operated mice, strong staining on day 1-4 post MI, and low signal on day 8 after coronary ligation, corroborating the in vivo MRI and tissue activity assay data. Fig 5E: PMID: 18268141



Immunofluorescence Microscopy
Immunofluorescence Microscopy of Mouse Anti-Glyceraldehyde 3-Phosphate Dehydrogenase Antibody. Tissue: Human neuroblastoma SH-SY5Y cells. Fixation: 0.5% PFA. Antigen retrieval: not required. Primary antibody: GAPDH antibody at 10 µg/mL for 1 h at RT. Secondary antibody: Fluorescein mouse secondary antibody at 1:10,000 for 45 min at RT. Localization: GAPDH is cytoplasmic. Staining: Anti-GAPDH (green), chicken antibody to neurofilament NF-H (red) and DNA (blue).



Western Blot
Western Blot of Mouse Anti-GAPDH Antibody. Lane 1: Molecular Weight. Lane 2: Glyceraldehyde-3-Phosphate-Dehydrogenase. Primary Antibody: Mouse Anti-GAPDH Antibody at 1µg/mL. Secondary Antibody: Anti-Mouse IgG HRP. Block: BlockOut Buffer (p/n MB-073). Predicted MW: ~38kDa.



Western blot of rat hippocampal lysate showing specific immunolabeling of the ~38k GAPDH protein.

Western Blot

Western Blot of Mouse anti-Glyceraldehyde-3-Phosphate Dehydrogenase antibody. Lane 1: rat hippocampal lysate. Lane 2: none. Load: 10 µg per lane. Primary antibody: GAPDH antibody at 1:400 for overnight at 4°C. Secondary antibody: IRDye800™ mouse secondary antibody at 1:10,000 for 45 min at RT. Block: 5% BLOTTO overnight at 4°C. Predicted/Observed size: ~38kDa/~38kDa for GAPDH protein. Other band(s): splice variants and isoforms.

References

- Matthias Nahrendorf et al. Activatable magnetic resonance imaging agent reports myeloperoxidase activity in healing infarcts and noninvasively detects the antiinflammatory effects of atorvastatin on ischemia-reperfusion injury. *Circulation.* (2008)

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