APN576Hu01 100μg
Active Fibronectin Type III Domain Containing Protein 5 (FNDC5)
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: Leu2~Ile137
Tags: N-terminal His-tag
Purity: >92%
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 1mM EDTA, 1mM DTT, 0.01% sarcosyl, 5% trehalose, and Proclin300.
Predicted isoelectric point: 6.2
Predicted Molecular Mass: 16.6kDa
Accurate Molecular Mass: 18kDa as determined by SDS-PAGE reducing conditions.
Applications: Cell culture; Activity Assays; In vivo assays.
(May be suitable for use in other assays to be determined by the end user.)

[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.

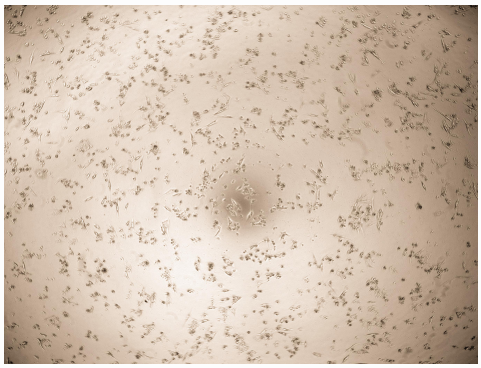
[SEQUENCE]

LRFIQEVNT TTRSCALWDL EEDTEYIVHV QAISIQGQSP ASEPVLFKTP REAEKMASKN KDEVTMKEMG RNQQLRTGEV LIIVVVLFMW AGVIALFCRQ YDIIKDNEPN NNKEKTKSAS ETSTPEHQGG GLLRSKI

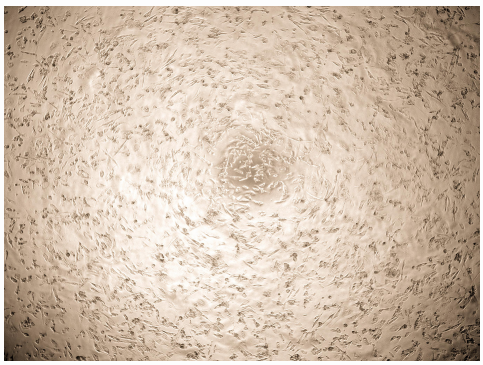
[ACTIVITY]

Fibronectin type III domain-containing protein 5, the precursor of irisin, is a protein that is encoded by the FNDC5 gene. It was reported that FNDC5 significantly decreased cell number, migration and viability through apoptosis in malignant MDA-MB-231 cells. Thus MDA-MB-231 cells were seeded overnight at a density of 5,000 cells/well, and treated with or without various concentrations of FNDC5 for 48h, then MDA-MB-231 cells were observed by inverted microscope and cell viability was measured by Cell Counting Kit-8 (CCK-8). Briefly, 10μL of CCK-8 solution was added to each well of the plate, then measure the absorbance at 450nm using a microplate reader after incubating the plate for 1-4 hours at 37°C.

Cell apoptosis of MDA-MB-231 cells after incubation with FNDC5 for 48h observed by inverted microscope was shown in Figure 1.



A



B

Figure 1. Cell apoptosis of MDA-MB-231 cells after stimulated with FNDC5 .

(A) MDA-MB-231 cells cultured in DMEM, stimulated with FNDC5(10ug/ml) for 48h;

(B) Unstimulated MDA-MB-231 cells cultured in DMEM for 48h.

The dose-effect curve of FNDC5 was shown in Figure 2. It was obvious that FNDC5 significantly decreased cell viability of MDA-MB-231 cells. The ED50 for this effect is typically 7.23~21.92ug/ml.

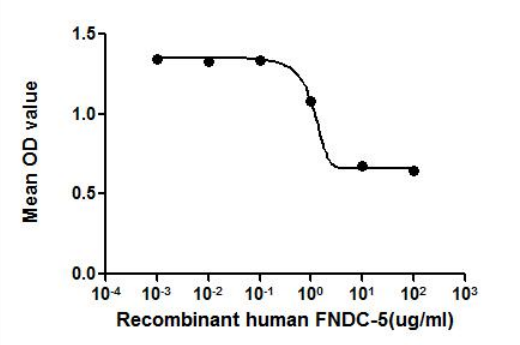


Figure 2. The dose-effect curve of FNDC5 on MDA-MB-231 cells

[IDENTIFICATION]

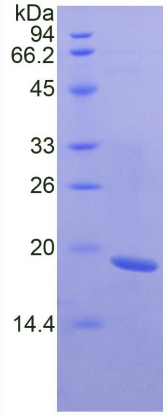


Figure 3. SDS-PAGE

Sample: Active recombinant FNDC5, Human

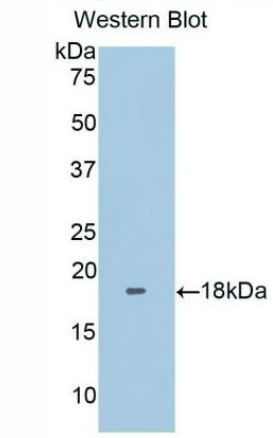


Figure 4. Western Blot

Sample: Recombinant FNDC5, Human;

Antibody: Rabbit Anti-Human FNDC5 Ab (PAN576Hu01)