

Product Specification Sheet

**CABP28K (Calbindin 1/CALB1/Spot35 protein/Cholecalciferin) Antibodies**

Cat. # D28K11-A	Rabbit Anti-Mouse D28K IgG # 1 (aff pure)	<b>SIZE:</b> 100 ug
Cat. # D28K11-P	Mouse D28K Control/blocking peptide # 1	<b>SIZE:</b> 100 ug

Calcium (Ca<sup>2+</sup> or Ca) is the most abundant cation and it is required for many physiological activities such as bone formation and it acts as a second messenger in signal transduction. However only 1% of Ca is present in ionic form in biological fluids. Ca concentration is regulated by calcitropic hormones that act on bone, kidney, and intestine. Extracellular Ca<sup>2+</sup>-levels are sensed and regulated by Calcium Sensing receptor (CASR). When Ca levels are limiting then it must be taken up by active, transcellular pathways comprising (1) Ca<sup>2+</sup> entry across apical membrane, (2) cytosolic transport of Ca<sup>2+</sup> across the cell from apical to basolateral membrane facilitated by a family of low mol wt Calcium binding proteins (CABPs) that include vitamin D3-dependent Ca<sup>2+</sup> binding proteins (calbindin-D9k, Calbindin-28k, Calretinin, Parvalbumin, S100, calmodulin) and finally (3) an active extrusion of Ca<sup>2+</sup> through basolateral membrane mediated by Ca<sup>2+</sup>-ATPase and Na<sup>+</sup>-Ca<sup>2+</sup> exchangers (NCX).

Calbindins are Ca-binding proteins belonging to the troponin C superfamily. There are two types of CaBPs: the "trigger"- and the "buffer"-CaBPs. The conformation of "trigger" type CaBPs changes upon Ca<sup>2+</sup> binding and exposes regions on protein that interact with target molecules, thus altering their activity. The buffer-type CABP are thought to control the intracellular calcium concentration. **CALB28K/Calbindin 1/CALB1 (D28K/Spot35 protein or cholecalciferin**, rat 261 aa; mouse 261 aa; human 261-aa, chromosome 8q21.3-q22.1) was originally described as 27-kDa induced by vitamin D in the duodenum of chicken. In mammals, it is expressed in the kidney, pancreatic islets, and brain. In brain, its synthesis is independent of vitamin D. CABP28K contains 4 active and 2 inactive EF-hand Ca-binding domains. The gene for CABP28K is clustered in the same region as carbonic anhydrase. The neurons in the brains of patients with Huntington disease are CAB28K depleted. Calbindin-D-28K is found predominantly in subpopulations of central and peripheral nervous system neurons, and in certain epithelial cells involved in Ca<sup>2+</sup> transport such as distal tubular cells and cortical collecting tubules of the kidney, and in enteric neuroendocrine cells.

**Source of Antigen and Antibodies**

<b>Antigen</b>	A 20-aa peptide sequence ( <b>designated D28K11-P or control peptide</b> ), within the C-terminal, non-calcium binding domain of mouse D28K (1) conjugated to KLH. Epitope location ~ C-terminal, non-calcium binding domain
<b>Ab Host/type</b>	Rabbit, Polyclonal IgG, purified over antigen-agarose (Cat # D28K11-A)
<b>2-Ab</b>	<b>Cat # 20320, goat anti-rabbit IgG-HRP (AP, biotin, FITC conjugates also available</b>
<b>-ve control</b>	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as -ve control

**Form & Storage of Antibodies/Peptide Control**

100ul solution lyophilized powder  
Supplied in Buffer: 0.05% azide  
**Reconstitute** powder in 100 ul PBS

**Affinity pure IgG**

100 ug/100ul solution lyophilized powder  
Supplied in **Buffer:** PBS+0.1% BSA  
**Reconstitute powder** in PBS at 1mg/ml

**Control/blocking peptide**

100 ug/100 ul solution lyophilized powder  
Supplied in Buffer: PBS pH 7.5,  
**Reconstitute powder in PBS at 1 mg/ml.**

**Storage**

**Short-term:** unopened, undiluted liquid vials at 20°C and powder at 4°C or -20°C..

**Long-term:** at -20°C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

**Stability:** 6-12 months at -20°C or below.

**Shipping:** 4°C for solutions and room temp for powder

**Recommended Usage**

**Western Blotting** (1-10 ug/ml for affinity pure using Chemiluminescence technique). D28K is ~28 kDa.

**ELISA** (1:10K-1:100K; using 50-100 ng of control peptide/well).

**Histochemistry & Immunofluorescence:** not tested. We recommend the use of affinity pure antibody at 2-20 ug/ml.

**Specificity & Cross-reactivity**

The D28K-P peptide is 95% conserved in rat, human, 90% in rabbit, 85% in chicken and bovine D28K. No significant sequence homology of D28K-P is seen with D9K, calretinin or other CABPs. Antibody reactivity in various species is not known. Control peptide, because of its low mol. Wt (<3 kDa), is not suitable for Western. It should be used for ELISA or antibody blocking experiments (use 5-10 ug control peptide per 1 ug of aff pure IgG or 1 ul antiserum) to confirm antibody specificity (see detailed protocol at:the web site).

**General References:** (1) Nordquist DT et al (1988) J. Neurochem. 8, 4780; Takeda T et al (1994) BBRC 204, 889; Wood TL et al (1988) DNA 7, 585; Parmentier M et al (1987) Eur. J. Biochem. 170, 207; Gross MD et al (1988) JBC 263, 14426; Lomri N et al (1989) Gene 80, 87-98

*\*This product is for In vitro research use only.*

**Related materials available from ADI**

Antibodies: CaT-1/2; Calbindins, S100, Parvalbumin, Calretinin

D28K11-A-P 71216S

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