

**APA309Hu01 200µg**

**Active Galectin 9 (GAL9)**

**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:** Met1~Thr323

**Tags:** N-terminal His-Tag.

**Purity:** >90%

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

**Buffer Formulation:** 20mM Tris, 150mM NaCl, pH8.0, containing 0.01% sarcosyl, 5%Trehalose.

**Applications:** Cell culture; Activity Assays; In vivo assays.

(May be suitable for use in other assays to be determined by the end user.)

**Predicted isoelectric point:** 8.3

**Predicted Molecular Mass:** 39.6kDa

**Accurate Molecular Mass:** 40kDa 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.

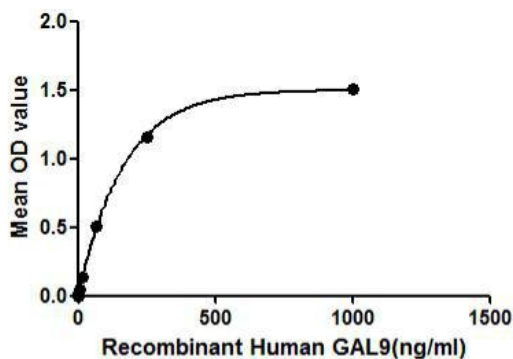
## [ SEQUENCE ]

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MAFSGSQAPY LSPAVPFSGT IQGGLQDGLQ ITVNGTVLSS SGTRFAVNFG  
TGFSGNDIAF HFNPRFEDGG YVVCNTRQNG SWGPEERKTH MPFQKGMPFD  
LCFLVQSSDF KVMVNGILFV QYFHRVPFHR VDTISVNGSV QLSYISFQPP  
GVWPANPAPI TQTVIHTVQS APGQMFSTPA IPPMMYPHPA YPMPFITIL  
GGLYPSKSIL LSGTVLPSAQ RFHINLCSGN HIAFHLNPRF DENAVVRNTQ  
IDNSWGSEER SLPRKMFPVR QQSFSVWILC EAHCLKVAVD GQHLFEYYHR  
LRNLPTINRL EVGGDIQLTH VQT
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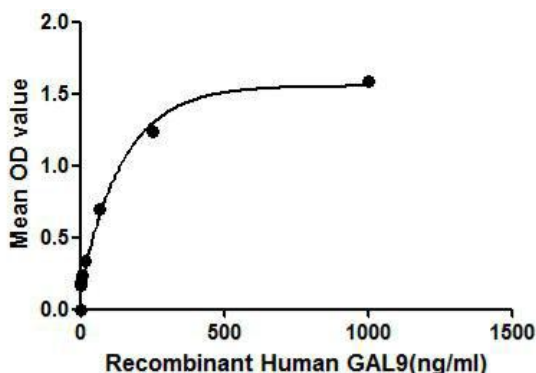
## [ ACTIVITY ]

GAL9 (Galectin-9) belongs to the galectin family, which is defined by their binding specificity for  $\beta$ -galactoside sugars, such as N-acetyllactosamine (Gal $\beta$ 1-3GlcNAc or Gal $\beta$ 1-4GlcNAc). It is reported that GAL9 induces T-helper type 1 lymphocyte (Th1) death by binding to HAVCR2 (Hepatitis A virus cellular receptor 2); besides, the interaction between GAL9 and PDI (Protein disulfide-isomerase) leads to disulfide reductase activity increasing at the plasma membrane, therefore alters the plasma membrane redox state and enhances cell migration. Thus a binding

ELISA assay was conducted to detect the interaction of recombinant human GAL9 with recombinant human HAVCR2 and recombinant human PDI separately. Briefly, GAL9 were diluted serially in PBS, with 0.01%BSA (pH 7.4). Duplicate samples of 100uL were then transferred to HAVCR2-coated and PDI-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-GAL9 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°C. Finally, add 50μL stop solution to the wells and read at 450nm immediately. The binding activity of GAL9 with HAVCR2 and PDI were shown in Figure 1 and Figure 2, and this effect was in a dose dependent manner.

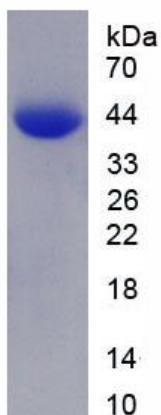


**Figure 1. The binding activity of GAL9 with HAVCR2.**



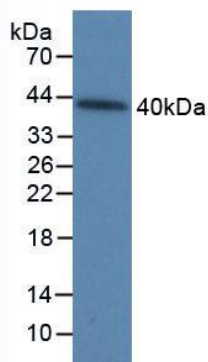
**Figure 2. The binding activity of GAL9 with PDI.**

## **[ IDENTIFICATION ]**



**Figure 3. SDS-PAGE**

**Sample: Active recombinant GAL9, Human**



**Figure 4. Western Blot**

**Sample: Recombinant GAL9, Human;**

**Antibody: Rabbit Anti-Human GAL9 Ab (PAA309Hu01)**

## **[ 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