

Piccolo

Cat.No. 142 008; Recombinant rabbit antibody, 100 µg recombinant IgG (lyophilized)

Data Sheet

Reconstitution/ Storage	100 µg purified recombinant IgG, lyophilized. Albumin and azide were added for stabilization. For reconstitution add 100 µl H ₂ O to get a 1mg/ml solution in PBS. Then aliquot and store at -20°C to -80°C until use. Antibodies should be stored at +4°C when still lyophilized. Do not freeze! For detailed information, see back of the data sheet.
Applications	WB: not recommended (see remarks) IP: not tested yet ICC: 1 : 500 IHC: 1 : 500 IHC-P: 1 : 100
Clone	Rb294E1
Subtype	IgG1 (κ light chain)
Immunogen	Recombinant protein corresponding to residues near the central region of rat Piccolo (UniProt Id: Q9JKS6)
Reactivity	Reacts with: mouse (Q9QYX7-1), rat (Q9JKS6). Other species not tested yet.
Remarks	This antibody is a chimeric antibody based on the monoclonal mouse antibody SY-294E1. The constant regions of the heavy and light chains have been replaced with rabbit specific sequences. The antibody can therefore be used with standard anti-rabbit secondary reagents. The antibody has been expressed in mammalian cells. WB: This antibody is less sensitive than the polyclonal product 142 113. We recommend 142 113 for westernblotting. Due to its large size, Piccolo requires special gel-electrophoresis and Western blot protocols for visualization by immunoblotting. Excellent results can be obtained with the 4-12% TRIS-glycine gradient gels from Anamed or NuPAGE 3-8% TRIS-Acetate gradient gels from Invitrogen.

Background

Piccolo, also referred to as **Aczonin**, is a large protein which consists of an N-terminal Zn²⁺ finger, several piccolo-bassoon homology domains (PBH-domains) and C-terminal PDZ and C2 domains. In general it is found together with bassoon, a related huge multi-domain protein of the CAZ (cytoskeletal matrix assembled at active zones). Piccolo is supposed to be a scaffolding protein for proteins involved in endo- and exocytosis of synaptic vesicles. Recently piccolo has been shown to interfere with clathrin mediated endocytosis by binding to the F-actin and dynamin binding protein Abp1.

Selected General References

- Unitary assembly of presynaptic active zones from Piccolo-Bassoon transport vesicles. Shapira M, Zhai RG, Dresbach T, Bresler T, Torres VI, Gundelfinger ED, Ziv NE, Garner CC *Neuron* (2003) 382: 237-52. .
- Interactions between Piccolo and the actin/dynamin-binding protein Abp1 link vesicle endocytosis to presynaptic active zones. Fenster SD, Kessels MM, Qualmann B, Chung WJ, Nash J, Gundelfinger ED, Garner CC *The Journal of biological chemistry* (2003) 27822: 20268-77. .
- Localization of the presynaptic cytomatrix protein Piccolo at ribbon and conventional synapses in the rat retina: comparison with Bassoon. Dick O, Hack I, Altmann WD, Garner CC, Gundelfinger ED, Brandstätter JH *The Journal of comparative neurology* (2001) 4392: 224-34. .
- An unusual C(2)-domain in the active-zone protein piccolo: implications for Ca(2+) regulation of neurotransmitter release. Gerber SH, Garcia J, Rizo J, Südhof TC *The EMBO journal* (2001) 207: 1605-19. .
- Piccolo, a presynaptic zinc finger protein structurally related to bassoon. Fenster SD, Chung WJ, Zhai R, Cases-Langhoff C, Voss B, Garner AM, Kaempf U, Kindler S, Gundelfinger ED, Garner CC *Neuron* (2000) 251: 203-14. .
- Piccolo, a novel 420 kDa protein associated with the presynaptic cytomatrix. Cases-Langhoff C, Voss B, Garner AM, Appeltauer U, Takei K, Kindler S, Veh RW, De Camilli P, Gundelfinger ED, Garner CC *European journal of cell biology* (1996) 693: 214-23. .
- Gene structure and genetic localization of the PCLO gene encoding the presynaptic active zone protein Piccolo. Fenster SD, Garner CC *International journal of developmental neuroscience : the official journal of the International Society for Developmental Neuroscience* () 203-5: 161-71. .

Access the online factsheet including applicable protocols at <https://sysy.com/product/142008> or scan the QR-code.



TO BE USED IN VITRO / FOR RESEARCH ONLY
NOT TOXIC, NOT HAZARDOUS, NOT INFECTIOUS, NOT CONTAGIOUS

FAQ - How should I store my antibody?

Shipping Conditions

- All our antibodies and control proteins / peptides are shipped lyophilized (vacuum freeze-dried) and are stable in this form without loss of quality at ambient temperatures for several weeks.

Storage of Sealed Vials after Delivery

- **Unlabeled** and **biotin-labeled antibodies** and **control proteins** should be stored at 4°C before reconstitution. **They must not be stored in the freezer when still lyophilized!** Temperatures below zero may cause loss of performance.
- **Fluorescence-labeled antibodies** should be reconstituted immediately upon receipt. Long term storage (several months) may lead to aggregation.
- **Control peptides** should be kept at -20°C before reconstitution.

Long Term Storage after Reconstitution (General Considerations)

- The storage freezer must not be of the frost-free variety ("no-frost freezer"). This cycle between freezing and thawing (to reduce frost-build-up), which is exactly what should be avoided. For the same reason, antibody vials should be placed in an area of the freezer that has minimal temperature fluctuations, for instance towards the back rather than on a door shelf.
- Aliquot the antibody and store frozen (-20°C to -80°C). Avoid very small aliquots (below 20 µl) and use the smallest storage vial or tube possible. The smaller the aliquot, the more the stock concentration is affected by evaporation and adsorption of the antibody to the surface of the storage vial or tube. Adsorption of the antibody to the surface leads to a substantial loss of activity.
- The addition of glycerol to a final concentration of 50% lowers the freezing point of your stock and keeps your antibody at -20°C in liquid state. This efficiently avoids freeze and thaw cycles.

Product Specific Hints for Storage

Control proteins / peptides

- Store at -20°C to -80°C.

Monoclonal Antibodies

- **Ascites** and **hybridoma supernatant** should be stored at -20°C up to -80°C. **Prolonged storage at 4°C is not recommended!** Unlike serum, ascites may contain proteases that will degrade the antibodies.
- **Purified IgG** should be stored at -20°C up to -80°C. Adding a carrier protein like BSA will increase long term stability. Many of our antibodies already contain carrier proteins. Please refer to the data-sheet for detailed information.

Polyclonal Antibodies

- **Crude antisera:** With anti-microbials added, they may be stored at 4°C. However, frozen storage (-20°C up to -80°C) is preferable.
- **Affinity purified antibodies:** Less robust than antisera. Storage at -20°C up to -80°C is recommended. Adding a carrier protein like BSA will increase long term stability. Most of our antibodies already contain carrier proteins. Please refer to the data-sheet for detailed information.

Fluorescence-labeled Antibodies

- Store as a liquid with 1 : 1 (v/v) glycerol at -20°C. Protect these antibodies from light exposure.

Avoid repeated freeze-thaw cycles for all antibodies!

FAQ - How should I reconstitute my antibody?

Reconstitution

- All our purified antibodies are lyophilized from PBS. To reconstitute the antibody in PBS, add the amount of deionized water given in the respective datasheet. If higher volumes are preferred, add water as mentioned above and then the desired amount of PBS and a stabilizing carrier protein (e.g. BSA) to a final concentration of 2%. Some of our antibodies already contain albumin. Take this into account when adding more carrier protein. For complete reconstitution, carefully remove the lid. After adding water, briefly vortex the solution. You can spin down the liquid by placing the vial into a 50 ml centrifugation tube filled with paper.
- If desired, add small amounts of azide or thimerosal to prevent microbial growth. This is especially recommended if you want to keep an aliquot a 4°C.
- After reconstitution of fluorescence-labeled antibodies, add 1 : 1 (v/v) glycerol to a final concentration of 50%. This lowers the freezing point of your stock and keeps your antibody in liquid state at -20°C.
- Glycerol may also be added to unlabeled primary antibodies. It is a suitable way to avoid freeze-thaw cycles.
- Please refer to our **tips and hints for subsequent storage** of reconstituted antibodies and control peptides and proteins.