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APA998Ra01 100µg Active Glypican 4 (GPC4) Organism Species: *Rattus norvegicus (Rat) Instruction manual*

FOR RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Prokaryotic expression. Host: *E. coli* Residues: Ala222~Leu468 Tags: N-terminal His-tag Purity: >90% Endotoxin Level: <1.0EU per 1µg (determined by the LAL method). Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 5%Trehalose . Original Concentration: 200µg/mL Applications: Cell culture; Activity Assays. (May be suitable for use in other assays to be determined by the end user.) Predicted isoelectric point: 7.1 Predicted Molecular Mass: 28.5kDa Accurate Molecular Mass: 27kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

Reconstitute in 10mM PBS (pH7.4) 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.

[SEQUENCE]

AQGLAVARD VVNRVSVVNP TAQCTQALLK MIYCSHCRGL VTVKPCYNYC SNIMRGCLAN QGDLDLEWNN FIDAMLMVAE RLEGPFNIES VMDPIDVKIS DAIMNMQDNS VQVSQKVFQG CGPPKPLPAG RISRSISESA FSARFRPYHP EQRPTTAAGT SLDRLVTDVK EKLKQAKKFW SSLPSTVCND ERMAAGNENE DDCWNGKSKS RYLFAVTGNG LANQGNNPEV QVDTSKPDIL ILRQIMAL

[ACTIVITY]

Glypican 4 (GPC4) is a cell surface heparan sulfate proteoglycans which composed of a membrane-associated protein core substituted with a variable number of heparan sulfate chains. Members of the glypican-related integral membrane proteoglycan family (GRIPS) contain a core protein anchored to the cytoplasmic membrane via a glycosyl phosphatidylinositol linkage. These proteins may play a role in the control of cell division and growth regulation. Besides, Fibroblast Growth Factor 2, Basic (FGF2) has been identified as an interactor of GPC4, thus a functional binding ELISA assay was conducted to detect the interaction of recombinant rat GPC4 and recombinant human FGF2. Briefly, GPC4 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100 μ I were then transferred to FGF2-coated microtiter wells and incubated for 1h at 37 °C. Wells were washed with PBST and incubated for 1h with anti-GPC4 pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated and washed 5 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 450 nm immediately. The binding activity of recombinant rat GPC4 and recombinant human FGF2 was shown in Figure 1, the

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EC50 for this effect is 2.64 ug/mL.



Figure 1. The binding activity of recombinant rat GPC4 and recombinant human FGF2

[IDENTIFICATION]

Figure 2. SDS-PAGE

Sample: Active recombinant GPC4, Rat

[IMPORTANT NOTE]

The kit is designed for research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.