

## MOUSE ANTI-DESMIN MONOCLONAL ANTIBODY

**CATALOG NUMBER:** MAB3430 – formerly Roche Catalog # 0811335001

**LOT NUMBER:**

**QUANTITY:** 40 µg

**CONCENTRATION:** 1 mg/mL

**SPECIFICITY:** The antibody reacts with desmin from human, pig, rat and taod. In tissue sections this antibody is used to stain skeletal, cardiac, visceral, and some vascular smooth muscle cells. Cell lines such as RD (ATCC CCL 136) and hamster BHK-21 are positive (1).

**IMMUNOGEN:** Purified desmin.

**ISOTYPE:** IgG<sub>1</sub>

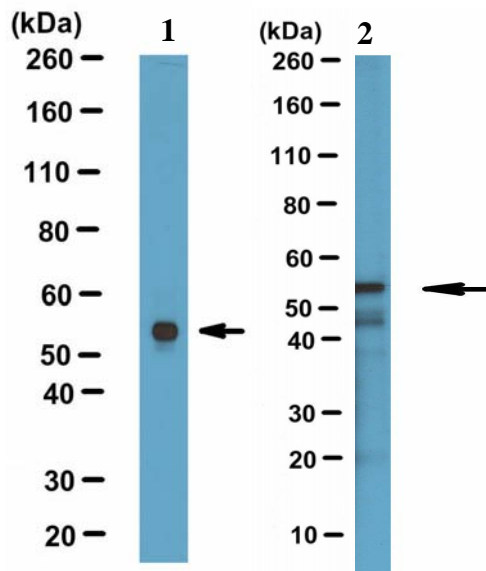
**CLONE NAME:** DE-B-5

**APPLICATIONS:** Western Blot Analysis  
 (1:1000 dilution)

Immunocytochemistry:  
 (5 µg/ml)

Immunohistochemistry:  
 (5 µg/ml)

Optimal working  
 dilutions must be  
 determined by end  
 user.



**Western Blot analysis:**  
 Mouse heart (lane 1)  
 and Human fetal  
 skeletal muscle (lane 2)  
 lysate was resolved by  
 electrophoresis,  
 transferred to PVDF  
 membranes and probed  
 with Desmin (1:1000  
 dilution). Proteins were  
 visualized using a Gt X  
 ms IgG conjugated to  
 HRP and visualized  
 using a  
 chemiluminescence  
 detection system.

Arrow indicates Desmin  
 (~53kDa).

**SPECIES REACTIVITIES:** Human, mouse, rat, porcine and toad.

**FORMAT:** Purified immunoglobulin (column chromatography.)

**PRESENTATION:** Liquid in 0.02 M phosphate buffer, 0.25M NaCl with 0.1% sodium azide, pH 7.6.



**STORAGE/HANDLING:** Maintain antibody refrigerated at 2-8°C in undiluted aliquots for up to 6 months.  
DO NOT FREEZE.

**REFERENCES:**

1. Debus, E., Weber, K., & Osborn, M. (1983) *EMBO J.* **2**, 2305-2312.
2. Altmannsberger, M. et al. (1982) *Lab. Invest.* **46**, 520-526.

### Immunohysto/cyto chemistry Protocols

Ideal specimens are obtained from frozen sections from shock-frozen tissue samples. The frozen sections are dried in the air and then fixed with acetone at -20°C for 10 min. Excess acetone is allowed to evaporate at 15-25°C. Material fixed in alcohol and embedded in paraffin can also be used (2). Formaldehyde fixation will reduce or eliminate the intensity of staining depending on the conditions under which it is performed. Other fixation conditions must be first tested by the investigator.

It is advantageous to block unspecific binding sites by overlaying the sections with fetal calf serum for 20–30 min at 15-25°C. Excess of fetal calf serum is removed by decanting before application of the antibody solution.

Cytocentrifuge preparations of single cells or cell smears are also fixed in acetone. These preparations should, however not be dried in the air. Instead, the excess acetone is removed by briefly washing in phosphate-buffered saline (PBS).

Further treatment is then as follows:

- Overlay the preparation with 10–20 µl antibody solution and incubate in a humid chamber at 37°C for 1 h.
- Dip the slide briefly in PBS and then wash 3 x in PBS for 3 min (using a fresh PBS bath in each case).
- Wipe the margins of the preparation dry and overlay the preparation with 10–20 µl of a solution of anti-mouse Ig-FITC or anti-mouse IgG-peroxidase solution and allow to incubate for 1 h at 37°C in a humid chamber.
- Wash the slide as described above.

The preparation must not be allowed to dry out during any of the steps.

If using an indirect immunofluorescence technique, the preparation should be overlaid with a suitable embedding medium (e.g. Moviol, Hoechst) and examined under the fluorescence microscope. If a POD-conjugate has been used as the secondary antibody, the preparation should be overlaid with a substrate solution (see below) and incubated at 15-25°C until a clearly visible redbrown color develops. A negative control (e.g. only the secondary antibody) should remain unchanged in color during this incubation period.

Subsequently, the substrate is washed off with PBS and the preparation is stained, if desired, with hemalum stain for about 1 min. The hemalum solution is washed off with PBS; the preparation is embedded and examined.

Substrate solutions:

Aminoethyl-carbazole: Dissolve 2 mg 3-amino-9-ethylcarbazole with 1.2 ml dimethylsulfoxide and add 28.8 ml 50 mM Tris-HCl, pH 7.3, and 20 µl 3% H<sub>2</sub>O<sub>2</sub> (w/v).

Prepare solution freshly each day. Diaminobenzidine: Dissolve 25 mg 3,3'-diaminobenzidine with 50 ml 50 mM Tris-HCl, pH 7.3, and add 40 µl 3% H<sub>2</sub>O<sub>2</sub> (w/v).

Prepare solution freshly each day.



**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  $\mu$ 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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PROCEDURES. NOT FOR HUMAN OR ANIMAL CONSUMPTION

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