

APA032Hu01 100µg
Active Fibroblast Growth Factor 1, Acidic (FGF1)
Organism Species: Homo sapiens (Human)
Instruction manual

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

1th Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Met1~Asp155

Tags: Two N-terminal Tags, His-tag and GST-tag

Purity: >98%

Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 0.05% sarcosyl and 5% trehalose.

Applications: Cell culture; Activity Assays; In vivo assays.

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

Predicted isoelectric point: 6.5

Predicted Molecular Mass: 47.3kDa

Accurate Molecular Mass: 55kDa 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]

MAEGEITTF ALTEKFNLP GNYKKPKLLY CSNGGHFLRI LPDGTVDGTR
DRSDQHIQLQ LSAESVGEVY IKSTETGQYL AMDTDGLLYG SQTPNEECLF
LERLEENHYN TYISKKHA EK NWFVGLKKN G SCKRGPRTHY GQKAILFLPL
PVSSD

[ACTIVITY]

FGF1 (Fibroblast growth factor 1) is a member of FGF family, which plays an important role in the regulation of cell survival, cell division, angiogenesis, cell differentiation and cell migration. FGF1 is thought to stimulate the proliferation of 3T3 fibroblasts, besides, human FGF1 and mouse FGF1 shares similarities in amino acid sequence with the identity of 96%. Thus, a cell proliferation assay was conducted to detect the bioactivity of recombinant human FGF1 using 3T3 fibroblasts. Briefly, 3T3 cells were seeded into triplicate wells of 96-well plates at a density of 2,000 cells/well and allowed to attach overnight, then the medium was replaced with serum-free standard DMEM prior to the addition of various

concentrations of FGF1. After incubated for 48h, cells were observed by inverted microscope and cell proliferation was measured by Cell Counting Kit-8 (CCK-8). Briefly, 10 μ L of CCK-8 solution was added to each well of the plate, then the absorbance at 450nm was measured using a microplate reader after incubating the plate for 1-4 hours at 37°C. Proliferation of 3T3 cells after incubation with FGF1 for 48h observed by inverted microscope was shown in Figure 1. Cell viability was assessed by CCK-8 (Cell Counting Kit-8) assay after incubation with recombinant FGF1 for 48h. The result was shown in Figure 2. It was obvious that FGF1 significantly increased cell viability of 3T3 cells.



Figure 1. Cell proliferation of 3T3 cells after stimulated with FGF1.

(A) 3T3 cells cultured in DMEM, stimulated with 100ng/ml FGF1 for 48h;

(B) Unstimulated 3T3 cells cultured in DMEM for 48h.

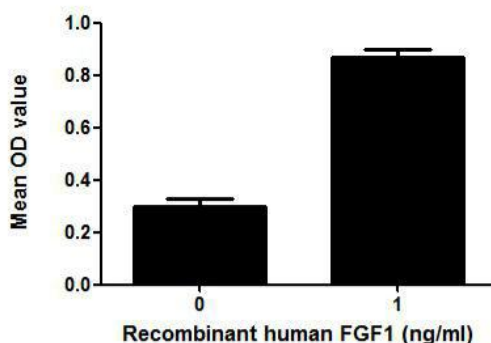


Figure 2. Cell proliferation of 3T3 cells after stimulated with FGF1.

[IDENTIFICATION]

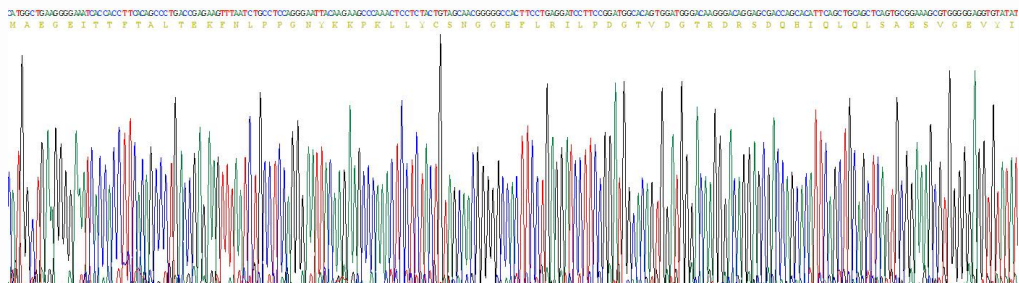


Figure 3. Gene Sequencing (extract)

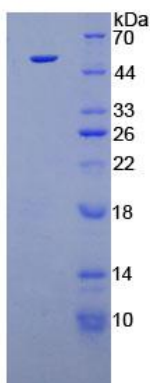


Figure 4. SDS-PAGE

Sample: Active recombinant FGF1, Human

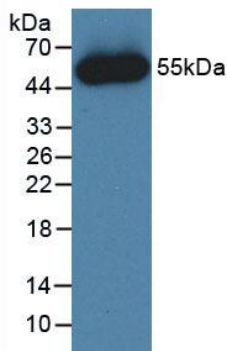


Figure 5. Western Blot

Sample: Recombinant FGF1, Human;

Antibody: Rabbit Anti-Human FGF1 Ab (PAA032Hu01)