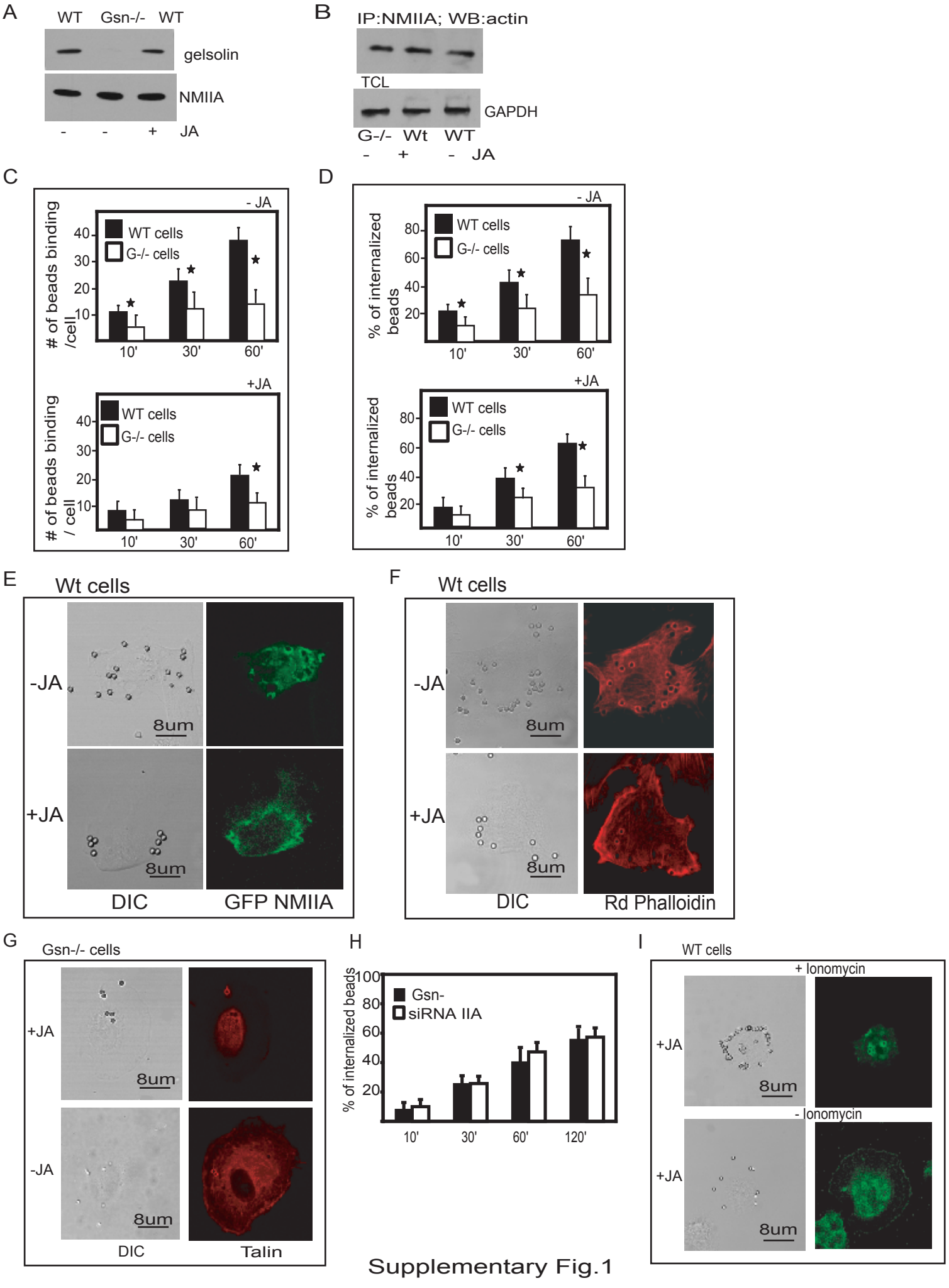
Supplementary Figure Legends

Fig. 1A Cell lysates from jasplakinolide (JA; 500 nM) treated or untreated gelsolin Wt and gelsolin null cells were collected and separated by SDS-PAGE gels. Blots were probed with gelsolin antibody or NMIIA antibody. We observed no difference in levels of gelsolin or NMIIA between Wt cells treated with or without JA.
Fig. 1B NMIIA immunoprecipitates from collagen bead treated samples were probed with β-actin antibody. JA treatment did not affect the interaction between NMIIA and actin significantly. TCL- total cell lysate.
Fig. 1C Histogram shows the **c**omparison of collagen bead binding in the presence or absence of JA in gelsolin Wt and gelsolin null cells.
Fig. 1D Comparison of the % collagen bead internalization in the presence or absence of JA in gelsolin Wt and gelsolin null cells.
Fig. 1E JA treated or untreated gelsolin Wt cells were incubated with collagen-coated beads. Fewer collagen beads bound to JA-treated Wt cells. The bound beads showed targeting of GFP-NMIIA to beads in the JA-treated cells.
Fig. 1F JA-treated Wt cells showed reduced bead binding but accumulation of actin around bound beads was not affected.
Fig. 1G Treatment with JA did not affect targeting of talin to bound beads in gelsolin null cells.
Fig. 1H Histogram shows comparison of % collagen bead internalization in gelsolin null cells and NMIIA siRNA knockdown cells.
Fig. 1I Gelsolin Wt cells were treated with JA and incubated with or without ionomycin. There were more beads bound in the presence of ionomycin compared to cells without ionomycin treatment. Localization of NMIIA was not affected in either sample.

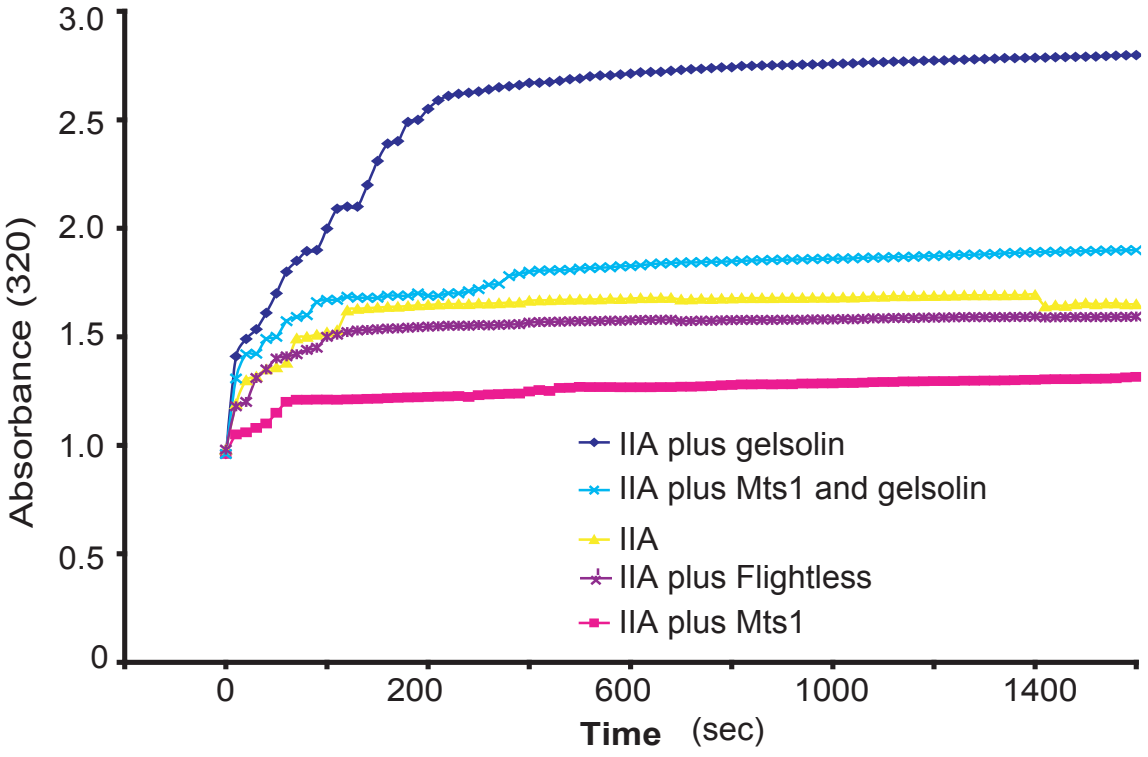
Fig. 2 In an *in vitro* study we determined the impact of gelsolin on non-muscle myosin filament assembly. The kinetics of myosin assembly were measured in a UV/visible spectrophotometer Ultrospec-3000 (Pharmacia, Biotech). Turbidity was measured at 320 nm at room temperature as described(1). Briefly, 200 μL samples were prepared and measured in a quartz cuvette for 30 min. The samples consisted of 10-20 μM NMIIA rods (in 20 mM Tris pH 7.5, 0.6 M NaCl, 1 mM DTT, 0.2% NaN_3) that were polymerized by 4-fold dilution into a buffer containing 20 mM Tris pH 7.5, 2.5 mM $MgCl_2$, 1 mM DTT, 0.02% NaN_3 and 0.4 $CaCl_2$ resulting into 150 mM NaCl, 2 mM $MgCl_2$ and 0.3 mM $CaCl_2$. We determined the effect of mts1(1) on myosin assembly. mts1, which is also known as S100A4, FSP1, CAPL, calvasculin, metastasin, p9Ka, 18A2, and pEL98, is a member of the S100 family of Ca^{2+}-binding proteins that may regulate cell migration and cancer metastasis and we used here to inhibit NMIIA filament formation(1). Myosin rods (18 μM) were incubated with 4-12 μM mts dimer for 20 min and then diluted four-fold as described above. Similarly, 6-14 μM gelsolin was incubated with 18 μM NMIIA rods in 20 mM Tris pH 7.5, 0.6 M NaCl, 1 mM DTT, 0.2% NaN_3, 0.3 mM $CaCl_2$ and then diluted 4-fold to induce polymerization.

1. Li, Z. H., Spektor, A., Varlamova, O., and Bresnick, A. R. (2003) *Biochemistry* **42**, 14258-14266



Supplementary Fig.1

A



Supplementry Fig2