Human Axl Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF154

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Axl in direct ELISAs and Western blots. In direct ELISAs and Western blots, approximately 25% cross-reactivity with recombinant mouse Axl is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant human Axl Glu33-Pro440 Accession # AAA61243
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.

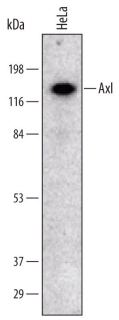
APPLICATIONS

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	1 µg/mL	See Below
Flow Cytometry	2.5 µg/10^6 cells	K562 human chronic myelogenous leukemia cell line
Immunohistochemistry	5-15 µg/mL	See Below
Blockade of Receptor-ligand Interaction	In a functional ELISA, 3-10 µg/mL of this antibody will block 50% of the binding of 20 ng/mL of biotinylated Recombinant Human Gas6, lacking the Gla domain, to immobilized Recombinant Human Axl Fc Chimera (Catalog # 154-AL) coated at 2 µg/mL (100 µL/well). At 100 µg/mL, this antibody will block >90% of the binding.	

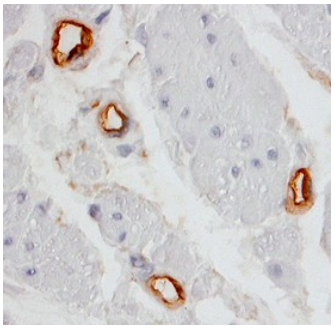
DATA

Western Blot



Detection of Human Axl by Western Blot. Western blot shows lysates of HeLa human cervical epithelial carcinoma cell line. PVDF membrane was probed with 1.0 µg/mL of Human Axl Antigen Affinity-purified Polyclonal Antibody (Catalog # AF154) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for Axl as an approximately 140 kDa glycoprotein (as indicated). This experiment was conducted using Immunoblot Buffer Group 2.

Immunohistochemistry



Axl in Human Stomach. Axl was detected in immersion fixed paraffin-embedded sections of human stomach using 10 µg/mL Human Axl Antigen Affinity-purified Polyclonal Antibody (Catalog # AF154) overnight at 4 °C. Before incubation with the primary antibody tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained with the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS008) and counterstained with hematoxylin (blue). Specific labeling was localized to endothelial cells in the vasculature of the longitudinal muscle layer. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

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.
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 from date of receipt, 2 to 8 °C, reconstituted. ● 6 months from date of receipt, -20 to -70 °C, reconstituted.

BACKGROUND

Axl (Ufo, Ark), Dtk (Sky, Tyro3, Rse, Brt), and Mer (human and mouse homologues of chicken c-Eyk) constitute a subfamily of the receptor tyrosine kinases (1, 2). The extracellular domains of these proteins contain two Ig-like motifs and two fibronectin type III motifs. This characteristic topology is also found in neural cell adhesion molecules and in receptor tyrosine phosphatases. The human Axl cDNA encodes an 887 amino acid (aa) precursor that includes an 18 aa signal sequence, a 426 aa extracellular domain, a 21 aa transmembrane segment, and a 422 aa cytoplasmic domain. The extracellular domains of human and mouse Axl share 81% aa sequence identity. A short alternately spliced form of human Axl is distinguished by a 9 aa deletion in the extracellular juxtamembrane region. These receptors bind the vitamin K-dependent protein growth arrest specific gene 6 (Gas6) which is structurally related to the anticoagulation factor protein S. Binding of Gas6 induces receptor autophosphorylation and downstream signaling pathways that can lead to cell proliferation, migration, or the prevention of apoptosis (3). This family of tyrosine kinase receptors is involved in hematopoiesis, embryonic development, tumorigenesis, and regulation of testicular functions.

References:

1. Yanagita, M. (2004) Curr. Opin. Nephrol. Hypertens. **13**:465.
2. Nagata, K. *et al.* (1996) J. Biol. Chem. **22**:30022.
3. Holland, S. *et al.* (2005) Canc. Res. **65**:9294.