

RABBIT ANTI-HERG (Kv11.1)
POTASSIUM CHANNEL

AFFINITY PURIFIED, POLYCLONAL ANTIBODY

CATALOG NUMBER: AB5908-50UL

LOT NUMBER:

QUANTITY: 50 μ L

CONCENTRATION: 0.6 mg/mL (after reconstitution)

SPECIFICITY: Recognizes HERG (Kv11.1, ether-a-go-go-related channel, KCNH2). The antibody

recognizes the HERG1b splice variant but not splice variants HERG1-3 and HERG-4. The

immunogen shares homology with rat erg2 (22/54) and rat erg3 (21/54).

IMMUNOGEN: GST fusion protein corresponding to amino acids 1106-1159 of human erg (HERG)

(Accession number Q12809).

APPLICATIONS: Western blot: 1:200-1:400 using ECL on HEK293-HERG transfected cells.

Immunoprecipitation: 2-5 µg

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 fusion protein. The stock solution of

the antigen can be made up using 100 μ L of PBS. For negative control, preincubate 3 μ g of fusion protein with 1 μ g of antibody for one hour at room temperature. For positive control, use 20 ng of fusion protein per lane. Optimal concentrations must be determined by the end

user.

SPECIES REACTIVITIES: Human. It has been reported that the antibody reacts with mouse and horse. It is expected

that the antibody may also react with rabbit (identical), canine (51/54), mouse (50/54) and

rat (50/54) 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, 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 date of receipt. After

reconstitution maintain at -20°C in undiluted aliquots for up to 6 months. Avoid repeated

freeze/thaw cycles.

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SUGGESTED WESTERN BLOT PROTOCOL

- 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.
- 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).

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 μ L 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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