APA257Hu62 10μg
Active Procollagen II N-Terminal Propeptide (PIINP)
Organism Species: *Homo sapiens* (Human)
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Eukaryotic expression.
Host: 293F cell
Residues: Gln26~Ala181
Tags: C-terminal His-tag
Purity: >95%
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: PBS, pH7.4, containing 5% Trehalose .
Original Concentration: 1500μg/mL
Applications: Activity Assays.
(May be suitable for use in other assays to be determined by the end user.)
Predicted isoelectric point: 4.2
Predicted Molecular Mass: 17.3kDa
Accurate Molecular Mass: 22&27kDa 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]

QDVQEAGSCVQDGQRYNDKDVWKPEPCRICVCDTGTVLCDDIICEDVKDCLSPEIPFGECCPICPTDLATASGQPGPKGQKGEPGDIK
DIVGPKGPPGPQGPAGEQGPRGDRGDKGEKGAPGPRGRDGEPGTPGNPGPPGPPGPPGPPGLGGNFAA

[ACTIVITY]

Procollagen II N-Terminal Propeptide (PIINP) is a specific proteolytic fragment generated during the biosynthesis of type II collagen, the major structural collagen in hyaline cartilage, intervertebral discs, and the vitreous body of the eye. During chondrocyte-mediated collagen maturation, the N-terminal propeptide is enzymatically cleaved from procollagen II and released into the extracellular matrix and biological fluids. As a direct byproduct of type II collagen synthesis, PIINP serves as a sensitive and specific biomarker for chondrocyte anabolic activity and cartilage formation. It is widely used in research and clinical assessments to evaluate cartilage turnover, monitor disease progression in osteoarthritis and rheumatoid arthritis, and reflect the integrity of cartilage metabolism. GP6, a platelet receptor, can bind to PIINP and mediate cell-matrix adhesion and downstream signaling events. Briefly, GP6 was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100 μL were then

transferred to PIINP-coated microtiter wells and incubated for 1h at 37℃. Wells were washed with PBST and incubated for 1h with anti-GP6 pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody for 1h at 37℃, wells were aspirated and washed 5 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37℃. Finally, add 50 μL stop solution to the wells and read at 450/630nm immediately. Measured by its binding ability in a functional ELISA. When recombinant human PIINP is Immobilized at 2 μg/mL(100 μLwell), the concentration of GP6 that produces 50% optimal bindingresponse is found to be approximately 0.378 μg/mL.

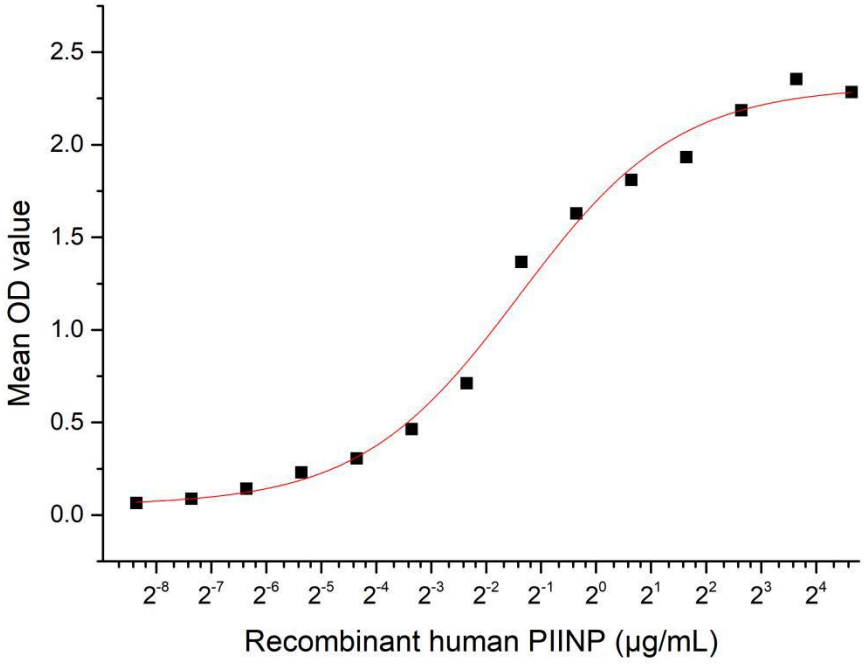

Figure 1. The binding activity of recombinant human PIINP and human GP6

[IDENTIFICATION]

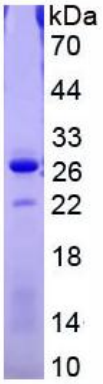

Figure 2. SDS-PAGE

Sample: Active recombinant PIINP, 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.