

## Product Information

### Monoclonal Anti-Myosin (Skeletal, Fast)-Alkaline Phosphatase antibody produced in mouse clone MY-32, purified from hybridoma cell culture

Catalog Number **A4335**

#### Product Description

Monoclonal Anti-Myosin (Skeletal, Fast) (mouse IgG1 isotype) is derived from the hybridoma produced by the fusion of mouse myeloma cells and splenocytes from an immunized mouse. Rabbit muscle myosin was used as the immunogen. The isotype is determined by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents, Catalog Number ISO2. Protein A purified Anti-Skeletal Myosin is conjugated to Alkaline Phosphatase using 0.2% glutaraldehyde.

Monoclonal Anti-Myosin (Skeletal, Fast) specifically recognizes an epitope in adult rat skeletal myosin heavy chains IIa, IIb and IIc (IIx) (in fast-twitch fibers).<sup>1</sup> It also reacts with neonatal but not with embryonal cardiac myosin heavy chains.<sup>1</sup> It does not cross react with non-muscle, and smooth muscle myosins. A similar reactivity pattern is observed in human, mouse and chicken skeletal muscle preparations.<sup>2,3,4,5,6,7</sup> Cross-reactivity has been observed with human, bovine, cat, rabbit, rat, mouse, guinea pig and chicken. The epitope recognized is stable to routine formalin-fixation and paraffin-embedding. The conjugated product is applicable on frozen sections and formalin-fixed sections using immunoenzymatic techniques.

Monoclonal Anti-Myosin (Skeletal, Fast)-Alkaline Phosphatase may be used for the detection and localization of skeletal muscle fast and neonatal myosins using immunohistochemistry. It may be used in muscle fiber typing, in studies of *in vivo* and *in vitro* muscle development and in the diagnosis of rhabdomyosarcomas.

Myosin is a 480 kDa protein that interacts with actin in muscle and non muscle cells.<sup>8</sup> Conventional myosin contains two identical heavy chains (MHC - 200 kDa each) and four light chains (MLC-15-26 kDa).

Conventional myosin molecules consist of two major regions: tail (rod) and globular heads; they aggregate into filaments through the tail region and interact with actin and with adenosine triphosphate (ATP) through the head region. Multiple forms of myosin heavy chains exist in skeletal, smooth and cardiac muscle

and in non-muscle tissues. A spectrum of skeletal muscle fiber types is found in adult skeletal muscles.

The phenotypic properties of these fiber types relate to the differential expression of myofibrillar proteins isoforms. The two major skeletal muscle fiber types are type I (slow-twitch) and type II (fast-twitch). Skeletal muscle type II fibers can be further subdivided into types IIa (fast red) and IIb (fast white). The different adult fiber types express three different isomyosin types. Fibers containing a mixture of slow myosins in variable amounts (e.g., human IIc) are found in certain muscles. In several mammalian species a IIc (IIx) myosin heavy chain isoform was demonstrated. In addition a superfast isoform was described in the jaw-closing muscles of most carnivores and of primates (except man).<sup>9</sup> The different isoforms are demonstrable by electrophoretic separation, analysis of peptide fragments, immunoblotting and immunohistochemical techniques.

The proportions of myosin isoforms present in different fiber types are largely determined by the activity pattern of the muscle, a result of the stimuli received from the nerve supply. Changes in thyroid state, denervation and chronic stimulation can induce changes in myosin heavy chain isoforms expression. Differentiation of muscle fiber types involves sequential replacement of developmental isoforms of myosin heavy chains such as embryonic and neonatal isoforms by specific adult isoforms. Fiber type analysis is a valuable procedure in the study of normal and diseased skeletal muscle. Discrimination of fiber types is routinely achieved by application of enzyme histochemical techniques on frozen sections. Anti-Myosin (Skeletal, Fast) is useful not only in fiber typing but also in detection of myogenic tumors.<sup>10</sup> It enables retrospective as well as prospective fiber typing using formalin-fixed, paraffin-embedded material.

#### Reagent

The conjugate is provided as a liquid in 0.05 M Tris buffer, pH 8.0, containing 1% BSA, 1 mM MgCl<sub>2</sub>, 50% glycerol and 0.1% sodium azide.

**Precautions**

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**

Store at 2-8 °C. Do not freeze.

**Product Profile**

Direct immunofluorescence: a minimum working dilution of 1:150 was determined using formalin-fixed, paraffin-embedded sections of human or animal skeletal muscle.

**Note:** In order to obtain best results, it is recommended that each individual user determine their optimal working dilution by titration assay.

**References**

1. Harris, A., et al., *Development*, **107**, 751 (1989).
2. Naumann, K., and Pette, D., *Differentiation*, **55**, 203 (1994).
3. Havenith, M., et al., *Histochemistry*, **93**, 497 (1990).
4. Schubert, W., *Euro. J. Cell Biol.*, **55**, 272 (1991).
5. Donoghue, M., et al., *J. Cell Biol.*, **115**, 423 (1991).
6. Maier, A., *Histochemistry*, **99**, 333 (1993).
7. Honda, H., and Rostami, A., *Proc. Nat. Acad. Sci. USA*, **86**, 7007 (1989).
8. Cooke, R., In: *Guidebook to the Cytoskeletal and Motor Proteins* Kreis, T., and Vale, R. (Eds.), Oxford University Press, p 207 (1993).
9. Rowlerson, A., et al., *J. Musc. Res. Cell Motility*, **2**, 415 (1981).
10. Carter, R., et al., *Histopathology*, **17**, 301 (1990).

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