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## Vascular Endothelial-Cadherin Regulates Cytoskeletal Tension, Cell Spreading, and Focal Adhesions by Stimulating RhoA

Mol. Biol. Cell Nelson et al. 15: 2943

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## Supplemental Material

**This article contains the following supporting material:**

- [Supplemental Table.pdf](#)
- [Supplemental Figure 1.pdf](#) - Cell-cell contact decreases cell spreading and FA formation. (A-D) Dot plots of total FA area (A, C) and number (B, D) as a function of cell spreading for vinculin (vin), talin (tal), FAK and phosphotyrosine (pY) for cells plated randomly (A, B) and at different seeding densities (C, D). (E, F) Histogram graphs of total FA area (E) and number (F) for vinculin (vin), talin (tal), FAK and phosphotyrosine (pY) in cells plated at different seeding densities. Colors of dot plots and histograms indicate seeding densities of 300 (black), 3000 (green), and 30,000 (red) cells/cm<sup>2</sup>. (\*) p<0.05, (\*\*) p<0.005 between cells cultured at 3000 (green) or 30,000 (red) cells/cm<sup>2</sup> compared to cells at 300 cells/cm<sup>2</sup> as calculated by non-parametric median test.
- [Supplemental Figure 2.pdf](#) - VE-cadherin-mediated decrease in cell spreading and FA formation is inhibited by blocking VE-cadherin engagement. (A, B) Dot plots of total FA area and number as a function of cell spreading for cells infected with Ad-GFP (A) or Ad-VEΔ (B) and plated at different seeding densities. (C, D) Histogram graphs of total FA area and number for cells infected with Ad-GFP (C) or Ad-VEΔ (D) plated at different seeding densities. (\*) p<0.005, (\*\*) p<0.0001 as calculated by non-parametric median test.
- [Supplemental Figure 3.pdf](#) - Cell-cell contact increases FA formation when spreading is controlled. Histogram graphs of total FA area (A) and number (B) for vinculin (vin), talin (tal), FAK, and phosphotyrosine (pY) in single cells (dotted lines) and pairs of cells (solid lines). (\*) p<0.05, (\*\*) p<0.005 as calculated by non-parametric median test.
- [Supplemental Figure 4.pdf](#) - Timing of localization of cell-cell adhesion components coincides with that of RhoA activation. (A, B) Fluorescence images of Ad-GFP- and Ad-VEΔ-infected cells stained for β-catenin (A) and actin (B) at different times after plating. Bar = 25 μm. (C) Higher magnification fluorescence images of Ad-GFP-infected cells stained for β-catenin and VE-cadherin at different times after plating. Bar = 20 μm.

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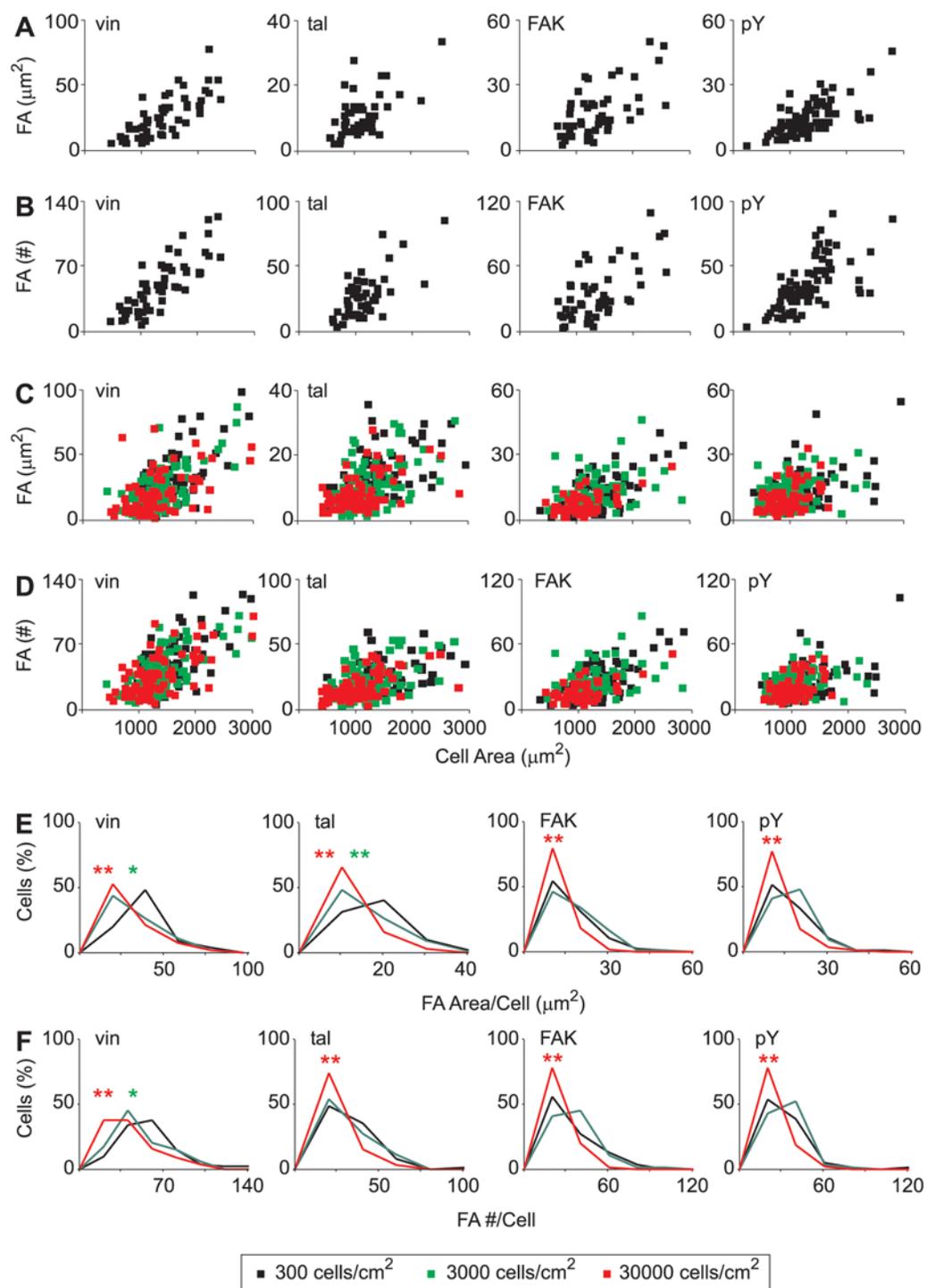
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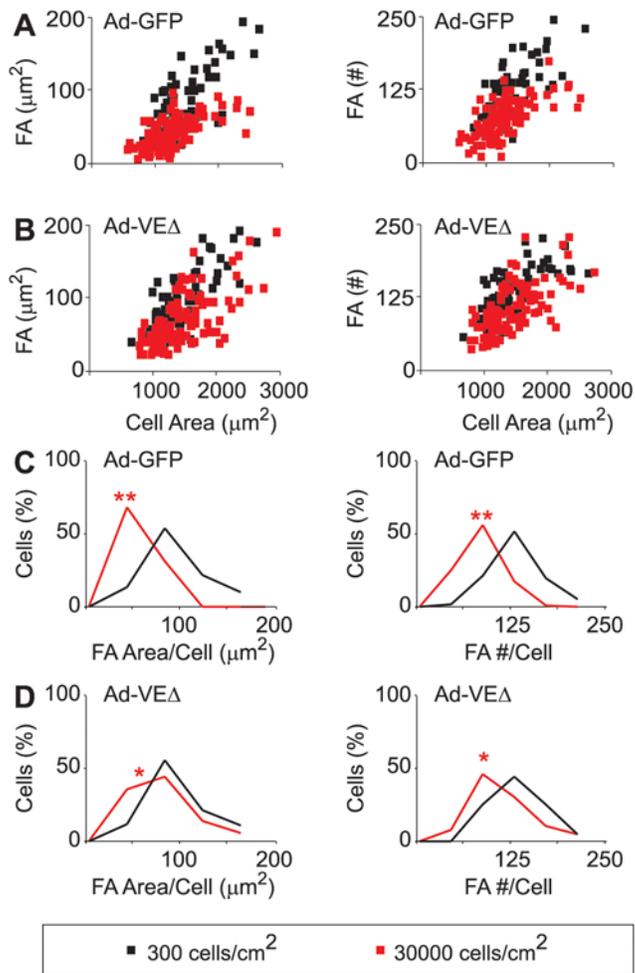
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	Average total FA area		Average # FA	
	$\mu\text{m}^2$ ( $\pm$ st. dev.)		( $\pm$ st. dev.)	
	singles	pairs	singles	pairs
vinculin	15 ( $\pm$ 1)	25 ( $\pm$ 2)	34 ( $\pm$ 3)	49 ( $\pm$ 3)
talin	6 ( $\pm$ 1)	9 ( $\pm$ 1)	15 ( $\pm$ 2)	22 ( $\pm$ 2)
FAK	5 ( $\pm$ 1)	8 ( $\pm$ 1)	13 ( $\pm$ 1)	19 ( $\pm$ 2)
pY	9 ( $\pm$ 1)	15 ( $\pm$ 1)	19 ( $\pm$ 2)	30 ( $\pm$ 2)

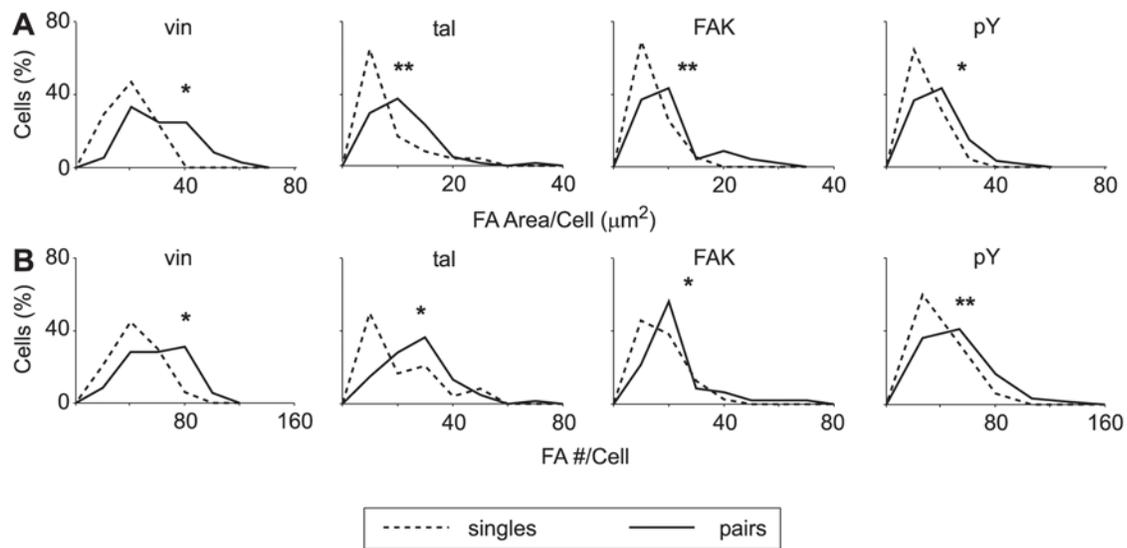
Supplemental Table 1. Quantitative analysis of total FA area and number for vinculin, talin, FAK, and phosphotyrosine (pY) in single cells and pairs of cells cultured in bowtie-shaped patterns. Data are represented graphically in Figure 4 and Supplemental Figure 4.



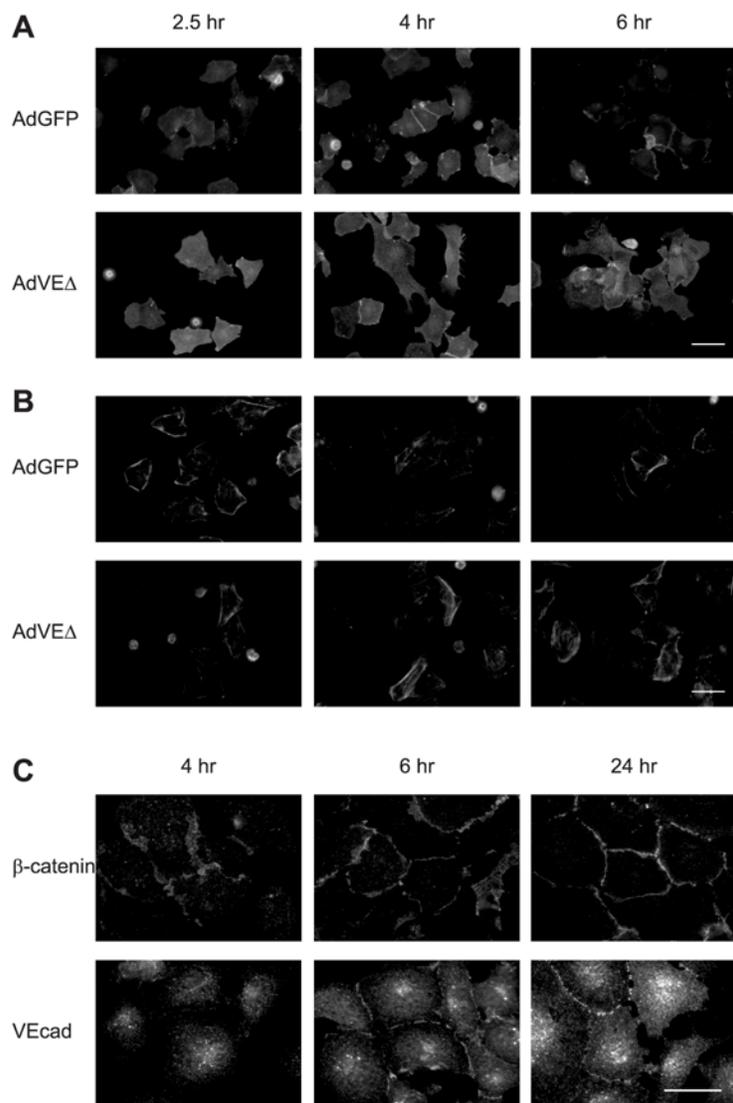
Supplemental Figure 1



Supplemental Figure 2



**Supplemental Figure 3**



Supplemental Figure 4