

Product Datasheet

CXCR4 Antibody NB100-74396SS

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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Updated 6/15/2014 v.20.1

NB100-74396SS

CXCR4 Antibody

Product Information	
Unit Size	0.025 ml
Concentration	This product is unpurified. The exact concentration of antibody is not quantifiable.
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.05% Sodium Azide
Purity	Whole antisera

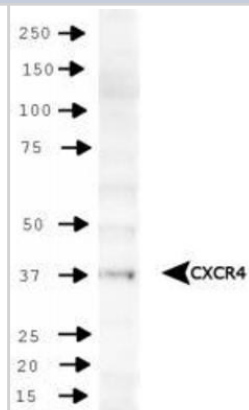
Product Description	
Host	Rabbit
Gene ID	7852
Gene Symbol	CXCR4
Species	Human, Mouse, Rat
Species Reactivity	Human, rat and mouse.
Immunogen	A synthetic peptide made to a C-terminal region of the human CXCR4 protein (within residues 300-352). [Swiss-Prot P61073]

Product Application Details	
Applications	Western Blot, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin
Recommended Dilutions	Immunocytochemistry/Immunofluorescence 1:50, Immunohistochemistry 1:200-1:1000, Immunohistochemistry-Paraffin 1:200-1:1000, Western Blot 1:5000
Application Notes	This CXCR4 antibody is useful for Immunohistochemistry-paraffin embedded sections, Immunocytochemistry/Immunofluorescence and Western Blot. In Western blot a band is observed ~ 40 kDa. Prior to immunostaining paraffin tissues, antigen retrieval with sodium citrate buffer (pH 6.0) is recommended.

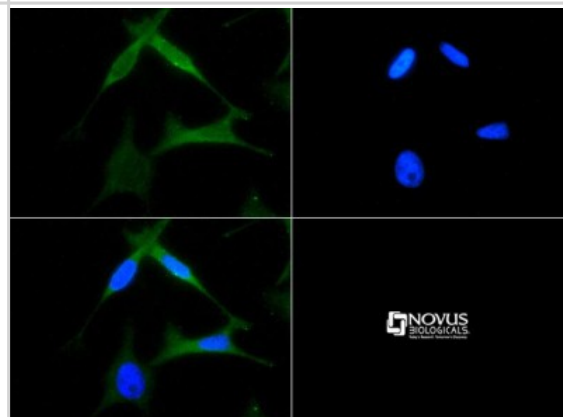


Images

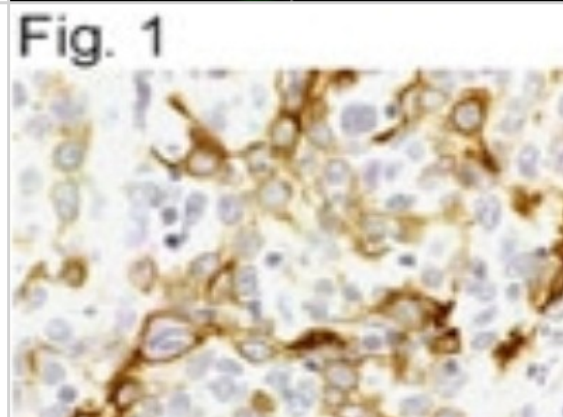
Western Blot: CXCR4 Antibody [NB100-74396] - Analysis of CXCR4 in HeLa whole cell extract.



Immunocytochemistry/Immunofluorescence: CXCR4 Antibody [NB100-74396] - CXCR4 antibody was tested in HeLa cells with FITC (green). Nuclei were counterstained with DAPI (blue).



Immunohistochemistry: CXCR4 Antibody [NB100-74396] - Figure 1 illustrates immunostaining of CXCR4 in human cervical carcinoma tissue sections.



Publications

Lee HW, Cho HJ, Lee SJ et al. Tpl2 induces castration resistant prostate cancer progression and metastasis. *Int. J. Cancer*. 2014 Oct 01 [PMID: 25274482] (WB, ICC/IF, IHC-P, Human)

Details:
CXCR4 antibody used for WB and ICC-IF on PC3 and 22Rv1 cells in relation to Tpl2 (i.e. cells with Tpl2 knockdown and with Tpl2 over-expression) . Antibody also used in IHC-P for the detection of CXCR4 on orthotopic xenograft tumors (Figure 6).

Gakhar G, Navarro VN, Jurish M et al. Circulating Tumor Cells from Prostate Cancer Patients Interact with E-Selectin under Physiologic Blood Flow. *PLoS One* 2013 Dec 27 [PMID: 24386459] (ICC/IF, Human)

Hoh BL, Hosaka K, Downes DP et al. Stromal cell-derived factor-1 promoted angiogenesis and inflammatory cell infiltration in aneurysm walls. *J Neurosurg*. 2013 Oct 25 [PMID: 24160472]

Huang Yf, Chen MJ, Wu MH, Hung SC. The use of hypoxic cultured mesenchymal stem cell for oncolytic virus therapy. *Cancer Gene Ther* 2013 Apr 26 [PMID: 23618949]

Mo W, Chen J, Patel A et al. CXCR4/CXCL12 Mediate Autocrine Cell- Cycle Progression in NF1-Associated Malignant Peripheral Nerve Sheath Tumors. *Cell* 2013 Feb 28 [PMID: 23434321] (WB, IHC-P, Mouse)

Chen C, Cao J, Song X et al. Adrenaline administration promotes the efficiency of granulocyte colony stimulating factor-mediated hematopoietic stem and progenitor cell mobilization in mice *Int J Hematol* 2012 Dec 8 [PMID: 23224606] (WB, Mouse)

Kumagai K, Takeuchi R, Ishikawa H et al. Low-intensity pulsed ultrasound accelerates fracture healing by stimulation of recruitment of both local and circulating osteogenic progenitors *J Orthop Res* 2012 Sep [PMID: 22419401] (IHC, ICC/IF, Mouse)

Hasegawa T, McLeod DS, Prow T, Merges C, Grebe R, Luttj GA. Vascular Precursors in Developing Human Retina. *Invest Ophthalmol Vis Sci*;49(5):2178-92. May 1 2008. [PMID: 18436851] (IHC, Human)

Stumm RK, Zhou C, Ara T et al. CXCR4 regulates interneuron migration in the developing neocortex. *J Neurosci*;23(12):5123-30. 2003 Jun 15. [PMID: 12832536] (IHC, Rat)

Stumm RK, Rummel J, Junker V et al. A dual role for the SDF-1/CXCR4 chemokine receptor system in adult brain: isoform-selective regulation of SDF-1 expression modulates CXCR4-dependent neuronal plasticity and cerebral leukocyte recruitment after focal ischemia. *J Neurosci*;22(14):5865-78. 2002 Jul 15. [PMID: 12122049] (IHC, ICC/IF, WB, Human, Rat)



Procedures

Western Blot Protocol for CXCR4 Antibody (NB100-74396)

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%.

Immunohistochemistry-Paraffin Embedded Sections

Antigen Unmasking:

Bring slides to a boil in 10 mM sodium citrate buffer (pH 6.0) then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench-top for 30 minutes.

Staining:

1. Wash sections in deionized water three times for 5 minutes each.
2. Wash sections in wash buffer for 5 minutes.
3. Block each section with 100-400 ul blocking solution for 1 hour at room temperature.
4. Remove blocking solution and add 100-400 ul diluted primary antibody. Incubate overnight at 4 C.
5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
6. Add 100-400 ul biotinylated diluted secondary antibody. Incubate 30 minutes at room temperature.
7. Remove secondary antibody solution and wash sections three times with wash buffer for 5 minutes each.
8. Add 100-400 ul Streptavidin-HRP reagent to each section and incubate for 30 minutes at room temperature.
9. Wash sections three times in wash buffer for 5 minutes each.
10. Add 100-400 ul DAB substrate to each section and monitor staining closely.
11. As soon as the sections develop, immerse slides in deionized water.
12. Counterstain sections in hematoxylin.
13. Wash sections in deionized water two times for 5 minutes each.
14. Dehydrate sections.
15. Mount coverslips.

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 1:25,000 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.

