APB979Hu61 100μg
Active Active Hepcidin (Hepc)
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: Ser25~Thr84
Tags: N-terminal His-tag
Purity: >95%
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: 10mM PBS, pH7.6, containing 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: 9.4
Predicted Molecular Mass: 34.1kDa
Accurate Molecular Mass: 34kDa as determined by SDS-PAGE reducing conditions.

[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 2-8ºC for one month.
Aliquot and store at -80ºC 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 37ºC 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]

SVFPQQ TGQLAELQPQ DRAGARASWM PMFQRRRRRD THFPICIFCC GCCHRSKCGM CCKT

[ACTIVITY]

Hepcidin (Hepc) is a regulator of iron metabolism. Hepcidin inhibits iron transport by binding to the iron export channel ferroportin which is located on the basolateral surface of gut enterocytes and the plasma membrane of reticuloendothelial cells (macrophages). Hepcidin ultimately breaks down the transporter protein in the lysosome. Inhibiting ferroportin prevents iron from being exported and the iron is sequestered in the cells. Besides, Ferroportin (FPN) has been identified as an interactor of Hepc, thus a binding ELISA assay was conducted to detect the interaction of recombinant human Hepc and recombinant human FPN. Briefly, Hepc were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100μL were then transferred to FPN-coated microtiter wells and incubated for 2h at 37℃. Wells were washed with PBST and incubated for 1h with anti-Hepc pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated and washed 3 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37℃. Finally, add 50μL stop solution to the wells and read at 450nm immediately. The binding activity of Hepc and FPN was shown in Figure 1, and this effect was in a dose dependent manner.

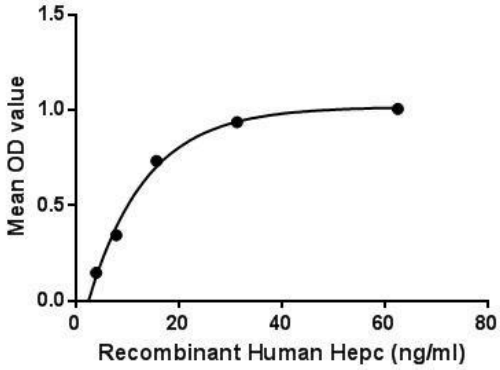

Figure 1. The binding activity of Hepc with FPN.

[IDENTIFICATION]

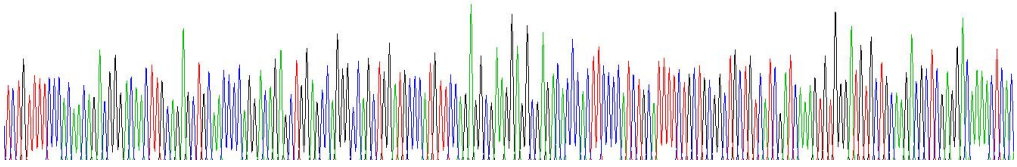

Figure 2. Gene Sequencing (extract)

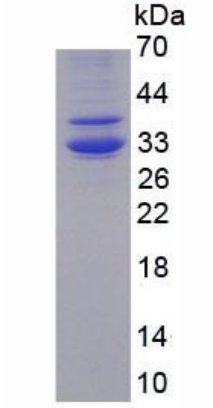

Figure 3. SDS-PAGE

Sample: Active recombinant Hepc, Human

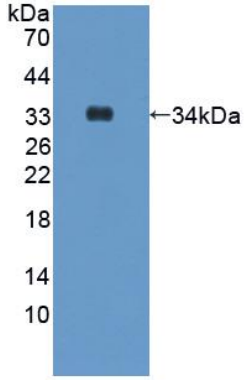

Figure 4. Western Blot

Sample: Recombinant Hepc, Human;

Antibody: Rabbit Anti-Human Hepc Ab (PAB979Hu06)

[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.