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**Supplementary Fig. 1. scrib\(^{KD}\) cells grow more slowly than wild-type cells and are compacted and eliminated when surrounded by wild-type cells.**

**a, b,** Time-course of cell competition assays between unlabelled wild-type (WT) and GFP labelled scrib\(^{KD}\) cells (a) and mock competition between unlabelled scrib\(^{KD}\) and GFP labelled scrib\(^{KD}\) cells (b), see corresponding Supplementary Movie 1. Inset below (a) shows the GFP channel from corresponding stills. **c,** Schematic representation of the experimental set-ups for experiments with competition-conditioned or mock-conditioned medium (as in Fig. 1b) and with transwells (as in Fig. 1c). **d,** Quantification of the growth rate of scrib\(^{KD}\) cells with or without the addition of tetracycline (TET). Each dot represents the average of 4 fields of cells (n). **e,** Average cell density of pure confluent scrib\(^{KD}\) cells and of subconfluent competing scrib\(^{KD}\) cells that are either entirely surrounded by WT cells (surrounded clones) or merely contacting WT clones (peripheral clones). Despite being subconfluent, scrib\(^{KD}\) cells in competing cultures have a higher density than in confluent pure cultures; n = cell number, mean ± sem. Scale bars: movie sequences = 100μm and immunofluorescence images = 50μm here and throughout all Supplementary Figures. **p** < 0.005, ***p** < 0.0005 by T-test.
Supplementary Fig. 2. E-cadherin is upregulated in scribKD cells and E-cadherin upregulation is not sufficient to induce directional migration.

**a-d**, Individual cell trajectories of wild-type (WT) (a) or scribKD (c) cells before or after contact as indicated, and corresponding average directionalities. (b, d). Error bars = SD; quantifications from pooled data from 3 biological replicates; **p<0.0005 by T-test.**

**e**, Anti-E-cadherin staining of co-cultures of WT and GFP labelled scribKD cells shows accumulation of E-cadherin in scribKD cells compared to WT cells (sum intensity projection).

**f**, Western blot against E-cadherin in WT and scribKD cells +/- doxycycline (DOX). β-tubulin was used as loading control.

**g**, Cell surface E-cadherin staining of RFP labelled WT and unlabelled scribKD co-cultures shows surface accumulation of E-cadherin in scribKD cells compared to WT cells.

**h**, Quantification of single cell E-cadherin intensities from one representative set of confocal images as in Fig. 2q, rightmost panel; black bars = median; * p<0.05 by KS test.

**j**, Stills from time-lapse movie of RFP labelled WT and unlabelled scribKD E-cadKD co-cultures showing that the latter are still eliminated by WT cells despite lack of contact-induced migration. Asterisks mark individual scribKD E-cadKD cells; black arrows = cell death events.

**k**, Immunofluorescence staining comparing E-cadherin levels between WT cells (within white dashed line), scribKD cells with nuclear GFP, and cells overexpressing GFP labelled E-cadherin (E-cad OE; within yellow dashed
line). Co-culture of RFP labelled WT cells and *E-cad* OE cells shows no directional migration upon contact (white dashed line = initial point of contact; black dashed line = final point of contact). n = number of cells.
**Supplementary Fig. 3. Characterisation of scrib<sup>RES</sup> cells and analysis of p21 and p53 expression in scrib<sup>KD</sup> cells.**

**a,** Western blot against Scribble for scrib<sup>KD</sup> and scrib<sup>RES</sup> cells +/- TET. **b,** Surface E-cadherin staining of GFP labelled scrib<sup>RES</sup> cells mixed with unlabelled scrib<sup>KD</sup> cells. **c,** d, Stills from time-lapse movies of wild-type (WT) and GFP labelled scrib<sup>RES</sup> cells to assess directional migration (c) and competition (d), see corresponding Supplementary Movie 10. White dashed line = initial point of contact; black dashed line = final point of contact. **e,** p21 staining of pure WT cells and pure scrib<sup>KD</sup> cells. Note widespread p21 elevation in scrib<sup>KD</sup> cells. **f,** Loss of Scribble leads to an overall
increase in nuclear p53 staining. n = cell number; ** p<0.005 by Wilcoxon rank sum test.
Supplementary Fig. 4. Loss of p53 activity blocks elimination of *scrib*<sup>KD</sup> cells, but does not prevent up-regulation of E-cadherin or contact-induced migration.

**a**, Anti-p53 staining of GFP labelled *scrib*<sup>KD</sup> or GFP labelled *scrib*<sup>KD</sup> *p53<sup>+/−</sup>* cells in competition with wild-type (WT) cells. Two fields of similar cell densities are shown for comparison.

**b**, Western blot against p53 and p21 in *scrib*<sup>KD</sup> and *scrib*<sup>KD</sup> *p53<sup>+/−</sup>* cells +/- UV-C irradiation; β-tubulin as loading control.

**c**, Anti-p21 staining of *scrib*<sup>KD</sup> *p53<sup>+/−</sup>* cells when cultured next to *scrib*<sup>KD</sup> or WT cells (all images are taken from the same coverslip using the fence system).

**d**, Single cell nuclear p21 intensity from confocal images as in (c); *** p<0.0005 by KS test.

**e**, Western blot against p21 in *scrib*<sup>KD</sup> and *scrib*<sup>KD</sup> *p53<sup>+/−</sup>* cells +/- TET; β-tubulin as loading control.

**f**, Stills from time-lapse movies of WT and GFP labelled *scrib*<sup>KD</sup> competition assays in the presence of Pifithrin-α (10μM).

**g**, Anti-E-cadherin staining of GFP labelled *scrib*<sup>KD</sup> or GFP labelled *scrib*<sup>KD</sup> *p53<sup>+/−</sup>* cells co-cultured with WT cells.

**h**, Stills from time-lapse movies with WT and GFP labelled *scrib*<sup>KD</sup> *p53<sup>+/−</sup>* cells. n = cell number. 2 independent repeats per experiment for (a, b, d).
Supplementary Fig. 5. *scrib*Δ/Δ clones are eliminated in a p53-independent manner in *Drosophila* wing discs.

a, Scribble immunostaining of *Drosophila* wing disc with GFP labelled clones expressing Scribble RNAi; 1 experimental repeat; n = 6 wing discs. The same result was obtained with several independent drivers. 

b, XY and XZ views of DE-cadherin staining of *Drosophila* wing disc with GFP labelled Scribble RNAi clones. XY view shows average projection of multiple z-sections acquired at the plane of the adherens junctions; 1 experimental repeat; n = 4 wing discs. The same result was obtained with several independent drivers. 

c, Representative images of 48 hour old GFP labelled *scrib*Δ/Δ clones (control) or *scrib*Δ/Δ clones overexpressing p53ΔN; 2 experimental repeats; n > 3 wing discs per condition/experiment. The same result was obtained with a validated p53RNAi line.
Supplementary Fig. 6. ROCK is required for P-myosin increase. 

(a, b) Active P-myosin II (phospho S20) immunofluorescence staining of GFP labelled scrib\textsuperscript{KD} and unlabelled wild-type (WT) co-cultures without (a) or with (b) addition of ROCK inhibitor (Y27632).
Supplementary Fig. 7 Neither S1P2 nor Piezo are required for scrib<sup>KD</sup> cell elimination.

**a, b.** Stills from time-lapse movies of WT and scrib<sup>KD</sup> co-cultures in the presence of a S1P2 inhibitor (JTE013) (a) or Piezo inhibitor (gadolinium III chloride) (b).
Supplementary Fig. 8. p53 activation is sufficient to cause flattening of wild-type MDCK cells.

a, Stills from time-lapse movies of co-cultures of GFP labelled wild-type (WT) and unlabelled p53⁻/⁻ MDCK cells. b, Stills from time-lapse movies of pure WT MDCK cells with Nutlin-3 (8μM, see Supplementary Movie 13 right).
Supplementary Fig. 9. Un-cropped original Western blots.

a, Western blots showing p21 and loading control actin from Fig. 3b. b, Western blots showing E-cadherin and loading control β-tubulin from Supplementary Fig. 2f. c, Western blots showing Scribble and loading control actin from Supplementary Fig. 3a. d, Western blots showing p53, p21 and loading control β-tubulin from Supplementary Fig. 4b. e, Western blots showing p21 and loading control β-tubulin from Supplementary Fig. 4e. Black box represents the area of each blot used in each Figure.