

APA051Hu01 50µg
Active Insulin Like Growth Factor 2 (IGF2)
Organism Species: *Homo sapiens* (Human)
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Ala25~Glu91

Tags: N-terminal His-tag

Purity: >90%

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

Buffer Formulation: PBS, pH7.4, containing 5%Trehalose .

Original Concentration: 200µg/mL

Applications: Activity Assays.

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

Predicted isoelectric point: 6.3

Predicted Molecular Mass: 11.2kDa

Accurate Molecular Mass: 14kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

Reconstitute in 10mM PBS (pH7.4) 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]

AYRPSETLCGGELVDTLQFVCGDRGFYFSRPASRVSRRSRGIVECCFRSCDLALLETYCATPAKSE

[ACTIVITY]

Insulin-like Growth Factor 2 (IGF2) is a mitogenic peptide hormone critical for fetal growth and development, predominantly synthesized by the liver and placenta. It mediates signaling through the IGF1 receptor (IGF1R) and insulin receptor isoform A (IR-A) to govern cell proliferation, differentiation, and metabolic processes. Dysregulated IGF2 expression is associated with oncogenesis and overgrowth disorders such as Beckwith-Wiedemann syndrome. IGF2 exhibits high-affinity binding to Insulin-like Growth Factor Binding Protein 5 (IGFBP5), which dynamically modulates IGF2 bioactivity by either enhancing receptor engagement or restricting ligand-receptor interaction, contingent on the cellular microenvironment. Thus a functional binding ELISA assay was conducted to detect the interaction of recombinant human IGF2 and recombinant rat IGFBP5. Briefly, biotin-linked IGF2 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100µl were then transferred to IGFBP5-coated microtiter wells and incubated for 1h at 37°C. Wells were washed with PBST 3 times and incubation with Streptavidin-HRP for 30min, then wells were aspirated and washed 5 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37°C. Finally, add 50µl stop solution to the wells and read at 450nm immediately. The binding activity of IGF2 and IGFBP5 was shown in Figure 1, the EC50 for this effect is 0.46ug/mL.

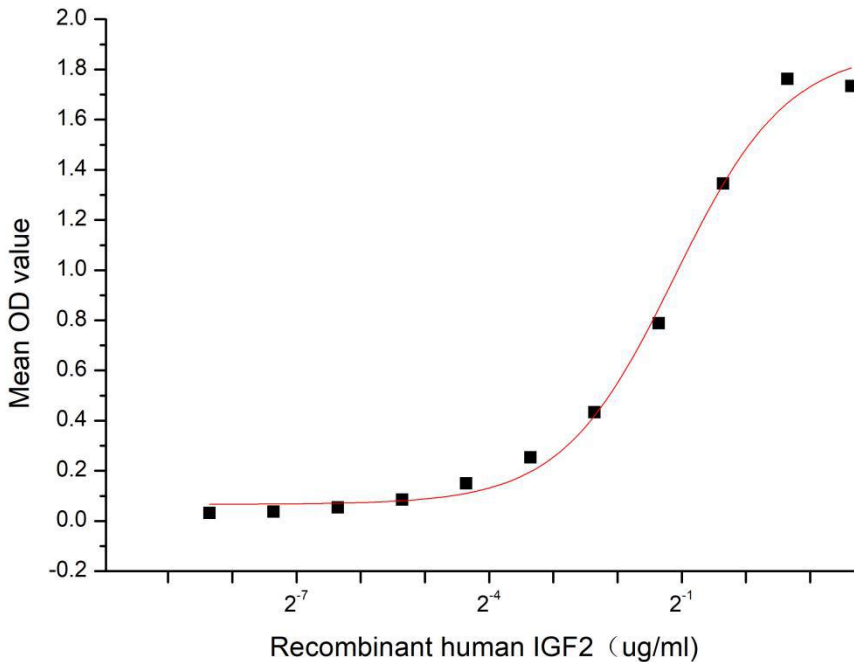


Figure 1. The binding activity of recombinant human IGF2 and recombinant rat IGFBP5

Insulin Like Growth Factor 2 (IGF2) is a key peptide hormone with critical roles in growth, development, and metabolism. It exerts its effects by binding to the IGF1 receptor (IGF1R) and insulin receptor (IR), activating downstream signaling pathways that promote cell proliferation, survival, and differentiation. As IGF2 has the function of cell proliferation, we measure the activity of recombinant human IGF2 by the ability of the protein to stimulate MCF-7 cells proliferation. MCF-7 cells were seeded into triplicate wells of 96-well plates and allowed to attach, replaced with various concentrations of recombinant human IGF2. After incubated for 72h, 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 450 nm was measured using a microplate reader after incubating the plate for 1-4 hours at 37 °C. Cell viability was assessed by CCK-8 assay after incubation with

recombinant human IGF2 for 72h. The result was shown in Figure 1. It was obvious that IGF2 significantly increased cell viability of MCF-7 cells. The ED50 of recombinant human IGF2 is 3.339 µg/ml.

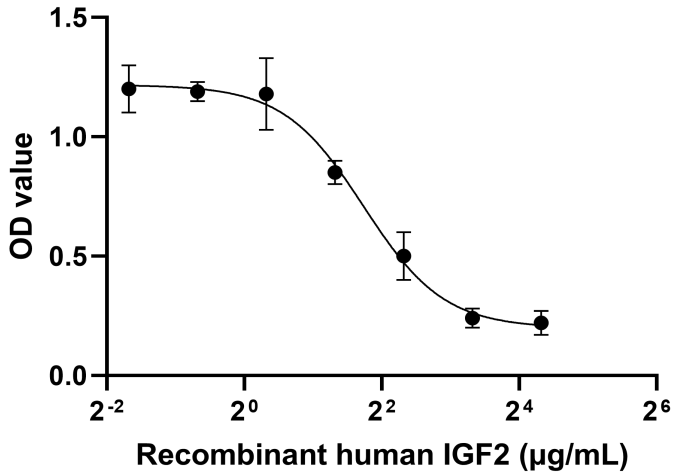


Figure2. The dose-effect curve of recombinant human IGF2 on MCF-7 cells

[IDENTIFICATION]

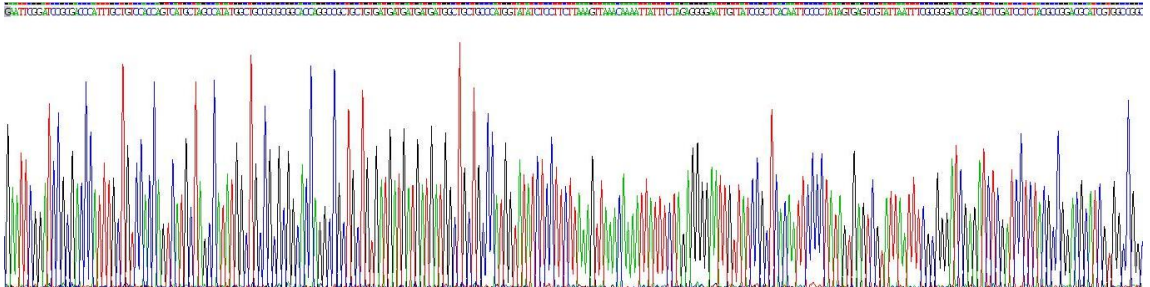


Figure 3. Gene Sequencing (extract)

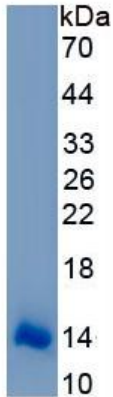


Figure 4. SDS-PAGE

Sample: Active recombinant IGF2, Human

[IMPORTANT NOTE]

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