

APG669Hu01 100μg

Active Chloride Channel Accessory 1 (CLCA1)

Organism Species: Homo sapiens (Human)

Instruction manual

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

1th Edition (Apr., 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: E. coli

Residues: Asn416~Arg670
Tags: N-terminal His-tag

Purity: >98%

Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 0.05% 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: 7.1

Predicted Molecular Mass: 28.4kDa

Accurate Molecular Mass: 28kDa as determined by SDS-PAGE reducing conditions.

[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]

NTISG CFNEVKQSGA IIHTVALGPS AAQELEELSK
MTGGLQTYAS DQVQNNGLID AFGALSSGNG AVSQRSIQLE SKGLTLQNSQ
WMNGTVIVDS TVGKDTLFLI TWTMQPPQIL LWDPSGQKQG GFVVDKNTKM
AYLQIPGIAK VGTWKYSLQA SSQTLTLTVT SRASNATLPP ITVTSKTNKD
TSKFPSPLVV YANIRQGASP ILRASVTALI ESVNGKTVTL ELLDNGAGAD
ATKDDGVYSR YFTTYDTNGR

[ACTIVITY]

CLCA1 (Calcium-activated chloride channel regulator 1) is a member of the calcium sensitive chloride conductance protein family. This protein is expressed as a precursor protein that is processed into two cell-surface-associated subunits. It has been reported that CLCA1 activates calcium-dependent chloride channel through the interaction with TMEM16A (anoctamin-1). Besides, there exits similarities between human and mouse TMEM16A in amino acid sequence with the identity of 89.66%. Thus, a functional ELISA assay was conducted to detect the association of recombinant human CLCA1 with recombinant mouse TMEM16A. Briefly, CLCA1 were diluted serially in PBS with 0.01%BSA (pH 7.4). Duplicate samples of 100uL were then transferred to TMEM16A-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-CLCA1 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 50µL stop solution to the wells and read at 450nm immediately. The binding

activity of CLCA1 with TMEM16A was shown in Figure 1 and this effect was in a dose dependent manner.

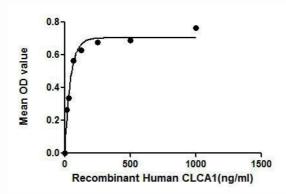


Figure 1. The binding activity of CLCA1 with TMEM16A

[IDENTIFICATION]

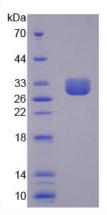


Figure 2. SDS-PAGE

Sample: Active recombinant CLCA1, Human

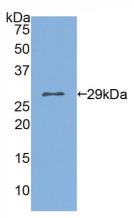


Figure 3. Western Blot

Sample: Recombinant CLCA1, Human;

Antibody: Rabbit Anti-Human CLCA1 Ab (PAG669Hu01)