Human LAP (TGF-β1) Antibody

Antigen Affinity-purified Polyclonal Goat IgG

Catalog Number: AF-246-NA

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human LAP TGF-β1 in direct ELISAs and Western blots.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	*S. frugiperda* insect ovarian cell line *Sf* 21-derived and Chinese hamster ovary cell line CHO-derived recombinant human LAP TGF-β1 Leu30-Ser390 Accession # P01137
Endotoxin Level	<0.10 EU per 1 µg of the antibody by the LAL method.
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.

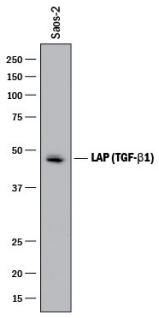
APPLICATIONS

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
Immunocytochemistry	5-15 µg/mL	Immersion fixed human peripheral blood mononuclear cells
Simple Western	20 µg/mL	See Below
Neutralization	Measured by its ability to neutralize LAP TGF-β1 inhibition of TGF-β1 growth inhibition in the HT-2 mouse T cell line. Tsang, M. *et al.* (1995) Cytokine **7**:389. The Neutralization Dose (ND_{50}) is typically 0.4-2 µg/mL in the presence of 500 ng/mL Recombinant Human LAP TGF-β1 and 1 ng/mL TGF-β1.	

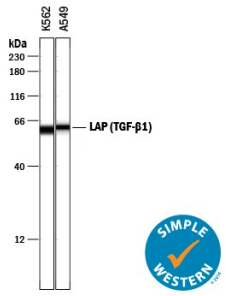
DATA

Western Blot



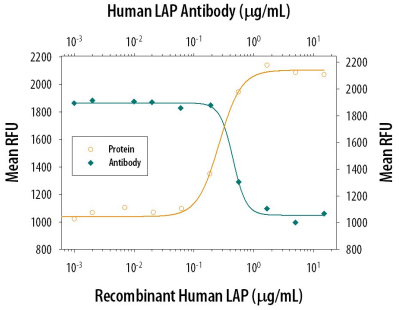
Detection of Human LAP (TGF-β1) by Western Blot. Western blot shows lysates of Saos-2 human osteosarcoma cell line. PVDF membrane was probed with 2 µg/mL of Goat Anti-Human LAP (TGF-β1) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF-246-NA) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for LAP (TGF-β1) at approximately 48 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Simple Western



Detection of Human LAP (TGF-β1) by Simple Western™. Simple Western lane view shows lysates of K562 human chronic myelogenous leukemia cell line and A549 human lung carcinoma cell line, loaded at 0.2 mg/mL. A specific band was detected for LAP (TGF-β1) at approximately 61 kDa (as indicated) using 20 µg/mL of Goat Anti-Human LAP (TGF-β1) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF-246-NA) followed by 1:50 dilution of HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

Neutralization



LAP TGF-β1 Inhibition of TGF-β1 Activity and Neutralization by Human LAP TGF-β1 Antibody. Recombinant Human LAP TGF-β1 (Catalog # 246-LP) inhibits Recombinant Human TGF-β1 (Catalog # 240-B) growth inhibition activity in the HT-2 mouse T cell line in a dose-dependent manner (orange line). Inhibition of Recombinant Human TGF-β1 (1 ng/mL) activity elicited by Recombinant Human LAP TGF-β1 (500 ng/mL) is neutralized (green line) by increasing concentrations of Goat Anti-Human LAP TGF-β1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF-246-NA). The ND_{50} is typically 0.4-2 µg/mL.

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Human LAP (TGF-β1) Antibody

Antigen Affinity-purified Polyclonal Goat IgG

Catalog Number: AF-246-NA

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 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

TGF-β1 (transforming growth factor beta 1) and the closely related TGF-β2 and -β3 are members of the large TGF-β superfamily. TGF-β proteins are highly pleiotropic cytokines that regulate processes such as immune function, proliferation and epithelial-mesenchymal transition (1-3). Human TGF-β1 cDNA encodes a 390 amino acid (aa) precursor that contains a 29 aa signal peptide and a 361 aa proprotein (4). A furin-like convertase processes the proprotein within the trans-Golgi to generate an N-terminal 249 aa (aa 30-278) latency-associated peptide (LAP) and a C-terminal 112 aa mature TGF-β1 (aa 279-390) (4-6). Disulfide-linked homodimers of LAP and TGF-β1 remain non-covalently associated after secretion, forming the small latent TGF-β1 complex (4-8). Purified LAP is also capable of associating with active TGF-β with high affinity, and can neutralize TGF-β activity (9). Covalent linkage of LAP to one of three latent TGF-β binding proteins (LTBPs) creates a large latent complex that may interact with the extracellular matrix (5-7). TGF-β activation from latency is controlled both spatially and temporally, by multiple pathways that include actions of proteases such as plasmin and MMP9, and/or by thrombospondin 1 or selected integrins (5, 8). The LAP portion of human TGF-β1 shares 91%, 92%, 85%, 86% and 88% aa identity with porcine, canine, mouse, rat and equine TGF-β1 LAP, respectively, while mature human TGF-β1 portion shares 100% aa identity with porcine, canine and bovine TGF-β1, and 99% aa identity with mouse, rat and equine TGF-β1. Although different isoforms of TGF-β are naturally associated with their own distinct LAPs, the TGF-β1 LAP is capable of complexing with, and inactivating, all other human TGF-β isoforms and those of most other species (9). Mutations within the LAP are associated with Camurati-Engelmann disease, a rare sclerosing bone dysplasia characterized by inappropriate presence of active TGF-β1 (10).

References:

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