Datasheet for ABIN487492

anti-IQGAP1 antibody

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Overview

Quantity:	0.1 mg
Target:	IQGAP1
Reactivity:	Human, Mouse, Rat
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This IQGAP1 antibody is un-conjugated
Application:	Western Blotting (WB), Immunoprecipitation (IP)

Product Details

Immunogen:	Purified native Bovine adrenal IQGAP1. Remarks: Hybridoma was established by fusion of mouse plasmacytoma ell NS-1 withBalb/c mouse lymph nodes.
Clone:	VIC1-IC11-IVB2
Isotype:	IgG2a
Specificity:	This antibody reacts with Human, Mouse and Rat IQGAP1.
Cross-Reactivity (Details):	Species reactivity (tested):Human, Mouse and Rat.
Characteristics:	Synonyms: KIAA0051, p195, Ras GTPase-activating-like protein IQGAP1
Purification:	Protein-A Agarose Chromatography of hybridoma supernatant.

Target Details

Target:	IQGAP1

Target Details

Alternative Name:	IQGAP1 (IQGAP1 Products)
Background:	IQ domain GTPase activating protein (IQGAP) is a ~170 kDa protein that is a target molecule for activated Cdc42 and Rac1. IQGAP binds actin and calmodulin and is an effector for the Rho family of GTPases, providing a direct link between the activated GTPase and the actin cytoskeleton. Additionally, the calmodulin-binding protein IQGAP1 binds stoichiometrically to beta-catenin, an oncoprotein integral to cell-cell adhesion and proliferative signaling, and regulates the association of beta-catenin with the cell-cell adhesion complex.Synonyms: KIAA0051, Ras GTPase-activating-like protein IQGAP1, p195
Gene ID:	8826
UniProt:	P46940
Pathways:	Signaling Events mediated by VEGFR1 and VEGFR2

Application Details

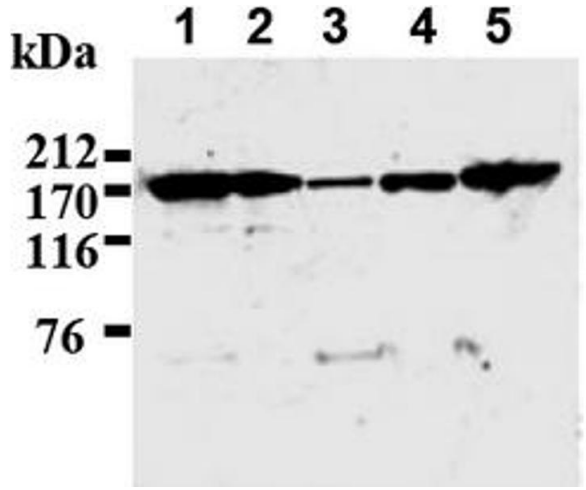
Application Notes:	Western blot: 1 µg/mL for chemiluminescence detection system. Positive Controls: HeLa, A431, NIH/3T3, C2C12, Rat-1. Immunoprecipitation: 2 µg/200-300 µL of cell extract. Positive Control: HeLa. Detailed procedure is provided in Protocols. Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
Protocol:	SDS-PAGE & Western Blotting1) Wash the cells 3 times with PBS and suspend with 10 volume of cold Lysis buffer (50 mMTris-HCl, pH 7. 2, 250 mM NaCl, 0. 1% NP-40, 2 mM EDTA, 10% glycerol) containingappropriate protease inhibitors. Incubate it at 4°C with rotating for 30 minutes, thensonicate briefly (up to 10 seconds). 2) Centrifuge the tube at 12,000 x g for 10 minutes at 4°C and transfer the supernatant toanother tube. Measure the protein concentration of the supernatant and add the Lysisbuffer to make 8 mg/mL solution. 3) Mix the sample with equal volume of Laemmli's sample buffer. 4) Boil the samples for 2 minutes and centrifuge. Load 10 µL of the sample per lane in a 1mm thick SDS-polyacrylamide gel for electrophoresis. 5) Blot the protein to a polyvinylidene difluoride (PVDF) membrane at 1 mA/cm2 for 1 hourin a semi-dry transfer system. (Transfer Buffer: 25 mM Tris, 190 mM glycine, 20% MeOH). See the manufacture's manual for the transfer procedure. 6) To reduce nonspecific binding, soak the membrane in 10% skimmed milk (in PBS, pH7. 2) for 1 hour at room temperature, or overnight at 4°C. 7) Incubate the membrane with primary antibody diluted with PBS, pH 7. 2 containing 1%skimmed milk as suggested in the APPLICATIONS for 1 hour at room temperature. (Theoptimal antibody concentration will depend on the experimental conditions.)8) Wash the

Application Details

	membrane with PBS (5 minutes x 6 times). 9) Incubate the membrane with the 1: 10000 POD-conjugated anti-mouse IgG diluted with1% skimmed milk (in PBS, pH 7. 2) for 1 hour at room temperature. 10) Wash the membrane with PBS (5 minutes x 6 times). 11) Wipe excess buffer from the membrane, then incubate it with appropriatechemiluminescence reagents for 1 minute. Remove extra reagent from the membrane bydabbing with a paper towel, and seal it in plastic wrap. 12) Expose to an X-ray film in a dark room for 5 minutes. Develop the film as usual. Theconditions for exposure and development may vary. Positive Controls for Western blotting: HeLa, A431, NIH/3T3, C2C12, Rat-1. Immunoprecipitation: 1) Wash the cells 3 times with PBS and suspend with 10 volume of cold Lysis buffer (50 mMTris-HCl, pH 7. 2, 250 mM NaCl, 0. 1% NP-40, 2 mM EDTA, 10% glycerol) containingappropriate protease inhibitors. Incubate it at 4°C with rotating for 30 minutes, thensonicate briefly (up to 10 seconds). 2) Centrifuge the tube at 12,000 x g for 10 minutes at 4°C and transfer the supernatant toanother tube. 3) Add 2 µg of the anti-IQGAP1 monoclonal antibody into 250 µL of the supernatant. Mixwell and incubate with gentle agitation for 30-120 minutes at 4°C. 4) Add 20 µL of 50% protein A agarose beads resuspended in the Lysis buffer. Mix well andincubate with gentle agitation for 60 minutes at 4°C. 5) Wash the beads 3-5 times with ice-cold Lysis buffer (centrifuge the tube at 2,500 x g for10 seconds). 6) Resuspend the beads in 20 µL of Laemmli's sample buffer, boil for 3-5 minutes, andcentrifuge for 5 minutes. Use 10 µL/lane for the SDS-PAGE analysis. (See SDS-PAGE & Western blotting.)Positive Control for Immunoprecipitation: HeLa.
Restrictions:	For Research Use only

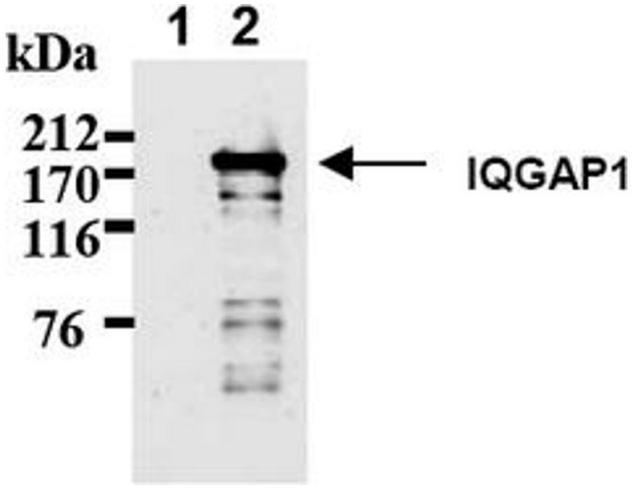
Handling

Concentration:	1.0 mg/mL
Buffer:	PBS, pH 7.2 containing 50 % Glycerol without preservatives.
Preservative:	Without preservative
Storage:	-20 °C
Storage Comment:	Store the antibody (in aliquots) at -20 °C. Avoid repeated freezing and thawing. Shelf life: one year from despatch.
Expiry Date:	12 months



Western Blotting

Image 1.



Western Blotting

Image 2.

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