

Novocastra™ Lyophilized Mouse Monoclonal Antibody Desmin

Product Code: NCL-DES-DERII

Intended Use	FOR RESEARCH USE ONLY.
Specificity	Human desmin, a 53 kD intermediate filament protein in muscle cells. Also reacts with rat, chicken, dog, guinea pig, mouse and hamster desmin.
Clone	DE-R-11
Ig Class	IgG1
Antigen Used for Immunizations	Purified porcine desmin.
Preparation	Lyophilized tissue culture supernatant containing 15 mM sodium azide. Reconstitute with the volume of sterile distilled water indicated on the vial label.
Effective on Frozen Tissue	Yes
Effective on Paraffin Wax Embedded Tissue	Yes
Recommendations on Use	Immunohistochemistry: Typical working dilution 1:50–1:100. Trypsin digestion of paraffin sections is recommended. 60 minutes primary antibody incubation at 25 °C. Standard ABC technique. Western Blotting: Typical working dilution 1:100–1:250.
Positive Controls	Immunohistochemistry: Bowel wall should show distinct staining of the muscularis mucosae and the muscularis propria. Western Blotting: Muscle.
Staining Pattern	Cytoplasmic.
Storage and Stability	Store unopened lyophilized antibody at 4 °C. Under these conditions, there is no significant loss in product performance up to the expiry date indicated on the vial label. The reconstituted antibody is stable for at least two months when stored at 4 °C. For long term storage, it is recommended that aliquots of the antibody are frozen at -20 °C (frost-free freezers are not recommended). Repeated freezing and thawing must be avoided. Prepare working dilutions on the day of use.
General Overview	NCL-DES-DERII reacts with an 18 kD rod piece of the intermediate filament protein desmin (53 kD) in muscle cells. The antibody does not appear to recognise other intermediate filament proteins. In normal tissues, NCL-DES-DERII reacts with both striated (skeletal and cardiac) and smooth muscle cells. The labelling is confined to the Z bands in skeletal and cardiac muscle giving a characteristic striated appearance.
General References	Altmanberger M, Weber K, Droste R, et al.. American Journal of Pathology. 118: 85–98 (1985). Osborn M, Altmanberger M, Debus E, et al.. Cancer Cells 1. The transformed phenotype. Cold Spring Harbor Lab. 191–200 (1984). Debus E, Weber K and Osborn M. The EMBO Journal. 2 (12): 2305–2312 (1983).



Instructions for Use

Trypsin Digestion for Immunohistochemical Demonstration on Paraffin Sections

1. Preheat the following to 37 °C using a water bath:
 - (i) 200 mL of TBS
 - (ii) 200 mL of distilled water.
2. Dissolve 0.2 g Trypsin 250 and 0.2 g Calcium chloride in the 200 mL of TBS.
3. Once the Trypsin solution is at 37 °C, pH to 7.8 with 1 M sodium hydroxide.
4. Place rehydrated paraffin sections in the distilled water to preheat the sections to 37 °C for a minimum of 5 minutes.
5. Incubate sections in Trypsin solution at 37 °C. The time required will depend on the antibody and tissue, however, 30 minutes is usually sufficient.
6. Rinse sections in running tap water.
7. Proceed with immunohistochemistry protocol.

Reagents Required but not Supplied

50 mM Tris-buffered saline

Trypsin 250: Difco order code 0152-13 (available from Becton Dickinson).

Calcium chloride

1 M Sodium Hydroxide

** Trypsin containing chymotrypsin should always be used. The enzyme activities can vary from a supplier and between batches. Such variations may affect the incubation time required.*