

APB166Hu61 10µg

Active VGF Nerve Growth Factor Inducible (VGF)

Organism Species: Homo sapiens (Human)

Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Eukaryotic expression.

Host: 293F cell

Residues: Asp330~Pro449 Tags: N-terminal His-tag

Purity: >95%

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

Buffer Formulation: PBS, pH7.4, containing 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: 3.8

Predicted Molecular Mass: 14.9kDa

Accurate Molecular Mass: 19kDa 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 ddH₂O to a concentration of 0.1-0.3mg/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]

D LLLQYLLQGG ARQRGLGGRG

LQEAAEERES AREEEEAEQE RRGGEERVGE EDEEAAEAEA EAEEAERARQ NALLFAEEED GEAGAEDKRS QEETPGHRRK EAEGTEEGGE EEDDEEMDP

[ACTIVITY]

Neurosecretory protein VGF is specifically expressed in a subpopulation of neuroendocrine cells, and is upregulated by nerve growth factor. Human VGF precursor is 615 amino acids (aa) in length. It contains an 22 aa signal sequence plus a 593 aa mature region. The structural organization of this gene is similar to that of the rat gene, and both the translated and the untranslated regions show a high degree of sequence similarity to the rat gene. The encoded secretory protein also shares similarities with the secretogranin/chromogranin family, however, its exact function is not known. Besides, Early Growth Response Protein 1 (EGR1) has been identified as an interactor of VGF, thus a binding ELISA assay was conducted to detect the interaction of recombinant human VGF and recombinant human EGR1. Briefly, EGR1 was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100µl were then transferred to VGF-coated microtiter wells and incubated for 1h at 37°C. Wells were washed with PBST and incubated for 1h with anti-EGR1 pAb, then aspirated and washed 3 times.

After incubation with HRP labelled secondary antibody for 1h at 37 $^{\circ}$ C, wells were aspirated and washed 5 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37 $^{\circ}$ C. Finally, add 50µL stop solution to the wells and read at 450/630nm immediately. The binding activity of VGF and EGR1 was shown in Figure 1, the EC50 for this effect is 0.11066 ug/mL.

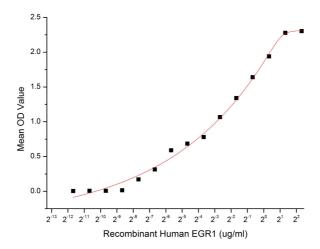


Figure 1. The binding activity of VGF with EGR1

[IDENTIFICATION]

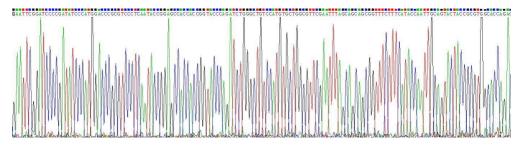


Figure 2. Gene Sequencing (extract)

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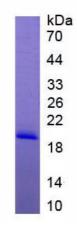


Figure 3. SDS-PAGE

Sample: Active recombinant VGF, 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.