

PRODUCT DATA SHEET

Revised: 12 November 2004, v.01
Printed: 28 January 2009; page 1 of 1



Product name(s):	Mouse monoclonal antibody to Calpain I, II (subunit p30)				
Catalogue number:	CG 1920	Batch number:	Ztemp	Expiry date:	12 months from receipt

Introduction:

The calpains constitute a superfamily of calcium-dependent cysteine proteases that are widely expressed in animal phyla. Both m-calpain (calpain II) and μ -calpain (calpain I) are composed of two subunits (80kDa and 30kDa). Whereas the small subunit is common to both enzymes, the larger catalytic subunits are encoded by different genes (*CAPN1* for μ -calpain and *CAPN2* for m-calpain) and exhibit distinct calcium ion requirements. m-Calpain requires millimolar levels of Ca^{2+} , and μ -calpain is active at micromolar concentrations of Ca^{2+} .

To date, 14 human calpain genes have been identified, and these are commonly divided into *typical* calpains and *atypical* calpains. Typical calpains are composed of 4 domains, whereas atypical calpains do not contain domain IV and are unable to interact with the p30 subunit.

Recent research has implicated calpain involvement in many age-related disease states, including neuronal degeneration, type 2 diabetes, cataract formation and tumour cell metastasis. This work is excellently reviewed elsewhere¹⁻³.

Product information:

Balb/c mice were immunised with calpain purified from human placenta. Splenocytes from mice giving a strong anti-calpain response were fused and the resulting hybridomas were tested and selected on the basis of solid-phase assays and Western blotting. The antibody (isotype IgG₁) has been purified by protein A chromatography, and contains 3mM w/v sodium azide as a preservative.

Clone 28F3 specifically recognises the 28-30kDa subunit common to both μ - and m-calpains. The antibody is known to react with human and bovine, but not rat or mouse calpains.

The calpain p30 antibody reacts strongly with a single major band (~28-30kDa) on denaturing Western blots of lysates prepared from MDBK (bovine kidney) and human fibroblast cell lines at dilutions up to 1:2,000* when used in combination with chemiluminescence procedures.

The antibody is also suitable for immunoprecipitation under native and denaturing conditions (2-5 μ L *per test*).

NOTES: *Optimal dilutions need to be determined by experimentation, but an initial dilution of 1:1000 is recommended when using the antibody in Western blotting (overnight incubation at 4°C) on test tissues in concert with a sensitive detection procedure such as enhanced chemiluminescence (ECL; Amersham).

Vial contents, Storage and Use:

See label for vial contents. Store unopened vial at -20°C until required for use. AVOID FREEZE-THAW CYCLES. Aliquot undiluted antibody into smaller volumes (not less than 10 μ L) prior to freezing if appropriate. The use of high quality 'antiserum-grade' plastic or glass vials is recommended. Store diluted antibody at 2-4°C (do not freeze) and use within 1 month.

Dilute to working strength with PBS (pH 7.2) containing 1.5% sodium chloride and 1% normal goat serum (if a goat anti-mouse IgG linker antibody is to be used).

References:

1. Carragher, N.O. and Frame, M.C. Calpain: A role in cell transformation and migration. *Int. J. Biochem. Cell Biol.*, **34**: 1539-1543, 2002.
2. Nixon, R.A. The calpains in aging and aging-related diseases. *Ageing Res. Rev.*, **2**: 407-418, 2003.
3. Suzuki, K. *et al.* Structure, activation, and biology of calpain. *Diabetes*, **53**: S12-S18, 2004.

A vial containing 50 μ g (at 1mg/mL) MDBK, PC12 or NIH3T3 cell lysate is supplied free-of-charge with this product. The protein is suspended in Laemmli-type sample buffer and the vial contents should be boiled for 5min prior to loading on to a SDS-PAGE gel. Approximately 10 μ L should be used *per lane*. Store the positive control lysate at -20°C until required for use. Do not aliquot. Additional vials are NOT available for purchase.