Human Serpin E1/PAI-1 Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF1786

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Serpin E1/PAI-1 in direct ELISAs and Western blots. In direct ELISAs, approximately 20% cross-reactivity with recombinant mouse Serpin E1 is observed and less than 2% cross-reactivity with recombinant human (rh) Serpin A1, rhSerpin A3, rhSerpin A4, rhSerpin A5, rhSerpin C1, rhSerpin F2, and rhSerpin F1/PEDF is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	*S. frugiperda* insect ovarian cell line *Sf* 21-derived recombinant human Serpin E1/PAI-1 Gly21-Pro402 Accession # P05121
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.

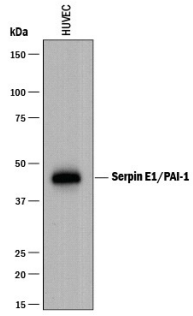
APPLICATIONS

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	1 µg/mL	See Below
Immunohistochemistry	5-15 µg/mL	See Below
Immunoprecipitation	25 µg/mL	Conditioned cell culture medium spiked with Recombinant Human Serpin E1/PAI-1 (Catalog # 1786-PI), see our available Western blot detection antibodies
Simple Western	2.5 µg/mL	See Below
Neutralization	Measured by its ability to neutralize Recombinant Human Serpin E1 (1.35 µg/mL, Catalog # 1786-PI) inhibition of Recombinant Human u-Plasminogen Activator (uPA)/Urokinase (0.1 µg/mL, Catalog # 1310-SE) cleavage of the fluorogenic peptide substrate Z-GGR-AMC (100 µM). The Neutralization Dose (ND_{50}) is typically 10 µg/mL.	

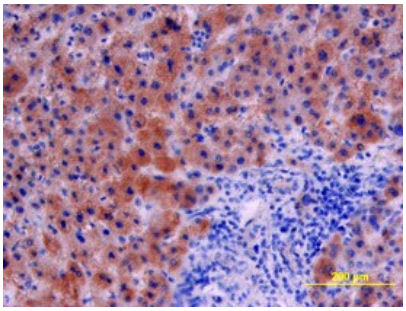
DATA

Western Blot



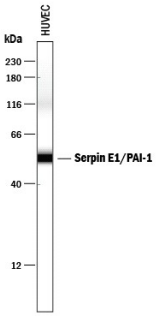
Detection of Human Serpin E1/PAI-1 by Western Blot. Western blot shows lysates of HUVEC human umbilical vein endothelial cells. PVDF membrane was probed with 1 µg/mL of Goat Anti-Human Serpin E1/PAI-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1786) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for Serpin E1/PAI-1 at approximately 45 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunohistochemistry



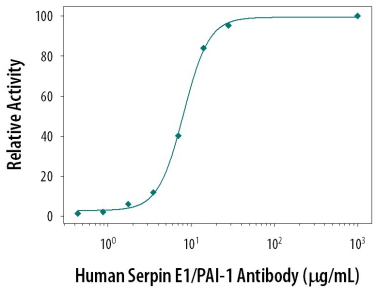
Serpin E1/PAI-1 in Human Liver Cancer Tissue. Serpin E1/PAI-1 was detected in immersion fixed paraffin-embedded sections of human liver cancer tissue using Goat Anti-Human Serpin E1/PAI-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1786) at 15 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS008) and counterstained with hematoxylin (blue). View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Simple Western



Detection of Human Serpin E1/PAI-1 by Simple Western™. Simple Western lane view shows lysates of HUVEC human umbilical vein endothelial cells, loaded at 0.2 mg/mL. A specific band was detected for Serpin E1/PAI-1 at approximately 54 kDa (as indicated) using 2.5 µg/mL of Goat Anti-Human Serpin E1/PAI-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1786) 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



Neutralization of Serpin E1/PAI-1 Activity by Human Serpin E1/PAI-1 Antibody. Recombinant Human u-Plasminogen Activator (uPA)/Urokinase (0.1 µg/mL, Catalog # 1310-SE) activity is measured in the presence of Recombinant Human Serpin E1 (1.35 µg/mL, Catalog # 1786-PI) that has been preincubated with increasing concentrations of Goat Anti-Human Serpin E1/PAI-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1786). The ND_{50} is typically 10 µg/mL.

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Human Serpin E1/PAI-1 Antibody

Antigen Affinity-purified Polyclonal Goat IgG

Catalog Number: AF1786

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

As a member of the Serpin superfamily of serine protease inhibitors, Serpin E1/PAI-1 is the principal inhibitor of urokinase-type plasminogen activator (uPA) and tissue-type PA (1, 2). As important regulators of extracellular matrix remodeling, uPA and PAI-1 play a major role in many processes such as angiogenesis, tumor invasion and obesity (2-4). For example, uPA and PAI-1 are the only tumor prognostic factors validated at the highest level of evidence with regard to their clinical utility in breast cancer (5). The human PAI-1 is initially synthesized as 402 amino acid precursor with a N-terminal signal peptide (6, 7). PAI-1 may exist in one of two possible conformations, designated as active or latent (8). The purified recombinant human (rh) PAI-1 is active against rhuPA. The heterogeneity at the N-terminus of the purified rhPAI-1 has been observed before for both the recombinant and native proteins (9).

References:

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