## Supplementary figure legends

Figure S1. Spindlin1 does not localize to the spindle or centromere in mitotic mouse fibroblast cells. (A) Co-staining for Spindlin1 and Tubulin in prometaphase cells; (B) co-staining for Spindlin1 and Tubulin in anaphase cells; (C) co-staining for Spindlin1 and Tubulin in telophase cells; (D) co-staining for Spindlin1 and Tubulin in interphase cells.

Figure S2. Quality control of the mono-nucleosomes. (A) Mono-nucleosomes on a 1.5% agarose gel; (B) Verification of the MLA reaction products with antibodies specific to H3K4me2 and H3K4me3.

Figure S3. Spindlin1 does not bind to H3K9me3, H3K27me3 and H3K4me0. (A) Pull-down experiments showed no specific interaction between Spindlin1 and mono-nucleosomes containing H3Kc9me3 and H3Kc27me3; (B) ITC experiments showed no interaction between Spindlin1 and H3K4me0 peptide; (C) ITC experiments showed no interaction between Spindlin1 and H3K9me3 or H3K27me3 peptides.

Figure S4. (A) Flag-Spindlin1 Y170A and F141A mutants displayed slightly reduced rDNA occupancy in ChIP experiments; (B) siRNA 3 for Spindlin1 specifically targets Spindlin1, but not Spindlin2A/2B (2A and 2B differ for only two bases in the coding region, which can be amplified using the same primer pair).

Figure S5. Nucleotide sequence alignment for Spindlin family proteins.

Figure S6. Spindlin1 does not appear to form a stable complex with other proteins in HeLa cells. Silver-stained SDS/PAGE gel of anti-Flag affinity-purified samples from HeLa cells expressing Flag-Spindlin1 and mock HeLa cells. * Co-purified common contaminants were verified using mass spectrometry analysis.

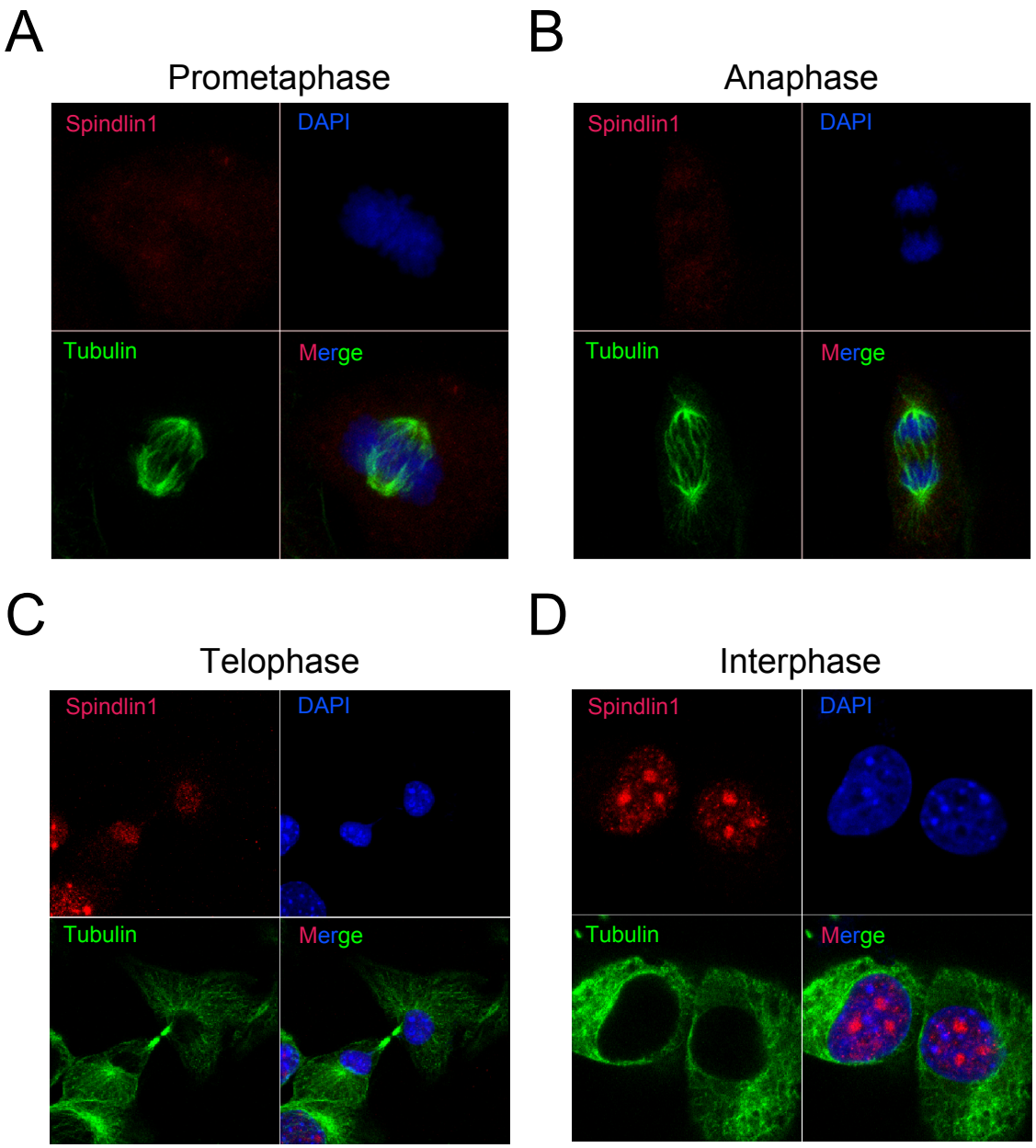

A
Prometaphase
Spindlin1
DAPI
Tubulin
Merge
B
Anaphase
Spindlin1
DAPI
Tubulin
Merge
C
Telophase
Spindlin1
DAPI
Tubulin
Merge
D
Interphase
Spindlin1
DAPI
Tubulin
Merge

Figure S1

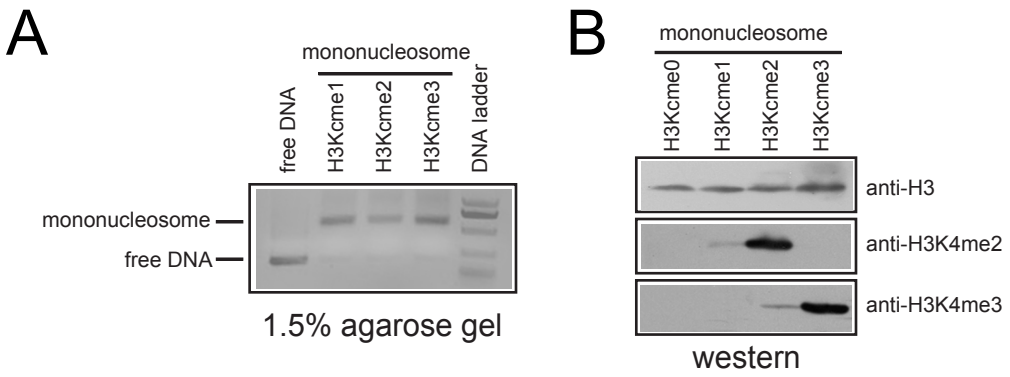

A
mononucleosome
free DNA
H3Kcme1
H3Kcme2
H3Kcme3
DNA ladder
mononucleosome
free DNA
1.5% agarose gel
B
mononucleosome
H3Kcme0
H3Kcme1
H3Kcme2
H3Kcme3
anti-H3
anti-H3K4me2
anti-H3K4me3
western

Figure S2

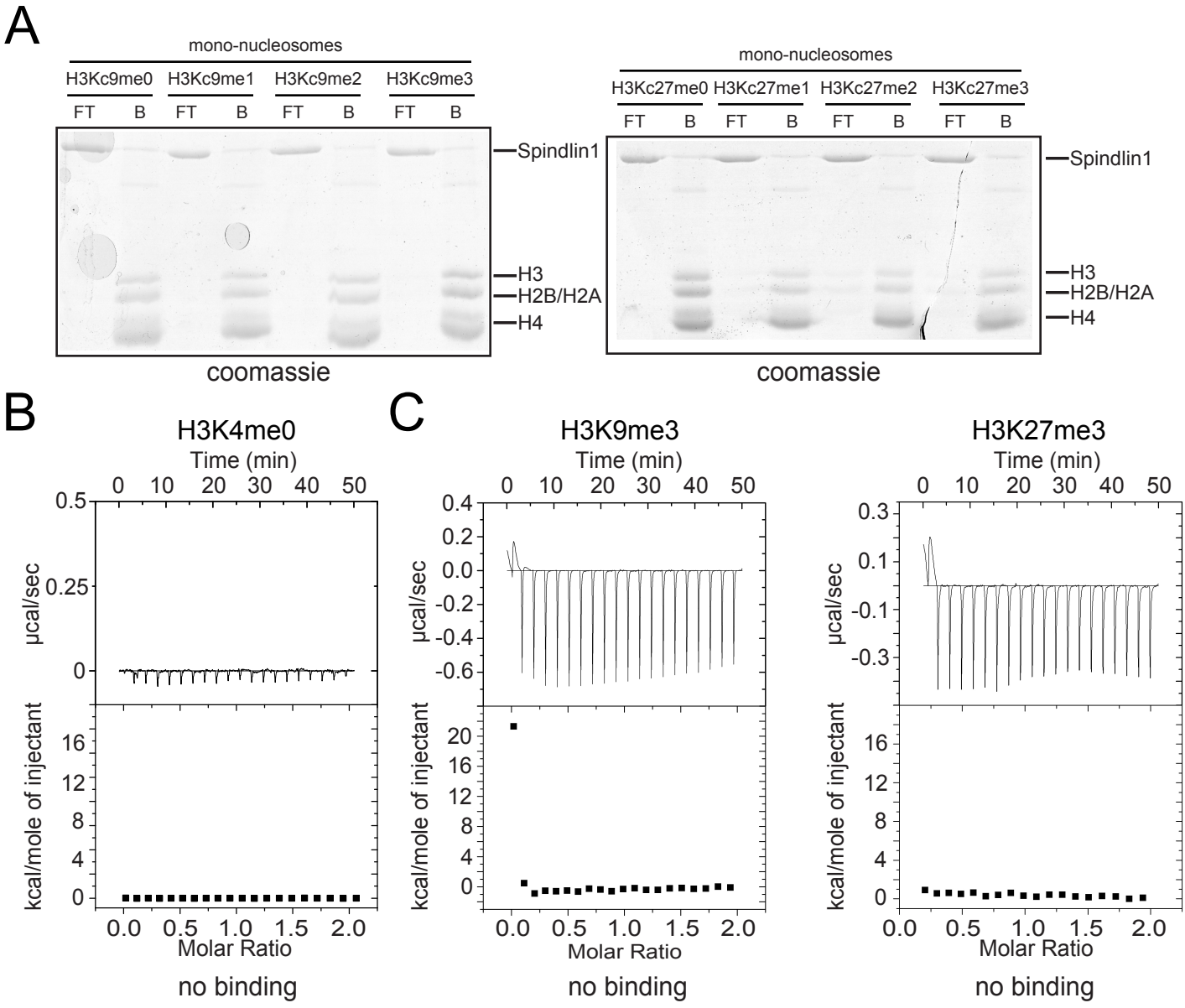
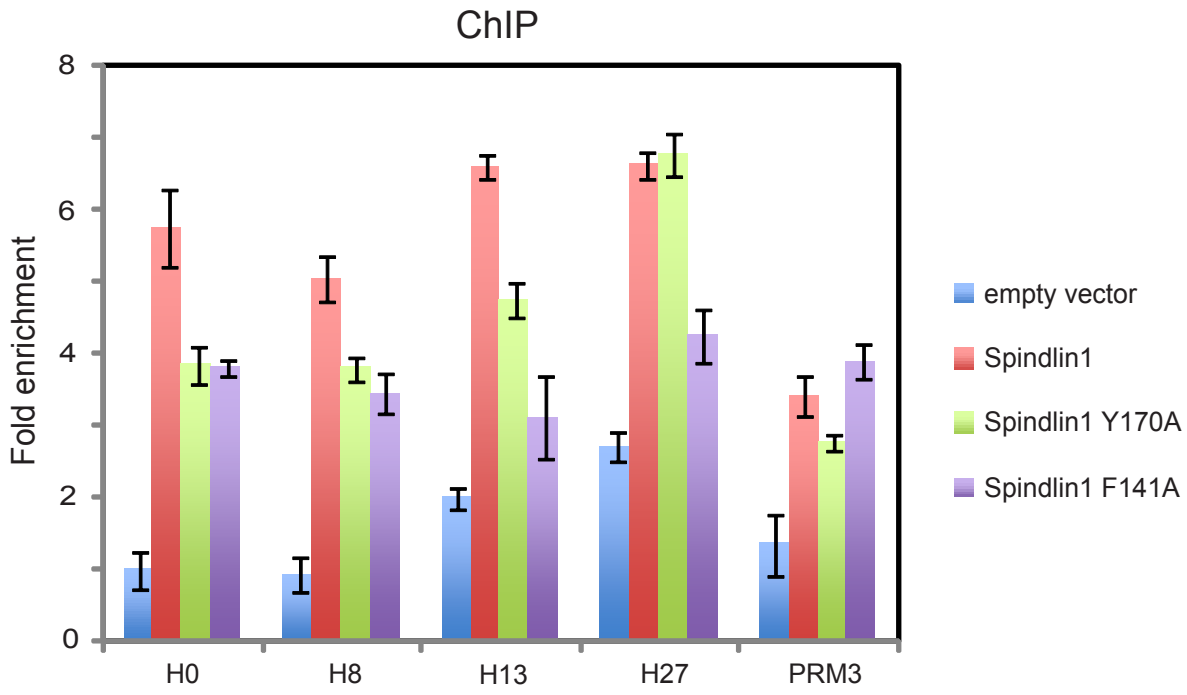

A
mono-nucleosomes
H3Kc9me0
H3Kc9me1
H3Kc9me2
H3Kc9me3
FT
B
Spindlin1
H3
H2B/H2A
H4
coomassie
mono-nucleosomes
H3Kc27me0
H3Kc27me1
H3Kc27me2
H3Kc27me3
coomassie
B
H3K4me0
C
H3K9me3
H3K27me3
Time (min)
µcal/sec
kcal/mole of injectant
Molar Ratio
no binding

Figure S3

A


ChIP
Fold enrichment
8
6
4
2
0
H0
H8
H13
H27
PRM3
empty vector
Spindlin1
Spindlin1 Y170A
Spindlin1 F141A

B

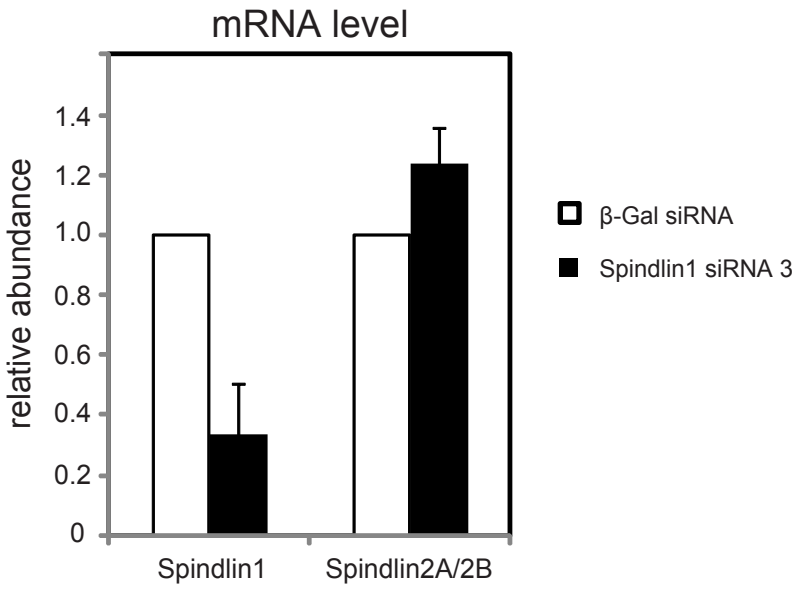

mRNA level
relative abundance
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0
Spindlin1
Spindlin2A/2B
β-Gal siRNA
Spindlin1 siRNA 3

Figure S4

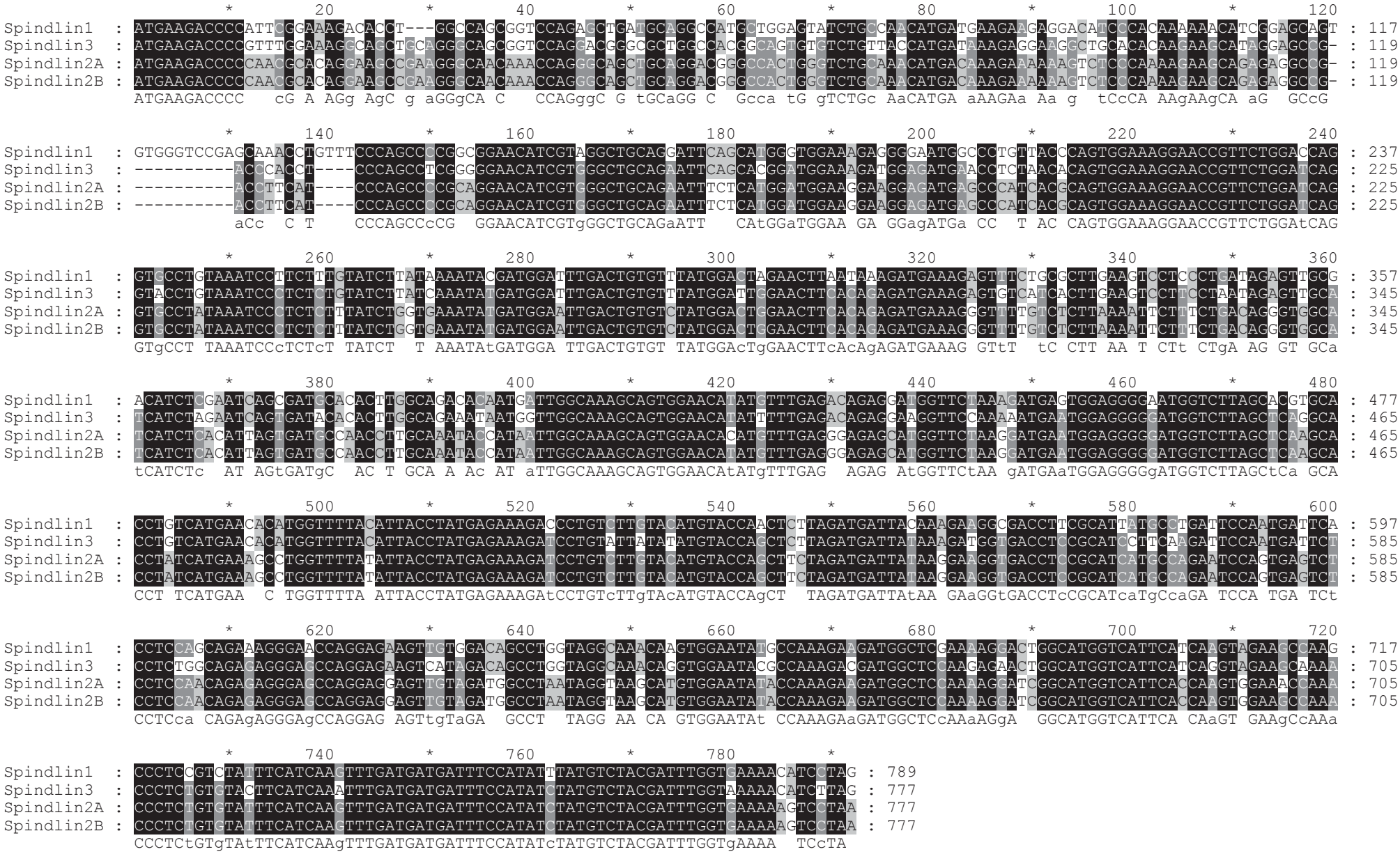



Figure S5

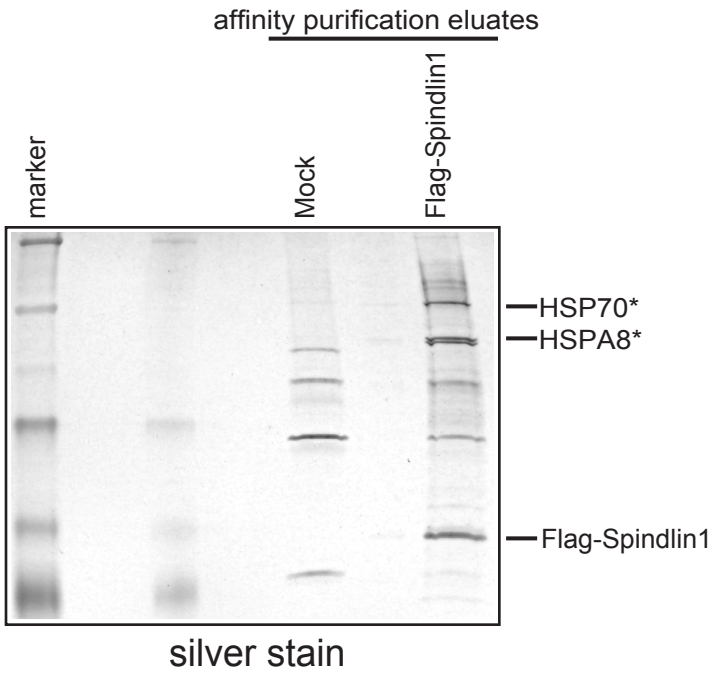

affinity purification eluates
marker
Mock
Flag-Spindlin1
HSP70*
HSPA8*
Flag-Spindlin1
silver stain

Figure S6