

APA899Mu61 100µg
Active Osteopontin (OPN)
Organism Species: *Mus musculus (Mouse)*
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

12th Edition (Revised in Aug, 2016)

[PROPERTIES]

Source: Eukaryotic expression.

Host: 293F cell

Residues: Leu17~Asn294

Tags: N-terminal His-tag

Purity: >95%

Traits: Freeze-dried powder

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

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

Original Concentration: 200µg/mL

Applications: Cell culture; Activity Assays.

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

Predicted isoelectric point: 4.3

Predicted Molecular Mass: 32.4kDa

Accurate Molecular Mass: 65kDa 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 (pH7.6) 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]

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                LPVK VTDSGSSEEK  LYSLHPDPIA  TWLVPDPSQK
QNL LAPQNAV  SSEEKDDFKQ  ETLPSNSNES  HDHMDDDDDD  DDDGDHAES
EDSVDSDESD  ESHHSDESDE  TVTASTQADT  FTPIVPTVDV  PNGRGDSLAY
GLRSKRSRFQ  VSDEQYPDAT  DEDLTSHMKS  GESKESLDVI  PVAQLLSMPS
DQDNNGKGS  H  ESSQLDEPSL  ETHRLEHSKE  SQESADQSDV  IDSQASSKAS
LEHQSHKFHS  HKDKLVLPDK  SKEDDRYLKF  RISHELESSS  SEVN
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[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 β which promote migration and recruitment of inflammatory cells. It has been reported that OPN induces MCP-1 expression through the NF-kappaB pathways in MCF-7 breast cancer cell line. To test the bioactivity of recombinant mouse OPN, MCF-7 cells were seeded into 24-well plate at a density of 1×10^5 cells/mL, and allowed to attach overnight before treated with certain concentrations of recombinant mouse OPN for 48h and MCP-1 levels in the cell supernatant were determined by ELISA

(SEA087Hu). MCP-1 levels in the cell supernatant of MCF-7 cells increased significantly after stimulated with OPN which was shown in Figure1, the EC50 was 398 ug/ml.

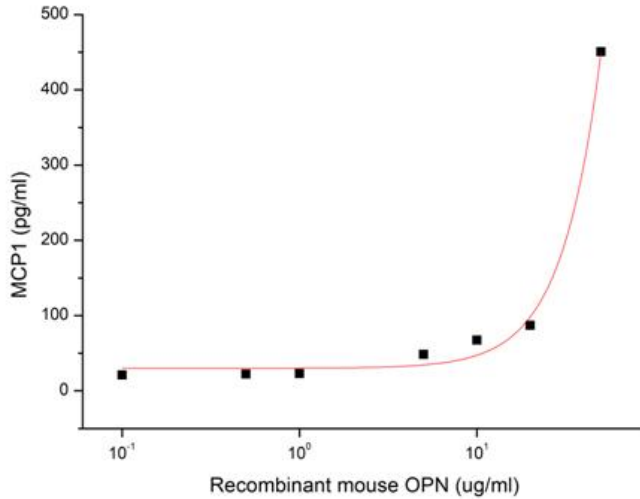


Figure1. MCP-1 levels in the cell supernatant of MCF-7 cells induced by OPN

[IDENTIFICATION]

TC TCCCGG TG AAGTCGAC TG ATTC TGGCAGC TCG AGGAG AAC TTC TCGAGC TG CAGCCGAT CCT TTAGCCAG TGG C TGGCTGGC TG AACGATCTG AAGCGAATCTCC TGGCCGCG CAG TCC TGTCTCTC TCG AAGAGCGTCG CTT TACCGAGAGCTC TTCAGGCA TTCGATG AAGG
 I P V K V T D S G S S E E K L V S I H P D P I A T W L V P D P S Q K Q N L I A P Q N A V S S E E K D D F K Q E T L P S N S N E S :

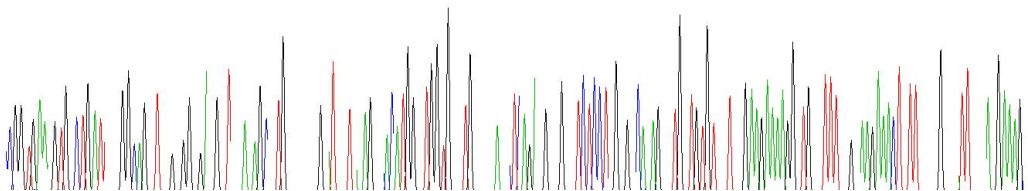


Figure 2. Gene Sequencing (extract)

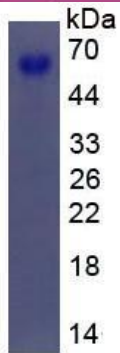


Figure 3. SDS-PAGE

Sample: Active recombinant OPN, Mouse

[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.