

Product Datasheet

Survivin Antibody (60.11) - BSA Free NB500-238

Unit Size: 0.1 ml

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

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NB500-238

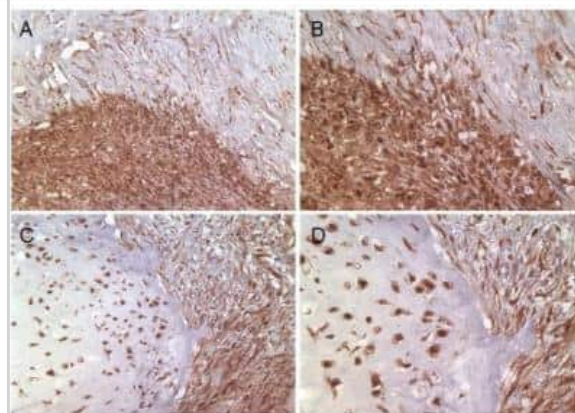
Survivin Antibody (60.11) - BSA Free

Product Information	
Unit Size	0.1 ml
Concentration	1.0 mg/ml
Storage	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	60.11
Preservative	0.1% Sodium Azide
Isotype	IgG2a Kappa
Purity	Protein G purified
Buffer	PBS
Target Molecular Weight	16 kDa
Product Description	
Host	Mouse
Gene ID	332
Gene Symbol	BIRC5
Species	Human, Mouse, Rat
Specificity/Sensitivity	Survivin Antibody (60.11) [NB500-238] is specific for the cytoplasmic form of Survivin.
Immunogen	This Survivin Antibody (60.11) [Alexa Fluor 350] was developed against full length recombinant human Survivin [UniProt# O15392]
Product Application Details	
Applications	Western Blot, ELISA, Flow Cytometry, Flow (Intracellular), Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Proximity Ligation Assay, Knockdown Validated
Recommended Dilutions	Western Blot 1:1000, Flow Cytometry 1 ug/mL. Use reported in scientific literature (PMID 25050620), ELISA reported in scientific literature (PMID 11861764), Immunohistochemistry 1:50-1:200, Immunocytochemistry/ Immunofluorescence 1:50-1:200, Immunohistochemistry-Paraffin 1:50-1:200, Immunohistochemistry-Frozen, Proximity Ligation Assay reported in scientific literature (PMID 28077791), Flow (Intracellular), Knockdown Validated
Application Notes	By WB, this antibody recognizes a band at ~16.5 kDa representing Survivin.

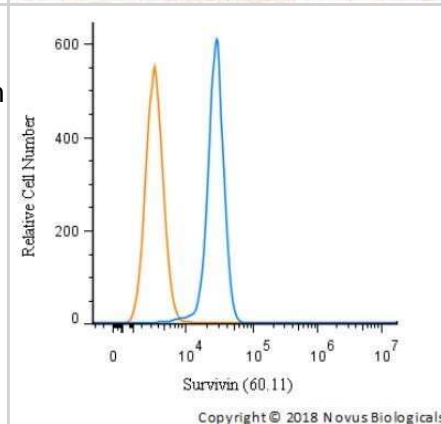


Images

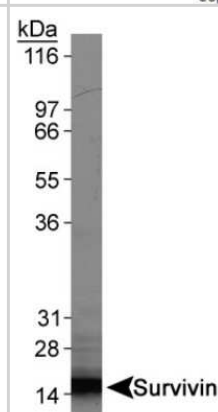
Immunohistochemistry: Survivin Antibody (60.11) [NB500-238] - Low-power image of human high-grade chondrosarcoma displays strong cellular expression of survivin protein (A). High-power magnification reveals the predominantly cytoplasmic staining, although strong nuclear signals are detectable (B). Other specimen of a grade III chondrosarcoma stained with monoclonal antibody, shows a similar pattern of staining (C and D). For A and B the polyclonal rabbit anti-survivin antibody [AF886] was used. For C and D the monoclonal mouse Survivin Antibody (60.11) [NB500-238] was used. Image collected and cropped by CiteAb from the following publication (<https://bmccancer.biomedcentral.com/articles/10.1186/1471-2407-11-120>), licensed under a CC-BY license.



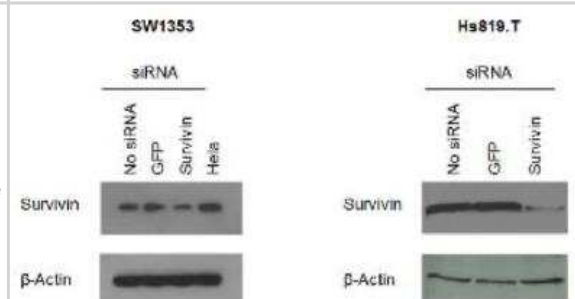
Flow (Intracellular): Survivin Antibody (60.11) [NB500-238] - An intracellular stain was performed on A549 cells with Survivin Antibody (60.11) [NB500-238] and a matched isotype control. Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 1 ug/mL for 30 minutes at room temperature, followed by mouse F(ab)₂ IgG (H+L) APC-conjugated secondary antibody (F0101B, R&D Systems). Image using the Unpurified form of this antibody.



Western Blot: Survivin Antibody (60.11) [NB500-238] - Survivin detection in 30ug of HeLa whole cell extract using [NB500-238] at 1ug/ml. Detected at predicted molecular weight of 16 kDa.



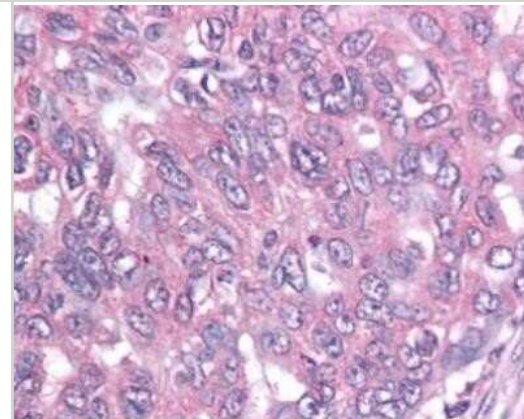
Knockdown Validated: Survivin Antibody (60.11) [NB500-238] - Knockdown Validation of Survivin Antibody (60.11) [NB500-238]. Suppression of survivin expression by transfection of siRNA. RNA interference was performed in SW1353 and Hs 819.T, either for GFP as control or for survivin. A: A pronounced decrease of survivin protein levels was measured by immunoblotting in SW1353 and Hs819.T. Image collected and cropped by CiteAb from the following publication (<https://bmccancer.biomedcentral.com/articles/10.1186/1471-2407-11-120>), licensed under a CC-BY license.



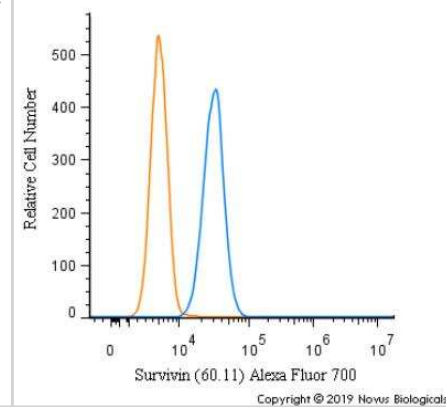
Immunocytochemistry/Immunofluorescence: Survivin Antibody (60.11) [NB500-238] - HeLa cells stained NB500-205 (Green) detected with DyLight Fluor 488 conjugated anti-mouse IgG secondary antibody. Nuclei are counterstained with Hoechst 33258 (Blue). Image using the Unpurified format of this antibody.



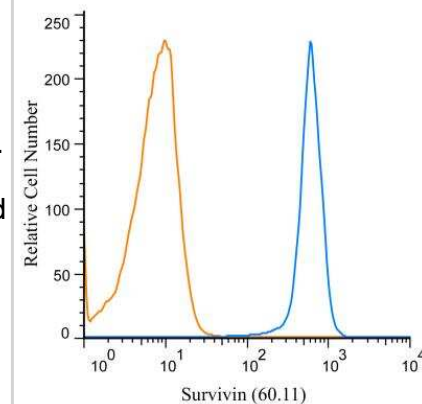
Immunohistochemistry-Paraffin: Survivin Antibody (60.11) [NB500-238] - Staining of ovary cancer. Image using the Unpurified format of this antibody.



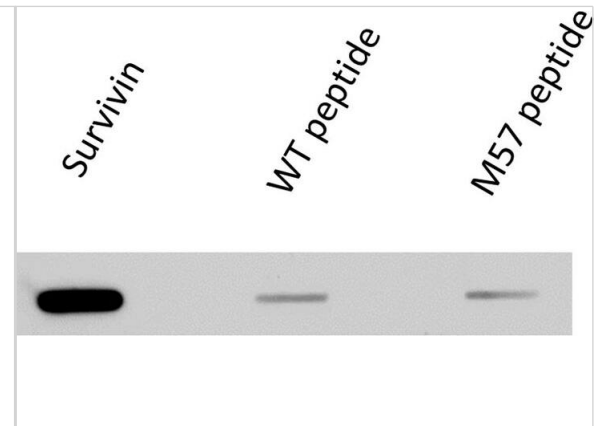
Flow Cytometry: Survivin Antibody (60.11) [NB500-238] - An intracellular stain was performed on U2OS cells with Survivin Antibody (60.11) [NB500-238AF700] (blue) and a matched isotype control (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 5 ug/mL for 30 minutes at room temperature. Both antibodies were conjugated to Alexa Fluor 700.



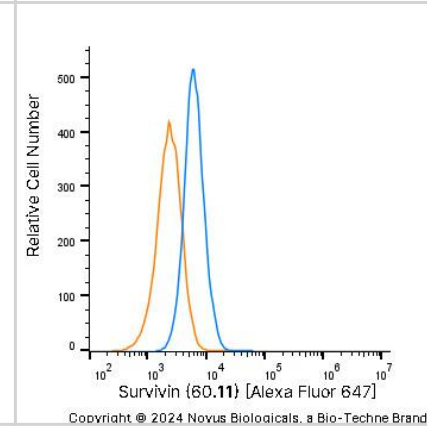
Flow (Intracellular): Survivin Antibody (60.11) [NB500-238] - An intracellular stain was performed on Daudi cells with Survivin Antibody (60.11)[NB500-238] (blue) and a matched isotype control, Mouse IgG2a Kappa Light Chain Isotype Control (MG2a-53) [NB600-986] (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 1 ug/mL for 30 minutes at room temperature, followed by mouse F(ab)2 IgG (H+L) APC-conjugated secondary antibody (F0101B, R&D Systems).



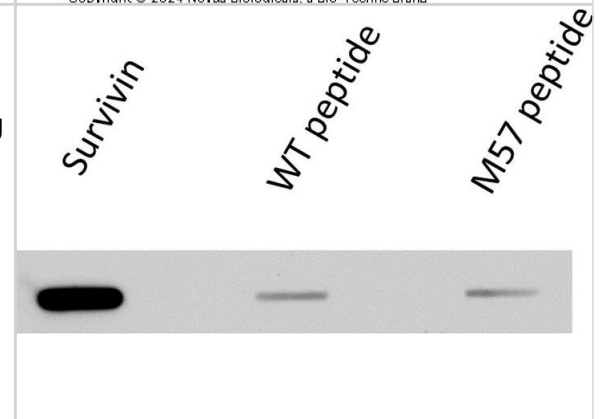
Binding of antibody (60.11) used in imaging flow cytometry to full-length survivin protein and to survivin vaccine peptide aa53-67/M57 and wild type peptide aa53-67



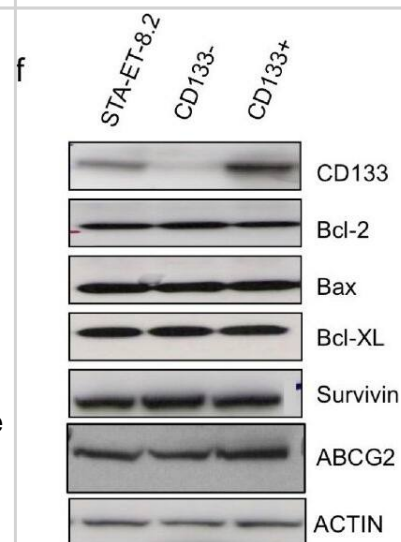
A431 human skin carcinoma cell line was stained with Mouse anti-Survivin (60.11) Protein-G purified Monoclonal Antibody conjugated to Alexa Fluor® 647 (Catalog # NB500-238AF647, blue histogram) or matched control antibody (orange histogram).



Western Blot: Survivin Antibody (60.11) - BSA Free [NB500-238] - Binding of antibody (60.11) used in imaging flow cytometry to full-length survivin protein & to survivin vaccine peptide aa53-67/M57 & wild type peptide aa53-67 Image collected & cropped by CiteAb from the following publication (<https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.21773>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Western Blot: Survivin Antibody (60.11) - BSA Free [NB500-238] - CD133+ cells are more resistant to chemotherapeutic agents. STA-ET-8.2 cells were FACS-sorted & CD133+ & CD133- fractions treated with increasing concentrations of (A) Doxorubicin (Doxo), (B) Etoposide (Etop), (C) Vincristine (Vinc) or (D) a combination of all three drugs at high (Doxo 10 μ M; Etop 10 μ g/ml; Vinc 100 ng/ml), medium (Doxo 2.5 μ M; 20 Etop 2.5 μ g/ml; Vinc 25 ng/ml) & low (Doxo 0.5 μ M; Etop 0.5 μ g/ml; Vinc 5 ng/ml) concentrations. Viability was assessed by MTS assay after 96 hrs (*p < 0.05) & CD133+ cells displayed increased drug resistance. (E): CD133+ & CD133- TC-71 cells are equally sensitive to Doxo, Etop, & Vinc. (F): Western blot reveals no difference in apoptosis & drug resistance protein expression between unsorted & CD133-sorted STA-ET-8.2 cell fractions. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/20346143>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Zhang X, Ciesielski M, Fenstermaker RA et al. The Presence of Survivin on B Cells from Myasthenia Gravis Patients and the Potential of an Antibody to a Modified Survivin Peptide to Alleviate Weakness in an Animal Model The Journal of Immunology 2020-10-01 [PMID: 32839239] (Flow Cytometry)

Jiang X, Gwee Y et al. CD133 expression in chemo-resistant Ewing sarcoma cells. BMC Cancer 2010-03-26 [PMID: 20346143] (WB, Human)

The integration of a Stat3 specific peptide aptamer into the thioredoxin scaffold protein strongly enhances its inhibitory potency. Schoneberger H, Weiss A, Brill B et al. Horm Mol Biol Clin Investig [PMID: 25961238]

Kim S, Shin M, Lee A et al. Improvement of Inflammation through Antioxidant Pathway of Gardeniae Fructus 50% EtOH Extract (GE) from Acute Reflux Esophagitis Rats Biomed Res Int. [PMID: 32185206] (WB, Rat)

Cho M, Lee OH, Chang EM et al. BIRC5 Expression is Regulated in Uterine Epithelium During the Estrous Cycle Genes (Basel) 2020-03-06 [PMID: 32155884] (IHC-P, Mouse)

Ring A, Nguyen C, Smbatyan G et al. CBP/beta-Catenin/FOXO1 Is a Novel Therapeutic Target in Triple Negative Breast Cancer Cancers (Basel). 2018-12-19 [PMID: 30572639] (WB, Human)

Details:

Citation used the Unpurified format of this antibody.

Fenstermaker RA, Figel SA, Qiu JX et al. Survivin Monoclonal Antibodies Detect Survivin Cell Surface Expression and Inhibit Tumor Growth in vivo Clin. Cancer Res. 2018-03-14 [PMID: 29540489] (WB, Mouse)

Gallagher SJ, Gunatilake D, Beaumont KA et al. HDAC inhibitors restore BRAF-inhibitor sensitivity by altering PI3K and survival signalling in a subset of melanoma Int J Cancer 2017-12-07 [PMID: 29210065] (WB, Human)

Details:

Citation using the Non-Recombinant Monoclonal format of this antibody.

Galbo P, Ciesielski MJ, Figel S et al. Circulating CD9+/GFAP+/survivin+ exosomes in malignant glioma patients following survivin vaccination. Oncotarget. [PMID: 29383115] (WB, Human)

Tsang TJ, Hsueh YC, Wei EI et al. Subcellular Localization of Survivin Determines Its Function in Cardiomyocytes Theranostics. 2017-10-13 [PMID: 29158846] (WB, Mouse)

Dheekollu J, Malecka K, Wiedmer A et al. Carcinoma-risk variant of EBNA1 deregulates Epstein-Barr Virus episomal latency Oncotarget 2017-01-31 [PMID: 28077791] (PLA)

Kapinas Kristina, Kim Heesun, Mandeville Matthew et al. microRNA-mediated survivin control of pluripotency. J Cell Physiol. [PMID: 24891298] (Human)

Details:

Citation using the Unpurified form of this antibody.

More publications at <http://www.novusbio.com/NB500-238>

Procedures

Serum protocol for Survivin Antibody (NB500-238)

Immunohistochemistry - FFPE sections

I. Deparaffinization:

- A. Treat slides with Xylene: 3 changes for 5 minutes each. Drain slides for 10 seconds between changes.
- B. Treat slides with 100% Reagent Alcohol: 3 changes for 5 minutes each. Drain slides for 10 seconds between changes.

II. Quench Endogenous Peroxidase:

- A. Place slides in peroxidase quenching solution: 15-30 minutes.

To Prepare 200 ml of Quenching Solution:

Add 3 ml of 30% Hydrogen Peroxide to 200 ml of Methanol.

Use within 4 hours of preparation

- B. Place slides in distilled water: 2 changes for 2 minutes each.

III. Retrieve Epitopes:

- A. Preheat Citrate Buffer. Place 200 ml of Citrate Buffer Working Solution into container, cover and place into steamer. Heat to 90-96 degrees Celcius.
- B. Place rack of slides into hot Citrate Buffer for 20 minutes. Cover.
- C. Carefully remove container with slides from steamer and cool on bench, uncovered, for 20 minutes.
- D. Slowly add distilled water to further cool for 5 minutes. E. Rinse slides with distilled water. 2 changes for 2 minutes each.

IV. Immunostaining Procedure:

- A. Remove each slide from rack and circle tissue section with a hydrophobic barrier pen (e.g. Liquid Blocker-Super Pap Pen).
- B. Flood slide with Wash Solution. Do not allow tissue sections to dry for the rest of the procedure.
- C. Drain wash solution and apply 4 drops of Blocking Reagent to each slide and incubate for 15 minutes.
- D. Drain Blocking Reagent (do not wash off the Blocking Reagent), apply 200 ul of primary antibody solution to each slide, and incubate for 1 hour.
- E. Wash slides with Wash Solution: 3 changes for 5 minutes each.
- F. Drain wash solution, apply 4 drops of Secondary antibody to each slide and incubate for 1 hour.
- G. Wash slides with Wash Solution: 3 changes for 5 minutes each.
- H. Drain wash solution, apply 4 drops of DAB Substrate to each slide and develop for 5-10 minutes. Check development with microscope.
- I. Wash slides with Wash Solution: 3 changes for 5 minutes each.
- J. Drain wash solution, apply 4 drops of Hematoxylin to each slide and stain for 1-3 minutes. Increase time if darker counterstaining is desired.
- K. Wash slides with Wash Solution: 2-3 changes for 2 minutes each.
- L. Drain wash solution and apply 4 drops of Bluing Solution to each slide for 1-2 minutes.
- M. Rinse slides in distilled water.
- N. Soak slides in 70% reagent alcohol: 3 minutes with intermittent agitation.
- O. Soak slides in 95% reagent alcohol: 2 changes for 3 minutes each with intermittent agitation.
- P. Soak slides in 100% reagent alcohol: 3 changes for 3 minutes each with intermittent agitation. Drain slides for 10 seconds between each change.
- Q. Soak slides in Xylene: 3 changes for 3 minutes each with intermittent agitation. Drain slides for 10 seconds between each change.
- R. Apply 2-3 drops of non-aqueous mounting media to each slide and mount coverslip.
- S. Lay slides on a flat surface to dry prior to viewing under microscope.

NOTES:

Use treated slides (e.g. HistoBond) to assure adherence of FFPE sections to slide.

Prior to deparaffinization, heat slides overnight in a 60 degrees Celcius oven.

All steps in which Xylene is used should be performed in a fume hood.

For Epitope Retrieval, a microwave or pressure cooker may be substituted for the steamer method. Adjust times as necessary depending on conditions.



For the initial IHC run with a new primary antibody, test tissues with and without Epitope Retrieval. In some instances, Epitope Retrieval may not be necessary.

200 ul is the recommended maximum volume to apply to a slide for full coverage. Using more than 200 ul may allow solutions to wick off the slide and create drying artifacts. For small tissue sections less than 200 ul may be used. 5 minutes of development with DAB Substrate should be sufficient. Do not develop for more than 10 minutes. If 5 minutes of development causes background staining, further dilution of the primary antibody may be necessary. Hematoxylin should produce a light nuclear counterstain so as not to obscure the DAB staining. Counterstain for 1-1 1/2 minutes for nuclear antigens. Counterstain for 2-3 minutes for cytoplasmic and membranous antigens. If darker counterstaining is desired increase time (up to 10 minutes).





Novus Biologicals USA

10730 E. Briarwood Avenue
Centennial, CO 80112
USA
Phone: 303.730.1950
Toll Free: 1.888.506.6887
Fax: 303.730.1966
nb-customerservice@bio-techne.com

Bio-Techne Canada

21 Canmotor Ave
Toronto, ON M8Z 4E6
Canada
Phone: 905.827.6400
Toll Free: 855.668.8722
Fax: 905.827.6402
canada.inquires@bio-techne.com

Bio-Techne Ltd

19 Barton Lane
Abingdon Science Park
Abingdon, OX14 3NB, United Kingdom
Phone: (44) (0) 1235 529449
Free Phone: 0800 37 34 15
Fax: (44) (0) 1235 533420
info.EMEA@bio-techne.com

General Contact Information

www.novusbio.com
Technical Support: nb-technical@bio-techne.com
Orders: nb-customerservice@bio-techne.com
General: novus@novusbio.com

Products Related to NB500-238

NB800-PC1	HeLa Whole Cell Lysate
HAF007	Goat anti-Mouse IgG Secondary Antibody [HRP]
NB720-B	Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]
NBP1-96981-0.5mg	Mouse IgG2a Kappa Isotype Control (M2AK)

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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