V 1.1

SUMO1 IP Control Beads

Cat. # CIG03-Beads
Lot: 021

Upon arrival, store at 4°C (desiccated)
See datasheet for storage after reconstitution

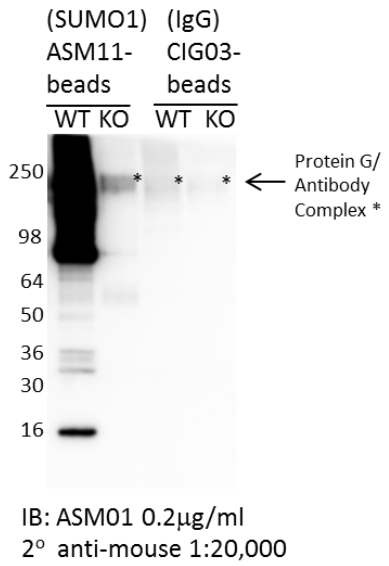
Form:	Lyophilized powder
Amount of material:	1 x 500 µl when reconstituted
Validated applications:	IP control for Ab affinity reagents
Species reactivity:	na
Host/Isotype:	Mouse/polyclonal
Clone:	na

Background Information
Many of Cytoskeleton Inc's Signal-Seeker™ affinity enrichment beads are based on mouse monoclonal antibody reagents covalently bound to beads. Mouse IgG IP Control Beads provide an ideal negative control and should be included in an IP experiment to control for non-specific binding in any antibody based affinity immune-precipitation reaction, see Figure 1.

Material
Normal whole mouse IgG from non-immunized animals has been covalently linked to agarose affinity beads. Antibody binding is in the range of 1.0-1.25 mg antibody per ml of bead slurry which is a similar range to select Signal-Seeker™ affinity reagents.

Storage and Reconstitution
Shipped at ambient temperature. The lyophilized IP control beads can be stored desiccated at 4°C for 6 months. For reconstitution, the product tube should be briefly centrifuged to collect the lyophilized beads at the bottom of the tube. Reconstitute each tube in 500 µl of 50% glycerol in water and store in –20°C. Allow beads to rehydrate completely before use (15-20 minutes). Final buffer composition is 200 mM PIPES, 5% sucrose, and 1% dextran. When stored and reconstituted as described, the product is stable for at least 6 months at –20°C.

Applications
Immuno-precipitation (IP) Application
Use an equivalent volume of control bead slurry as that being used for SUMO1 enrichment IP assay. This is generally in the region of 40 µl per IP. Sufficient for 12 IP reactions. See Figure 1 for representative data.



HAP1 wildtype (WT) or SUMO1 knockout (KO) lysate, was obtained using BlastR lysis and filter system. 1 mg of each lysate were incubated with 40 µg of SUMO1 affinity beads (ASM11-beads (Cytoskeleton)), and conjugated SUMO1 IP control beads (CIG03). Samples were separated by SDS-PAGE and transferred to PVDF. Enriched SUMO1 samples were analyzed by western blot using SUMO1 (ASM01 (Cytoskeleton)) antibody at 1:5000. Mouse-HRP secondary at 1:20,000 in 5% milk was used.

Fig. 1: Isolation and detection of SUMO1 modified proteins from HAP1 wildtype and SUMO1 knockout cells

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