Human Osteoactivin/GPNMB Antibody

Monoclonal Mouse IgG_{2B} Clone # 303822

Catalog Number: MAB25501

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Osteoactivin/GPNMB in direct ELISAs and Western blots. In direct ELISAs, no cross-reactivity with recombinant mouse Osteoactivin is observed.
Source	Monoclonal Mouse IgG_{2B} Clone # 303822
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Mouse myeloma cell line NS0-derived recombinant human Osteoactivin/GPNMB isoform 2 Ala22-Asn486 Accession # NP_002501
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 μm filtered solution in PBS.

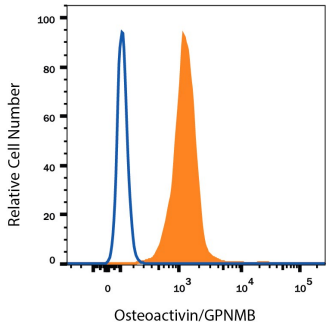
APPLICATIONS

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
Western Blot	1 µg/mL	Recombinant Human Osteoactivin/GPNMB Fc Chimera (Catalog # 2550-AC) under non-reducing conditions only
Immunocytochemistry	8-25 µg/mL	Immersion fixed MG-63 human osteosarcoma cell line
Intracellular Staining by Flow Cytometry	0.25 µg/10^6 cells	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

DATA

Intracellular Staining by Flow Cytometry



Detection of Osteoactivin/GPNMB in MG-63 Human Cell Line by Flow Cytometry. MG-63 human osteosarcoma cell line was stained with Mouse Anti-Human Osteoactivin/GPNMB Monoclonal Antibody (Catalog # MAB25501, filled histogram) or isotype control antibody (Catalog # MAB004, open histogram), followed by Phycoerythrin-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0102B). To facilitate intracellular staining, cells were fixed with Flow Cytometry Fixation Buffer (Catalog # FC004) and permeabilized with Flow Cytometry Permeabilization/Wash Buffer I (Catalog # FC005). View our protocol for Staining Intracellular Molecules.

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. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
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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Human Osteoactivin/GPNMB Antibody

Monoclonal Mouse IgG_{2B} Clone # 303822

Catalog Number: MAB25501

BACKGROUND

Osteoactivin (also GPNMB and DC-HIL) is a variably glycosylated 75-125 kDa member of the NMB/pMEL-17 family of molecules. It is found in multiple subcellular sites, but is most often associated with the endosomal/lysosomal compartment (1-3). Human Osteoactivin is a 560 amino acid (aa) type I transmembrane protein. Its precursor contains a 21 aa signal sequence, a 465 aa luminal/extracellular domain, a 21 aa transmembrane segment and a 53 aa cytoplasmic tail (4, 5). The luminal region contains an N-terminal heparin-binding motif (aa 23-26), multiple glycosylation sites, an RGD motif (aa 64-66) and an 88 aa PKD domain (aa 240-327). The intracellular tail has an ITAM (Y-x-x-I) and lysosomal targeting (L-L) motif (4, 5). The extracellular/luminal region shares 74% and 77% aa identity with the equivalent regions in mouse and canine, respectively. Multiple isoforms would appear to exist. There is one alternate splice form known that shows a 12 aa insert between aa 339-340 (6). An addtional 206 aa isoform shows a mutation at position # 181 that results in a 26 aa substitution for the C-terminal 380 amino acids (7, 8). This has the potential to be soluble and may represent a counterpart to a secreted isoform of rat Osteoactivin (9). Cells known to express Osteoactivin include macrophages/Kupffer cells, fibroblasts, osteoblasts, myeloid dendritic cells, retinal pigment epithelial cells and melanocytes, plus fetal chondrocytes and stratum basale keratinocytes (3-5, 10-12). In mice, Osteoactivin is reported to bind to heparan sulfate-proteoglycan, possibly on the surface of endothelial cells and may also interact with integrins (13). It also appears to act as an inflammatory suppressor gene, as its expression downregulates the macrophage inflammatory response by inhibiting IL-6 and IL-12 p40 production (3).

References:

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