

SUPPLEMENTAL DATA

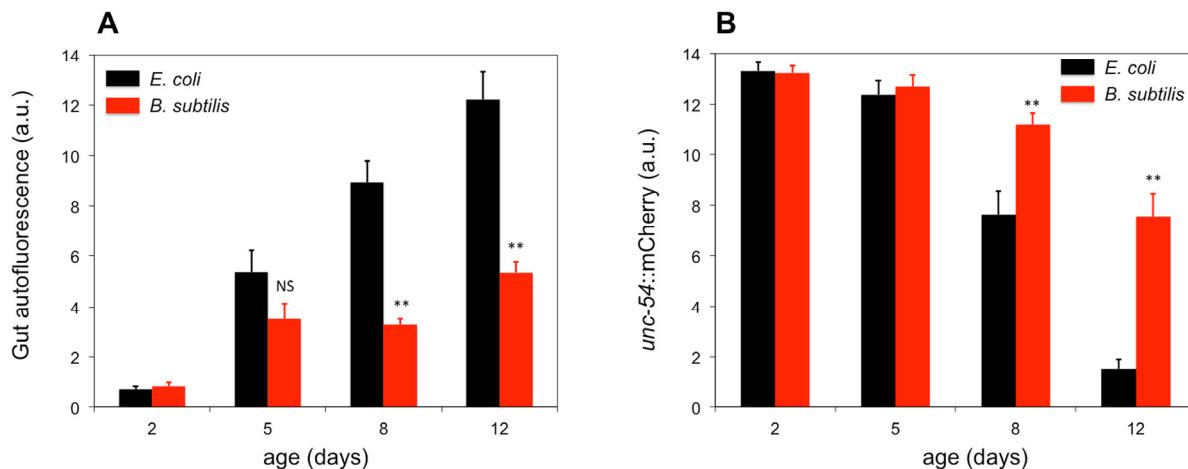


Figure S1. Gut autofluorescence and *unc-54::mCherry* reporter as indicators of the overall health of *B. subtilis* and *E. coli* fed worms. (A) Gut autofluorescence intensity for *B. subtilis* and *E. coli* fed adult hermaphrodites during aging. y-axis shows levels of gut autofluorescence in arbitrary units. x-axis shows age of worms. (B) *unc-54::H1::mCherry* fluorescent marker expression for *B. subtilis* and *E. coli* fed adult hermaphrodites during aging. y-axis shows levels of fluorescent expression in arbitrary units. x-axis shows age of worms. (A-B) Bars indicate the mean fluorescent marker expression ± S.E.M. n = 10-15 for each group (**p < 0.01; NS p > 0.05; Student's t test).

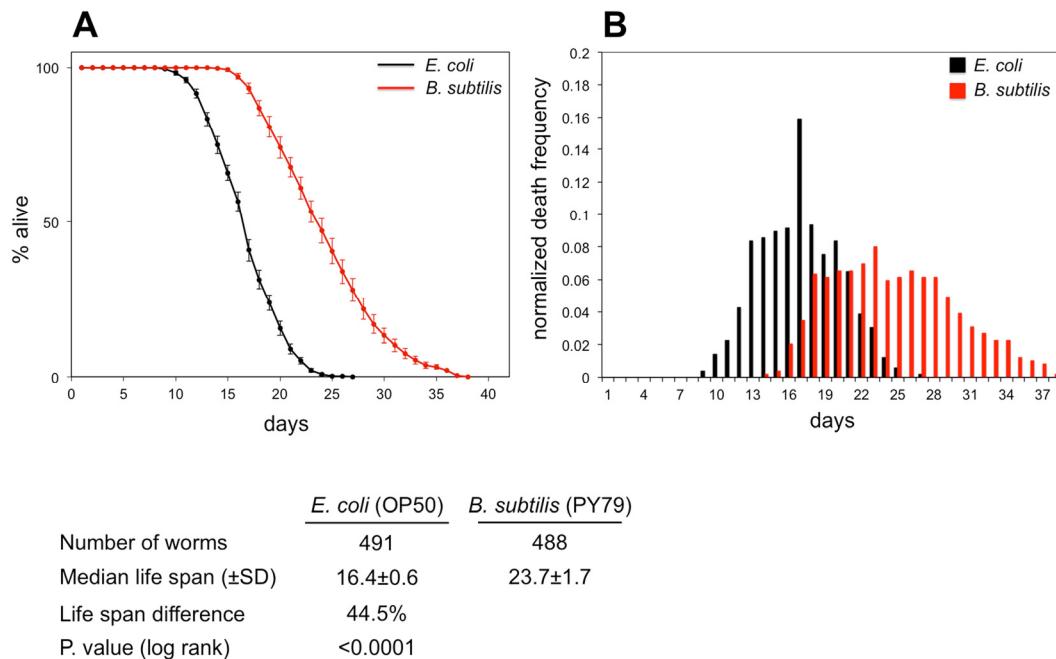


Figure S2. *B. subtilis* fed worms live longer than *E. coli* fed worms and display different demographics of death. (A) Represented are the life span curves for adult hermaphrodite worms maintained on *E. coli* (OP50) or on *B. subtilis* (PY79). The number of dead worms was counted on a daily basis. The worm populations were divided into 10 plates. The points in the life span curves represent the daily mean percentage of alive worms in the 10 plates \pm S.E.M. y-axis indicates percentage of worms that are alive. x-axis indicates day of adulthood. (B) Represented are the daily death frequencies normalized to their respective sample size.

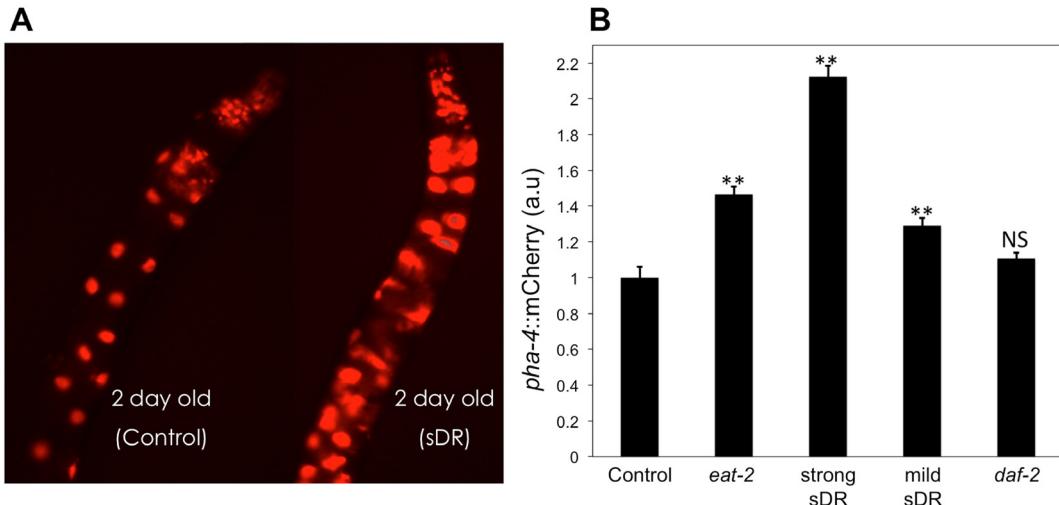


Figure S3. *pha-4::mCherry* reporter as a worm nutritional indicator. (A) Representative pictures of *pha-4::H1::mCherry* fluorescent marker expression for 2 day old WT adult hermaphrodites, which were exposed to plenty of food (Control) or dietary restricted (sDR). (B) *pha-4::H1::mCherry* fluorescent marker expression for 2 day old WT adult hermaphrodites, which were exposed to plenty of food (Control) or dietary restricted (sDR); and for 2 day old *eat-2(ad1116)* and *daf-2(e1370)* mutant adult hermaphrodites, which were exposed to plenty of food. For details on dietary restriction conditions see the Materials and Methods section. y-axis shows levels of fluorescent expression in arbitrary units. x-axis shows the type of worms and the diet condition. Bars indicate the mean fluorescent marker expression \pm S.E.M. n = 15 for each group (**p < 0.01, Student's t test). (A-B) In all cases worms were fed *E. coli*.

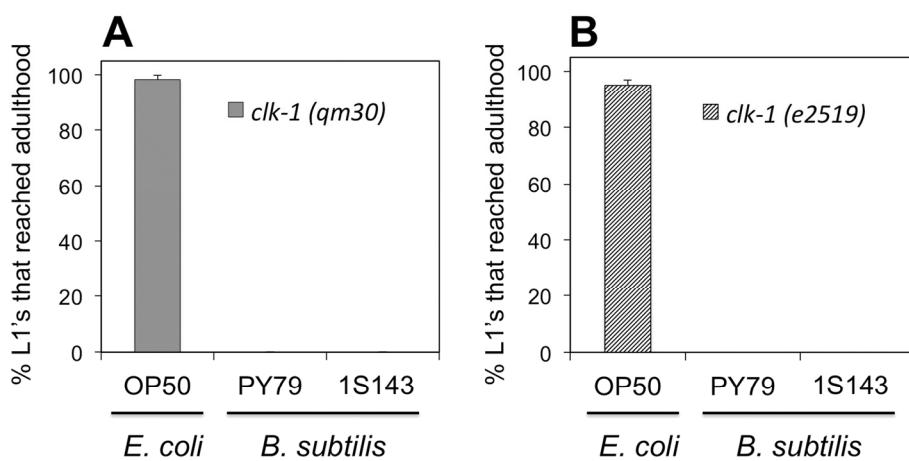


Figure S4. *clk-1* mutant worms are unable to develop into adults when fed the *B. subtilis* diet. (A) Percentage of *clk-1(qm30)* L1 larvae that developed into adults after 3.9 days of feeding on *E. coli* (OP50), *B. subtilis* (PY79) or the spore-less *B. subtilis* (1S143). (B) Percentage of *clk-1(e2519)* L1 larvae that developed into adults after 4.0 days of feeding on *E. coli* (OP50), *B. subtilis* (PY79) or the spore-less *B. subtilis* (1S143). (A-B) y-axis shows percentage of L1 larvae that reached adulthood. x-axis shows the diet used. Bars indicate the mean value \pm S.E.M. n = 140-160 worms per group.

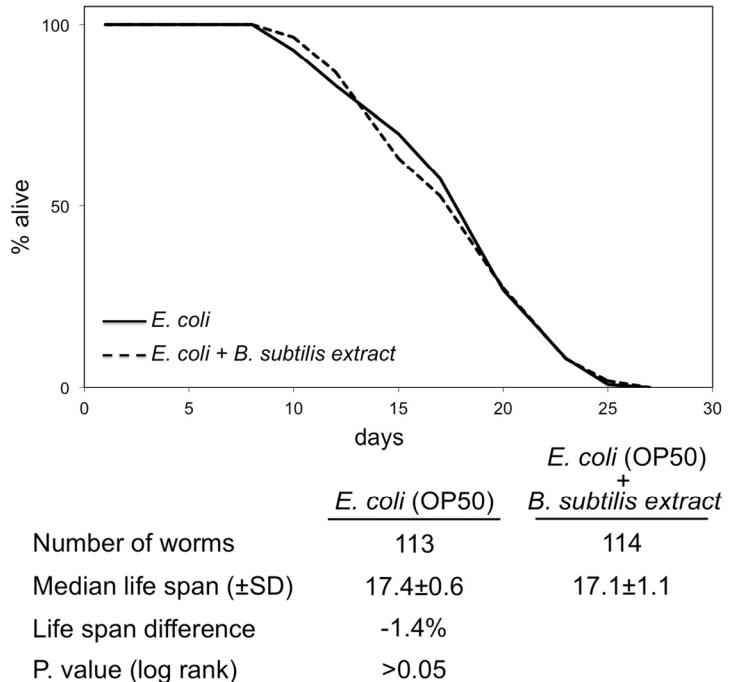


Figure S5. *E. coli* fed worms supplemented with coQ-less *B. subtilis* extract do not alter their longevity. Represented are the life span curves for adult hermaphrodite worms maintained on *E. coli* with or without supplementation of coQ-less *B. subtilis* extract. The spore-less *B. subtilis* (1S143) strain was used to prepare the *B. subtilis* extract with the purpose of avoiding the presence of *B. subtilis* spores in the extract. y-axis indicates percentage of worms that are alive. x-axis indicates day of adulthood.

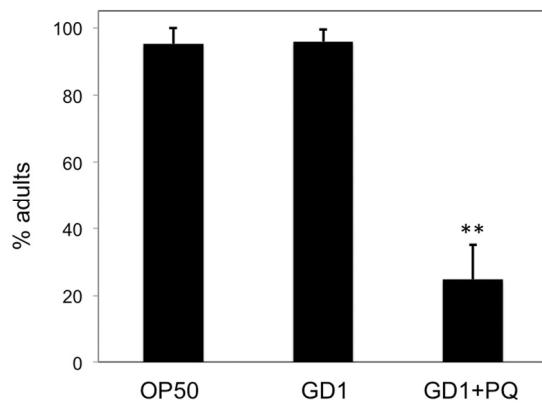


Figure S6. Treatment with a mild dose of paraquat severely affects the development of worms feeding on coQ-defective *E. coli* (GD1) bacteria. Development of synchronized wild type N2 L1 larvae after 3 days feeding on *E. coli* OP50 and *E. coli* GD1 with or without PQ treatment (0.1 mM). y-axis shows percentage of individuals that reached adulthood after 3 days. x-axis shows the type of *E. coli* and the treatment. Bars indicate the mean value \pm S.D. n = 114-127 worms per group. **p < 0.001, Student's t test.

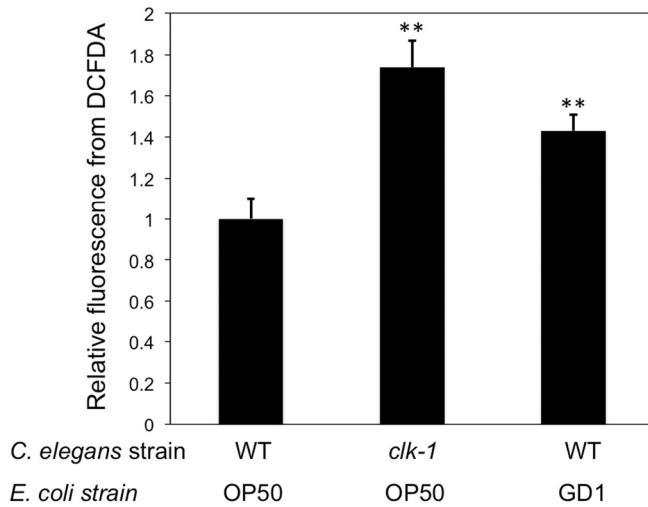


Figure S7. *E. coli* (GD1) fed wild type worms and *E. coli* (OP50) fed *clk-1(qm30)* worms have higher ROS levels than *E. coli* fed WT worms. Bars indicate the relative mean fluorescent marker expression \pm S.E.M difference relative to the *E. coli* fed WT worms. n = 14-20 for each group (** $p < 0.01$, Student's t test). y-axis indicates relative fluorescence from DCFDA. x-axis indicates diet and type of worms.

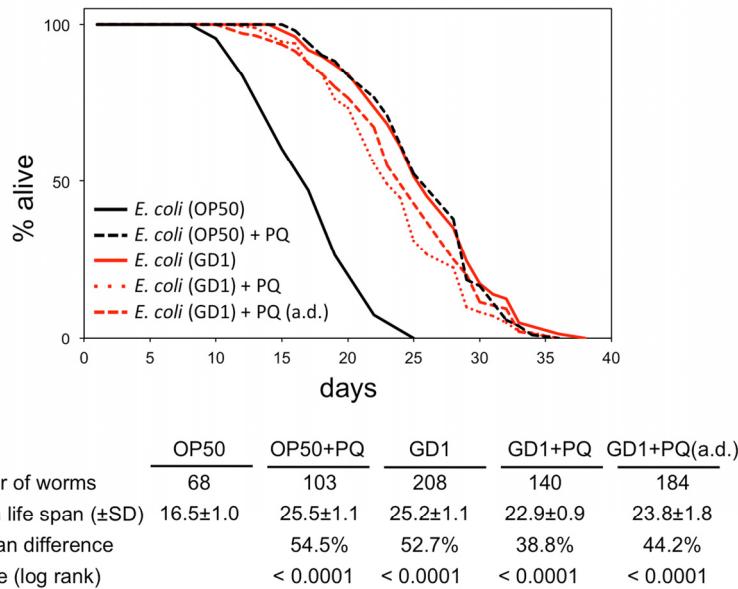


Figure S8. Life span extension of paraquat-treated worms depends on the presence of coQ in the *E. coli* diet. Represented are the life span curves for adult worms maintained on coQ-active *E. coli* OP50 or coQ-deficient *E. coli* GD1 with or without PQ treatment (0.1 mM). Worms were subjected to PQ treatment since L1, or since adulthood (a.d.). y-axis indicates percentage of worms that are alive. x-axis indicates day of adulthood.

Table S1. Supporting life span data for Figure 1

| <i>C. elegans</i> strain | Bacterial food strain | Sample size | Median life span ± SD | % Life span change | P. value (log rank) |
|--------------------------|---------------------------|-------------|-----------------------|--------------------|---------------------|
| Wild type (N2) | <i>E. coli</i> (OP50) | 178 | 14.2±0.8 | | |
| Wild type (N2) | <i>B. subtilis</i> (168) | 169 | 20.4±0.5 | +43.1% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (3610) | 148 | 22.2±1.5 | +56.0% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) | 170 | 22.6±1.0 | +58.5% | <0.0001 |

| <i>C. elegans</i> strain | Bacterial food strain | Sample size | Median life span ± SD | % Life span change | P. value (log rank) |
|--------------------------|----------------------------|-------------|-----------------------|--------------------|---------------------|
| Wild type (N2) | <i>E. coli</i> (OP50) | 107 | 15.5±0.3 | | |
| Wild type (N2) | <i>B. subtilis</i> (PY79) | 118 | 25.2±0.4 | +62.6% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (1S143) | 94 | 22.7±1.2 | +46.5% | <0.0001 |

| <i>C. elegans</i> strain | Bacterial food strain | Sample size | Median life span ± SD | % Life span change | P. value (log rank) |
|--------------------------|-------------------------------------|-------------|-----------------------|--------------------|---------------------|
| Wild type (N2) | <i>E. coli</i> (OP50) | 76 | 15.9±0.4 | | |
| Wild type (N2) | <i>B. subtilis</i> (PY79) | 132 | 23.0±0.7 | +44.7% | <0.0001 |
| Wild type (N2) | UV killed <i>B. subtilis</i> (PY79) | 81 | 22.7±0.8 | +42.8% | <0.0001 |
| Wild type (N2) | UV killed <i>E. coli</i> (OP50) | 96 | 19.2±0.4 | +20.8% | <0.001 |

| <i>C. elegans</i> strain | Bacterial food strain | Sample size | Median life span ± SD | % Life span change | P. value (log rank) |
|--------------------------|---|-------------|-----------------------|--------------------|---------------------|
| Wild type (N2) | <i>E. coli</i> (OP50) | 107 | 15.5±0.3 | | |
| Wild type (N2) | <i>B. subtilis</i> (PY79) | 118 | 25.2±0.4 | +62.6% | <0.0001 |
| Wild type (N2) | Worms developed from L1 to late L4 on <i>E. coli</i> (OP50), then switched to <i>B. subtilis</i> (PY79) | 111 | 19.3±2.4 | +24.5% | <0.0001 |

Table S2. Nutritional composition of *E. coli* vs. *B. subtilis*

| | <i>E. coli</i> (OP50) | <i>B. subtilis</i> (PY79) |
|-----------------|-----------------------|---------------------------|
| % Water content | 78.02 | 78.75 |
| Protein* | 84.72 | 77.04 |
| Fat* | 0.41 | 0.28 |
| Carbohydrate* | 4.51 | 9.79 |
| Ashes** | 10.37 | 12.89 |
| Calories** | 360.57 | 349.82 |

* Grams per 100 g of dry bacteria

** Calories per 100 g of dry bacteria

Table S3. Development of *clk-1* mutant L1 larvae after 7 days on the *B. subtilis* diet with *E. coli* extract supplementation

| <i>C. elegans</i> strain | Bacterial food strain | Extract supplemented* | Extract dilution | Outcome upon extract supplementation |
|--------------------------|---------------------------|-----------------------|------------------|--|
| <i>clk-1(qm30)</i> | <i>B. subtilis</i> (PY79) | <i>E. coli</i> (OP50) | Undiluted | L1s developed into adults and these produced progeny that arrested at L1-L2 stages |
| <i>clk-1(qm30)</i> | <i>B. subtilis</i> (PY79) | <i>E. coli</i> (OP50) | 1:2 | L1s developed into adults and some of these produced progeny that arrested at L1-L2 stages |
| <i>clk-1(qm30)</i> | <i>B. subtilis</i> (PY79) | <i>E. coli</i> (OP50) | 1:5 | L1s developed into adults and these laid dead embryos |
| <i>clk-1(qm30)</i> | <i>B. subtilis</i> (PY79) | <i>E. coli</i> (OP50) | 1:10 | Some L1s developed into adults |
| <i>clk-1(qm30)</i> | <i>B. subtilis</i> (PY79) | <i>E. coli</i> (OP50) | 1:20 | Most L1s developed into L4s |

* single dose of *E. coli* extract was used

Table S4. Supporting life span data for Figure 3

| <i>C. elegans</i> strain | Bacterial food strain | Sample size | Median life span ± SD | % Life span change | P. value (log rank) |
|--------------------------|---|-------------|-----------------------|--------------------|---------------------|
| Wild type (N2) | <i>E. coli</i> (OP50) | 236 | 16.4±1.1 | | |
| Wild type (N2) | <i>E. coli</i> (OP50) + <i>E. coli</i> (OP50) extract | 118 | 17.3±0.4 | +5.5% | >0.05 |
| Wild type (N2) | <i>E. coli</i> (OP50) + <i>E. coli</i> (OP50) extract (1:5 dilution) | 188 | 16.7±0.8 | +1.8% | >0.05 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) | 247 | 24.0±1.0 | +46.3% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) + <i>E. coli</i> (OP50) extract | 117 | 18.4±1.0 | +12.2% | <0.005 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) + <i>E. coli</i> (OP50) extract (1:5 dilution) | 187 | 20.6±1.2 | +25.6% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) + <i>E. coli</i> (OP50) extract (1:10 dilution) | 79 | 20.7±1.4 | +26.2% | <0.0001 |
| Wild type (N2) | <i>E. coli</i> (OP50) | 120 | 16.4±0.8 | | |
| Wild type (N2) | <i>E. coli</i> (OP50) + <i>E. coli</i> (GD1) extract | 84 | 16.7±0.3 | +1.8% | >0.05 |
| Wild type (N2) | <i>E. coli</i> (OP50) + <i>E. coli</i> (GD1) extract (1:5 dilution) | 108 | 16.4±0.4 | +0.2% | >0.05 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) | 92 | 24.1±0.9 | +47.0% | <0.0001 |

| | | | | | |
|----------------|--|-----|----------|--------|---------|
| Wild type (N2) | <i>B. subtilis</i> (PY79) + <i>E. coli</i> (GD1) extract | 80 | 22.4±0.5 | +36.6% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) + <i>E. coli</i> (GD1) extract (1:5 dilution) | 113 | 24.1±0.6 | +47.0% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) + <i>E. coli</i> (GD1) extract (1:10 dilution) | 122 | 24.1±1.0 | +47.0% | <0.0001 |

Table S5. Supporting life span data for Figure 5

| <i>C. elegans</i> strain | Bacterial food strain | Sample size | Median life span ± SD | % Life span change | P. value (log rank) |
|--------------------------|---------------------------------------|-------------|-----------------------|--------------------|---------------------|
| Wild type (N2) | <i>E. coli</i> (OP50) | 202 | 17.4±0.5 | | |
| Wild type (N2) | <i>E. coli</i> (OP50) + 10mM NAC | 195 | 17.5±1.0 | +0.6% | >0.05 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) | 235 | 24.5±0.6 | +40.8% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) + 10 mM NAC | 102 | 22.1±1.0 | +27.0% | <0.001 |

| <i>C. elegans</i> strain | Bacterial food strain | Sample size | Median life span ± SD | % Life span change | P. value (log rank) |
|--------------------------|---|-------------|-----------------------|--------------------|---------------------|
| Wild type (N2) | <i>E. coli</i> (OP50) | 150 | 17.2±0.6 | | |
| Wild type (N2) | <i>E. coli</i> (OP50) + 0.1 mM PQ | 146 | 25.1±1.5 | +45.9% | <0.0001 |
| Wild type (N2) | <i>E. coli</i> (OP50) + 0.1 mM PQ (after development) | 180 | 23.6±1.6 | +37.2% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) | 169 | 23.2±1.2 | +34.9% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) + 0.1 mM PQ | 151 | 27.2±2.0 | +58.1% | <0.0001 |
| Wild type (N2) | <i>B. subtilis</i> (PY79) + 0.1 mM PQ (after development) | 180 | 25.1±1.8 | +45.9% | <0.0001 |

Table S6. Supporting life span data for Figure 6

| <i>C. elegans</i> strain | Molecular pathway or process affected | Bacterial food strain | Sample size | Median life span ± SEM | Bacterial food strain | Sample size | Median life span ± SEM | % Life span change | P. value (log rank) |
|-------------------------------------|---|-----------------------|-------------|------------------------|---------------------------|-------------|------------------------|--------------------|---------------------|
| Wild type (N2) | | <i>E. coli</i> (OP50) | 512 | 16.0±0.3 | <i>B. subtilis</i> (PY79) | 465 | 23.2±0.5 | +45.1% | <0.0001 |
| <i>daf-2(e1370)</i> | Insulin-like pathway | <i>E. coli</i> (OP50) | 159 | 41.9±1.1 | <i>B. subtilis</i> (PY79) | 90 | 48.0±2.8 | +14.5% | <0.001 |
| <i>daf-16(mu86)</i> | Insulin-like pathway | <i>E. coli</i> (OP50) | 346 | 13.9±0.3 | <i>B. subtilis</i> (PY79) | 229 | 15.7±0.5 | +13.2% | <0.0005 |
| <i>daf-16(mgDf50)</i> | Insulin-like pathway | <i>E. coli</i> (OP50) | 117 | 11.7±0.1 | <i>B. subtilis</i> (PY79) | 115 | 14.9±0.2 | +27.4% | <0.0001 |
| <i>daf-2(e1370); daf-16(mgDf50)</i> | Insulin-like pathway | <i>E. coli</i> (OP50) | 107 | 11.2±0.1 | <i>B. subtilis</i> (PY79) | 114 | 12.3±0.4 | +9.9% | <0.001 |
| <i>glp-1(e2141)</i> | Germ Line Proliferation | <i>E. coli</i> (OP50) | 189 | 24.5±0.8 | <i>B. subtilis</i> (PY79) | 203 | 29.8±0.5 | +22.0% | <0.0001 |
| <i>isp-1(qm150)</i> | Electron transport chain | <i>E. coli</i> (OP50) | 102 | 24.6±0.8 | <i>B. subtilis</i> (PY79) | 174 | 32.0±2.4 | +30.2% | <0.0001 |
| <i>nuo-6(qm200)</i> | Electron transport chain | <i>E. coli</i> (OP50) | 213 | 31.9±0.5 | <i>B. subtilis</i> (PY79) | 312 | 38.7±1.8 | +21.3% | <0.0001 |
| <i>eat-2(ad1116)</i> | Dietary restriction | <i>E. coli</i> (OP50) | 355 | 24.0±0.6 | <i>B. subtilis</i> (PY79) | 408 | 28.2±0.9 | +17.3% | <0.0001 |
| <i>hif-1(ia4)</i> | Hypoxia-induced factor | <i>E. coli</i> (OP50) | 211 | 18.2±0.6 | <i>B. subtilis</i> (PY79) | 249 | 22.3±1.4 | +22.1% | <0.0001 |
| <i>hsf-1(sy441)</i> | Heat-shock transcription factor | <i>E. coli</i> (OP50) | 107 | 13.1±0.3 | <i>B. subtilis</i> (PY79) | 112 | 15.1±0.4 | +15.6% | <0.0005 |
| <i>jnk-1(gk7)</i> | Jun N-terminal Kinase | <i>E. coli</i> (OP50) | 118 | 15.0±0.3 | <i>B. subtilis</i> (PY79) | 102 | 21.6±0.6 | +44.5% | <0.0001 |
| <i>dbl-1(nk3)</i> | Transforming growth factor beta pathway | <i>E. coli</i> (OP50) | 93 | 14.7±0.4 | <i>B. subtilis</i> (PY79) | 94 | 21.0±0.8 | +42.5% | <0.0001 |
| <i>pmk-1(km25)</i> | Mitogen-activated protein kinase | <i>E. coli</i> (OP50) | 118 | 14.5±0.5 | <i>B. subtilis</i> (PY79) | 128 | 19.1±0.1 | +32.0% | <0.0001 |
| <i>tol-1(nr2033)</i> | Toll-like receptor | <i>E. coli</i> (OP50) | 227 | 14.9±0.3 | <i>B. subtilis</i> (PY79) | 222 | 29.7±1.2 | +98.9% | <0.0001 |

Table S7. Calculations for proportional life span differences of mutants with respect to controls

| * <i>C. elegans</i> strain | Median life span ± SD on <i>B. subtilis</i> diet | Number of life span assays (total number of worms) | **Projected median life span ± SD on <i>B.</i> <i>subtilis</i> diet | ***p. value (T test) indicating probability that median life span is not proportionally similar to median life span of N2 controls |
|-------------------------------------|--|---|--|---|
| Wild type (N2) | 23.2±1.7 | 13 (465) | | |
| <i>daf-16(mu86)</i> | 15.7±1.3 | 6 (229) | 18.0±1.5 | p<0.0001 |
| <i>daf-16(mgDf50)</i> | 14.9±0.3 | 3 (115) | 19.2±0.3 | p<0.005 |
| <i>daf-2(e1370); daf-16(mgDf50)</i> | 12.3±0.8 | 3 (114) | 17.0±1.0 | p<0.0001 |
| <i>glp-1(e2141)</i> | 29.8±1.1 | 4 (203) | 21.2±0.8 | p<0.05 |
| <i>eat-2(ad1116)</i> | 24.3±1.6 | 9 (408) | 20.0±1.3 | p<0.0005 |
| <i>hif-1(ia4)</i> | 22.3±2.2 | 6 (249) | 19.8±1.9 | p<0.005 |
| <i>hsf-1(sy441)</i> | 15.1±0.8 | 3 (112) | 18.0±0.9 | p<0.0001 |
| <i>pmk-1(km25)</i> | 19.1±0.2 | 3 (128) | 20.4±0.2 | p<0.05 |
| <i>tol-1(nr2033)</i> | 29.7±3.0 | 6 (222) | 30.8±3.1 | p<0.0001 |

* Strains used in Figure 6, which are not represented in this table, displayed median life spans that were proportionally similar to the median life span of N2 controls (p>0.05, T test)

** Life span data for *B. subtilis* fed mutant worms after normalizing with respect to median life span reference point obtained for N2 worms fed *E. coli* (16.0 days) and *B. subtilis* (23.2 days)

*** T test compares normalized median life spans of *B. subtilis* fed mutant worms with median life spans of *B. subtilis* fed N2 wild type worms