Product Datasheet

PUMA Antibody
NB110-81760

Unit Size: 0.1 ml

Store at 4C. Do not freeze.

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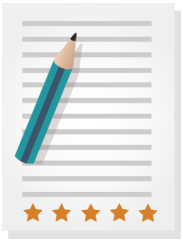


NB110-81760

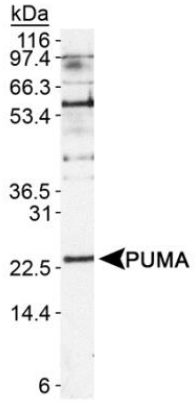
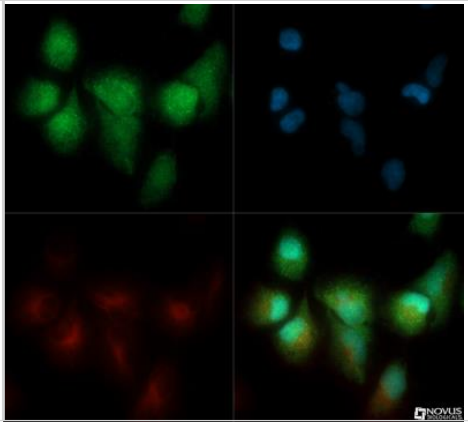
PUMA Antibody

Product Information	
Unit Size	0.1 ml
Concentration	1 mg/ml
Storage	Store at 4C. Do not freeze.
Clonality	Polyclonal
Preservative	0.1% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	Tris-Citrate/Phosphate (pH 7.0 - 8.0)
Target Molecular Weight	24 kDa

Product Description	
Host	Rabbit
Gene ID	27113
Gene Symbol	BBC3
Species	Human, Mouse
Reactivity Notes	Human and mouse.
Specificity/Sensitivity	This is specific for isoform 1 of PUMA.
Immunogen	Synthetic peptide made to an N-terminal portion of human PUMA (within residues 1-100). [NCBI Sequence NP_001120712]

Product Application Details	
Applications	Western Blot, Immunocytochemistry/Immunofluorescence
Recommended Dilutions	Western Blot 1 ug/ml, Immunocytochemistry/Immunofluorescence 1:500 - 1:2000
Application Notes	This PUMA antibody is useful for Immunocytochemistry/Immunofluorescence and Western blot, where a band is seen at ~24 kDa. In ICC/IF mitochondrial staining was observed. 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.

Images

Western Blot: PUMA Antibody [NB110-81760] - Detection of PUMA in HL-60 whole cell lysates using NB110-81760.	
Immunocytochemistry/Immunofluorescence: PUMA Antibody [NB110-81760] - PUMA antibody was tested in Hela cells with DyLight 488 (green). Nuclei and alpha-tubulin were counterstained with DAPI (blue) and Dylight 550 (red).	

Procedures

Western Blot protocol for PUMA Antibody (NB110-81760)
PUMA Antibody: https://www.novusbio.com/products/puma-antibody_nb110-81760
Western Blot Protocol

1. Perform SDS-PAGE (4-12% MOPS) on samples to be analyzed, loading 30 ug of total protein per lane.
2. Transfer proteins to Nitrocellulose according to the instructions provided by the manufacturer of the transfer apparatus.
3. Rinse membrane with dH2O and then 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% NFDM + 1% BSA in TBS + Tween, 1 hour at RT.
6. Rinse the membrane in dH2O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.
7. Dilute the rabbit anti-PUMA primary antibody (NB 110-81760) in blocking buffer and incubate 1 hour at room temperature.
8. Rinse the membrane in dH2O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.
9. Apply the diluted rabbit-IgG 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 [TBS + 0.1% Tween] 3 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 (Pierce ECL).

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

Immunocytochemistry/Immunofluorescence protocol for PUMA Antibody (NB110-81760)
PUMA Antibody: https://www.novusbio.com/products/puma-antibody_nb110-81760
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 NB110-81760

NB800-PC3	HL-60 Whole Cell Lysate
HAF008	Goat anti-Rabbit IgG Secondary Antibody [HRP (Horseradish Peroxidase)]
NB7156	Goat anti-Rabbit IgG (H+L) Secondary Antibody
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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