Human Klotho β Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF5889

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Klotho β in direct ELISAs and Western blots. In direct ELISAs, approximately 35% cross-reactivity with recombinant mouse Klotho β is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant human Klotho β Phe53-Leu997 Accession # Q86Z14
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.

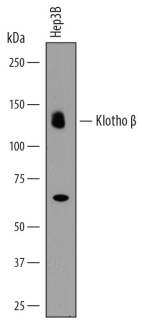
APPLICATIONS

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	0.5 µg/mL	See Below
Immunocytochemistry	5-15 µg/mL	See Below

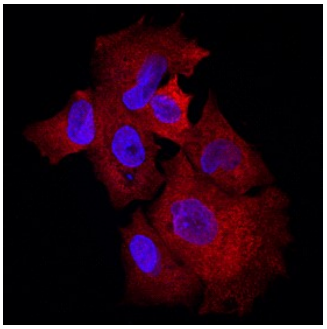
DATA

Western Blot



Detection of Human Klotho β by Western Blot. Western blot shows lysates of Hep3B human hepatocellular carcinoma cell line. PVDF membrane was probed with 0.5 µg/mL of Goat Anti-Human Klotho β Antigen Affinity-purified Polyclonal Antibody (Catalog # AF5889) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for Klotho β at approximately 130 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry



Klotho β in HepG2 Human Cell Line. Klotho β was detected in immersion fixed HepG2 human hepatocellular carcinoma cell line using Goat Anti-Human Klotho β Antigen Affinity-purified Polyclonal Antibody (Catalog # AF5889) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red; Catalog # NL001) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

PREPARATION AND STORAGE

Reconstitution	Sterile PBS to a final concentration of 0.2 mg/mL.
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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Global bio-techne.com info@bio-techne.com techsupport@bio-techne.com TEL +1 612 379 2956
USA TEL 800 343 7475 Canada TEL 855 668 8722 China TEL +86 (21) 52380373
Europe | Middle East | Africa TEL +44 (0)1235 529449

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BACKGROUND

Klotho β, a divergent structural member of the glycosidase I superfamily, is expressed primarily in the liver and pancreas, with lower expression in adipose tissue (1, 2). Like Klotho, Klotho β facilitates binding between FGF19 subfamily members and their receptors via formation of a ternary complex (3). The Klotho β mediated interaction of human FGF19 (mouse FGF15) with FGF Receptor 4 in the liver negatively regulates bile acid synthesis by controlling the secretion of two key bile acid synthase genes, cholesterol 7-α hydroxylase (Cyp7a1) and sterol 12-α hydroxylase (Cyp8b1) (2-5). Klotho β is also a cofactor for the interaction of FGF21 with FGF Receptor 1c in adipocytes, which allows FGF21 to stimulate GLUT1 expression, upregulating adipocyte insulin-dependent glucose uptake (2-4, 6). The 1043 amino acid (aa) type I transmembrane protein is composed of a 51 aa signal sequence, a 943 aa extracellular domain (ECD) containing two glycosidase-like regions, a 21 aa transmembrane domain, and 28 aa intracellular tail. Since Klotho-related proteins lack critical active site Glu residues present in β-glycosidases, it was initially unclear whether they were functional enzymes (1, 7). However, glucuronidase activity has since been demonstrated for Klotho, indicating that physiologically relevant enzymatic activity for Klotho β is also possible (8). The extracellular domain shares 79%, 87%, 87% and 67% identity with mouse, equine, canine and rat Klotho β, respectively. The low identity with rat reflects aa discordance within rodent ECD.

References:

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3. Ito, S. *et al.* (2005) J. Clin. Invest. **115**:2202.
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8. Goetz, R. *et al.* (2007) Mol. Cell. Biol. **27**:3417.

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