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Monoclonal Antibody to 6xHistidine Epitope Tag (HHHHHH) - Purified

Alternate names: 6xHis-Tag, HHHHHH Tag, HIS6 Tag, His Tag

Catalog No.: AM33047PU-N

Quantity: 0.1 mg

Concentration: 0.5 mg/ml

Background: Plasmid vectors for the expression of coding regions of eukaryotic genes in bacterial, insect and mammalian hosts are in common usage; such expression vectors are frequently used to encode hybrid fusion proteins consisting of a eukaryotic target protein and a specialized region designed to aid in the purification and visualization of the target protein. A system that has proven to be very successful relies on the insertion of a six histidine (6xHis) sequence in the N-terminus of the encoded protein, allowing for efficient coupling to Ni -chelating resins and purification by single step affinity chromatography. Visualization of such fusion proteins can be achieved by utilizing antibodies generated against specific peptide sequences downstream from the multiple cloning site.

Host / Isotype: Mouse / IgG1

Recommended Isotype Controls: AM03095PU-N

Clone: J099B12

Immunogen: KLH-conjugated His-tag peptide.

Format:
State: Liquid purified IgG fraction
Purification: Affinity Chromatography
Buffer System: PBS, pH 7.2
Preservatives: 0.09% Sodium Azide

Applications:
Western blotting: Each lot of this antibody is quality control tested.
Use 0.1-0.2 µg/ml antibody dilution buffer for each mini-gel.
Immunofluorescence: Use 0.5-1.0 µg/ml antibody dilution buffer.
Flow Cytometric analysis of intracellularly-stained cells.
Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.

Specificity: This *J099B12* Monoclonal antibody was raised against linear 8XHis tag. It reacts with His tag protein, either C, N, or internal His6 and up repeats.
It can be used for Western blotting to detect linear His repeat (His6 or more). It can also be used for Intracellularly Flow Cytometry and Immunofluorescence staining of His-Tagged protein.

Storage:

Store undiluted at 2-8°C.
DO NOT FREEZE!
Shelf life: one year from despatch.

Product Citations:

Originator or purchased from resellers:

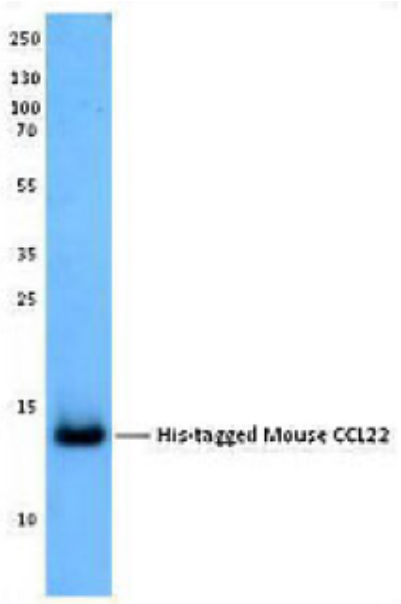
1. Burritt JB, Quinn MT, Jutila MA, Bond CW, Jesaitis AJ. Topological mapping of neutrophil cytochrome b epitopes with phage-display libraries. J Biol Chem. 1995 Jul 14;270(28):16974-80. PubMed PMID: 7622517.
2. Taylor RM, Jesaitis AJ. Immunoaffinity purification of human phagocyte flavocytochrome b and analysis of conformational dynamics. Methods Mol Biol. 2007;412:429-37. doi: 10.1007/978-1-59745-467-4_26. PubMed PMID: 18453126.

General Readings:

1. Tymms MJ, Ng AY, Thomas RS, Schutte BC, Zhou J, Eyre HJ, et al. A novel epithelial-expressed ETS gene, ELF3: human and murine cDNA sequences, murine genomic organization, human mapping to 1q32.2 and expression in tissues and cancer. Oncogene. 1997 Nov 13;15(20):2449-62. PubMed PMID: 9395241.
2. Hou CL, Tang Cj, Roffler SR, Tang TK. Protein 4.1R binding to eIF3-p44 suggests an interaction between the cytoskeletal network and the translation apparatus. Blood. 2000 Jul 15;96(2):747-53. PubMed PMID: 10887144.

Pictures:

Insect cell supernanant expressing His-tagged Mouse CCL22 was resolved by electrophoresis, transferred to nitrocellulose, and probed with anti-His Tag antibody (clone *J099B12*). Proteins were visualized using a Goat anti-Mouse IgG secondary conjugated to HRP and chemiluminescence detection.



293E cells transfected with His-tagged human pro-IL1β were probed with anti-His Tag antibody (clone *J099B12*). The cells were then detected by DyLight™ 594 conjugated anti-Mouse IgG1 secondary (cytosol, red) and DAPI (nuclei, blue).