APA567Hu01 100µg Active S100 Calcium Binding Protein B (S100B) Organism Species: *Homo sapiens* (Human) *Instruction manual*

FOR RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1th Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression. Host: *E. coli* Residues: Met1~Glu92 Tags: N-terminal His-tag Purity: >92% 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: 5.3 Predicted Molecular Mass: 13.5kDa

Accurate Molecular Mass: 14kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

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.

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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.

[<u>SEQUENCE</u>]

MSELEKAMVA LIDVFHQYSG REGDKHKLKK SELKELINNE LSHFLEEIKE QEVVDKVMET LDNDGDGECD FQEFMAFVAM VTTACHEFFE HE

[ACTIVITY]

Protein S100B is a member of the S100 family. S100 proteins are EF-hand calcium-binding proteins and involved in the regulation of a number of cellular processes such as cell cycle progression and differentiation. Experimental results suggest that the receptor for advanced glycation end products (RAGE) plays important roles in mediating S100 protein-induced cellular signaling. Thus a binding ELISA assay was conducted to detect the interaction of recombinant human S100B and recombinant human RAGE. Briefly, S100B were diluted serially in PBS, with 0.01%BSA (pH 7.4). Duplicate samples of 100uL were then transferred to RAGE-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-S100B 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 of S100B and RAGE was shown in Figure 1, and this effect was in a dose dependent manner.

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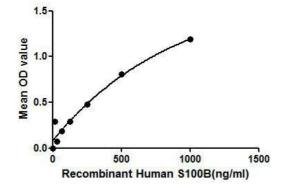


Figure 1. The binding activity of S100B with RAGE.

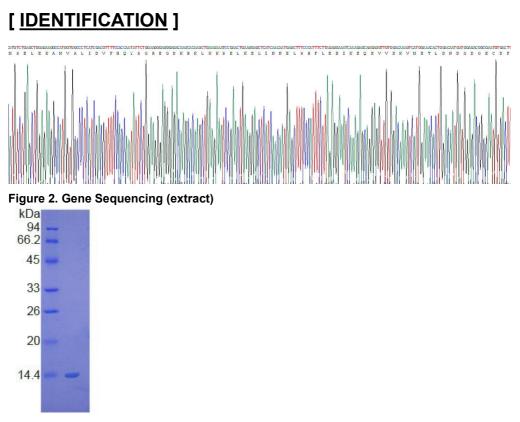


Figure 3. SDS-PAGE Sample: Active recombinant S100B, Human

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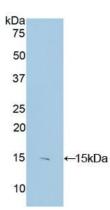


Figure 4. Western Blot

Sample: Recombinant S100B, Human;

Antibody: Rabbit Anti-Human S100B Ab (PAA567Hu01)

[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.