APB039Hu61 100µg Active Surfactant Associated Protein D (SPD) 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: Ala21~Phe375 Tags: N-terminal His-tag Purity: >90% 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: 6.5

Predicted Molecular Mass: 37.1kDa

Accurate Molecular Mass: 44kDa 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.

Cloud-Clone Corp.

[<u>USAGE</u>]

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]

AEMKTYSHRT MPSACTLVMC SSVESGLPGR DGRDGREGPR GEKGDPGLPG AAGQAGMPGQ AGPVGPKGDN GSVGEPGPKG DTGPSGPPGP PGVPGPAGRE GPLGKQGNIG PQGKPGPKGE AGPKGEVGAP GMQGSAGARG LAGPKGERGV PGERGVPGNT GAAGSAGAMG PQGSPGARGP PGLKGDKGIP GDKGAKGESG LPDVASLRQQ VEALQGQVQH LQAAFSQYKK VELFPNGQSV GEKIFKTAGF VKPFTEAQLL CTQAGGQLAS PRSAAENAAL QQLVVAKNEA AFLSMTDSKT EGKFTYPTGE SLVYSNWAPG EPNDDGGSED CVEIFTNGKW NDRACGEKRL VVCEF

[ACTIVITY]

SP-D (surfactant protein-D) is a 43 kDa member of the collectin family of innate immune modulators. It is constitutively secreted by alveolar lining cells and epithelium associated with tubular structures. Its principal components consist of a collagen-like region and a C-terminal carbohydrate recognition domain (CRD), a structure that further places it in a subset of an expanded group of proteins termed defense collagens. SP-D is known to bind both SIRP alpha and the calreticulin/CD91 complex on macrophages. When the ratio of antigen/pathogen to available CRDs is low, antigen can be bound without occupying all available

Cloud-Clone Corp.

CRDs. The free CRDs will bind to SIRP alpha, generating a signal that downmodulates the inflammatory response. When virtually all CRDs are occupied by ligand, however, free CRDs are not available for SIRP alpha binding. The activity of the recombinant human SPD was measured by its ability to bind fluorescein-conjugated E. coli bioparticles. The rhSPD was diluted to 10 ug/ml in assay buffer of 20 mM Tris, 137 mM NaCl, 1 mM CaC12, pH7.4 and fluorescein-conjugated E. coli was diluted to 7*108 cells/ml. Equal volume of 10 ug/ml rhSPD and fluorescein-conjugated E. coli was took onto the slide and observed under the fluorescence microscopy. The result was shown in figure 1, the rhSPD could bind fluorescein-conjugated E. coli bioparticles.



Figure 1. Fluorescence micrograph of FITC-labeled E. coli DH5a after incubation with SP-D. (A) FITC-labeled E. coli DH5a (7 x 10^8 cells/ml) were incubated with an equal volume of SP-D (10 ug/ml).

(B) FITC-labeled E. coli DH5a without treatment of SP-D.

Cloud-Clone Corp.

[IDENTIFICATION]

Figure 2. SDS-PAGE

Sample: Active recombinant SPD, Human

[<u>IMPORTANT NOTE</u>]

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.