

**Product No. F-1043**

**Lot 036H4828**

**Monoclonal Anti-Filensin**

Mouse Ascites Fluid

Clone FIL-7B10

Monoclonal Anti-Filensin (mouse IgG1 isotype) is derived from the FIL-7B10 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from immunized BALB/c mice. Human and bovine lens filament enriched fraction (plasma membrane-cytoskeleton complex) was used as the immunogen.<sup>1</sup> The isotype is determined using Sigma ImmunoType™ Kit (Sigma ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Sigma ISO-2). The product is provided as ascites fluid with 0.1% sodium azide (see MSDS)\* as a preservative.

**Specificity**

Monoclonal Anti-Filensin recognizes an epitope located within either the rod or the N-terminal non-( $\alpha$ )-helical domain of the filensin molecule.<sup>1</sup> The product specifically detects both the full length filensin (115 kD) and the processed 53 kD fragment of filensin applying immunoblotting.<sup>1</sup> It also reacts with a variety of breakdown products of filensin in the M.W. range of 28-69 kD. In immunofluorescence confocal microscopy of paraformaldehyde-fixed bovine lens tissue, the antibody shows mostly membrane staining in the peripheral fiber cells, but a greater cytoplasmic distribution in the corneal fiber cells.<sup>1</sup> The antibody does not react with cultured lens epithelial cells, nor with normal human skin, psoriatic skin or tumor skin (basal cell carcinoma). Cross-reactivity is observed with human, bovine and sheep, but not with chicken, rat and mouse.

**Description**

The ocular lens is ectodermally derived and consists of two cell types. These are the epithelial cells, which underlie only the anterior portion of the collagenous lens capsule, and the highly elongated fiber cells which comprise the bulk of the lens.<sup>1,2</sup> Fiber cells arise continuously from the undifferentiated lens epithelial cells, rapidly during embryonic and early post-natal life, and more slowly during adult life. As fiber cells

differentiate, they are added to the surface of the existing fiber cell mass. Lens fiber cells have two discrete and independent cytoskeletal networks, composed of different intermediate filament proteins. Both networks undergo significant remodelling with progressive maturation of the fiber cells.<sup>2</sup> Intermediate filament proteins serve multiple functions in cells, with at least some of these functions likely to be tissue-specific. In addition to intermediate filaments and microfilaments, a cytoskeletal structure called the beaded filament has also been reported in the fiber cells of the lens.<sup>3</sup> Biochemical and immunocytochemical studies have implicated two fiber cell-specific proteins as the principal components of this beaded filament. These two proteins, referred to as filensin (formerly CP95 or CP115, M.W. 115 kD) and CP49 (also phakinin and phakosin) appear to be unique to the differentiated fiber cell. Thus, filensin/CP 49 are found at all stages of lens fiber cell differentiation, from the outer cortical fiber cells at the center of the lens, in contrast to vimentin which is only abundant in the outer cortical fiber cells. Filensin and CP49 have been shown to be members of the intermediate filament protein family on the basis of primary sequence data.<sup>4,6</sup> However, both have not only unusual primary sequence features such as the absence of a tail (CP49) and a foreshortened central rod domain (filensin), but also show divergence in those highly conserved motifs which flank the central  $\alpha$ -helical domain of all intermediate filament proteins. Filensin is extensively processed during lens fiber cell differentiation to give protein fragments derived from distinct protein domains, one corresponding to the N-terminal non- $\alpha$ -helical/and rod domain and the other to the C-terminal non- $\alpha$ -helical tail domain.<sup>1</sup> The extensive C-terminal domain of filensin forms a range of stable fragments during differentiation from 51 to 62 kD, and this domain is believed to be important for binding to lens plasma membranes as well as to vimentin.<sup>4</sup> The N-terminal portion of filensin, which includes the  $\alpha$ -helical rod domain, is also present as a stable product in the lens fiber cells, known as the 53

kD polypeptide (or fragment).<sup>5</sup> Since filensin is specific to lens fiber cells, it may serve as an excellent marker for assessing the differentiation and maturation of these cells. The availability of an antibody which reacts specifically with filensin,<sup>1,2,7</sup> enables the detection and localization of this protein, by using various immunochemical techniques, including immunoblotting and confocal microscopy.

### Uses

Monoclonal Anti-Filensin may be used for the localization of filensin using various immunochemical assays including immunoblotting and immunohistochemistry.

**Titer:** 1:400

The antibody titer was determined by indirect immunofluorescent labeling of bovine lens frozen sections.

In order to obtain best results, it is recommended that each user determine the optimal working dilution for individual applications by titration assay.

### Storage

For continuous use, store at 2-8°C. For extended storage freeze in working aliquots. Repeated freezing and thawing is not recommended. Storage in "frost-free" freezers is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

\* Due to the sodium azide content a material safety sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazardous and safe handling practices.

### References

1. Sandilands, A., et al., *Eur. J. Cell Biol.*, **67**, 238 (1995).
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3. Georgatos, S., et al., *Bio Essays*, **16**, 413 (1994).
4. Merdes, A., et al., *J. Cell Biol.*, **115**, 397 (1991).
5. Quinlan, R., et al., *Curr. Eye Res.*, **11**, 909 (1992).
6. Remington, S., *J. Cell Sci.*, **105**, 1057 (1993).
7. Fitzgerald, P., *Curr. Eye Res.*, **7**, 1255 (1988).