# **Product Datasheet**

# MICALL1 Antibody - Azide and BSA Free H00085377-B01P

Unit Size: 0.05 mg

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

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# H00085377-B01P

MICALL1 Antibody - Azide and BSA Free

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Product Information	
Unit Size	0.05 mg
Concentration	Concentrations vary lot to lot. See vial label for concentration. If unlisted please contact technical services.
Storage	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	No Preservative
Isotype	IgG
Purity	Protein A purified
Buffer	PBS (pH 7.4)
Product Description	
Host	Mouse
Gene ID	85377
Gene Symbol	MICALL1
Species	Human, Mouse, Rat
Specificity/Sensitivity	MICAL-L1 - MICAL-like 1,
Immunogen	MICAL-L1 (AAI48805.1, 1 a.a 863 a.a.) full-length human protein. MAGPRGALLAWCRRQCEGYRGVEIRDLSSSFRDGLAFCAILHRHRPDLLDFDS LSKDNVFENNRLAFEVAEKELGIPALLDPNDMVSMSVPDCLSIMTYVSQYYNHF CSPGQAGVSPPRKGLAPCSPPSVAPTPVEPEDVAQGEELSSGSLSEQGTGQT PSSTCAACQQHVHLVQRYLADGRLYHRHCFRCRRCSSTLLPGAYENGPEEGT FVCAEHCARLGPGTRSGTRPGPFSQPKQQHQQQLAEDAKDVPGGGPSSSAP AGAEADGPKASPEARPQIPTKPRVPGKLQELASPPAGRPTPAPRKASESTTPA PPTPRPRSSLQQENLVEQAGSSSLVNGRLHELPVPKPRGTPKPSEGTPAPRK DPPWITLVQAEPKKKPAPLPPSSSPGPPSQDSRQVENGGTEEVAQPSPTASLE SKPYNPFEEEEEDKEEEAPAAPSLATSPALGHPESTPKSLHPWYGITPTSSPKT KKRPAPRAPSASPLALHASRLSHSEPPSATPSPALSVESLSSESASQTAGAELL EPPAVPKSSSEPAVHAPGTPGNPVSLSTNSSLASSGELVEPRVEQMPQASPG LAPRTRGSSGPQPAKPCSGATPTPLLLVGDRSPVPSPGSSSPQLQVKSSCKE NPFNRKPSPAASPATKKATKGSKPVRPPAPGHGFPLIKRKVQADQYIPEEDIHG EMDTIERRLDALEHRGVLLEEKLRGGLNEGREDDMLVDWFKLIHEKHLLVRRE SELIYVFKQQNLEQRQADVEYELRCLLNKPEKDWTEEDRAREKVLMQELVTLIE QRNAIINCLDEDRQREEEEDKMLEAMIKKKEFQREAEPEGKKKGKFKTMKMLK LLGNKRDAKSKSPRDKS
Notes	This product is produced by and distributed for Abnova, a company based in Taiwan.
Product Application Details	
Applications	Western Blot, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin, Immunoprecipitation
Recommended Dilutions	Western Blot 1:100-1:2000, Immunohistochemistry 1:10-1:500, Immunocytochemistry/ Immunofluorescence, Immunoprecipitation 1:10-1:500, Immunohistochemistry-Paraffin 1:400



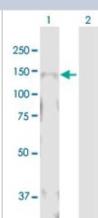
## **Application Notes**

Antibody reactive against Recombinant Protein with GST tag on ELISA and Western Blot and also on transfected lysate in western blot. GST tag alone is used as a negative control. Antibody is also useful for Immunohistochemistry on paraffin-embedded sections. Immunocytochemistry/Immunofluorescence, Immunohistochemistry and Immunoprecipitation were reported in scientific literature, but Immunocytochemistry/Immunofluorescence has been experiencing lot-to-lot variations. Use in Immunocytochemistry/immunofluorescence reported in scientific literature (PMID: 24019528).

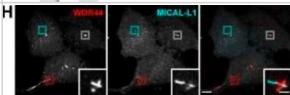
# **Images**

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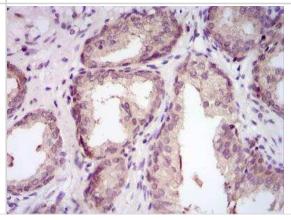
Western Blot: MICALL1 Antibody [H00085377-B01P] - Analysis of MICALL1 expression in transfected 293T cell line by MICALL1 polyclonal antibody. Lane 1: MICAL-L1 transfected lysate(94.93 KDa). Lane 2: Non-transfected lysate.



Immunocytochemistry/Immunofluorescence: MICALL1 Antibody [H00085377-B01P] - GRAF/WDR44 label a subset of tubular endosomes. Insets show magnifications of the boxed areas. Scale bars: 10 um; scale bars of insets: 2 um. Image collected and cropped by CiteAb from the following publication (https://rupress.org/jcb/article/doi/10.1083/jcb.201811014/151714/GRAF 2-WDR44-and-MICAL1-mediate-Rab81011dependent) licensed under a



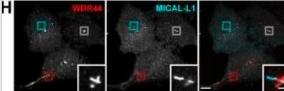
Immunohistochemistry-Paraffin: MICALL1 Antibody [H00085377-B01P] - IHC staining of MICALL1 in human prostate cancer using DAB with hematoxylin counterstain.



Western Blot: MICALL1 Antibody [H00085377-B01P] - Analysis of MICALL1 expression in HepG2.



Immunocytochemistry/ Immunofluorescence: MICALL1 Antibody [H00085377-B01P] - GRAF/WDR44 label a subset of tubular endosomes. (A-C) HeLa cells were incubated with methanol (vehicle) or BFA (5 μg/ml) for 5 or 15 min. (A) Percentage of cells with endogenous (endo.) WDR44 tubules. n = 7-25. (B) Confocal images of cells stained with α-WDR44 & α-TGN46 or preincubated with Alexa Fluor 546–Transferrin (10 µg/ml, 1 h). (C) Manders colocalization coefficients for the indicated proteins with endogenous WDR44 structures. n = 10-50cells. (D) Confocal images of transfected HeLa cells showing colocalization of RFP-WDR44 with GFP-STX16 & GFP-VAMP3. (E) Immunoprecipitation (IP) of transfected 293T cells with α-GFP. uGRAF2 was coimmunoprecipitated by GFP-MICAL1, but not by any other member of the MICAL family. (F) Confocal images of transfected HeLa cells stained with α-WDR44. In the case of MICAL3pF1KA0819 & MICAL-L2, boxed areas show tubules positive only for WDR44 (red), only for MICAL3pF1KA0819/MICAL-L2 (cyan), or shared by the two proteins (white). (G) Percentage of transfected HeLa cells with endogenous (endo.) WDR44 tubules. n = 4. (H) Confocal images of untransfected HeLa cells stained with α-WDR44 & α-MICAL-L1. Boxed areas show tubules positive only for WDR44 (red), only for MICAL-L1 (cyan), or shared by the two proteins (white). (I) Internalized Integrin-β1 in shRNA-expressing HeLa cells after uptake of α-Integrin-β1 & following a 4-h chase. n = 4. (A, C, G, & I) Data are means  $\pm$  SEM; \*, P < 0.05; \*\*, P < 0.01; \*\*\*, P < 0.001; & \*\*\*\*, P < 0.0001. (B, D, F, & H) Insets show magnifications of the boxed areas. Scale bars: 10 µm; scale bars of insets: 2 µm. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32344433), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



#### **Publications**

R Sakai, R Fukuda, S Unida, M Aki, Y Ono, A Endo, S Kusumi, D Koga, T Fukushima, M Komada, T Okiyoneda The integral function of endocytic recycling compartment is regulated by RFFL-mediated ubiquitination of Rab11 effectors J. Cell. Sci., 2019-02-07;0(0):. 2019-02-07 [PMID: 30659120]

Bali L, Christopher H, Rose W et al. Disruptions in endocytic traffic contribute to the activation of the NLRP3 inflammasome. Sci Signal. 2023-02-21 [PMID: 36809026]

Finicle BT Targeting endolysosomal trafficking with synthetic sphingolipid analogs to improve the delivery of oligonucleotide therapeutics Thesis 2023-01-01 (ICC/IF)

Rangaraj N, Vaibhava V, Sudhakar C et al. Transferrin-induced signaling through transferrin receptor and AKT kinase mediates formation of Rab8- and MICAL-L1-positive tubules involved in receptor recycling bioRxiv 2023-02-07 (ICC/IF, Human)

Dhawan K, Naslavsky N, Caplan S Coronin2A links actin-based endosomal processes to the EHD1 fission machinery Molecular biology of the cell 2022-08-03 [PMID: 35921168]

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Siddiqa A, Massimi P, Pim D, Banks L Diverse Papillomavirus Types Induce Endosomal Tubulation Front Cell Infect Microbiol 2019-05-28 [PMID: 31192164] (ICC/IF, Human)

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Mukadam AS, Breusegem SY, Seaman MNJ. Analysis of novel endosome-to-Golgi retrieval genes reveals a role for PLD3 in regulating endosomal protein sorting and amyloid precursor protein processing Cell. Mol. Life Sci. 2018-01-24 [PMID: 29368044] (WB, Human)

More publications at <a href="http://www.novusbio.com/H00085377-B01P">http://www.novusbio.com/H00085377-B01P</a>



#### **Procedures**

# Immunohistochemistry-Paraffin Protocol Specific for H00085377-B01P: MICALL1 Antibody 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'
- Normal blocking serum, 20'at RT
- 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'





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### Products Related to H00085377-B01P

HAF007 Goat anti-Mouse IgG Secondary Antibody [HRP]

NB720-B Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]

NBP1-97019-5mg Mouse IgG Isotype Control

NBP2-70017PEP MICALL1 Antibody Blocking Peptide

#### 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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