Revision 1

#8351 Store at -20C

NKCC1 (D13A9) Rabbit mAb

Cell Signaling TECHNOLOGY®

Orders: 877-616-CELL (2355)
orders@cellsignal.com

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Web: info@cellsignal.com
cellsignal.com

3 Trask Lane | Danvers | Massachusetts | 01923 | USA

For Research Use Only. Not for Use in Diagnostic Procedures.

Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
W, IP, IF-IC	H	Endogenous	160-200	Rabbit IgG	#P55011	6558

Product Usage Information

Application	Dilution
Western Blotting	1:2000
Immunoprecipitation	1:50
Immunofluorescence (Immunocytochemistry)	1:100

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. Do not aliquot the antibody.

Specificity/Sensitivity

NKCC1 (D13A9) Rabbit mAb recognizes endogenous levels of total NKCC1 protein. This antibody does not cross-react with NKCC2.

Species predicted to react based on 100% sequence homology

Bovine, Pig

Source / Purification

Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Arg80 of human NKCC1 protein.

Background

The electroneutral cation-chloride-coupled co-transporter (SLC12) gene family comprises bumetanide-sensitive $Na^+/K^+/Cl^-$ (NKCC), thiazide-sensitive Na^+/Cl^-, and K^+/Cl^- (KCC) co-transporters. SLC12A1/NKCC2 and SLC12A2/NKCC1 regulate cell volume and maintain cellular homeostasis in response to osmotic and oxidative stress (1). The broadly expressed NKCC1 is thought to play roles in fluid secretion (i.e. salivary gland function), salt balance (i.e. maintenance of renin and aldosterone levels), and neuronal development and signaling (2-7). During neuronal development, NKCC1 and KCC2 maintain a fine balance between chloride influx (NKCC1) and efflux (KCC2), which regulates γ-aminobutyric acid (GABA)-mediated neurotransmission (3). Increased NKCC1 expression in immature neurons maintains high intracellular chloride levels that result in inhibitory GABAergic signaling; KCC2 maintains low intracellular chloride levels and excitatory GABAergic responses in mature neurons (4,5,8). Deletion of NKCC1 impairs NGF-mediated neurite outgrowth in PC-12D cells while inhibition of NKCC1 with bumetanide inhibits re-growth of axotomized dorsal root ganglion cells (6,7). Defective chloride homeostasis in neurons is linked to seizure disorders that are ameliorated by butemanide treatment, indicating that abnormal NKCC1 and NKCC2 expression or signaling may play a role in neonatal and adult seizures (9-12). NKCC1 is found as a homodimer or within heterooligomers with other SLC12 family members. This transport protein associates with a number of oxidative- and osmotic-responsive kinases that bind, phosphorylate, and activate NKCC1 co-transporter activity (13-16). In response to decreased intracellular chloride concentrations, Ste20-related proline-alanine-rich kinase (SPAK) phosphorylates NKCC1 to increase co-transporter activity and promote chloride influx (16-19). Oxidative stress response kinase 1 (OSR1) also phosphorylates and activates NKCC1 in response to oxidative stress (14).

Background References

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Species Reactivity

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

Western Blot Buffer

IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v nonfat dry milk, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

Applications Key

W: Western Blotting **IP:** Immunoprecipitation **IF-IC:** Immunofluorescence (Immunocytochemistry)

Cross-Reactivity Key

H: Human

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Limited Uses