

**Anti-Actin pan (6-isoforms of actin) and controls**

Cat. # ACTB20-M      Monoclonal Anti-Actin pan (6-isoforms of actin) IgG, ascites      **SIZE:** 100 ul

Muscle (from Latin musculus "little mouse") is contractile tissue of the body and is derived from the mesodermal layer of embryonic germ cells. There are three types of muscles (skeletal, cardiac and smooth): Skeletal muscle or "voluntary muscle" is anchored by tendons to bone. Smooth muscle or "involuntary muscle" is found within the walls of organs and structures such as the esophagus, stomach, intestines, bronchi, uterus, urethra, bladder, and blood vessels, and unlike skeletal muscle, smooth muscle is not under conscious control. Cardiac muscle is also an "involuntary muscle" but is a specialized kind of muscle found only within the heart. Cardiac and skeletal muscle are "striated" in that they contain sarcomeres and are packed into highly-regular arrangements of bundles; smooth muscle has neither. Muscle is mainly composed of muscle cells. Within the cells are myofibrils; myofibrils contain sarcomeres, which are composed of actin and myosin. All three muscles use the movement of actin against myosin to create contraction.

Smooth muscle cells are generally arranged in sheets or bundles and connected by gap junctions. In order to contract the cells contain intracellular contractile filamentous proteins called actin and myosin. Smooth muscle does not contain the proteins troponin or titin, and caldesmon and calponin are significant proteins expressed within smooth muscle. Actin filaments attach to the sarcolemma by focal adhesions or attachment plaques and attach to other actin filaments via dense bodies (acting much like Z-lines in striated muscle).

Actin and myosin are the two major cytoskeleton proteins implicated in cellular movement, secretion, phagocytosis, and kinesis. Actin is one of the most conserved cellular protein. At least 6 actin isoforms have been identified by protein sequence analyses. Four actin isoforms represent the differentiation markers of muscle tissues. There are three  $\alpha$ -actins:  $\alpha$ -skeletal,  $\alpha$ -cardiac, and  $\alpha$ -smooth muscle), one  $\beta$ -actin ( $\beta$ -non-muscle), and two  $\gamma$ -actins ( $\gamma$ -smooth muscle and  $\gamma$ -non-muscle). Actin isoform are >90% conserved, except in the N-terminal 18-aa (50-60% homology). Beta-actin protein and mRNA levels are often used as a reference for comparing changes in cellular protein/mRNA levels by Western or Northern blots.

**Source of Antigen and Antibodies**

<b>Antigen</b>	Purified chicken actin protein
<b>Ab Host/type</b>	Mouse, monoclonal unpurified ascites # <b>ACTB20-M</b> (isotype IgG1k)
<b>2-Ab</b>	<b>Goat Anti-mouse IgG-HRP conjugate</b> Cat # 40320 (AP, biotin, FITC conjugates also available)
<b>-ve</b>	Cat # 20008-1, Mouse (non-immune) Serum IgG, purified, suitable for ELISA, Western, IHC as -ve control

Chicken gizzard smooth muscle actin (cat # ACTB20-N) was purified (>99%, mol wt ~43 kda) and used for control. For Western blot +ve control (Cat # **ACTB20-C**) is supplied in SDS-PAGE sample buffer (reduced). Load 10 ul/lane of ACTB20-C for good visibility with antibody Cat # ACTB20-M or other antibodies. Store at -20oC in suitable size aliquots. SDS may crystallize in cold conditions. It should redissolve by warming before taking it from the stock. It should be heated once prior to loading on gels. If the product has been stored for several weeks, then it may be preferable to add 5 ul of fresh 2x sample buffer per 10 ul of the ACTB20-C solution prior to heating and loading on gels. This preparation is not biologically active. It is not suitable for ELISA or other applications where native protein is required. Do not freeze, thaw, or heat repeatedly

**Recommended Usage**

Western Blotting: monoclonal antibodies at 1:1500-1:2K using Chemiluminescence technique)..

**ELISA** (1:10-50K; using 50-200 ng control antigen/well).

**IHC/IF:** Immunohistochemistry (paraffin, formalin, deparaffinized methacarn-fixed tissue: 1:2000-1:5,000 and alcohol fixation).

**Storage**

**Short-term:** unopened, undiluted liquid vials for less than a week at 4oC.

**Long-term:** at -20C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

**Stability:** 6-12 months at -20oC or below.

**Specificity & Cross-reactivity**

ACTB20-M antibody is a pan-actin antibody that binds to an epitope in a highly conserved region of actin; therefore, this antibody reacts with all six isoforms of vertebrate actin as well as plants (as well as with Dictyostelium discoideum and Physarum polycephalum actins). The epitope recognized by the antibody appears to be located ~50-70 aa. The antibody reacts with both globular (G) and filamentous (F) forms of actin and does not interfere with actin polymerization to form filaments. Human beta-actin non muscle (ACTB11-C) or ACTB15-C (muscle actin) or smooth muscle actin (#ACTB19-C) can use as controls for western.

**References:** (1). Ohmuri H (1995) Gene Accession # S38782; Vandekerchove, J et al (1978) Eur. J. Biochem. 90, 451; Lessard J et al (1988) Cell. Motil Cytoskel. 10, 349; North JA et al (1994) J. Cell Sci. 107, 437;

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