

DESCRIPTION

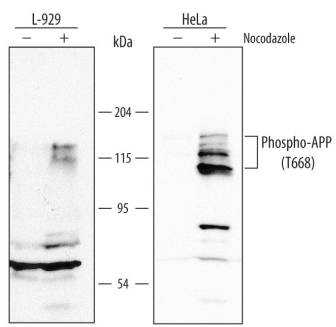
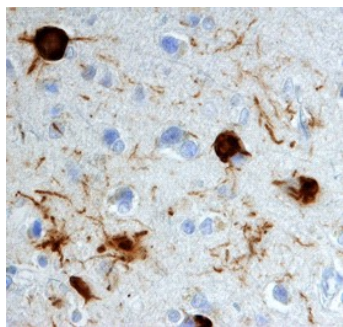
Species Reactivity	Human/Mouse
Specificity	Detects multiple isoforms of human and mouse APP/Protease Nexin II when phosphorylated at sites corresponding to T668 of the human APP695 isoform.
Source	Polyclonal Rabbit IgG
Purification	Antigen Affinity-purified
Immunogen	Phosphopeptide containing the human APP/Protease Nexin II T668 site
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Western Blot	0.5 µg/mL	See Below
Immunohistochemistry	5-15 µg/mL	See Below

DATA

<p>Western Blot</p>  <p>Detection of Human and Mouse Phospho-APP/Protease Nexin II (T668) by Western Blot. Western blot shows lysates of L-929 mouse fibroblast cell line and HeLa human cervical epithelial carcinoma cell line untreated (-) or treated (+) with 1 µg/mL nocodazole for 16 hours. PVDF membrane was probed with 0.5 µg/mL of Rabbit Anti-Human/Mouse Phospho-APP/Protease Nexin II (T668) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2508), followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.</p>	<p>Immunohistochemistry</p>  <p>Phospho-APP/Protease Nexin II (T668) in Human Brain. APP/Protease Nexin II phosphorylated at T668 was detected in immersion fixed paraffin-embedded sections of human Alzheimer's brain (cortex) using Rabbit Anti-Human/Mouse Phospho-APP/Protease Nexin II (T668) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2508) at 10 µg/mL overnight at 4 °C. Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using the Anti-Rabbit HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS005) and counterstained with hematoxylin (blue). View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.</p>
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PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Amyloid precursor protein (APP) is a type I membrane protein with several human isoforms due to alternative splicing. APP-770, -751, and -733 contain a Kunitz protease inhibitor (KPI) domain (residue 291-342) and APP-695 does not. APP is a cell surface molecule with many functions. It can be processed proteolytically in two different pathways. In one pathway, β- and γ-secretase cleave at the β site between residue 670 and 671 and the γ site between residue 711 and 714 to produce β-amyloid peptide (Aβ40 and Aβ42), a major component in plaques found in brains of patients with Alzheimer's disease (1). The other pathway involves α-secretase that cleaves residues between 687 and 688. It is anti-amyloidogenic due to its benign character and the prevention of the Aβ peptide formation (2). Soluble APP containing the KPI domain, also referred to as protease nexin II, is a potent inhibitor of serine proteases and may have additional functions. For example, it may regulate the contact face of blood coagulation and limit thrombosis specially in brain due to its localization and coagulation factor XI inhibiting activity (3, 4).

References:

1. Haass, C. (2004) EMBO J. **23**:483.
2. Lichtenthaler, S. F. and C. Haass (2004) J. Clin. Invest. **113**:1384.
3. Badellino, K.O. and P.N. Walsh (2000) Biochemistry **39**:4769.
4. Xu, F. *et al.* (2005) Proc. Natl. Acad. Sci USA. **102**:18135.