

**APA899Hu61 100µg**  
**Active Osteopontin (OPN)**  
**Organism Species: Homo sapiens (Human)**  
***Instruction manual***

FOR RESEARCH USE ONLY  
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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1st Edition (Apr, 2016)

## **[ PROPERTIES ]**

**Source:** eukaryotic expression.

**Host:** 293 cell

**Residues:** Ile17~Asn287

**Tags:** N-terminal His-tag

**Purity:** >95%

**Buffer Formulation:** 20mM Tris, 150mM NaCl, pH8.0, containing 1mM EDTA, 1mM DTT, 0.01% sarcosyl, 5% trehalose, and Proclin300.

**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:** 4.3

**Predicted Molecular Mass:** 32.2kDa

**Accurate Molecular Mass:** 60kDa 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 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 ]**

```
IPVK QADSGSSEEK QNAVSSEETN DFKQETLPSK
SNESHDMDD MDEDDDDHV DSQDSIDSND SDDVDDTDDS HQSDESHSD
ESDELVTDFP TDLPADEVFT PVVPTVDTYD GRGDSVVYGL RSKSKKFRFP
DIQYPDATDE DITSHMESEE LNGAYKAIPV AQDLNAPSDW DSRGKDSYET
SQLDDQSAET HSHKQSRLYK RKANDESNEH SDVIDSQELS KVSREFHSHE
FHSHEMLVV DPKSKEEDKH LKFRISHELD SASSEVN
```

## **[ ACTIVITY ]**

Osteopontin (OPN), a multifunctional phosphorylated glycoprotein, plays an important role in neutrophil recruitment and was found to induce the expression of proinflammatory chemokines including MCP-1 and MIP-1 $\beta$  which promote migration and recruitment of inflammatory cells. It has been reported that OPN induces MCP-1 expression through the NF-kappa B pathways in MCF-7 breast cancer cell line. Briefly, MCF-7 cells were seeded overnight at a density of 1x10<sup>5</sup> cells/mL, and treated with or without 200ng/mL OPN for 24h and MCP-1 levels in the cell supernatant were determined by ELISA.

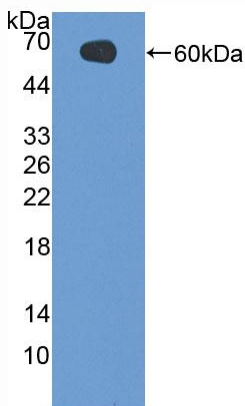
Result: MCP-1 levels in the cell supernatant of MCF-7 cells increased significantly after stimulated with OPN, the data was shown in Table 1 and Figure 1.





**Figure 3. SDS-PAGE**

**Sample: Active recombinant OPN, Human**



**Figure 4. Western Blot**

**Sample: Recombinant OPN, Human;**

**Antibody: Rabbit Anti-Human OPN Ab (PAA899Hu06)**

### **[ 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.