

**DESCRIPTION**

<b>Species Reactivity</b>	Human/Mouse/Rat
<b>Specificity</b>	Detects human $\alpha$ -Smooth Muscle Actin.
<b>Source</b>	Monoclonal Mouse IgG <sub>2A</sub> Clone # 1A4
<b>Purification</b>	Protein A or G purified from hybridoma culture supernatant
<b>Immunogen</b>	Peptide corresponding to residues EEEDSTALVC of the N-term of human $\alpha$ -smooth muscle actin (1)
<b>Formulation</b>	Lyophilized from a 0.2 $\mu$ m filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 $\mu$ m filtered solution in PBS.

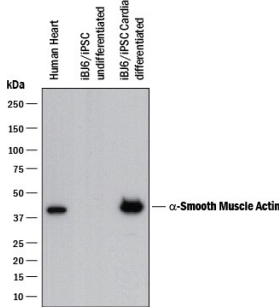
**APPLICATIONS**

**Please Note:** Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	<b>Recommended Concentration</b>	<b>Sample</b>
<b>Western Blot</b>	0.1-0.25 $\mu$ g/mL	See Below
<b>Immunocytochemistry</b>	8-25 $\mu$ g/mL	See Below
<b>Immunohistochemistry</b>	8-25 $\mu$ g/mL	See Below
<b>Intracellular Staining by Flow Cytometry</b>	0.25 $\mu$ g/10 <sup>6</sup> cells	See Below
<b>Simple Western</b>	10 $\mu$ g/mL	See Below
<b>CyTOF-ready</b>	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

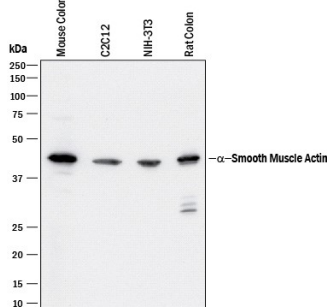
**DATA**

**Western Blot**



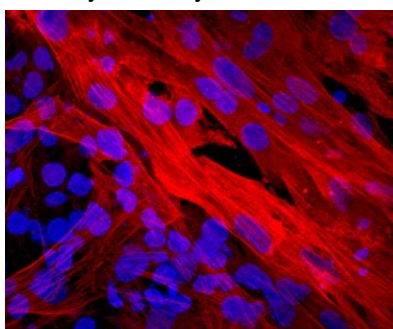
**Detection of Human  $\alpha$ -Smooth Muscle Actin by Western Blot.** Western blot shows lysates of human heart tissue and IBJ6 human induced pluripotent stem cell line undifferentiated or cardiac differentiated. PVDF membrane was probed with 0.25  $\mu$ g/mL of Mouse Anti-Human/Mouse/Rat  $\alpha$ -Smooth Muscle Actin Monoclonal Antibody (Catalog # MAB1420) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for  $\alpha$ -Smooth Muscle Actin at approximately 40 kDa (as indicated). This experiment was conducted under reducing conditions and using [Immunoblot Buffer Group 1](#).

**Western Blot**



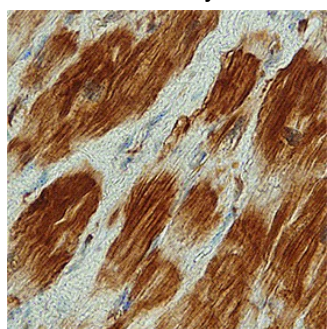
**Detection of Mouse and Rat  $\alpha$ -Smooth Muscle Actin by Western Blot.** Western blot shows lysates of mouse colon tissue, C2C12 mouse myoblast cell line, NIH-3T3 mouse embryonic fibroblast cell line, and rat colon tissue. PVDF membrane was probed with 0.1  $\mu$ g/mL of Mouse Anti-Human/Mouse/Rat  $\alpha$ -Smooth Muscle Actin Monoclonal Antibody (Catalog # MAB1420) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for  $\alpha$ -Smooth Muscle Actin at approximately 42 kDa (as indicated). This experiment was conducted under reducing conditions and using [Immunoblot Buffer Group 1](#).

**Immunocytochemistry**



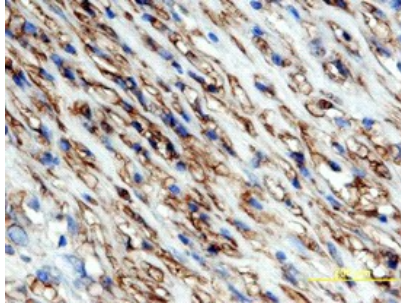
**$\alpha$ -Smooth Muscle Actin in BG01V Human Embryonic Stem Cells.**  $\alpha$ -Smooth Muscle Actin was detected in immersion fixed BG01V human embryonic stem cells differentiated to cardiomyocytes using Mouse Anti-Human/Mouse/Rat  $\alpha$ -Smooth Muscle Actin Monoclonal Antibody (Catalog # MAB1420) at 10  $\mu$ g/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NLO07) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm and cytoskeleton. View our protocol for [Fluorescent ICC Staining of Cells on Coverslips](#).

**Immunohistochemistry**



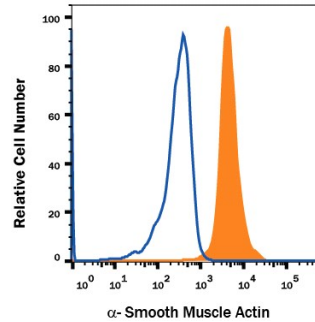
**$\alpha$ -Smooth Muscle Actin in Human Heart.**  $\alpha$ -Smooth Muscle Actin was detected in immersion fixed paraffin-embedded sections of human heart using Mouse Anti-Human/Mouse/Rat  $\alpha$ -Smooth Muscle Actin Monoclonal Antibody (Catalog # MAB1420) at 15  $\mu$ g/mL overnight at 4 °C. Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using the Anti-Mouse HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS002) and counterstained with hematoxylin (blue). View our protocol for [Chromogenic IHC Staining of Paraffin-embedded Tissue Sections](#).

**Immunohistochemistry**



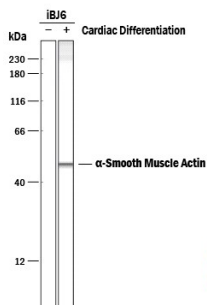
**$\alpha$ -Smooth Muscle Actin in Human Breast Cancer Tissue.**  $\alpha$ -Smooth Muscle Actin was detected in immersion fixed paraffin-embedded sections of human breast cancer tissue using Mouse Anti-Human/Mouse/Rat  $\alpha$ -Smooth Muscle Actin Monoclonal Antibody (Catalog # MAB1420) at 8  $\mu$ g/mL overnight at 4 °C. Tissue was stained using the Anti-Mouse HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS002) and counterstained with hematoxylin (blue). View our protocol for [Chromogenic IHC Staining of Paraffin-embedded Tissue Sections](#).

**Intracellular Staining by Flow Cytometry**



**Detection of  $\alpha$ -Smooth Muscle Actin in Human Blood Monocytes by Flow Cytometry.** Human peripheral blood monocytes were stained with Mouse Anti-Human/Mouse/Rat  $\alpha$ -Smooth Muscle Actin Monoclonal Antibody (Catalog # MAB1420, filled histogram) or isotype control antibody (Catalog # MAB003, open histogram), followed by Allophycocyanin-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0101B). To facilitate intracellular staining, cells were fixed with Flow Cytometry Fixation Buffer (Catalog # FC004) and permeabilized with Flow Cytometry Permeabilization/Wash Buffer I (Catalog # FC005). View our protocol for [Staining Intracellular Molecules](#).

**Simple Western**



**Detection of Human  $\alpha$ -Smooth Muscle Actin by Simple Western™.** Simple Western lane view shows lysates of iBJ6 human induced pluripotent stem cell line untreated (-) or differentiated to cardiomyocytes (+), loaded at 0.2 mg/mL. A specific band was detected for  $\alpha$ -Smooth Muscle Actin at approximately 49 kDa (as indicated) using 10  $\mu$ g/mL of Mouse Anti-Human/Mouse/Rat  $\alpha$ -Smooth Muscle Actin Monoclonal Antibody (Catalog # MAB1420). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.



**PREPARATION AND STORAGE**

<b>Reconstitution</b>	Reconstitute at 0.5 mg/mL in sterile PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
<b>Stability &amp; Storage</b>	<b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b> <ul style="list-style-type: none"> <li>● 12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>● 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>● 6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>

**BACKGROUND**

$\alpha$ -Smooth Muscle Actin has been frequently used as a marker of smooth muscle differentiation (1, 2).

**References:**

1. Skalli, O. *et al.* (1986) J. Cell Biol. **103**:2787.
2. Oishi, K. *et al.* (2002) J. Physiol. **540**:139.