

Novocastra™ Ready-to-Use Mouse Monoclonal Antibody Alpha Smooth Muscle Actin

Product Code: RTU-SMA

Intended Use	FOR RESEARCH USE ONLY.
Specificity	Human alpha smooth muscle actin. Reactive with smooth muscle cells in blood vessel walls, gut wall, myometrium and arrectores pili of skin. Myoepithelial cells such as those in breast and salivary gland also contain actin.
Clone	asm-1
Ig Class	IgG2a
Antigen Used for Immunizations	Synthetic amino terminal decapeptide of alpha smooth muscle isoform of actin.
Hybridoma Partner	Mouse myeloma (p3-NS1-Ag4-1).
Preparation	Tissue culture supernatant diluted in 5% horse serum in PBS containing 12 mM sodium azide. Volume as indicated on vial label.
Effective on Frozen Tissue	Yes
Effective on Paraffin Wax Embedded Tissue	Yes. Trypsinization of paraffin sections may be required.
Recommendations on Use	Immunohistochemistry: Typical working dilution: NEAT. 15 minutes primary antibody incubation at 25 °C when used in conjunction with the Novostain Universal Detection Kit (Ready to Use), code NCL-RTU-D. Recommendations on use will differ if other detection systems are used eg Standard ABC technique. Western Blotting: Not recommended. Not recommended for use on Ventana automated staining systems (Ventana Medical Systems Inc., USA).
Positive Controls	Immunohistochemistry: Bowel wall, with distinct staining of the muscularis mucosae and the muscularis propria.
Staining Pattern	Cytoplasmic.
Storage and Stability	Store ready-to-use prediluted liquid antibody at 4 °C. Return to 4 °C immediately after use. Under these conditions, there is no significant loss in product performance up to the expiry date indicated on the vial label.
General Overview	Clone asm-1 is reported to label alpha smooth muscle actin found in smooth muscle cells. These cells can be found in vascular walls, intestinal muscularis mucosae and muscularis propria and in the stroma of various tissues. Clone asm-1 is also reported to react with myoepithelial cells of various glands, notably salivary and mammary glands.
General References	Mason D and Gatter K. Journal of Clinical Pathology. 40: 1042–1054 (1987). Skalli O, Ropraz P, Trzeciak A, et al.. The Journal of Cell Biology. 103: 2787–2796 (1986).



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.*