upstate | CHEMICON | Linco

THE EXPERTISE OF UPSTATE®, CHEMICON® AND LINCO® IS NOW A PART OF MILLIPORE

RABBIT ANTI-Kir4.2
AFFINITY PURIFIED, POLYCLONAL ANTIBODY

CATALOG NUMBER: AB5880-50UL

LOT NUMBER:

QUANTITY: 50 µL

CONCENTRATION: 0.6 mg/mL (after reconstitution)

SPECIFICITY: Recognizes Kir4.2 (Kir1.3, Kcnj15). The epitope does not share homology with any other known proteins.

IMMUNOGEN: Synthetic peptide corresponding amino acids 347-366 of mouse Kir4.2 (Accession number O88932).

APPLICATIONS: Western blot: 1:200 using ECL on rat kidney membranes.
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. The stock solution of the antigen can be made up using 100 µL of sterile distilled water. For negative control, preincubate 1 µg of peptide with 1 µg of antibody for one hour at room temperature. Optimal concentrations must be determined by the end user.

SPECIES REACTIVITIES: Rat. It is expected that the antibody will also react with mouse, human (18/20) and guinea pig (19/20) due to sequence homology. Other species have not been tested.

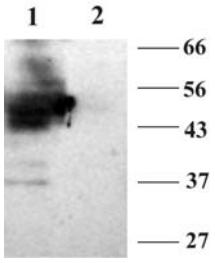
FORMAT: Affinity purified immunoglobulin.

PRESENTATION: Lyophilized from phosphate buffered saline, pH 7.4, containing 1% BSA, 5% sucrose as a stabilizer and 0.025% sodium azide as a preservative. Reconstitute with 50 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 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 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)
3. Transfer in semi-dry system under standard conditions (3 h 200 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 4°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 15 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 15 min. with PBS-0.1% tween 20.
11. Perform ECL with commercial kits (*Chemi*LUCENT, Millipore Catalog Number 2600).



Western blotting of rat kidney membranes:
1. AB5880, 1:200
2. AB5880, preincubated with the control peptide.

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

28820 Single Oak Drive • Temecula, CA 92590
Technical Support: T: 1-800-MILLIPORE (1-800-645-5476) • F: 1-800-437-7502
www.millipore.com