Product Datasheet

Survivin Antibody
NB500-201SS

Unit Size: 0.025 ml

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

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NB500-201SS

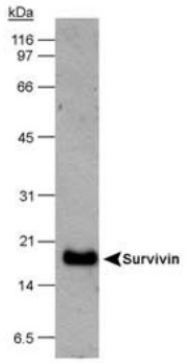
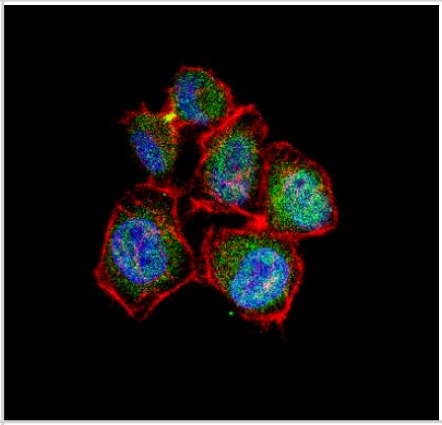
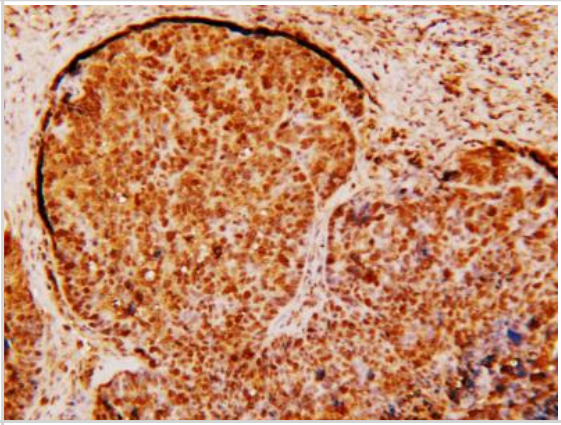
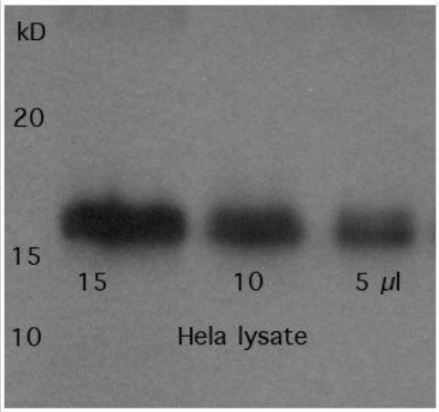
Survivin Antibody

Product Information	
Unit Size	0.025 ml
Concentration	1 mg/ml
Storage	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.02% Sodium Azide
Purity	Immunogen affinity purified
Buffer	PBS
Target Molecular Weight	16.5 kDa

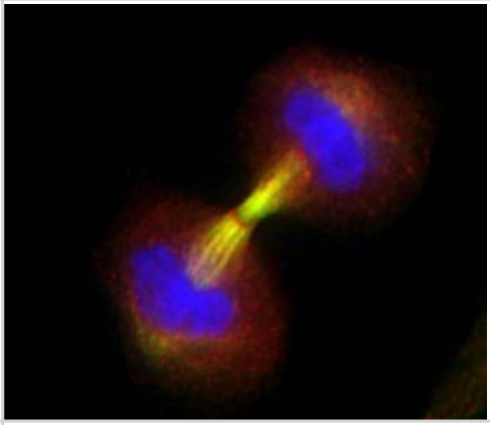
Product Description	
Host	Rabbit
Gene ID	332
Gene Symbol	BIRC5
Species	Human, Mouse, Rat, Canine, Feline, Guinea Pig, Hamster
Species Reactivity	Human, mouse, rat, feline, hamster, canine and guinea pig.
Immunogen	Full length recombinant human Survivin [UniProt# O15392]

Product Application Details	
Applications	Western Blot, Simple Western, Chromatin Immunoprecipitation, ELISA, Flow Cytometry, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation
Recommended Dilutions	Chromatin Immunoprecipitation 1:10-1:500, Immunocytochemistry/Immunofluorescence 1:50-1:250, Immunohistochemistry 1:50-1:100, Immunohistochemistry-Paraffin 1:50-1:500, Immunoprecipitation 1:10-1:500, Western Blot 1 ug/ml, ELISA, Immunohistochemistry-Frozen, Flow Cytometry, Simple Western 1:25
Application Notes	This Survivin antibody is useful for Chromatin Immunoprecipitation, Immunocytochemistry/Immunofluorescence, Immunohistochemistry on paraffin-embedded sections, Immunoprecipitation and Western Blot. In WB, a band at ~16.5 kDa can be seen. For IHC, prior antigen retrieval (pressure cooking) is recommended for cytoplasmic and nuclear detection of Survivin. Immunohistochemistry-Frozen and Flow Cytometry were reported in scientific literature.In Simple Western only 10-15 uL of the recommended dilution is used per data point.

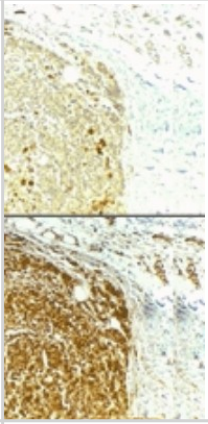
Images

Western Blot: Survivin Antibody [NB500-201] - Detection of survivin in HeLa whole cell lysate (30 ug) using NB 500-201 at 1ug/ml (1 minute exposure).	
Immunocytochemistry/Immunofluorescence: Survivin Antibody [NB500-201] - IF Confocal analysis of HeLa cells using Survivin antibody (NB500 -201, 1:10). An Alexa Fluor 488-conjugated Goat to rabbit IgG was used as secondary antibody (green). Actin filaments were labeled with Alexa Fluor 568 phalloidin (red). DAPI was used to stain the cell nuclei (blue).	
Immunohistochemistry-Paraffin: Survivin Antibody [NB500-201] - analysis of Survivin in ovarian cancer tissue using anti-Survivin antibody. Image from verified customer review.	
Western Blot: Survivin Antibody [NB500-201] - Analysis of Survivin in Hela cell lysate. Image courtesy of anonymous customer review.	

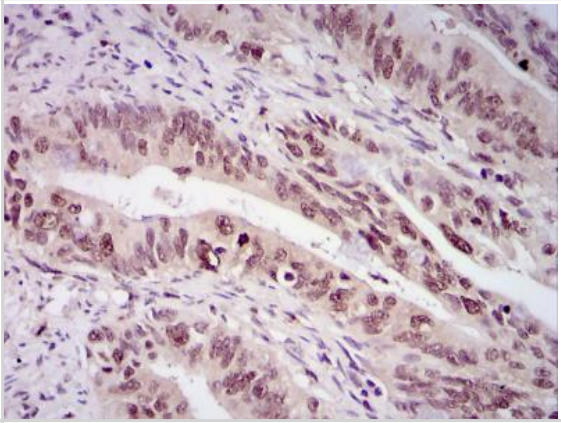
Immunocytochemistry/Immunofluorescence: Survivin Antibody [NB500-201] - Telophase with accumulation of survivin in the midbodies of two daughter cells.



Immunohistochemistry: Survivin Antibody [NB500-201] - The top photo is a no primary antibody control stain and the bottom photo is anti-survivin staining of melanoma. Photo courtesy of Dr. Dario Altieri, Yale University.



Immunohistochemistry-Paraffin: Survivin Antibody [NB500-201] - IHC staining of Survivin in human rectal cancer using DAB with hematoxylin counterstain.



Simple Western: Survivin Antibody [NB500-201] - Simple Western lane view shows a specific band for Survivin in 1.0 mg/ml of HeLa lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.

Publications

Yi XP, Han T, Li YX et al. Simultaneous silencing of XIAP and survivin causes partial mesenchymal-epithelial transition of human pancreatic cancer cells via the PTEN/PI3K/Akt pathway Mol Med Rep. 2015 Feb 20 [PMID: 25707849]

Cornago Protomartir M. Caracterizacion de los efectos de los inhibidores de las histona desacetilasas en lineas celulares de glioma Thesis. 2014 Sep 01 (WB, ICC/IF, Human)

Yamazaki H, Takagi S, Hosoya K. Survivin suppressor (YM155) enhances chemotherapeutic efficacy against canine histiocytic sarcoma in murine transplantation models Research in Veterinary Science. 2015 Feb 11 [PMID: 25744435] (IHC-P, Canine)

Details:
Survivin antibody used at 1:500 dilution in IHC-P application on sections of canine histiocytic sarcoma/HS xenografts from mice treated or not with survivin suppressor sepantronium bromide / YM155. Canine HS cell lines used in these experiments were CHS-4, CTT, and LHS and their derivatives CHS-4-CR, CTT-CR, and LHS-CR as CCNU-resistant HS cells (see full text for detailed protocols, data images shown in Fig 5a).

Lu H, Cao X, Chen Q et al. The Expression and Role of MEKK3 in Renal Clear Cell Carcinoma. ANat Rec (Hoboken). 2014 Nov 12 [PMID: 25388155]

Vogt N, Abramov D, Koch K et al. No evidence of cell cycle dysregulation in mantle cell lymphoma in vivo. Leuk. Lymphoma. 2014 Oct 15 [PMID: 25315075] (ICC/IF, Human)

Cornago M, Garcia-Alberich C, Blasco-Angulo N et al. Histone deacetylase inhibitors promote glioma cell death by G2 checkpoint abrogation leading to mitotic catastrophe. Cell Death Dis. 2014 Oct 03 [PMID: 25275596] (WB, ICC/IF, Human)

Details:
Survivin antibody used for ICC-IF on U251-MG cells in metaphase after HDAC inhibitor suberanilohydroxamic acid/SAHA treatment (Figure 7a). Antibody also used for WB on lysates of glioma cell lines U87, U251-MG, LN229 cells as well as human GBM primary cultures C55, C65, C48 and C60 treated or not with HDAC inhibitor SAHA and valproic acid/VPA (Figure 7b and c)

Grazia G, Vegetti C, Benigni F et al. Synergistic anti-tumor activity and inhibition of angiogenesis by cotargeting of oncogenic and death receptor pathways in human melanoma. Cell Death Dis. 2014 Oct 03 [PMID: 25275595] (WB, Human)

Details:
Survivin antibody used for WB on lysates of melanoma/mesothelioma cells lines Me13 and Me41 treated or not with AZD6244 (A), BEZ235 (B) TRAIL (T) and their combinations (AB, ABT). Blots shown in Figure 5a

Artesi M, Kroonen J, Bredel M et al. Connexin 30 expression inhibits growth of human malignant gliomas but protects them against radiation therapy. Neuro-oncology. 2014 Aug 25 [PMID: 25155356] (WB, Human)

Fu DR, Kato D, Watabe A et al. Prognostic Utility of Apoptosis Index, Ki-67 and Survivin Expression in Dogs with Nasal Carcinoma Treated with Orthovoltage Radiation Therapy. J Vet. Med. Sci. 2014 Aug 25 [PMID: 25151945] (IHC-P, Canine)

Althoff K, Lindner S, Odersky A et al. miR-542-3p exerts tumor suppressive functions in neuroblastoma by downregulating Survivin. Int. J. Cancer 2014 Jul 21 [PMID: 25046253] (ICC/IF, Human)

Details:
Survivin antibody used in ICC-IF on IMR-32, SHEP, SK-N-BE and WAC II human neuroblastoma cell lines after miR-542-3p or control expression - Cells fixed in 4% paraformaldehyde, permeabilized in PBS containing 0.1% Triton X-100 for 10 min, primary used at 1:250 dilution with 1 hour incubation (Figure 3C).

Cao XQ, Lu HS, Zhang L et al. MEKK3 and Survivin Expression in Cervical Cancer: Association with Clinicopathological Factors and Prognosis. Asian Pac. J. Cancer Prev. 2014 Jul 21 [PMID: 25040987] (WB, IHC-P, Human)

Details:
Survivin antibody used for WB (1:1000 dilution/ 4C ON incubation) on tissues from patients of cervical cancer and chronic cervicitis (Figure 2). Antibody also used for IHC-P at 1:100 dilution - 4C ON incubation on cases of cervical cancer, cervical intraepithelial neoplasia and chronic cervicitis (Figure 4).

Ramirez-Labrada A, Lopez-Royuela N, Jarauta V et al. Two death pathways induced by sorafenib in myeloma cells: Puma-mediated apoptosis and necroptosis. Clin Transl Oncol 2014 Jul 19 [PMID: 25037851] (WB, Human)

Details:
Survivin antibody used for WB on lysates of multiple myeloma (MM) cell lines - MM.1S, H929, U266 and RPMI 8226 cells treated or not with 7 uM sorafenib for 24 h at 4 different time points (Fig. 5a).

More publications at http://www.novusbio.com/NB500-201

Procedures

Western Blot Protocol Specific for Survivin Antibody (NB500-201)
Western Blot Procedure
1) Cells were pelleted, washed in 1XPBS, suspended in ice water (~ 5 x 10(6) cells/ml), and placed on ice
2) Lysates were prepared with the addition of 2X lysis buffer [2% SDS/ 50mM Tris-HCl / 10% glycerol]
3) Lysates were heated to 95 degrees C for 3 minutes and then microfuged at room temperature for 10 minutes
4) 50 ug of lysate were electrophoresed (150 V) through a 4-15% PAGE
5) Proteins were transferred (60 V) onto an Immobilon-P membrane (Millipore Corp.) for 45 minutes
6) The blot was blocked overnight at 4 degrees C in blocking buffer [1XPBS, pH 7 / 5% nonfat milk / 0.1% Tween-20]
7) Washed the blot in 1XPBS / 0.1% Tween-20
8) Incubated the blot with 1 ug/ml of (NB500-201) anti-Survivin antibody, diluted in blocking buffer, for 2 hours at room temperature
9) Washed the blot in 1XPBS / 0.1% Tween-20
10) Reacted the blot with HRP-conjugated donkey anti-rabbit Ig, diluted in 1XPBS / 0.1% Tween-20, for 30 minutes at room temperature
11) Washed the blot in 1XPBS / 0.1% Tween-20
12) Visualized blot by ECL and autoradiography
NOTE: HeLa whole cell extracts (NB800-PC1) were used as a positive control for this antibody.

Immunohistochemistry-Paraffin Protocol Specific for Survivin Antibody (NB500-201)

Materials

1) 1 Phosphate buffered saline (pH 7.6): NaCl 137mmol/L, KCl 2.7mmol/L, Na2HPO4 4.3mmol/L, KH2PO4 1.4 mmol/L
2) Citrate buffer, 0.01 M, pH6.0, Sodium Citrate 3g, Citric acid 0.4g
3) 3% Hydrogen peroxide
4) Primary antibody
5) Blocking serum (normal serum)
6) Biotinylated secondary antibody
7) DAB staining kit

Methods

1. Dewax and hydration of slides using xylene and EtOH:
Dry slides for 20 min in a 60 C oven
Add Xylene, 2 x 10 min
100%, 95%, 80%, and 70% EtOH, 5 min each EtOH concentration
Rinse in PBS, 5'

2 Antigen retrieval method (only for paraffin slides)
1a. High-pressure antigen retrieval procedure (recommended method)
Place slides in a glass slide holder (ensure that the slide holder is completely filled with slides, slides without sections if necessary, to ensure even heating. The entire slide holder is immersed in 1000 ml of Citrate buffer (0.01M, pH6.0) within a pressure cooker
Once steam is produced, and ONLY when steam is visible, from the pressure cooker (usually 15-20 min), the required high-pressure will have been reached, and slides will be incubated for 2 min.
Turn off heat, and allow buffer and slides to cool to room temperature
Slides are then rinsed in PBS for 5 minutes
2. Add 3% hydrogen peroxide solution, 10'at RT, then PBS, 3X5'
3. Normal blocking serum, 20'at RT
4. Incubate with Primary Ab, 4C overnight or 1.5 hours at 37C
5. Rinse with PBS, 3 X 5' each rinse
6. Add Biotin-conjugated second antibody, 10'at RT
7. Rinse with PBS, 3 X 5' each rinse
8. Add Streptavidin-Peroxidase, 10'at RT
9. Rinse with PBS, 3 X 5' each rinse
10. Staining with DAB solution, 2-5'under microscope
11. Stop the reaction by washing in tap water
12. Counterstain in Haematoxylin for 3-5 minutes
13. 75%, 80%, 95% and 100% ethanol, 5x2', xylene 2 x 10'

Immunoprecipitation Protocol Specific for Survivin Antibody (NB500-201)

Immunoprecipitation Procedure

1) Lyse cells plated in a 60mm dish:
a) 300 ul CHAPS buffer [50mM Tris-HCl, pH 7.5/50mM NaCl/1mM EDTA/1% NP-40/0.1% CHAPS/1mM NaVO4/1mM PMSF]
b) Rock for 20 minutes at 4 degrees C
2) Harvest lysate and spin down the insoluble material at 14K rpm
3) Collect soluble fraction
4) Pre-clear lysate with 40 ul of 50:50 slurry of Protein A beads, rocking for 1 hour at 4 degrees C
5) Spin down beads at 2K rpm, at 4 degrees C
6) Collect pre-cleared lysate
7) Incubate lysate with 5-7ug of anti-Survivin (NB 500-201) overnight, rocking at 4 degrees C
8) Add 50 ul of Protein A 50:50 slurry for 2 hours, rocking at 4C
9) Wash beads with 200 ul of CHAPS buffer, three times
10) Denature immune complex by adding 2x Sample Buffer, containing 2-ME
11) Boil for 10 minutes and load onto an SDS-gel.

Novus Biologicals USA
8100 Southpark Way, A-8
Littleton, CO 80120
USA
Phone: 303.730.1950
Toll Free: 1.888.506.6887
Fax: 303.730.1966
novus@novusbio.com

Novus Biologicals Canada
461 North Service Road West, Unit B37
Oakville, ON L6M 2V5
Canada
Phone: 905.827.6400
Toll Free: 855.668.8722
Fax: 905.827.6402
canada@novusbio.com

Novus Biologicals Europe
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@bio-techne.com

General Contact Information
www.novusbio.com
Technical Support: technical@novusbio.com
Orders: orders@novusbio.com
General: novus@novusbio.com

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.

For more information on our guarantee, please visit www.novusbio.com/guarantee.