ALPHA DIAGNOSTIC INTERNATIONAL

Product Specification Sheet

Chloride Channel-K1/K2 (CLC-K1/K2 or CLCN-KA/Kb) Antibodies

Cat. # CLCK11-P	Rat CLC-K1 Control Peptide	**SIZE**: 100 ug
Cat. # CLCK11-S	Rabbit Anti-rat CLC-K1 antiserum	**SIZE**: 100 ul
Cat. # CLCK11-A	Rabbit Anti-rat CLC-K1 IgG (aff pure)	**SIZE**: 100 ug

Chloride is a critical component of all living cells. Voltage-gated chloride channels regulate cellular traffic of chloride ion. The chloride channels (CIC or CLC) performs several functions including the regulation of cell volume, membrane potential stabilization, signal transduction, and transepithelial transport. Mutations in CIC genes have been linked with several human diseases including myotonias (Thomsen's disease), cystic fibrosis, Bartters syndrome type III, Dent's disease, and X-linked recessive nephrolithiasis. In mammals, CLC proteins form a superfamily of at least 9 different genes (CLC1-7 also known as CLCN1-7 and CLK1-2 or CLCKa and CLCKb). Additional forms of these proteins are obtained by alternative splicing. All CLC proteins (~700-1000 aa) are predicted to contain 10 (possibly 12) transmembrane domains. Except CLC-1 and CLC-K1/K2 that are specific for kidney, most other CLC are widely distributed in various tissues.

Rat CLC-K1 is 687 aa membrane protein (human 687 aa) (1). CLC-K1 is ~84% identical with CLC-K2 (2). CLC-K1 is specific for kidney (cortical thick ascending limb and the distal convoluted tubule with minor expression in the S3 segment of the proximal tubule and the cortical collecting tubule). It may play a role in urine concentrating mechanism.

Source of Antigen and Antibodies

Antigen	17-aa peptide of rat CLC-K1; **Designated (CLCK11-P or control peptide /blocking peptide**) conjugated to KLH; epitope location ~C-terminus, extracellular domain
Ab Host/type	Rabbit, Polyclonal unpurified antiserum (#**CLCK11**-S) and IgG, purified over antigen-agarose (Cat # **CLCK11**-A)
2-Ab	Cat # 20320, goat anti-rabbit IgG-HRP (AP, biotin, FITC conjugates also available).
-ve control IgG	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as –ve control

Form & Storage of Antibodies/Peptide Control

Antiserum (unpurified)
100ul solution lyophilized powder
Supplied 0.05% azide, **Reconstitute** powder in 100 ul PBS

Affinity pure IgG
100 ug/100ul solution lyophilized powder
Supplied in **Buffer**: PBS+0.1% BSA
Reconstitute powder in PBS at 1mg/ml

Control/blocking peptide
100 ug/100 ul solution lyophilized powder
Supplied in Buffer: PBS pH 7.5,
Reconstitute powder in PBS at 1 mg/ml.

Storage
Short-term: unopened, undiluted liquid vials at -20OC and powder at 4oC or -20oC..
Long-term: at –20C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

Stability: 6-12 months at –20oC or below.
Shipping: 4oC for solutions and room temp for powder

Recommended Usage

Western Blotting (1:1K-5K for neat serum and 1-10 ug/ml for affinity pure antibody using ECL technique). Antibodies made to CLC-K1 C-terminal regions has detected proteins of ~70 kDa in kidney (2).

ELISA: Control peptide can be used to coat ELISA plates at 1 ug/ml and detected with antibodies (1:10-50K for neat serum and 0.5-1 ug/ml for affinity pure).

Histochemistry & Immunofluorescence: Not tested. We recommend the use of affinity purified antibody at 1-20 ug/ml in paraformaldehyde fixed sections of tissues. CLC-k1 has been localized to the thin limb of Henle's loop in rat inner medulla (2).

Specificity & Cross-reactivity

Rat CLCK11-P control peptide is 100% conserved in mouse, 93% in human CLC-K1 or CLC-Ka. It also has high homology with rat CLC-K2 and 81% with human CLC-K2. Anti-rat CLCK11 should crossreact with both CLC-K1 and CLC-K2. No significant sequence homology is detected with other CLCs or other proteins. Antibody crossreactivity in various other species is not established. Control peptide, because of its low mol. Wt (<3 kDa), is not suitable for Western. It should be used for ELISA or antibody blocking experiments (use 5-10 ug control peptide per 1 ug of aff pure IgG or 1 ul antiserum) to confirm antibody specificity (see detailed protocol at the web site).).

General References: Uchida S et al (1993) JBC 268, 3821-3824; Uchida S et al (1994) JBC 269, 19192; Uchida S et al (1998) Am. J. Physiol. 274, F602; Keifferle S et al (1994) PNAS 91, 6943-6947, Adachi S et al (1994) JBC 269, 17677-17683; Uchida S et al (1995) J Clin. Invest. 95, 104-113.

*This product is for In vitro research use only.

Related material available from ADI

Antibodies CLC1-7 and CLC-K1; KCCL1-3; AQP-9 and RUT; OCT and OAT, AE-3, and NACX

CLCK11-S 50415A

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