Supplemental Materials

Molecular Biology of the Cell

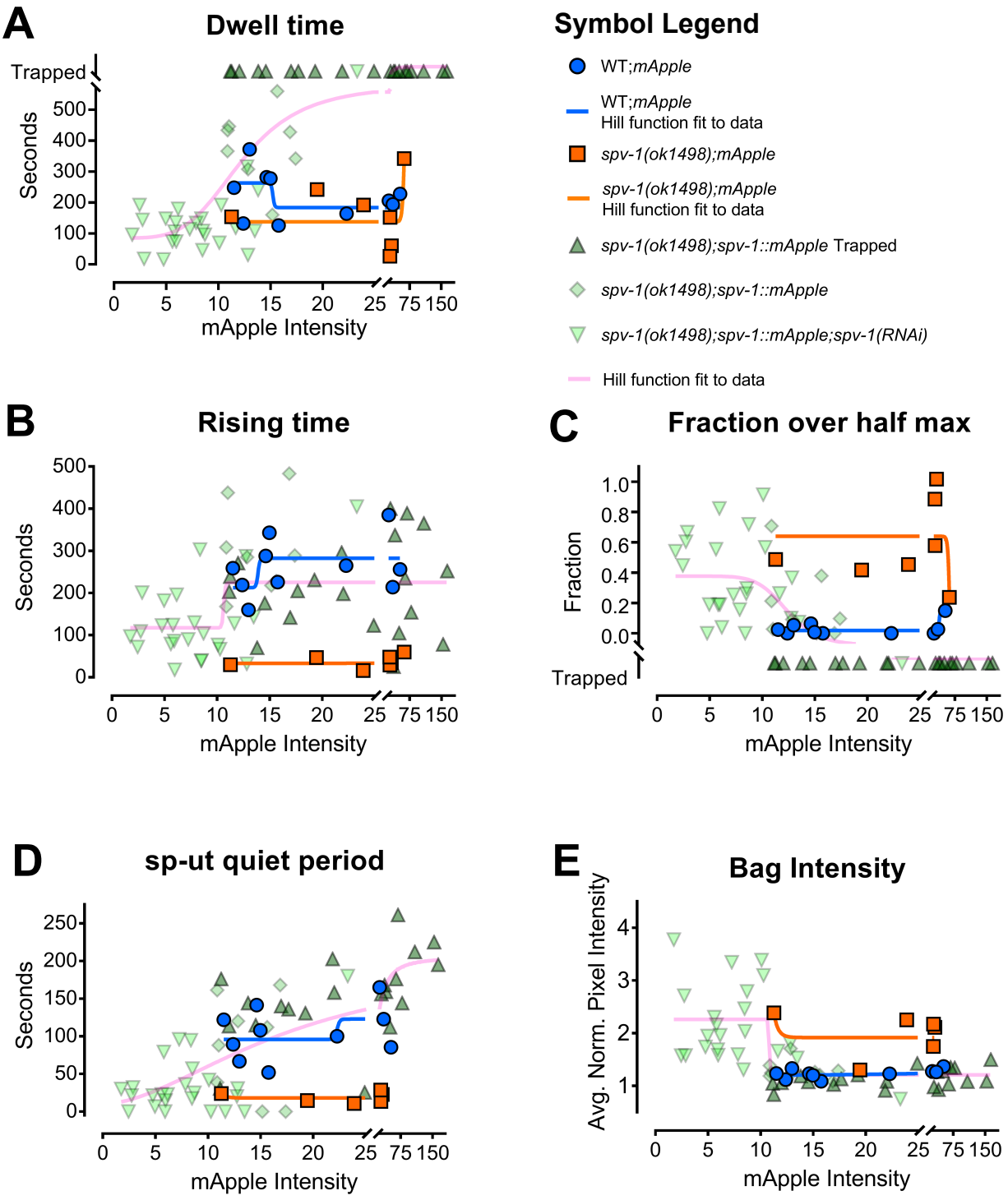
Bouffard et al.

Supplemental Figure 1. mApple alone does not induce embryo trapping or low calcium signaling. (A) Dwell times plotted as a function of mApple intensity. (B) Rising times plotted as a function of mApple intensity. (C) Fractions over half max plotted as a function of mApple intensity. (D) sp-ut quiet periods plotted as a function of mApple intensity. (E) Bag intensities plotted as a function of mApple intensity. In all panels, *spv-1(ok1498);spv-1*::mApple data are shown as slightly transparent to emphasize the mApple controls. Hill function fits to the mApple control data are plotted solely as a guide for the eye.

Supplemental Figure 2. Tissue function and calcium signaling metrics all show thresholds covering SPV-1::mApple intensity values from ~10 to ~15. (A) Dwell times plotted as a function of mApple intensity. Annotations display the R2 value of the Hill function fit, and the Threshold value from that Hill function. The 95 percent confidence interval for the Threshold value is displayed below the data points, with numbers indicating the upper and lower values. (B) Rising times plotted as in panel A. (C) Fractions over half max plotted as in panel A. (D) sp-ut quiet periods plotted as in panel A. (E) Bag intensities plotted as in panel A. The n. d. on the confidence interval indicates that the program could not determine the confidence interval for the Threshold value of the bag intensity.

Supplemental Figure 3. Heat shock treatment does not alter wildtype tissue function or calcium signaling in any of the metrics. (A) Dwell times plotted by condition. (B) Rising times plotted by condition. (C) Fractions over half max plotted by condition. (D) sp-ut quiet periods plotted by condition. (E) Bag intensities plotted by condition. In all panels, error bars display standard deviation, p-values were calculated using Welch's t-test: ns, $p \geq 0.05$

Supplemental Figure 4. Heat shock treatment and activation of constitutively active GTPases does not alter actin bundle alignment. (A) Max intensity projections of representative fixed, dissected spermathecae, stained with phalloidin-conjugated Texas Red. All spermathecae are oriented with the sp-ut valve on the right. Scale bars are 10 μm, brightness is enhanced for presentation. (B) Anisotropy values provide a measure of actin bundle alignment. Each data point is a single cell, 3 cells were quantified in each of 4 different spermathecae for each condition. Error bars display standard deviation, p-values were calculated using Welch's t-test: ns, $p \geq 0.05$



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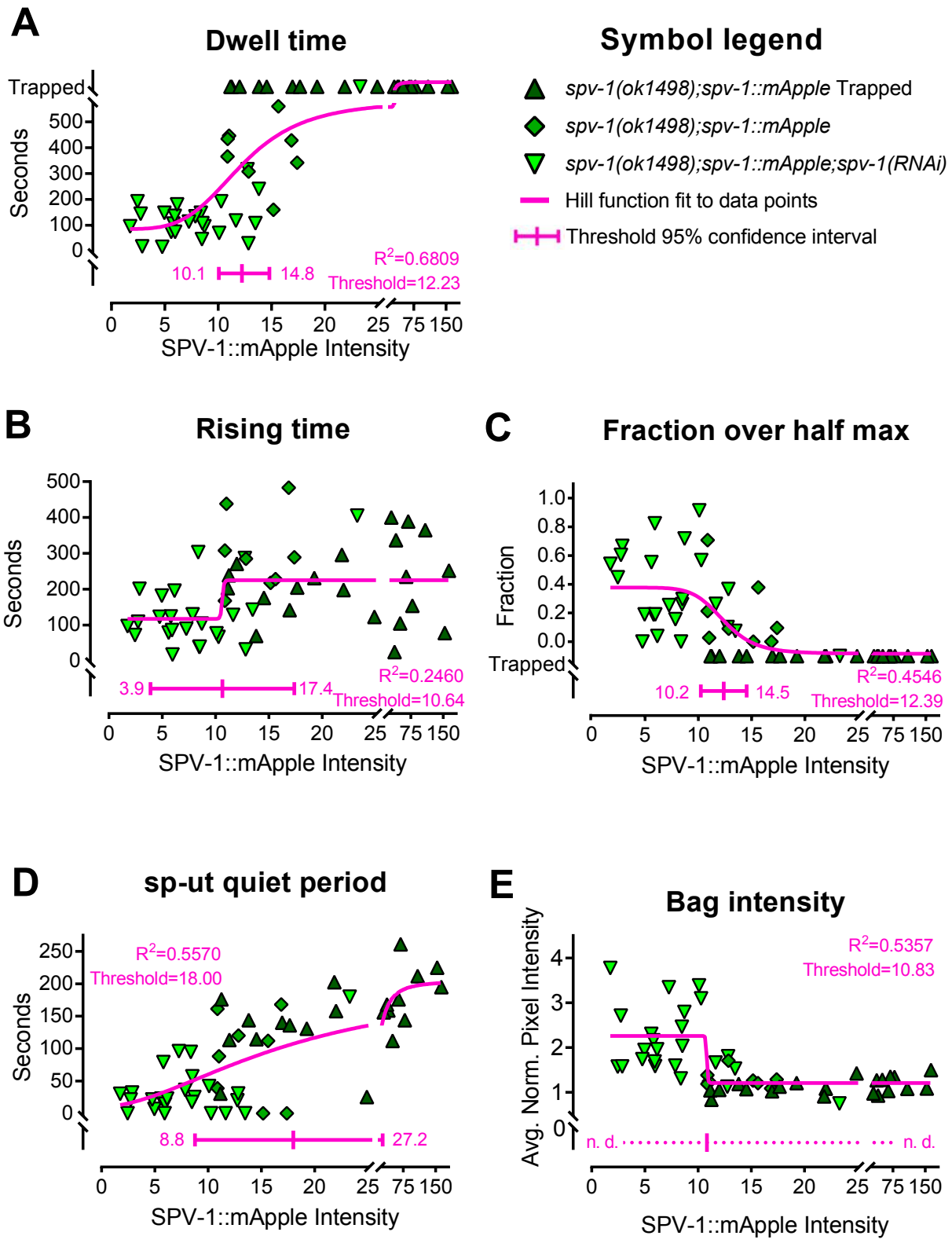

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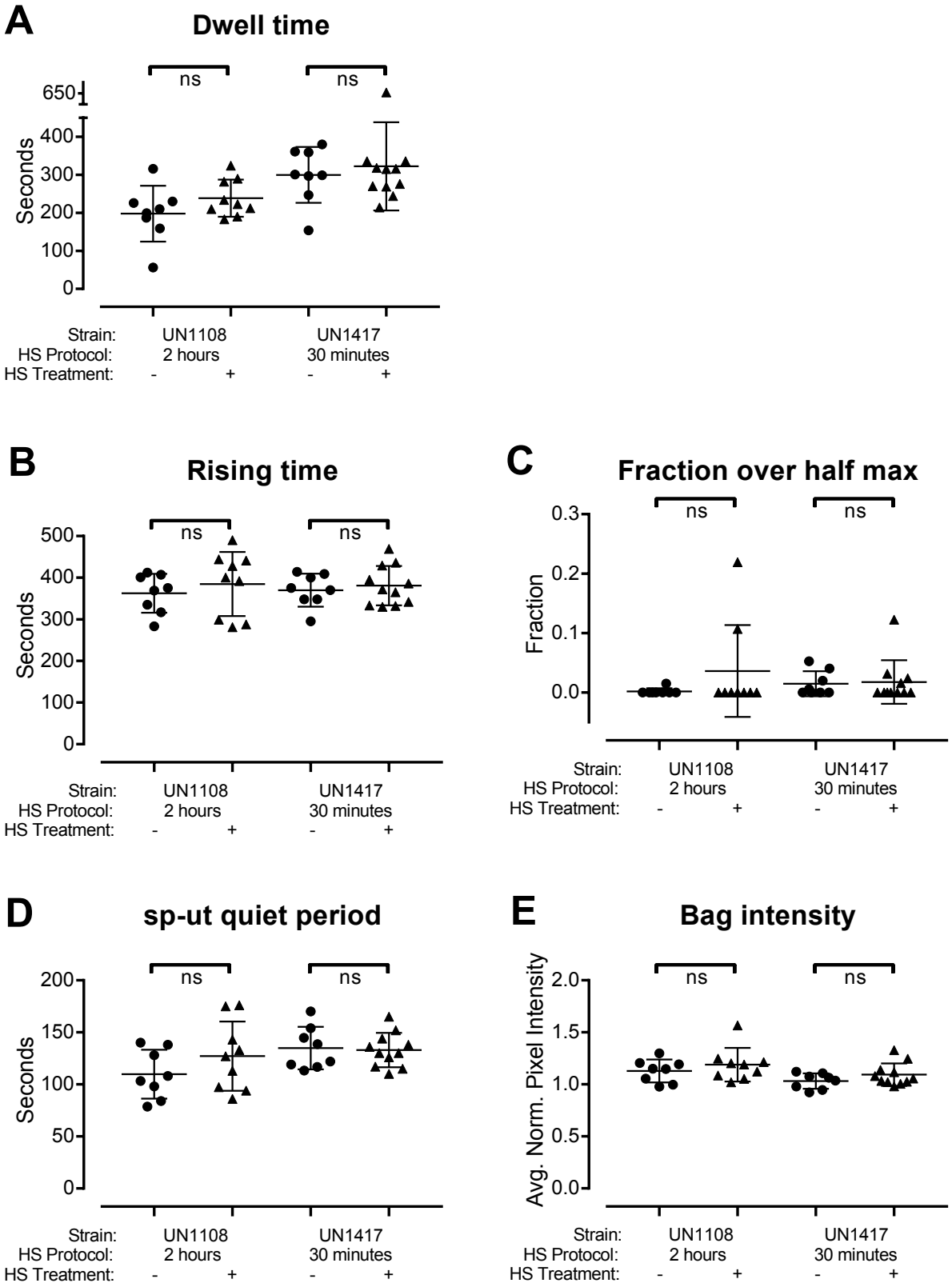

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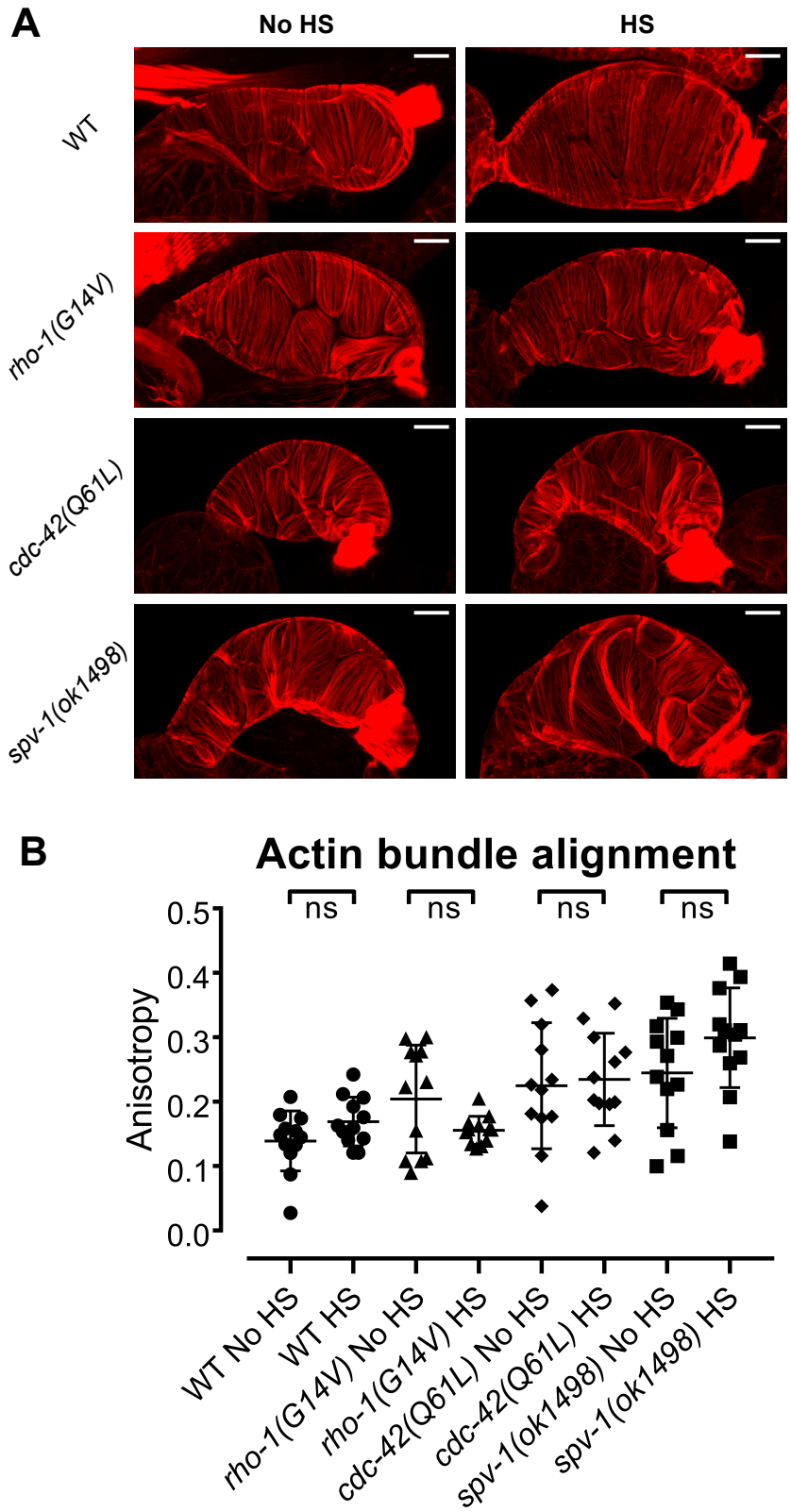

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