# **Product Datasheet**

# S5a/Angiocidin Antibody NBP2-19952

Unit Size: 0.1 ml

Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.

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#### NBP2-19952

S5a/Angiocidin Antibody

Product Information	
Unit Size	0.1 ml
Concentration	Concentrations vary lot to lot. See vial label for concentration. If unlisted please contact technical services.
Storage	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.01% Thimerosal
Isotype	IgG
Purity	Antigen Affinity-purified
Buffer	0.1M Tris (pH 7), 0.1M Glycine, 20% Glycerol
Target Molecular Weight	41 kDa
Product Description	
Host	Rabbit
Gene ID	5710

	human S5a/Angiocidin. The exact sequence is proprietary.
<b>Product Application Details</b>	
Applications	Western Blot, Immunocytochemistry/ Immunofluorescence, Knockdown Validated
Recommended Dilutions	Western Blot 1:500-1:3000, Immunocytochemistry/ Immunofluorescence 1:100-1:1000, Knockdown Validated

Recombinant protein encompassing a sequence within the center region of

## **Images**

**Gene Symbol** 

**Immunogen** 

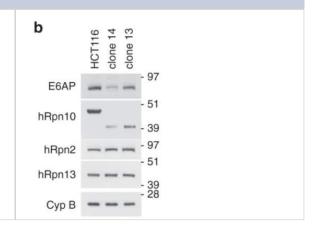
**Reactivity Notes** 

**Species** 

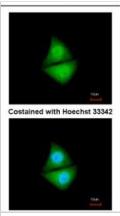
Western Blot: S5a/Angiocidin Antibody [NBP2-19952] - Immunoblots of parental HCT116 cells and two clonal cell lines (clone 14 and clone 13) generated by CRISPR-mediated truncation of hRpn10. Image collected and cropped by CiteAb from the following publication (nature.com/articles/s41467-020-15073-7), licensed under a CC-BY license.

PSMD4 Human

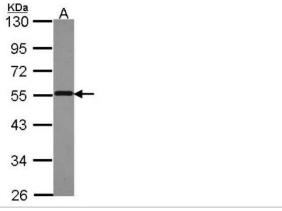
Xenopus laevis (90%).



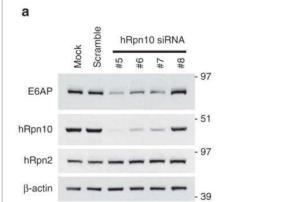
Immunocytochemistry/Immunofluorescence: Proteasome 19S S5A Antibody [NBP2-19952] - Immunofluorescence analysis of paraformaldehyde-fixed A549, using antibody at 1:200 dilution.



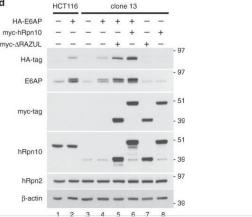
Western Blot: Proteasome 19S S5A Antibody [NBP2-19952] - Sample (30 ug of whole cell lysate) A: Hela 10% SDS PAGE gel, diluted at 1:1000.



Knockdown Validated: S5a/Angiocidin Antibody [NBP2-19952] - hRpn10 was knocked down in HCT116 cells by four different siRNAs and the cell lysates immunoprobed as indicated. Mock and scrambled control samples are included. b-actin is used as a loading control. Image collected and cropped by CiteAb from the following publication (nature.com/articles/s41467-020-15073-7), licensed under a CC-BY license.



Western Blot: S5a/Angiocidin Antibody [NBP2-19952] - E6AP levels depend on hRpn10.hRpn10 (a) or E6AP (b) was knocked down in HCT116 cells by four different siRNAs & the cell lysates immunoprobed as indicated. Mock & scrambled control samples are included. β-actin is used as a loading control in a & d. c Lysates from HCT116 or clone 14 cells expressing myc-hRpn10 constructs were immunoprobed as indicated. d Lysates from HCT116 or clone 13 cells expressing HA-E6AP and/or myc-hRpn10 constructs were immunoprobed as indicated. a–d Antibodies used for immunoprobing are indicated to the left of each panel. Source data are provided as a Source Data file. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32157086), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Western Blot: S5a/Angiocidin Antibody [NBP2-19952] - E6AP levels depend on hRpn10.hRpn10 (a) or E6AP (b) was knocked down in HCT116 cells by four different siRNAs & the cell lysates immunoprobed as indicated. Mock & scrambled control samples are included. β-actin is used as a loading control in a & d. c Lysates from HCT116 or clone 14 cells expressing myc-hRpn10 constructs were immunoprobed as indicated. d Lysates from HCT116 or clone 13 cells expressing HA-E6AP and/or myc-hRpn10 constructs were immunoprobed as indicated. a–d Antibodies used for immunoprobing are indicated to the left of each panel. Source data are provided as a Source Data file. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32157086), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

C HCT116 clone 14

myc-hRpn10 - - - + myc-ΔRAZUL - + - - +

E6AP - - 51

myc-tag - 39

hRpn10 - 39

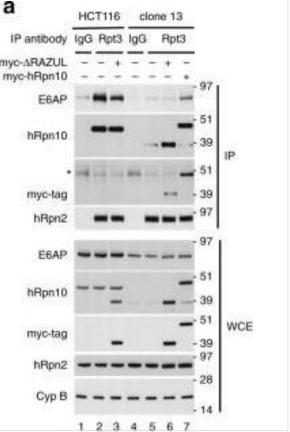
hRpn2 - 97

Cyp B - - 28

Western Blot: S5a/Angiocidin Antibody [NBP2-19952] - E6AP levels depend on hRpn10.hRpn10 (a) or E6AP (b) was knocked down in HCT116 cells by four different siRNAs & the cell lysates immunoprobed as indicated. Mock & scrambled control samples are included. β-actin is used as a loading control in a & d. c Lysates from HCT116 or clone 14 cells expressing myc-hRpn10 constructs were immunoprobed as indicated. d Lysates from HCT116 or clone 13 cells expressing HA-E6AP and/or myc-hRpn10 constructs were immunoprobed as indicated. a–d Antibodies used for immunoprobing are indicated to the left of each panel. Source data are provided as a Source Data file. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32157086), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

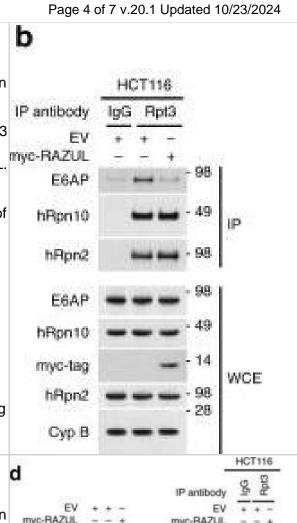


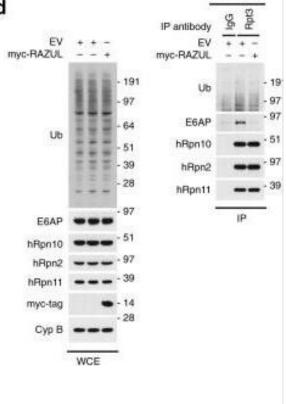
Western Blot: S5a/Angiocidin Antibody [NBP2-19952] - hRpn10 RAZUL contributes E6AP to the proteasome.a Immunoblots of Rpt3 immunoprecipitates or WCE from HCT116 or clone 13 lysates expressing myc-hRpn10 constructs. An asterisk "\*" indicates heavy chain myc-ARAZUL antibody. Cyclophilin B (Cyp B) is used as a loading control for WCE samples in a-c & hRpn2 as a positive control for the immunoprecipitation. IgG controls are included. b, d Immunoblots of Rpt3 or IgG (control) immunoprecipitates or WCE of lysates from HCT116 cells transfected with empty vector (as a control) or myc-hRpn10 RAZUL. c Immunoblots of Rpt3 immunoprecipitates or WCE from lysates of HCT116 cells transfected with a scrambled control or siRNA against E6AP. a-d Antibodies used for immunoprobing are indicated to the left of each panel, e Pull-down assay for a commercially available mixture of His6-tagged, non-cleavable K48-linked Ub2/Ub4 with incubation of human 26S proteasome (lane 6), 26S proteasome with equimolar E6AP (lane 7), or just E6AP (lane 8). E6AP or 26S proteasome was added to Ni-NTA agarose resin as negative controls (lanes 4 & 5). K48-linked Ub2/Ub4, E6AP, & 26S proteasome were loaded directly in lanes 1–3, as indicated, f Selected regions from 1D 13C-edited, 1H NMR experiments acquired at 850 MHz & 25 °C for free 13C-AZUL (black) or mixtures with equimolar unlabeled RAZUL (blue) or 26S proteasome (red). The concentration of each sample was 0.3 µM & 200,000 scans were recorded for each experiment. Source data are provided as a Source Data file. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32157086), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



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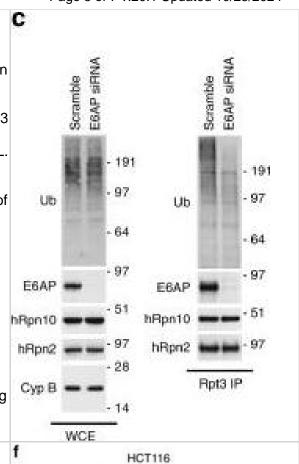
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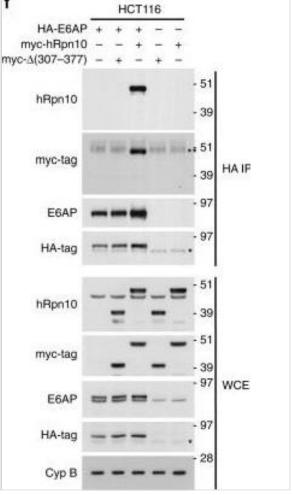




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Western Blot: S5a/Angiocidin Antibody [NBP2-19952] - A C-terminal domain in hRpn10 binds E6AP AZUL.a Positions of known functional domains within hRpn10 (top) & E6AP isoform II (bottom). Question mark "?" indicates hRpn10 uncharacterized region & the E6AP catalytic cysteine C843 is indicated, b Pull-down assay of His-tagged hRpn10fulllength (full), hRpn10196–377 or hRpn10196–306 without (-) or with (+)incubation of E6AP. c1H, 15N HSQC spectra of 0.2 mM 15NhRpn10305-377 (black) & with twofold molar excess unlabeled AZUL (green). Shifted signals are labeled. d Table summarizing Kd, kon, & koff average values with standard deviations for the hRpn10305-377: AZUL interaction measured by ITC and/or SPR. N/A, not applicable. e HCT116 lysates expressing empty vector, myc-hRpn10 full length, or myc-Rpn10 with RAZUL deleted (Δ307–377) were subjected to mycimmunoprecipitation with anti-myc-tag nanobody-coupled agarose. Whole cell extracts (WCE) & myc-immunoprecipitates were immunoprobed with the indicated antibodies. Cyclophilin B (Cyp B) is used as a loading control in e & f. f Lysates from HCT116 cells expressing HA-E6AP & the myc-hRpn10 constructs of e were subjected to HA IP followed by immunoblotting with the indicated antibodies. An asterisk "\*" indicates non-specific interaction; double asterisk "\*\*" indicates heavy chain antibody, e-f All antibodies used for immunoprobing are indicated to the left of the images. Note that the hRpn10 & E6AP antibodies recognize both endogenous & exogenously expressed protein, causing these panels to show both tagged & endogenous protein. g Schematic representation highlighting interaction domains of hRpn10 including newly identified RAZUL. Source data are provided as a Source Data file. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32157086), licensed under a CC-BY







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### **Publications**

Osei-Amponsa V, Sridharan V, Tandon M et al. Impact of losing hRpn13 Pru or UCHL5 on proteasome clearance of ubiquitinated proteins and RA190 cytotoxicity Mol. Cell. Biol. 2020-07-06 [PMID: 32631902]

Structure of E3 ligase E6AP with a proteasome-binding site provided by substrate receptor hRpn10 Buel GR, Chen X, Chari R et al. Nat Commun [PMID: 32157086] (KD, WB, Human)





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## **Products Related to NBP2-19952**

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

NBP2-24891 Rabbit IgG Isotype Control

NBP2-37888PEP S5a/Angiocidin Recombinant Protein Antigen

#### 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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