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Product Information

Monoclonal Anti-MYOD1, clone 5.2F
produced in mouse, purified immunoglobulin

Catalog Number **M6190**

Product Description
Monoclonal Anti-MYOD1 (mouse IgG2a isotype) is derived from the hybridoma produced by the fusion of mouse myeloma Sp2/0-Ag14 cells with splenocytes from BALB/c mice immunized with recombinant mouse MYOD1 protein. The antibody is purified by protein A chromatography.

Monoclonal Anti-MYOD1 recognizes human, rat, chicken and mouse phosphoprotein (45 kDa), identified as MYOD1 (Myogenic Differentiation Antigen 1). The epitope of this antibody maps between amino acids 3 and 56 in the N-terminus of mouse MyoD1 protein. It does not crossreact with myogenin, Myf-5, or Myf-6. The antibody labels nuclei of myoblasts in developing muscle tissues. MyoD1 is not detected in normal adult tissues, but is highly expressed in the tumor cell nuclei of rhabdomyosarcomas. Occasionally, nuclear expression of MyoD1 is seen in ectomesenchyoma and a subset of Wilm's tumors. Weak cytoplasmic staining is seen in several non-muscle tissues including glandular epithelium, rhabdomyosarcomas, neuroblastomas, Ewing's sarcomas and alveolar soft part sarcomas. Monoclonal Anti-MyoD1 may be used for the detection of MyoD1 by immunohistochemistry on frozen and formalin–fixed, paraffin embedded tissue sections, immunoprecipitation, Western blot, and immunocytochemistry.

MyoD1 (also known as Rhabdomyosarcoma Marker), a transcriptional activator, is a member of a family of myogenic regulatory proteins called muscle determination factors (MDFs). This unique family of basic helix-loop-helix proteins includes MyoD, Myf-5, myogenin, and MRF-4. These proteins play important roles in skeletal muscle development. Binding sites for these proteins are found in the promoters of a number of genes whose expression is specific to muscle cells. Tight control of the gene expression, however, is dependent on the interaction of various factors

Skeletal myogenesis may be viewed as a two-step process in which Myf-5 and MyoD act early to establish myoblasts. Available evidence suggests that activation of Myf-5 and MyoD determines commitment of the cells to myogenic lineage. Subsequent expression of myogenin triggers terminal muscle cell differentiation and activates muscle-specific genes. MRF4 is also assumed to exert late functions, similar to myogenin.[1]

MyoD1 is not detected in normal adult tissues, but is highly expressed in the tumor cell nuclei of rhabdomyosarcomas. The mouse MyoD1 gene is capable of inducing the myogenic phenotype in embryonic C3H mouse fibroblasts. Experiments on knockout mice have shown that embryos lacking MyoD or Myf-5 alone did not show any impairment in skeletal muscle formation. In contrast, embryos that lacked both MyoD and Myf-5 failed to develop detectable skeletal muscle cells. Mouse embryos lacking myogenin died immediately after birth and exhibited severe skeletal muscle abnormalities.[2,3]

A transcription factor, Pax3, has been identified as one of the key genes in the development of skeletal muscle. Pax3 is expressed in the embryo and is an essential upstream regulator for MyoD expression in precursors of body muscle, but not in head muscles.

Peptide growth factors, such as TGF-β, BMPs, and FGFs, are known to inhibit myogenesis by blocking the expression and transcriptional activity of myogenic factors. TNF plus interferon-γ (IFN-γ) signaling was required for NF-κB-dependent down-regulation of MyoD and dysfunction of skeletal myofibers. MyoD mRNA was also down-regulated by TNF and IFN-γ expression in mouse muscle *in vivo*. These data elucidate a possible mechanism that may underlie the skeletal muscle decay in cachexia.[4,5]

Reagent
Supplied at 1.0 mg/ml in a solution of phosphate buffered saline containing 0.08% sodium azide as a preservative.

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
Store at –20 °C. Upon initial thawing freeze the solution in working aliquots for extended storage. Avoid repeated freezing and thawing to prevent denaturing the antibody. Do not store in a frost-free freezer. The antibody is stable for at least 12 months when stored appropriately. Working dilutions should be discarded if not used within 12 hours.

Product Profile
Immunohistochemistry: a recommended working concentration of 2-4 µg/mL is determined using frozen and formalin-fixed, paraffin- embedded tissues.

Immunoprecipitation: a working concentration of 2 µg/mg of protein lysate is suggested.

Note: In order to obtain best results using different techniques and preparations we recommend determining optimal working concentration by titration.

References

1. Arnold, H.H. and Winter, B., Muscle differentiation: more complexity to the network of myogenic regulators. *Curr. Opin. Gen. Develop*., **8,** 539-544 (1998).
2. Lassar, A. and Munsterberg, A., Wiring diagrams: regulatory circuits and the control of skeletal myogenesis. *Current Opinion Cell Biol*., **6,** 432-442 (1994).
3. Buckingham, M., Skeletal muscle formation in vertebrates. *Curr. Opinion Genet. Develop*. **11**, 440-448 (2001).
4. Kim, Y.–J., et al., The product of an oculopharyngeal muscular dystrophy gene, poly(A)-binding protein 2, interacts with SKIP and stimulates muscle-specific gene expression. Hum. Molec. Genet., **10**, 1129-1139 (2001).
5. Guttridge**,** D. C., et al., NF-κ-B-induced loss of MyoD messenger RNA: possible role in muscle decay and cachexia. *Science*, **289**, 2363-2366 (2000).

AA,PHC 08/14-1