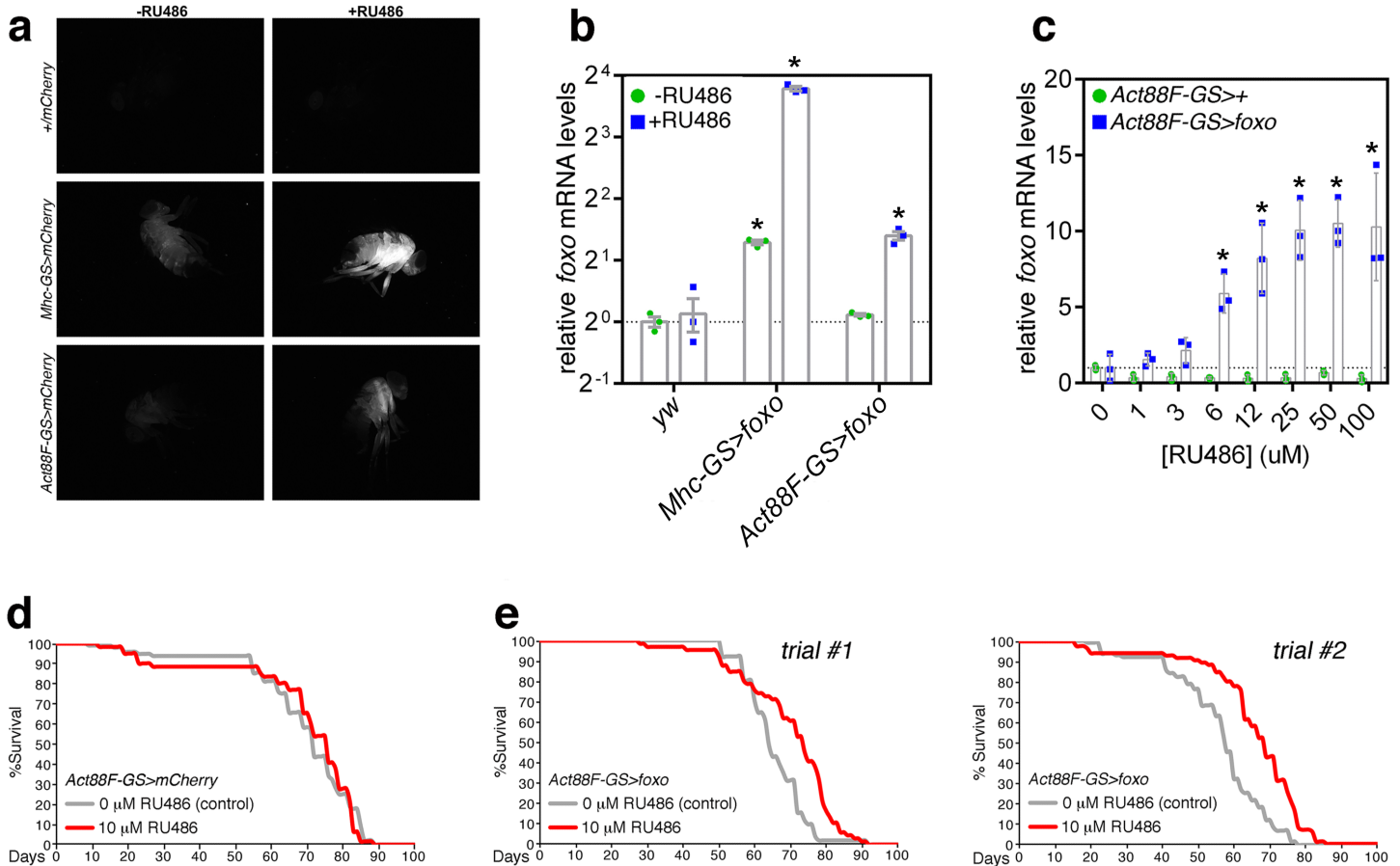


**SUPPLEMENTARY INFORMATION (Robles-Murguia et al.)**

**Tissue-specific alteration of gene expression and function by RU486 and the GeneSwitch system**

## SUPPLEMENTARY FIGURES



**Supplementary Figure 1.** Characterization of the *Act88F-GS-Gal4* driver for drug-induced transgenic expression in *Drosophila*. **a** Compared to the *Mhc-GS-Gal4*, which is leaky<sup>1</sup>, *Act88F-GS-Gal4* drives expression of *mCherry* transgenes only in presence of RU486 but not in its absence. As inferred from the fluorescence of *mCherry* transgenes, *Act88F-GS-Gal4* drives transgenic expression specifically in thoracic flight muscles and to a lesser extent in leg muscles. No expression is detected in non-muscle tissues. **b** Consistently, *foxo* expression is induced by *Act88F-GS-Gal4* only in presence of RU486 but not in its absence, whereas leaky *foxo* expression occurs with *Mhc-GS-Gal4* even in the absence of RU486. **c** Transgenic *foxo* expression with *Act88F-GS-Gal4* is dose-dependent. In **b-c**,  $n=3$  and the error bars represent the SD.

**d** *mCherry* expression induced with 10  $\mu$ M RU486 and *Act88F-GS-Gal4* does not extend lifespan ( $P=0.25$ ;  $n[\text{control}]=72$  and  $n[\text{RU486}]=76$ ). **e** RU486-induced overexpression of wild-type *foxo* with *Act88F-GS-Gal4* significantly extends lifespan (trial #1:  $P=0.0014$ ;  $n[\text{control}]=64$  and  $n[\text{RU486}]=74$ ); trial #2:  $P<0.0001$ ;  $n[\text{control}]=58$  and  $n[\text{RU486}]=69$ ), consistent with previous results<sup>2</sup>, obtained with a constitutive *Mhc-Gal4* driver.

## SUPPLEMENTARY REFERENCES

- 1 Poirier, L., Shane, A., Zheng, J. & Seroude, L. Characterization of the *Drosophila* gene-switch system in aging studies: a cautionary tale. *Aging Cell* **7**, 758-770, doi:10.1111/j.1474-9726.2008.00421.x (2008).
- 2 Demontis, F. & Perrimon, N. FOXO/4E-BP signaling in *Drosophila* muscles regulates organism-wide proteostasis during aging. *Cell* **143**, 813-825, doi:10.1016/j.cell.2010.10.007 (2010).

## SUPPLEMENTARY TABLES

Control					
<i>WT strains</i>	Non-flyers	Flyers	Percentage		
w1118	8	109	6.84		
B3	2	112	1.75		
<i>GeneSwitch strains</i>					
ACT5C	0	66	0.00		
WB-FB	0	85	0.00		
MHC	3	59	4.84		
ACT88F	5	79	5.95		

1uM RU486					
<i>WT strains</i>	Non-flyers	Flyers	Percentage	Fisher P=	Chi+Yates P=
w1118	8	117	6.40	1.0000	0.8911
B3	9	130	6.47	0.1179	0.1280
<i>GeneSwitch strains</i>					
ACT5C	4	60	6.25	0.0559	0.1199
WB-FB	4	92	4.17	0.1236	0.1626
MHC	2	48	4.00	1.0000	0.8308
ACT88F	61	8	88.41	0.0001	0.0001

10uM RU486					
<i>WT strains</i>	Non-flyers	Flyers	Percentage	Fisher P=	Chi+Yates P=
w1118	14	135	9.40	0.5076	0.5977
B3	6	103	5.50	0.1636	0.2522
<i>GeneSwitch strains</i>					
ACT5C	6	56	9.68	0.0113	0.0300
WB-FB	1	87	1.14	1.0000	0.3243
MHC	11	58	15.94	0.0492	0.0766
ACT88F	73	5	93.59	0.0001	0.0001

100uM RU486					
<i>WT strains</i>	Non-flyers	Flyers	Percentage	Fisher P=	Chi+Yates P=
w1118	23	116	16.55	0.0207	0.0293
B3	3	138	2.13	1.0000	0.8307
<i>GeneSwitch strains</i>					
ACT5C	4	61	6.15	0.0578	0.1238
WB-FB	2	89	2.20	0.4977	0.5073
MHC	33	42	44.00	0.0001	0.0001
ACT88F	76	5	93.83	0.0001	0.0001

**Supplementary Table 1.** Statistical analysis of flight assays.