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Supplemental information

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SCALE SPACE DETECTOR FOR ANALYZING SPATIOTEMPORAL VENTRICULAR CONTRACTILITY AND NUCLEAR MORPHOGENESIS IN ZEBRAFISH

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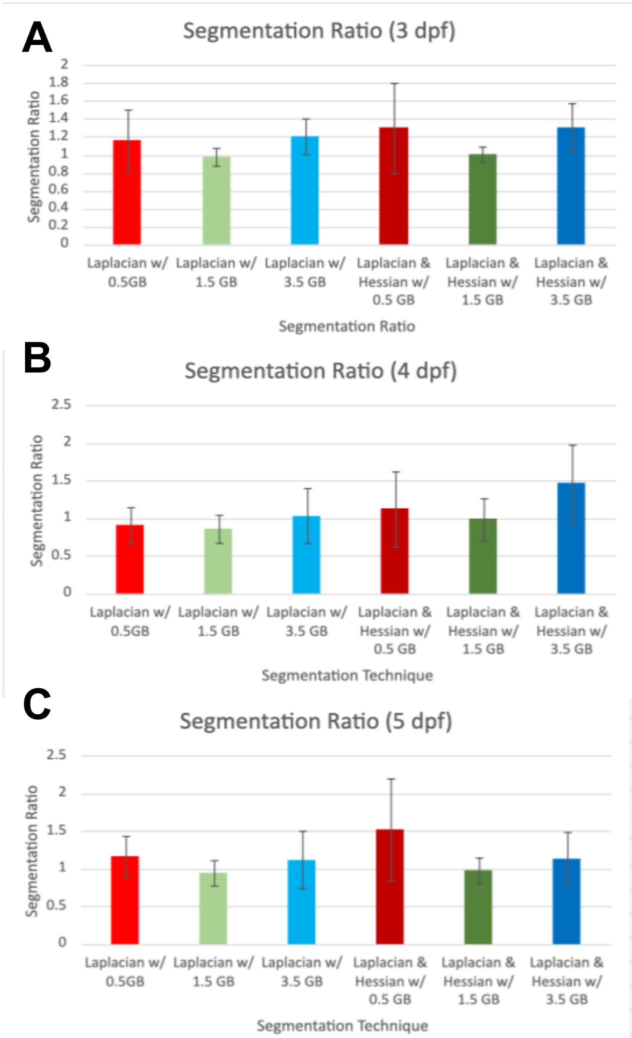

Figure S1: Quantifying segmentation accuracy, related to Figure 3. We tested the precision of the proposed framework to assess false positives or negatives that may be introduced as markers. We reconstructed scale spaces for the Difference of Gaussian Filter and the hessian + DoG respectively , by blurring volumes at different degrees of blur (0.5 – 3.5 gaussian kernel). By identifying the characteristic scale space representation (1.5 gaussian blur), we observed a perfect segmentation result with respect to visually inspected nuclei count across different developmental stages. (GB = gaussian blurring degree)

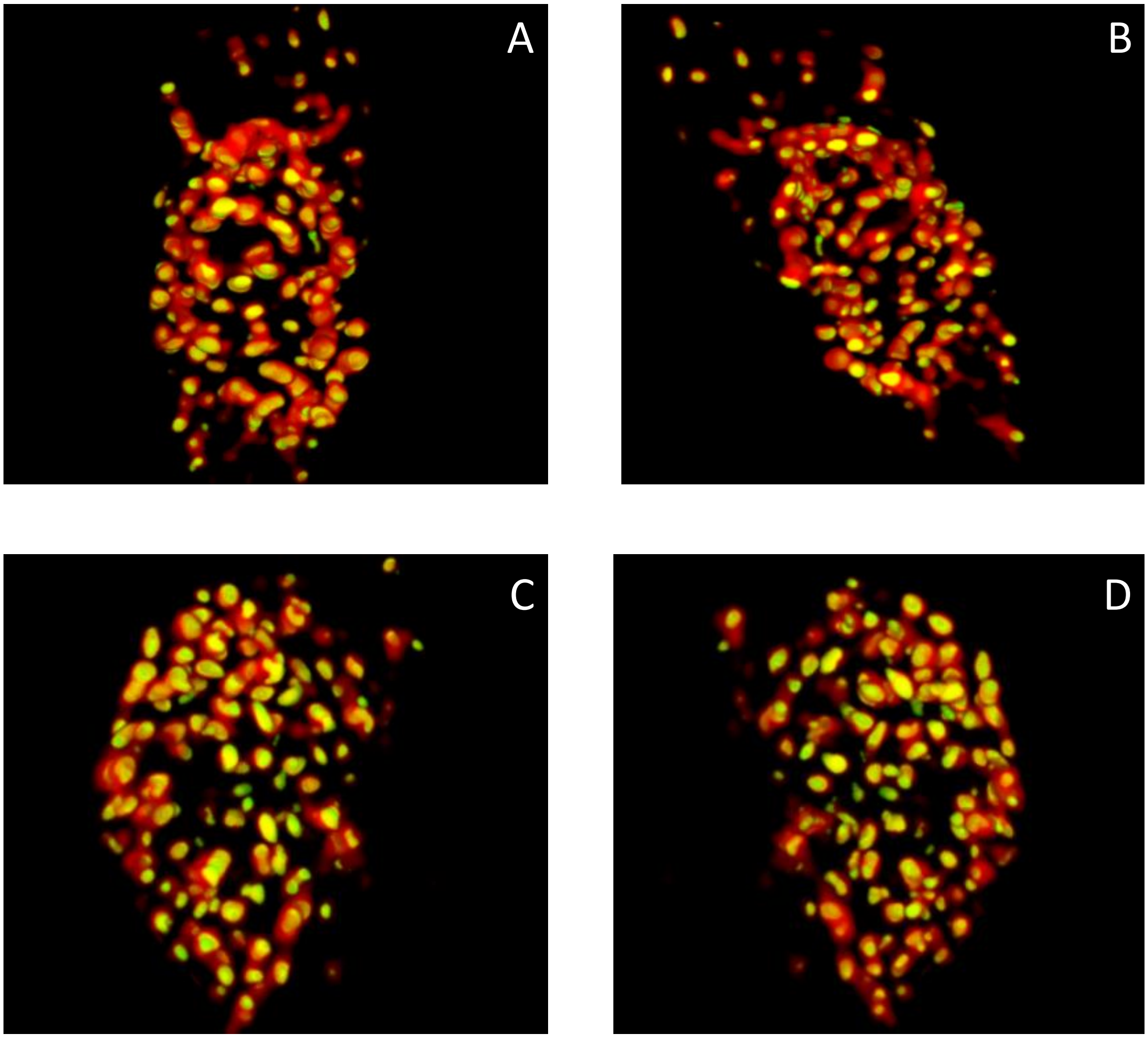

Figure S2. Raw nuclei volumes (red) and Hessian DoG segmented nuclei (green) overlay, related to Figure 3. Yellow represents the common pixel coverage between the two volumes. (A) and (B) represent the 3 days post fertilization ventricular outer curvature and inner curvature respectively (C) and (D) represent the 4 days post fertilization ventricular outer curvature and inner curvature respectively

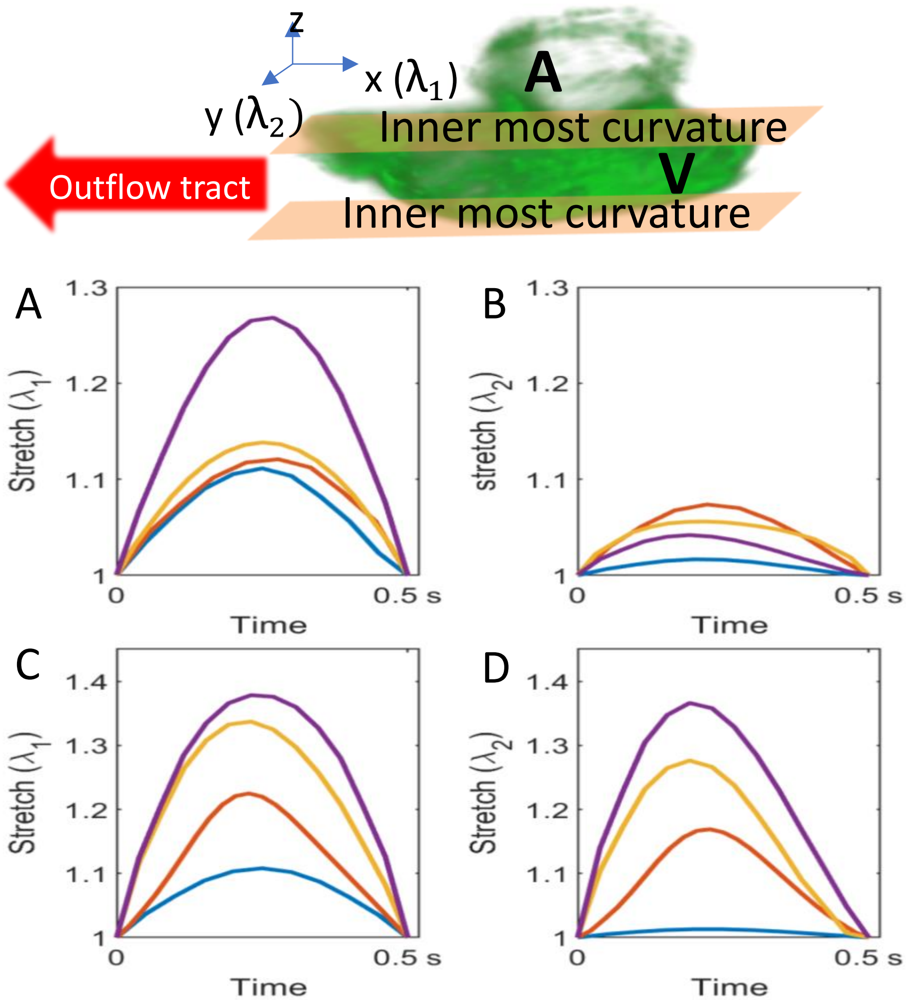

Figure S3: The graphs represent the time course of strain in principal directions one and two for the ventricle's outermost and innermost curvature, related to Figure 4. Epsilon 1 and Epsilon 2 are relative to the changing orientation of the triangular plane utilized in analysis. Relative to each configuration λ_1 lies in the longitudinal direction and λ_2 lies along the circumferential direction. The start of ventricular filling is the reference point for analysis. At this stage, the ventricle has the lowest volume, and the change in stretch through the cardiac cycle is more evident. The graphs correspond to the following values: (A) Innermost curvature stretch in longitudinal direction, (B) innermost curvature stretch in circumferential direction, (C) outermost curvature in longitudinal direction, and (D) Outermost curvature in circumferential direction.

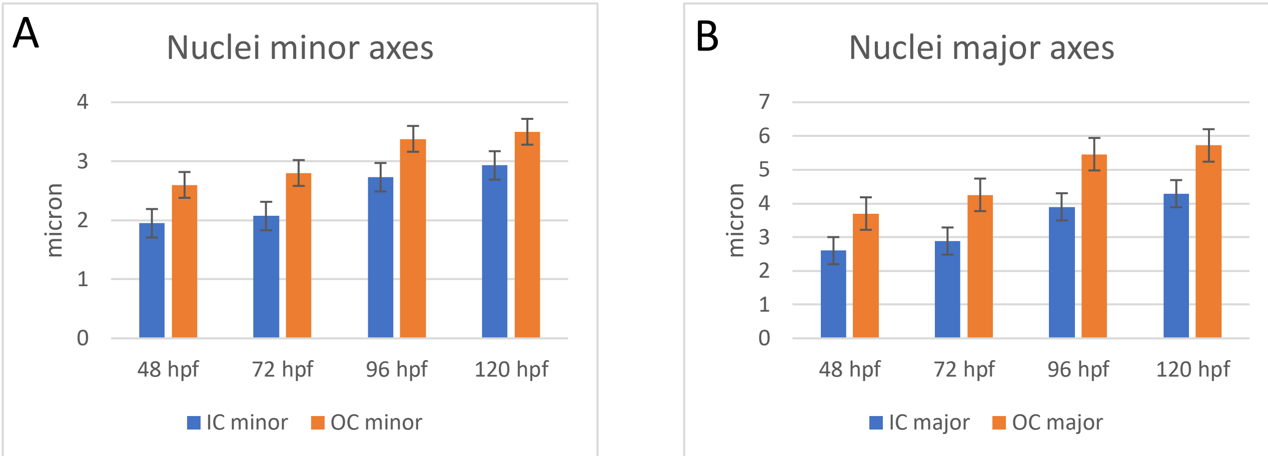

Figure S4: Systolic and diastolic ellipsoid major and minor axis comparisons, **related to Figure 5** (A) Nuclei ellipsoid minor axis reported in micron (B) Nuclei ellipsoid major axis reported in micron. No statistical testing was performed

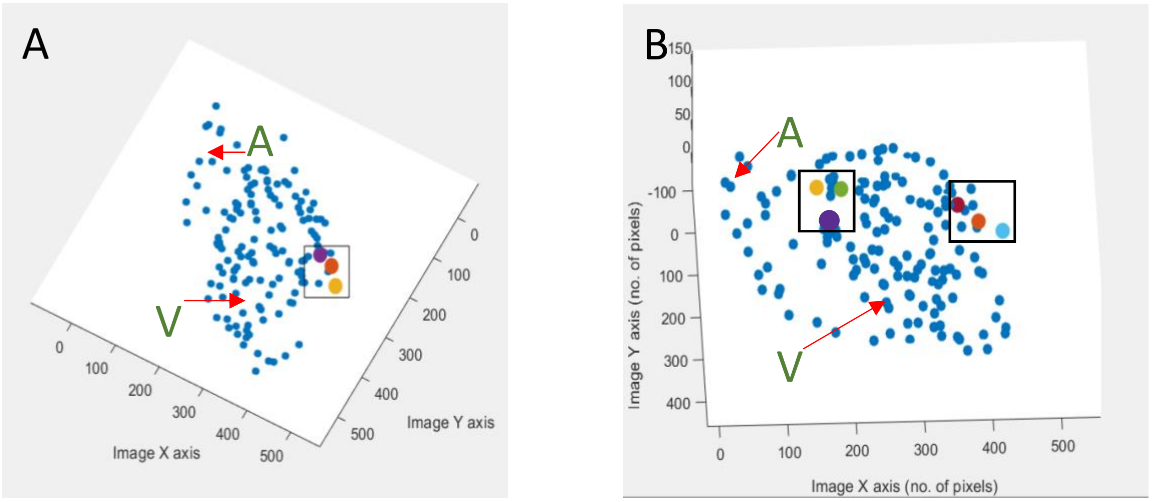

Figure S5. Snapshot of 4D nuclei tracking performed using MATLAB, related to STAR methods (A) 3 dpf ventricle nuclei volumetric stack reconstructed using Matlab. Yellow, orange and purple blobs highlight centers of mass localized for nuclei tracking in the outer curvature (B) 4 dpf ventricle volumetric stack reconstructed using MATLAB. Yellow, green and purple centers of mass localized for nuclei tracking in the inner curvature. Red, orange and blue centers of mass localized for nuclei tracking in the outer curvature. A= atrium, V = ventricle

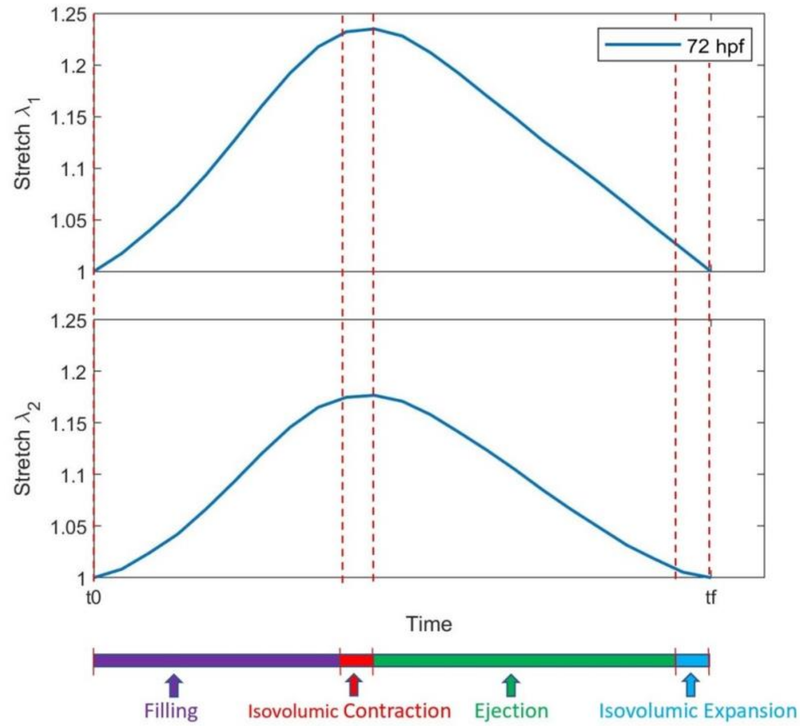

Figure S6: Time course of the principal stretch values for 3dpf zebrafish taken from the outermost curvature with a reference state being the lowest ventricular volume, **related to Figure 4.** This figure displays key steps in the cardiac cycle superimposed on the time course of principal stretch values lambda 1 and lambda 2. Critical points of interest include ventricular: filling, isovolumic contraction, ejection, and isovolumic expansion.

	Nucleus systolic volume	Nucleus diastolic volume
	Inner curvature vs Outer curvature	Inner curvature vs Outer curvature
48 hpf	P = 0.2	P = 0.0003
72 hpf	P = 0.4	P = 0.01*
96 hpf	P = 0.002*	P = 0.03*
120 hpf	P = 0.008*	P = 0.04*

	Nucleus systolic surface area	Nucleus diastolic surface area
	Inner curvature vs Outer curvature	Inner curvature vs Outer curvature
48 hpf	P = 0.3	P = 0.01
72 hpf	P = 0.3	P = 0.03*
96 hpf	P = 0.03*	P = 0.001*
120 hpf	P = 0.02*	P = 0.01*

Table S1: Statistical significance of ventricular cardiomyocyte nucleus at systole and diastole for the inner and outermost curvature region, related to Figure 4. Asterisk denotes significant change from previous value