# **Product Datasheet**

# Serpin E1/PAI-1 Antibody - BSA Free NBP1-19773

Unit Size: 0.1 mg

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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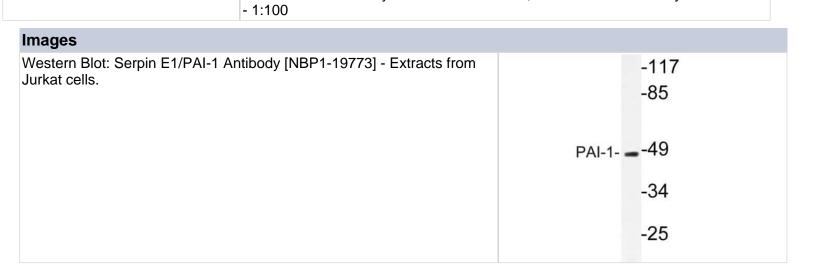
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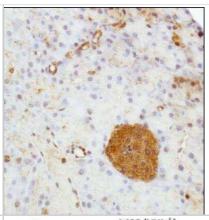
#### NBP1-19773

Serpin E1/PAI-1 Antibody - BSA Free

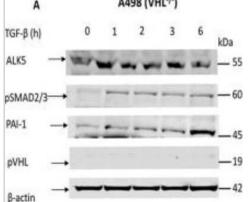
Serpin E1/PAI-1 Antibody - BSA Free	
Product Information	
Unit Size	0.1 mg
Concentration	1.0 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.02% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	PBS
Target Molecular Weight	45 kDa
Product Description	
Host	Rabbit
Gene ID	5054
Gene Symbol	SERPINE1
Species	Human, Mouse, Rat
Reactivity Notes	Use in Rat reported in scientific literature (PMID:35386330).
Specificity/Sensitivity	PAI-1 (N315) pAb detects endogenous levels of PAI-1 protein.
Immunogen	A synthetic peptide made to an internal portion of the human PAI1/Serpine 1 protein (between residues 300-400) [UniProt P05121]
Product Application Details	
Applications	Western Blot, ELISA, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin
Recommended Dilutions	Western Blot 1.0 ug/ml, ELISA, Immunohistochemistry 1:50 - 1:100, Immunocytochemistry/ Immunofluorescence 5 - 10 ug/ml, Immunohistochemistry-Paraffin 1:50 - 1:100, Immunohistochemistry-Frozen 1:50



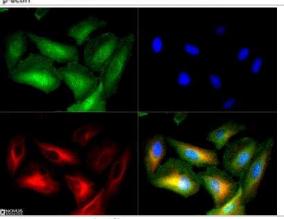
Immunohistochemistry-Paraffin: Serpin E1/PAI-1 Antibody [NBP1-19773] - Staining of PAI1/Serpine 1 in mouse pancreas.



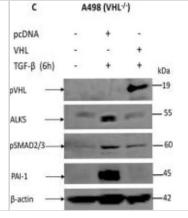
Western Blot: Serpin E1/PAI-1 Antibody - BSA Free [NBP1-19773] - Serpin E1/PAI-1 Antibody [NBP1-19773] - Western Blot: Serpin E1/PAI-1 Antibody [NBP1-19773] - Immunoblots showing protein expression of ALK5, pSMAD2/3, PAI-1, pVHL, andI2-actin in A498 cells after treatment with TGF-i2 at given point of time. Image collected and cropped by Citeab from the following publication (VHL status regulates transforming growth factor B signaling pathways in renal cell carcinoma. Oncotarget (2018)) licensed under a CC-BY license.



Immunocytochemistry/Immunofluorescence: Serpin E1/PAI-1 Antibody [NBP1-19773] - PAI1/Serpine1 antibody was tested in Hela cells with DyLight 488 (green). Nuclei and alpha-tubulin were counterstained with DAPI (blue) and Dylight 550 (red).

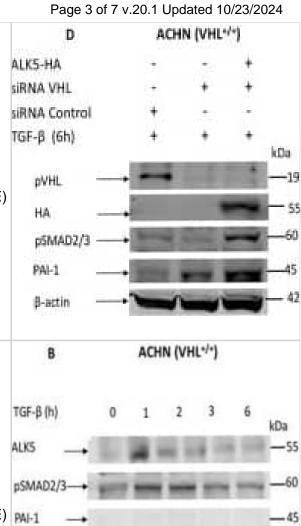


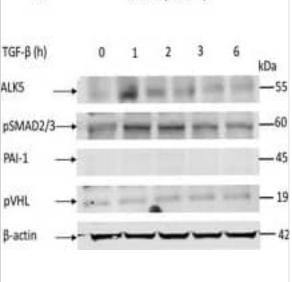
Western Blot: Serpin E1/PAI-1 Antibody - BSA Free [NBP1-19773] - Serpin E1/PAI-1 Antibody [NBP1-19773] - Immunoblots showing protein expression of pVHL, ALK5, pSMAD2/3, PAI-1, andI2-actin after transfection of indicated vectors in A498 cells followed by TGF-i2 treatment for 6 hours. Image collected and cropped by Citeab from the following publication (VHL status regulates transforming growth factor-B signaling pathways in renal cell carcinoma. Oncotarget (2018)) licensed under a CC-BY license.



Western Blot: Serpin E1/PAI-1 Antibody - BSA Free [NBP1-19773] - (A) Immunoblots showing protein expression of ALK5, pSMAD2/3, PAI-1, pVHL, & β-actin in A498 cells after treatment with TGF-β at given point of time; (B) Immunoblots showing protein expression of ALK5. pSMAD2/3, PAI-1, pVHL, & β-actin in ACHN cells after treatment with TGF-β at given point of time; (C) Immunoblots showing protein expression of pVHL, ALK5, pSMAD2/3, PAI-1, & β-actin after transfection of indicated vectors in A498 cells followed by TGF-B treatment for 6 hours; (D) Immunoblots showing protein expression of pVHL, HA, pSMAD2/3, PAI-1, & β-actin after transfection of indicated vectors in ACHN cells (48h) followed by TGF-β treatment for 6 hours; (E) Invasion assay showing the invasiveness induced by TGF-β in A498 cells after re-introduction of VHL (n=3 independent experiments \*P< 0. 05); (F) Invasion assay showing invasiveness by TGF-β in ACHN cells after siRNA VHL knockdown (n=3 independent experiments \*P< 0.05). Image collected & cropped by CiteAb from the following publication (https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.24631). licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Western Blot: Serpin E1/PAI-1 Antibody - BSA Free [NBP1-19773] - (A) Immunoblots showing protein expression of ALK5, pSMAD2/3, PAI-1, pVHL, & β-actin in A498 cells after treatment with TGF-β at given point of time; (B) Immunoblots showing protein expression of ALK5. pSMAD2/3, PAI-1, pVHL, & β-actin in ACHN cells after treatment with TGF-β at given point of time; (C) Immunoblots showing protein expression of pVHL, ALK5, pSMAD2/3, PAI-1, & β-actin after transfection of indicated vectors in A498 cells followed by TGF-β treatment for 6 hours; (D) Immunoblots showing protein expression of pVHL, HA, pSMAD2/3, PAI-1, & β-actin after transfection of indicated vectors in ACHN cells (48h) followed by TGF-β treatment for 6 hours; (E) Invasion assay showing the invasiveness induced by TGF-β in A498 cells after re-introduction of VHL (n=3 independent experiments \*P< 0. 05); (F) Invasion assay showing invasiveness by TGF-β in ACHN cells after siRNA VHL knockdown (n=3 independent experiments \*P< 0.05). Image collected & cropped by CiteAb from the following publication (https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.24631). licensed under a CC-BY license. Not internally tested by Novus Biologicals.





#### **Publications**

Liu B, Kong Y, Alimi OA et al. Multifunctional Microgel-Based Cream Hydrogels for Postoperative Abdominal Adhesion Prevention ACS nano 2023-02-28 [PMID: 36779870]

Mallikarjuna P, Raviprakash TS, Aripaka K et al. Interactions between TGF-beta type I receptor and hypoxia-inducible factor-α mediates a synergistic crosstalk leading to poor prognosis for patients with clear cell renal cell carcinoma Cell Cycle 2019-09-01 [PMID: 31339433]

Wang X, Guo L, Huang J et al. Plasminogen Activator Inhibitor-1 Potentiates Neutrophil Infiltration and Tissue Injury in Colitis International Journal of Biological Sciences 2023-04-18 [PMID: 37151884]

Qu F, Brough SC, Michno W et al. Crosstalk between small-cell lung cancer cells and astrocytes mimics brain development to promote brain metastasis Nature cell biology 2023-10-01 [PMID: 37783795]

Samarkina A, Youssef MK, Ostano P et al. Androgen receptor is a determinant of melanoma targeted drug resistance Nature communications 2023-10-14 [PMID: 37838724] (Western Blot, Immunocytochemistry/ Immunofluorescence, Human)

Bertelli PM, Pedrini E, Hughes D et al. Long term high glucose exposure induces premature senescence in retinal endothelial cells Frontiers in Physiology 2022-08-26 [PMID: 36091370]

Hasuike Y, Mochizuki H, Nakamori M. Expanded CUG Repeat RNA Induces Premature Senescence in Myotonic Dystrophy Model Cells Frontiers in Genetics 2022-03-25 [PMID: 35401669] (Western Blot)

Ghosh AK, Kalousdian AA, Shang M et al. Cardiomyocyte PAI-1 influences the cardiac transcriptome and limits the extent of cardiac fibrosis in response to left ventricular pressure overload Cellular signalling 2022-12-27 [PMID: 36584735] (IHC, Mouse)

Zhu X, Schrader JM, Irizarry BA et al. Impact of Aβ40 and Aβ42 Fibrils on the Transcriptome of Primary Astrocytes and Microglia Biomedicines 2022-11-19 [PMID: 36428550] (IHC-Fr, Rat)

#### Details:

Fifteen-month-old rTgF344-AD rat brain sections, dilution used 1:200

Gao J, Wen J, Hu D et al. Bottlebrush inspired injectable hydrogel for rapid prevention of postoperative and recurrent adhesion Bioactive Materials 2022-02-01 [PMID: 35386330] (IF/IHC, Rat)

Hu BA, Sai WW, Yuan J et al. PGF2 alpha-FP Receptor Ameliorates Senescence of VSMCs in Vascular Remodeling by Src/PAI-1 Signal Pathway Oxidative medicine and cellular longevity 2022-01-27 [PMID: 35126810] (WB, Rat, Mouse)

Zhang S, Cao Y, Du J Et al. Neutrophil extracellular traps contribute to tissue plasminogen activator resistance in acute ischemic stroke FASEB journal: official publication of the Federation of American Societies for Experimental Biology 2021-09-01 [PMID: 34449927] (ICC/IF, IF/IHC, Human)

More publications at <a href="http://www.novusbio.com/NBP1-19773">http://www.novusbio.com/NBP1-19773</a>



#### **Procedures**

## Western blot Protocol specific for PAI1/Serpine1 antibody (NBP1-19773)

#### Western Blot Protocol

- 1. Perform SDS-PAGE on samples to be analyzed, loading 40 ug of total protein per lane.
- 2. Transfer proteins to membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.
- 3. Stain according to standard Ponceau S procedure (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.
- 4. Rinse the blot.
- 5. Block the membrane using standard blocking buffer for at least 1 hour.
- 6. Wash the membrane in wash buffer three times for 10 minutes each.
- 7. Dilute primary antibody in blocking buffer and incubate 1 hour at room temperature.
- 8. Wash the membrane in wash buffer three times for 10 minutes each.
- 9. Apply the diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturers instructions) and incubate 1 hour at room temperature.
- 10. Wash the blot in wash buffer three times for 10 minutes each (this step can be repeated as required to reduce background).
- 11. Apply the detection reagent of choice in accordance with the manufacturers instructions.

Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%.

#### Immunohistochemistry Protocol specific for PAI1/Serpine1 antibody (NBP1-19773)

Immunohistochemistry-Paraffin Embedded Sections

#### Antigen Unmasking:

Bring slides to a boil in 10 mM sodium citrate buffer (pH 6.0) then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench-top for 30 minutes.

#### Staining:

- 1. Wash sections in deionized water three times for 5 minutes each.
- 2. Wash sections in wash buffer for 5 minutes.
- 3. Block each section with 100-400 ul blocking solution for 1 hour at room temperature.
- 4. Remove blocking solution and add 100-400 ul diluted primary antibody. Incubate overnight at 4 C.
- 5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
- 6. Add 100-400 ul biotinylated diluted secondary antibody. Incubate 30 minutes at room temperature.
- 7. Remove secondary antibody solution and wash sections three times with wash buffer for 5 minutes each.
- 8. Add 100-400 ul Streptavidin-HRP reagent to each section and incubate for 30 minutes at room temperature.
- 9. Wash sections three times in wash buffer for 5 minutes each.
- 10. Add 100-400 ul DAB substrate to each section and monitor staining closely.
- 11. As soon as the sections develop, immerse slides in deionized water.
- 12. Counterstain sections in hematoxylin.
- 13. Wash sections in deionized water two times for 5 minutes each.
- 14. Dehydrate sections.
- 15. Mount coverslips.



#### Immunocytochemistry/Immunofluorescence Protocol for PAI1/Serpine1 Antibody (NBP1-19773)

Immunocytochemistry Protocol

Culture cells to appropriate density in 35 mm culture dishes or 6-well plates.

- 1. Remove culture medium and add 10% formalin to the dish. Fix at room temperature for 30 minutes.
- 2. Remove the formalin and add ice cold methanol. Incubate for 5-10 minutes.
- 3. Remove methanol and add washing solution (i.e. PBS). Be sure to not let the specimen dry out. Wash three times for 10 minutes.
- 4. To block nonspecific antibody binding incubate in 10% normal goat serum from 1 hour to overnight at room temperature.
- 5. Add primary antibody at appropriate dilution and incubate at room temperature from 2 hours to overnight at room temperature.
- 6. Remove primary antibody and replace with washing solution. Wash three times for 10 minutes.
- 7. Add secondary antibody at appropriate dilution. Incubate for 1 hour at room temperature.
- 8. Remove antibody and replace with wash solution, then wash for 10 minutes. Add Hoechst 33258 to wash solution at 1:25,0000 and incubate for 10 minutes. Wash a third time for 10 minutes.
- 9. Cells can be viewed directly after washing. The plates can also be stored in PBS containing Azide covered in Parafilm (TM). Cells can also be cover-slipped using Fluoromount, with appropriate sealing.

\*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow safe laboratory procedures.





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# **Products Related to NBP1-19773**

HAF008 Goat anti-Rabbit IgG Secondary Antibody [HRP]

NB7160 Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]

NBP2-24891 Rabbit IgG Isotype Control

NBP1-19773B Serpin E1/PAI-1 Antibody [Biotin]

#### Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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