#### APB166Hu01 100µg 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: Prokaryotic expression. Host: *E. coli* Residues: Asp330~Pro449 Tags: N-terminal His-tag Purity: >98% Endotoxin Level: <1.0EU per 1µg (determined by the LAL method). Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 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: 4.8 Predicted Molecular Mass: 43.3kDa Accurate Molecular Mass: 43kDa as determined by SDS-PAGE reducing conditions.

## [ <u>USAGE</u> ]

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.

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**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 °C, 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

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450/630nm immediately. The binding activity of VGF and EGR1 was shown in Figure 1, the EC50 for this effect is 0.60912 ug/mL.



Figure 1. The binding activity of VGF with EGR1



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Figure 3. SDS-PAGE Sample: Active recombinant VGF, Human

## [<u>IMPORTANT NOTE</u>]

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.