APJ103Hu61 100μg
Active Filaggrin (FLG)
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: CHO
Residues: Glu1452~Ser1777
Tags: N-terminal His-tag
Purity: >80%
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: PBS, pH7.4, containing 5% Trehalose .
Original Concentration: 200μg/mL
Applications: Activity Assays.
(May be suitable for use in other assays to be determined by the end user.)
Predicted isoelectric point: 8.3
Predicted Molecular Mass: 36.2kDa
Accurate Molecular Mass: 55kDa 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]

ESTHGQTAPSTGGRQGSRHEQARNSSRHSASQDGQDTIRGHPGSSRGGRQGSYHEQSVDRSG
HSGYHHSHTTPQGRSDASHGQSGPRSASRQTRNEEQSGDGSRHSGSRHHEPSTRAGSSRHSQ
VGQGESAGSKTSRRQGSSVSQDRDSEGHSEDSERRSESASRNHYGSAREQSRHGSRNPRSHQE
DRASHGHSAESSRQSGTRHAETSSGGQAASSQEQARSSPGERHGSRHQQSADSSTDSGTGRR
QDSSVVGDSGNRGSSGSQASDSEGHSEESDTQSVSAHGQAGPHQQSHQESTRGQSGERSGRS
GSFLYQVSTHEQSES

[ACTIVITY]

Filaggrin (FLG), a key keratinocyte differentiation protein encoded by the FLG gene, is critical for skin barrier formation. Synthesized as a large precursor, profilaggrin, it is cleaved into functional filaggrin monomers in the stratum granulosum of the epidermis. These monomers bind to keratin filaments, promoting their aggregation to form a dense, protective cytoskeletal structure. Filaggrin also contributes to skin hydration by breaking down into natural moisturizing factors. FLG interacts with SPINK5, which encodes the serine protease inhibitor LEKTI; SPINK5 regulates proteases that process profilaggrin into filaggrin, maintaining proper barrier function.Briefly, SPINK5 was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100 μl were then

transferred to FLG-coated microtiter wells and incubated for 1h at 37℃. Wells were washed with PBST and incubated for 1h with anti-SPINK5 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 FLG is Immobilized at 2 μg/mL(100 uLwell), the concentration of SPINK5 that produces 50% optimal bindingresponse is found to be approximately11.830 μg/mL.

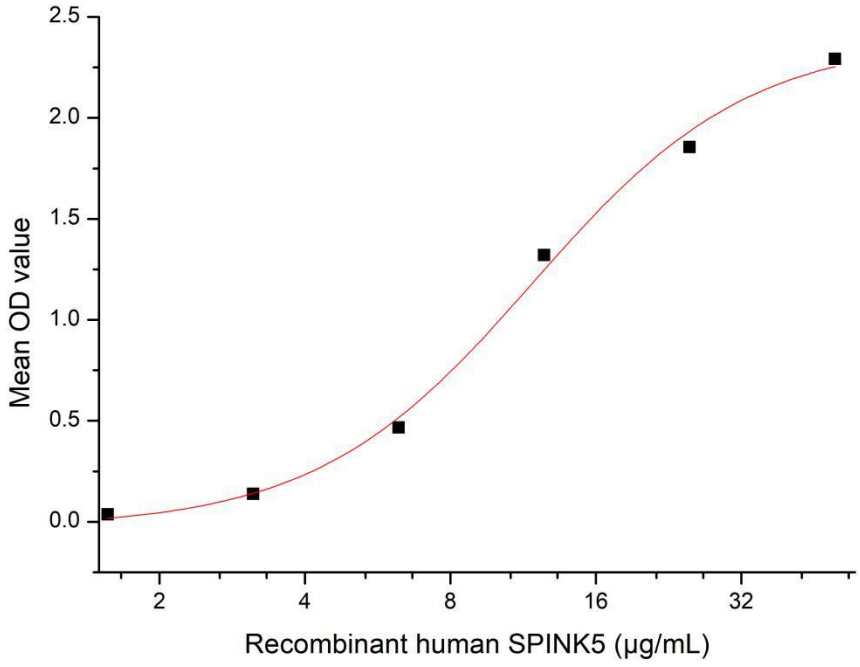


Figure 1. The binding activity of recombinant SPINK5 and FLG

[IDENTIFICATION]

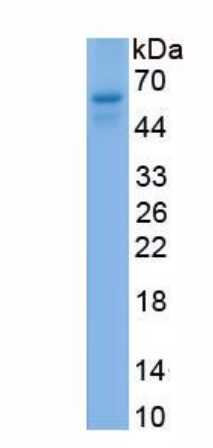


Figure 2. SDS-PAGE

Sample: Active recombinant FLG, 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.