

**APG886Hu01 10µg**  
**Active Cold Inducible RNA Binding Protein (CIRBP)**  
**Organism Species: Homo sapiens (Human)**  
***Instruction manual***

FOR IN VITRO USE AND RESEARCH USE ONLY  
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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1th Edition (Apr, 2016)

## **[ PROPERTIES ]**

**Source:** Prokaryotic expression.

**Host:** *E. coli*

**Residues:** Met1~Glu172

**Tags:** N-terminal His-tag

**Purity:** >95%

**Buffer Formulation:** 20mM Tris, 150mM NaCl, pH8.0, containing 0.01% sarcosyl and 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:** 9.7

**Predicted Molecular Mass:** 21.7kDa

**Accurate Molecular Mass:** 22&27kDa 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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MASDEGKLFV  GGLSFDTNEQ  SLEQVFSKYG  QISEVVVVKD  RETQRSRGFG  
FVTFENIDDA  KDAMMAMNGK  SVDGRQIRVD  QAGKSSDNRS  RGYRGGGSAGG  
RGFFRGGGRGR  GRGFSRGGGD  RGYGGNRFES  RSGGYGGSRD  YYSSRSQSGG  
YSDRSSGGSY  RDSYDSYATH  NE
```

## **[ ACTIVITY ]**

CIRBP (Cold-inducible RNA-binding protein) is considered to play a protective role in the genotoxic stress response by stabilizing transcripts of genes involved in cell survival and act as a translational activator. Besides, ATXN1 (Ataxin-1) has been proven as an interactor of CIRBP. Thus a binding ELISA assay was conducted to detect the interaction of recombinant human CIRBP and recombinant human ATXN1. Briefly, CIRBP were diluted serially in PBS, with 0.01%BSA (pH 7.4). Duplicate samples of 100uL were then transferred to ATXN1-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-CIRBP mAb, 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 CIRBP and ATXN1 was shown in Figure 1, and this effect was in a dose dependent manner.

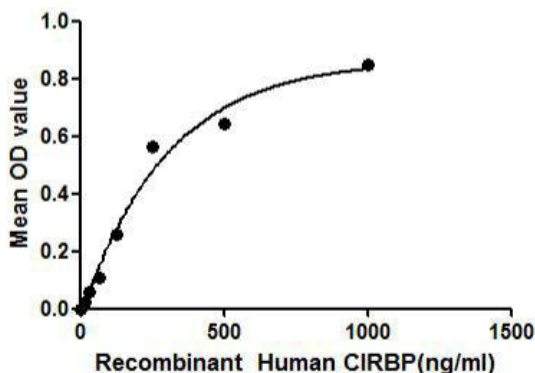


Figure 1. The binding activity of CIRBP with ATXN1.

## [ IDENTIFICATION ]

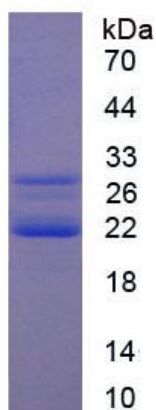
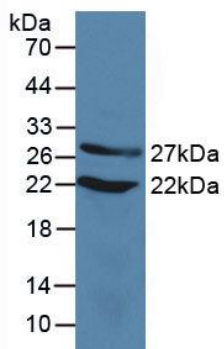


Figure 2. SDS-PAGE

Sample: Active recombinant CIRBP, Human



**Figure 3. Western Blot**

**Sample: Recombinant CIRBP, Human;**

**Antibody: Rabbit Anti-Human CIRBP Ab (PAG886Hu01)**