

DESCRIPTION

Specificity	Detects TGF-β3 in direct ELISAs and Western blots. In direct ELISAs, less than 10% cross-reactivity with recombinant amphibian TGF-β5 is observed and less than 5% cross-reactivity with recombinant human (rh) TGF-β1, rhTGF-β2 and rhLAP (TGF-β1) is observed.
Source	Polyclonal Goat IgG
Purification	Protein A or G purified
Immunogen	<i>S. frugiperda</i> insect ovarian cell line Sf 21-derived recombinant chicken TGF-β3 and recombinant human TGF-β3 Ala301-Ser412 (Tyr340Phe) Accession # P10600
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.

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
Simple Western	50 μg/mL	See Below
Neutralization	Measured by its ability to neutralize TGF-β3 inhibition of IL-4-dependent proliferation in the HT-2 mouse T cell line. Tsang, M. <i>et al.</i> (1995) Cytokine 7:389. The Neutralization Dose (ND ₅₀) is typically 1-3 μg/mL in the presence of 0.1 ng/mL Recombinant Human TGF-β3 and 7.5 ng/mL Recombinant Mouse IL-4.	

DATA

Western Blot

Detection of Human TGF-β3 by Western Blot. Western blot shows lysates of human heart tissue and human breast cancer tissue. PVDF membrane was probed with 2 μg/mL of Goat Anti-TGF-β3 Polyclonal Antibody (Catalog # AB-244-NA) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for TGF-β3 at approximately 67 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Simple Western

Detection of Human TGF-β3 by Simple Western™. Simple Western lane view shows lysates of human heart tissue and human breast cancer tissue, loaded at 0.2 mg/mL. A specific band was detected for TGF-β3 at approximately 64 kDa (as indicated) using 50 μg/mL of Goat Anti-TGF-β3 Polyclonal Antibody (Catalog # AB-244-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

TGF-β3 Inhibition of IL-4-dependent Cell Proliferation and Neutralization by TGF-β3 Antibody. Recombinant Human TGF-β3 (Catalog # 243-B3) inhibits Recombinant Mouse IL-4 (Catalog # 404-ML) induced proliferation in the HT-2 mouse T cell line in a dose-dependent manner (orange line). Inhibition of Recombinant Mouse IL-4 (7.5 ng/mL) activity elicited by Recombinant Human TGF-β3 (0.1 ng/mL) is neutralized (green line) by increasing concentrations of Goat Anti-TGF-β3 Polyclonal Antibody (Catalog # AB-244-NA). The ND₅₀ is typically 1-3 μg/mL.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 1 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> • 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-β3 (transforming growth factor beta 3) is one of three closely related mammalian members of the large TGF-β superfamily that share a characteristic cystine knot structure (1-7). TGF-β1, -2 and -3 are highly pleiotropic cytokines that are proposed to act as cellular switches that regulate processes such as immune function, proliferation and epithelial-mesenchymal transition (1-4). Each TGF-β isoform has some non-redundant functions; for TGF-β3, mice with targeted deletion show defects palatogenesis and pulmonary development (2). Human TGF-β3 cDNA encodes a 412 amino acid (aa) precursor that contains a 20 aa signal peptide and a 392 aa proprotein (8). A furin-like convertase processes the proprotein to generate an N-terminal 220 aa latency-associated peptide (LAP) and a C-terminal 112 aa mature TGF-β3 (8, 9). Disulfide-linked homodimers of LAP and TGF-β3 remain non-covalently associated after secretion, forming the small latent TGF-β3 complex (8-10). 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 (9, 10). TGF-β is activated from latency by pathways that include actions of the protease plasmin, matrix metalloproteases, thrombospondin 1 and a subset of integrins (10). Mature human TGF-β3 shows 100%, 99% and 98% aa identity with mouse/dog/horse, rat and pig TGF-β3, respectively. It demonstrates cross-species activity (1). TGF-β3 signaling begins with high-affinity binding to a type II ser/thr kinase receptor termed TGF-β RII. This receptor then phosphorylates and activates a second ser/thr kinase receptor, TGF-β RI (also called activin receptor-like kinase (ALK)-5), or alternatively, ALK-1. This complex phosphorylates and activates Smad proteins that regulate transcription (3, 11, 12). Contributions of the accessory receptors betaglycan (also known as TGF-β RIII) and endoglin, or use of Smad-independent signaling pathways, allow for disparate actions observed in response to TGF-β in different contexts (11).

References:

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