



# Certificate of Analysis

**Product:** Anti-SUMO-2/3 Mouse MAb V 1.0

**Catalog #:** ASM24

**Product Description:** Mouse monoclonal IgG1-kappa  
Clone 11G2  
Purified by protein G affinity chromatography

**Amount:** 200 µl per tube when reconstituted (ASM24)  
150 µl per tube when reconstituted (ASM24-S)

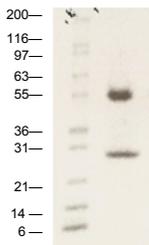
**Lot:** 011

TEST	SPECIFICATON	LOT RESULTS
<b>Appearance</b>	White lyophilized powder	White lyophilized powder
<b>Purity</b>	20 µg sample shows heavy (55kD) & light chains (22kD) represent >80% of proteins by coomassie staining under denaturing conditions.	>80%
<b>Protein Quantitation</b> Protein quantitated with Precision Red protein assay reagent	120-360 µg per tube (ASM24) 90-270 µg per tube (ASM24-S)  Aliquot size is determined by biological activity therefore µg amounts per tube will vary between lots.	190 µg 150 µg
<b>Sensitivity/Specificity</b>	See IP data	Signal strength; heat shock>untreated>SUMO-2 knockdown. Signals comparable to previous Lot of ASM24.
<b>Immunolocalization</b>	Chromosomal localization (enrichment) of SUMO-2/3 during mitosis in HeLa cells. Nuclear staining, dispersed cytoplasmic localization and filamentous structures visible in non-mitotic HeLa cells. ASM23 was used at 1:500 dilution.	Chromosomal localization confirmed in mitotic cells  Nuclear, dispersed and filamentous cytoplasmic structures confirmed.
<b>Immunoprecipitation</b>	Immunoprecipitation of free SUMO-2/3 and SUMOylated proteins from HeLa cell lysates. Signal strength from equivalent amounts of lysate; Heat shocked cells>untreated cells>SUMO-2 knockdown. Signals are comparable to previous Lot of ASM24.	Signal strength; heat shock>untreated>SUMO-2 knockdown. Signals comparable to previous Lot of ASM24.
<b>QC Release Date</b>	5/25/15	

Kim Middleton  
QC Manager

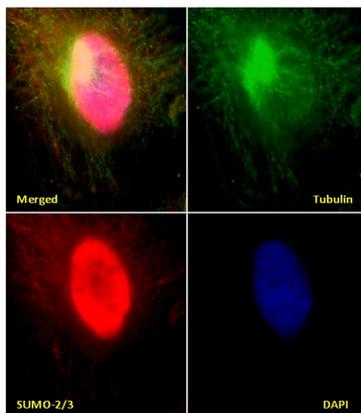
## Lot specific QC Data

### Purity Analysis: Lot 011



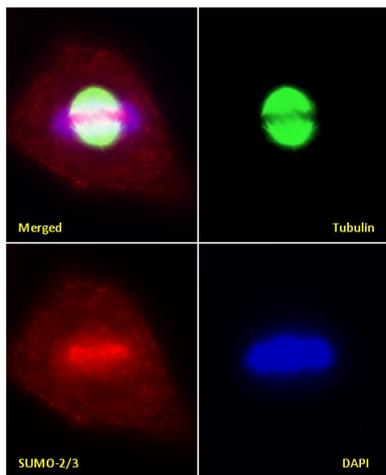
Purity analysis of protein G purified ASM24. 20 µg of ASM23 was run on 4-20% SDS-PAGE. Proteins were stained with coomassie blue. Bands at 22 kD and 55 kD represent antibody heavy and light chains respectively.

### Immunolocalization interphase cells: Lot 011



HeLa cells were stained and visualized by fluorescence microscopy as described in the product data sheet for interphase cells. The cells were stained against  $\alpha/\beta$ -tubulin (sheep anti-tubulin Ab, Cat# ATN02, green) and SUMO-2/3 (11G2, red). DNA was stained with DAPI. Nuclear staining and cytoplasmic staining of filamentous structures are visible.

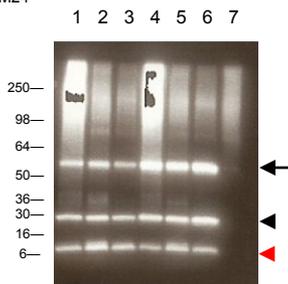
### Immunolocalization mitotic cells: Lot 011



HeLa cells were stained and visualized by fluorescence microscopy as described in the product data sheet for mitotic cells. The cells were stained against  $\alpha/\beta$ -tubulin (sheep anti-tubulin Ab, Cat# ATN02, green) and SUMO-2/3 (11G2, red). DNA was stained with DAPI. Enrichment of SUMO 2/3 at chromosomes can be observed during mitosis.

### Immunoprecipitation: Lot 011

IP ASM24



2 mg of cell lysate from heat shocked HeLa cells (Lanes 1 & 4), untreated HeLa cells (Lanes 2 & 5) and SUMO-2 shRNA knock-down HeLa cells (Lanes 3 & 6) were immunoprecipitated (IP) by incubated with 30 µl of ASM24 anti-SUMO-2/3 antibody and protein G-beads according to the protocol in the product data sheet. Lanes 1-3 represent IP data using ASM24 Lot 009, Lanes 4-6 represent IP data from ASM23 Lot 011. Immunoprecipitated SUMO-2/3 modified proteins were run on SDS-PAGE, transferred to PVDF membranes and probed with a 1:500 dilution of ASM23. Arrow indicates heavy chain, black arrowhead indicates light chain, red arrowhead indicates free SUMO-2/3. Lane 7 is 10 µg of input lysate from heat shocked HeLa cells.