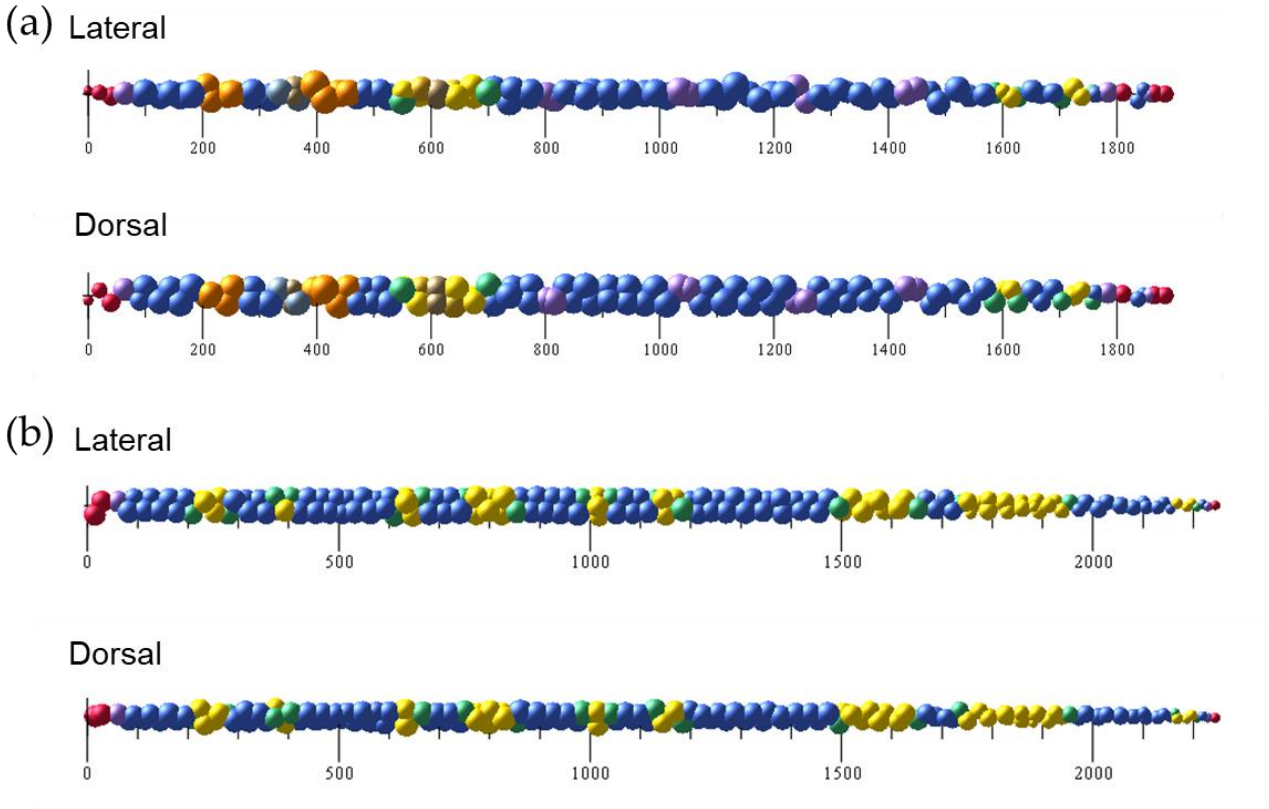
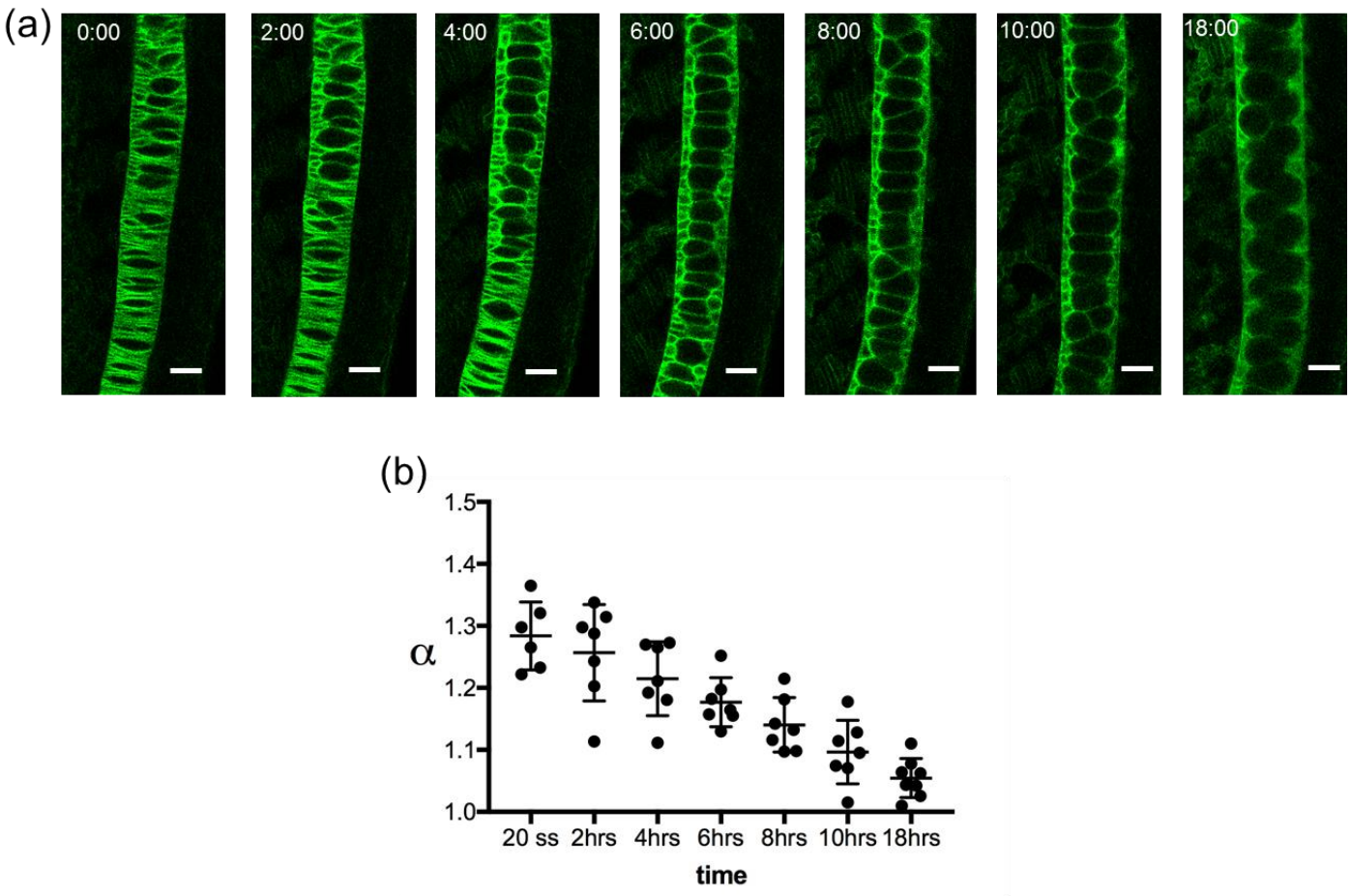
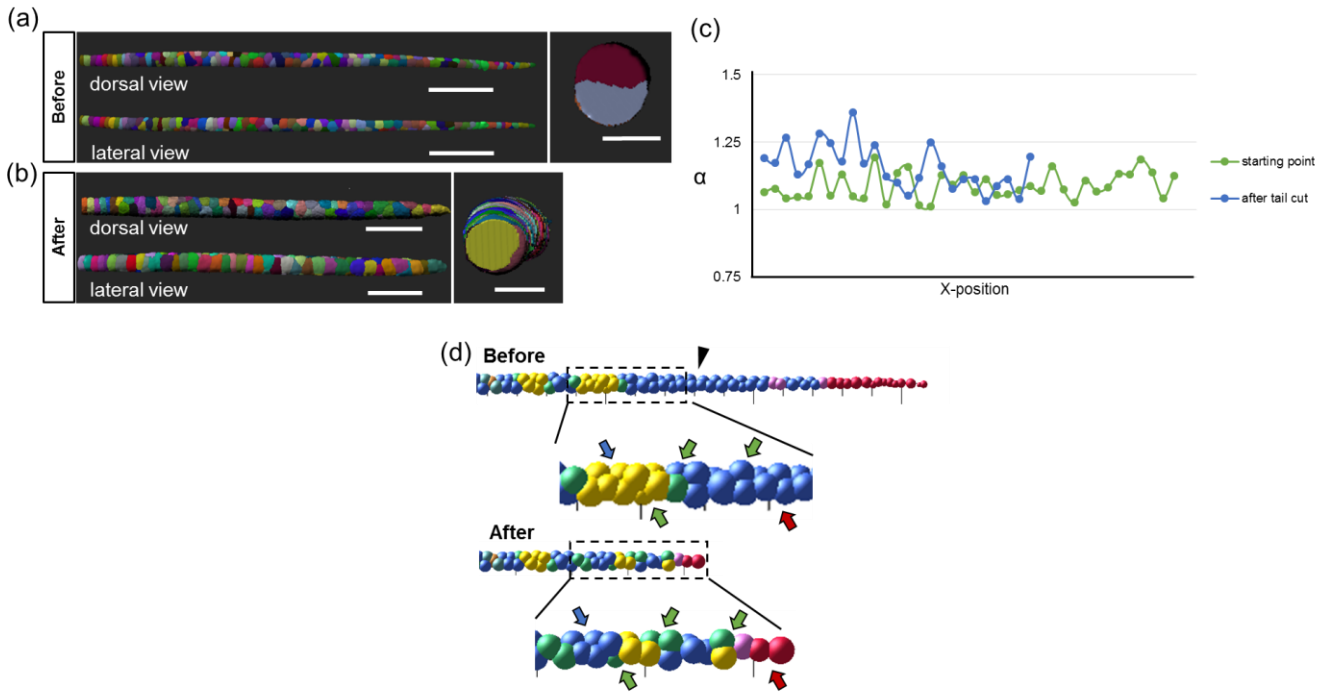


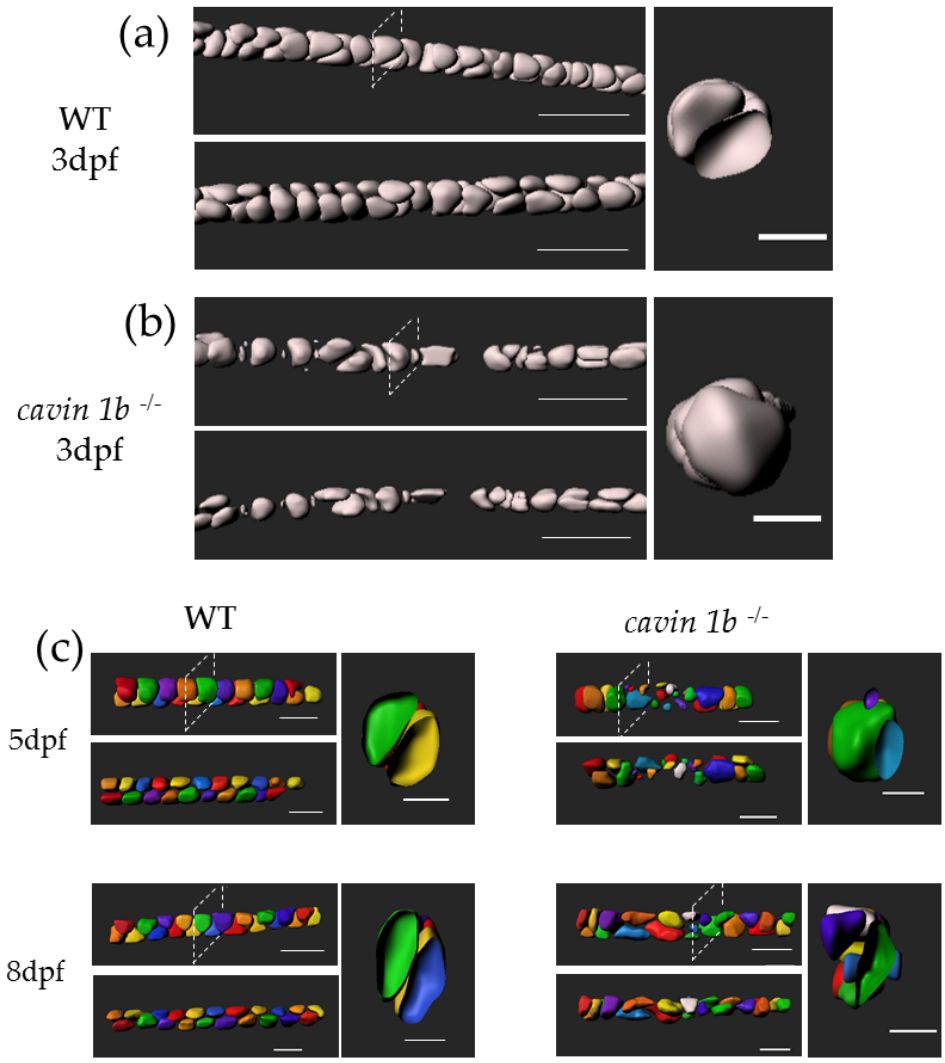
Supplemental Figure 1: Distinct arrangements of vacuolated cells present in the notochord of WT zebrafish. (a-b) Lateral and dorsal views of a sphere plot showing specific arrangements of vacuolated cells present in the notochord of 48hpf WT zebrafish. The patterns observed are indicated as follows: blue (2,1,1), yellow (3,2,1), red (1,1,0) and orange (4,2,2).



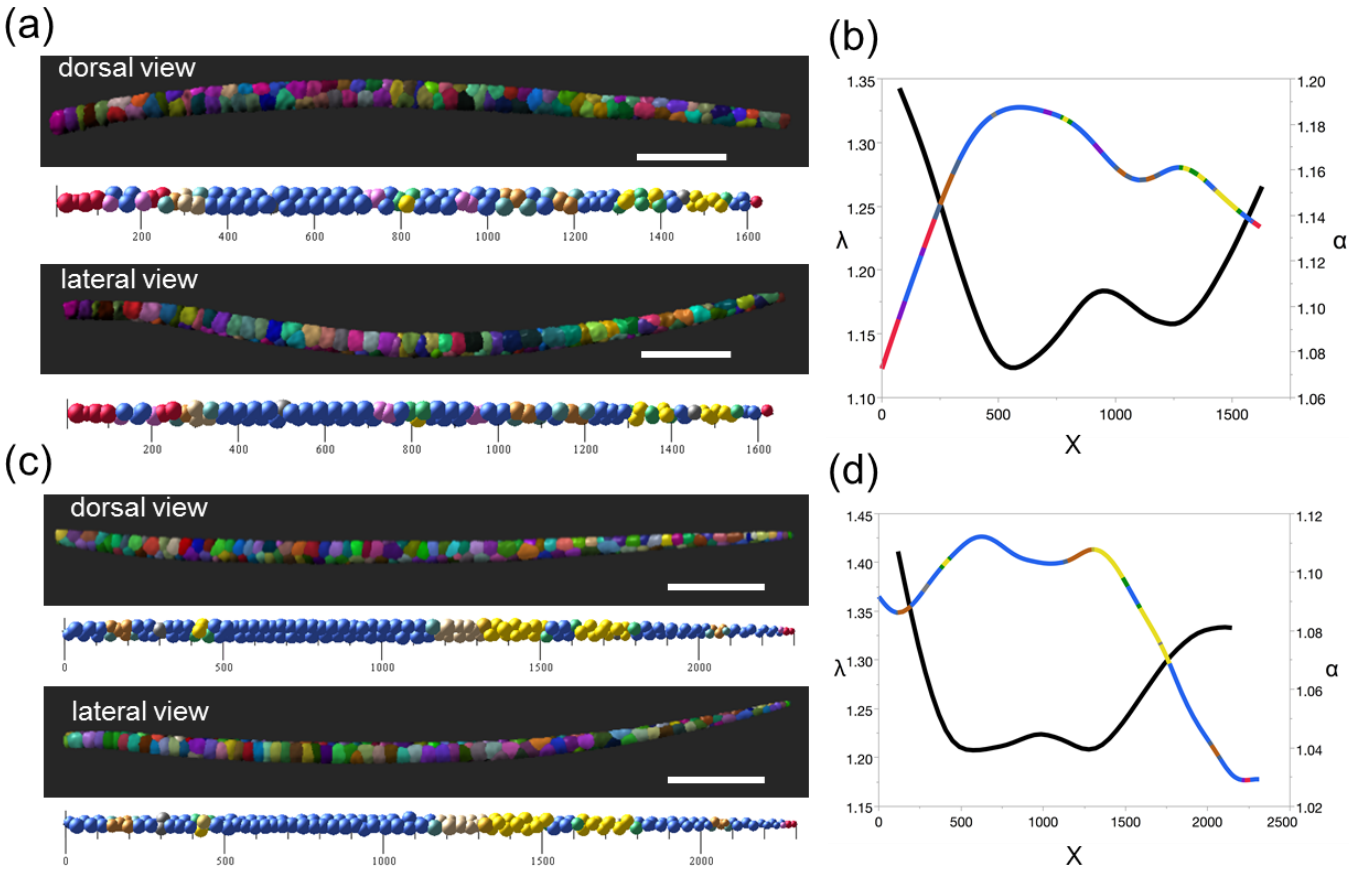
Supplemental Figure 2: The notochord of WT zebrafish presents a highly elliptical aspect ratio during morphogenesis. (a) Still images from a confocal time-lapse movie, starting at the 20-somite stage (time 0), of vacuolated cells in the notochord of a zebrafish as they settle in their final arrangement at room temperature. Scale bar=25μm. **(b)** Local aspect ratio (α) measurements of the notochord at seven time points (20ss -18hrs). The α value decreases significantly as vacuolated cells swell. One-way ANOVA, $p<0.0001$.



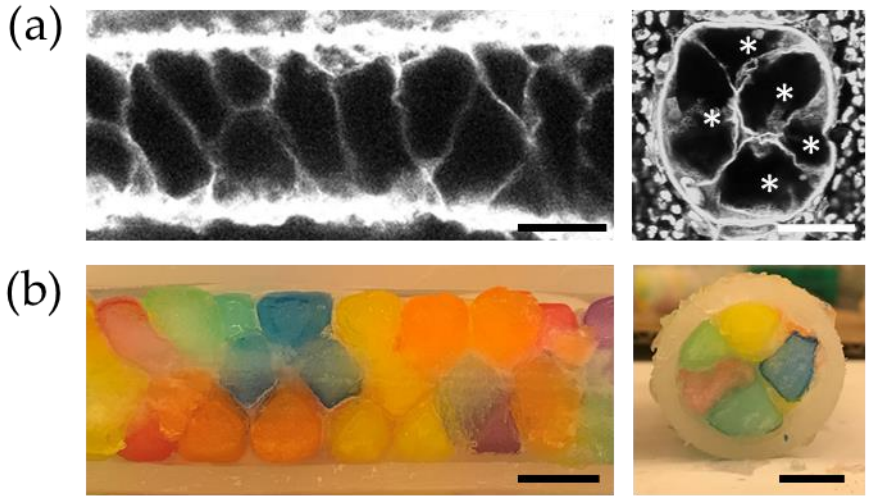
Supplemental figure3. The aspect ratio of the notochord is held by the internal pressure of the structure. **(a)** Lateral, dorsal and cross section views of 3D renderings (top) depicting the morphology and arrangement of vacuolated cells in a 24hpf WT zebrafish embryo before cutting off the tail. Scale bar=300µm. **(b)** Lateral, dorsal and cross section views of 3D renderings (top) depicting the morphology and arrangement of vacuolated cells in a 24hpf WT zebrafish embryo after cutting off the tail. Scale bar=200µm. Scale bars=50µm for cross sections. **(c)** Local aspect ratio (α) measured along the length of the notochord from a 24hpf WT embryo, before and after having the tail was cut. Note the increase in α after the cut. **(d)** Sphere plots corresponding to the 3D renderings depicting vacuolated cells before and after cutting off the tail. The cut site is indicated by the black arrow head. Zoomed in area of sphere plots shows where re-arrangement of patterns occurred. Cells that were arranged in a chiral (3,2,1) pattern before the cut turn into a staircase (2,1,1) pattern after the cut, labeled by the blue arrows. Cells that were arranged in either a staircase or chiral pattern before the cut turn into a transition pattern (green) after the cut, shown by the green arrows. Cells that were staircase before the cut then became bamboo (1,1,0) after the cut are marked by magenta arrows.



Supplemental Figure 4. Dynamic re-arrangement of vacuolated cells following collapse and regeneration in *cavin1b*$^{-/-}$ mutants. (a) Lateral, dorsal, and orthogonal views of 3D rendering of vacuolated cells in the notochord of a 3 days post fertilization (dpf) WT larva. **(b)** Lateral, dorsal, and orthogonal views of 3D rendering of vacuolated cells in the notochord of a 3dpf *cavin1b*$^{-/-}$ larva following motion-dependent collapse of vacuolated cells. Note the re-arrangement of remaining vacuolated cells in in a linear pattern. For (a) and (b) scale bars are 200μm for lateral and dorsal views, and Scale 50μm for orthogonal views (right panels). **(c)** Lateral, dorsal, and orthogonal views of wild type and *cavin1b*$^{-/-}$ notochord 3D renderings at 5 and 8dpf. The same fish was followed over the three-day time period. Following sheath cell invasion and trans-differentiation the new and more numerous vacuolated cells have filled the available space and arranged in a complex pattern (compare with Fig. 6B). Scale bars are 100μm for lateral and dorsal views, and 50μm for orthogonal views.



Supplemental Figure 5: Mechanical buffering of vacuolated cell arrangement in the notochord of WT zebrafish. (a,c) Lateral and dorsal views of 3D renderings (top) and corresponding sphere plots (bottom) depicting the morphology and arrangement of vacuolated cells in a 48hpf WT zebrafish embryo. Scale bars for a=200µm and c=300µm. **(b,d)** Lambda values (coloured lines) and corresponding aspect ratio values (black lines) along the length of a 48hpf zebrafish notochord. Note the mirroring behavior at all points.



Supplemental Figure 6: Arrangement of vacuolated cells in the notochord of *Xenopus laevis*. (a) Lateral and cross section views of the notochord from a fixed *Xenopus* larva (stage 33-34) stained with Alexa568-phalloidin. Asterisks label the vacuole in vacuolated cells. Scale bars are 50μm. **(b)** Large silicone tube (1.91cm inner diameter) filled with 60 JBs (λ=1.85). To visualize the JB arrangement the tube was frozen and cut for imaging. Scale bars are 1cm.