

# APC618Hu01 100µg Active Microfibrillar Associated Protein 2 (MFAP2) Organism Species: Homo sapiens (Human) Instruction manual

FOR IN VITRO USE AND RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

#### [PROPERTIES]

Source: Prokaryotic expression.

Host: E. coli

Residues: Leu6~Val162 Tags: N-terminal His-tag

**Purity: >95%** 

Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 0.05% sarcosyl

and 5% trehalose.

**Applications:** Cell culture; Activity Assays.

(May be suitable for use in other assays to be determined by the end user.)

**Predicted isoelectric point:** 5.5

Predicted Molecular Mass: 22.0kDa

Accurate Molecular Mass: 38kDa as determined by SDS-PAGE reducing conditions.

#### Phenomenon explanation:

The possible reasons that the actual band size differs from the predicted are as follows:

- 1. Splice variants: Alternative splicing may create different sized proteins from the same gene.
- 2. Relative charge: The composition of amino acids may affects the charge of the protein.
- 3. Post-translational modification: Phosphorylation, glycosylation, methylation etc.
- 4. Post-translation cleavage: Many proteins are synthesized as pro-proteins, and then cleaved to give the active form.
- 5. Polymerization of the target protein: Dimerization, multimerization etc.

#### [USAGE]

Reconstitute in 20mM Tris, 150mM NaCl (pH8.0) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

#### [STORAGE AND STABILITY]

Storage: Avoid repeated freeze/thaw cycles.

Store at 2-8°C for one month.

Aliquot and store at -80°C for 12 months.

**Stability Test:** The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37°C for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date under appropriate storage condition.

#### [SEQUENCE]

LFLLF LPAGLLAQGQ YDLDPLPPFP DHVQYTHYSD QIDNPDYYDY QEVTPRPSEE QFQFQSQQV QQEVIPAPTP EPGNAELEPT EPGPLDCREE QYPCTRLYSI HRPCKQCLNE VCFYSLRRVY VINKEICVRT VCAHEELLRA DLCRDKFSKC GV

### [ACTIVITY]

Microfibrillar-associated protein 2 (MFAP2) is an O-glycosylated protein which excreted to the extracellular space and the extracellular matrix. MFAP2 combine biglycan and elastin to form a ternary complex. MFAP2 plays a key role in the support and distensibility of the juxtacanalicular region of these collector channels. It also can inhibit LTB-1 binding to fibrillin-1, stimulate the phosphorylation of Smad2, and thereby mediate the subsequent extracellular deposition of latent TGFbeta. Besides, Fibrillin 1 (FBN1) has been identified as an interactor of MFAP2, thus a binding ELISA assay was conducted to detect the interaction of recombinant human MFAP2 and recombinant human FBN1. Briefly, MFAP2 were

diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100uL were then transferred to FBN1-coated microtiter wells and incubated for 2h at  $37^{\circ}$ C. Wells were washed with PBST and incubated for 1h with anti-MFAP2 pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated and washed 3 times. With the addition of substrate solution, wells were incubated 15-25 minutes at  $37^{\circ}$ C. Finally, add  $50\mu$ L stop solution to the wells and read at 450nm immediately. The binding activity of MFAP2 and FBN1 was shown in Figure 1, and this effect was in a dose dependent manner.

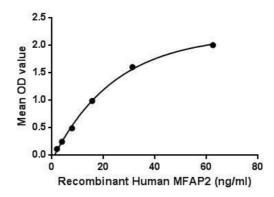


Figure 1. The binding activity of MFAP2 with FBN1.

#### [IDENTIFICATION]

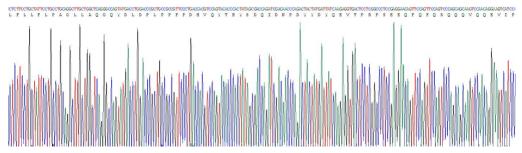


Figure 2. Gene Sequencing (extract)

## Coud-Clone Corp.

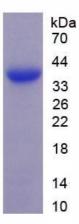


Figure 3. SDS-PAGE

Sample: Active recombinant MFAP2, Human

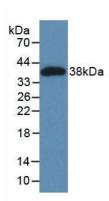


Figure 4. Western Blot

Sample: Recombinant MFAP2, Human;

Antibody: Rabbit Anti-Human MFAP2 Ab (PAC618Hu01)