

Technical Data Sheet

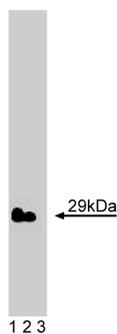
Purified Mouse Anti-Cellugyrin

Product Information

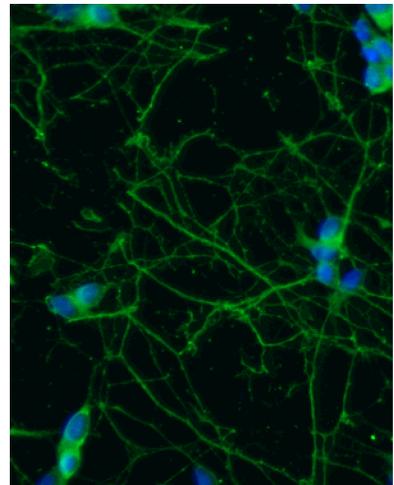
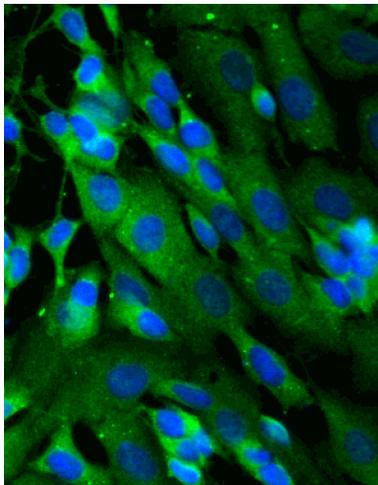
Material Number:	611128
Size:	50 µg
Concentration:	250 µg/ml
Clone:	24/Cellugyrin
Immunogen:	Rat Cellugyrin aa. 95-204
Isotype:	Mouse IgG1
Reactivity:	QC Testing: Rat Tested in Development: Mouse
Target MW:	29 kDa
Storage Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09% sodium azide.

Description

Neurotransmitter release is mediated by the synaptic vesicle cycle at the presynaptic nerve terminal. This exocytic process involves vesicle docking at the plasma membrane, which is followed by priming and fusion. Following exocytosis, the empty vesicles are recycled for continued neurotransmitter release. Vesicle fusion is mediated by a protein complex consisting of both synaptic vesicle and synaptic plasma membrane components, such as synaptotagmin, synaptobrevin, and synaptogyrin. It is thought that synaptic vesicle-mediated exocytosis is very similar to other exocytic pathways. In line with this idea, cellugyrin is a synaptogyrin-like protein that is widely expressed in non-neuronal tissues. Cellugyrin and synaptogyrin share 47% amino acid sequence identity. Additionally, both cellugyrin and synaptogyrin are phosphorylated in their cytoplasmic tails by pp60c-src. This suggests a role for phosphorylation in the regulation of membrane trafficking. Thus, cellugyrin is a ubiquitously expressed exocytic protein of which synaptogyrin is a specialized neuronal version. However, the exact function of either protein in exocytosis remains to be determined.



Western blot analysis of cellugyrin on a rat liver lysate. Lane 1: 1:250, lane 2: 1:500, lane 3: 1:1000 dilution of the mouse anti-cellugyrin antibody.



Immunofluorescent staining of undifferentiated (left) and differentiated (right) SH-SY5Y cells. Undifferentiated cells were seeded in a collagen coated 384 well imaging plate (Material # 353962) at ~ 8,000 cells per well. After overnight incubation, cells were stained using the methanol fix/permeabilization protocol (see Recommended Assay Procedure; Bioimaging protocol link) and the anti-Cellugyrin antibody. Differentiated cells: cells were seeded in a 96 well, collagen coated imaging plate (Material # 353219) at ~ 5,000 cells per well. Cells were incubated with 50 nM ATRA (Sigma, R2625) for 5 days, followed by 50 ng/ml BDNF (Sigma, B3795) for 5 days. Differentiated cells were fixed and stained using the methanol fix/permeabilization protocol, and the anti-Cellugyrin antibody. The second step reagent in both cases was Alexa Fluor® 488 goat anti mouse Ig (Invitrogen)(pseudo colored green). Cell nuclei were counter stained with Hoechst 33342 (pseudo colored blue). The images were 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 undifferentiated SH-SY5Y, SK-N-SH, C6, U87 and U373 cells using both the Triton X100 and methanol fix/permeabilization protocols (see Recommended Assay Procedure; Bioimaging protocol link).

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Preparation and Storage

Store undiluted at -20°C.

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

Application Notes

Application

Western blot	Routinely Tested
Immunofluorescence	Tested During Development
Bioimaging	Tested During Development

Suggested Companion Products

Catalog Number	Name	Size	Clone
611467	Rat Liver 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. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
7. Triton is a trademark of the Dow Chemical Company.
8. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Janz R, Sudhof TC. Cellugyrin, a novel ubiquitous form of synaptogyrin that is phosphorylated by pp60c-src. *J Biol Chem.* 1998; 273(5):2851-2857. (Biology)

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