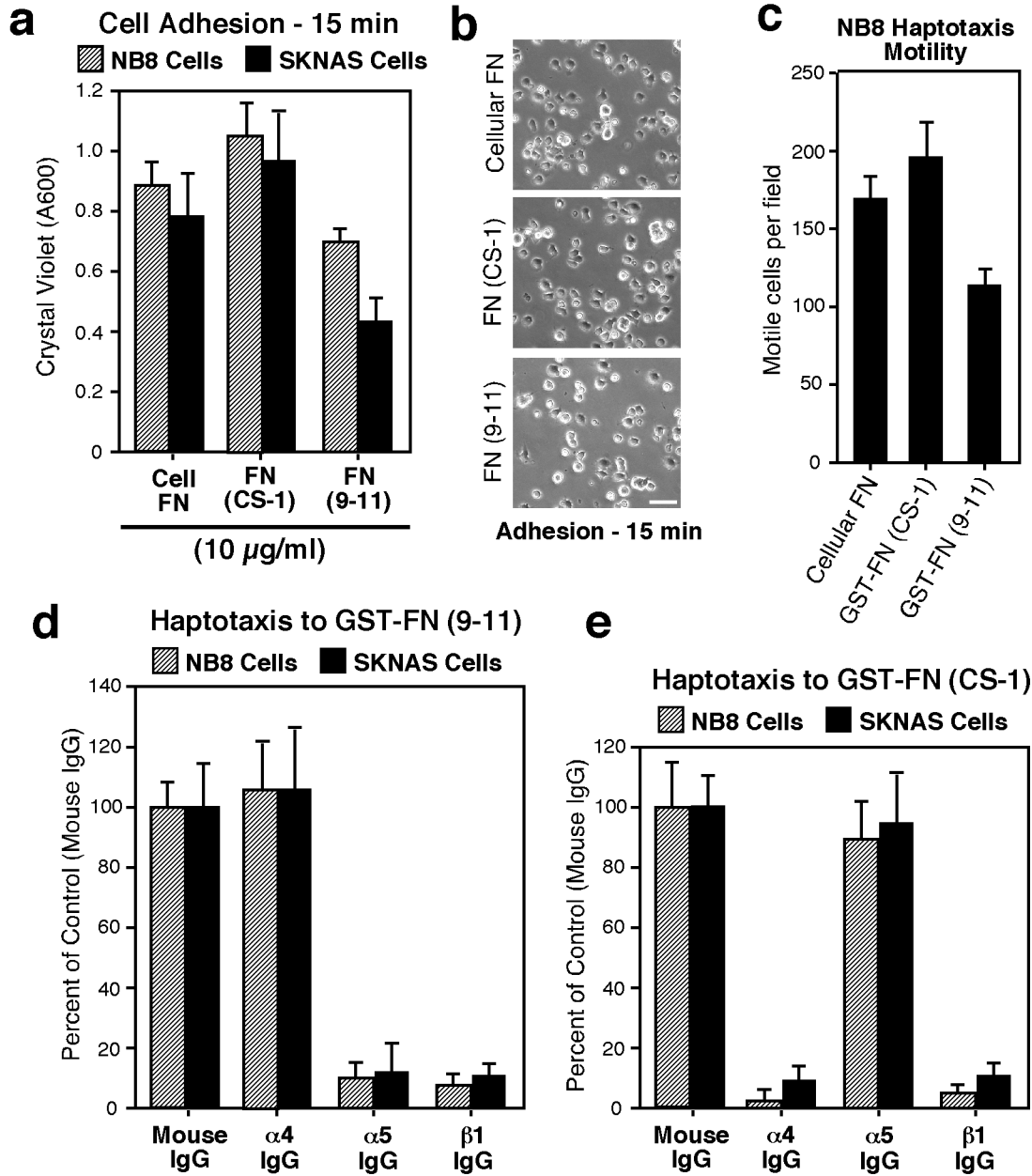
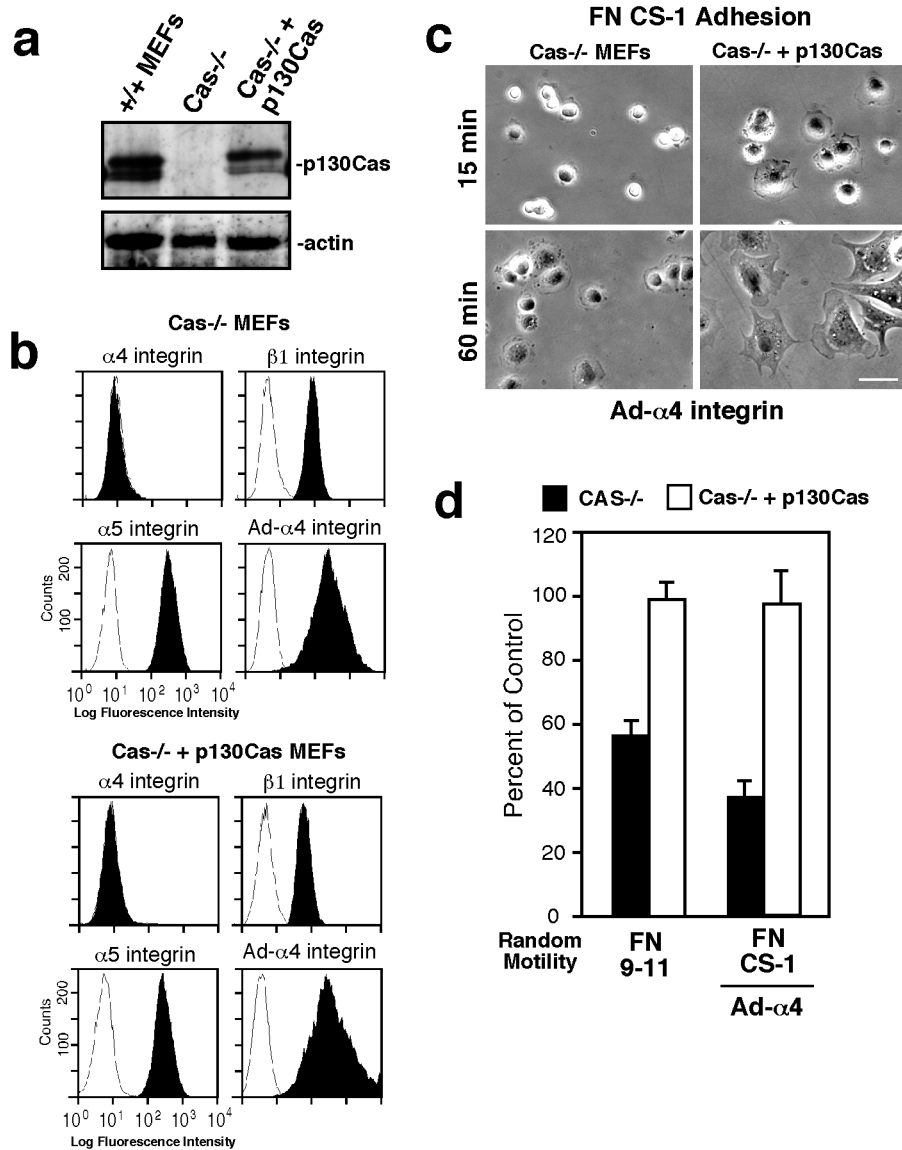


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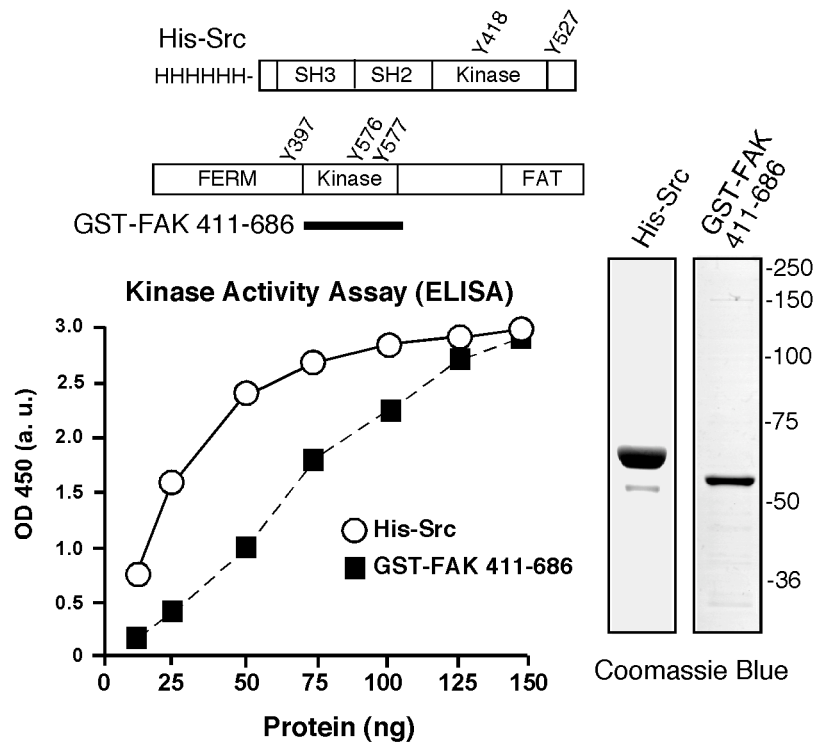
**Supplemental Figure 1.** Recombinant FN (CS-1) or FN (9-11) stimulate  $\alpha 4\beta 1$ - or  $\alpha 5\beta 1$ -mediated NB cell motility, respectively. (a) Quantitative analysis of NB8 and SKNAS cell adhesion after 15 min to cellular FN, FN (CS-1), and FN (9-11). (b) Phase contrast images of NB8 cells adhesion on cellular FN, FN (CS-1), and FN (9-11) after 15 min. Scale bar is 25  $\mu\text{m}$ . (c) NB8 haptotaxis motility performed on cellular FN, FN (CS-1), and FN (9-11). (d) NB cell haptotaxis motility on FN (CS-1) is blocked by anti- $\alpha 4$  mAb (HP2/1, 10  $\mu\text{g}/\text{ml}$ ) and anti- $\beta 1$  mAb (P4C10, 10  $\mu\text{g}/\text{ml}$ ) but not by anti- $\alpha 5$  mAb (P1D6, 10  $\mu\text{g}/\text{ml}$ ). (e) NB haptotaxis cell motility on FN (9-11) is blocked by anti- $\alpha 5$  mAb (P1D6, 10  $\mu\text{g}/\text{ml}$ ) and anti- $\beta 1$  mAb (P4C10, 10  $\mu\text{g}/\text{ml}$ ), but not by anti- $\alpha 4$  mAb (HP2/1, 10  $\mu\text{g}/\text{ml}$ ). (a, c, d, and e) Values are means  $\pm$  SD of triplicates from at least two separate experiments.

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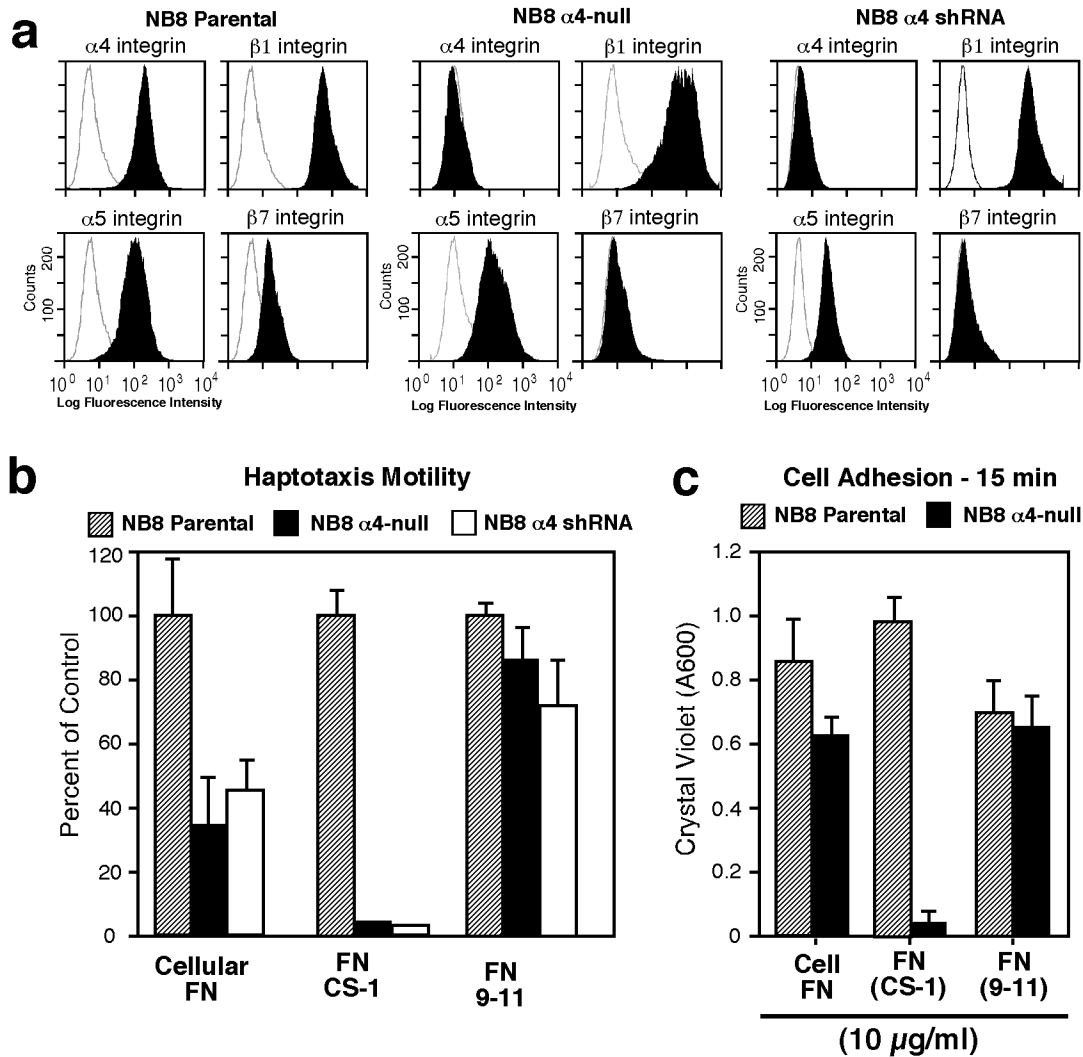
**Supplemental Figure 2.** Human  $\alpha 4$  integrin expression in Cas $^{-/-}$  and Cas $^{-/-}$  + p130Cas fibroblasts. **(a)** Expression of p130Cas in normal mouse embryonic fibroblasts (MEFs), Cas $^{-/-}$ , and Cas $^{-/-}$  + p130Cas fibroblasts were analyzed anti-p130Cas immunoblotting. Actin blotting verified equal loading. **(b)** Flow cytometry analyses of endogenous murine  $\alpha 4$ ,  $\beta 1$ ,  $\alpha 5$ , and adenovirus (Ad)-expressed human  $\alpha 4$  integrin in Cas $^{-/-}$  and Cas $^{-/-}$  + p130Cas cells (shaded peaks). Staining with control mAb (open peaks). **(c)** Phase contrast images showing adhesion and spreading on FN (CS-1) of Cas $^{-/-}$  and Cas $^{-/-}$  + p130Cas cells with human  $\alpha 4$  integrin expression at indicated time points. Scale bar is 15  $\mu$ m. **(d)** Random motility assay on FN (9-11) or FN (CS-1) of Cas $^{-/-}$  and Cas $^{-/-}$  + p130Cas cells with or without human  $\alpha 4$  integrin expression revealed that p130Cas promotes  $\alpha 5\beta 1$ - and  $\alpha 4\beta 1$ -mediated cell motility.

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**Supplemental Figure 3.** Activity profile of recombinant His-Src and GST-FAK 411-686. Schematic of His-tagged Src and the FAK kinase domain region (411-686) expressed in baculovirus and initially isolated by Ni-NTA and glutathione agarose, respectively. After size fractionation chromatography, the purity of His-Src and GST-FAK 411-686 was >90% as visualized by SDS-PAGE and Coomassie Blue staining. Shown is an ELISA-based activity assay measuring the phosphorylation of poly Glu:Tyr (4:1).

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**Supplemental Figure 4.** Abolishing  $\alpha4$  integrin expression inhibits  $\alpha4\beta1$ - but not  $\alpha5\beta1$ -stimulated NB motility. **(a)** Flow cytometry analyses of  $\alpha4$ ,  $\alpha5$ ,  $\beta1$ , and  $\beta7$  integrin expression (shaded peaks) in NB8 parental, NB8  $\alpha4$ -null sorted, and  $\alpha4$  shRNA-expressing cells. Open peaks show staining with control mAb. **(b)** Haptotaxis motility assays show that loss of  $\alpha4$  integrin expression prevents FN (CS-1) but not FN (9-11)-stimulated NB cell motility and inhibits migration on cellular FN. Values presented are percent of NB8 parental cell motility on cellular FN (Control). **(c)** Quantitative analysis of NB8 parental and  $\alpha4$ -null cell adhesion to cellular FN, FN (CS-1), and FN (9-11) after 15 min. **(b and c)** Values are means  $\pm$  SD of triplicates from two separate experiments.

Wu et. al., Supplemental Figure 5

