

# Product Datasheet

## Calponin 1 Antibody (EP798Y)

### NB110-55650

Unit Size: 0.1 ml

Store at 4C. Do not freeze.

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**NB110-55650**

Calponin 1 Antibody (EP798Y)

Product Information	
Unit Size	0.1 ml
Concentration	Please see the vial label for concentration. If unlisted please contact technical services.
Storage	Store at 4C. Do not freeze.
Clonality	Monoclonal
Clone	EP798Y
Preservative	No Preservative
Isotype	IgG
Purity	Protein A or G purified
Buffer	Culture supernatant in buffered aqueous solution
Product Description	
Host	Rabbit
Gene ID	1264
Gene Symbol	CNN1
Species	Human
Immunogen	A synthetic peptide corresponding to residues near the C-term of human Calponin was used as an immunogen.
Product Application Details	
Applications	Western Blot, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin
Recommended Dilutions	Western Blot 1:1000-10000, Immunohistochemistry 1:10-1:500, Immunocytochemistry/Immunofluorescence 1:100-250, Immunohistochemistry-Paraffin 1:50-1:200, Immunohistochemistry-Frozen 1:50-1:200
Application Notes	This is a high affinity, high titer antibody. Optimal dilutions should be determined by the end user. (Provided dilutions serve only as a suggestive starting point.)



## Publications

Galli D, Carubbi C, Masselli E et al. PKC-epsilon is a negative regulator of PVAT-derived vessel formation. *Exp. Cell Res.* 2015 Jan 15 [PMID: 25433270] (WB, Mouse)

### Details:

rabbit anti-CALPONIN/Calponin 1 used at 1:1000 dilution for WB on protein extract from cardiomyocyte negative fractions (CMnegF) of control animals/C, of animals injected with mutant PKC epsilon K522M /rPKCEm and of animals injected with PKC-Epsilon over-expressing vectors/rPKCE (Fig. 5D)

Papke CL, Cao J, Kwartler CS et al. Smooth muscle hyperplasia due to loss of smooth muscle alpha-actin is driven by activation of focal adhesion kinase, altered p53 localization and increased levels of platelet-derived growth factor receptor-beta. *Hum Mol Genet* 2013 Apr 24 [PMID: 23591991] (WB, Mouse)

Guo X, Oshima H, Kitmura T et al. Stromal fibroblasts activated by tumor cells promote angiogenesis in mouse gastric cancer. *J Biol Chem.* 2008 Jul. [PMID: 18495668]

Jeyasuria P, Wetzel J, Bradley M et al. Progesterone-Regulated Caspase 3 Action in the Mouse May Play a Role in Uterine Quiescence During Pregnancy Through Fragmentation of Uterine Myocyte Contractile Proteins. *Biol Reprod*;80(5):928-934. 2009 [PMID: 19144964]

Elia A, Charalambous F, Georgiades P. New phenotypic aspects of the decidual spiral artery wall during early post-implantation mouse pregnancy. *Biochemical and Biophysical Research Communications.* 2011 Nov 12. [PMID: 22100651]



## Procedures

### Immunohistochemistry Protocol for Calponin Antibody (NB110-55650)

Immunohistochemistry Protocol for Calponin Antibody (NB110-55650):

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Immunohistochemistry Protocol for Paraffin-embedded Tissues:

#### 1. Solutions and reagents

##### 1.1. Xylene

##### 1.2. Ethanol, anhydrous denatured, histological grade (100%, 95%, 70%)

##### 1.3. Washing buffer:

TBST washing buffer: 1XTBS/0.1% Tween-20

To prepare stock solution of 10X TBS: add 24.2 g Trizma base and 80 g sodium chloride to 1L of dH<sub>2</sub>O. Adjust pH to 7.6.

Working solution. 1XTBST/0.1% Tween-20: add 100ml 10XTBS to 900 ml dH<sub>2</sub>O. Add 1 ml Tween-20 and mix well.

##### 1.4. Distilled water (dH<sub>2</sub>O)

##### 1.5. Antigen Retrieval Solution:

0.01M Sodium Citrate Buffer, pH 6.0

To prepare stock solutions:

Solution A. 0.1 M citric acid solution: dissolve 21.0 g of citric acid, monohydrate (C<sub>6</sub>H<sub>8</sub>O<sub>7</sub>·H<sub>2</sub>O) in 1 liter of dH<sub>2</sub>O.

Solution B. 0.1M sodium citrate solution: dissolve 29.4 g trisodium citrate dihydrate (C<sub>6</sub>H<sub>5</sub>Na<sub>3</sub>O<sub>7</sub>·2H<sub>2</sub>O) in 1 liter of dH<sub>2</sub>O.

Working solution: Add 9 ml of Stock solution A and 41 ml of stock solution B to 450 ml of dH<sub>2</sub>O. Adjust pH to 6.0.

##### 1.6. 3% Hydrogene Peroxide

##### 1.7. Blocking buffer:

PBS (Dulbeccos Phosphate Buffered Salts, 1X, catalog #21-031-CV from Mediatech, Inc.) + 10% serum (serum origin depends on the host of the secondary antibody)

##### 1.8. Hematoxylin QS (catalog #H-3404 from Vector Laboratories, Inc.)

##### 1.9. Permanent Mounting medium (VectaMount, catalog# H-5000 Vector Laboratories, Inc.)

#### 2. Protocol

##### 2.1. Deparaffinization/Rehydration

###### 2.1.1. Heat slides in an oven at 65C for 1 hour.

2.1.2. De-paraffinize/hydrate using the following series of washes: two Xylene washes (5 min each), followed by two 100% ethanol rinses (5 min each), followed by 95% ethanol, 70% ethanol, 50% ethanol, 30% ethanol, followed by H<sub>2</sub>O and a TBST wash for 5 min on a shaker.

##### 2.2. Antigen Retrieval

2.2.1. Immerse slides into staining dish containing Antigen Retrieval Solution.

2.2.2. Place covered staining dish into the rice cooker. Add 120 ml of dH<sub>2</sub>O.

2.2.3. When cook is turned to warm (about 20 to 30 min), unplug the cooker and remove the staining dish to the bench top.

2.2.4. Allow to cool down, without cover, for 20 min.

### 2.3. Staining

2.3.1. Wash slides with TBST for 5 min on a shaker.

2.3.2. Inactivate endogenous peroxidase by covering tissue with 3% hydrogen peroxide for 10 min.

2.3.3. Wash slides three times with TBST (3 min each on a shaker).

2.3.4. Block slides with the blocking solution for 1 hour.

2.3.5. Dilute primary antibody in the blocking buffer per recommendation on the data sheet.

2.3.6. Apply primary antibody to each section and incubate overnight in the humidified chamber (4C).

2.3.7. Wash slides three times with TBST (3 min each on a shaker).

2.3.8. Apply to each section secondary HRP-conjugated anti-rabbit antibody diluted in the blocking solution per manufacturers recommendation; incubate for 1 hour at room temperature.

2.3.9. Wash slides three times with TBST (3 min each on a shaker).

2.3.10. Add freshly prepared DAB substrate to the sections.

2.3.11. Incubate tissue sections with the substrate at room temperature until suitable staining develops (generally 2 to 5 min).

2.3.12. Rinse sections with water.

2.3.13. Counterstain with Hematoxylin.

2.3.14. Rinse sections with water.

2.3.15. Dehydrate samples using two rinses with 100% Ethanol (20 dips per rinse) followed by two rinses with Xylene (30 dips per rinse).

2.3.16. Mount coverslips on slides using Permount medium.





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### **Limitations**

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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