Technical Data Sheet

Purified Mouse Anti-Calcineurin

Product Information

Material Number: 556350
Size: 0.1 mg
Concentration: 0.5 mg/ml
Clone: G182-1847
Immunogen: Human β1 isoform of calcineurin A Subunit aa. 349-505
Isotype: Mouse IgG1
Reactivity: QC Testing: Human
Target MW: 61 kDa
Storage Buffer: Aqueous buffered solution containing ≤0.09% sodium azide.

Description
Calcineurin is a Ca2+- and calmodulin-dependent serine/threonine phosphatase which consists of two subunits. The A subunit (~61 kDa) is catalytic and has calmodulin-binding activity; the B subunit (~19 kDa), which is myristylated at its N-terminus, has Ca2+-binding activity. Calcineurin is expressed ubiquitously in eukaryotic cells, including yeast. The protein is highly expressed in mammalian brain, and has been implicated in a number of calcium-dependent pathways in the nervous system, including regulation of neurotransmitter release. Calcineurin also plays a key role in T cell receptor-mediated signal transduction pathways. Calcineurin dephosphorylates components of NF-AT (Nuclear Factor of Activated T cells), allowing NF-AT to translocate to the nucleus where it participates with AP-1 in activation of multiple cytokine and surface receptor genes. Inhibition of calcineurin by immunosuppressive agents such as cyclosporin A and FK506 correlates with the suppression of early T cell activation events. These immunosuppressive agents bind to specific intracellular proteins, called immunophilins, to form complexes that inhibit calcineurin’s phosphatase activity. In the presence of these complexes, NF-AT remains phosphorylated and thus cannot translocate to the nucleus. A novel protein, Cain (calcineurin inhibitor), has been described which displays a similar pattern of neuronal and brain expression and has been shown to bind and inhibit calcineurin activity in vivo.

G182-1847 recognizes the α and β isoforms of the A subunit of calcineurin (~61 kDa). It reacts with an epitope in the autoinhibitory domain. It does not recognize the B subunit of calcineurin. A recombinant tag-fusion protein containing amino acids 349-505 of the human β1 isoform of calcineurin A subunit was used as immunogen.

Western blot analysis of calcineurin. Lysate from A-431 cells was probed with anti-calcineurin (clone G182-1847) at concentrations of 1.0 (lane 1), 0.2 (lane 2), and 0.04 µg/ml (lane 3). The 61 kDa A subunit of calcineurin is identified.

Preparation and Storage
The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at 4°C.

Application Notes

<table>
<thead>
<tr>
<th>Application</th>
<th>Status</th>
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<tbody>
<tr>
<td>Western blot</td>
<td>Routinely Tested</td>
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<tr>
<td>Immunoprecipitation</td>
<td>Tested During Development</td>
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Recommended Assay Procedure:
Applications include immunoprecipitation (1-2 µg/ml) and western blot analysis (1-2 µg/ml). Jurkat human T cells (ATCC TIB-152), HeLa human carcinoma cells (ATCC CCL-2), and A-431 human epidermal carcinoma cells (ATCC CRL-1555) are suggested as positive controls. A431 cell lysate (Cat. No. 611447) and HeLa cell lysate (Cat. No. 611449) are also available as ready-to-use positive western blot control.

Suggested Companion Products

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<tr>
<th>Catalog Number</th>
<th>Name</th>
<th>Size</th>
<th>Clone</th>
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<tbody>
<tr>
<td>554002</td>
<td>HRP Goat Anti-Mouse Ig</td>
<td>1.0 ml</td>
<td>(none)</td>
</tr>
<tr>
<td>611447</td>
<td>A431 Cell Lysate</td>
<td>500 µg</td>
<td>(none)</td>
</tr>
<tr>
<td>611449</td>
<td>HeLa Cell Lysate</td>
<td>500 µg</td>
<td>(none)</td>
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Product Notices
1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

References
Fruman DA, Klee CB, Bierer BE, Burakoff SJ. Calcineurin phosphatase activity in T lymphocytes is inhibited by FK 506 and cyclosporin A. Proc Natl Acad Sci U S A. 1992; 89(9):3686-3690. (Biology)