Supplemental material

Weng et al., http://www.jcb.org/cgi/content/full/jcb.201508056/DC1



Figure S1. Adherens junctions show intermediate position during the change from subapical to apical junction configurations. (A) Fixed embryos were hand-sectioned and stained with anti-Arm antibody to reveal the endogenous adherens junctions, showing intermediate states of junction morphology and position. Bar, 10 µm. (B) Projection of E-Cad::GFP images at different depth from confocal stacks of living wild-type embryos at different time points during apical constriction. A stack of images was taken from the apical surface of the ventral cells to 5 µm deep. Shown images are projection of two consecutive optical slices. Bar, 5 µm.



Figure S2. Peak density of E-Cad at adherens junctions increases during junction remodeling. (A) Peak density of E-Cad at edge junctions. (B) Peak density of E-Cad at vertex junctions. Peak density is the sum of the pixels with 90th to 100th percentile intensity. The values at different time points of each edge or vertex are normalized by the maximum value of that edge or vertex during the time period presented.



Figure S3. Temporal correlation between junctional features and myosin activity in ventral cells. (A) Mean correlation coefficients between junction features and randomly matched junctional myosin intensity from other junctions in ventral cells. n = 30. (B) Mean correlation coefficients between junction features and myosin intensity in dorsal cells. n = 20.



Figure S4. **Myosin knockdown suppresses junction shift phenotype induced by Fog overexpression.** (A) Midsagittal section of dorsal region of an embryo with Fog overexpression and GFP RNAi driven by maternal driver shows that adherens junctions appear on the very apical edge of lateral membranes. (B) Expression of *sqh* RNAi prevents apical localization of adherens junctions despite overexpression of Fog. Bar, 10 µm.



Video 1. Adherens junctions (E-Cad::GFP) in a wild-type embryo during apical constriction. A stack of images was taken from the apical surface of the ventral cells to 6 µm deep at 0.5-µm increments. Shown images are projection of two consecutive optical slices. At the beginning, junctions most reside at 3- to 4-µm slices. As the apical constriction progress, junctions first move into 2-µm position and then move into 1-µm position while disappearing from the more basal positions. Images were taken at 5-s intervals using an SP5 laser-scanning confocal microscope (Leica).

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Video 2. Lateral view of junction movement along apical-basal axis in ventral mesodermal cells. Maximum projection of the resliced stacks from Video 1. Apical-basal dimension is 6 µm. Images were taken at 5-s intervals using an SP5 laser-scanning confocal microscope (Leica).



Video 3. Examples of single junction cluster that directly moves apically or shrinks along the apical-basal axis and of two clusters that fuse. Wild-type embryos expressing ubi-ECad::GFP were imaged from the apical surface of the ventral cells to 6 µm deep at 0.5-µm increments, and the tracked clusters were 3D reconstructed at each time point. Images were acquired every 5 s using an SP5 laser-scanning confocal microscope (Leica).

EDGE program was developed by Michael Gelbart and is publicly available at https://code.google.com/p/ embryo-development-geometry-explorer/. Provided online are two ImageJ plugins ROI_coordinates and 3D_Tile. ROI_coordinates functions to output the x, y coordinates of the manually drawn regions of interest to a txt file. ImageJ plugin 3D_Tile functions to generate 3D projections of the stacks using the coordinates of the regions of interest and tile the projections by their temporal sequences.