

APA560Ra01 10µg
Active Epidermal Growth Factor (EGF)
Organism Species: *Rattus norvegicus (Rat)*
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Asn974~Arg1026

Tags: N-terminal His and GST Tag

Purity: >98%

Endotoxin Level: <1.0EU per 1µg (determined by the LAL method).

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: 6.7

Predicted Molecular Mass: 36.1kDa

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

[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]

NSNTGCP PSYDGYCLNG GVCMYVESVD
RYVCNCVIGY IGERCQHRDL RWWKLR

[ACTIVITY]

Epidermal growth factor (EGF) binding to EGFR, results in cellular proliferation, differentiation, and survival. To test the effect of EGF on cell proliferation, 3T3 fibroblasts cells were seeded into triplicate wells of 96-well plates at a density of 2,000 cells/well and allowed to attach, replaced with serum-free overnight, then the medium was replaced with 1% serum standard DMEM prior to the addition of various concentrations of recombinant rat EGF. After incubated for 96h, 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 EGF for 96h 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 EGF for 96h. The result was shown in Figure 2. It was obvious that EGF significantly increased cell viability of 3T3 cells.

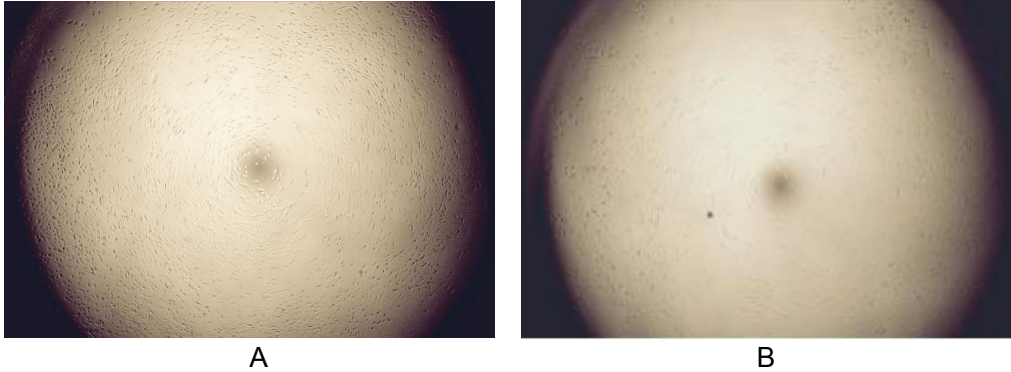


Figure 1. Cell proliferation of 3T3 cells after stimulated with EGF.

- (A) 3T3 cells cultured in DMEM, stimulated with 1ng/mL EGF for 96h;
- (B) Unstimulated 3T3 cells cultured in DMEM for 96h.

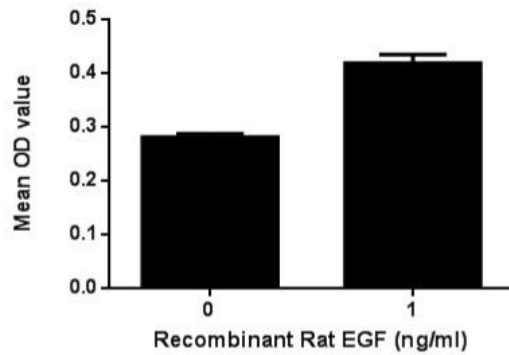


Figure 2. Cell proliferation of 3T3 cells after stimulated with EGF.

[IDENTIFICATION]

iMAGTAAQACAGGATGCGDGGGCTGTCAGATGGACTGCCTCAATGGTGGGGTGGATGTAATGTTGAATCCGTGGACCGCTAGCGTGTGCATGTTGTCATTTGGGCTATAATTGGAGMGGATGTGACGACGACCTTAGCTGGTGGAGCTGGCC
 N S N T G C P P S Y D G Y C L N G G V C H Y V E S V D R Y V C N C V I G Y I G E R C Q H R D L R W W K L R

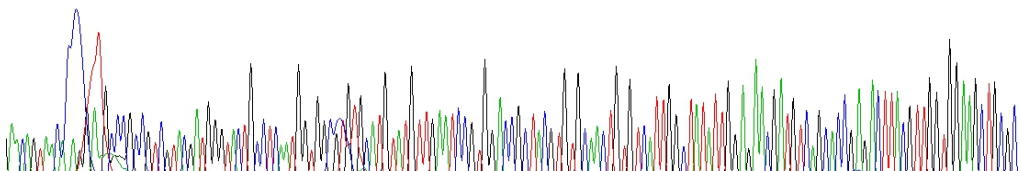


Figure 3 . Gene Sequencing (extract)

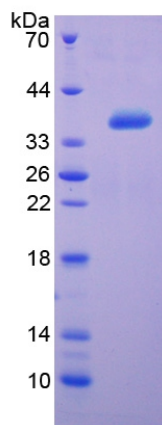


Figure 4. SDS-PAGE

Sample: Active recombinant EGF, Rat

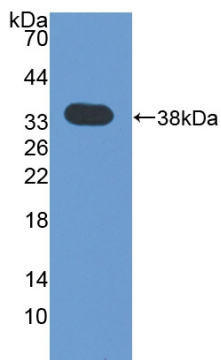


Figure 5. Western Blot

Sample: Recombinant EGF, Rat;

Antibody: Rabbit Anti-Rat EGF Ab (PAA560Ra01)

[IMPORTANT NOTE]

The kit is designed for in vitro and research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.