Revision 1

Store at -20C

#8575

Calcium Ion Regulation Antibody Sampler Kit

1 Kit (6 x 20 microliters)

Cell Signaling TECHNOLOGY®

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

Product Includes	Product #	Quantity	Mol. Wt	Isotype/Source
ATP2A2/SERCA2 (D51B11) Rabbit mAb	9580	20 µl	114, 140 kDa	Rabbit IgG
Phospho-Phospholamban (Ser16/Thr17) Antibody	8496	20 µl	6 (monomer); 12, 24 (oligomers) kDa	Rabbit
Phospholamban (D9W8M) Rabbit mAb	14562	20 µl	12, 24 kDa	Rabbit IgG
Phospho-PKA C (Thr197) (D45D3) Rabbit mAb	5661	20 µl	42 kDa	Rabbit IgG
PKA C-α (D38C6) Rabbit mAb	5842	20 µl	42 kDa	Rabbit IgG
ATP2A1/SERCA1 (D54G12) Rabbit mAb	12293	20 µl	100 kDa	Rabbit IgG
Anti-rabbit IgG, HRP-linked Antibody	7074	100 µl		Goat

Please visit cellsignal.com for individual component applications, species cross-reactivity, dilutions, protocols, and additional product information.

Description

The Calcium Ion Regulation Antibody Sampler Kit provides an economical way to investigate the regulation of calcium ions within the cell. The kit contains enough primary and secondary antibodies to perform two western blot experiments per primary antibody.

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.

Background

Sarcoplasmic and endoplasmic reticulum Ca^{2+} ATPases (SERCA) are members of a highly conserved family of Ca^{2+} pumps (1). ATP2A1 (SERCA1) is a fast-twitch, skeletal muscle sarcoplasmic reticulum (SR) Ca^{2+} ATPase (2). Multiple ATP2A2 (SERCA2) isoforms have been isolated, with ATP2A2a (SERCA2a) found predominantly in the SR of muscle cells and ATP2A2b (SERCA2b) more ubiquitously expressed in the ER of most cell types (3). Post-translational modification of ATP2A2, including phosphorylation and tyrosine nitration, modify Ca^{2+} -ATPase activity and calcium transport (4,5).

Phospholamban (PLN) was identified as a major phosphoprotein component of the SR (6). Despite very high expression in cardiac tissue, phospholamban is also expressed in skeletal and smooth muscle (7). Localization of PLN is limited to the SR, where it serves as a regulator of the sarco-endoplasmic reticulum calcium ATPase, SERCA (8). PLN binds directly to SERCA and effectively lowers its affinity for calcium, thus reducing calcium transport into the SR. Phosphorylation of PLN at Ser16 by PKA or myotonic dystrophy protein kinase and/or phosphorylation at Thr17 by Ca^{2+}/calmodulin-dependent protein kinase results in release of PLN from SERCA, relief of this inhibition, and increased calcium uptake by SR (reviewed in 9,10). It has long been held that phosphorylation at Ser16 and Thr17 occurs sequentially, but increasing evidence suggests that phosphorylation, especially at Thr17, may be differentially regulated (reviewed in 11,12).

The second messenger cyclic AMP (cAMP) activates cAMP-dependent protein kinase (PKA or cAPK) in mammalian cells and controls many cellular mechanisms such as gene transcription, ion transport, and protein phosphorylation (13). Inactive PKA is a heterotetramer composed of a regulatory subunit (R) dimer and a catalytic subunit (C) dimer. In this inactive state, the pseudosubstrate sequences on the R subunits block the active sites on the C subunits. Three C subunit isoforms (C-α, C-β, and C-γ) and two families of the regulatory subunits (RI and RII) with distinct cAMP binding properties have been identified. Upon binding of cAMP to the R subunits, the auto-inhibitory contact is eased and active monomeric C subunits are released. PKA shares substrate specificity with Akt (PKB) and PKC, which are characterized by an arginine at position -3 relative to the phosphorylated serine or threonine residue (14). PKA phosphorylation is involved in the regulation of Ca^{2+} channels, including $Ca_v1.1$ in skeletal muscle and $Ca_v1.2$ in the heart (reviewed in 15).

Background References

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Limited Uses