



Colorado Biosciences Park
12635 East Montview Boulevard, #213
Aurora, CO 80045-7337
Tel: (888) 442-7100

Anti-Phospho-Tyr¹⁴⁷² NMDA NR2B-Subunit

Catalog Number: p1516-1472 Size: 100 µl \$310.00

Product Description: Affinity purified rabbit polyclonal antibody

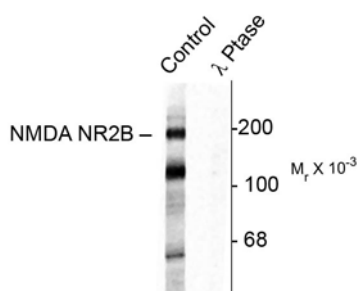
Applications: **WB:** 1:1000

Antigen: Phosphopeptide corresponding to amino acid residues surrounding the phospho-Tyr¹⁴⁷² of NMDA NR2B.

Species reactivity: The antibody has been directly tested for reactivity in Western blots with rat tissue. It is anticipated that the antibody will react with bovine, canine, chicken, human, mouse, non-human primate and zebra fish based on the fact that these species have 100% homology with the amino acid sequence used as antigen.

Biological Significance: The ion channels activated by glutamate that are sensitive to N-methyl-D-aspartate (NMDA) are designated NMDA receptors (NMDAR). The NMDAR plays an essential role in memory, neuronal development and it has also been implicated in several disorders of the central nervous system including Alzheimer's, epilepsy and ischemic neuronal cell death (Grosshans et al., 2002; Wenthold et al., 2003; Carroll and Zukin, 2002). The NMDA receptor is also one of the principal molecular targets for alcohol in the CNS (Lovinger et al., 1989; Alvestad et al., 2003; Snell et al., 1996). Channels with physiological characteristics are produced when the NR1 subunit is combined with one or more of the NMDAR2 (NR2 A-D) subunits (Ishii et al., 1993). Overexpression of the NR2B-subunit of the NMDA Receptor has been associated with increases in learning and memory while aged, memory impaired animals have deficiencies in NR2B expression (Clayton et al., 2002a; Clayton et al., 2002b). Recent work suggests that phosphorylation of Tyr¹⁴⁷² on NR2B may regulate the functional expression the receptor in LTP and other forms of plasticity (Nakazawa et al., 2001; Roche et al., 2001).

Anti-Phospho Tyr¹⁴⁷² NMDA NR2B-Subunit



Western blot of rat hippocampal lysate showing specific immunolabeling of the ~180k NR2B subunit of the NMDAR phosphorylated at Tyr¹⁴⁷² (Control). The phosphospecificity of this labeling is shown in the second lane (*lambda*-phosphatase: λ-Ptase). The blot is identical to the control except that it was incubated in λ-Ptase (1200 units for 30 min) before being exposed to the phospho-Tyr¹⁴⁷² NMDA NR2B antibody. The immunolabeling is completely eliminated by treatment with λ-Ptase.

Page 1 of 2

WB = Western Blot **IF** = Immunofluorescence **IHC** = Immunohistochemistry **IP** = Immunoprecipitation

Packaging: 100 µl in 10 mM HEPES (pH 7.5), 150 mM NaCl, 100 µg per ml BSA and 50% glycerol. Adequate amount of material to conduct 10-mini Western Blots.

Storage and Stability. For long term storage -20°C is recommended. Stable at -20°C for at least 1 year.

Shipment: Domestic - Blue Ice; International - Blue Ice or Dry Ice.

Purification Method: Prepared from rabbit serum by affinity purification via sequential chromatography on phospho- and dephosphopeptide affinity columns.

Antibody Specificity: Specific for the ~180k NMDAR NR2B-subunit protein phosphorylated at Tyr¹⁴⁷² in Western blots. The antibody also labels proteins of ~65k and ~115k. Immunolabeling is completely blocked by either λ -Ptase or by the phosphopeptide used as the antigen but not by the corresponding dephosphopeptide.

Quality Control Tests: Western blots performed on each lot.

References:

- Alvestad RM, Grosshans DR, Coultrap SJ, Nakazawa T, Yamamoto T, Browning MD (2003) Tyrosine dephosphorylation and ethanol inhibition of N-methyl-D-aspartate receptor function. *J Biol Chem* 278:11020-11025.
- Carroll RC, Zukin RS (2002) NMDA-receptor trafficking and targeting: implications for synaptic transmission and plasticity. *Trends Neurosci* 25:571-577.
- Clayton DA, Grosshans DR, Browning MD (2002a) Aging and surface expression of hippocampal NMDA receptors. *J Biol Chem* 277:14367-14369.
- Clayton DA, Mesches MH, Alvarez E, Bickford PC, Browning MD (2002b) A hippocampal NR2B deficit can mimic age-related changes in long-term potentiation and spatial learning in the Fischer 344 rat. *J Neurosci* 22:3628-3637.
- Grosshans DR, Clayton DA, Coultrap SJ, Browning MD (2002) LTP leads to rapid surface expression of NMDA but not AMPA receptors in adult rat CA1. *Nat Neurosci* 5:27-33.
- Ishii T, Moriyoshi K, Sugihara H, Sakurada K, Kadotani H, Yokoi M, Akazawa C, Shigemoto R, Mizuno N, Masu M, Nakanishi S (1993) Molecular characterization of the family of the N-methyl-D-aspartate receptor subunits. *J Biol Chem* 268:2836-2843.
- Lovinger DM, White G, Weight FF (1989) Ethanol inhibits NMDA-activated ion current in hippocampal neurons. *Science* 243:1721-1724.
- Nakazawa T, Komai S, Tezuka T, Hisatsune C, Umemori H, Semba K, Mishina M, Manabe T, Yamamoto T (2001) Characterization of Fyn-mediated tyrosine phosphorylation sites on the NR2B subunit of the N-methyl-D-aspartate receptor. *J Biol Chem* 276:693-699.
- Roche KW, Standley S, McCallum J, Dune LC, Ehlers MD, Wenthold RJ (2001) Molecular determinants of NMDA receptor internalization. *Nat Neurosci* 4:794-802.
- Snell LD, Nunley KR, Lickteig RL, Browning MD, Tabakoff B, Hoffman PL (1996) Regional and subunit specific changes in NMDA receptor mRNA and immunoreactivity in mouse brain following chronic ethanol ingestion. *Mol Brain Res* 40:71-78.
- Wenthold RJ, Prybylowski K, Standley S, Sans N, Petralia RS (2003) Trafficking of NMDA receptors. *Annu Rev Pharmacol Toxicol* 43:335-358.

Note: Dr. Michael Browning, an author of three of the cited papers, is President and founder of PhosphoSolutions.

WB = Western Blot **IF** = Immunofluorescence **IHC** = Immunohistochemistry **IP** = Immunoprecipitation

Packaging: 100 μ l in 10 mM HEPES (pH 7.5), 150 mM NaCl, 100 μ g per ml BSA and 50% glycerol. Adequate amount of material to conduct 10-mini Western Blots.

Storage and Stability. For long term storage -20°C is recommended. Stable at -20°C for at least 1 year.

Shipment: Domestic - Blue Ice; International - Blue Ice or Dry Ice.