

APG886Mu01 100µg Active Cold Inducible RNA Binding Protein (CIRBP) Organism Species: Mus musculus (Mouse) 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~Glu172
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: 9.6

Predicted Molecular Mass: 22.3kDa

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]

MASDEGKLFV GGLSFDTNEQ ALEQVFSKYG QISEVVVVKD RETQRSRGFG FVTFENIDDA KDAMMAMNGK SVDGRQIRVD QAGKSSDNRS RGYRGGSAGG RGFFRGGRSR GRGFSRGGGD RGYGGGRFES RSGGYGGSRD YYASRSQGGS YGYRSSGGSY RDSYDSYATH NE

[ACTIVITY]

Cold-inducible RNA-binding protein (CIRBP) plays a critical role in controlling the cellular response upon confronting a variety of cellular stresses, including short wavelength ultraviolet light, hypoxia, and hypothermia. Besides, Heterogeneous Nuclear Ribonucleoprotein A1 (HNRPA1) has been identified as an interactor of CIRBP, thus a binding ELISA assay was conducted to detect the interaction of recombinant mouse CIRBP and recombinant mouse HNRPA1. Briefly, CIRBP were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100µL were then transferred to HNRPA1-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-CIRBP pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary

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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 50μ L stop solution to the wells and read at 450nm immediately. The binding activity of CIRBP and HNRPA1 was shown in Figure 1, and this effect was in a dose dependent manner.

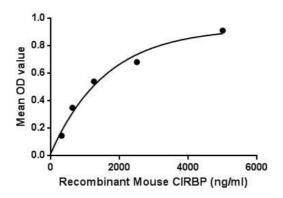


Figure 1. The binding activity of CIRBP with HNRPA1.

[IDENTIFICATION]

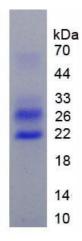


Figure 2. SDS-PAGE

Sample: Active recombinant CIRBP, Mouse

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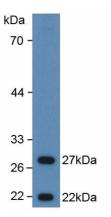


Figure 3. Western Blot

Sample: Recombinant CIRBP, Mouse;

Antibody: Rabbit Anti-Mouse CIRBP Ab (PAG886Mu01)

[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.