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RABBIT ANTI-P2Y1 RECEPTOR (P2RY1) AFFINITY PURIFIED, POLYCLONAL ANTIBODY

CATALOG NUMBER: AB5814-200UL

LOT NUMBER:

QUANTITY: 200 µL

CONCENTRATION: 0.8 mg/mL (after reconstitution)

SPECIFICITY: Recognizes P2Y1 Receptor. The epitope does not share homology with any other known proteins.

IMMUNOGEN: Synthetic peptide corresponding to amino acids 242-258 of rat or human P2Y1 (SwissProt accession numbers P49651, P47900).

APPLICATIONS: Western blot: 1:200-1:400 using ECL on rat brain membranes or human platelets.
Immunohistochemistry on rat brain sections
Dilutions should be made using a carrier protein such as BSA (1-3%)
Optimal working dilutions must be determined by the end user.

CONTROL ANTIGEN: Included free of charge with the antibody is XX µg of control antigen (lyophilized powder). The stock solution of the antigen can be made up using 100 µL of sterile distilled water. For negative control, preincubate 1 µg of protein with 1 µg of antibody for one hour at room temperature. Optimal concentrations must be determined by the end user.

SPECIES REACTIVITIES: Human and rat. Other species have not been tested. It is expected that the antibody may also react with bovine (17/17), chicken and turkey (16/17) and possibly mouse (15/17) due to sequence homology.

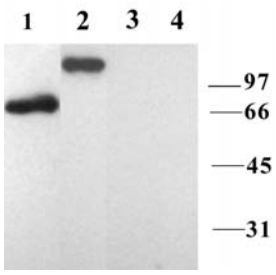
FORMAT: Affinity purified immunoglobulin.

PRESENTATION: Lyophilized from phosphate buffered saline, pH 7.4, containing 1% BSA, and 0.05% sodium azide as a preservative. Reconstitute with 200 µL of sterile deionized water. Centrifuge antibody preparation before use (10,000 xg for 5 min).

STORAGE/HANDLING: Maintain lyophilized material at -20°C for up to 12 months after date of receipt. After reconstitution maintain at -20°C in undiluted aliquots for up to 6 months. Avoid repeated freeze/thaw cycles.

SUGGESTED WESTERN BLOT PROTOCOL

1. Mix the samples (organ membranes: 50 μg/lane; transfected cells: 500,000 cells/lane) with sample-buffer X 2, and heat 10 min at 70°C.
2. 5-50 μL applied to Minigel lane (0.75-1.5 mm width) and run at standard conditions. (60 mA for 2 1.5 mm Minigel gels, 1.4 h). It is suggested that you run 5-15% acrylamide (37.5:1 acrylamide:bisacrysmide) minigel (1.5 mm width) at 30 mA/gel ~1-1.5 hours.
3. Transfer in semi-dry system under standard conditions (3 h 100 mA for two minigel gels)
4. Stain the transferred bands with Millipore BLOT-*FastStain* (Catalog Number 2076).
5. Destain with deionized water.
6. Block with 5% non-fat milk (Marvel or Carnation) in PBS, and 0.025 % sodium azide, overnight at 2-8°C. The non-fat milk should be dissolved freshly, centrifuged 10,000 rpm for 10 min, and filtered through glass filter (Gelman Acrodisc).
7. Incubation with first antibody 2 h at room temperature or overnight at 4°C in blocking solution. The antibody preparation should be centrifuged before use (10,000 g 5 min.). Optimal working dilutions and incubation time will need to be determined by the end user.
8. Wash 4 x 10 min. with PBS-0.1% tween 20. From this stage, azide should be omitted.
9. Incubation with the secondary antibody (HRP-conjugated goat anti-rabbit antibody, for example Millipore Catalog Number AP132P, diluted appropriately) 1 h at room temperature.
10. Wash 4 x 10 min. with PBS-0.1% tween 20.
11. Perform ECL with commercial kits (Chemilucent, Millipore Catalog Number 2600).



Western blotting of rat brain membranes (lanes 1,3) or human platelets lysate (lanes 2,4).
1, 2. AB5814 (1:200)
3, 4. AB5814, preincubated with the control peptide antigen.

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.*

FOR RESEARCH USE ONLY; NOT FOR USE IN DIAGNOSTIC PROCEDURES. NOT FOR HUMAN OR ANIMAL CONSUMPTION

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