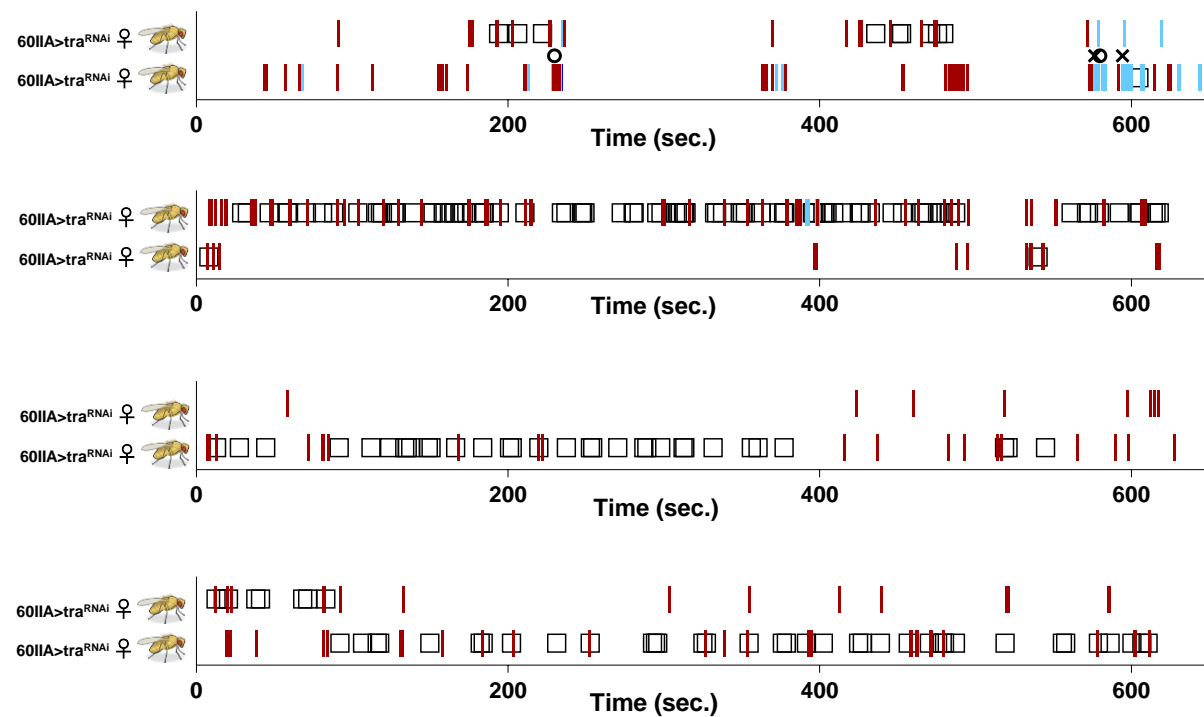


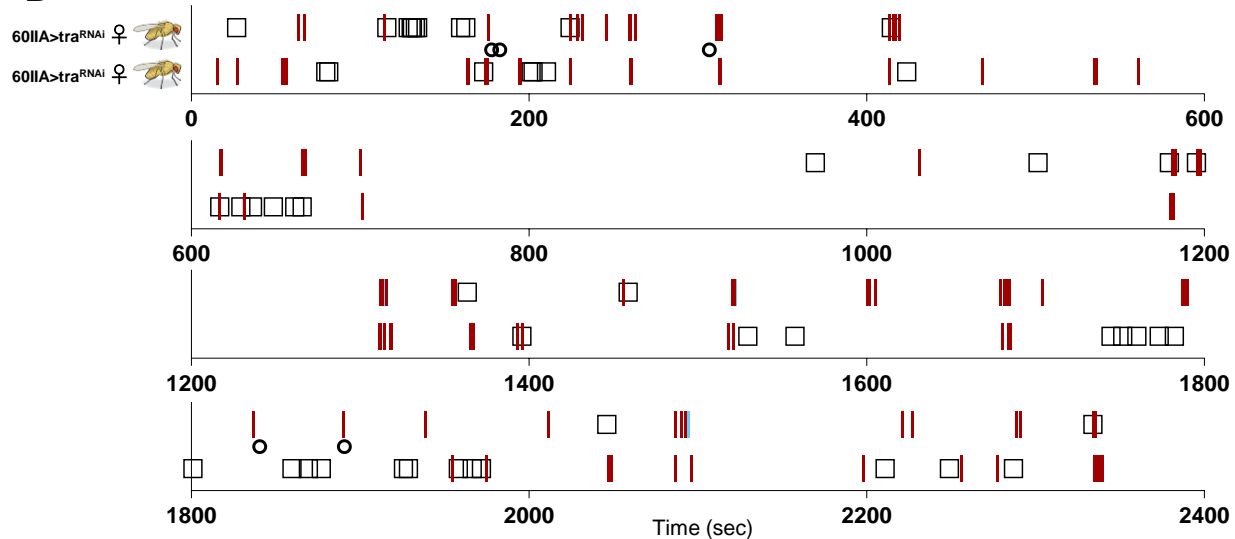


**Movie 1. A pair of 60IIA>tra<sup>RNAi</sup> females exhibiting high intensity aggressive behavior and circling.**

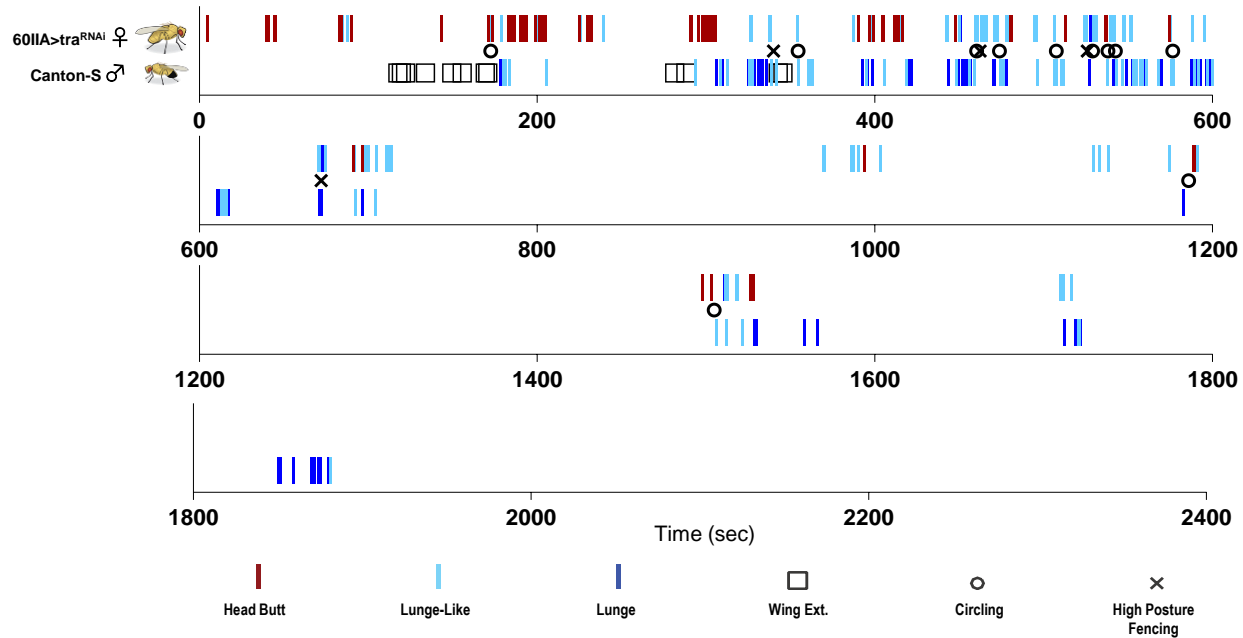
**A**



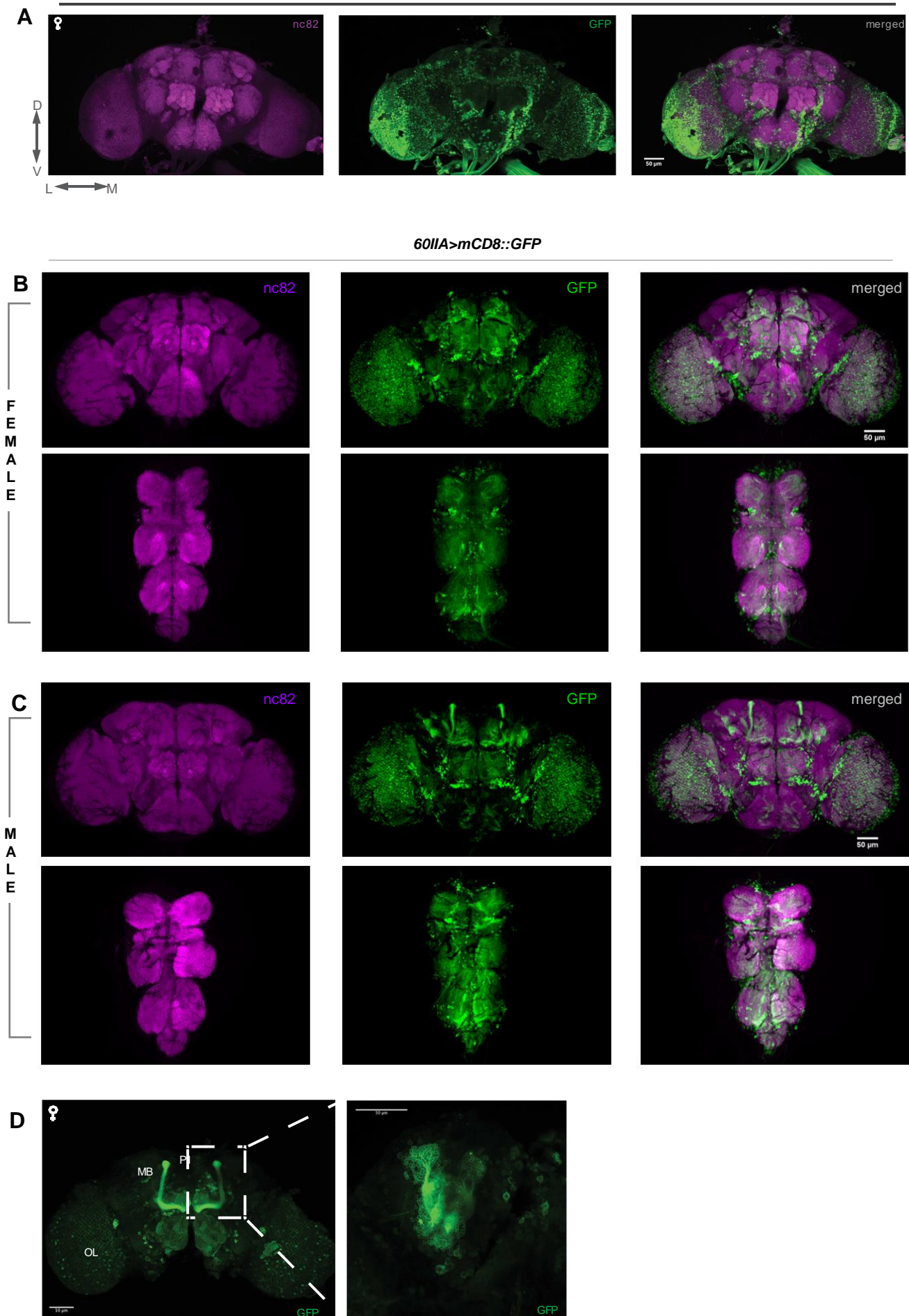
**B**



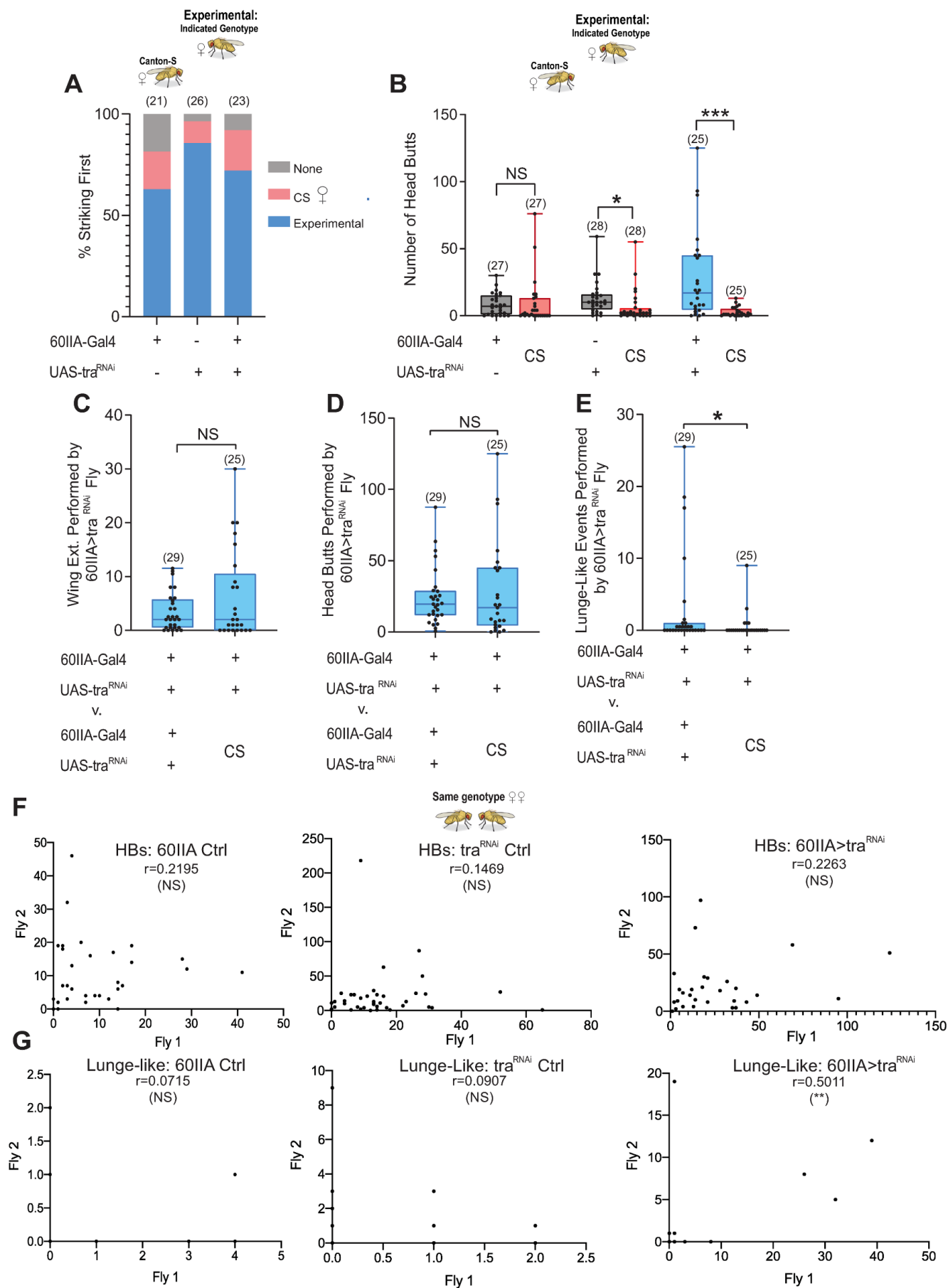
**Fig. S1. Behavioral switching in masculinized females paired with females of the same genotype.** (A-B) Time course of fight dynamics between a pair of *60IIA>tra<sup>RNAi</sup>* females selected for their use of both female aggression and male courtship. Behaviors scored include head butts, lunge-like behavior (Lunge-Like), lunging (Lunge), wing extensions (Wing Ext.), circling (Circling), and high posture fencing. Fights were scored for (A) 10 minutes from the first encounter or (B) 40 minutes from the first encounter. Circling was scored when flies rapidly chased each other in a tight circle. High posture fencing was scored when flies fought on their hind legs.



**Fig. S2. Behavioral switching in a masculinized female paired with a Canton-S males for 40 minutes.** Time course of fight dynamics between a *60IIA>tra<sup>RNAi</sup>* female and a Canton-S male selected because it contained high intensity aggressive behavior. Behaviors scored include head butts, lunge-like behavior (Lunge-Like), lunging (Lunge), wing extensions (Wing Ext.), circling (Circling), and high posture fencing. Fights were scored for 40 minutes from the first encounter. Circling was scored when flies rapidly chased each other in a tight circle. High posture fencing was scored when flies fought on their hind legs.



**Fig. S3. *60IIA-Gal4* expression is widespread in the brain and VNC and appears similar in males and females.** Confocal images of adult brains and VNC immunolabeled for nc-82 (magenta) and GFP (green). Scale bars = 50µm (A) Confocal reconstruction of a *60IIA>nls::GFP* female brain. (B) Maximum projection of a *60IIA>mCD8::GFP* female brain and VNC. (C) Maximum projection of a *60IIA>mCD8::GFP* male brain and VNC. (D) Confocal reconstruction of a *60IIA>mCD8::GFP* female brain. Magnification shows strong expression in the mushroom bodies (right panel).



**Fig. S4. *60IIA>tra<sup>RNAi</sup>* females show less male aggression toward less aggressive Canton-S opponents.** First strikes (A) and individual behaviors (B-E) in various pairings between *60IIA>tra<sup>RNAi</sup>*, Canton-S and *60IIA>tra<sup>RNAi</sup>* females or genetic controls. Behaviors were scored for 10 minutes from the first encounter. (A) Percent breakdown of first strikes in fights between Canton-S males and experimental genotypes *60IIA>tra<sup>RNAi</sup>*, *60IIA-Gal4/+* or *tra<sup>RNAi</sup>/+*. (B) Number of head butts performed by indicated genotype (Kruskal-Wallis test with Dunn's multiple comparisons test). (C) Number of wing-extensions performed by *60IIA>tra<sup>RNAi</sup>* females (Mann-Whitney test). (D) Number of head butts performed by *60IIA>tra<sup>RNAi</sup>* females (Mann-Whitney test). (E) Number of lunge-like behaviors performed by *60IIA>tra<sup>RNAi</sup>* females (Mann-Whitney test). (F) Correlation between number of head butts performed by each fly in same-genotype pairings (Spearman test). (G) Correlation between number of lunge-like behaviors performed by each fly in same-genotype pairings (Spearman test). For all histograms, \*  $p < 0.05$ , \*\*\*  $p < 0.001$ , and NS indicates not significantly different.