Datasheet for ABIN1951823

anti-Doublecortin antibody

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Overview

Quantity:	0.1 mL
Target:	Doublecortin (DCX)
Reactivity:	Human, Mouse, Rat, Cow, Pig, Horse, Chicken, Primate
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This Doublecortin antibody is un-conjugated
Application:	Western Blotting (WB), Immunofluorescence (IF), Immunocytochemistry (ICC)

Product Details

Immunogen:	Recombinant GFAP expressed in bacteria and highly purified. Subsequent boosts were performed with GFAP purified from a Triton X-100 extract of myelin associated material from bovine spinal cord.
Isotype:	IgG1
Specificity:	Immunostaining Cell Cultures 1. Draw of culture medium with aspirator and add 1 ml of 3.7 % formalin in PBS solution to the dish. (make up from 10mls Fisher 37% formalin plus 90mls PBS, the Fisher formalin contains 37% formaldehyde plus about 1% methanol which may be relevant sometimes). Let sit at room temp for 1 minute. (can add 0.1% Tween 20 to PBS used here and all subsequent steps to reduce background, probably best not to do this first time round though as it may extract your antigen or help wash your

cells off the dish).

2. Take off the formalin/PBS and add 1ml of cold methanol (-20°C, kept in well sealed bottle in fridge). Let sit for no more

than 1 minute.

3. Take off methanol and add 1ml of PBS, not letting the specimen dry out. To block nonspecific antibody binding can add

~10ml (=1%) of goat serum (Sigma), and can incubate for 30 minutes. Can then add antibody reagents. Typically 100ml of

hybridoma tissue culture supernatent or 1ml of mouse ascites fluid or crude serum. Incubate for 1 hour at room temp. (or

can go at 37°C for 30 minutes to 1 hour, or can do 4°C overnight, exact time not too critical). Can do very gentle shaking for

well adherent cell lines (3T3, Hek293 etc.).

4. Remove primary antibody and replace with 1 ml of PBS. Let sit for 5-10 minutes, replace PBS and repeat twice, to give

three washes in PBS.

5. Add 0.5 mls of secondary antibody. These are fluorescently labeled Goat anti mouse or rabbit antibodies and are

conjugated to ALEXA dyes and are from Molecular probes (Eugene Oregon, the ALEXA dyes are sulphonated rhodamine

compounds and are much more stable to UV than FITC, TRITC, Texas red etc.). Typically make 1:2,000 dilutions of these

secondaries in PBS plus 1% goat serum, BSA or non fat milk carrier. Incubate for 1 hour at room temp. (or can go at 37°C

for 30 minutes to 1 hour, or can do 4°C overnight). Can do gentle shaking for well adherent cell lines (3T3, HEK293 etc.).

6. Remove secondary antibody and replace with 1 ml of PBS. Let sit for 5-10 minutes, replace PBS and repeat twice, to give

three washes in PBS.

7. Drop on one drop of Fisher mounting medium onto dish and apply 22mm square coverslip. View in the microscope!

Immunostaining Tissue

Solutions

PBS - sodium phosphate-buffered (100 mM, pH 7.2) isotonic (0.9% NaCl, w/v) saline Antibody dilution buffer (PBS with

0.1% non-ionic detergent, such as Triton X-100 or Tween-20) fluorescein anti-fading reagent --

Product Details

	Make up a 2 mg/mL phenylene diamine solution in PBS (phenylene diamine requires extensive vortexing to put it into solution). Once the
Purification:	Purified

Target Details

Target:	Doublecortin (DCX)
Alternative Name:	Doublecortin (DCX Products)

Application Details

Application Notes:	Immunofluorescence: 1:1,000 , Immunohistochemistry: 1:1,000 , Western Blot: 1:5,000. Dilutions listed as a recommendation. Optimal dilution should be determined by investigator.
Restrictions:	For Research Use only

Handling

Format:	Liquid
Buffer:	Liquid with 10mM Sodium Azide
Preservative:	Sodium azide
Precaution of Use:	WARNING: Reagents contain sodium azide. Sodium azide is very toxic if ingested or inhaled. Avoid contact with skin, eyes, or clothing. Wear eye or face protection when handling. If skin or eye contact occurs, wash with copious amounts of water. If ingested or inhaled, contact a physician immediately. Sodium azide yields toxic hydrazoic acid under acidic conditions. Dilute azide-containing compounds in running water before discarding to avoid accumulation of potentially explosive deposits in lead or copper plumbing.
Handling Advice:	Avoid repeated freeze-thaw cycles.
Storage:	4 °C
Storage Comment:	Antibody can be aliquotted and stored frozen at -20° C to -70° C in a manual defrost freezer for six , months without detectable loss of activity. The antibody can be stored at 2° - 8° C for 1 month without , detectable loss of activity.