

## Supplementary Electronic Material

*Or47b*-neurons promote male-mating success in *Drosophila*

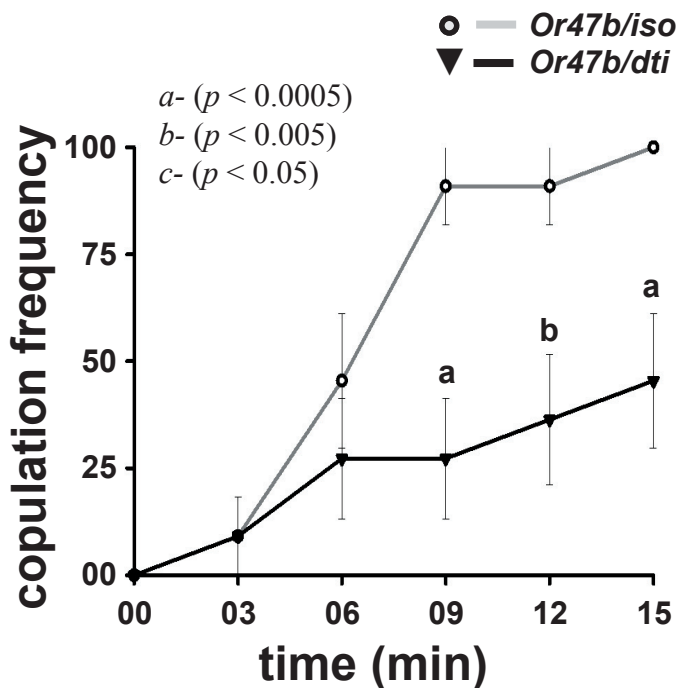
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Running title: *Or47b*-neurons promote male-mating success.

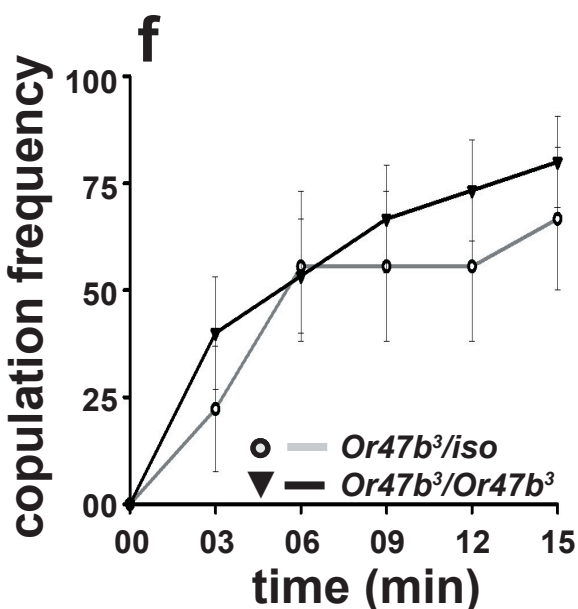
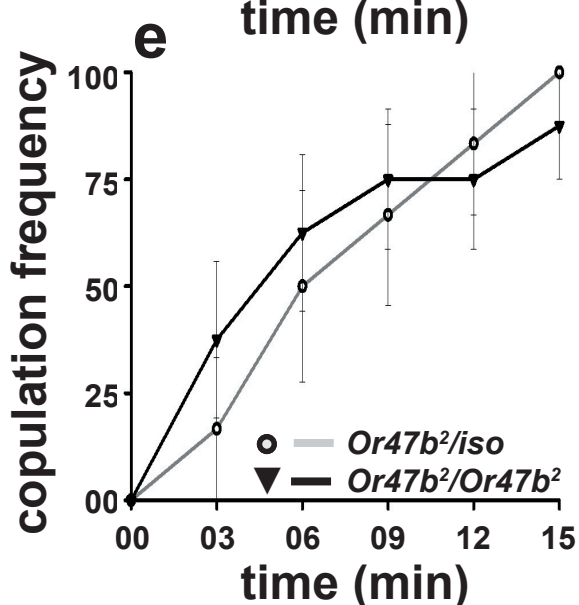
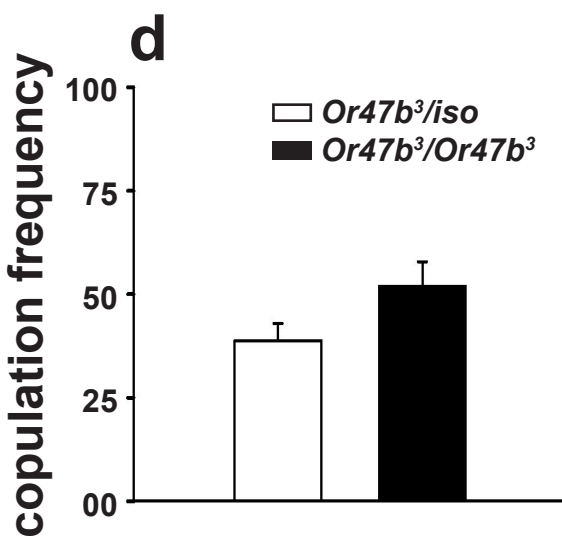
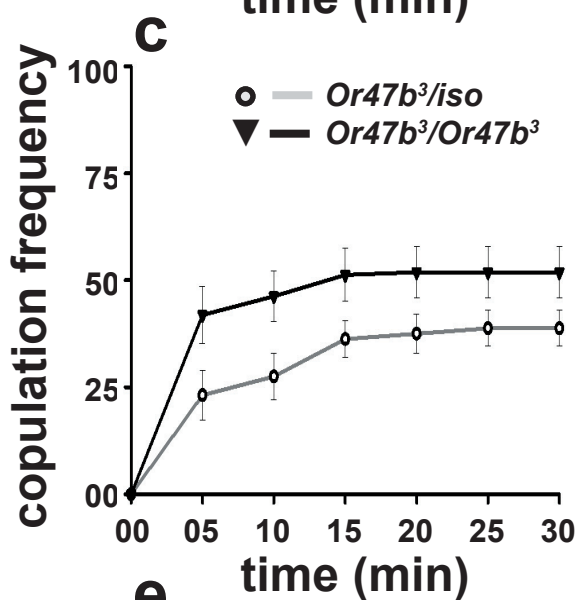
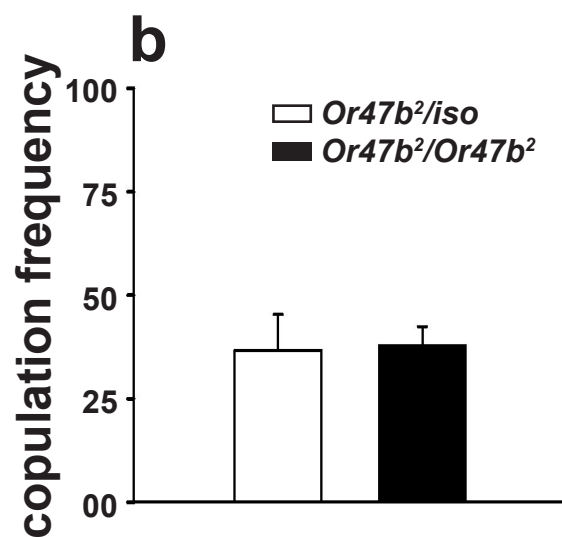
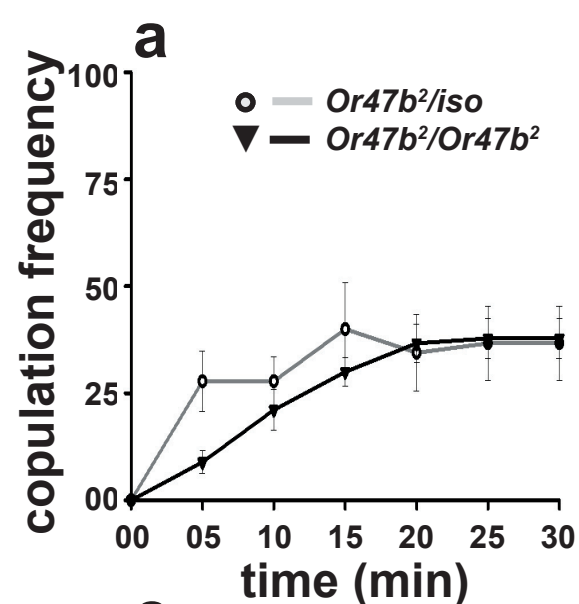
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## Supplementary methods 1:

**Immunocytochemistry protocols:** 20-25 brains of each genotype i.e. *Or47b-GAL4, UAS-GFP* and *Or47b-GAL4, UAS-GFP; UAS-dti* were dissected in 1X phosphate buffer saline (1X PBS). Brains were fixed in 4% paraformaldehyde for 30 min at room temperature (RT) following which they were washed with 0.5% Triton X in PBS (0.5% PBT) thrice for 10 min each. The brains were blocked in 10% horse serum for 1 h at RT after which they were incubated in antibody against GFP ( $\alpha$ -GFP, chicken, 1:1000) for 24 h at 4 °C. Following 6 washes of 10 min each with 0.5% PBT, brains were incubated with secondary antibody ( $\alpha$ -chicken Alexa Fluor 488, 1:3000) for 24 h at 4 °C. Excess was washed away using 0.5% PBT 6 times each for 10 min. 4-5 brains were mounted using 7:3 PBS: glycerol as mounting medium on each slide. Brains were imaged using epifluorescence inverted motorised microscope (Carl Zeiss, Axio observer Z1).



**Figure S1: *Or47b* ablation affects copulation frequency of single male-female pairs:** Copulation frequency profiles of *Or47b*-ablated flies when assayed in single male-female pairs. Flies were monitored for the formation of mating pairs, and number of such pairs formed in 3-min bins over a period of 15-min was used to estimate copulation frequency. All other details same as in Figure 1.



**Figure S2: *Or47b* loss-of-function mutation does not affect copulation frequency:** Profiles of copulation frequency of flies maintained in groups (a) *Or47b<sup>2/2</sup>* and *Or47b<sup>2/+</sup>*, (c) *Or47b<sup>3/3</sup>* and *Or47b<sup>3/+</sup>*, inverted triangle represent mutants, and hollow circles represent heterozygote controls. Bar graphs of copulation frequency of (b) *Or47b<sup>2/2</sup>* and *Or47b<sup>2/+</sup>*, (d) *Or47b<sup>3/3</sup>* and *Or47b<sup>3/+</sup>*, dark bar represents mutants and light bar represents heterozygote controls. ANOVA suggest that there is no detectable effect of mutation in *Or47b* receptor, and copulation frequency of the null mutants (*Or47b<sup>2/2</sup>* and *Or47b<sup>3/3</sup>*) maintained in groups did not differ ( $p > 0.05$ ) from the heterozygote controls (*Or47b<sup>2/+</sup>* and *Or47b<sup>3/+</sup>*). Copulation frequency profiles of flies maintained in single male-female pairs of (e) *Or47b<sup>2/2</sup>* and *Or47b<sup>2/+</sup>*, and (f) *Or47b<sup>3/3</sup>* and *Or47b<sup>3/+</sup>*. ANOVA revealed that the copulation frequency of null mutants did not differ from the controls ( $p > 0.05$ ). In heterozygote, + refers to *iso31*. All other details are similar to Figure 1.