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# **ProductInformation**

MONOCLONAL ANTI-b-ACTIN Clone AC-74 Mouse Ascites Fluid

Product Number A 5316

## **Product Description**

Monoclonal Anti-β-Actin (mouse IgG2a isotype) is derived from the AC-74 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from an immunized mouse. A slightly modified synthetic β-cytoplasmic actin N-terminal peptide, Ac-Asp-Asp-Asp-Ile-Ala-Ala-Leu-Val-<u>Ile</u>-Asp-Asn-Gly-Ser-Gly-<u>Lys</u>, conjugated to KLH¹ was used as the immunogen. The isotype is determined using Sigma ImmunoType<sup>™</sup> Kit (Product Code ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Product Code ISO-2).

Monoclonal Anti-β-Actin recognizes an epitope located on the N-terminal end of the  $\beta$ -isoform of actin. <sup>1</sup> The antibody specifically labels β-actin in a wide variety of tissues and species using immunoblotting (42 kDa), immunofluorescent staining of cultured cell lines and immunohistochemistry. In immunofluorescent staining of chicken gizzard ultra-thin tissue cryosections, the antibody labels the dense bodies and the longitudinal channels linking consecutive dense bodies that are also occupied by desmin, and the membrane-associated dense plaque. 1 It does not stain adult cardiac and skeletal muscles (except traces due to contaminations of the sample with non-muscle cells). The antibody cross reacts with β-actin expressing cells in human, bovine, sheep, pig, rabbit, cat, dog, mouse, rat, guinea pig, chicken, carp, leech, and fruit fly tissues, but not in amoeba. It can be used for staining of acetone-fixed, frozen sections and EM preparations. The epitope recognized by the antibody is resistant to formalinfixation and paraffin-embedding. Ethanol, methacarn, or Bouin's solutions may also be used as fixatives.

The two major cytoskeletal proteins implicated in cell motility are actin and myosin. Actin and myosin are constituents of many cell types and are involved in a myriad of cellular processes including locomotion, secretion, cytoplasmic streaming, phagocytosis, and

cytokinesis. Although actin is one of the most conserved eukaryotic proteins, it is expressed in mammals and birds as at least six isoforms characterized by electrophoresis and amino acid sequence analysis.<sup>2,3</sup> Four isoforms represent the differentiation markers of muscle tissues and two are found practically in all cells. There are three  $\alpha$ -actins ( $\alpha$ -skeletal,  $\alpha$ -cardiac and  $\alpha$ -smooth muscle), one β-actin (β-non-muscle) and two  $\gamma$ -actins ( $\gamma$ -smooth muscle and  $\gamma$ -non-muscle). Actin isoforms show >90% overall sequence homology, but only 50-60% homology in their 18 NH<sub>2</sub>-terminal residues.<sup>4</sup> The NH<sub>2</sub>-terminal region of actin appears to be a major antigenic region, and may be involved in the interaction of actin with other proteins such as myosin. The actin in cells of various species and tissue origin are very similar in their immunological and physical properties. As a consequence, it has been difficult to produce potent antisera to this protein. The availability of monoclonal antibody to β-actin provides a specific and useful tool in studying the intracellular distribution of  $\beta$ -actin and the static and dynamic aspects of the cytoskeleton.

# Reagents

The product is provided as ascites fluid containing 0.1% sodium azide as a preservative.

#### **Precautions and Disclaimer**

Due to the sodium azide content a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS 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, solution may be frozen 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 by centrifugation before use.

#### **Product Profile**

- A minimum dilution of 1:1,000 was determined by indirect immunofluorescent staining of cultured human or chicken fibroblasts.
- 2. A minimum dilution of 1:5,000 was determined by indirect immunoblotting of cultured human or chicken fibroblast cell extract.

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

#### References

- 1. North, A. J., et al., J. Cell Biol., **120**, 1559 (1993).
- 2. Vandekerckhove, J., and Weber, K., Eur. J. Biochem., **90**, 451 (1978).
- 3. Drew, J. S., et al., Am. J. Physiol., **260**, C1332 (1991).
- 4. Lessard, J. L., Cell Motil. Cytoskel. ETON, **10**, 349 (1988).

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