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Supplementary material

Exposure to males, but not receipt of sex peptide, accelerates functional aging in female fruit flies

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Table S1 AIC values from GLMM/GLMs assessing the effect of exposure to males on senescence in female traits. Wild type females were assigned at random to one of three male exposure treatments (treatment). Females either encountered no males during their lifetime and remained virgin, were continuously held with males or experienced an intermittent exposure regime. Responses were then assayed at different female ages. We initially assessed whether a linear or quadratic effect of female age within treatments was most appropriate, and where this was the case for at least one treatment used the quadratic term in the full model. We first tested the overall effect of exposure treatment including the effect of mating (comparison across all treatments) and then the effect of amount of exposure by comparing only the treatments that had mated. In the case of climbing activity this was measured multiple times on the same fly, hence the inclusion of identity (ID) as a random factor. The model highlighted in bold is the model which minimized AIC and from which removing a term significantly altered the model (as tested using Analysis of Deviance).

Response	Treatments	Model	#Parameter	AIC
Climbing time	all	treatment * age * lifespan + (1 ID)	7 + (1 ID)	9569.9
treatm	treatment + age + lifespan + (1 ID) + treatment:age + treatment:lifespan + age:lifespan	6+ (1 ID)	9575.2	
	null mated only treatment * age * lifespan + (1 ID)		0+ (1 ID)	9783.5
			7+ (1 ID)	4742.5
		treatment + age + lifespan + (1 ID) + treatment:age + treatment:lifespan + age:lifespan	6+ (1 ID)	4748.9
		null	0+ (1 ID)	4816.8

Starvation survival	all	treatment*age*I(age^2)	7	1942.0
		treatment + age + I(age^2) + treatment:I(age^2) + age:I(age^2) + treatment:I(age^2)	6	1938.6
		<pre>treatment + age + I(age^2) + treatment:age + treatment:I(age^2)</pre>	5	1937.6
		treatment + age + $I(age^{2})$ + treatment: age	4	1945.6
		null	0	2052.7
	mated only	treatment*age*I(age^2)	7	1340.2
		$treatment + age + I(age^{2}) + treatment:I(age^{2}) + age:I(age^{2}) + treatment:I(age^{2})$	6	1338.2
		treatment + age + I(age^2) + treatment:age + treatment:I(age^2)	5	1336.7
		<pre>treatment + age + I(age^2) + treatment:age</pre>	4	1335.5
		treatment + age + $I(age^{2})$	3	1340.6
		null	0	1397.7
Body mass	all	treatment*age	3	201.9
		treatment + age	2	205.7
		null	0	206.4
TAGs/mg	all	treatment*age	3	893.9
		treatment + age	2	892.3
		treatment	1	891.1
		age	1	890.7
		null	0	889.5

Model	Term	Estimate	S.E.
Climbing time, all treatments.	intercept	0.968	25.301
treatment * age * lifespan + (1 ID)	treatment	-5.347	12.672
	age	0.227	1.102
	lifespan	-0.342	0.493
	treatment:age	2.022	0.740
	treatment:lifespan	0.223	0.336
	age:lifespan	0.042	0.022
	treatment:age:lifespan	-0.052	0.019
Climbing time, mated treatments only	intercept	98.439	79.655
treatment * age * lifespan + (1 ID)	treatment	-44.040	32.049
	age	-12.796	5.730
	lifespan	-2.728	2.360
	treatment:age	7.200	2.343
	treatment:lifespan	1.195	0.994
	age:lifespan	0.382	0.152
	treatment:age:lifespan	-0.190	0.065
Starvation survival, all treatments,	intercept	2.224	0.075
treatment + age + I(age^2) + treatment:age + treatment:I(age^2)	treatment(intermittent)	0.122	0.084
	treatment(constant)	0.103	0.083
	age	-0.017	0.008
	I(age^2)	0.000	0.000
	treatment(intermittent):age	-0.003	0.005
	treatment(constant):age	0.008	0.004
Starvation survival, mated treatments only	intercept	2.255	0.078
treatment + age + $I(age^2)$ + treatment:age	treatment(constant)	-0.025	0.081
	age	-0.004	0.009
	I(age^2)	-0.001	0.000
	treatment(constant):age	0.011	0.004
Body mass, all treatments	intercept	5.916	0.444
treatment*age	age	0.054	0.021
	treatment	0.629	0.203
	age:treatment	-0.023	0.010

Table S2 Model parameter estimates (and S.E.) for terms in the best supported models as reported in Table S1.

Table S3 AIC values from GLMM/GLMs assessing the effect of receipt of sex peptide on senescence in female traits. Wild type females were assigned at random to be continuously exposed to either males lacking sex peptide (SP) or SP transferring control males (treatment). Responses were then assayed at different female ages. We initially assessed whether a linear or quadratic effect of female age within treatments was most appropriate, and where this was the case for at least one treatment used the quadratic term in the full model. In the case of climbing activity this was measured multiple times on the same fly, hence the inclusion of identity (ID) as a random factor. The model highlighted in bold is the model which minimized AIC and removing a term significantly altered the model (as tested using Analysis of Deviance).

Response	Model	# Parameters	AIC
Climbing time	treatment * age * lifespan + (1 ID)	7+ (1 ID)	2142.5
	treatment + age + lifespan + (1 ID) + treatment:age + treatment:lifespan + age:lifespan	6+ (1 ID)	2141.3
	treatment + age + lifespan + (1 ID) + treatment:age + treatment:lifespan	5+ (1 ID)	2139.4
	treatment + age + lifespan + $(1 ID)$ + treatment:age	4+ (1 ID)	2137.5
	treatment + age + lifespan + $(1 ID)$	3+ (1 ID)	2136.0
	treatment + age + $(1 ID)$	2+ (1 ID)	2134.2
	treatment + $(1 ID)$	1+ (1 ID)	2136.1
	age + (1 ID)	1+ (1 ID)	2132.3
	null	0+ (1 ID)	2640.0
Starvation survival	treatment*age*I(age^2)	7	1290.8
	treatment + age + I(age ²) + treatment:I(age ²) + age:I(age ²) + treatment:age:I(age ²)	6	1291.9
	treatment + age + I(age^2) + treatment:age + treatment:I(age^2)	5	1289.9
	treatment + age + $I(age^{2})$ + treatment:age	4	1288.6
	treatment + age + $I(age^{2})$	3	1288.3
	treatment + age	2	1303.0
	age + I(age^2)	2	1288.8
	age	1	1303.3
	null	0	1375.6

Table S4 Parameter estimates (and S.E.) for terms in the best supported models from GLMM/GLMs assessing the effect of receipt of sex peptide on senescence in female traits as reported in Table S3.

Model	Term	Estimate	S.E.
			0.0.57
Climbing time	intercept	2.145	0.065
	age	0.031	0.004
Starvation survival	intercept	23.874	1.322
	age	-1.401	0.224
	I(age^2)	0.033	0.008



Figure S1 Adult lifespan and the change in climbing time between the first and last assay (day 32 – day 4). A larger positive value indicates a greater decline in climbing ability. Females were kept singly as virgins (black dots, solid line), exposed to one male for 3 days per week (grey dots, dashed line), or constantly exposed to one male (white dots, dotted line) and their climbing ability was assessed weekly and measured as the time taken to reach 8cm. Trend lines are fitted for illustration.