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# **Product Information**

Anti-m-Calpain (Domain III), Large Subunit produced in rabbit, affinity isolated antibody

Catalog Number C0728

## **Product Description**

Anti-m-Calpain (Domain III), Large Subunit is produced in rabbit using as immunogen a synthetic peptide corresponding to the N-terminal of domain-III in the large subunit of rat m-calpain (E.C. 3.4.22.17, calpain-II, protein kinase-C activating factor). The antibody is affinity purified using peptide agarose.

Anti-m-Calpain (Domain III), Large Subunit specifically binds to latent and active m-Calpain and does not cross-react with other calpain family members, e.g.,  $\mu$ -calpain, calpain-94, ncl-2, ncl-3, etc. The antibody reacts with bands at 80 kDa and 58 kDa, and a series of further cleaved active forms, by immunoblotting against the reduced protein. It also reacts with non-reduced m-calpain. Species reactivity includes rat, mouse and human. It also reacts with non-reduced m-calpain. The antibody may be used for immuno-precipitation, immunohistochemistry and ELISA.

The calpains are calcium-activated non-lysosomal thiolproteases. m-Calpain, also termed calpain-II, is an intracellular, Ca<sup>2+</sup>-dependent cysteine protease. m-Calpain has a millimolar sensitivity as compared to the micromolar calcium sensitivity of u-calpain (calpain-I). The calpains have papain-like activity, thus the -pain nomenclature. Both m- and μ-calpain are ubiquitously expressed and are countered by the endogenous calpain inhibitor calpastatin. The latent large subunit of m-calpain is 80 kDa and a N-terminal truncation at activation yields an approximately 58 kDa form. Other calpain family members (calpain-94, ncl-2, ncl-3, etc) have more limited tissue distribution, and perhaps different functions. The calpain family members are heterodimers and consist of a common small subunit (regulatory) of 30 kDa, and a large variable subunit (catalytic) of 80 kDa. Domains in the large subunit include the N-terminal domain-I, the proteinase domain-II, domain-II and the EF-hand (Ca2+-binding) domain-IV.

Calpains are present in all mammalian tissues and are involved in a variety of processes including cell proliferation,<sup>2,3</sup> differentiation,<sup>4-6</sup> vesicle secretion and others.<sup>7,8</sup>

### Reagent

Supplied in 0.01 M phosphate buffered saline, pH 7.4, containing 50% glycerol, 1% BSA and 0.02% sodium azide as preservative.

Antibody concentration: ~1 mg/ml.

#### **Precautions and Disclaimer**

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

#### Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage, the solution may be stored at -20 °C. The antibody is supplied with 50% glycerol to prevent freezing. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

### **Product Profile**

Immunoblotting: a working dilution of 1:1,000 is determined using an alkaline phosphatase conjugated secondary antibody and BCIP/NBT as substrate. Higher antibody concentrations may be necessary for non-human samples.

**Notes**: Most cell types produce m-calpain. Since it is a cellular protein, cell lysates work well for immunoblotting.

In order to obtain best results and assay sensitivity in different techniques and preparations we recommend determining optimum working dilutions by titration assay.

#### References

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