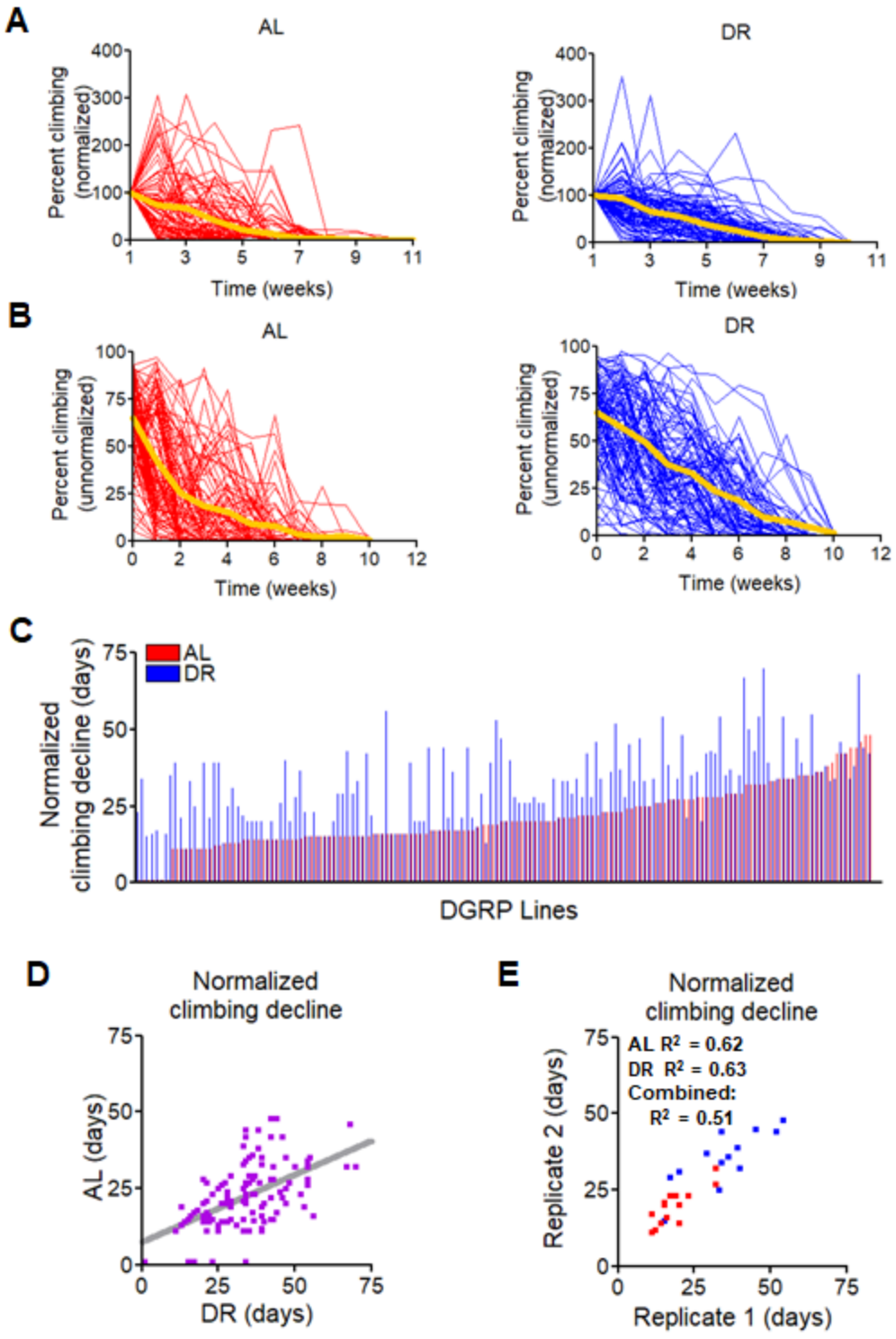

Figure S1. Climbing ability over the course of life varies by strain and diet. Related to Figure 1. (A) Climbing ability of each DGRP line over the course of life normalized to climbing ability after 7 days on diet. AL on left (red), DR on right (blue). Yellow line represents average climbing value across all strains for a given timepoint. (B) Absolute climbing ability of each line over the course of life. AL on left, DR on right. Yellow line as in A. (C) The age (in days) at

which a line declines to half of its initial percent of climbing flies. Data are arranged in ascending order of the strains' AL phenotypes (red). Adjacent lines in blue represent the same strain under DR diet. (D) Comparison of days below 50% maximal climbing capacity of each strain under AL versus DR diet. Grey bar represents best-fit trendline. (E) Comparison of biological replicates of 25 tested DGRP lines under AL (red) or DR (blue) for 50% decline in initial climbing ability.

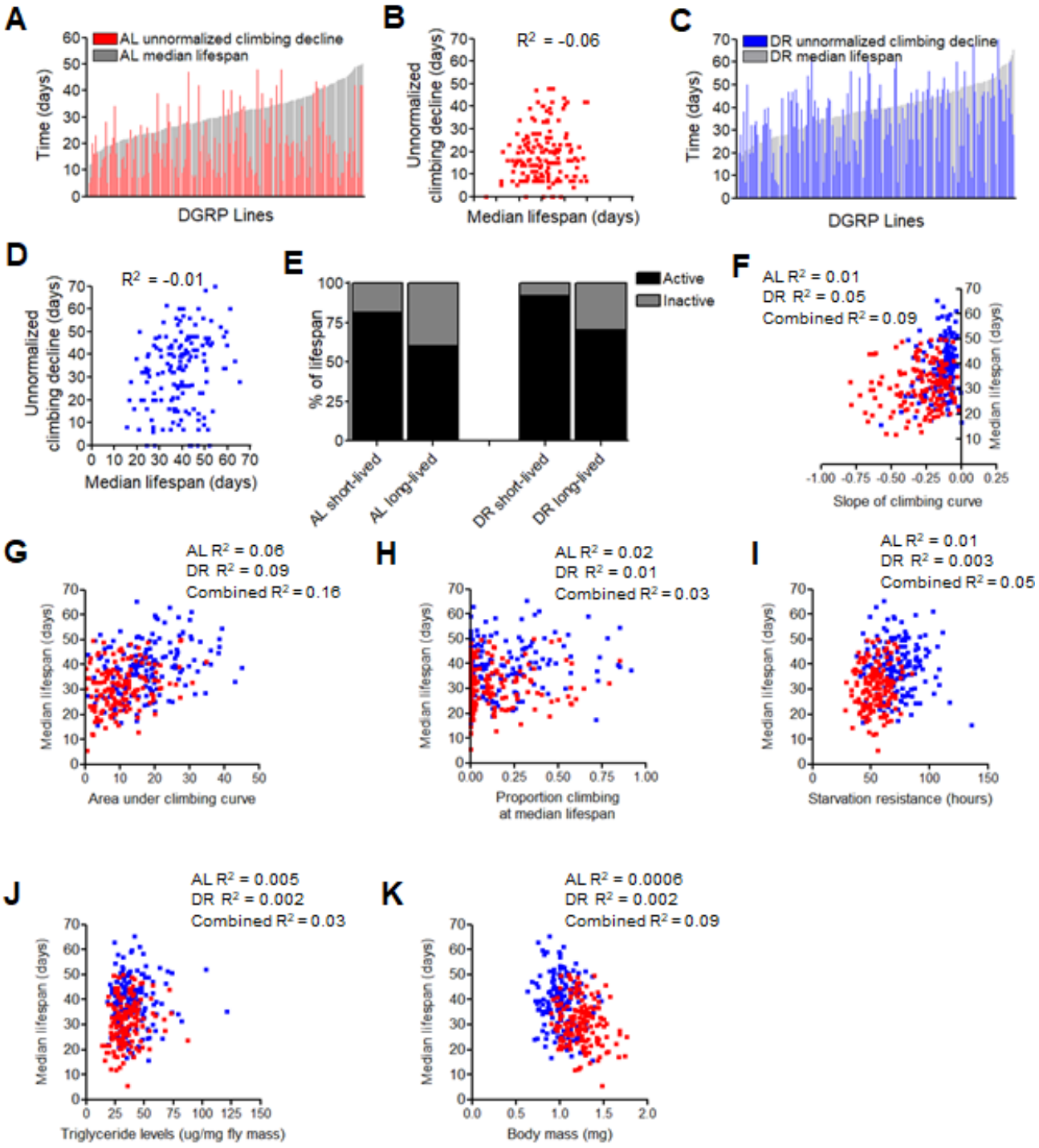

Figure S2. No correlation between median lifespan and the day below 20% climbing. Related to Figure 2. (A-D) Comparison of each tested strain's day less than 20% of surviving flies are able to climb, on (A-B) AL or (C-D) DR. Each bar represents a DGRP strain, ordered by median lifespan on each diet. Colored bars represent climbing half-life and white bars represent median lifespan. (B and D) Scatter plots depicting climbing ability compared to median lifespan on the (C) AL diet or (D) DR. Each dot represents a single DGRP strain. (E) Comparison of the length of time above or below 50% of maximal climbing ability across short-lived half of DGRP strains or long-lived half of DGRP strains under AL conditions (left) or DR (right). (F-H)

Comparison of median lifespan across DGRP strains on AL (red) or DR (blue) with (F) slope of the climbing curve, (G) area under the climbing curve, and (H) proportion climbing at median lifespan. n = 200 flies per strain per diet. (F-H) Scatter plots comparing median lifespan across all DGRP strains and (F) slope of the climbing curve, (G) area under the climbing curve, and (H) proportion of flies climbing at the population's median lifespan. (I-K) Scatter plots comparing median lifespan across all DGRP strains and (I) mean starvation resistance, (J) triglyceride levels normalized to body mass, and (K) body mass. Red is AL, blue is DR.

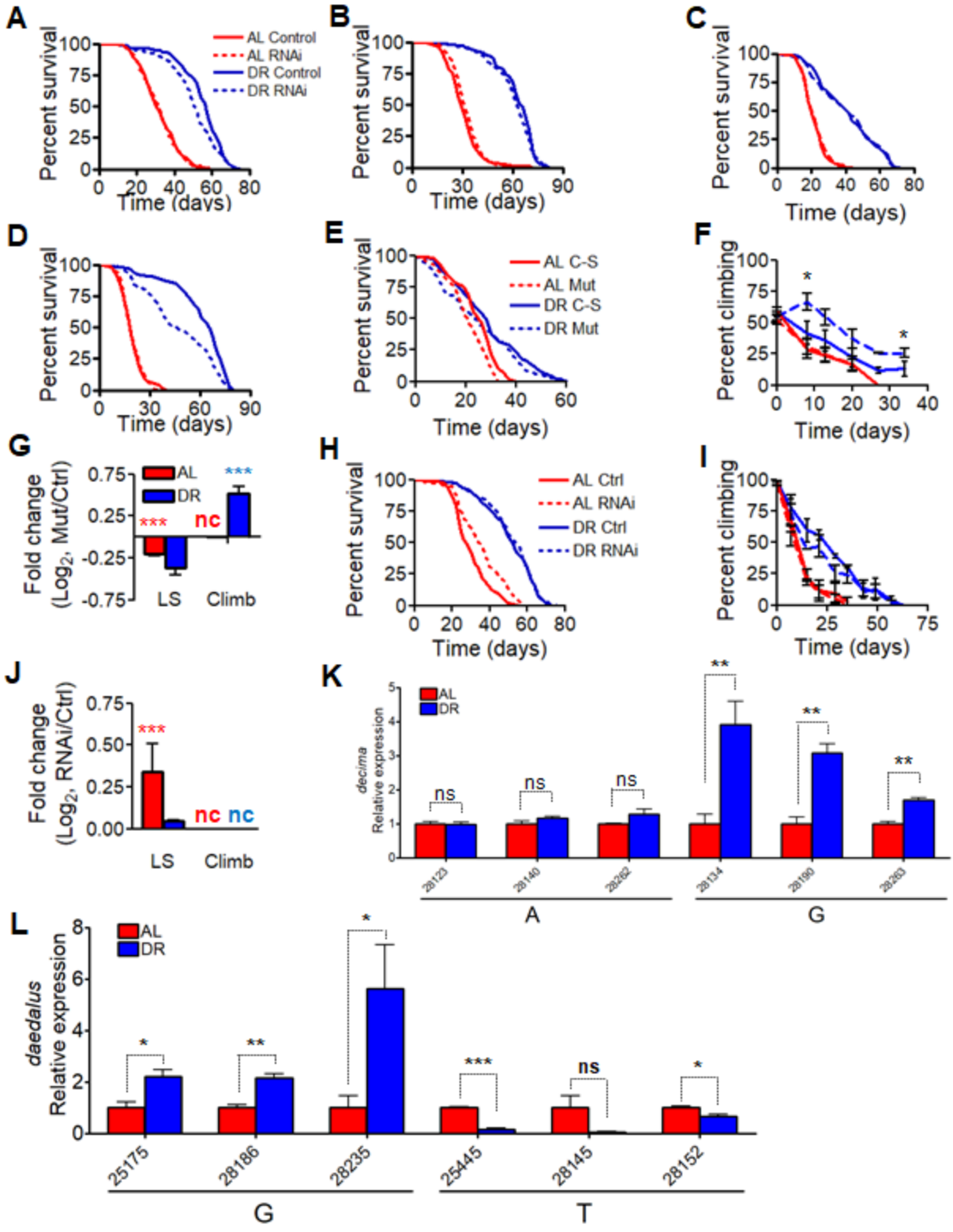

Figure S3. GWAS candidate screening. Related to Figures 3-4. The effects on lifespan of ubiquitous RNAi against (A) *CR32111*, (B) *CG8312*, (C) *CG5888*, and (D) *CG31221*. (E-G) The effect of *Minos* element insertion on (E) lifespan and (F) climbing ability over the course of life in a *Canton-S* background, and (G) the $\log_2$ difference between mutant and controls. (H-J) The

effects of neuronal *dcma* RNAi using the v30163 transgenic line on (H) lifespan, (I) climbing ability, and (J) the changes between RNAi and control. (K) mRNA expression of *decima* across DGRP strains with either nucleotide polymorphism, A on left or G on right. (L) mRNA expression of *daedalus* across DGRP strains with either nucleotide polymorphism, G on left or T on right. Significant differences between mutant or RNAi and controls are indicated by *. * = $p < 0.05$, ** = $p < 0.005$, *** = $p < 0.0005$. p values shown in Data S3. nc = no change, ns = not significant. n = 200 flies per condition for each mutant experiment. n = 50 heads for qRT-PCR data or 5 whole bodies. Data show collective results from three biological replicates. Error bars represent SD between replicates.

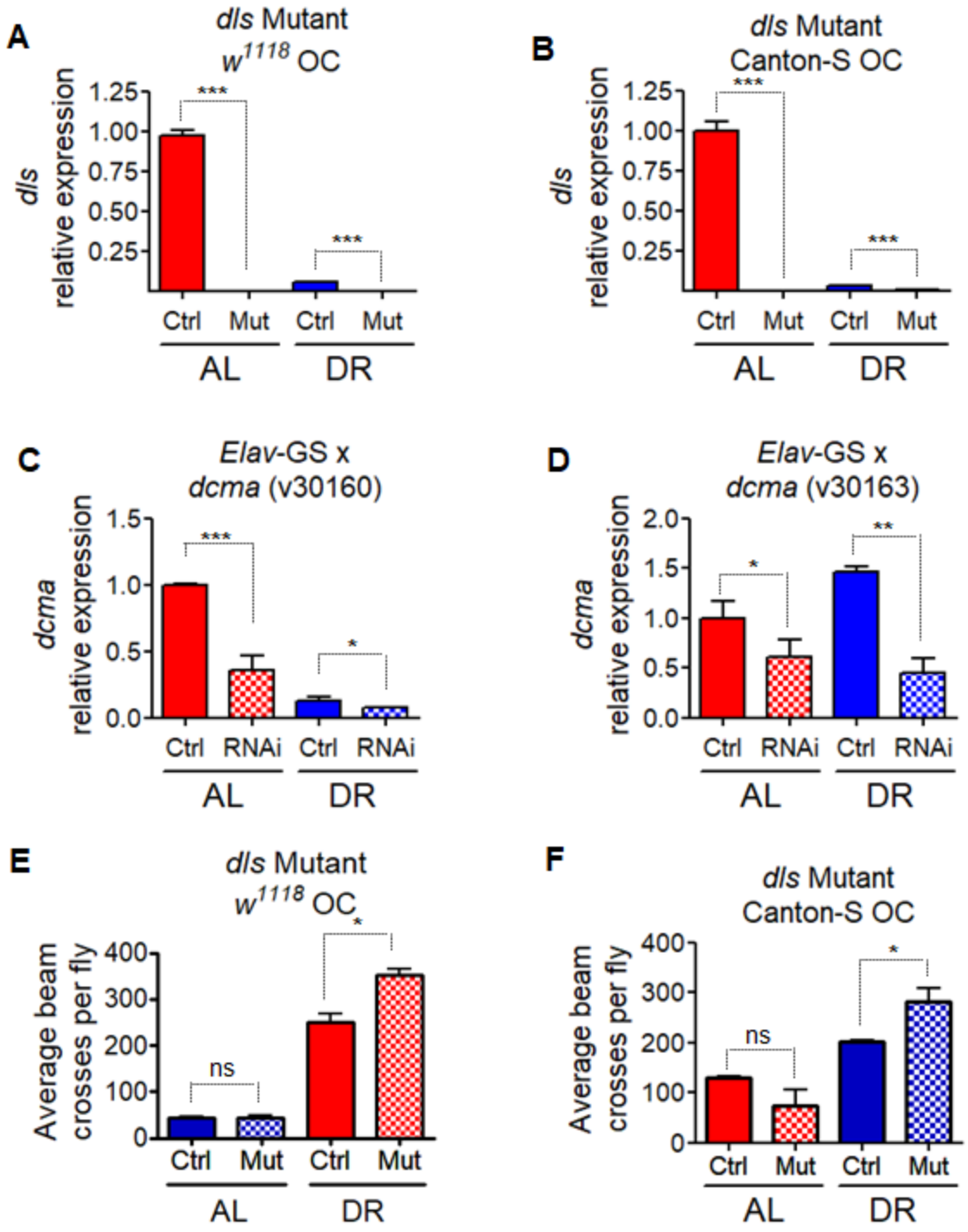

Figure S4. Knockdown validation of *dls* mutant crosses and *Elav*-GS-driven *dcma* crosses and *dls* spontaneous activity. Related to Figures 3-4. All qRT-PCR data normalized to AL control and expression levels of *rp49* housekeeping gene. (A and B) Relative expression of *dls* outcrossed to (A) w^{1118} or (B) *Canton-S* (right) control lines under AL or DR, and expression in a mutant line in w^{1118} or Canton-S background. (C and D) Relative expression of 2 separate neuronal *dcma* RNAi crosses under AL (red) or DR (blue). (E-F) Spontaneous activity recordings over a 24-period for flies with a *Minos* element insertion in *dls* in a (E) w^{1118} or (F)

Canton-S background. Significant differences between mutant and controls are indicated by *. * = $p < 0.05$, ** = $p < 0.005$, *** = $p < 0.0005$. ns = not significant. p values shown in Data S3. AL is shown in red, DR in blue. Error bars represent SD between replicates. RNAi and mutant strains represented by checkered bars, controls by solid bars. Significant differences between RNAi or mutant and controls are indicated by *. * = $p < 0.05$, ** = $p < 0.005$, *** = $p < 0.0005$. AL is shown in red, DR in blue.

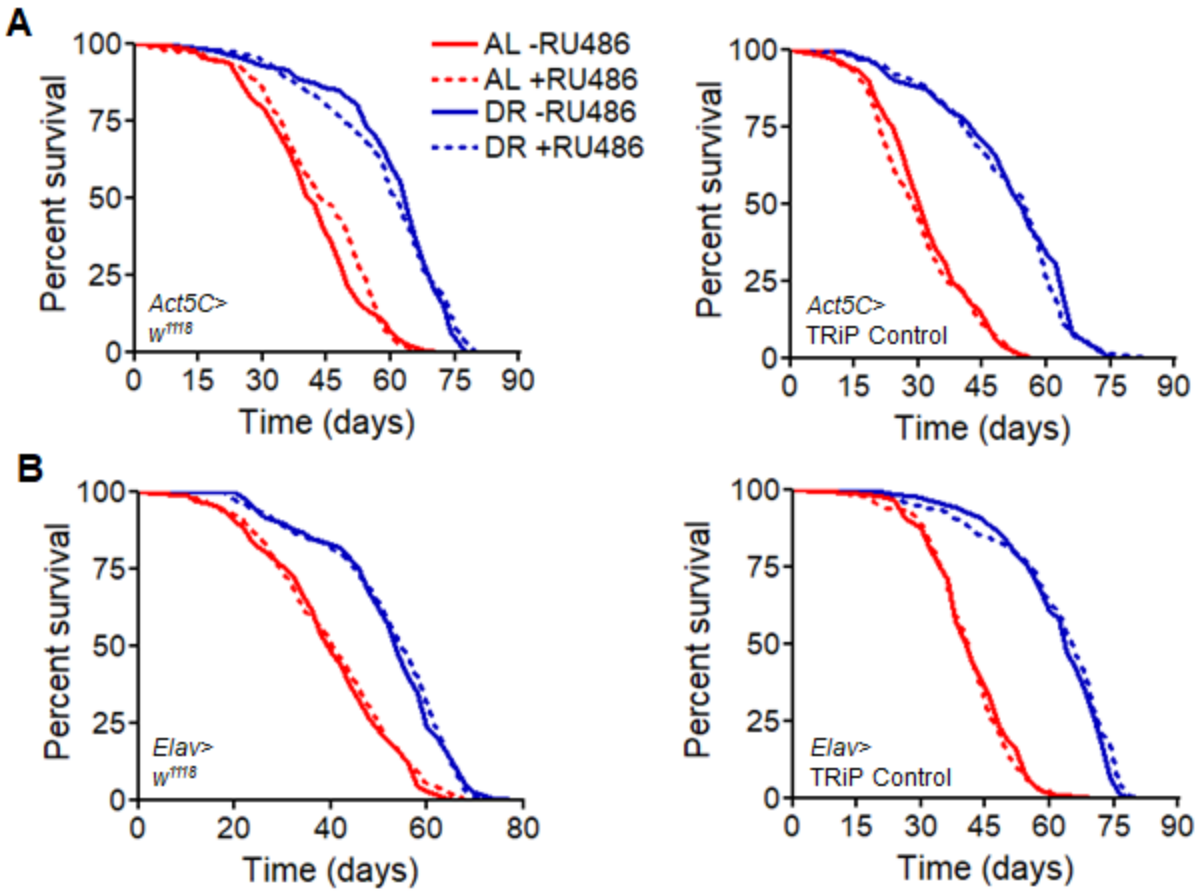

Figure S5. The effect of RU486 on control lines crossed with different GeneSwitch drivers. Related to Figure 4. (A) Lifespan of ubiquitous expression driver *Act5C*-GS-Gal4 crossed with w^{1118} control strain (top) and crossed with TRiP control line (bottom). (B) Lifespan of pan-neuronal expression driver *Elav*-GS-Gal4 crossed with w^{1118} control strain (top) and crossed with TRiP control line (bottom).

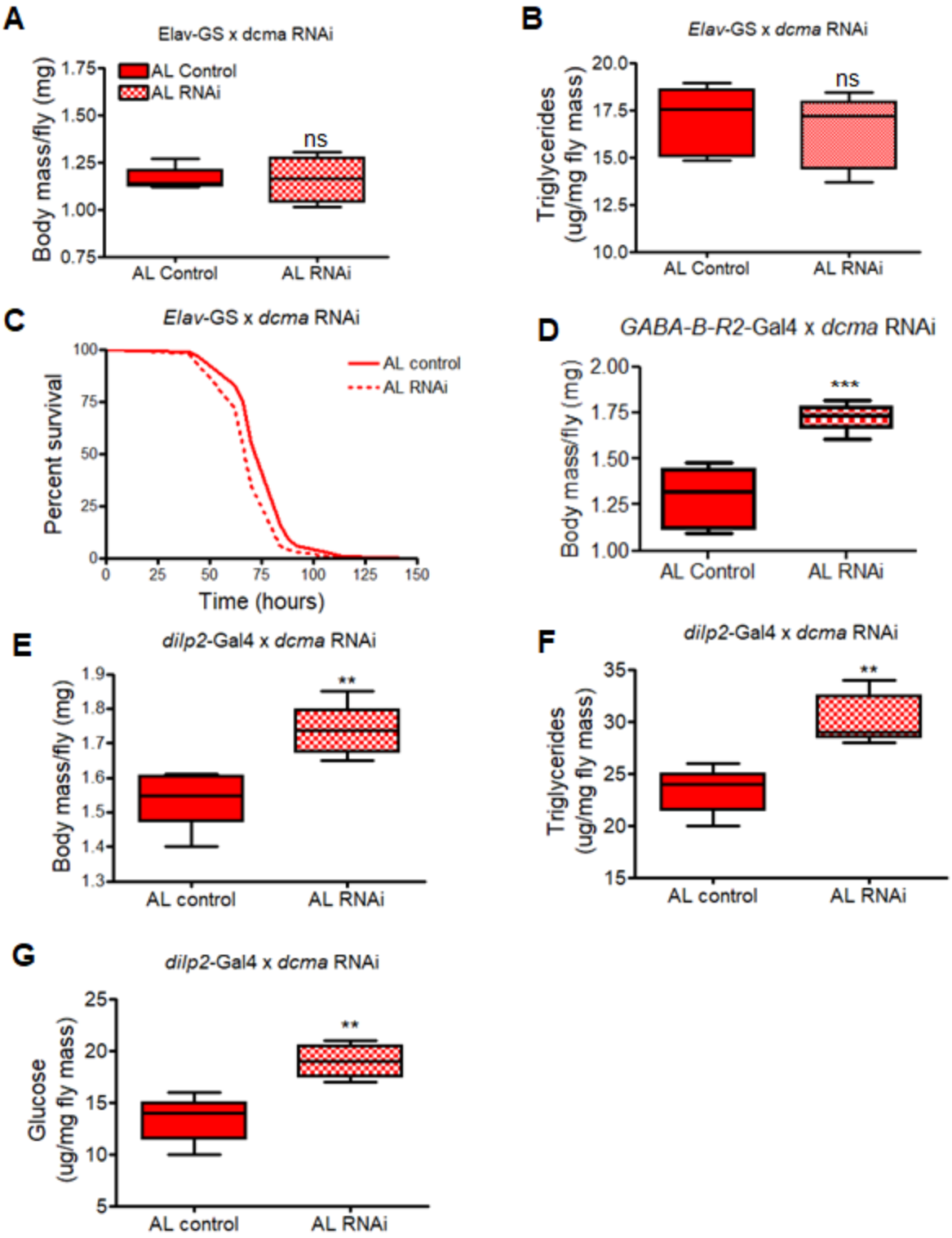

Figure S6. Phenotypic analysis of *decima*. Related to Figure 5. (A) Body mass of flies with pan-neuronal RNAi against *dcma* after 7 days under AL diet. (B) Triglyceride levels of flies with pan-neuronal *dcma* RNAi after 7 days under AL diet, normalized to body mass. (C) Starvation resistance of flies with pan-neuronal *dcma* RNAi after rearing under AL diet. (D) Body mass of flies with *dcma* RNAi driven in GABA receptor neurons after 7 days under AL diet. (E) Body

mass of flies after 7 days of *dilp2*-driven *dcma* RNAi. (F) Triglyceride levels of flies with *dilp2*-driven *dcma* RNAi after 7 days under AL diet, normalized to body mass. (G) Glucose levels in whole flies after 7 days of *dilp2*-driven *dcma* RNAi, normalized to body mass. For body mass and triglyceride experiments, N = 15. For starvation experiments, N = 200 flies per condition. All data represents 3 biological replicates. Significant differences indicated by *. * = $p < 0.05$, ** = $p < 0.005$, *** = $p < 0.0005$. ns = not significant. p values shown in Data S3. All flies were maintained on AL diet. Checkered boxes represent RNAi. Error bars represent SD between replicates.

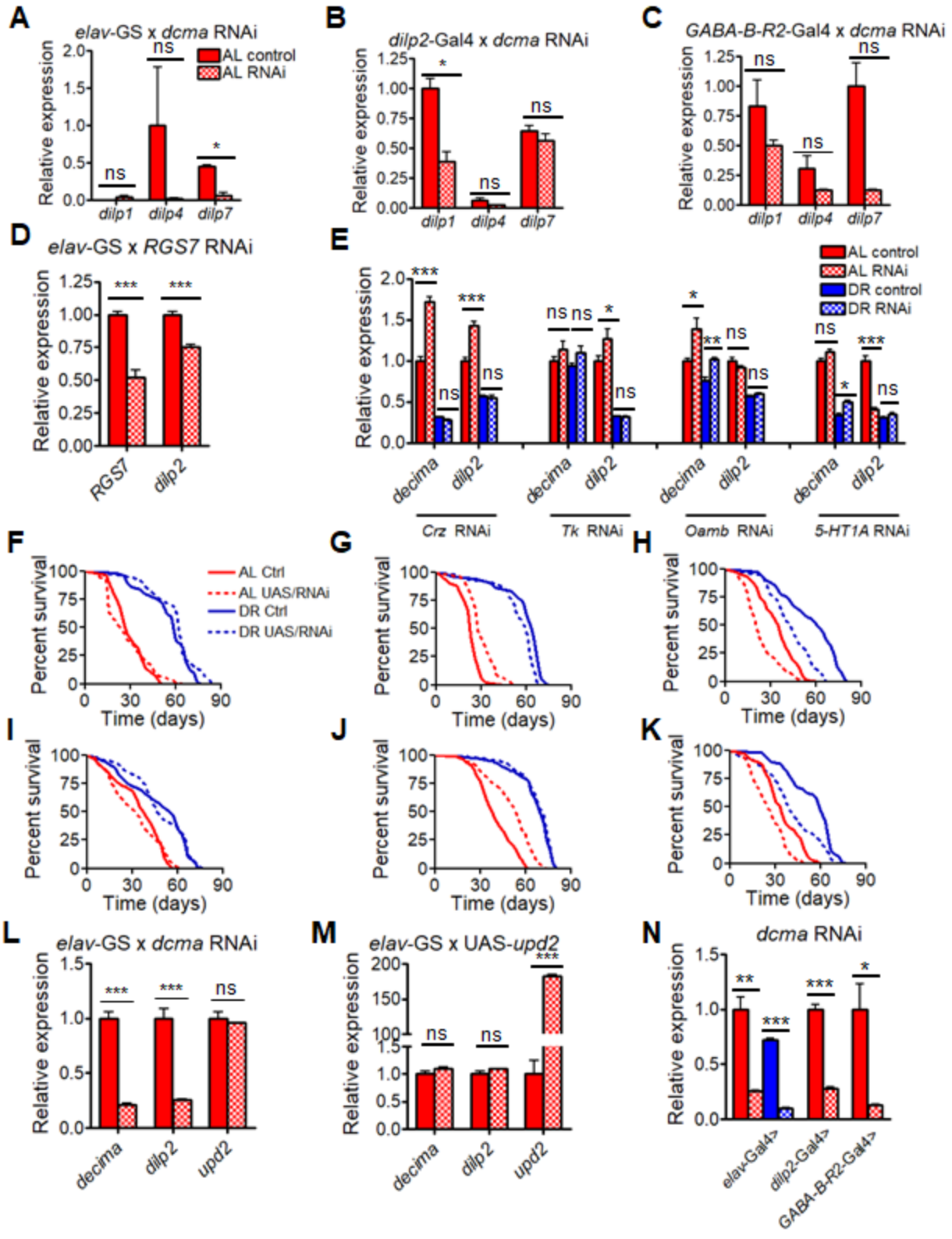

Figure S7. Pathway analysis of *decima*. Related to Figure 6. (A-C) Expression of non-neuronal dilps in the heads of flies under AL with *dcma* RNAi driven (A) pan-neuronally, (B) in IPCs, (C) in GABA receptor neurons. (D) Expression of *RGS7* (left) and *dilp2* (right) in heads of

flies under AL with pan-neuronal RNAi against *RGS7*. (E) Expression of *dcma* and *dilp2* with pan-neuronal RNAi against *Crz* (left), *Tk* (2nd to left), *Oamb* (2nd to right), and *5-HT1A* RNAi (right). (F-K) Lifespan data for *dilp2*-driven (F) RNAi for *dcma* and overexpression of *dilp2*, (G) *dcma* RNAi alone, (H) or *dilp2* overexpression, or pan-neuronally-driven (I) *dcma* RNAi and overexpression of *dilp2*, (J) *dcma* RNAi alone, or (K) overexpression of *dilp2*. (L) Expression of *decima* (left), *dilp2* (middle), and *upd2* (right) in heads under AL with pan-neuronal RNAi against *dcma*. (M) Expression of *decima* (left), *dilp2* (middle), and *upd2* (right) in heads from flies under AL with pan-neuronal overexpression of *upd2*. (N) RNAi against *dcma* driven with *elav*-Gal4 (left), *dilp2*-Gal4 (middle), and *GABA-B-R2*-Gal4 (right). All expression relative to *rp49* housekeeping gene. N = 50 heads per condition for qRT-PCR data. N = 200 flies per condition for lifespan experiments. All data represents 3 biological replicates. Significant differences indicated by *. * = $p < 0.05$, ** = $p < 0.005$, *** = $p < 0.0005$. ns = not significant. Error bars represent SD between replicates. In lifespan graphs, red indicates AL and blue indicates DR. Solid lines represent control conditions and dotted lines represent experimental conditions.