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# Calbindin D28k

Cat.No. 214 003; Polyclonal rabbit antibody, 50 µg specific antibody (lyophilized)

#### **Data Sheet**

Reconstitution/ Storage	50 µg specific antibody, lyophilized. Affinity purified with the immunogen. Albumin and azide were added for stabilization. For <b>reconstitution</b> add 50 µl H <sub>2</sub> 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: 1: 1000 up to 1: 5000 (AP staining) IP: yes ICC: 1: 5000 IHC: 1: 500 IHC-P: 1: 500 Clarity: external data (see remarks)
Immunogen	Full-length recombinant human Calbindin D28k protein (UniProt Id: P05937)
Reactivity	Reacts with: human (P05937), rat (P07171), mouse (P12658), monkey, ape, cow. Other species not tested yet.
Matching control	214-0P
Remarks	<b>Clarity</b> : This antibody has been successfully used for CLARITY application in human brain (Woelfle et al., 2023; PMID: 37221592).

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

### Background

Calbindin D28k (CALB1, D28K or CAB27) and Calretinin (Calbindin D29k) are members of the EF-hand calcium-binding protein family, playing essential roles in intracellular calcium homeostasis, neuroprotection, and signal transduction (1). Calbindin D28k is widely expressed in tissues that experience high calcium influx, such as the brain, sensory organs, endocrine tissues, and the enteric nervous system (ENS). In the central nervous system (CNS), Calbindin D28k is an important marker for specific neuronal populations, particularly in Purkinje cells of the cerebellum and GABAergic and glutamatergic neurons of the cortex (2). Outside the CNS, Calbindin D28k plays crucial roles in nonneuronal tissues. In the retina, it contributes to photoreceptor function and calcium signaling (3). In the pancreas, it is present in  $\alpha$ - and  $\beta$ -cells (4). Additionally, it was reported that 31% of myenteric neurons and up to 95% of submucosal neurons in the human ENS express calbindin, suggesting its involvement in both motor and secretory functions (5).

#### Selected References for 214 003

CLARITY increases sensitivity and specificity of fluorescence immunostaining in long-term archived human brain tissue. Woelfle S, Deshpande D, Feldengut S, Braak H, Del Tredici K, Roselli F, Deisseroth K, Michaelis J, Boeckers TM, Schön M BMC biology (2023) 211: 113. . CLARITY; tested species: human

#### Selected General References

'New' functions for 'old' proteins: the role of the calcium-binding proteins calbindin D-28k, calretinin and parvalbumin, in cerebellar physiology. Studies with knockout mice.

Schwaller B et al. Cerebellum (2002) PubMed:12879963

Calbindin D28k-Immunoreactivity in Human Enteric Neurons. Zetzmann K et al. Int J Mol Sci (2018) PubMed:29316719

Types and density of calbindin D28k-immunoreactive ganglion cells in mouse retina. Gu YN et al. Exp Eye Res (2016) PubMed:26874036

Calbindin-D28K immunoreactivity in the human cerebellar cortex. Flace P et al. Anat Rec (Hoboken) (2014) PubMed:24719368

Expression of the novel epithelial Ca2+ channel ECaC1 in rat pancreatic islets. Janssen SW et al. J Histochem Cytochem (2002) PubMed:12019295

Access the online factsheet including applicable protocols at <a href="https://sysy.com/product/214003">https://sysy.com/product/214003</a> or scan the QR-code.



# FAQ - How should I store my antibody?

## **Shipping Conditions**

 All our antibodies and control proteins / peptides are shipped lyophilized (vacuum freezedried) 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 freezethaw cycles.
- Please refer to our tips and hints for subsequent storage of reconstituted antibodies and control peptides and proteins.