Human Indoleamine 2,3-dioxygenase/IDO Antibody

Monoclonal Mouse IgG_{2B} Clone # 998736

Catalog Number: MAB60302

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Indoleamine 2,3-dioxygenase/IDO in direct ELISAs.
Source	Monoclonal Mouse IgG_{2B} Clone # 998736
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	*E. coli*-derived human Indoleamine 2,3-dioxygenase/IDO Ala2-Gly403 Accession # P14902
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 either lyophilized or as a 0.2 μm filtered solution in PBS.

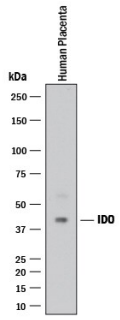
APPLICATIONS

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	2 µg/mL	See Below
Immunohistochemistry	5-25 µg/mL	See Below
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.	

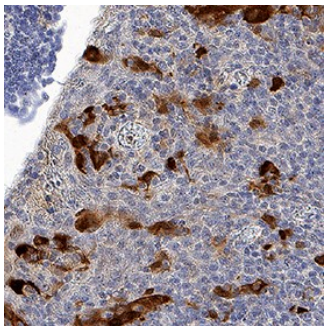
DATA

Western Blot



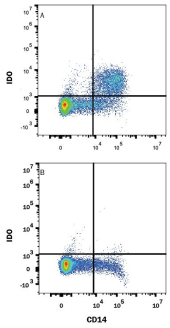
Detection of Human Indoleamine 2,3-dioxygenase/IDO by Western Blot. Western blot shows lysates of human placenta tissue. PVDF membrane was probed with 2 µg/mL of Mouse Anti-Human Indoleamine 2,3-dioxygenase/IDO Monoclonal Antibody (Catalog # MAB60302) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for Indoleamine 2,3-dioxygenase/IDO at approximately 45 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunohistochemistry



Indoleamine 2,3-dioxygenase/IDO in Human Tonsil Tissue. Indoleamine 2,3-dioxygenase/IDO was detected in immersion fixed paraffin-embedded sections of human tonsil tissue using Mouse Anti-Human Indoleamine 2,3-dioxygenase/IDO Monoclonal Antibody (Catalog # MAB60302) at 5 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Mouse IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC001). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to cytoplasm. View our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

Intracellular Staining by Flow Cytometry



Detection of Indoleamine 2,3-dioxygenase/IDO in Human Monocytes by Flow Cytometry. Human Monocytes were selected from PBMC using MagCellect Human CD14+ Cell Isolation Kit (Catalog # MAGH105) and cultured overnight with (A) 50 ng/mL Recombinant Human MCSF (Catalog # 216-MC), 50 ng/mL Recombinant Human IFNg (Catalog # 285-IF) and 50 ng/mL LPS, or (B) Recombinant Human MCSF alone. Cells were stained with Mouse Anti-Human Indoleamine 2,3-dioxygenase/IDO Monoclonal Antibody (Catalog # MAB60302) followed by APC-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0101B) and Mouse Anti-Human CD14 PE-conjugated Monoclonal Antibody (Catalog # FAB3832P). Quadrant markers were set based on Mouse IgG2B Isotype Control (Catalog # MAB0041). To facilitate intracellular staining, cells were fixed with 1% paraformaldehyde and permeabilized with saponin. View our protocol for Staining Intracellular Molecules.

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Monoclonal Mouse IgG_{2B} Clone # 998736

Catalog Number: MAB60302

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.

BACKGROUND

Indoleamine 2,3-dioxygenase (IDO) is a heme-containing intracellular dioxygenase catalyzing the degradation of the essential amino acid L-tryptophan to N-formyl-kynurenine (1). This degradation is the first and rate-limiting step of the L-kynurenine pathway (2). IDO is widely expressed in dendritic cells, macrophages, microglia, eosinophils, fibroblasts, endothelial cells, and most tumor cells. In immune cells, its expression is mainly induced by cytokines such as IFN-γ, IFN-α, IFN-β, and IL-10. IDO has an antimicrobial function due to its decreasing the availability of the essential amino acid tryptophan in inflammatory environments (3). Recent studies have demonstrated that IDO induces immunosuppression during infection, pregnancy, transplantation, autoimmunity, and neoplasia (3-5).

References:

1. Lewis-Ballester, A. *et al.* (2009) Proc. Natl. Acad. Sci. USA. **106**:17371.
2. Costantino, G. (2009) Expert Opin. Ther. Targets **13**:247.
3. Xu, H. *et al.* (2008) Immunol. Lett. **121**:1.
4. Lob, S. *et al.* (2009) Nat. Rev. Cancer **9**:445.
5. Curti, A. *et al.* (2009) Blood **113**:2394.

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