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Product Description

300

Monoclonal anti Calbindin D-28k

Product: Monoclonal anti Calbindin D-28k

Code No.: 300

Lot No.: 07 (F)

Form: Lyophilized concentrated supernatant (no preservatives).

Quantity: 200 µl

Reconstitution: with 200 µl / 500 µl or 1 ml bidistilled water.

Description
Monoclonal anti-calbindin D-28k is a mouse IgG1 produced by hybridization of mouse myeloma cells with spleen cells from mice immunized with calbindin D-28k purified from chicken gut (1). The antibody was evaluated for specificity and potency: a) by indirect immunofluorescent or immunoperoxidase labelling as well as Biotin-Avidin labeling of cryostate or vibratome-sections of 4 % paraformaldehyde fixed tissue; b) by immunoenzymatic labelling of immunoblots; c) by radioimmunoassay.

The product is a monoclonal antibody (McAB) against calbindin D-28k, a calcium-binding protein of the EF-hand family related to calmodulin and troponin-C.

McAB 300 reacts specifically with calbindin D-28k on immunoblots of extracts of tissue originating from human, monkey, guinea pig, rabbit, rat, mouse and chicken (but probably not fish, Fig. 1). McAB 300 does not cross-react with calretinin or other known calcium binding-proteins. McAB 300 specifically stains the ^{45}Ca-binding spot of calbindin D-28k (MW 28'000, IEP 4.8) in a two-dimensional gel. In radioimmunoassay it detects calbindin D-28k with a sensitivity of 10 ng/assay and an affinity of 1.6×10^{12} L/M.
McAB 300 against calbindin D-28k specifically localizes calbindin using free-floating or mounted sections of brain, kidney and pancreas of probably all vertebrates with the exception of fishes.

Immunoblot of brain extracts

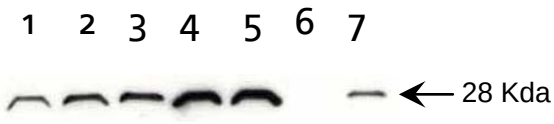

Fig. 1 Immunoblot of brain homogenate of various species with the monoclonal antibody CB300
1: Mouse; 2: Rat; 3. Guinea pig; 4. Rabbit; 5. Macaca fascicularis; 6. Zebrafish; 7. recombinant calbindin D-28k. In all species, only a band at 28 KDa is detected.

Immunohistochemistry on Calbindin D-28k knock-out mice

Antibody CB300 immunolabels a subpopulation of neurons in the normal brain with high efficiency (Fig. 2a) but does not stain in the brain of calbindin D-28k knock out mice (Fig. 2).

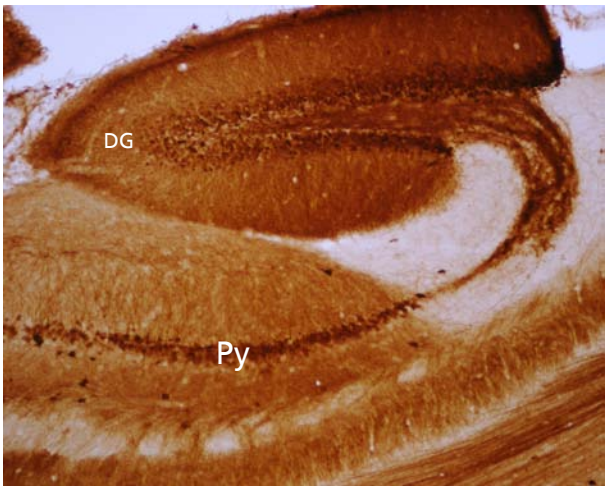

Fig 2a: immunohistochemical staining with McAB300 in the hippocampus of a control mouse. Notice the strong staining of the dentate gyrus (DG) and of the pyramidal cells (Py) of the hippocampus. X100

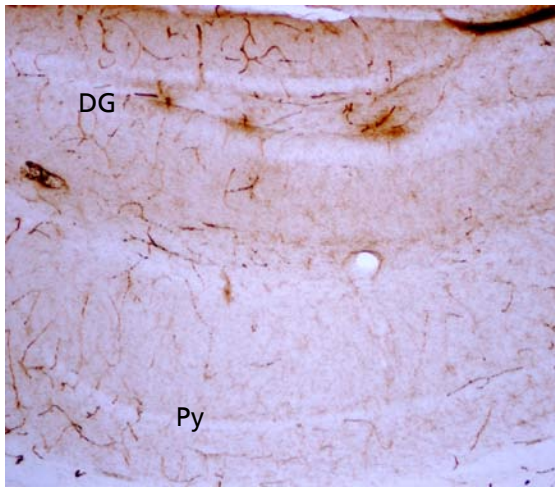

Fig 2b: Lack of specific immunohistochemical staining with McAB 300 in the hippocampus of a Calbindin D-28k knock-out mouse (6). Non-specific staining of the vessel walls due to the anti-mouse secondary antibody. DG: dentate gyrus, Py: pyramidal cells X 100

Working dilution

Immunohistochemistry: 1:5'000 - 1:10'000 with the avidin-biotin method.
Immunoblots: 1:100 - 1:1'000 with an indirect peroxidase method.
We recommend that the optimal dilutions be determined by titration experiments.

Storage

For long storage, keep at - 80°C (or at least - 20°C). For continuous use, keep at 4°C (with 0.01% Na-azide). Avoid repeated freezing and thawing.

References

1. Celio M.R. et al., Cell Calcium 11:599-602, 1990
2. Kretsinger R.H. (1981) Neurosci. Res. Progr. Bull. 19/8, MIT-Press
3. Garcia-Segura L.M. et al. (1984) Brain Rews. 296: 75-86.
4. Airaksinen M.S., et al, (1997), PNAS 94(4) : 1488-1493