

APA156Hu01 100µg
Active Active Carbohydrate Antigen 19-9 (CA19-9)
Organism Species: *Homo sapiens (Human)*
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Arg35~Thr361

Tags: N-terminal His-tag

Purity: >95%

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

Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 0.05% sarcosyl and 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: 9.0

Predicted Molecular Mass: 39.5kDa

Accurate Molecular Mass: 43kDa 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 20mM Tris, 150mM NaCl (pH8.0) 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]

```
RVSRDD ATGSPRAPSG
SSRQDTTPTR PTLILLWTW PFHIPVALSR CSEMVPGTAD CHITADRKVY
PQADTVIVHH WDIMSNPKSR LPPSPRPQQQ RWIWFNLEPP PNCQHLEALD
RYFNLTSYR SDSDIFTPYG WLEPWSGQPA HPPLNLSAKT ELVAWAVSNW
KPDSARVRY YQSLQAHKVD VYGRSHKPLP KGTMMETLSR YKFYLAFENS
LHPDYITEKL WRNALEAWAV PVVLGSPRSN YERFLPPDAF IHVDDFQSPK
DLARYLQELD KDHARYLSYF RWRETLRPRS FSWALDFCKA CWKLQQESRY
QTVRSIAAWF T
```

[ACTIVITY]

Galactoside 3 (4) -L-fucosyltransferase also known as Carbohydrate Antigen 19-9 (CA19-9) is an enzyme that in humans is encoded by the FUT3 gene. This gene is a member of the fucosyltransferase family, which catalyzes the addition of fucose to precursor polysaccharides in the last step of Lewis antigen biosynthesis. It encodes an enzyme with alpha (1,3) fucosyltransferase and alpha (1,4)-fucosyltransferase activities. Mutations in this gene are responsible for the majority

of Lewis antigen-negative phenotypes. Besides, Lipase maturation factor 2 (LMF2) has been identified as an interactor of CA19-9, thus a binding ELISA assay was conducted to detect the interaction of recombinant human CA19-9 and recombinant human LMF2. Briefly, CA19-9 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100 μ L were then transferred to LMF2-coated microtiter wells and incubated for 2h at 37 $^{\circ}$ C. Wells were washed with PBST and incubated for 1h with anti-CA19-9 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 $^{\circ}$ C. Finally, add 50 μ L stop solution to the wells and read at 450nm immediately. The binding activity of CA19-9 and LMF2 was shown in Figure 1, and this effect was in a dose dependent manner.

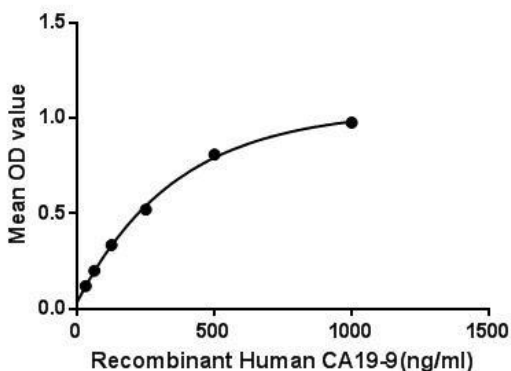


Figure 1. The binding activity of CA19-9 with LMF2.

[IDENTIFICATION]

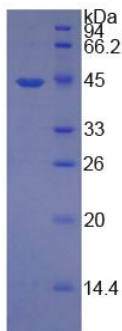


Figure 2. SDS-PAGE

Sample: Active recombinant CA19-9, Human

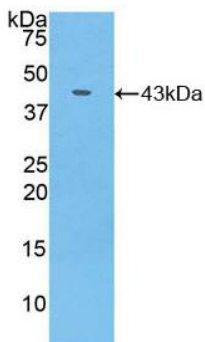


Figure 3. Western Blot

Sample: Recombinant CA19-9, Human;

Antibody: Rabbit Anti-Human CA19-9 Ab (PAA156Hu01)

[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.