



GUINEA PIG ANTI-P2X2 RECEPTOR POLYCLONAL ANTIBODY

CATALOG NUMBER:	AB5894
LOT NUMBER:	
QUANTITY:	50 µL
SPECIFICITY:	P2X2 Receptor.
IMMUNOGEN:	A 13 amino acid peptide corresponding to amino acids 460-472 from the carboxy-terminus of the rat P2X2 Receptor protein (Catalog Number AG354).
APPLICATIONS:	Immunoblotting: 1:500 Immunohistochemistry: 1:500. Immunocytochemistry: 1:500. Optimal working dilutions must be determined by end user.
SPECIES REACTIVITIES:	Rat and human. Other species have not been tested.
FORMAT:	Serum
PRESENTATION:	Liquid. Contains 0.05% sodium azide.
STORAGE/HANDLING:	Maintain at -20°C in undiluted aliquots for up to 6 months. Avoid repeated freeze/thaw cycles.
RELATED REFERENCES:	Brake, A.J., et al., (1994) New structure motif for ligand-gated ion channels defined by ionotropic ATP receptor. <i>Nature</i> 371 :519-523. Vulchanova, L.L., et al., (1996) Differential distribution of two ATP-gated channels (P2X receptors) determined by immunocytochemistry. <i>PNAS.USA</i> 93 :8063-8067. Vulchanova, L., et al., (1997) Immunohistochemical study of the P2X2 and P2X3 receptor subunits in rat and monkey sensory neurons and their central terminals. <i>Neuropharmacology</i> 36 :1229-1242.



APPLICATION NOTES FOR AB5894

IMMUNOHISTOCHEMISTRY

Male Sprague-Dawley rats (b.wt. 100-150g) were anesthetized with sodium pentobarbital and perfused via the ascending aorta with: 1) 50 mL of Ca²⁺-free Tyrode+s solution followed by 2) a formalin-picric acid fixative (4% paraformaldehyde with 0.4% picric acid in 0.16 M phosphate buffer, pH 6.9) and 3) 10% sucrose in PBS as a cryo-protectant. Tissues were rapidly dissected out and stored overnight in 0.1 M phosphate buffer (pH 7.4) containing 10% sucrose.

Slide-mounted tissue sections were incubated with blocking buffer for 1 hour at room temperature. Primary antibody was diluted in blocking buffer to the appropriate working dilution. Blocking buffer was removed and the slides were then incubated at 2-8°C for 18-24 hours with AB5894 (1:500). After rinsing in PBS 3 times sections were incubated for 60 minutes at room temperature with Cy3-conjugated secondary antibodies. After mounting in a mixture of PBS and glycerol (1:3) containing 0.1% p-phenylenediamine, sections were examined with a Nikon Microphot-SA epifluorescence microscope.

IMMUNOCYTOCHEMISTRY

P2X2 transfected cells were processed for indirect immunofluorescence. Media was removed and cells were gently washed 3 times with serum-free media. Following fixation procedure, cells were processed for indirect immunofluorescence as above.

WESTERN BLOTTING

Cell membrane extracts were examined by electrophoresis (8% acrylamide) with SDS under reducing conditions and transferred to a nylon membrane. Membranes were blocked for 1 hour at 2-8°C with 0.1% Tween 20 and 2.5% milk powder (w/v) in PBS. Membranes were incubated with AB5894 diluted 1:500 with same buffer overnight at 2-8°C. Membranes were rinsed and incubated with HRP conjugated secondary antibody for 1 hour at room temperature. Following rinsing the membranes were processed using enhanced chemiluminescence.

Important Note: *During shipment, small volumes of product will occasionally become entrapped in the seal of the product vial. For products with volumes of 200 µL or less, we recommend gently tapping the vial on a hard surface or briefly centrifuging the vial in a tabletop centrifuge to dislodge any liquid in the container's cap.*

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PROCEDURES. NOT FOR HUMAN OR ANIMAL CONSUMPTION

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