

Rat SFRP1 protein, His tag (active)

Cat No. GTX00065-pro**Application** Functional Assay**Species** Rat**Package**

10 µg

APPLICATION

Application Note

Secreted frizzled-related protein 1, also known as SFRP1 is a member of the SFRP family that contains a cysteine-rich domain homologous to the putative Wnt-binding site of Frizzled proteins. It acts as a biphasic modulator of Wnt signaling, counteracting Wnt-induced effects at high concentrations and promoting them at lower concentrations. As a tumor suppressor, SFRP1 expression markedly inhibited tumor cell growth in culture, soft agar and xenografts in athymic nude mice. Besides, Wingless Type MMTV Integration Site Family, Member 2 (WNT2) has been identified as an interactor of SFRP1, thus a binding ELISA assay was conducted to detect the interaction of recombinant rat SFRP1 and recombinant rat WNT2. Briefly, SFRP1 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100 µl were then transferred to WNT2-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-SFRP1 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 50 µl stop solution to the wells and read at 450nm immediately. The binding activity of SFRP1 and WNT2 was in a dose dependent manner.

To measure the ability of SFRP1 on tumor suppression, liver cancer HepG2 cells were seeded into triplicate wells of 96-well plates at a density of 5000 cells/well and allowed to attach, replaced with serum-free overnight, then the medium was replaced with 2% serum standard DMEM prior to the addition of various concentrations of recombinant rat SFRP1. After incubated for 96h, cells were observed by inverted microscope and cell proliferation was measured by Cell Counting Kit-8 (CCK-8). Briefly, 10 µl of CCK-8 solution was added to each well of the plate, then the absorbance at 450nm was measured using a microplate reader after incubating the plate for 1-4 hours at 37°C. Suppression of HepG2 cells after incubation with SFRP1 for 96h observed by inverted microscope. Cell viability was assessed by CCK-8 assay after incubation with recombinant SFRP1 for 96h. And SFRP1 significantly decreased cell viability of HepG2 cells.

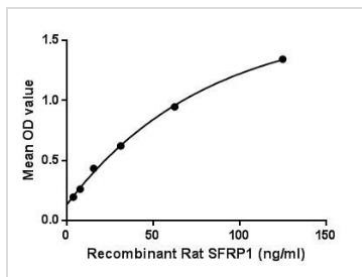
Observed MW 18 kDa.

PROPERTIES

Form Lyophilized powder**Buffer** Lyophilized from 20 mM Tris (pH 8.0) with 150 mM NaCl, 1 mM EDTA, 1 mM DTT, 0.01% SKL, 5% Trehalose, Proclin300. Reconstitute with 20 mM Tris and 150 mM NaCl (pH 8.0) to a concentration of 0.1-1.0 mg/mL. Do not vortex.**Storage** For short-term storage (1-2 weeks), store at 4°C. For long-term storage, store at -20°C or below. After reconstitution, keep as concentrated solution. Avoid freeze-thaw cycles.**Region/Sequence** N-terminal His-Tag; Val8~Asp153**Expression System** E. coli**Purity** > 95%**Endotoxin** < 1 EU/µg**Conjugation** Unconjugated**Note** For laboratory use only. Not for any clinical, therapeutic, or diagnostic use in humans or animals. Not for animal or human consumption.

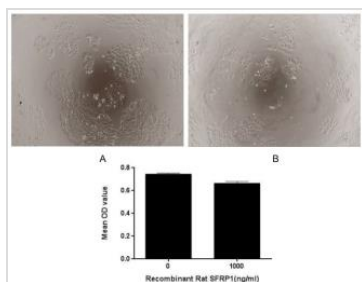
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DATA IMAGES



GTX00065-pro Functional Assay Image

Functional ELISA analysis of GTX00065-pro Rat SFRP1 protein (active) which can bind immobilized WNT2 protein.

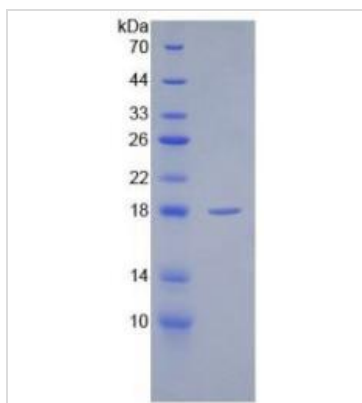


GTX00065-pro Functional Assay Image

Cell suppression effect of GTX00065-pro Rat SFRP1 protein (active). Cell viability was measured by Cell Counting Kit-8 (CCK-8).

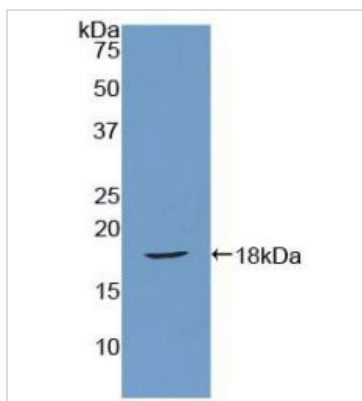
(A) Unstimulated HepG2 cells cultured in DMEM for 96hrs.

(B) HepG2 cells cultured in DMEM, stimulated with 1000 ng/ml SFRP1 for 96hrs.



GTX00065-pro Image

SDS-PAGE analysis of GTX00065-pro Rat SFRP1 protein (active).



GTX00065-pro Image

WB analysis of GTX00065-pro Rat SFRP1 protein (active).



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