abcam

Product datasheet

Anti-Calbindin antibody ab25085

★★★★★ 6 Abreviews 23 References 5 Images

Overview

Product name Anti-Calbindin antibody

Description Rabbit polyclonal to Calbindin

Host species Rabbit

Tested applications Suitable for: IP, WB, IHC-P, IHC-Fr, ICC/IF

Species reactivity Reacts with: Mouse, Human

Predicted to work with: Rat, Rabbit, Horse, Chicken, Hamster, Cow

Immunogen Synthetic peptide within Human Calbindin aa 150-250. The exact sequence is proprietary.

Database link: P05937

Positive control Recombinant Human Calbindin protein (<u>ab117207</u>) can be used as a positive control in WB. Rat

brain

Properties

Form Liquid

Storage instructions Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

Storage buffer pH: 7.6

Preservative: 0.1% Sodium azide Constituents: PBS, 1% BSA

Purity Immunogen affinity purified

Clonality Polyclonal

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab25085 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

IP Use a concentration of 5 μg/ml.	Application	Abreviews	Notes
	IP		Use a concentration of 5 µg/ml.

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Application	Abreviews	Notes
WB	★★★★ ★ (2)	Use a concentration of 1 - 2 μg/ml. Predicted molecular weight: 28 kDa.
IHC-P	**** <u>(2)</u>	1/400. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
IHC-Fr	★★★★ <u>(1)</u>	Use at an assay dependent concentration.
ICC/IF	★★★★ <u>(1)</u>	Use a concentration of 1 - 5 μg/ml.

Target

Function Buffers cytosolic calcium. May stimulate a membrane Ca(2+)-ATPase and a 3',5'-cyclic

nucleotide phosphodiesterase.

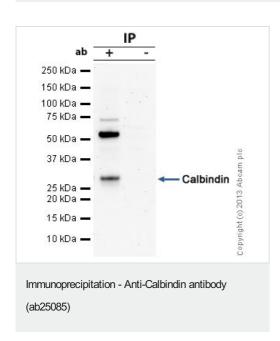
Sequence similarities Belongs to the calbindin family.

Contains 5 EF-hand domains.

DomainThis protein has four functional calcium-binding sites; potential sites II and VI have lost affinity for

calcium.

Images



Calbindin was immunoprecipitated using 0.5mg Rat Brain tissue extract, 5µg of Rabbit polyclonal to Calbindin and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).

The antibody was incubated under agitation with Protein G beads for 10min, Rat Brain tissue extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of $40\mu l$ SDS loading buffer and incubated for 10min at $70^{\circ}C$; $10\mu l$ of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab25085.

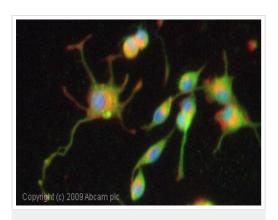
Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697).<

Band: 28kDa; Calbindin



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Calbindin antibody (ab25085)

ab25085 staining human Calbindin D28 in Human cerebellum by immunohistochemistry formalin fixed, paraffin embedded.



Immunocytochemistry/ Immunofluorescence - Anti-Calbindin antibody (ab25085)

ICC/IF image of ab25085 stained PC12 cells. The cells were 100% methanol fixed (5 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab25085, 1ug/ml) overnight at +4øC. The secondary antibody (green)ÿwas Alexa Fluor© 488 goat anti-rabbit lgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor© 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43uM.



Western blot - Anti-Calbindin antibody (ab25085)

Anti-Calbindin antibody (ab25085) at 1 μ g/ml + Brain (Rat) Tissue Lysate at 10 μ g

Secondary

Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

Developed using the ECL technique.

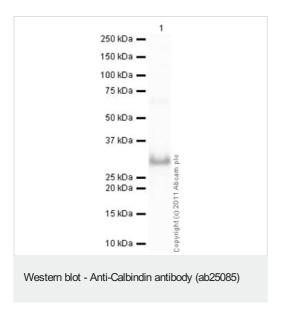
Performed under reducing conditions.

Predicted band size: 28 kDa Observed band size: 30 kDa

Additional bands at: 67 kDa. We are unsure as to the identity of

these extra bands.

Exposure time: 4 minutes



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