

1 **Supplementary Material**

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3 *Methods - Bacterial Growth Curves*

4 To initiate the culture, bacteria were taken from the spaceflight or ground control 50% glycerol
5 stocks that were stored at -80⁰C in 50 µl aliquots, and then placed in a 50 mL subculture
6 containing fresh LB media with 100 µg/mL streptomycin at ~1x10⁸ CFU/ml density (as estimated
7 by spectrophotometry). Samples were then grown at 37°C in a shaker rotating at 225 rpm.
8 Estimates of growth (colony forming units) were taken every 2 hours from the above culture for
9 30 hours total by diluting 20 µl of the sample in 180 µl of fresh LB media, then reading
10 immediately at 600 nm on a NanoDrop 2000c (Thermo Scientific) blanked with the initial
11 measurement. Both Space and Ground treatments were run in triplicate and averaged at each
12 timepoint, and the entire experiment was repeated independently three times with spaceflight
13 and ground samples. Data was analyzed in JMP Pro 12 using an ANOVA with repeated
14 measures.

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16 *Results – Bacterial growth curves*

17 *In vitro* growth was significantly higher in the spaceflight bacteria compared to the ground
18 bacteria of the same strain ($t=4.66$, $P<0.0001$).

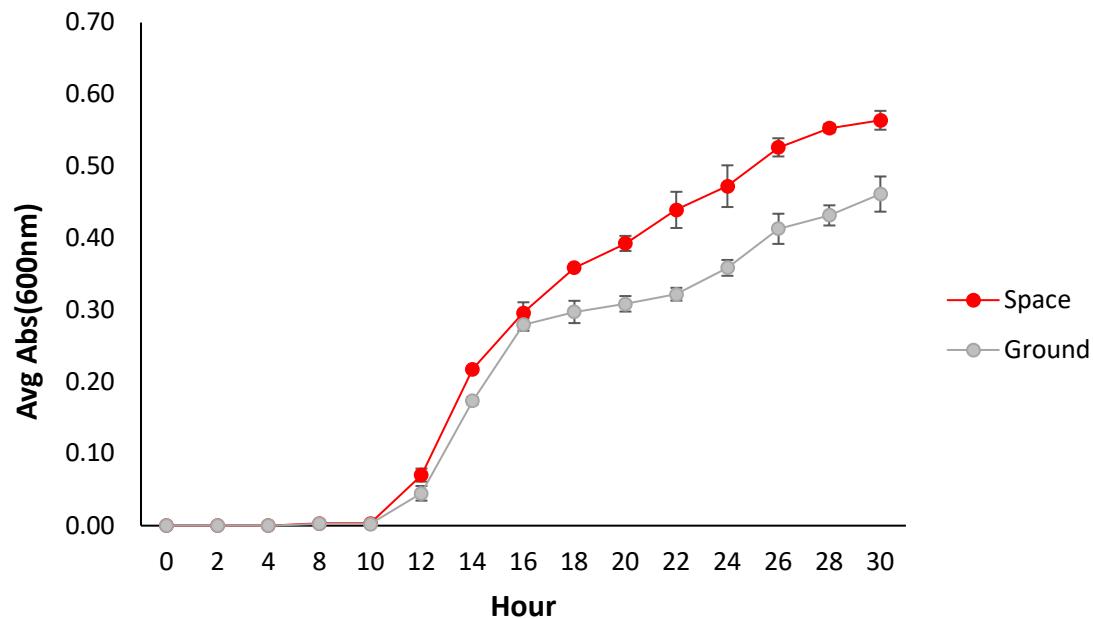
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Supplementary Figure 1. Growth curves for spaceflight bacteria compared to ground controls

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in vitro. Spaceflight and ground samples were both stored in 50% glycerol stock and diluted to starting concentrations of $\sim 1 \times 10^8$ CFU. Growth at 37°C was then measured by taking a sample every 2 hours for 30 hours total, diluting the sample 10x, and measuring ABS at 600nm on a NanoDrop spectrophotometer. Error bars represent 1 standard error.

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37 **Supplementary Table 1.** Transcript-level differential expression from RNAseq of ground-reared
 38 flies infected with either space or ground control *Serratia marcescens*, calculated using sleuth
 39 package Wald test. Transcripts with q-values < 0.05 were considered significantly differentially
 40 expressed, these are the only values included in this table. All other values were >0.05.

target_id	ens_gene	ext_gene	pval	qval	[-log10(qval)]	beta
FBtr0310667	FBgn0033062	Ars2	8.64E-09	0.00017907	3.746974596	-5.8759894
FBtr0085912	FBgn0000559	eEF2	2.61E-06	0.01355726	1.867828177	-7.2844011
FBtr0088122	FBgn0010357	betaTry	2.62E-06	0.01355726	1.867828177	0.76007008
FBtr0075412	FBgn0014163	fax	2.11E-06	0.01355726	1.867828177	-1.238556
FBtr0305083	FBgn0023172	RhoGEF2	4.92E-06	0.01700121	1.769520087	-7.1835147
FBtr0333107	FBgn0260442	rhea	4.84E-06	0.01700121	1.769520087	5.2835701
FBtr0087437	FBgn0012042	AttA	6.01E-06	0.01779894	1.749605913	4.65161273
FBtr0077042	FBgn0035666	Jon65Aii	7.16E-06	0.01853873	1.731919905	0.87658333
FBtr0089350	FBgn0283521	lola	8.76E-06	0.01859388	1.730629941	7.20501561
FBtr0076398	FBgn0045823	vsg	8.98E-06	0.01859388	1.730629941	6.99749765
FBtr0079035	FBgn0031673	CG31650	2.58E-05	0.04856692	1.31365946	-6.1667293

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48 **Supplementary Table 2.** Statistical output for proportional hazards analysis of *dif¹* mutant *D. melanogaster* line after infection with ground and space bacteria samples.

Level 1	Level 2	Risk Ratio	P>ChiSq
Space	Ground	28.63	<0.0001
Space	12.5% Glycerol	75.5	<0.0001
Ground	12.5% Glycerol	1.89	0.052

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51 **Supplementary Table 3.** Statistical output for proportional hazards analysis of PGRP-SA^{semI}
52 mutant *D. melanogaster* line after infection with ground and space bacteria samples.

Level 1	Level 2	Risk Ratio	P>ChiSq
Space	Ground	14.6	<0.0001
Space	12.5% Glycerol	45.51	<0.0001
Ground	12.5% Glycerol	2.11	0.068

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54 **Supplementary Table 4.** Statistical output for proportional hazards analysis of PGRP-LC^{Δ5}
55 mutant *D. melanogaster* line after infection with ground and space bacteria samples.

Level 1	Level 2	Risk Ratio	P>ChiSq
Space	Ground	8.62	<0.0001
Space	12.5% Glycerol	13.6	<0.0001
Ground	12.5% Glycerol	8.22	<0.0001

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57 **Supplementary Table 5.** Statistical output for proportional hazards analysis of *Imd*¹ mutant *D.*
58 *melanogaster* line after infection with ground and space bacteria samples.

Level 1	Level 2	Risk Ratio	P>ChiSq
Space	Ground	3.04	0.036
Space	12.5% Glycerol	14.6	<0.0001
Ground	12.5% Glycerol	11.21	<0.0001

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60 **Supplementary Table 6.** Statistical output for proportional hazards analysis of *ref*^{E20} mutant *D.*
61 *melanogaster* line after infection with ground and space bacteria samples.

Level 1	Level 2	Risk Ratio	P>ChiSq
Space	Ground	3.67	0.027
Space	12.5% Glycerol	16.44	<0.0001
Ground	12.5% Glycerol	9.7	<0.0001

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