Product Datasheet

Vanilloid R1/TRPV1 Antibody NBP1-97417SS

Unit Size: 0.025 ml

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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Publications: 1

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Updated 6/15/2014 v.20.1

NBP1-97417SS

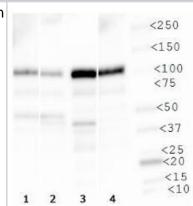
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Images

Western Blot: TRPV1 Antibody [NBP1-97417] - WB analysis of TRPV1 in 1. Ntera2 cell lysate, 2. HepG2 cell lysate, 3. MCF7 cell lysate and 4. Cos7 cell lysate.



Immunocytochemistry/Immunofluorescence: TRPV1 Antibody [NBP1-97417] - TRPV1 antibody was tested in Ntera-2 cells with FITC (green). Nuclei and alpha-tubulin were counterstained with DAPI (blue) and Dylight 550 (red).

Publications

Schwab AJ, Ebert AD. Sensory neurons do not induce motor neuron loss in a human stem cell model of spinal muscular atrophy. PLoS ONE 2014 Jul 24 [PMID: 25054590] (ICC/IF, Human)

Details:

TRPV1 antibody used for ICC-IF on induced pluripotent stem cells (iPSCs) - 4% paraformaldehyde /PFA -20 min RT fixation, 0.2% Triton X-100 - 30 min RT permeablization, blocking with 5% normal goat serum.



Procedures

Protocol specific for TRPV1 antibody (NBP1-97417)

Western Blot Protocol

1. Perform SDS-PAGE on samples to be analyzed, loading 40 ug of total protein per lane.

2. Transfer proteins to membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.

3. Stain according to standard Ponceau S procedure (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.

4. Rinse the blot.

5. Block the membrane using standard blocking buffer for at least 1 hour.

6. Wash the membrane in wash buffer three times for 10 minutes each.

7. Dilute primary antibody in blocking buffer and incubate 1 hour at room temperature.

8. Wash the membrane in wash buffer three times for 10 minutes each.

9. Apply the diluted 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 three 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.

Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%.

Immunocytochemistry Protocol

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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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.

