## Localized Microinjection of Intact *Drosophila melanogaster* Larva to Investigate the Effect of Serotonin on Heart Rate

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## S1. Illustration of the microneedle filling technique



Figure S1: Illustration of the microneedle filling technique. A) The glass capillary with an internal filament was pulled and its blunt end was placed inside the injection liquid. After few minutes, the liquid was drawn up through the filament via capillary action and filled the tip of the microneedle. B) A MicroFil made up of a flexible fused silica capillary coated with polyimide was connected to a syringe containing the injection liquid and used to backfill the rest of the microneedle.

## S2. 3<sup>rd</sup> instar *Drosophila* larva heart rate in the first five minutes after immobilization



Figure S2:  $3^{rd}$  instar Drosophila larva heart rate in the first five minutes after immobilization (N=10). Right after immobilization, the average heart rate was  $201 \pm 8$  (SEM). The average heart rate gradually decreased in the next 4 minutes post-immobilization and stabilized around  $180 \pm 4$  (SEM). In the box plot, the lower limit shows the first quartile, the horizontal line shows the median, and the upper limit shows the third quartile of the data. The whiskers connect each quartile to the minimum and maximum data points. The dashed line connects the average heart rates of each data set.