Datasheet for ABIN968007

anti-Clathrin antibody (AA 4-171)

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Overview

Quantity:	150 µg
Target:	Clathrin
Binding Specificity:	AA 4-171
Reactivity:	Human, Mouse, Rat, Chicken, Dog
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This Clathrin antibody is un-conjugated
Application:	Immunohistochemistry (IHC), Western Blotting (WB), Immunofluorescence (IF), Immunoprecipitation (IP), BioImaging (BI)

Product Details

Immunogen:	Rat Clathrin Heavy Chain aa. 4-171
Clone:	23-Clathrin Heavy Chain
Isotype:	IgG1
Cross-Reactivity:	Human, Chicken, Dog (Canine), Mouse (Murine)
Characteristics:	1. Since applications vary, each investigator should titrate the reagent to obtain optimal results. 2. Please refer to us for technical protocols. 3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing. 4. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

Product Details

Purification:	The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

Target Details

Target:	Clathrin
Abstract:	Clathrin Products
Background:	Clathrin is the major protein component in the coat formed around pits and vesicles involved in receptor-mediated endocytosis. Clathrin forms a non-covalently bound triskelion structure composed of three heavy chains (192 kDa each) and three light chains (23-25 kDa each). Each leg of the triskelion structure contains one heavy and one light chain. The three heavy chains forming the triskelion structure are attached at their respective proximal ends like spokes on a wheel. Clathrin heavy chain is composed of a terminal globular domain, a distal segment containing several areas sensitive to enzymatic cleavage, and a proximal segment which contains a light chain binding site. The proximal and distal domains are connected by a joint segment at which there is a sharp bend in the heavy chains of fully-assembled triskelia. Although the calculated molecular weight is 192 kDa, clathrin heavy chain migrates at approximately 180 kDa.
Molecular Weight:	180 kDa

Application Details

Application Notes:	Immunofluorescent staining and bioimaging Methanol Procedure for a 96 well plate: Remove media from wells. Add 100 µl/well fresh 3.7% Formaldehyde in PBS. Incubate for 10 minutes at room temperature (RT). Flick out and add 100 µl/well 90% methanol. Incubate for 5 minutes at RT. Flick out and wash twice with PBS. Flick out PBS and add 100 µl/well blocking buffer (3% FBS in PBS). Incubate for 30 minutes at RT. Flick out and add diluted antibody (diluted in blocking buffer). Incubate for 1 hour at RT. Wash three times with PBS. Flick out PBS and add second step reagent. Incubate for 1 hour at RT. Wash three times with PBS. Triton-X 100 Procedure for a 96 well plate: Remove media from wells. Add 100 µl/well fresh 3.7% Formaldehyde in PBS. Incubate for 10 minutes at room temperature (RT). Flick out and add 100 µl/well 0.1% Triton-X 100. Incubate for 5 minutes at RT. Flick out and wash twice with PBS. Flick out PBS and add 100 µl/well blocking buffer (3% FBS in PBS). Incubate for 30 minutes at RT. Flick out and add diluted antibody (diluted in blocking buffer). Incubate for 1 hour at RT. Flick out and wash three times with PBS. Flick out and add second step reagent. Incubate for 1 hour at RT. Flick out and wash three times with PBS.

Application Details

Comment:	Related Products: ABIN968535
Restrictions:	For Research Use only

Handling

Format:	Liquid
Concentration:	250 µg/mL
Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09 % sodium azide.
Preservative:	Sodium azide
Precaution of Use:	This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.
Storage:	-20 °C
Storage Comment:	Store undiluted at -20° C.

Publications

Product cited in:

Padilla, Chang, Pacheco-Rodriguez, Adamik, Moss, Vaughan: "Interaction of FK506-binding protein 13 with brefeldin A-inhibited guanine nucleotide-exchange protein 1 (BIG1): effects of FK506." in: **Proceedings of the National Academy of Sciences of the United States of America**, Vol. 100, Issue 5, pp. 2322-7, (2003) (PubMed).

Kalthoff, Groos, Kohl, Mahrhold, Ungewickell: "Clint: a novel clathrin-binding ENTH-domain protein at the Golgi." in: **Molecular biology of the cell**, Vol. 13, Issue 11, pp. 4060-73, (2002) (PubMed).

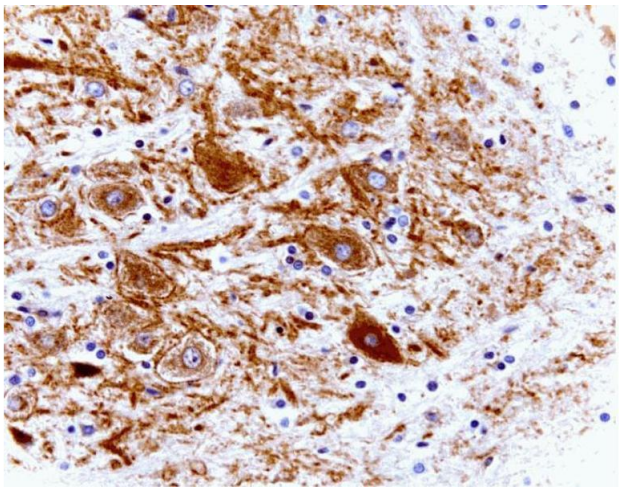
van Kerkhof, Sachse, Klumperman, Strous: "Growth hormone receptor ubiquitination coincides with recruitment to clathrin-coated membrane domains." in: **The Journal of biological chemistry**, Vol. 276, Issue 6, pp. 3778-84, (2001) (PubMed).

Okamoto, Karam, Jeng, Forte, Goldenring: "Identification of clathrin and clathrin adaptors on tubulovesicles of gastric acid secretory (oxyntic) cells." in: **The American journal of physiology**, Vol. 274, Issue 4 Pt 1, pp. C1017-29, (1998) (PubMed).

Liu, Wong, Craik, Brodsky: "Regulation of clathrin assembly and trimerization defined using

recombinant triskelion hubs." in: **Cell**, Vol. 83, Issue 2, pp. 257-67, (1995) (PubMed).

Images

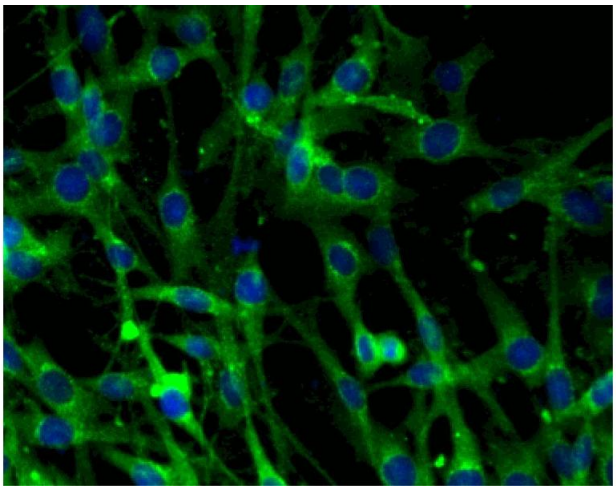


Immunohistochemistry (Paraffin-embedded Sections)

Image 1. Rat cerebellum, zinc-fixed paraffin embedded tissue (left), 40X

Western Blotting

Image 2. Western blot analysis of Clathrin Heavy Chain on HeLa cell lysate (center). Lane 1: 1:1000, lane 2: 1:2000, lane 3: 1:4000 dilution of anti-Clathrin Heavy Chain.



Immunofluorescence

Image 3. Immunofluorescent staining of C6 cells (right). Cells were seeded in a collagen coated 384 well imaging plate at ~ 6,000 cells per well. After overnight incubation, cells were stained using the methanol fix/perm protocol and the anti-Clathrin Heavy chain antibody. The second step reagent was Alexa Fluor® 488 goat anti mouse Ig (Invitrogen). The image was taken on a Pathway 855 or 435 imager using a 20x objective. This antibody also stained SH-SY5Y, SK-N-SH cells using both the Triton X100 and methanol fix/perm protocols.