

Antigen Affinity-purified Polyclonal Goat IgG Catalog Number: AF1819

DESCRIPTION

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Species Reactivity	Mouse	
Specificity	Detects mouse Klotho in direct ELISAs and Western blots.	
Source	Polyclonal Goat IgG	
Purification	Antigen Affinity-purified	
Immunogen	Mouse myeloma cell line NS0-derived recombinant mouse Klotho Arg31-His550 Accession # BAA25307	
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	0.2 µg/mL	See Below
Immunohistochemistry	2-15 μg/mL	See Below
SA         This antibody functions as an ELISA detection antibody when paired with Rat Anti-Mouse Klotho Monoclor           Antibody (Catalog # MAB1819).		paired with Rat Anti-Mouse Klotho Monoclonal

This product is intended for assay development on various assay platforms requiring antibody pairs.





Detection of Mouse Klotho by Western Blot. Western blot shows lysates of mouse kidney tissue. PVDF membrane was probed with 0.2 µg/mL of Goat Anti-Mouse Klotho Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1819) followed by HRPconjugated Anti-Goat IgG Secondary Antibody (Catalog # Catalog # HAF109). A specific band was detected for Klotho at approximately 145 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

## Immunohistochemistry



Klotho in Mouse Kidney. Klotho was detected in perfusion fixed frozen sections of mouse kidney using 1.7 µg/mL Goat Anti-Mouse Klotho Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1819) overnight at 4 °C. Tissue was stained with the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # Catalog # CTS008) and counterstained with hematoxylin (blue). View our protocol for Chromogenic IHC Staining of Frozen Tissue Sections.



#### Mouse Klotho ELISA

Standard Curve. Recombinant Mouse Klotho protein was serially diluted 2-fold and captured by Rat Anti-Mouse Klotho Monoclonal Antibody (Catalog # Catalog # MAB1819) coated on a Clear Polystyrene Microplate (Catalog # Catalog # DY990). Goat Anti-Mouse Klotho Antigen Affinitypurified Polyclonal Antibody (Catalog # AF1819) was biotinylated and incubated with the protein captured on the plate. Detection of the standard curve was achieved by incubating Streptavidin-HRP (Catalog # Catalog # DY998) followed by Substrate Solution (Catalog # Catalog # DY999) and stopping the enzymatic reaction with Stop Solution (Catalog # Catalog # DY994)

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# **Mouse Klotho Antibody**

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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	<ul> <li>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</li> <li>12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>		

## BACKGROUND

Klotho, also called Klotho- $\alpha$ , is the founding member of the Klotho family within the glycosidase-1 superfamily. Klotho is expressed in areas concerned with calcium regulation, predominantly in the kidney distal convoluted tubules, but also in the brain choroid plexus (which produces cerebrospinal fluid) and the parathyroid. The 1014 amino acid (aa) type I transmembrane protein contains a 34 aa signal sequence, a 948 aa extracellular domain (ECD) containing two extracellular glycosidase-like domains, a 21 aa transmembrane domain and an 11 aa intracellular domain. Within the ECD, mouse Klotho shares 95%, 87%, and 87% aa identity with rat, human, and equine Klotho, respectively. Although a truncated 554 aa isoform predicts a soluble 70 kDa form, the soluble form found in plasma and cerebrospinal fluid is a 130 kDa form produced by proteolytic cleavage of the glycosylated 135 kDa full-length Klotho. A prominent intracellular 120 kDa form of Klotho is localized to endoplasmic reticulum and Golgi membranes. Klotho is named for the Greek goddess who spins the thread of life. The phenotype of Klotho-deficient mice resembles premature aging, including arteriosclerosis, osteoporosis, skin atrophy, infertility, emphysema, and premature death. Conversely, excess Klotho extends lifespan. Klotho acts as a cofactor for interaction of FGF-23 with FGF R1. This interaction negatively regulates 1a-hydroxylase, the rate-limiting enzyme in the synthesis of 1,25(OH)<sub>2</sub>D<sub>3</sub> (vitamin D). Klotho-deficient mice show severe hyperphosphatemia and ectopic calcification of soft tissues due to excess vitamin D. Both Klotho and β-Klotho are co-factors for FGF-19 binding. Klotho also shows glucuronidase activity which activates the renal ion channel TRPV5 to reabsorb urinary calcium. Klotho has been reported to downregulate insulin or IGF-I signaling in adipocytes, to bind and antagonize Wnt molecules, and to facilitate release of parathyroid hormone.

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