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Place your order with CEDARLANE[®] or your local distributor. Please contact CEDARLANE[®] for lot specific information.

Mouse Anti-Human α-Actin [alpha Smooth Muscle isoform] Monoclonal Antibody

CLT9000 LOT: 0608036820

DESCRIPTION: This monoclonal antibody recognizes only the α -actin isoform of smooth muscle cells from human, bovine, rat, mouse and chicken. It reacts with all types of smooth muscle cells e.g. in arterial walls, in lamina muscularis mucosae and propria of the intestinum, in myometrium and in stroma cells of different tissues. The α -actin antibody positively stains microfilaments in smooth muscle cells and in some fibroblast-subtypes derived from smooth muscle cells. It also reacts positively with myoepithelial cells of different glands such as mammary and salivary glands. (It shows no cross-reactivity with striated muscle cells of heart and skeletal muscle).

SPECIFICITY: This antibody is specific for the alpha-smooth-muscle isoform of actin (MW 43 kDa)

<u>PRESENTATION</u>: This product is provided in a 50 μ g size at a concentration of 0.1 mg/ml. The antibody was purified by protein A affinity chromatography and is provided in PBS, pH 7.4, containing 0.5% BSA and 0.09 % sodium azide (NaN₃) as a preservative.

STORAGE/STABILITY: This antibody is stable at 4°C for up to 12 months from date of receipt.

APPLICATIONS:

- Western Blot
- Immunohistochemistry: 1:200; Frozen and formalin-fixed, paraffin-embedded tissues;
- Immunofluorescence

<u>SPECIES REACTIVITY</u>: Human, mouse, rat, bovine, equine, and chicken.

<u>CONTROL</u>: Positive reaction using stress fibres of smooth muscle-derived cells and some smooth muscle subtype fibroblasts.

Continued Overleaf...

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SPECIFICATIONS:

Clone: ASM -1

<u>Hybridoma Production</u>: BALB/c mice were immunized with the synthetic amino-terminal decapeptide of α -actin of smooth muscle cells (1).

Ig Class: mouse IgG2a

Immunogen: Synthetic peptide corresponding to the 10 N-terminal amino acids of the alpha smooth muscle subtype fibroblast

<u>RECOMMENDED PROCEDURES</u>:

<u>Frozen Sections</u>: Ideal frozen sections (4-5 μ m) are obtained from shock frozen tissue samples. Air dry frozen sections and then fix with acetone for 10 minutes at -15 to -25°C. Remove excess acetone by drying or by washing with PBS (phosphate buffered saline).

<u>Cell suspensions</u>: Cytocentrifuge preparations of single cells or cell smears. These should also be fixed in acetone as above. However, these preparations should not be air-dried. Remove excess acetone by rinsing with PBS.

<u>Coverslip</u>, chamber slide preparation: Remove liquid carefully by suction. Wash cells for a short time in PBS and fix the cells in acetone or methanol for 10 minutes at -15 to -25°C. Wash out fixative with PBS or air dry.

Further treatment as follows:

- 1. Block unspecific binding sites by overlaying the preparation with 20 μl fetal calf serum (FCS) and incubate for 30 minutes at 37°C in a humid chamber.
- 2. Cover the section according to the size with 10-20 μ l of antibody solution and incubate for 60 minutes in a humid chamber at 15-25°C.
- 3. Immerse the slides in PBS and wash 3 times for 3 minutes each in PBS.
- 4. Wipe slides dry except area of sections, cover the section with 10-20 μl of a solution of FITC anti-mouse IgG (cat. no. CLCC30001) and incubate for 60 minutes at 15-25°C or at 37°C in a humid chamber.
- 5. Wash slides 3 times for 3 minutes each in PBS.
- 6. Cover the preparation with a suitable mounting medium (e.g. Mowlol, Hoechst) and examine under a fluorescence microscope.

NOTE 1: The section should not be allowed to dry out during any of the steps.

NOTE 2: The antibody solution should be completely free of precipitate. If necessary, centrifuge the solution at high speed prior to use.

REFERENCES:

- 1. Skalli, O. et al. (1986) J.Cell.Biol. 103 : 2787-2796.
- 2. Schürch, W. et al. (1987) Am. J. Pathol. 128: 91-103
- 3. Ehler, E. et al. (2004) Developmental Dynamics 229: 745-755
- 4. Demirkesen, C et al. (1995) J Cutan Pathol 22: 518-535

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