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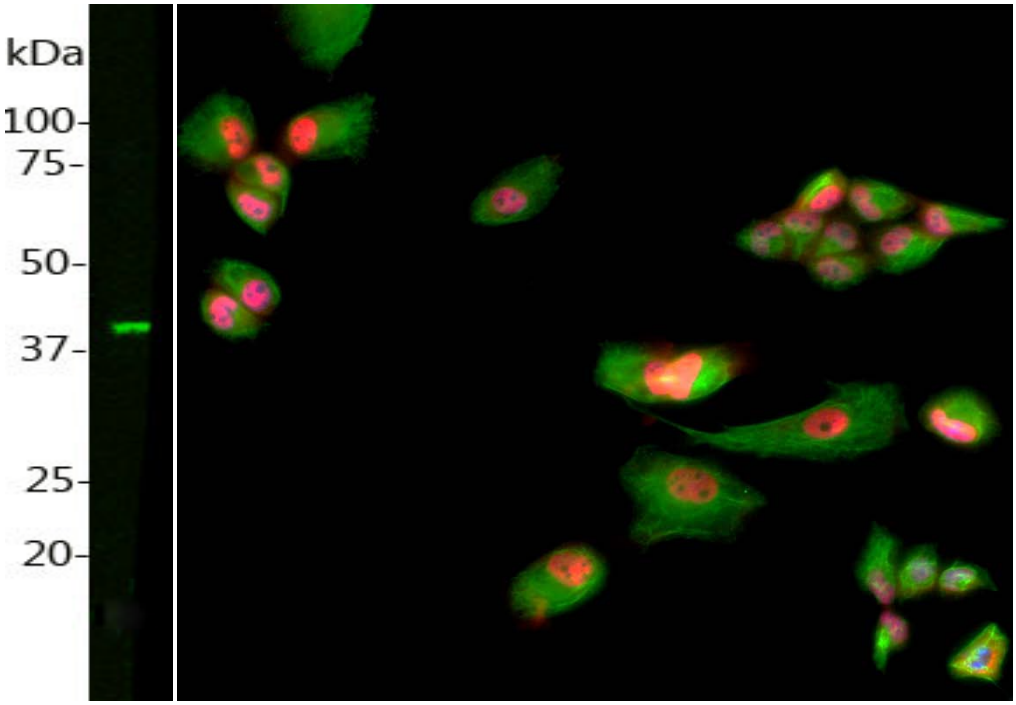
Catalogue# MCA-1H1: Monoclonal antibody to Muscleblind-like 1

The Immunogen: Muscleblind was originally isolated following studies of *Drosophila*, since inactivation of the muscleblind (mbl) gene in this species resulted in defects the development of both muscles and the visual system (1). The *Drosophila* Muscleblind protein contains 4 so called Zn-CCCH Motifs (a.k.a. Cys3His or C3H motifs), members of the Zinc finger family of nucleic acid binding motifs. The Zn-CCCH motifs are about 26 amino-acids long, and each coordinate one Zinc ion and may bind RNA. Human homologues of the single *Drosophila* muscleblind gene were discovered from cDNA and genomic sequencing, but were also discovered as a result of experiments aimed at finding proteins which bind to polynucleotide repeated sequences. Several important human diseases are associated with expansion of polynucleotide sequences, in most cases trinucleotide repeats. Myotonic dystrophy (DM1) is one of these diseases, and is associated with increases in the number of CTG repeats in the 3′ UTR of the gene encoding myotonin (a.k.a. DM Kinase, DMK or myotonin protein kinase), a ser/thr kinase expressed specifically in muscle. Normal humans have 5-37 tandem CTG repeats in this gene while those with myotonic dystrophy have from 50 to as many as 1,000 CTG repeats, with individuals with the most serious forms of the disease generally having the most repeats. Since the increased repeats were found in the 3′ untranslated region (UTR) of the myotonin gene and so do not encode protein, it was puzzling as to how the increased number repeats caused the disease, especially since knock out of the myotonin gene has a relatively mild phenotype (2, 3). One theory is that the repeated sequences sequester important RNA binding and processing proteins and hence reduced the efficiency of RNA processing which would have a generally deleterious effect. To test this theory Miller et al. (4) looked for proteins that would bind specifically to the product of genomic CTG repeats in the 3′ UTR, double stranded CUG RNA repeats. They found several which they named EXP proteins, for triplet repeat expansion dsRNA-binding proteins. These proteins were then identified as the mammalian homologues of the *Drosophila* Muscleblind protein. Mammalian genomes contain three muscleblind-like proteins, generally referred to as MBNL1, MBNL2 and MBNL3, and all three were found to associate with long double stranded CUG RNA repeats. In situ hybridization studies in mouse show that all three genes are widely expressed especially in development and show significant overlap with myotonin expression (5). Recent studies show that transgenic knock out of MBNL1 in mice results in a phenotype similar to that of myotonic dystrophy, and also reduced efficiency of mRNA processing similar to those seen in this disease, both observations supporting the RNA sequestration theory (6). Although the exact function of Muscleblind proteins is not known, this and other evidence suggests that they have a role in RNA splice site selection (6).

Antibody characteristics: MCA-1H1 is a mouse IgG1 class antibody. It was raised against the full-length recombinant Muscleblind-like 1 protein expressed in and purified from *E. coli.*

Suggestions for use: The antibody solution is affinity purified from tissue culture supernatant and is at concentration of 1mg/ml in phosphate buffered saline. The antibody solution can be used at dilutions of at least 1:1,000 in immunofluorescence experiments. In western blotting using chemiluminescence it can be used at dilutions of 1:1,000-1:5,000. Antibody preparation contains 10mM sodium azide preservative (Link to http://www.encorbio.com/MSDS/azide.htm for Material Safety Data Sheet). Avoid repeated freezing and thawing, store at 4°C or -20°C.



Left: Western blot analysis of MCA-1H1. Blot of HL60 cell lysate was probed with MCA-1H1. The MCA-1H1 monoclonal binds Muscleblind-like protein 1 at ~40kDa, as expected. **Right:** HeLa cells were stained with monoclonal antibody MCA-1H1. Muscleblind-like protein 1 is present in the nucleus of HeLa cells (red). Co-staining is with our rabbit polyclonal antibody against vimentin (RPCA-Vim, in red) reveals cytoplasmic intermediate filaments. Blue is a DNA stain which largely colocalizes with the MBNL1 protein in the nucleus.

References:

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Limitations: This product is for research use only and is not approved for use in humans or in clinical diagnosis.