Technical Data Sheet

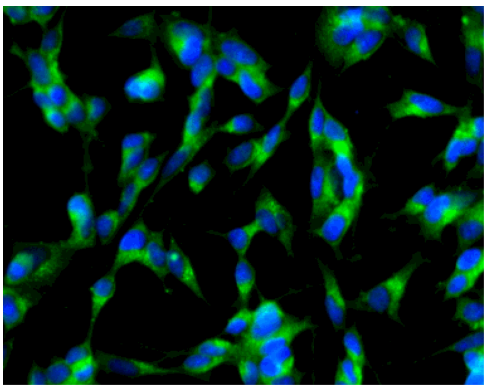
Purified Mouse Anti-Adaptin α

Product Information

Material Number:	**610502**
Size:	150 µg
Concentration:	250 µg/ml
Clone:	8/Adaptin α
Immunogen:	Mouse Adaptin α [A] aa. 38-215
Isotype:	Mouse IgG1
Reactivity:	QC Testing: Rat Tested in Development: Human, Mouse, Dog
Target MW:	112 kDa
Storage Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09% sodium azide.

Description

Sorting of integral membrane proteins at various stages of the endocytic and secretory pathways is mediated by vesicular trafficking between a variety of organelles. Two sorting signals are tyrosine-based and dileucine-based signals that interact with heterotetrameric adaptor protein complexes (AP-1, AP-2, AP-3, and AP-4), which are associated with the vesicle coats. These coatomers contain two large Adaptin proteins (α, γ, δ, or ε and β1, β2, β3, or β4, respectively) that are noncovalently linked to one medium chain (µ1, µ2, µ3, or µ4) and one small chain (σ1, σ2, σ3, or σ4). The AP-1 and AP-3 complexes are involved in protein sorting from the TGN and endosomes, while AP-2 adaptor complexes are involved in clathrin-mediated endocytosis. In the AP-2 complex, Adaptin α is expressed in two very similar isoforms from two different genes. Adaptin α [A] (112kDa) is expressed primarily in brain, while Adaptin α [C] (105kDa) is expressed in brain, liver and other tissues.



Western blot analysis of Adaptin α on a rat cerebrum lysate (left). *Lane 1: 1:1000, lane 2: 1:2000, lane 3: 1:4000 dilution of the anti-Adaptin α antibody.*

Immunofluorescent staining of SH-SY5Y cells (right). *Cells were seeded in a 384 well collagen coated imaging plate (Material # 353962) at ~ 8,000 cells per well. After overnight incubation, cells were stained using the methanol fix/perm protocol (see Recommended Assay Procedure; Bioimaging protocol link) and the anti-Adaptin-α antibody. The second step reagent was Alexa Fluor® 488 goat anti mouse Ig (Invitrogen)(pseudo colored green). Cell nuclei were counter stained with Hoechst 33342 (pseudo colored blue). The image was taken on a BD Pathway™ 855 or 435 Bioimager System using a 20x objective and merged using the BD AttoVison ™ software.This antibody also stained SK-N-SH, C6, U87 and U373 cells using both the Triton X100 and methanol fix/perm protocols (see Recommended Assay Procedure; Bioimaging protocol link).*

Preparation and Storage

Store undiluted at -20°C.

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

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Application Notes

Application

Western blot	Routinely Tested
Immunofluorescence	Tested During Development
Immunohistochemistry-formalin (antigen retrieval required)	Tested During Development
Immunoprecipitation	Tested During Development
Bioimaging	Tested During Development

Suggested Companion Products

Catalog Number	Name	Size	Clone
611463	Rat Cerebrum Lysate	500 µg	(none)
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)
554001	FITC Goat Anti-Mouse Ig	0.5 mg	Polyclonal

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. 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.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
4. Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.
5. This antibody has been developed and certified for the bioimaging application. However, a routine bioimaging test is not performed on every lot. Researchers are encouraged to titrate the reagent for optimal performance.
6. Triton is a trademark of the Dow Chemical Company.
7. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
8. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

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Robinson MS. Cloning of cDNAs encoding two related 100-kD coated vesicle proteins (alpha-adaptins). *J Cell Biol.* 1989; 108(3):833-842. (Biology)

Santolini E, Puri C, Salcini AE, et al. Numb is an endocytic protein. *J Cell Biol.* 2000; 151(6):1345-1351. (Biology: Immunoprecipitation, Western blot)

Teo M, Tan L, Lim L, Manser E. The tyrosine kinase ACK1 associates with clathrin-coated vesicles through a binding motif shared by arrestin and other adaptors. *J Biol Chem.* 2001; 276(21):18392-18398. (Biology: Immunofluorescence, Western blot)

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