

# Novocastra™ Lyophilized Mouse Monoclonal Antibody Alpha Smooth Muscle Actin

## Product Code: NCL-SMA

<b>Intended Use</b>	FOR RESEARCH USE ONLY.
<b>Specificity</b>	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.
<b>Clone</b>	asm-1
<b>Ig Class</b>	IgG2a
<b>Antigen Used for Immunizations</b>	Synthetic amino terminal decapeptide of alpha smooth muscle isoform of actin.
<b>Hybridoma Partner</b>	Mouse myeloma (p3-NS1-Ag4-1).
<b>Preparation</b>	Lyophilized tissue culture supernatant containing 15 mM sodium azide. Reconstitute with the volume of sterile distilled water indicated on the vial label.
<b>Effective on Frozen Tissue</b>	Yes
<b>Effective on Paraffin Wax Embedded Tissue</b>	Yes
<b>Recommendations on Use</b>	Immunohistochemistry: Typical working dilution 1:50. Trypsinisation of paraffin sections may be required. 60 minutes primary antibody incubation at 25 °C. Standard ABC technique. Western Blotting: Typical working dilution 1:100–1:250.
<b>Positive Controls</b>	Immunohistochemistry: Bowel wall, with distinct staining of the muscularis mucosae and the muscularis propria. Western Blotting: Smooth muscle.
<b>Staining Pattern</b>	Cytoplasmic.
<b>Storage and Stability</b>	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.
<b>General Overview</b>	NCL-SMA 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. NCL-SMA is also reported to react with myoepithelial cells of various glands, notably salivary and mammary glands.
<b>General References</b>	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.*