

APA049Hu61 100μg

**Active Interferon Gamma (IFNg)** 

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: Gln24~Gln166 Tags: N-terminal His-tag

**Purity: >95%** 

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

Buffer Formulation: 10mM PBS, pH7.4, containing 5% trehalose, 0.01%

sarcosyl.

**Applications:** Cell culture; Activity Assays; In vivo assays.

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

Predicted isoelectric point: 9.7

Predicted Molecular Mass: 18.4kDa

Accurate Molecular Mass: 22&25kDa 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 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]

QDPYVKE AENLKKYFNA GHSDVADNGT LFLGILKNWK EESDRKIMQS QIVSFYFKLF KNFKDDQSIQ KSVETIKEDM NVKFFNSNKK KRDDFEKLTN YSVTDLNVQR KAIHELIQVM AELSPAAKTG KRKRSQMLFR GRRASQ

### [ACTIVITY]

IFN- $\gamma$  is an important activator of macrophages, it promotes production of inducible Nitric Oxide Synthase (iNOS) in macrophages. After stimulated with IFN- $\gamma$ , morphological changes will occur in murine macrophage cell line (Raw 246.7 cells), and inducible nitric-oxide synthase (iNOS) in the cells will increase. Raw 246.7 cells were incubated in DMEM with IFN- $\gamma$  (2ng/mL) for 24h, then cells were observed by inverted microscope and iNOS in cell lysates was detected by ELISA.

Effect of IFN-y on morphological change of Raw 246.7 cells is shown in Figure 1.

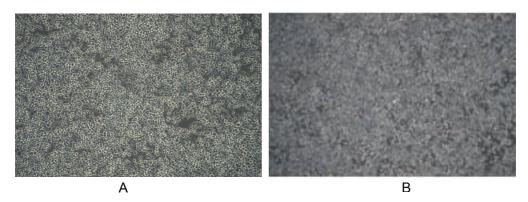


Figure 1. Morphological change of Raw 246.7 cells after stimulation of IFNg.

- (A) Raw 264.7 cells cultured in DMEM, stimulated with IFNg;
- (B) Unstimulated Raw 246.7 cells cultured in DMEM (negative control).

Effect of IFN-y on the expression of iNOS is shown in Table 1.

Table 1. ELISA detection of iNOS expression from RAW 246.7 cells stimulated by IFNg.

Sample	Concentration of iNOS
(cell lysates of Raw 246.7 cells)	(ng/mL)
Stimulated with IFNg (2ng/mL)	8.62
Unstimulated	2.71

## [IDENTIFICATION]

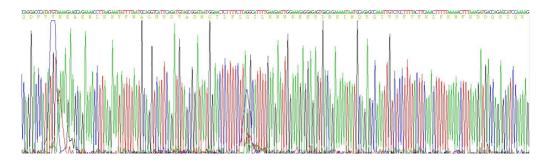


Figure 2. Gene Sequencing (extract)

# Cloud-Clone Corp.

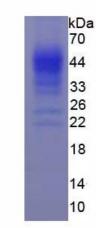


Figure 3. SDS-PAGE

Sample: Active recombinant IFNg, Human

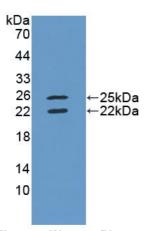


Figure 4. Western Blot

Sample: Recombinant IFNg, Human;

Antibody: Rabbit Anti-Human IFNg Ab (PAA049Hu06)

# [ 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.