A

siRNA	CD41+ (%)	CD41 + (MFI)	HA + (%)	HA (MFI)	CD41+HA+ (%)
CTL	87.4 ± 2.2	5422 ± 339.4	78.2 ± 2.8	4430.3 ± 133.2	62.9 ± 1.3
TLN1	88.4 ± 1.6	5384 ± 307.2	74.2 ± 2.6	3774.0 ± 264.5	61.8 ± 1.2

The mean percentage of CD41+, HA+, CD41+HA+ cells and mean fluorescence intensity (MFI) ± SD from 3 independent experiments are represented.

B

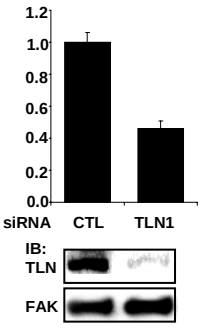

Fig. S7. Additional controls demonstrating that transfected cells adhered to fibrinogen are depleted of talin1 and express HA-FlLGAP$^{\Delta GAP}$ and the αIIb-ΔL β3-1A WT integrin. (A-B) CHO K1 cells were transiently transfected with control or talin1 siRNA for 48 hours and then co-transfected for 24 hours with expression vectors for the αIIb-ΔL and β3-1A WT integrin subunits ± the expression vector for HA-FilGAP$^{\Delta GAP}$. Transfected cells were replated on fibrinogen for 1 hour in CCM1 and then assayed for chimeric integrin, HA expression and talin knockdown. (A) FACS analysis using an antibody to CD41 (PC5), which recognizes the αIIb-β3 extracellular complex, indicated that recombinant integrin expression was not altered by depleting cells of talin1 and that adhering transiently transfected cells to fibrinogen enriches for cells expressing the chimeric integrin (>85% positive). Data represents the percentage (%) and mean fluorescence intensity (MFI) for each condition. Furthermore ~ 60% of these cells expressed HA-FilGAP$^{\Delta GAP}$. Only cells positive for HA were included in our assays for cell spreading. (B) Talin1 knockdown in replated cells was confirmed by western blotting. Plotted are the average expression levels for talin1 ± s.d. from the 3 independent experiments shown in A, together with representative western blots. Effects on spreading were similar to those presented in Fig. 7 (data not shown). Thus, replating cells on fibrinogen enriched for cells expressing both subunits of the chimeric integrins and these adherent cells showed significant inhibition of talin expression when talin1 siRNA was also transfected.