OriGene Technologies, Inc.
9620 Medical Center Drive, Ste 200
Rockville, MD 20850, US
Phone: +1-888-267-4436
techsupport@origene.com
EU: info-de@origene.com
CN: techsupport@origene.cn

Product datasheet for RA10108-50

P2X2 (P2RX2) Rabbit Polyclonal Antibody

Product data:

Product Type:	Primary Antibodies
Applications:	IF, IHC, WB
Recommend Dilution:	Immunohistochemistry: 1:1000. Immunocytochemistry: 1:1000. Western Blot: 1:1000.
Reactivity:	Human, Monkey, Rat
Host:	Rabbit
Clonality:	Polyclonal
Immunogen:	Corresponding to residues 460-472 of the carboxy-terminus of rat P2X2.
Specificity:	This antibody reacts to P2X2.
Formulation:	State: Serum State: Liquid serum containing 0.05% sodium azide
Gene Name:	purinergic receptor P2X 2
Database Link:	Entrez Gene 22953 Human
Synonyms:	P2X purinoceptor 2, ATP receptor, Purinergic receptor

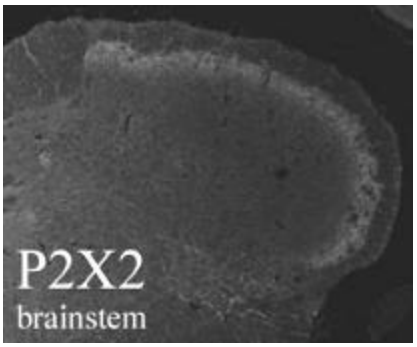
Note: Sodium azide (NaN3) interferes with peroxidase reactions and should not be used with peroxidase methodologies. If sodium azide is present in any steps of the staining procedure, the tissue should thoroughly be rinsed with sodium azide-free buffer before performing the peroxidase reaction.

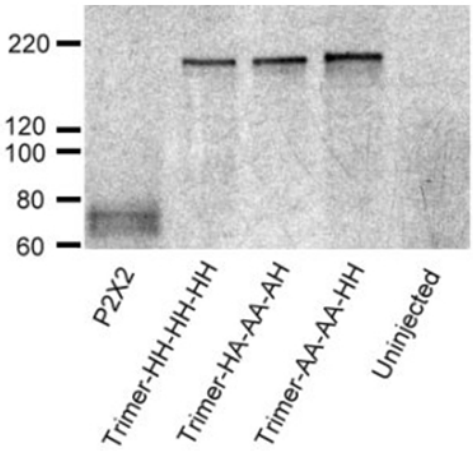
Protocol: Immunohistochemistry:
Antiserum was used on perfusion fixed tissue. Perfusion:
1) calcium-free Tyrodes solution,
2) paraformaldehyde-picric acid fixative, and
3) 10% sucrose in PBS as a cryo-protectant. Desired tissues were dissected and stored overnight in 10% sucrose in PBS.
Slide-mounted tissue sections were processed for indirect immunofluorescence. Slides were incubated with blocking buffer for 1 hour at room temperature. Primary antiserum was diluted with blocking buffer to the appropriate working concentration. Blocking buffer was removed and slides were incubated for 18-24 hours at 4ºC with primary antiserum. Slides were rinsed 3 times and then incubated with secondary antibodies for 1 hour at room temperature. Slides were again rinsed 3 times and coverslipped. Staining was examined using fluorescence microscopy.

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 described 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 4°C with 0.1% Tween 20 and 2.5% milk powder (w/v) in PBS. Membranes were incubated with primary antiserum (1:1000) in the same buffer overnight at 4°C. Membranes were rinsed and incubated with horseradish peroxidase conjugated secondary antibody for 1 hour at room temperature. Following rinsing, the membranes were processed using enhanced chemiluminescence.

Product images:





Western blot analysis indicated that concatamerized trimers remain intact. Total protein from oocytes injected with RNA for wild type P2X2 monomer, or trimer HH-HH-HH, HA-AA-AH, and AA-AA-HH was separated by SDS-PAGE under reducing conditions on a 4–12% gradient gel along with protein from uninjected oocytes. Immunoblot analysis was performed using a polyclonal antibody directed to an extracellular epitope of the human P2X2 receptor and ECL. The position of molecular mass standards (kDa) are shown on the left. In Western blots of similar gels, trimer HA-AH-HH gave a single band of the same size as the trimers shown here. DOI 10.1074/jbc.