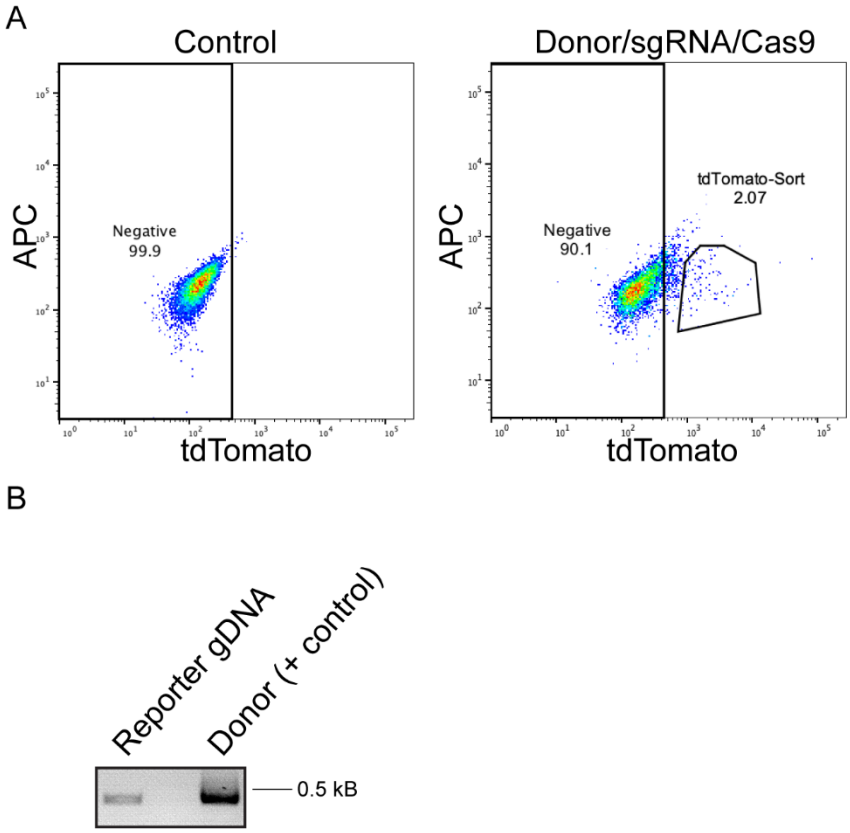


Fig. S1. Generation of integrin β4 reporter 4T1 cells. (A) 4T1 cells were transfected with donor plasmid and sgRNA #2/Cas9 and processed for flow cytometry to quantify tdTomato positive cells, which were subsequently processed for single-cell sorting. (B) Genomic DNA from clones described (A) was isolated and processed for PCR to determine the correct genomic insert of tdTomato.

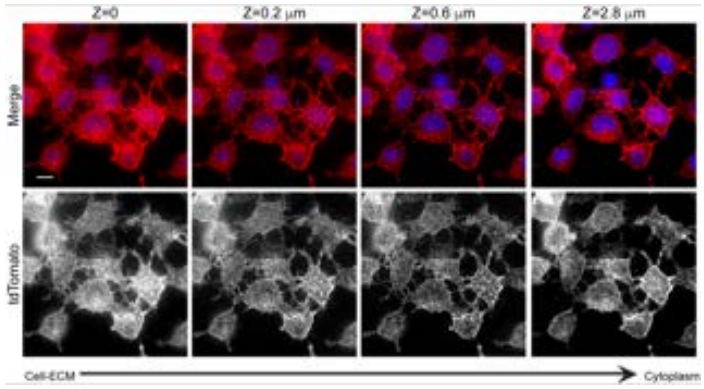
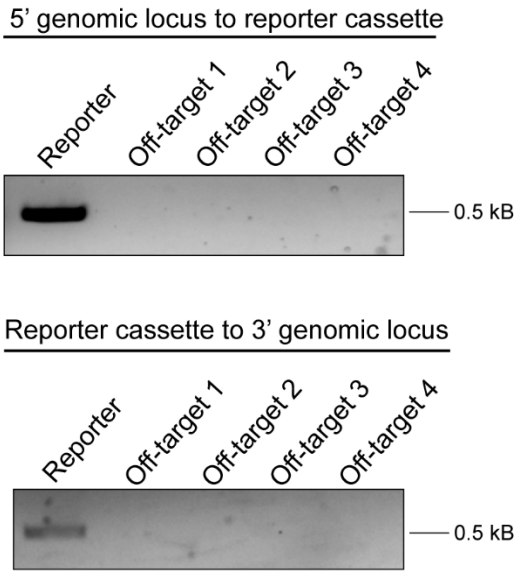


Fig. S2. β4-tdTomato is expressed on the surface of comma-d1 reporter cells. A Z-stack was performed to refine the localization of the tdTomato signal in adherent reporter cells. Z-stack slices moving from the basal surface (Z=0 microns) towards the apical surface (Z=2.8 microns) demonstrate that the tdTomato signal is predominately on the cell surface. Top panel: Live cell fluorescence images show the distribution of tdTomato (red), and nuclei (blue). Bottom panel: Grey scale images show tdTomato only. Scale bar: 10 microns.



Rank	Genomic Locus	Target Sequence	Primer Sequences
1	chr12: 100944782-100944804	AGGGGGCGGGGGGGAGGTTC	F: agagcagcacccacaagtct
			R: caggggaaaacatctcagga
2	chr13:4621969-4621991	AAGGGGCGTGGGGGAGGTTC	F: ccaaaggctgctagtggaag
			R:gagtagcggccagagaaatg
3	chr6: 70352481-70352503	CTCGGGGGGGGGGGAGGTTC	F: atcccaaaagtcccccatac
			R: acccctgtctccatctgttg
4	chr11:119990864-119990886	CTGGAGCACTGGGGAGGTTC	F: ggaatgagtgggatccaaga
			R: accagtctgagaaggcctga

Fig. S3. Off-target analysis of integrin β4 reporter comma-d1 cells. Genomic DNA was isolated from comma-d1 reporter cells and the indicated potential off-target sites were screened by PCR to determine reporter cassette integration. The four genomic loci screened, and the primer sequences utilized are also shown.

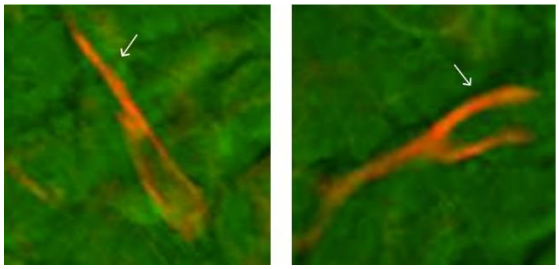
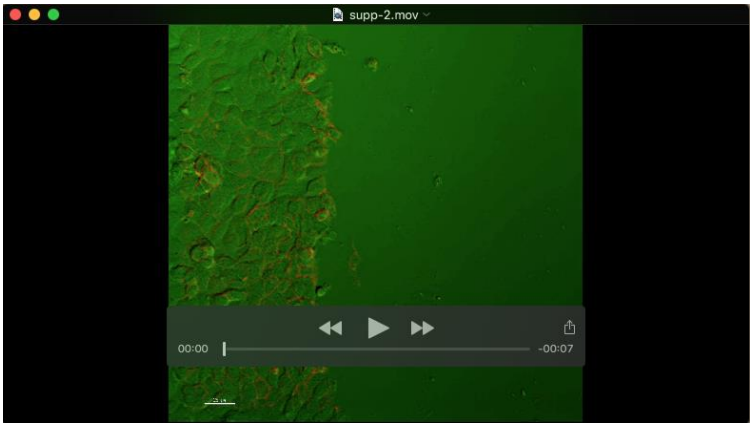
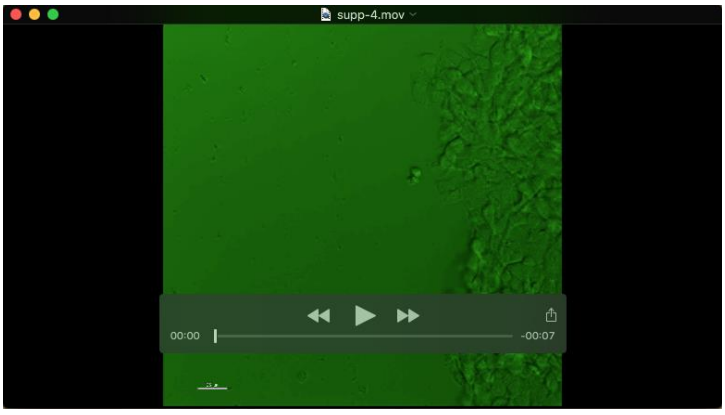


Fig. S4. Integrin β4 is present in cell protrusions of migrating cells. Inset of still images of migrating cells from Movies 3 and 5 that show tdTomato/β4 in cell protrusions formed by migrating cells. Arrows indicate cell protrusions.

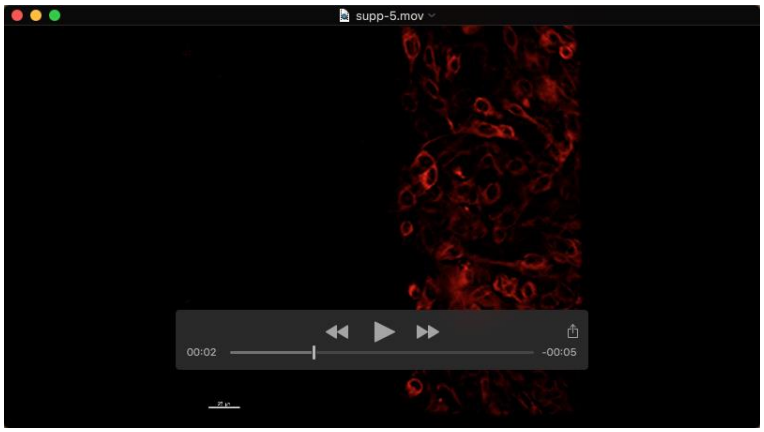


Movie 1. 18-hour movie of control β4 reporter comma-d1 cells in response to a scratch wound using a green differential interference contrast (DIC) background. Scale bar represents 25 micrometers.

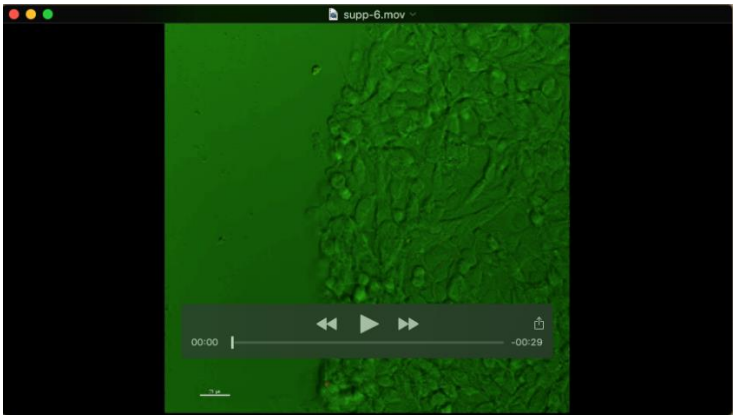
Movie 2. Movie 1 without a DIC background. Scale bar represents 25 micrometers.



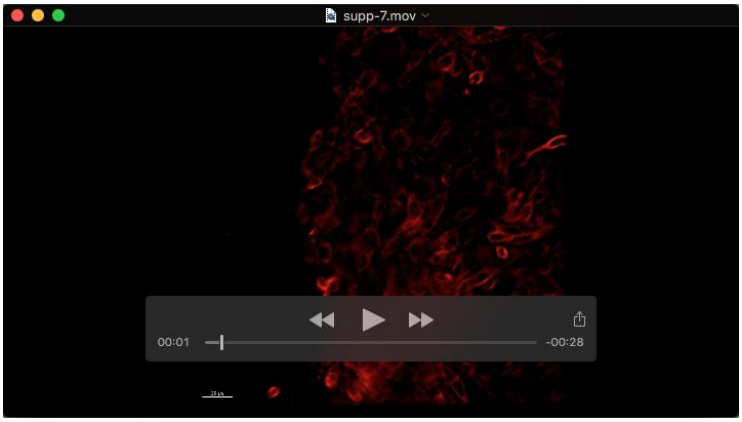
Movie 3. An 18-hour movie of YAP-transformed β4 reporter comma-d1 cells in response to a scratch wound using a green DIC background. Scale bar represents 25 micrometers.



Movie 4. Movie 3 without a DIC background. Scale bar represents 25 micrometers.



Movie 5. 72-hour movie of YAP-transformed β4 reporter comma-d1 cells in response to a scratch wound using a green DIC background. Scale bar represents 25 micrometers.



Movie 6. Movie 5 without a DIC background. Scale bar represents 25 micrometers.