## **Product Datasheet**

# Actin Gamma 1 Antibody - BSA Free NB600-533

Unit Size: 0.1 mg

Store at 4C. Do not freeze.

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**Publications: 6** 

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## NB600-533

Actin Gamma 1 Antibody - BSA Free	
Product Information	
Unit Size	0.1 mg
Concentration	1.0 mg/ml
Storage	Store at 4C. Do not freeze.
Clonality	Polyclonal
Preservative	0.09% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	Tris-Citrate/Phosphate (pH 7.0 - 8.0)
Target Molecular Weight	45 kDa
Product Description	
Host	Rabbit
Gene ID	71
Gene Symbol	ACTG1
Species	Human, Mouse
Reactivity Notes	Human and mouse.
Immunogen	The epitope recognized by this antibody maps to the N-terminus of human Actin Gamma 1 [UniProt# P63261]. The N-terminus of Actin Gamma 1 is highly conserved with Beta Actin and preliminary indications are that NB600-533 also recognizes Beta Actin (GeneID 60).
Product Application Details	
Applications	Western Blot, Simple Western, Flow Cytometry, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunoprecipitation
Recommended Dilutions	Western Blot, Simple Western 1:100, Flow Cytometry, Immunohistochemistry, Immunocytochemistry/ Immunofluorescence 1:100, Immunoprecipitation 1:10-1:500
Application Notes	This Actin Gamma 1 antibody is useful for Immunocytochemistry/Immunofluorescence, Immunoprecipitation and Western Blot.  In Simple Western only 10 - 15 uL of the recommended dilution is used per data point.  See Simple Western Antibody Database for Simple Western validation: Tested in HeLa lysate 1.0 mg/mL, separated by Size, antibody dilution of 1:100, apparent MW was 49 kDa. Separated by Size-Wes, Sally Sue/Peggy Sue.  The observed molecular weight of the protein may vary from the listed predicted molecular weight due to post translational modifications, post translation cleavages, relative charges, and other experimental factors. Use in FLOW reported in scientific literature (PMID 28501006).



## **Images**

Western Blot: Actin Gamma 1 Antibody [NB600-533] - Knockdown of DLK in differentiated Neuro-2a cells. Neuro-2a cells were infected with an empty lentiviral vector (pLKO.1) or with lentivirus expressing mouse DLK shRNAs (sh73 and sh69). After infection and selection with puromycin, cells were subjected to differentiation for 24 h before being processed for total RNA extraction and whole-cell extracts. Representative Western blots showing levels of DLK, phospho-JNK (p-JNK), total JNK, phospho-c-Jun (p-c-Jun) and actin in infected Neuro-2a cells. Image collected and cropped by CiteAb from the following publication

(https://neuraldevelopment.biomedcentral.com/articles/10.1186/s13064-016-0068-8), licensed under a CC-BY license.

Immunocytochemistry/Immunofluorescence: Actin Gamma 1 Antibody [NB600-533] - Detection of Actin Gamma 1 (Green) in Hela cells using NB600-533 at a 1:50 dilution. Nuclei (Blue) were counterstained using Hoechst 33258.

B

pukC<sup>2,2</sup> shr<sup>2,3</sup> sh6<sup>9</sup>

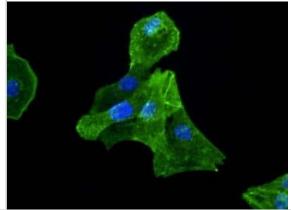
DLK

p-JNK

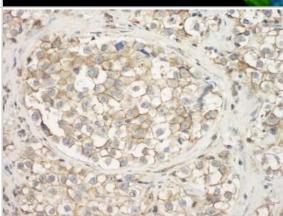
JNK

p-c-jun

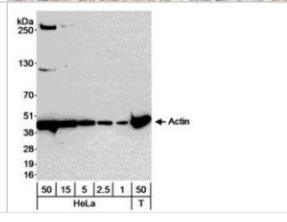
actin



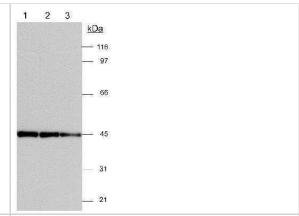
Immunohistochemistry: Actin Gamma 1 Antibody [NB600-533] - Sample: FFPE section of human testicular seminoma. Antibody: Affinity purified rabbit anti-Actin used at a dilution of 1:1,000 (1ug/ml). Detection: DAB



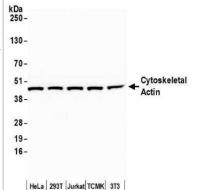
Western Blot: Actin Gamma 1 Antibody [NB600-533] - Whole cell lysate from HeLa (1, 2.5, 5, 15 and 50 ug) and mouse NIH3T3 cells (50 ug), probed with diluted at 0.04 ug/ml.



Western Blot: Actin Gamma 1 Antibody [NB600-533] - Actin Antibody [NB600-533]-Detection of actin in 3T3 (20 ug) lysates. ECL detection 30 seconds. Lane 1 - 1:5,000 dilution Lane 2 - 1:10,000 dilution Lane 3 - 1:15,000 dilution



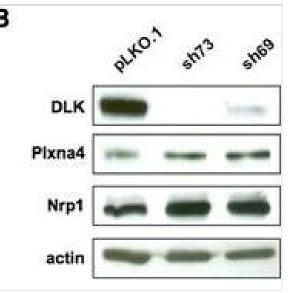
Western Blot: Actin Gamma 1 Antibody [NB600-533] - Detection of human and mouse Cytoskeletal Actin by western blot. Samples: Whole cell lysate (15 ug) from HeLa, HEK293T, Jurkat, mouse TCMK-1, and mouse NIH 3T3 cells prepared using NETN lysis buffer. Antibody: Affinity purified rabbit anti-Cytoskeletal Actin antibody NB600-533 used for WB at 0.1 ug/ml. Detection: Chemiluminescence with an exposure time of 10 seconds.



Simple Western: Actin Gamma 1 Antibody [NB600-533] - Simple Western lane view shows a specific band for Actin Gamma 1 in 1.0 mg/ml of HeLa lysate. This experiment was performed under reducing conditions using the 12-230kDa separation system.



Western Blot: Actin Gamma 1 Antibody [NB600-533] - Validation of RNAseq data by qRT-PCR & Western blot analyses, a The relative mRNA level of DLK & axon guidance genes in infected cells was analyzed by gRT-PCR, normalized to three housekeeping genes & calculated with the  $\Delta\Delta$ CT method. The value of mRNA expression for each gene in control cells (pLKO.1) was arbitrarily set to 1. Data are the mean ± SEM (error bars) from three independent experiments carried out in triplicate.  $\hat{x}$ , p < 0.05; \*\*, p < 0.01; \*\*\*, p < 0.001; \*\*\*\*, p < 0.0001; ns, p > 0.05. b Representative Western blots showing levels of DLK, Plxna4, Nrp1 & actin in control & DLK-depleted Neuro-2a cells. c Quantitative densitometric measurements of DLK, Plxna4 & Nrp1 protein levels in infected cells. Results are normalized to actin level in control cells, which were set to 1, & represent mean ± SEM (error bars) from three independent experiments. \*, p < 0.05; \*\*, p < 0.01; \*\*\*, p < 0.001 Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/27468987), licensed under a CC-BY license. Not internally tested by Novus Biologicals.





### **Publications**

Moore HR, Alspach E, Hirsch JL et al. The p38MAPK-MK2-HSP27 Pathway Regulates the mRNA Stability of the Senescence-Associated Secretory Phenotype bioRxiv 2019-06-08 (WB, Human)

Flanagan KC, Alspach E, Pazolli E, Parajuli S. c-Myb and C/EBPb regulate OPN and other senescence-associated secretory phenotype factors. Oncotarget. 2018-01-02 [PMID: 29416593] (WB, Human)

Luo H, Yao L, Zhang Y, Li R. Liquid chromatography-mass spectrometry-based quantitative proteomics analysis reveals chondroprotective effects of astragaloside IV in interleukin-1b-induced SW1353 chondrocyte-like cells. Biomed. Pharmacother. 2017-05-10 [PMID: 28501006] (FLOW, WB, Human)

Blondeau A, Lucier JF, Matteau D et al. Dual leucine zipper kinase regulates expression of axon guidance genes in mouse neuronal cells. Neural Dev 2016-07-28 [PMID: 27468987] (WB)

Duxin JP, Moore HR, Sidorova J et al. Okazaki fragment processing-independent role for human Dna2 enzyme during DNA replication. J Biol Chem 2012-06-01 [PMID: 22570476] (WB, Human)

Duxin JP, Dao B, Martinsson P et al. Human Dna2 Is a Nuclear Mitochondrial DNA Maintenance Protein. Mol Cell Biol;29(15):4274-4282. 2009-01-01 [PMID: 19487465]



#### **Procedures**

## Serum protocol for Actin Gamma 1 Antibody (NB600-533)

Actin Gamma 1 Antibody:

Western Blot Protocol

- 1. Perform SDS-PAGE (4-12%) on samples to be analyzed, loading 20 ug of total protein per lane.
- 2. Transfer proteins to Nitrocellulose according to the instructions provided by the manufacturer of the transfer apparatus.
- 3. Stain the blot using ponceau S for 1-2 minutes to access the transfer of proteins onto the nitrocellulose membrane. Rinse the blot in water to remove excess stain and mark the lane locations and locations of molecular weight markers using a pencil.
- 4. Rinse the blot in TBS for approximately 5 minutes.
- 5. Block the membrane using 5% non-fat dry milk + 1% BSA in TBS for 1 hour.
- 6. Dilute the rabbit anti-actin primary antibody (NB 600-533) in blocking buffer and incubate 2 hours at room temperature.
- 7. Wash the membrane in water for 5 minutes and apply the diluted rabbit-IgG HRP-conjugated secondary antibody in blocking buffer (as per manufacturer's instructions) and incubate 1 hour at room temperature.
- 8. Wash the blot in TBS containing 0.05-0.1% Tween-20 for 10-20 minutes.
- 9. Wash the blot in type I water for an additional 10-20 minutes (this step can be repeated as required to reduce background).
- 10. Apply the detection reagent of choice in accordance with the manufacturer's instructions (Amersham's ECL is the standard reagent used at Novus Biologicals).

Note: Tween-20 can be added to the blocking buffer at a final concentration of 0.05-0.2%, provided it does not interfere with antibody-antigen binding.





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## **Products Related to NB600-533**

NB800-PC1 HeLa Whole Cell Lysate

HAF008 Goat anti-Rabbit IgG Secondary Antibody [HRP]

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

NBP2-24891 Rabbit IgG Isotype Control

#### 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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