Fernandez-Gonzalez et al.
Figure S1. Wounded cells extrude basally in early *Drosophila* embryos.

Epidermal cells expressing GFP:Rho1 in a wounded stage 7 embryo. Columns correspond to different time points with respect to the wound, rows indicate different depths with respect to the vitelline membrane. The red target indicates the interface ablated to wound the cells. The yellow lines delineate the wounded cells. Anterior left, dorsal up. Bar = 5 μm.

Figure S2. Hemocytes phagocytose wound debris in the late embryo.

Epidermal cells expressing GFP:Rho1 in a stage 14 embryo. The yellow arrow points to one of the wounded cells. A macrophage (vacuolated cell, arrowhead) comes into the field of view. A new vacuole appears inside the hemocyte (yellow arrow at 8.50 min), corresponding to part of the debris generated at the wound (Stramer *et al.*, 2005). Bar = 5 μm.

Figure S3. Cells around the wound reduce their apical surface before the onset of wound closure in the early embryo.

(A) Apical cell area normalized to the area before wounding for cells immediately adjacent to the wound (adjacent cells, red, \( n = 25 \) cells in three embryos) and cells 1-5 cell diameters away from the wound (distant cells, blue, \( n = 154 \) cells in three embryos) in early embryos. Within 4 sec of wounding, the apical area of adjacent cells increased by 7.3±4% (apical area increased in 64% of the adjacent cells) and the apical area of distant cells decreased by 3.5±1% (apical area decreased in 60% of the distant cells). Shortly after, the apical areas of adjacent cells decreased by 20.5±6% of their initial values within 3 min of wounding (apical area decreased in 80% of the adjacent cells) and the apical areas of distant cells decreased by 9.6±2% of their initial values within 5 min of wounding (apical area decreased in 75% of distant cells).

(B) Same data as in A, plotted with the normalized area of the corresponding wounds (green, \( n = 3 \)).

(A-B) Time is with respect to wounding. Error bars, s.e.m..

Figure S4. Apical area and medial myosin in epidermal cells of early and late *Drosophila* embryos.

(A and B) Epidermal cells expressing E-cadherin:GFP in a stage 7 (A, early) and a stage 14 (B, late) embryo. Bars = 10 μm.
(C) Average apical cell area in early and late embryos. Apical cell area is greater in the early embryo (42.6±0.5 μm²/cell in early embryos, n = 1,468 cells, and 30.0±0.4 μm²/cell in late embryos, n = 1,812 cells, P < 0.001). Error bars, s.e.m.

(D and E) Epidermal cells expressing myosin:GFP in a stage 7 (D) and a stage 14 (E) embryo. Myosin levels were lower in the late embryo, both at cell-cell junctions and at the medial surface of cells, but medial myosin was present in the late embryonic epidermis. The bright structures in the center of E are denticle precursors. Anterior left, dorsal up. Bar = 10 μm.

(F) Ratio of medial-to-junctional myosin in intact cells in the early embryo (red bar, n = 50 cells in 5 embryos) and the late embryo (blue bar, n = 50 cells in 5 embryos). Only smooth cells (outside of denticle belts) were analyzed in the late embryo. The ratio of medial-to-junctional myosin per cell was greater in late embryos (P = 1.8x10⁻⁷). Error bars, s.e.m..

**Movie legends**

**Movie S1. Myosin accumulates in medial networks in wounded epidermal cells of the early Drosophila embryo.**

Epidermal cells expressing E-cadherin:GFP (green) and myosin:mCherry (red) in a stage 7 embryo. The time after wounding is indicated. Anterior left, dorsal up.

**Movie S2. Myosin forms a purse string around wounded epidermal cells in the late Drosophila embryo.**

Epidermal cells expressing E-cadherin:GFP (green) and myosin:mCherry (red) in a stage 14 embryo. The time after wounding is indicated. Anterior left, dorsal up.

**Movie S3. Low energy ablations induce assembly of a myosin purse string around the wound in late embryos.**

Epidermal cells expressing E-cadherin:GFP (green) and myosin:mCherry (red) in a stage 14 embryo. Red circular structures are vacuoles inside macrophages basal to the epidermis. The laser used for wounding was configured to the minimum power that elicited a wound response and was fired 3 times (a single pulse each time) along a 6-μm line across the cell interface. Conversely, we increased the power used to wound early embryos, but this resulted in the formation of large cavitation bubbles that prevented controlled tissue ablation. Anterior left, dorsal up.

**Movie S4. Single cell wound repair in late embryos is driven by the assembly of a myosin purse string around the wound.**
Epidermal cells expressing E-cadherin:GFP (green) and myosin:mCherry (red) in a stage 14 embryo. The laser used for wounding was configured to the minimum power that elicited a wound response and the laser was fired 3 times (a single pulse each time) along a 6-μm line on the medial cortex of a cell. Subcellular wound repair occurred rapidly in late embryos, as previously shown in syncytial Drosophila embryos prior to cellularization (Abreu-Blanco et al., 2011). Anterior left, dorsal up.

**Movie S5. F-actin accumulates on the medial surface of wounded epidermal cells in the early Drosophila embryo forming protrusions and blebs.**

Epidermal cells expressing GFP:Moesin in a stage 7 embryo. The time after wounding is indicated. Anterior left, dorsal up.

**Movie S6. Livewire-based segmentation of the wound outline.**

Epidermal cells expressing E-cadherin:GFP around a wound. The user moves the mouse around the wound and the algorithm finds, automatically, the brightest set of pixels following the trajectory of the mouse.
