

Human CTLA-4 Antibody

Monoclonal Mouse IgG_{2B} Clone # 922101

Catalog Number: MAB3251

| DESCRIPTION | | |
|--------------------|---|--|
| Species Reactivity | Human | |
| Specificity | Detects human CTLA-4 in direct ELISAs. | |
| Source | Monoclonal Mouse IgG _{2B} Clone # 922101 | |
| Purification | Protein A or G purified from hybridoma culture supernatant | |
| Immunogen | Chinese hamster ovary cell line CHO-derived recombinant human CTLA-4 Ala37-Phe162 Accession # P16410 | |
| 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. | |

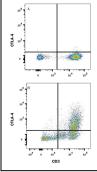
APPLICATIONS

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 |
|--|-------------------------------|-----------|
| Intracellular Staining by Flow Cytometry | 0.25 μg/10 ⁶ cells | See Below |

DATA

Intracellular Staining by Flow Cytometry



Detection of CTLA-4 in Human PBMCs by Flow Cytometry. Human peripheral blood mononuclear cells (PBMCs) either (A) untreated or (B) treated with 5 ug/mL PHA for 2 days were stained with Mouse Anti-Human CTLA-4 Monoclonal Antibody (Catalog # MAB3251) followed by Allophycocyanin-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0101B) and Mouse Anti-Human CD3s PE-conjugated Monoclonal Antibody (Catalog # FAB100P). Quadrant markers were set based on control antibody staining (Catalog # MAB002). 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

| 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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BACKGROUND

CTLA-4 (cytotoxic T-lymphocyte associated protein-4, designated CD152), is a type I transmembrane T cell inhibitory molecule that is a member of the Ig superfamily (1, 2). Human or mouse CTLA-4 cDNA encodes 223 amino acids (aa) including a 35 aa signal sequence, a 126 aa extracellular domain (ECD) with one Iglike V-type domain, a 21 aa transmembrane (TM) sequence, and a 41 aa cytoplasmic sequence. It is found as a covalent homodimer of 41-43 kDa (2) Within the ECD, human CTLA-4 shares 68%, 71% and 83-86% aa sequence identity with mouse, rat and porcine/bovine/rabbit/feline/canine CTLA-4, respectively. A 174 aa form that lacks TM and cytoplasmic sequences (sCTLA-4) is possibly secreted (3-5). Isoforms of 56-79 aa that mainly contain parts of the cytoplasmic domain are reported. In mouse, an isoform lacking the Ig-like domain has ligand-independent inhibitory activity and is termed liCTLA-4 (6). CD28, which is structurally related to CTLA-4, is constitutively expressed on naïve T cells and promotes T cell activation when engaged by B7-2 on antigen-presenting cells (APC) within the immunological synapse (IS) (1, 7, 8). In contrast, CTLA-4 is recruited from intracellular vesicles to the IS beginning 1-2 days after T cell activation (2, 7, 8). It forms a linear lattice with B7-1 on APC, inducing negative regulatory signals and ending T cell activation (9). Abatacept, a therapeutic human CTLA-4-Ig fusion protein (trade name Orencia), competes with CD28 for B7-1 and B7-2 binding and has been used to antagonize T cell activation in autoimmune conditions and to enhance transplant survival (10). Mice deleted for CTLA-4 show no abnormalities until after birth, but then develop lethal autoimmune reactions due to continued T cell activation and poor control by regulatory T cells, which constitutively express CTLA-4 in wild-type mice and humans (11-13).

References:

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