Different Mechanisms of Longevity in Long-Lived Mouse and *Caenorhabditis elegans* Mutants Revealed by Statistical Analysis of Mortality Rates

Bryan G. Hughes and Siegfried Hekimi[1]
Department of Biology, McGill University, Montreal, Quebec H3A 1B1, Canada

ABSTRACT Mouse and *Caenorhabditis elegans* mutants with altered life spans are being used to investigate the aging process and how genes determine life span. The survival of a population can be modeled by the Gompertz function, which comprises two parameters. One of these parameters ("*G*") describes the rate at which mortality accelerates with age and is often described as the "rate of aging." The other parameter ("*A*") may correspond to the organism's baseline vulnerability to deleterious effects of disease and the environment. We show that, in mice, life-span-extending mutations systematically fail to affect the age-dependent acceleration of mortality (*G*), but instead affect only baseline vulnerability (*A*). This remains true even when comparing strains maintained under identical environmental conditions. In contrast, life-span-extending mutations in *C. elegans* were associated with decreases in *G*. These observations on mortality rate kinetics suggest that the mechanisms of aging in mammals might fundamentally differ from those in nematodes.

KEYWORDS aging; *Caenorhabditis elegans*; Gompertz; longevity; mice

THE aging process can be studied by investigating genetic variants that alter life span in model organisms (Finch and Ruvkun 2001; Hekimi 2006). For example, the fact that mutations of genes involved in the insulin/insulin-like signaling pathway can extend life span in *Caenorhabditis elegans*, *Drosophila*, and mice is considered to imply a role for this pathway in the aging process (Kenyon 2010). Likewise, a role for mitochondrial function in aging is suggested by the finding that impairments to mitochondrial function can extend lifespan in *C. elegans* and mice (Ewbank *et al.* 1997; Feng *et al.* 2001; Dillin *et al.* 2002; Lee *et al.* 2003; Liu *et al.* 2005; Hughes and Hekimi 2011; Wang and Hekimi 2015).

Another point of view is provided by the study of mutations in a number of genes that induce segmental progeroid syndromes and shorten life span in mice. The short life span of these mutant mice is accompanied by the accelerated expression of some of the phenotypes commonly encountered in aging (Mounkes *et al.* 2003; Wong *et al.* 2003; Baker *et al.* 2004; Chang *et al.* 2004; Trifunovic *et al.* 2004). While these have often been presented as representing alterations to the aging process, it remains possible that their shorter life spans are caused by the induction of specific pathologies that only mimic aspects of the actual aging process (Harrison 1994; Miller 2004).

It has also been argued that an extension of life span may not necessarily be concrete evidence of a retardation of the aging process (Orr *et al.* 2003; De Magalhaes *et al.* 2005; Ladiges *et al.* 2009). In this view, a life-span-extending intervention may simply remedy deficiencies in the environment or in the genetic makeup of one particular strain. The intervention would therefore extend life span by correcting specific flaws rather than altering the aging process. These considerations create a conundrum: If life span is not a reliable measure of aging, how can we confirm that a particular manipulation truly affects the aging process? One approach is to assess physiological phenotypes that are known to deteriorate with age, such as cognition or the functioning of the cardiovascular or immune systems, to detect similarities or discrepancies with the patterns observed in control strains. An alternative criterion is to consider whether a particular manipulation changes how mortality rates increase with age (Sacher 1977; Finch *et al.* 1990; De Magalhaes *et al.* 2005; Yen and Mobbs 2010). This is based on the hypothesis that the increased incidence of the age-related pathological changes that characterize the aging process is reflected in changing mortality rates.

doi: 10.1534/genetics.116.192369
Manuscript received June 6, 2016; accepted for publication August 28, 2016; published Early Online September 15, 2016.
Supplemental material is available online at www.genetics.org/lookup/suppl/doi:10.1534/genetics.116.192369/-/DC1.
[1]Corresponding author: Department of Biology, McGill University, 1205 Dr. Penfield Ave., Montreal, QC H3A 1B1, Canada. E-mail: siegfried.hekimi@mcgill.ca

Human mortality rates increase exponentially with age, as first noted and quantified by Benjamin Gompertz in 1825 (described by Olshansky and Carnes 1997). This property has subsequently been observed for the mortality rates of model organisms including mice, *Drosophila*, and *C. elegans* (Johnson 1987; Gavrilov and Gavrilova 1991; De Magalhaes *et al.* 2005). The Gompertz model of mortality is commonly expressed by the following equation, where the mortality rate (R) can be represented at any age (t) for a given population by

$$R(t) = Ae^{Gt}.$$

"G" describes the rate at which mortality rates accelerate with age and "A" represents the initial mortality rate at time 0 (Finch 1990). "A" is strictly theoretical as a mortality rate, since there can be no actual mortality at time 0. Instead, it can be determined by extrapolation from mortality rates at greater ages and does not necessarily correspond to true mortality rates at birth or during youth. Figure 1 shows how changes to the Gompertz parameters affect the survival curve of a hypothetical population of mice with a median life span of 2 years (solid black line). Decreasing A (solid blue lines) extends life span by shifting the inflection point of the curve rightward, such that it occurs proportionally later in age, relative to maximum life span. There is no change in the apparent "slope" of the curve. In contrast, decreasing G (dashed blue lines) decreases the slope.

A has been described as measuring the vulnerability to disease unrelated to the onset of aging (Sacher 1977) or the effect of the environment on mortality (Finch *et al.* 1990). Changes to A will alter mortality rates evenly across the life span of the population. In contrast, since the parameter G can be considered a rate constant for the age-related increase of mortality of a sample or population, it is often given a preeminent role as an indicator of the "rate of aging" (Sacher 1977; Finch *et al.* 1990). This is a logical hypothesis, since an increased or decreased G would likely reflect the rate at which physiological conditions are declining with age. Therefore it is often assumed that interventions that extend life span by slowing aging, rather than by alleviating some age-independent pathology, will be associated with a decreased G; likewise, those that accelerate aging would be associated with an increased G (Takeda *et al.* 1981; Finch *et al.* 1990; Honda *et al.* 1993; Pletcher *et al.* 2000; De Magalhaes *et al.* 2005; Merry 2005; Yen *et al.* 2008; Tricoire and Rera 2015). It should be noted, however, that some have argued against this viewpoint and suggest that G should not be assigned a dominant role as a measure of aging (Driver 2001; Masoro 2006).

Although the Gompertz model seems to fit survival curves for many human and model organisms, it can be modified to provide a better fit. It can include additional terms that account for mortality rates that plateau at later ages (logistic model), deaths due to exogenous, non-aging-related environmental causes (Makeham model), or both (*i.e.*, logistic–Makeham) (Wilson 1994). Some caution is necessary, however, when interpreting survival curves that appear to be better fitted by more complex variants. For example, if the survival curve of a population is not optimally fitted by a particular model, then the addition of further parameters will naturally allow for more flexibility to adjust the model to the survival curve, without necessarily being informative about the underlying biology (Wilson 1994). Furthermore, a population of ≥100 is necessary to reliably determine which model best fits a particular distribution (Wilson 1994). Since most life span studies in mice use fewer animals than this, it is reasonable to use the simplest possible model in these cases, *i.e.*, the Gompertz model.

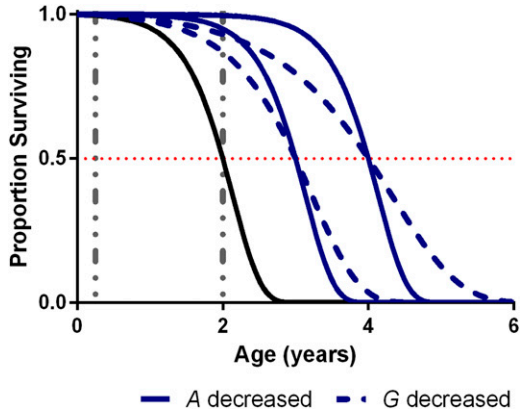


Figure 1 The effect of changing Gompertz parameters on the appearance of survival curves. A "baseline" survival curve with a median life span of 2 years is shown in black [G = 2.6, ln(A) = −4.6]. Increased life spans (median life span of 3 and 4 years) obtained by decreasing A [ln(A) of −7.2 and −9.8] while keeping G fixed are shown as solid blue lines. Increased life spans obtained by decreasing G (to 1.6 and 1.1) while keeping A fixed are shown as dashed blue lines. Vertical dashed lines show typical ages used for assessment of changes to age-dependent pathologies in mice.

Using this model, some studies observed a decreased G in cohorts of long-lived mice (Lapointe *et al.* 2009; Hughes and Hekimi 2011). However, a recent analysis of 29 published life span studies found this to be relatively rare in mice subjected to life-span-extending dietary or genetic manipulations (Yen *et al.* 2008). Instead, it was changes in A, not G, that were most frequently observed, with 4 of 12 long-lived strains having a statistically significant increase in A, but only 1 of 12 having a decreased G. Strikingly, the remainder did not exhibit any statistically significant changes in Gompertz parameters. Life-span-shortening interventions were somewhat more likely to be associated with decreases in G, with 6 of 15 short-lived strains associated with a statistically significantly decrease in G and 5 with a decrease in A. We theorized that actual changes in the parameters are being masked by the relatively small sample sizes that often characterize life span studies in mice and that looking at individual studies by this method will fail to reveal systemic relationships between changes to life span and Gompertz parameters.

Since a substantial number of studies reporting changes in mouse life span resulting from genetic manipulations have now been published, we hypothesized that a correlation-based approach may be a more powerful technique to search for patterns in Gompertz parameter shifts. For example, a negative correlation between life span and *G* across long-lived lines of mice would suggest that their extended longevity was due to a decreased rate of aging.

Dramatic changes in mortality rate trajectories have been reported across different species (Jones *et al.* 2014), demonstrating the importance of studying these phenomena in diverse model organisms. The nematode roundworm *C. elegans* has been used to identify many life-span-extending and -shortening mutations. The strength of invertebrate model systems such as *C. elegans* for aging research is their short life span and the possibility to repeatedly carry out experiments with very large cohorts, as well as the possibility to identify mutants with very large increases in life span. Early studies with this organism established that *C. elegans* mortality patterns apparently followed the Gompertz model (Johnson 1990; Honda and Matsuo 1992; Vanfleteren *et al.* 1998), with variations depending on growth conditions (Vanfleteren *et al.* 1998). Later studies investigated whether the environment and mutations could alter Gompertz parameters in this organism (Lenaerts *et al.* 2007; Wu *et al.* 2009; Yen and Mobbs 2010). We therefore elected to analyze short- and long-lived *C. elegans* as a comparator to our analysis of mice.

By the straightforward method of plotting Gompertz parameters against life span we found that most of the genetically driven variability in life span between normal- and long-lived groups of mice was due to changes in *A*, not *G*. In fact, *G* remained remarkably invariant for different groups of wild-type mice as well as for mice with genetic variations that extend life span. The only exceptions to this trend were some interventions that acutely shortened life span. We also found this to be true for a collection of inbred mouse strains studied under uniform conditions as part of the Mouse Phenome Database at The Jackson Laboratory (Yuan *et al.* 2009). Thus, with the exception of some severe life-span-shortening interventions, life span in laboratory mice is largely determined by factors that affect initial vulnerability, rather than age-dependent mortality rate acceleration. In contrast to mice, we found life span to be associated with changes in *G*, not *A*, among long-lived *C. elegans* mutants. This was true as a trend across long-lived mutants and was also observed by analyzing changes to Gompertz parameters among numerous replicate studies of the well-characterized *daf-2*, *isp-1*, and *eat-2* mutants.

Materials and Methods

Estimation of Gompertz parameters

To estimate Gompertz parameters, numerical survival data were extracted from published Kaplan–Meier survival curves, using Engauge Digitizer 6.2 (Mark Mitchell, http://markummitchell.github.io/engauge-digitizer/). Survival data were divided into 10- or 1-day intervals for mice and *C. elegans*, respectively, and Gompertz parameters were determined by maximum-likelihood estimation (MLE) using WinModest 1.0.2 (Pletcher 1999), according to the Gompertz or logistics hazard functions (Pletcher *et al.* 2000):

$$\text{Gompertz:}\quad R(t) = Ae^{Gt}$$

$$\text{Logistic:}\quad R(t) = \left(Ae^{Gt}\right)/\left[1 + L(A/G)\left(e^{Gt} - 1\right)\right].$$

All estimates had associated inform values of "0," indicating that the maximum-likelihood procedure was able to successfully resolve parameters within the given range and that asymptotic confidence intervals could be calculated. For all groups, the accuracy of the estimation was graphically confirmed by overlaying the resulting Gompertz survival function on the raw survival data, using Prism 6 (GraphPad Software).

For *C. elegans*, initial analysis using the logistic model yielded a clear bimodal distribution of values across strains for the logistic parameter "*L*", with clusters of value >0.01 and $<1 \times 10^5$. Parameters in this lower group were more difficult to determine by MLE, with many appearing to resolve to arbitrarily small values without resulting in further changes to *A* or *G*. We therefore set these values to 0.

Model comparison

We used WinModest's likelihood theory-based tools to determine which model (among Gompertz, logistic, Makeham, and logistic–Makeham) provided the best fit for *C. elegans* survival curves. This methodology takes into account the fact that models with additional parameters will naturally be less constrained when attempting to fit to a data set and therefore identifies the mortality model with the fewest parameters that fits the data sufficiently. This method is therefore superior to simple comparison of correlation coefficients obtained by regression-based methods.

General statistics

Nonparametric Spearman correlation was used to quantify the relationship between life span and Gompertz parameters. Prism was used to determine Spearman correlation coefficients and perform significance tests. For all tests a *P*-value <0.05 was taken to indicate statistical significance.

For pairwise comparisons between values of mutant and control parameters, differences (mutant minus control) were used rather than ratios because of the presence of negative or extremely low values. The nonparametric Wilcoxon signed-rank test was used to determine whether the average effect size was significantly different from 0. As an additional method of investigating the role of Gompertz parameters in alterations to life span, we used WinModest's longevity decomposition tool to determine the extent to which the

Table 1 Maximum-likelihood estimations of the Gompertz parameter values determined from published survival curves (set 2)

			Control				Mutant				
Gene and allele or strain	Sex	Background[a]	Median life span (*D*)	*n*	ln(*A*)	*G*	Median life span (*D*)	*n*	ln(*A*)	*G*	Source
AC5$^{-/-}$	M+F	129/SvJ-C57BL/6	755	25	−6.74	3.46	990	13	−11.73	4.72	Yan *et al.* (2007)
cIGF-1^{tg}	M	FVB/N	705	39	−3.54	1.73	880	38	−4.05	1.72	Li and Ren (2007)
IRS1$^{-/-}$	F	C57BL/6	750	21	−5.11	2.73	970	14	−6.85	2.83	Selman *et al.* (2008)
	M		785	35	−6.84	3.50	860	13	−20.25	9.28	
Klotho^{+46}	M	C3H	785	29	−4.09	2.18	890	22	−5.61	2.53	Kurosu *et al.* (2005)
Klotho^{+48}	M						1000	22	−6.24	2.55	
Klotho^{+46}	F		735	25	−4.30	2.41	840	28	−6.54	3.13	
Klotho^{+48}	F						825	29	−5.91	2.83	
MT^{tg}	M	FVB	850	55	−4.78	2.28	980	55	−4.26	1.58	Yang *et al.* (2006)
FGF21^{tg}	M	C57Bl/6J	840	32	−4.56	1.94	1100	37	−7.24	2.57	Zhang *et al.* (2012)
S6K1$^{-/-}$	F	C57BL/6	820	23	−4.43	2.15	975	29	−6.07	2.45	Selman *et al.* (2009)
Hcrt-UCP2	F	C57BL/6	550	31[b]	−2.91	2.07	660	26[b]	−4.06	2.42	Conti *et al.* (2006)
	M		720	36[b]	−3.24	1.75	815	53[b]	−4.85	2.37	
MIF$^{-/-}$	F	C57BL/6J-129/SvJ	740	24	−4.60	2.51	900	39	−5.01	2.17	Harper *et al.* (2010)
IRS1$^{-/-}$	F	C57BL/6	780	16	−6.57	3.41	870	15	−4.18	1.86	Selman *et al.* (2011)
	M		770	37	−6.27	3.22	890	12	−5.88	2.58	
GHRH$^{-/-}$	M	C57BL6-129SV	610	56	−2.24	1.38	920	39	−6.18	2.67	Sun *et al.* (2013)
	F		660	52	−3.55	2.16	960	58	−4.67	1.90	
IGF-1R$^{+/-}$	F	C57BL/6J	800	38	−5.35	2.62	870	34	−6.45	2.84	Xu *et al.* (2014)
bIGF-1$^{+/-}$	M	129/Sv×C57BL/6	830	20	−5.15	2.32	975	9	−13.98	5.70	Kappeler *et al.* (2008)
	F		850	22	−5.54	2.63	880	18	−6.70	2.99	
Pten^{tg}	M	C57BL6-CBA (75%:25%)	780	49	−4.77	2.56	880	32	−5.35	2.40	Ortega-Molina *et al.* (2012)
	F		790	63	−6.17	3.11	910	32	−7.82	3.45	
Akt1$^{+/-}$	F	C57BL/6	780	79	−5.40	2.85	870	80	−6.42	2.99	Nojima *et al.* (2013)
	M		840	101	−5.84	2.77	895	103	−5.17	2.29	
mTOR$^{\Delta/\Delta}$	M	129S1-C57BL/6Ncr	680	10	−5.44	3.19	830	17	−5.39	2.57	Wu *et al.* (2013)
	F		800	24	−3.59	1.74	960	26	−6.95	2.89	
bSirt1^{tg}	F	C57BL/6J	795	43	−5.81	2.91	930	34	−7.40	3.25	Satoh *et al.* (2013)
	M		855	47	−5.73	2.71	925	33	−8.35	3.53	
Sirt6$^{tg\text{-}55}$	M	C57BL/6J-BALB/c	865	35	−6.15	2.86	985	23	−6.41	2.54	Kanfi *et al.* (2012)
Sirt6$^{tg\text{-}108}$	M		730	36	−4.08	2.18	790	25	−4.86	2.23	
Gpx4$^{+/-}$	M	C57BL/6	960	50	−6.33	2.68	1030	50	−8.81	3.57	Ran *et al.* (2007)
mGsta4$^{-/-}$	F	C57BL/6	740	50	−4.12	2.18	840	50	−6.36	3.06	Singh *et al.* (2010)
hMTH1^{tg}	M+F	C57BL/6	790	42	−7.89	4.02	910	34	−6.78	2.94	De Luca *et al.* (2013)
TRX^{tgc}	M	C57BL/6	890	60	−4.15	1.73	950	60	−5.20	2.09	Pérez *et al.* (2011)
TgTert^{tgd}	?	C57BL6-DBA/2	690	68	−3.91	2.24	1010	27	−7.83	3.14	Tomás-Loba *et al.* (2008)
UCP1^{tg}	M+F	C57BL/6	820	53	−4.85	2.29	940	51	−6.30	2.76	Gates *et al.* (2007)
Dgat$^{-/-}$	F	C57BL/6J	750	30	−4.34	2.30	940	30	−7.38	3.20	Streeper *et al.* (2012)
PKA RIIβ$^{-/-}$	M	C57BL/6	900	20	−9.53	4.33	970	20	−6.68	2.79	Enns *et al.* (2009)
IκB-α^{DN}	M	C57BL/6	880	23	−11.26	5.28	965	31	−9.31	3.92	Zhang *et al.* (2013)
BubR1^{tg}	M+F	C57BL/6-SV129	630	60	−3.12	2.00	730	57	−3.36	1.70	Baker *et al.* (2013)
AT1A$^{-/-}$	M	C57BL/6×129/SvEv	760	20[b]	−9.98	5.36	940	20	−12.57	5.30	Benigni *et al.* (2009)
ETA$^{-/-}$	M	C57BL/6J	730	34	−3.82	1.74	920	28	−3.90	1.49	Ceylan-Isik *et al.* (2013)
AgRP$^{-/-}$	M+F	C57BL/6J-129Sv	650	16	−5.03	3.45	710	21	−5.66	3.36	Redmann and Argyropoulos (2006)
Arf/p53^{tg}	?	C57BL/6J	840	111	−5.12	2.47	950	25	−8.16	3.51	Matheu *et al.* (2007)
Mclk1$^{+/-}$	M+F	129/Svj-BALB/c	764	14	−8.87	4.61	900	54	−5.2	2.35	Lapointe *et al.* (2009)
PAPP-A$^{-/-}$	F	C57BL/6-129Sv/E	670	50	−3.48	2.00	880	38	−4.77	2.08	Conover *et al.* (2010)
	M		680	45	−2.71	1.45	830	40	−3.98	1.75	

M, male; F, female.
[a] Dash denotes a mix of the indicated backgrounds; "×" denotes F_1 cross.
[b] Sample size approximated from survival curve.
[c] Cohort 2, male.
[d] Sp53/Sp16/SArf/TgTert^{tg} *vs.* Sp53.

differences in Gompertz parameters between control and mutant groups contributed to changes in life span (Pletcher *et al.* 2000).

VBA code used to generate the simulated data described in supplemental figures is provided in File S2, as a macro-enabled workbook for Microsoft Excel 2010.

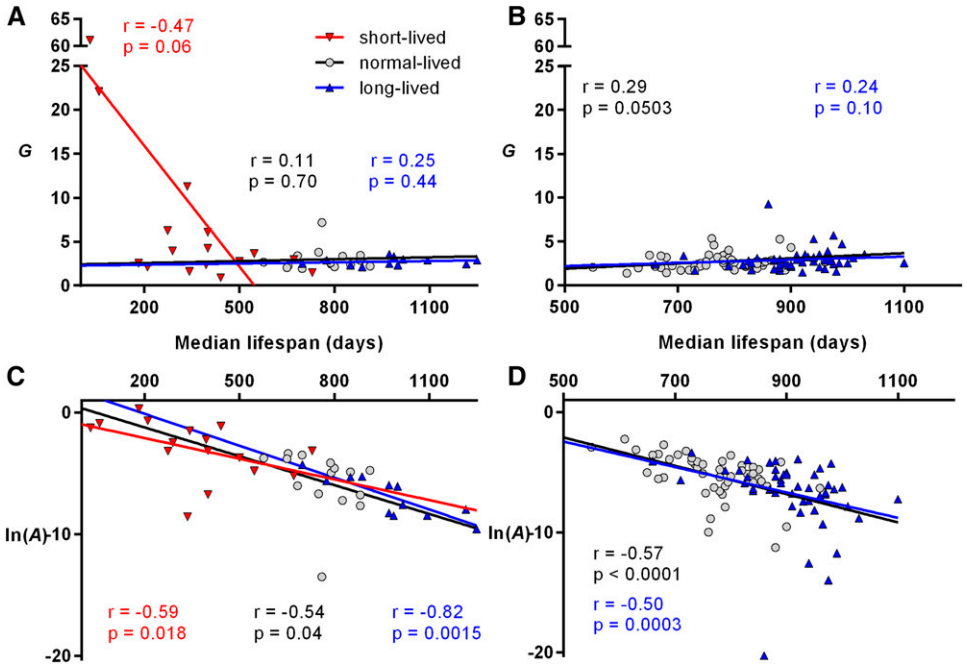


Figure 2 MLE estimations of the Gompertz parameter values plotted against median life span for lines of mice with varying life spans (the symbols are as presented in the key in A). Lines of best fit, Spearman correlation coefficients (*r*), and associated *P*-values are shown in colors corresponding to the data points. Parameters determined previously (set 1) are shown in A and C; parameters determined as part of the current study (set 2) are shown in B and D. Note that the line of best fit (determined by linear regression) is shown as an aid for the reader, and the *P*-values shown were determined separately by nonparametric methods.

Data availability

The authors state that all data necessary for confirming the conclusions presented in the article are represented fully within the article or Supplemental Material (File S1).

Results

Life span of normal or genetically modified long-lived mice is largely determined by changes to A, not G

To calculate Gompertz parameters, one requires numerical survival data beyond what is typically provided in published reports. Gompertz parameters for a number of short- and long-lived strains have previously been determined (Yen *et al.* 2008), and we included those in our analysis (set 1, summarized in Table S1; note that we did not include calorically restricted groups in our analysis). We divided the mice into groups based on their life spans. Short-lived mice were defined as those subjected to genetic manipulations that shortened life span relative to that of controls. We also included in this group lines of mice known to have average life spans markedly shorter than those of conventional lines of laboratory mice, namely NZB/W mice, which are known to suffer from severe autoimmune disease (Partridge *et al.* 2005), and senescence-accelerated mice (SAM) as well as their "senescence-resistant" (SRM) controls (Avraam *et al.* 2013). Normal-lived mice were defined as those of the control strains, with life spans typical of laboratory mouse strains. Median "normal" life spans varied considerably (from 550 to 960 days), presumably dependent upon genetic background or husbandry. Lines of mice were defined as long lived if their life span was extended relative to that of normal-lived controls.

This compilation was published in 2008, and the increasing ease of mouse genetic manipulations and continued interest in understanding the causes of aging have meant that the number of long-lived strains has continued to grow. We therefore obtained numerical survival data from the published survival curves of an additional 32 separate studies comparing 31 separate long-lived mutants (set 2, summarized in Table 1). We did not search for additional short-lived strains of mice.

The two sets together therefore encompass a diverse collection of genetic manipulations that have been shown to extend life span and likely comprise a majority of published reports of long-lived strains of mice (Yuan *et al.* 2009; Selman and Withers 2011; Liao and Kennedy 2014). The affected genes include those playing a role in oxidative stress response, signaling (mTOR, insulin/insulin-like, or growth hormone), metabolism, genomic integrity, mitochondrial function, and cellular proliferation. One strain of mice, *Atg5* transgenics (Pyo *et al.* 2013), was excluded from further analysis because the parameter estimates [*G* of 8–14 and a ln(*A*) of −14 to −25] were so markedly different from those of the remainder of the strains (Table 1).

To determine whether either Gompertz parameter changed systematically with changes in longevity, we examined the relationship between median life spans and Gompertz parameters for each cohort. In short-lived mice the correlation of *G* with life span approached statistical significance (Figure 2A: $r = -0.47$, $P = 0.06$). However, this was largely due to the two shortest-lived lines (*Klotho* and *Lmna* mutants), with median life spans <2 months (if they were excluded, $r = -0.22$, $P = 0.46$). These two lines also had by far the largest *G* (22 and 61, respectively, compared to a median value of 3.3 for short-lived mice). There was no correlation between life span and *G* for normal- or long-lived

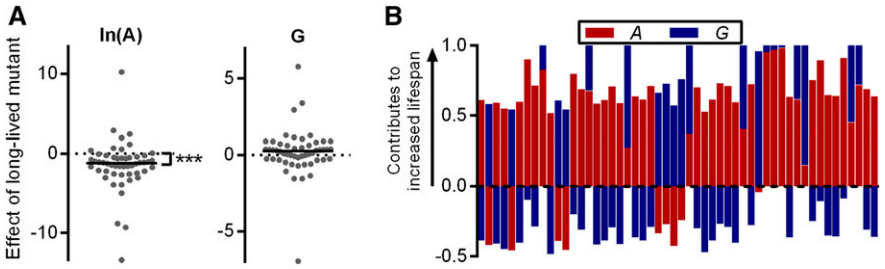


Figure 3 Pairwise comparison within studies of long-lived mouse strains relative to normal-lived controls (set 2). (A) Difference in Gompertz parameter values between long-lived and control groups within studies (males and females analyzed separately). A positive value corresponds to an increase in parameter value for long-lived mice and a negative value to a decrease. (B) Fractional contribution of each parameter change to the extended life span of long-lived groups of mice. Each bar is 1 unit long. A positive value indicates that the change in parameter value contributes to the increased life span. Negative values indicate that the parameter change acts to shorten life span. *** $P < 0.0001$ *vs.* 0 by Wilcoxon signed-rank test.

mice in set 1 (Figure 2A: $r = 0.11$, $P = 0.70$ and $r = 0.25$, $P = 0.44$, respectively). For set 2 (Figure 2B: $r = 0.29$, $P = 0.0503$ and $r = 0.24$, $P = 0.10$ for normal- and long-lived groups, respectively), we saw no negative correlation and a borderline statistically significant positive correlation (underscoring that the changes in G in this set are not responsible for the increased life spans).

In contrast to G, there was a clear negative correlation between life span and ln(A). This was apparent for short-, normal-, and long-lived lines in set 1 (Figure 2C: $r = -0.59$, $P = 0.018$; $r = -0.54$, $P = 0.04$; and $r = -0.82$, $P = 0.0015$, respectively) and for normal- and long-lived lines in set 2 (Figure 2D: $r = -0.57$, $P < 0.0001$ and $r = -0.50$, $P = 0.0003$, respectively).

Combining sets 1 and 2 together yields similar results to those when they are analyzed separately, with no statistically significant effect on G (Figure S1A) and a negative correlation between ln(A) and lifespan ($r = -0.65$, $P < 0.0001$) (Figure S1B). Likewise, analyzing males and females separately (Figure S2) does not affect the Gompertz parameter–life span relationship, with a negative correlation between ln(A) and life span ($r = -0.69$ and -0.55 for females and males, respectively, $P < 0.0001$). The same relationship was also apparent when restricting the analysis to those studies carried out in the most commonly used background strain, C57BL/6, where there was a weak positive correlation between G and life span ($r = 0.29$, $P = 0.04$) and a strong negative correlation between ln(A) and life span ($r = -0.60$, $P < 0.0001$) (Figure S3).

Estimation of Gompertz parameters can be subject to biases resulting in systematic under- or overestimations (Promislow *et al.* 1999), especially for smaller samples sizes, potentially introducing statistical artifacts into our analysis. Correction for potential systematic biases in parameter estimations (determined using standard resampling techniques), as well as removal of possible outliers, did not change the effects described above (see Figure S4 and its legend).

Genetic differences underlie the relationship between A and life span in a panel of inbred mice

The results shown in Figure 2 imply that the biological mechanisms that determine life span in laboratory mice are largely those associated with changes to A, rather than G. As described above, A has been interpreted as representing the effect of age-independent factors on life span. It is therefore possible that the systematic decrease in A with increased life span that is apparent in Figure 2 could be due to environmental differences between studies, with more beneficial environments resulting in decreased aging-independent mortality rates and, consequently, longer life spans. Indeed, differences in the quality of husbandry have been raised as a potential confounding variable in aging studies (Liang *et al.* 2003; Ladiges *et al.* 2009).

To address this question, we conducted a pairwise comparison, within studies, of Gompertz parameters for normal- and long-lived lines in set 2 of mice. Among 52 long-lived lines (males and females analyzed separately), A was decreased relative to normal-lived controls in 43 lines. Within-study ln(A) values decreased by an average of -1.504 units in the long-lived group ($P < 0.0001$ *vs.* 0 by Wilcoxon signed-rank test). In contrast, G was increased by an average of 0.26 units ($P = 0.08$ *vs.* 0) (Figure 3A).

The above analysis considers A and G separately, when, in reality, changes in both parameters cooperate to establish a new survival trajectory when the survival curve of a population is shifted. It is possible to determine the contribution of each parameter to changes in average longevity (longevity decomposition), revealing the extent to which each parameter is responsible for the shift in life span (Pletcher *et al.* 2000). Thus, among the long-lived strains of set 2, changes to A account for the majority of the life span increase in 38 of 52 long-lived strains (Figure 3B; $P = 0.0009$ *vs.* expected by chance, by chi-square test).

To further differentiate between environmental and genetic effects we determined the relationship between the Gompertz parameters and life span among a group of 31 inbred strains of mice of various average life spans that were maintained under uniform conditions. Complete survival data were obtained from the Mouse Phenome Database, maintained by The Jackson Laboratory (Bogue *et al.* 2016). The 31 inbred strains used in this study were selected to encompass the greatest possible genetic diversity (Yuan *et al.* 2009, 2012) and included wild-derived strains as well as representatives from the seven genetically related groups that comprise laboratory mice (Yuan *et al.* 2009).

In this data set (Figure 4, Table S2), there was no correlation between median life span and G ($r = 0.16$, $P = 0.23$), but life span was correlated with ln(A) ($r = -0.58$, $P < 0.0001$).

These relationships were unchanged when we corrected for systematic bias in parameter estimations, as well as upon removal of possible outliers (Figure S5).

As an additional test, we determined the relationship between the Gompertz parameters and life span among a group of 44 recombinant inbred strains of mice of various average life spans that were maintained under uniform conditions (males and females analyzed separately) (Liao *et al.* 2010). Although the small sample size of these groups ($n = 5$) limits the reliability of Gompertz parameter estimation, we still observed the same pattern of relationships between Gompertz parameters and life span, with no correlation between G and life span ($r = 0.14$, $P = 0.2$) and a negative correlation between ln(A) and life span ($r = -0.38$, $P = 0.0006$) (Figure S6, Table S3).

Life span in C. elegans mutants is associated with changes to G, not A

We wondered whether other classic model organisms commonly used for aging research would demonstrate a similar invariance for G with increased life span. The nematode *C. elegans* is one of the most widely used model organisms for the study of aging (Antebi 2007; Li and Ren 2007; Van Raamsdonk and Hekimi 2010). We calculated Gompertz parameters from published survival curves of 39 long-lived and 8 short-lived mutants, along with the 20 associated wild-type (N2) controls (Table 2). These mutants were chosen to affect a diverse collection of biological pathways including, but not limited to, mitochondrial function, insulin/insulin-like signaling, nutrient uptake, stress resistance, sensory perception, and autophagy. All life span experiments were carried out at 20° on agar plates and had an associated N2 control. Because of the large number of long-lived worm mutants that have been identified, this represents only a limited subset of potentially usable strains. For comparison, the GenAge database of aging-related genes (Tacutu *et al.* 2013) currently lists 112 long-lived *C. elegans* mutants (although survival experiments for these mutants were not all conducted in a manner that would have satisfied our inclusion criteria).

Initial observations showed that the standard Gompertz model poorly fitted the survival of many groups of worms. In line with this, an automated life span analysis performed on populations of *C. elegans* with very large sample sizes (>200 animals) has recently shown that the exponential increase in mortality rates that characterize the Gompertz survival model largely ceases at later ages (Stroustrup *et al.* 2016). Thus, logistic models that account for late-life mortality rate deacceleration provide a better fit for *C. elegans* survival data. Indeed, of the 11 groups of N2 worms with $n > 100$ [at which sample size the actual population model can be reliably determined from the sample (Wilson 1994)] all but 1 were best described by the logistic model (see *Materials and Methods* for details of the model comparison method). Of the 18 mutant strains with $n > 100$, 16 were best fitted by the logistic model. We therefore used this model to analyze all *C. elegans* survival curves.

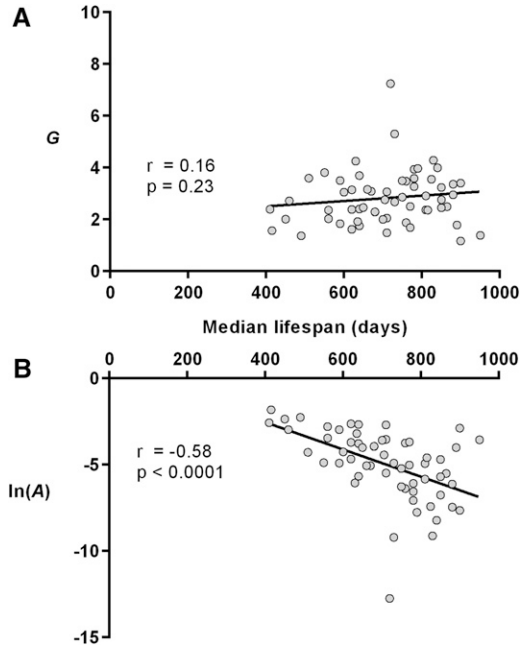


Figure 4 Maximum-likelihood estimations of the Gompertz parameter values plotted against median life span for strains maintained as part of the mouse phenome project at The Jackson Laboratories (see Table S2 for numeric values and descriptions of lines). Lines of best fit, Spearman correlation coefficients (r), and associated P-values are shown. (A) Gompertz parameter G, acceleration of mortality rates with age. (B) Natural logarithm of A, baseline mortality. Note that the line of best fit (determined by linear regression) is shown as an aid for the reader and that the P-values shown were determined separately by nonparametric methods. n = 13–32, average = 30.1.

For *C. elegans*, median life span for short-lived strains exhibited a trend toward a negative correlation with G ($r = -0.55$, $P = 0.16$), and there was a statistically significant negative correlation between G and lifespan for control N2 or long-lived worms ($r = -0.53$, $P = 0.02$ and $r = -0.70$, $P < 0.0001$, respectively) (Figure 5A). In contrast to what we repeatedly observed in mice, there was no statistically significant relationship between ln(A) and life span (Figure 5B). There was an inverse relationship between the logistic parameter L and median life span for N2 worms ($r = -0.54$, $P = 0.01$), but this relationship did not reach statistical significance for short- or long-lived strains (Figure 5C). These relationships were unchanged when we corrected for systematic bias in parameter estimations, as well as upon removal of possible outliers (Figure S7). Thus, changes to the rate-of-aging parameter G seem to account for the bulk of the life span increase in *C. elegans*.

Within studies, pairwise comparisons between mutant and N2 control worms reveal that G is increased and that changes to G make the greatest contribution to the increase in average life span, for 6 of 7 short-lived strains (Figure 6, A and B). Among long-lived mutant strains, ln(A) values were actually increased by an average of 1.24 ($P = 0.0044$ *vs.* 0 by Wilcoxon signed-rank test) and the value of the L parameter changed by an average of −1.38 ($P = 0.0022$ *vs.* 0). These

Table 2 Maximum-likelihood estimates of the logistic model mortality parameters for short- and long-lived *C. elegans* mutant and N2 control strains

		N2					Mutant						
Gene(s)	Allele(s)	Median life span (*D*)	*n*	ln(*A*)	*G*	*L*	Median life span (*D*)	*n*	ln(*A*)	*G*	*L*	Effect on life span	Source
hsf-1	*sy441*	20	42	−8.00	0.342	0.282	12	42	−7.98	0.633	1.441	0.600	Hajdu-Cronin *et al.* (2004)
gas-1	*fc21*	19	100	−8.61	0.426	1.358	12	100	−7.87	0.668	2.442	0.632	Suthammarak *et al.* (2013)
skn-1	*zu129*	15	75	−6.49	0.371	1.535	10	75	−24.72	3.093	11.184	0.667	An and Blackwell (2003)
mev-1	*kn1*	19	100	−8.61	0.426	1.358	13	100	−5.35	0.291	0.226	0.684	Suthammarak *et al.* (2013)
jnk-1	*gk7*	16	386	−8.90	0.537	1.997	12	333	−14.39	1.270	4.723	0.750	Ezekowitz (2014)
daf-16	*m26*	20	19	−7.54	0.345	1.186	15	37	−14.57	0.995	3.603	0.750	Kenyon *et al.* (1993)
jkk-1	*km2*	16	386	−8.90	0.537	1.997	13	189	−13.51	1.063	2.651	0.813	Ezekowitz (2014)
sir-2.1	*ok434*	19	70	−5.63	0.188	0.000	16	94	−7.28	0.358	0.014	0.842	Berdichevsky *et al.* (2006)
clk-2	*qm37*	18	50[a]	−14.95	0.839	2.666	18	50[a]	−14.93	0.945	6.160	1.000	Van Raamsdonk *et al.* (2010)
ctbp-1	*ok498*	19	254	−7.35	0.325	0.659	22	236	−8.52	0.330	0.492	1.158	Chen *et al.* (2009)
cep-1	*gk138*	17	115	−9.33	0.493	0.856	20	116	−10.08	0.496	2.446	1.176	Tavernarakis *et al.* (2008)
age-1	*hx546*	34	50	−7.18	0.142	0.000	42	50	−6.94	0.110	0.000	1.235	Yanase *et al.* (2002)
clk-5	*qm152*	18	50[a]	−14.95	0.839	2.666	24	50[a]	−14.83	0.585	2.479	1.333	Van Raamsdonk *et al.* (2010)
clk-8	*qm162*	18	50[a]	−14.95	0.839	2.666	24	50[a]	−8.57	0.325	1.383	1.333	Van Raamsdonk *et al.* (2010)
daf-19	*m86*	18	347	−8.45	0.475	2.471	24	35	−6.96	0.203	0.258	1.333	Apfeld and Kenyon (1999)
mec-8	*e398*	18	347	−8.45	0.475	2.471	24	46	−8.08	0.219	0.996	1.333	Apfeld and Kenyon (1999)
egl-30	*ad806*	18	50	−19.95	1.502	9.977	25	62	−6.12	0.146	0.000	1.389	Ch'ng *et al.* (2008)
arr-1	*ok401*	12	180	−5.38	0.331	0.457	17	210	−4.83	0.176	0.075	1.417	Palmitessa and Benovic (2010)
cdc-48.1; *atx-3*	*tm544*; *gk193*	19	312	−7.24	0.317	0.704	27	222	−6.95	0.184	0.382	1.421	Kuhlbrodt *et al.* (2011)
clk-3	*qm38*	18	50[a]	−14.95	0.839	2.666	26	50[a]	−10.41	0.341	0.884	1.444	Van Raamsdonk *et al.* (2010)
clk-1	*qm30*	18	309	−8.91	0.451	0.982	26	112	−21.52	0.903	7.037	1.444	Van Raamsdonk and Hekimi (2009)
che-13	*e1805*	18	347	−8.45	0.475	2.471	26	36	−5.89	0.142	0.206	1.444	Apfeld and Kenyon (1999)
che-2	*e1033*	18	347	−8.45	0.475	2.471	26	33	−25.00	1.560	19.645	1.444	Apfeld and Kenyon (1999)
daf-6	*e1377*	18	347	−8.45	0.475	2.471	26	75	−5.83	0.138	0.193	1.444	Apfeld and Kenyon (1999)
osm-1	*p808*	18	347	−8.45	0.475	2.471	26	17	−7.21	0.195	0.000	1.444	Apfeld and Kenyon (1999)
daf-4	*e1364*	15	60	−13.21	0.851	1.699	22	71	−11.14	0.479	1.927	1.467	Shaw *et al.* (2007)
nuo-6	*qm200*	22	150	−12.51	0.535	0.818	33	120	−10.38	0.288	1.743	1.500	Yang and Hekimi (2010)
clk-10	*qm169*	18	50[a]	−14.95	0.839	2.666	28	50[a]	−6.66	0.154	0.292	1.556	Van Raamsdonk *et al.* (2010)
clk-6	*qm158*	18	50[a]	−14.95	0.839	2.666	28	50[a]	−11.78	0.398	2.622	1.556	Van Raamsdonk *et al.* (2010)
clk-9	*qm164*	18	50[a]	−14.95	0.839	2.666	28	50[a]	−10.79	0.354	1.282	1.556	Van Raamsdonk *et al.* (2010)
sod-2	*ok1030*	18	309	−8.91	0.451	0.982	28	373	−8.44	0.255	0.805	1.556	Van Raamsdonk and Hekimi (2009)
che-11	*e1810*	18	347	−8.45	0.475	2.471	28	27	−6.02	0.126	0.000	1.556	Apfeld and Kenyon (1999)
daf-10	*e1387*	18	347	−8.45	0.475	2.471	29	30	−8.89	0.252	0.877	1.611	Apfeld and Kenyon (1999)
osm-6	*p811*	18	347	−8.45	0.475	2.471	30	30	−8.53	0.220	0.000	1.667	Apfeld and Kenyon (1999)
clk-7	*qm159*	18	50[a]	−14.95	0.839	2.666	32	50[a]	−8.03	0.212	1.109	1.778	Van Raamsdonk *et al.* (2010)
osm-3	*p802*	18	347	−8.45	0.475	2.471	32	133	−7.86	0.195	0.556	1.778	Apfeld and Kenyon (1999)
egl-3	*nr2090*	18	46	−11.13	0.704	4.043	32	63	−7.27	0.163	0.452	1.778	Ch'ng *et al.* (2008)
clk-4	*qm151*	18	50[a]	−14.95	0.839	2.666	34	50[a]	−5.99	0.095	0.033	1.889	Van Raamsdonk *et al.* (2010)
eat-2	*ad1116*	18	309	−8.91	0.451	0.982	35	156	−13.18	0.366	2.838	1.944	Van Raamsdonk and Hekimi (2009)

(continued)

Table 2, *continued*

Gene(s)	Allele(s)	N2					Mutant					Effect on life span	Source
		Median life span (*D*)	*n*	ln(*A*)	*G*	*L*	Median life span (*D*)	*n*	ln(*A*)	*G*	*L*		
tax-4	*p678*	18	347	−8.45	0.475	2.471	37	105	−8.90	0.186	0.299	2.056	Apfeld and Kenyon (1999)
che-3	*p801*	18	347	−8.45	0.475	2.471	40	31	−9.17	0.192	0.509	2.222	Apfeld and Kenyon (1999)
isp-1	*qm150*	18	309	−8.91	0.451	0.982	42	114	−6.66	0.097	0.487	2.333	Van Raamsdonk and Hekimi (2009)
clk-1; sod-2	*ok1030*	18	309	−8.91	0.451	0.982	44	147	−9.36	0.169	0.687	2.444	Van Raamsdonk and Hekimi (2009)
osm-5	*p813*	18	347	−8.45	0.475	2.471	44	46	−6.74	0.092	0.000	2.444	Apfeld and Kenyon (1999)
daf-2; clk-9	*e1370; qm164*	21	200[b]	−7.72	0.303	0.265	59	200[b]	−8.43	0.130	0.429	2.810	Van Raamsdonk *et al.* (2010)
daf-2; clk-2	*e1370; qm37*	15	200[b]	−10.90	0.720	2.747	48	200[b]	−6.08	0.055	0.313	3.200	Van Raamsdonk *et al.* (2010)
daf-2	*e1370*	18	309	−8.91	0.451	0.982	63	168	−7.38	0.074	0.000	3.500	Van Raamsdonk and Hekimi (2009)

Note that some mutant strains were compared to the same N2.
[a] Twenty worms per plate were examined with three trials per strain.
[b] Sum of at least three independent trials and an initial number of 80 worms per strain per trial.

Figure 5 Logistic parameter values for short-lived, long-lived, and wild-type control (N2) lines of *C. elegans*. Lines of best fit, Spearman correlation coefficients, *r*, and associated *P*-values are shown. (A) MLE estimations of the Gompertz parameter *G*, describing the age-related acceleration of mortality rates. (B) Natural logarithms of the MLE estimations of the Gompertz parameter *A*, the baseline mortality rate. (C) MLE estimations for the late-stage mortality rate deacceleration parameter *L*. Note that the line of best fit (determined by linear regression) is shown as an aid for the reader, and the *P*-values shown were determined separately by nonparametric methods.

effects (which by themselves would decrease life span) were offset by a decrease in *G* (−0.28, $P < 0.0001$ *vs.* 0) (Figure 6C). Accordingly, changes to *G* made the greatest contribution to the increased average life span in 33 of 39 long-lived strains (Figure 6D).

Due to the inherent variability in any one study, it would be difficult to reliably conclude whether a particular long-lived strain has a certain characteristic effect on the Gompertz parameters. For example, unique environmental or methodological issues could subtly influence the pattern of mortality in a particular study. We therefore analyzed survival results from multiple studies for three long-lived mutants that have been widely studied. We chose *daf-2(e1370)* as a model of impaired insulin/insulin-like signaling (Kenyon *et al.* 1993; Murphy and Hu 2013), *isp-1(qm150)* as a model of impaired mitochondrial function (Feng *et al.* 2001; Dancy *et al.* 2015; Wang and Hekimi 2015), and the feeding-impaired *eat-2(ad1116)* mutant as a model of caloric restriction (Lakowski and Hekimi 1998; Lan *et al.* 2015). We identified 18 *daf-2* studies, 13 *isp-1* studies, and 23 *eat-2* studies and determined the Gompertz parameters from published

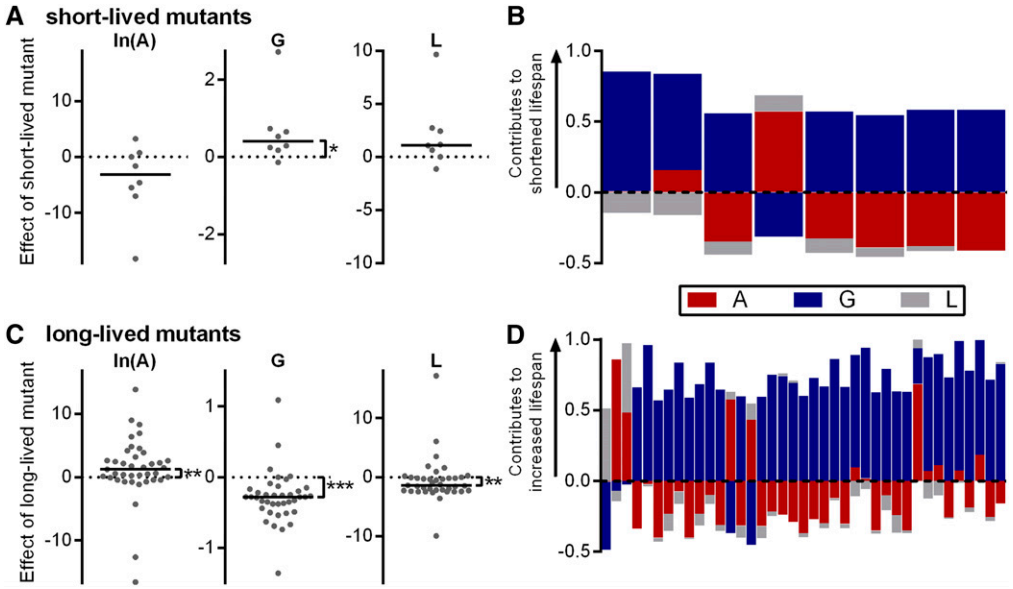


Figure 6 Pairwise comparison within studies of short- and long-lived *C. elegans* strains relative to their normal-lived N2 controls. (A) Differences in Gompertz parameter values between short-lived and N2 groups within studies. A positive value corresponds to an increase in parameter value for the mutant and a negative value to a decrease. (B) Fractional contribution of each parameter change to the shortened life span of short-lived lines of worms. Each bar is 1 unit long. A positive value indicates that the change in parameter value contributes to the shortened life span of the mutant. Negative values indicate that the parameter change acts to lengthen life span. (C) Differences in Gompertz parameter values between long-lived and N2 groups within studies. (D) Fractional contribution of each parameter change to the lengthened life span of long-lived lines of worms. A positive value indicates that the change in parameter value contributes to the increased life span of the mutant. Negative values indicate that the parameter change acts to shorten life span.

survival data (Table S4). There were decreases in *G* for the long-lived mutants relative to normal-lived N2 controls for each group of mutants (Figure 7, A–C, $P \leq 0.0012$ *vs.* a change of 0 by Wilcoxon signed-rank test) and no statistically significant effects on *A* or *L* (Figure 7, A–C). Likewise, decomposition of the contribution of each parameter to the increased longevity of each mutant revealed that, in the majority of studies for each mutant, changes to *G* were the dominant contributor to the increased average life span (Figure 7, D–F). We therefore conclude that *daf-2*, *isp-1*, and *eat-2* mutations can be reliably said to extend life span through decreases to the age-dependent acceleration of mortality rates.

Discussion

We found that the age-dependent acceleration of mortality rate, *G*, remained essentially invariant throughout the wide range of life spans that characterized normal and genetically long-lived mice (Figure 2). This is consistent with earlier findings that *G* is reasonably constant between different human populations and among a small number of inbred strains of laboratory mice (Finch 1990), as well as among wild-caught strains of *Drosophila* (Spencer and Promislow 2005), and that life-span-extending interventions in mice had a tendency to be associated with statistically significant changes in *A*, rather than *G* (Yen *et al.* 2008). We have shown that this effect is systematic, rather than sporadic, and can be observed even in homogenous environmental conditions, where the genetic makeup of the strains is the only variable (Figure 4). Thus it appears that most variation in mouse life span—save for extreme shortenings—is largely due to mechanisms that affect initial vulnerability. Importantly, this is true among populations of both wild-type control strains and those with experimentally introduced or spontaneous single-gene mutations that extend life span.

The apparent invariance of *G* for mice (Figure 2 and Figure 4) suggests that it is fixed within a relatively narrow band, with both increases and decreases likely to be associated with dramatically shortened life spans. Indeed, for the data set containing short-lived mice, the three lowest *G* values, as well as the seven largest, were associated with shortened life spans (Table S1 and Figure 2A). Interestingly, even decreases in *G* associated with extended life span may be accompanied by signs of early frailty relative to wild type. For example, young *Mclk1*$^{+/-}$ mice have impaired mitochondrial function and increased mitochondrial oxidative stress that is not apparent in aged *Mclk1*$^{+/-}$ mice (Lapointe and Hekimi 2008; Lapointe *et al.* 2009). This is associated with increased mortality relative to that of wild-type siblings that is reversed at ~2 years of age (made dramatically apparent by a crossing over of the survival curves at midlife), coupled with an increased *A* (Lapointe *et al.* 2009). Similarly, young *Prop1*$^{df/df}$ mice (with decreased *G*, Table S1) exhibit some marked physiological deficiencies in addition to their dwarf stature, including infertility, decreased ambulatory activity, and early frailty exemplified by a requirement for a prolonged nursing period and group housing (Conover and Bale 2007), along with a trend toward an increased *A*. Thus, observations in these two strains are consistent with decreases to *G* in mice being associated with detrimental effects of varying severity. These may be counterbalanced by protective effects over the long term, resulting in an increased life span.

Short-lived lines of mice demonstrated great variability in terms of the relationship between the Gompertz parameters and life span (Figure 2). This is consistent with the view that some or all of these lines may be short lived due to sicknesses

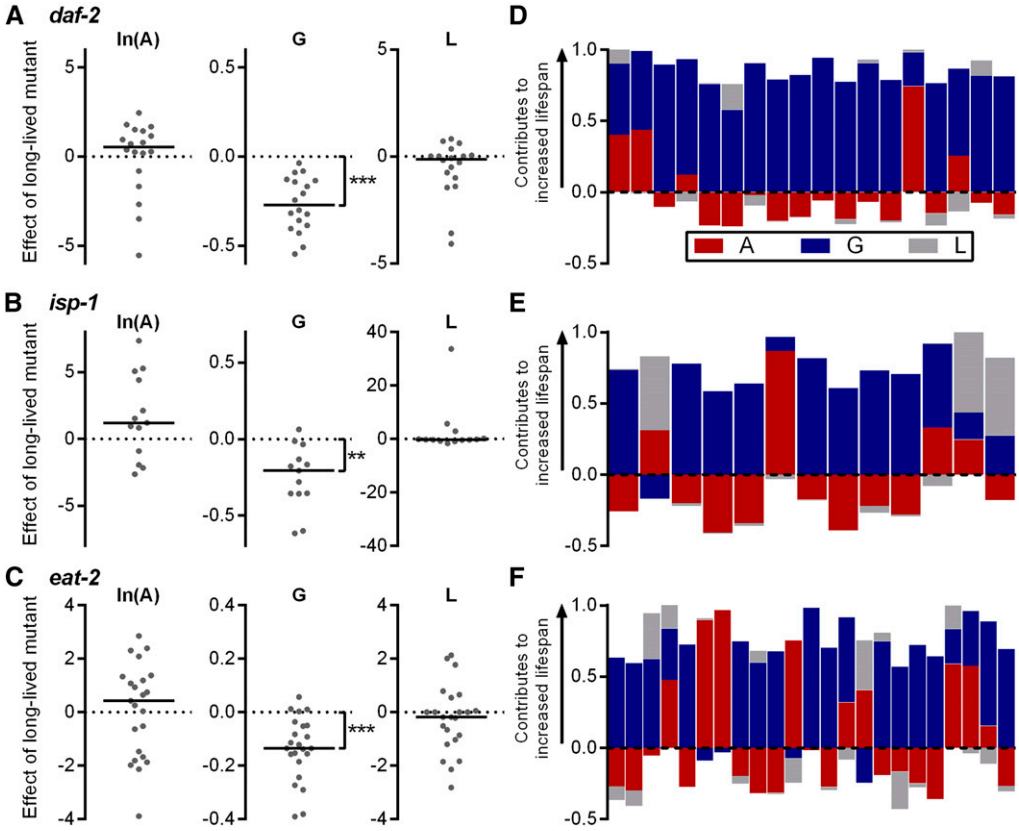


Figure 7 Pairwise comparison of Gompertz parameters within studies of long-lived *daf-2*, *isp-1*, and *eat-2* mutants (tabulated data in Table S4). (A–C) Differences in Gompertz parameter values between N2 and (A) *daf-2(e1370)*, (B) *isp-1(qm150)*, and (C) *eat-2(ad1116)* mutants within studies. A positive value corresponds to an increase in parameter value for the mutant and a negative value to a decrease. (D–F) Fractional contribution of each parameter change to the lengthened life span of (D) *daf-2*, (E) *isp-1*, and (F) *eat-2* mutant worms. Each bar is 1 unit long. A positive value indicates that the change in parameter value contributes to the increased life span of the mutant. Negative values indicate that the parameter change acts to shorten life span. ** $P = 0.0012$, *** $P < 0.0001$ *vs.* 0 by Wilcoxon signed-rank test.

distinct from aging and that the diversity of possible causes of mortality in this group combined to prevent the establishment of any clear pattern.

The remarkable invariance of *G* across a great range of mouse life spans would imply that these differences in life span do not reflect changes to the underlying biological aging process. This would seem to suggest that the vast majority of variation to life span seen in normal or long-lived mice—whether due to single-gene mutations or the more complex genetic heterogeneity among different strains—is not associated with any change in the aging process, but rather to aging-independent physiological features. Thus, *Bub1b* mutants ($G = 2.55$; median life span = 6 months), C57Bl/6 controls for *Trx* transgenic mice ($G = 2.67$; life span = 19 months), and $Prop1^{df/df}$ mice ($G = 2.89$; life span = 41 months) could be said to be aging at essentially the same rate. This is a surprising finding, since several long-lived strains of mice have been found to be resistant to the development of age-dependent pathologies (Flurkey *et al.* 2001; Kinney *et al.* 2001a,b; Ladiges *et al.* 2009), including several strains in which *G* was not decreased. For example, long-lived $AC5^{-/-}$ mice (*G* of 4.72 *vs.* 3.46 in controls) were protected from aging-induced cardiomyopathy (Yan *et al.* 2007), and both long-lived female $Irs1^{-/-}$ mice (*G* of 2.83 *vs.* 2.73 in controls) and $S6K1^{-/-}$ mice (*G* of 2.45 *vs.* 2.15 in controls) exhibited superior maintenance of motor skills and immune function into old age relative to their wild-type controls (Selman *et al.* 2008, 2009). Such signs of delayed biological aging have also been observed in FIRKO (Katic *et al.* 2007) and αMUPA (Gutman *et al.* 2011; Yanai *et al.* 2011) mice, both of which were found to be long lived with a statistically significant decrease in *A* and a trend toward an increased *G* (Table S1).

It would seem surprising that a population that is not aging slower would consist of individuals showing a slower rate of biological aging. How can we explain long-lived mice with fewer age-dependent pathologies but an unchanged rate of population aging? One possible explanation for this apparent paradox is that these interventions are delaying the age of onset of age-related pathologies, rather than slowing their progression. Pathological analysis carried out at a single young and old age (represented by the vertical dashed lines in Figure 1), as is common in murine life span studies, would not differentiate between delays in pathology onset and a change in the rate at which they worsen. Interestingly, a delayed onset of age-related pathology would also mirror the changes to mortality patterns for populations where life span increases due to changes to *A*, in which the rapid increase in mortality rates that characterizes mid- to old-age animals is delayed (Figure 1, solid blue lines). It has also been suggested that such a "rectangularization of the survival curve" in human populations would be associated with decreased durations of morbidity and hence beneficial (Fries 1980).

It is also worth noting that, although the theory behind the Gompertz model has been well explored (Gavrilov and Gavrilova 2001; Ricklefs and Scheuerlein 2002; Milne 2008), the role of *G* as a measure of the rate of aging does not seem

to have been subject to experimental validation (Driver 2001; Masoro 2006). As an alternative to the traditional view of *G* as a measure of aging, it has been suggested that a reduction in age-specific mortality throughout most of adult life would be sufficient evidence of a slower rate of aging, even if the rate at which it increased with age was unaffected (Masoro 2006). If future studies were to find that long-lived lines of mice display convincing evidence of a slower rate of accumulation of age-dependent pathology (*i.e.*, slower biological aging), despite alterations to *A* rather than *G*, this may be cause to rethink our conventional understanding of the meaning behind the Gompertz parameters.

The extended life spans of long-lived *C. elegans* mutants were found to be associated with decreases in the Gompertz parameter *G*, the rate of aging. This is in striking contrast to what we observed in mice. Such species-specific differences should perhaps not be unexpected: Although *C. elegans* has important advantages as a model organism for the study of aging, they are (unlike mammals) poikilothermic and self-fertilizing hermaphrodites (Brenner 1974; Hekimi *et al.* 2001), and aspects of their aging process are clearly distinct from those of mammals (Gruber *et al.* 2014). For example, the last portion of worm life span is often spent lying immobile on its plate, moving only rarely or if prodded (in some long-lived mutants, they can spend one-quarter of their life in this state) (Van Raamsdonk *et al.* 2010). This is not observed in mice, where immobility is cause for immediate euthanasia. Nematodes are also tolerant of physiological states that would be lethal in mammals, such as extreme hypoxia and hyperoxia (Van Voorhies and Ward 2000).

Another striking difference between long-lived worm and mouse mutants is the degree to which life span can be extended. Among the studies analyzed here, the average percentage of increase in life span was 76% for worms [with a maximum of 250% for *daf-2(e1370)* mutants and eight other strains with a >100% increase] *vs.* 20% for mice (including sets 1 and 2). Among mice, the two strains showing the greatest increase in life span showed increases of 92% and 51%, respectively, substantially less than observed for long-lived *C. elegans* (Figure S8A). It is possible that dramatic physiological shifts are required for the greatest increases in life span and that these are associated with changes to *G* rather than *A*. Thus we could predict that long-lived strains of *C. elegans* or mice with equivalent increases in life span relative to their controls might show similar changes to their Gompertz parameters. This is difficult to test because of the limited overlap in the degree of life span extension between the two species (Figure S8B). However, at the point of greatest overlap (*C. elegans* and mice with median life spans extended between 1.3 and 1.4 times relative to their controls), while there does not appear to be a difference in the effects on *G*, the effects on ln(*A*) are significantly different between the two species [$P = 0.0317$ by Mann–Whitney test, with ln(*A*) tending to be increased in long-lived worms and decreased in long-lived mice]. This suggests that the interspecies difference in parameter effects is not simply due to the degree of life span extension.

The trends that we have described in this study do not rule out the existence of long-lived mouse mutants with decreases in *G* rather than *A* or long-lived worms with changes to *A* rather than *G*. Such exceptions would in fact be valuable comparators to lines showing the more stereotyped pattern of changes and may help relate changes in mortality trends to underlying biological mechanisms. Interestingly, one environmental intervention, caloric restriction, seems to increase life span in mice via decreases to *G*, rather than *A* (Simons *et al.* 2013).

C. elegans, in particular, are attractive model organisms for identification of short- or long-lived strains that exhibit atypical Gompertzian behavior because their short life span and minimal requirements for uptake allow for higher-throughput life span experiments with greater sample sizes. Indeed, a recent study used a novel automated imaging system to collect high-precision survival data for multiple replicate populations of ≥500 animals (Stroustrup *et al.* 2016). Intriguingly, the authors found that life-span-shortening or -extending mutations resulted in survival curves that could be mapped onto control survival curves by application of a single temporal scaling parameter. The mortality rate models used here were constructed using a parameterization that is incompatible with such investigations of temporal scaling (Stroustrup *et al.* 2016), but it is tempting to imagine using this tool to identify strains of *C. elegans* that have extended life spans characterized by decreases to *A*, rather than *G*.

In conclusion, our principal finding is the interspecies variation in mortality rate kinetics in response to genetically driven changes to life span. In normal and long-lived mutant mice, there was a remarkable invariance of the age-dependent acceleration of mortality rate, represented by the Gompertz parameter *G*, across a wide range of median life spans. Although genetic manipulations are capable of increasing *G* in mice, such changes are more likely to result in shortened than in lengthened life span. Genetic alterations that extend life span, or affect life span within the normal range, almost invariably act through changes to the age-independent Gompertz parameter *A*. This appears to be true for single-gene mutations, as well as for the more complex changes that affect life span in various laboratory strains. This indicates that the vast majority of mouse life span extensions achieved via genetic manipulation are due to a delay in the onset of age-dependent mortality, rather than a slowing of the aging rate itself. This was not, however, conserved across species, with long-lived *C. elegans* exhibiting a decreased *G*. It is perhaps not surprising that the nature of life span extension may be fundamentally different between these species, given the substantial differences in physiology and environmental niche occupied, as well as life spans that differ by orders of magnitude.

Acknowledgments

S.H. is funded by past and current grants from the Canadian Institutes of Health Research, MOP-114891, MOP-123295, MOP-97869, and MOP-97869, as well as by McGill University.

Literature Cited

An, J. H., and T. K. Blackwell, 2003 SKN-1 links C. elegans mesendodermal specification to a conserved oxidative stress response. Genes Dev. 17: 1882–1893.

Antebi, A., 2007 Genetics of aging in Caenorhabditis elegans. PLoS Genet. 3: 1565–1571.

Apfeld, J., and C. Kenyon, 1999 Regulation of lifespan by sensory perception in Caenorhabditis elegans. Nature 402: 804–809.

Avraam, D., J. P. De Magalhaes, and B. Vasiev, 2013 A mathematical model of mortality dynamics across the lifespan combining heterogeneity and stochastic effects. Exp. Gerontol. 48: 801–811.

Baker, D. J., K. B. Jeganathan, J. D. Cameron, M. Thompson, S. Juneja *et al.*, 2004 BubR1 insufficiency causes early onset of aging-associated phenotypes and infertility in mice. Nat. Genet. 36: 744–749.

Baker, D. J., M. M. Dawlaty, T. Wijshake, K. B. Jeganathan, L. Malureanu *et al.*, 2013 Increased expression of BubR1 protects against aneuploidy and cancer and extends healthy lifespan. Nat. Cell Biol. 15: 96–102.

Benigni, A., D. Corna, C. Zoja, A. Sonzogni, R. Latini *et al.*, 2009 Disruption of the Ang II type 1 receptor promotes longevity in mice. J. Clin. Invest. 119: 524–530.

Berdichevsky, A., M. Viswanathan, H. R. Horvitz, and L. Guarente, 2006 C. elegans SIR-2.1 interacts with 14–3-3 proteins to activate DAF-16 and extend life span. Cell 125: 1165–1177.

Bogue, M. A., L. L. Peters, B. Paigen, R. Korstanje, R. Yuan *et al.*, 2016 Accessing data resources in the mouse phenome database for genetic analysis of murine life span and health span. J. Gerontol. A Biol. Sci. Med. Sci. 71: 170–177.

Brenner, S., 1974 The genetics of *Caenorhabditis elegans*. Genetics 77: 71–94.

Ceylan-Isik, A. F., M. Dong, Y. Zhang, F. Dong, S. Turdi *et al.*, 2013 Cardiomyocyte-specific deletion of endothelin receptor A rescues aging-associated cardiac hypertrophy and contractile dysfunction: role of autophagy. Basic Res. Cardiol. 108: 1–19.

Chang, S., A. S. Multani, N. G. Cabrera, M. L. Naylor, P. Laud *et al.*, 2004 Essential role of limiting telomeres in the pathogenesis of Werner syndrome. Nat. Genet. 36: 877–882.

Chen, S., J. R. Whetstine, S. Ghosh, J. A. Hanover, R. R. Gali *et al.*, 2009 The conserved NAD(H)-dependent corepressor CTBP-1 regulates Caenorhabditis elegans life span. Proc. Natl. Acad. Sci. USA 106: 1496–1501.

Ch'ng, Q., D. Sieburth, and J. M. Kaplan, 2008 Profiling synaptic proteins identifies regulators of insulin secretion and lifespan. PLoS Genet. 4: e1000283.

Conover, C. A., and L. K. Bale, 2007 Loss of pregnancy-associated plasma protein A extends lifespan in mice. Aging Cell 6: 727–729.

Conover, C. A., L. K. Bale, J. R. Mader, M. A. Mason, K. P. Keenan *et al.*, 2010 Longevity and age-related pathology of mice deficient in pregnancy-associated plasma protein-A. J. Gerontol. A Biol. Sci. Med. Sci. 65A: 590–599.

Conti, B., M. Sanchez-Alavez, R. Winsky-Sommerer, M. C. Morale, J. Lucero *et al.*, 2006 Transgenic mice with a reduced core body temperature have an increased life span. Science 314: 825–828.

Dancy, B. M., M. M. Sedensky, and P. G. Morgan, 2015 Mitochondrial bioenergetics and disease in Caenorhabditis elegans. Front. Biosci. 20: 198–228.

De Luca, G., I. Ventura, V. Sanghez, M. T. Russo, M. A. Ajmone-Cat *et al.*, 2013 Prolonged lifespan with enhanced exploratory behavior in mice overexpressing the oxidized nucleoside triphosphatase hMTH1. Aging Cell 12: 695–705.

De Magalhaes, J. P., J. A. Cabral, and D. Magalhaes, 2005 The influence of genes on the aging process of mice: a statistical assessment of the genetics of aging. Genetics 169: 265–274.

Dillin, A., A. L. Hsu, N. Arantes-Oliveira, J. Lehrer-Graiwer, H. Hsin *et al.*, 2002 Rates of behavior and aging specified by mitochondrial function during development. Science 298: 2398–2401.

Dorman, J. B., B. Albinder, T. Shroyer, and C. Kenyon, 1995 The age-1 and daf-2 genes function in a common pathway to control the lifespan of *Caenorhabditis elegans*. Genetics 141: 1399–1406.

Driver, C., 2001 The Gompertz function does not measure ageing. Biogerontology 2: 61–65.

Enns, L. C., J. F. Morton, P. R. Treuting, M. J. Emond, N. S. Wolf *et al.*, 2009 Disruption of protein kinase A in mice enhances healthy aging. PLoS One 4: e5963.

Ewbank, J. J., T. M. Barnes, B. Lakowski, M. Lussier, H. Bussey *et al.*, 1997 Structural and functional conservation of the Caenorhabditis elegans timing gene clk-1. Science 275: 980–983.

Ezekowitz, J. A., 2014 Time to energize coenzyme Q10 for patients with heart failure? JACC Heart Fail. 2: 650–652.

Feng, J. L., F. Bussiere, and S. Hekimi, 2001 Mitochondrial electron transport is a key determinant of life span in Caenorhabditis elegans. Dev. Cell 1: 633–644.

Finch, C. E., 1990 *Longevity, Senescence, and the Genome*. University of Chicago Press, Chicago.

Finch, C. E., and G. Ruvkun, 2001 The genetics of aging. Annu. Rev. Genomics Hum. Genet. 2: 435–462.

Finch, C. E., M. C. Pike, and M. Witten, 1990 Slow mortality rate accelerations during aging in some animals approximate that of humans. Science 249: 902–905.

Flurkey, K., J. Papaconstantinou, R. A. Miller, and D. E. Harrison, 2001 Lifespan extension and delayed immune and collagen aging in mutant mice with defects in growth hormone production. Proc. Natl. Acad. Sci. USA 98: 6736–6741.

Fries, J. F., 1980 Aging, natural death, and the compression of morbidity. N. Engl. J. Med. 303: 130–135.

Gates, A. C., C. Bernal-Mizrachi, S. L. Chinault, C. Feng, J. G. Schneider *et al.*, 2007 Respiratory uncoupling in skeletal muscle delays death and diminishes age-related disease. Cell Metab. 6: 497–505.

Gavrilov, L. A., and N. S. Gavrilova (Editors), 1991 *The Biology of Life Span: A Quantitative Approach*. Harwood Academic Publishers, Chur, Switzerland.

Gavrilov, L. A., and N. S. Gavrilova, 2001 The reliability theory of aging and longevity. J. Theor. Biol. 213: 527–545.

Gruber, J., C.-B. Chen, S. Fong, L. F. Ng, E. Teo *et al.*, 2014 Caenorhabditis elegans: what we can and cannot learn from aging worms. Antioxid. Redox Signal. 23: 256–279.

Gutman, R., Y. Genzer, N. Chapnik, R. Miskin, and O. Froy, 2011 Long-lived mice exhibit 24h locomotor circadian rhythms at young and old age. Exp. Gerontol. 46: 606–609.

Hajdu-Cronin, Y. M., W. J. Chen, and P. W. Sternberg, 2004 The L-type cyclin CYL-1 and the heat-shock-factor HSF-1 are required for heat-shock-induced protein expression in *Caenorhabditis elegans*. Genetics 168: 1937–1949.

Harper, J. M., J. E. Wilkinson, and R. A. Miller, 2010 Macrophage migration inhibitory factor-knockout mice are long lived and respond to caloric restriction. FASEB J. 24: 2436–2442.

Harrison, D. E., 1994 Potential misinterpretations using models of accelerated aging. J. Gerontol. 49: B245–B246.

Hekimi, S., 2006 How genetic analysis tests theories of animal aging. Nat. Genet. 38: 985–991.

Hekimi, S., C. Benard, R. Branicky, J. Burgess, A. K. Hihi *et al.*, 2001 Why only time will tell. Mech. Ageing Dev. 122: 571–594.

Honda, S., and M. Matsuo, 1992 Lifespan shortening of the nematode Caenorhabditis elegans under higher concentrations of oxygen. Mech. Ageing Dev. 63: 235–246.
Honda, S., N. Ishii, K. Suzuki, and M. Matsuo, 1993 Oxygen-dependent perturbation of life span and aging rate in the nematode. J. Gerontol. 48: B57–B61.
Hughes, B. G., and S. Hekimi, 2011 A mild impairment of mitochondrial electron transport has sex-specific effects on lifespan and aging in mice. PLoS One 6: e26116.
Johnson, T. E., 1987 Aging can be genetically dissected into component processes using long-lived lines of *Caenorhabditis elegans*. Proc. Natl. Acad. Sci. USA 84: 3777–3781.
Johnson, T. E., 1990 Increased life-span of age-1 mutants in Caenorhabditis elegans and lower Gompertz rate of aging. Science 249: 908–912.
Jones, O. R., A. Scheuerlein, R. Salguero-Gomez, C. G. Camarda, R. Schaible *et al.*, 2014 Diversity of ageing across the tree of life. Nature 505: 169–173.
Kanfi, Y., S. Naiman, G. Amir, V. Peshti, G. Zinman *et al.*, 2012 The sirtuin SIRT6 regulates lifespan in male mice. Nature 483: 218–221.
Kappeler, L., C. D. M. Filho, J. Dupont, P. Leneuve, P. Cervera *et al.*, 2008 Brain IGF-1 receptors control mammalian growth and lifespan through a neuroendocrine mechanism. PLoS Biol. 6: e254.
Katic, M., A. R. Kennedy, I. Leykin, A. Norris, A. Mcgettrick *et al.*, 2007 Mitochondrial gene expression and increased oxidative metabolism: role in increased lifespan of fat-specific insulin receptor knock-out mice. Aging Cell 6: 827–839.
Kenyon, C., J. Chang, E. Gensch, A. Rudner, and R. Tabtiang, 1993 A C. elegans mutant that lives twice as long as wild type. Nature 366: 461–464.
Kenyon, C. J., 2010 The genetics of ageing. Nature 464: 504–512.
Kinney, B. A., C. J. Meliska, R. W. Steger, and A. Bartke, 2001a Evidence that Ames dwarf mice age differently from their normal siblings in behavioral and learning and memory parameters. Horm. Behav. 39: 277–284.
Kinney, B. A., K. T. Coschigano, J. J. Kopchick, R. W. Steger, and A. Bartke, 2001b Evidence that age-induced decline in memory retention is delayed in growth hormone resistant GH-R-KO (Laron) mice. Physiol. Behav. 72: 653–660.
Kuhlbrodt, K., P. C. Janiesch, E. Kevei, A. Segref, R. Barikbin *et al.*, 2011 The Machado-Joseph disease deubiquitylase ATX-3 couples longevity and proteostasis. Nat. Cell Biol. 13: 273–281.
Kurosu, H., M. Yamamoto, J. D. Clark, J. V. Pastor, A. Nandi *et al.*, 2005 Suppression of aging in mice by the hormone klotho. Science 309: 1829–1833.
Ladiges, W., H. Van Remmen, R. Strong, Y. Ikeno, P. Treuting *et al.*, 2009 Lifespan extension in genetically modified mice. Aging Cell 8: 346–352.
Lakowski, B., and S. Hekimi, 1998 The genetics of caloric restriction in *Caenorhabditis elegans*. Proc. Natl. Acad. Sci. USA 95: 13091–13096.
Lan, J., X. Zhang, and D. Chen, 2015 Molecular mechanisms of dietary restriction in aging—insights from Caenorhabditis elegans research. Sci. China Life Sci. 58: 352–358.
Lapointe, J., and S. Hekimi, 2008 Early mitochondrial dysfunction in long-lived Mclk1+/− mice. J. Biol. Chem. 283: 26217–26227.
Lapointe, J., Z. Stepanyan, E. Bigras, and S. Hekimi, 2009 Reversal of the mitochondrial phenotype and slow development of oxidative biomarkers of aging in long-lived Mclk1+/− mice. J. Biol. Chem. 284: 20364–20374.
Larsen, P. L., and C. F. Clarke, 2002 Extension of life-span in Caenorhabditis elegans by a diet lacking coenzyme Q. Science 295: 120–123.
Lee, S. S., R. Y. Lee, A. G. Fraser, R. S. Kamath, J. Ahringer *et al.*, 2003 A systematic RNAi screen identifies a critical role for mitochondria in C. elegans longevity. Nat. Genet. 33: 40–48.
Lenaerts, I., S. Van Eygen, and J. Van Fleteren, 2007 Adult-limited dietary restriction slows gompertzian aging in Caenorhabditis elegans. Ann. N. Y. Acad. Sci. 1100: 442–448.
Li, Q., and J. Ren, 2007 Influence of cardiac-specific overexpression of insulin-like growth factor 1 on lifespan and aging-associated changes in cardiac intracellular Ca2+ homeostasis, protein damage and apoptotic protein expression. Aging Cell 6: 799–806.
Liang, H., E. J. Masoro, J. F. Nelson, R. Strong, C. A. Mcmahan *et al.*, 2003 Genetic mouse models of extended lifespan. Exp. Gerontol. 38: 1353–1364.
Liao, C.-Y., and B. K. Kennedy, 2014 Mouse models and aging: longevity and progeria, pp. 249–285 in *Current Topics in Developmental Biology*, edited by L. S. Colin. Academic Press, San Diego.
Liao, C. Y., B. A. Rikke, T. E. Johnson, V. Diaz, and J. F. Nelson, 2010 Genetic variation in the murine lifespan response to dietary restriction: from life extension to life shortening. Aging Cell 9: 92–95.
Liu, X., N. Jiang, B. Hughes, E. Bigras, E. Shoubridge *et al.*, 2005 Evolutionary conservation of the clk-1-dependent mechanism of longevity: loss of mclk1 increases cellular fitness and lifespan in mice. Genes Dev. 19: 2424–2434.
Masoro, E. J., 2006 Caloric restriction and aging: controversial issues. J. Gerontol. A Biol. Sci. Med. Sci. 61: 14–19.
Matheu, A., A. Maraver, P. Klatt, I. Flores, I. Garcia-Cao *et al.*, 2007 Delayed ageing through damage protection by the Arf/p53 pathway. Nature 448: 375–379.
Merry, B. J., 2005 Dietary restriction in rodents—Delayed or retarded ageing? Mech. Ageing Dev. 126: 951–959.
Miller, R. A., 2004 'Accelerated aging': A primrose path to insight? Aging Cell 3: 47–51.
Milne, E. M. G., 2008 The natural distribution of survival. J. Theor. Biol. 255: 223–236.
Mounkes, L. C., S. Kozlov, L. Hernandez, T. Sullivan, and C. L. Stewart, 2003 A progeroid syndrome in mice is caused by defects in A-type lamins. Nature 423: 298–301.
Murphy, C. T., and P. J. Hu, 2013 Insulin/insulin-like growth factor signaling in C. elegans (December 26, 2013), *WormBook*, ed. The *C. elegans* Research Community, WormBook, doi/10.1895/wormbook.1.164.1, http://www.wormbook.org.
Nojima, A., M. Yamashita, Y. Yoshida, I. Shimizu, H. Ichimiya *et al.*, 2013 Haploinsufficiency of Akt1 prolongs the lifespan of mice. PLoS One 8: e69178.
Olshansky, S. J., and B. A. Carnes, 1997 Ever since Gompertz. Demography 34: 1–15.
Orr, W. C., R. J. Mockett, J. J. Benes, and R. S. Sohal, 2003 Effects of overexpression of copper-zinc and manganese superoxide dismutases, catalase, and thioredoxin reductase genes on longevity in Drosophila melanogaster. J. Biol. Chem. 278: 26418–26422.
Ortega-Molina, A., A. Efeyan, E. Lopez-Guadamillas, M. Muñoz-Martin, G. Gómez-López *et al.*, 2012 Pten positively regulates brown adipose function, energy expenditure, and longevity. Cell Metab. 15: 382–394.
Palmitessa, A., and J. L. Benovic, 2010 Arrestin and the multi-PDZ domain-containing protein MPZ-1 interact with phosphatase and tensin homolog (PTEN) and regulate Caenorhabditis elegans longevity. J. Biol. Chem. 285: 15187–15200.
Partridge, L., S. D. Pletcher, and W. Mair, 2005 Dietary restriction, mortality trajectories, risk and damage. Mech. Ageing Dev. 126: 35–41.
Pérez, V. I., L. A. Cortez, C. M. Lew, M. Rodriguez, C. R. Webb *et al.*, 2011 Thioredoxin 1 overexpression extends mainly the earlier part of life span in mice. J. Gerontol. A Biol. Sci. Med. Sci. 66A: 1286–1299.
Pletcher, S. D., 1999 Model fitting and hypothesis testing for age-specific mortality data. J. Evol. Biol. 12: 430–439.
Pletcher, S. D., A. A. Khazaeli, and J. W. Curtsinger, 2000 Why do life spans differ? Partitioning mean longevity differences in terms of age-specific mortality parameters. J. Gerontol. A Biol. Sci. Med. Sci. 55: B381–B389.

Promislow, D. E. L., M. Tatar, S. Pletcher, and J. Carey, 1999 Below-threshold mortality: implications for studies in evolution, ecology and demography. J. Evol. Biol. 12: 314–328.

Pyo, J.-O., S.-M. Yoo, H.-H. Ahn, J. Nah, S.-H. Hong *et al.*, 2013 Overexpression of Atg5 in mice activates autophagy and extends lifespan. Nat. Commun. 4: 2300.

Ran, Q., H. Y. Liang, Y. Ikeno, W. B. Qi, T. A. Prolla *et al.*, 2007 Reduction in glutathione peroxidase 4 increases life span through increased sensitivity to apoptosis. J. Gerontol. Ser. A Biol. Sci. Med. Sci. 62: 932–942.

Redmann, Jr., S. M., and G. Argyropoulos, 2006 AgRP-deficiency could lead to increased lifespan. Biochem. Biophys. Res. Commun. 351: 860–864.

Ricklefs, R. E., and A. Scheuerlein, 2002 Biological implications of the Weibull and Gompertz models of aging. J. Gerontol. A Biol. Sci. Med. Sci. 57: B69–B76.

Sacher, G. A., 1977 Life table modification and life prolongation, pp. 582–637 in *Handbook of the Biology of Aging*, edited by C. E. Finch, and L. Hayflick. Van Nostrand Reinhold, New York.

Satoh, A., S. C. Brace, N. Rensing, P. Cliften, D. F. Wozniak *et al.*, 2013 Sirt1 extends life span and delays aging in mice through the regulation of Nk2 homeobox 1 in the DMH and LH. Cell Metab. 18: 416–430.

Selman, C., and D. J. Withers, 2011 Mammalian models of extended healthy lifespan. Philos. Trans. R. Soc. Lond. B Biol. Sci. 366: 99–107.

Selman, C., S. Lingard, A. I. Choudhury, R. L. Batterham, M. Claret *et al.*, 2008 Evidence for lifespan extension and delayed age-related biomarkers in insulin receptor substrate 1 null mice. FASEB J. 22: 807–818.

Selman, C., J. M. A. Tullet, D. Wieser, E. Irvine, S. J. Lingard *et al.*, 2009 Ribosomal protein S6 kinase 1 signaling regulates mammalian life span. Science 326: 140–144.

Selman, C., L. Partridge, and D. J. Withers, 2011 Replication of extended lifespan phenotype in mice with deletion of insulin receptor substrate 1. PLoS One 6: e16144.

Shaw, W. M., S. Luo, J. Landis, J. Ashraf, and C. T. Murphy, 2007 The C. elegans TGF-β Dauer pathway regulates longevity via insulin signaling. Curr. Biol. 17: 1635–1645.

Simons, M. J. P., W. Koch, and S. Verhulst, 2013 Dietary restriction of rodents decreases aging rate without affecting initial mortality rate – a meta-analysis. Aging Cell 12: 410–414.

Singh, S. P., M. Niemczyk, D. Saini, V. Sadovov, L. Zimniak *et al.*, 2010 Disruption of the mGsta4 gene increases life span of C57BL mice. J. Gerontol. A Biol. Sci. Med. Sci. 65: 14–23.

Spencer, C. C., and D. E. Promislow, 2005 Age-specific changes in epistatic effects on mortality rate in Drosophila melanogaster. J. Hered. 96: 513–521.

Streeper, R. S., C. A. Grueter, N. Salomonis, S. Cases, M. C. Levin *et al.*, 2012 Deficiency of the lipid synthesis enzyme, DGAT1, extends longevity in mice. Aging 4: 13–27.

Stroustrup, N., W. E. Anthony, Z. M. Nash, V. Gowda, A. Gomez *et al.*, 2016 The temporal scaling of Caenorhabditis elegans ageing. Nature 530: 103–107.

Sun, L. Y., A. Spong, W. R. Swindell, Y. Fang, C. Hill *et al.*, 2013 Growth hormone-releasing hormone disruption extends lifespan and regulates response to caloric restriction in mice. eLife 2: e01098.

Suthammarak, W., B. H. Somerlot, E. Opheim, M. Sedensky, and P. G. Morgan, 2013 Novel interactions between mitochondrial superoxide dismutases and the electron transport chain. Aging Cell 12: 1132–1140.

Tacutu, R., T. Craig, A. Budovsky, D. Wuttke, G. Lehmann *et al.*, 2013 Human ageing genomic resources: integrated databases and tools for the biology and genetics of ageing. Nucleic Acids Res. 41: D1027–D1033.

Takeda, T., M. Hosokawa, S. Takeshita, M. Irino, K. Higuchi *et al.*, 1981 A new murine model of accelerated senescence. Mech. Ageing Dev. 17: 183–194.

Tavernarakis, N., A. Pasparaki, E. Tasdemir, M. C. Maiuri, and G. Kroemer, 2008 The effects of p53 on whole organism longevity are mediated by autophagy. Autophagy 4: 870–873.

Tomás-Loba, A., I. Flores, P. J. Fernández-Marcos, M. L. Cayuela, A. Maraver *et al.*, 2008 Telomerase reverse transcriptase delays aging in cancer-resistant mice. Cell 135: 609–622.

Tóth, M. L., T. Sigmond, É. Borsos, J. Barna, P. Erdélyi *et al.*, 2008 Longevity pathways converge on autophagy genes to regulate life span in Caenorhabditis elegans. Autophagy 4: 330–338.

Tricoire, H., and M. Rera, 2015 A new, discontinuous 2 phases of aging model: lessons from *Drosophila melanogaster*. PLoS One 10: e0141920.

Trifunovic, A., A. Wredenberg, M. Falkenberg, J. N. Spelbrink, A. T. Rovio *et al.*, 2004 Premature ageing in mice expressing defective mitochondrial DNA polymerase. Nature 429: 417–423.

Vanfleteren, J. R., A. De Vreese and B. P. Braeckman, 1998 Two-parameter logistic and Weibull equations provide better fits to survival data from isogenic populations of Caenorhabditis elegans in axenic culture than does the Gompertz model. J. Gerontol. A Biol. Sci. Med. Sci. 53**:** B393–B403; discussion B404–B398.

Van Raamsdonk, J. M., and S. Hekimi, 2009 Deletion of the mitochondrial superoxide dismutase sod-2 extends lifespan in Caenorhabditis elegans. PLoS Genet. 5: e1000361.

Van Raamsdonk, J. M., and S. Hekimi, 2010 Reactive oxygen species and aging in Caenorhabditis elegans: Causal or casual relationship? Antioxid. Redox Signal. 13: 1911–1953.

Van Raamsdonk, J. M., Y. Meng, D. Camp, W. Yang, X. Jia *et al.*, 2010 Decreased energy metabolism extends life span in *Caenorhabditis elegans* without reducing oxidative damage. Genetics 185: 559–571.

Van Voorhies, W. A., and S. Ward, 2000 Broad oxygen tolerance in the nematode Caenorhabditis elegans. J. Exp. Biol. 203: 2467–2478.

Wang, Y., and S. Hekimi, 2015 Mitochondrial dysfunction and longevity in animals: untangling the knot. Science 350: 1204–1207.

Wilson, D. L., 1994 The analysis of survival (mortality) data: fitting Gompertz, Weibull, and logistic functions. Mech. Ageing Dev. 74: 15–33.

Wong, K.-K., R. S. Maser, R. M. Bachoo, J. Menon, D. R. Carrasco *et al.*, 2003 Telomere dysfunction and Atm deficiency compromises organ homeostasis and accelerates ageing. Nature 421: 643–648.

Wu, D., J. R. Cypser, A. I. Yashin, and T. E. Johnson, 2009 Multiple mild heat-shocks decrease the Gompertz component of mortality in Caenorhabditis elegans. Exp. Gerontol. 44: 607–612.

Wu, J. J., J. Liu, E. B. Chen, J. J. Wang, L. Cao *et al.*, 2013 Increased mammalian lifespan and a segmental and tissue-specific slowing of aging after genetic reduction of mTOR expression. Cell Reports 4: 913–920.

Xu, J., G. Gontier, Z. Chaker, P. Lacube, J. Dupont *et al.*, 2014 Longevity effect of IGF-1R+/− mutation depends on genetic background-specific receptor activation. Aging Cell 13: 19–28.

Yan, L., D. E. Vatner, J. P. O'connor, A. Ivessa, H. Ge *et al.*, 2007 Type 5 adenylyl cyclase disruption increases longevity and protects against stress. Cell 130: 247–258.

Yanai, H., A. Budovsky, R. Tacutu, and V. E. Fraifeld, 2011 Is rate of skin wound healing associated with aging or longevity phenotype? Biogerontology 12: 591–597.

Yanase, S., K. Yasuda, and N. Ishii, 2002 Adaptive responses to oxidative damage in three mutants of Caenorhabditis elegans

(age-1, mev-1 and daf-16) that affect life span. Mech. Ageing Dev. 123: 1579–1587.

Yang, W., and S. Hekimi, 2010 Two modes of mitochondrial dysfunction lead independently to lifespan extension in Caenorhabditis elegans. Aging Cell 9: 433–447.

Yang, X., T. A. Doser, C. X. Fang, J. M. Nunn, R. Janardhanan *et al.*, 2006 Metallothionein prolongs survival and antagonizes senescence-associated cardiomyocyte diastolic dysfunction: role of oxidative stress. FASEB J. 20: 1024–1026.

Yen, K., and C. V. Mobbs, 2010 Evidence for only two independent pathways for decreasing senescence in Caenorhabditis elegans. Age 32: 39–49.

Yen, K., D. Steinsaltz, and C. V. Mobbs, 2008 Validated analysis of mortality rates demonstrates distinct genetic mechanisms that influence lifespan. Exp. Gerontol. 43: 1044–1051.

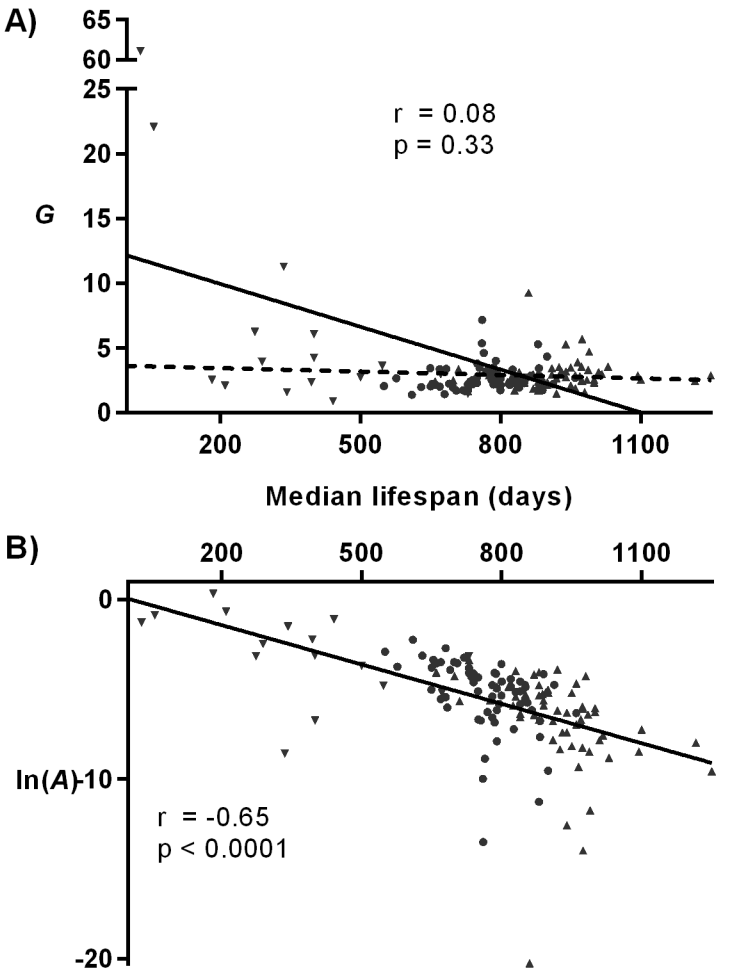
Yuan, R., S. W. Tsaih, S. B. Petkova, C. M. De Evsikova, S. Q. Xing *et al.*, 2009 Aging in inbred strains of mice: study design and interim report on median lifespans and circulating IGF1 levels. Aging Cell 8: 277–287.

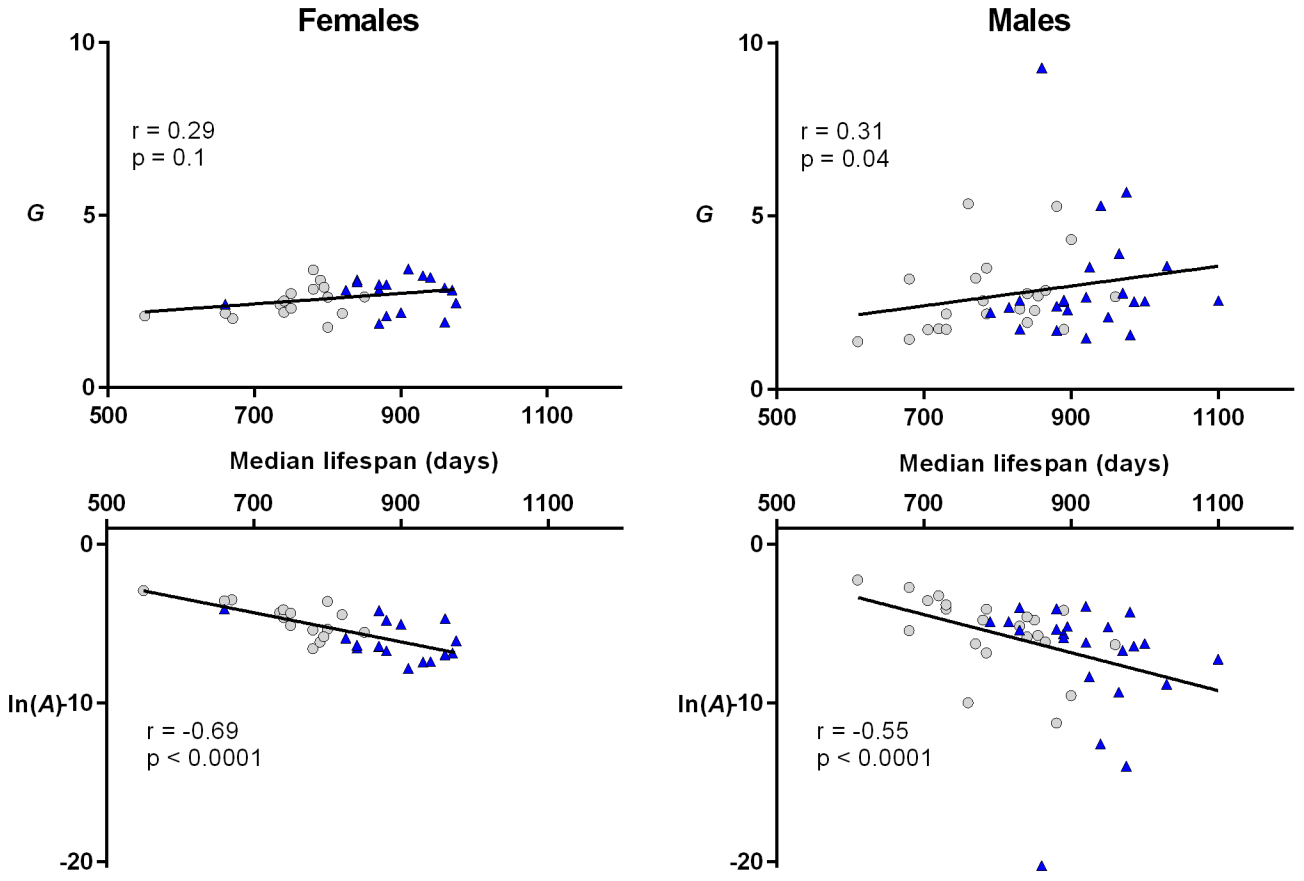
Yuan, R., Q. Meng, J. Nautiyal, K. Flurkey, S.-W. Tsaih *et al.*, 2012 Genetic coregulation of age of female sexual maturation and lifespan through circulating IGF1 among inbred mouse strains. Proc. Natl. Acad. Sci. USA 109: 8224–8229.

Zhang, G., J. Li, S. Purkayastha, Y. Tang, H. Zhang *et al.*, 2013 Hypothalamic programming of systemic ageing involving IKK-B, NF-kB and GnRH. Nature 497: 211–216.

Zhang, Y., Y. Xie, E. D. Berglund, K. C. Coate, T. T. He *et al.*, 2012 The starvation hormone, fibroblast growth factor-21, extends lifespan in mice. eLife 1: e00065.

Communicating editor: G. A. Churchill


A)
65
60
25
20
15
10
5
0
G
r = 0.08
p = 0.33
200
500
800
1100
Median lifespan (days)
B)
200
500
800
1100
0
-10
-20
ln(A)
r = -0.65
p < 0.0001


Females
Males
r = 0.29
p = 0.1
r = 0.31
p = 0.04
G
10
5
0
500
700
900
1100
Median lifespan (days)
ln(A)
-10
-20
r = -0.69
p < 0.0001
r = -0.55
p < 0.0001

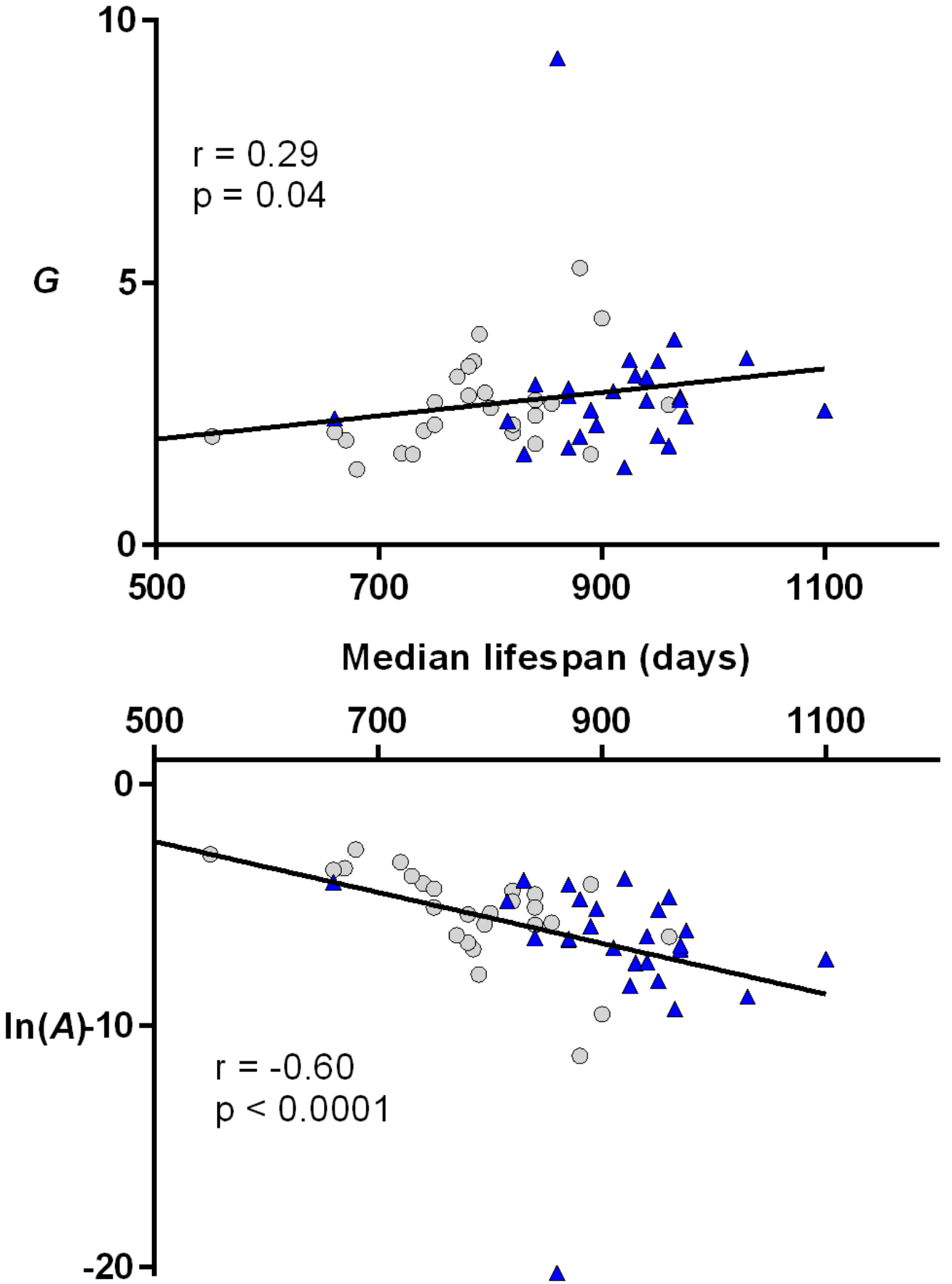

10
5
0
G
r = 0.29
p = 0.04
500
700
900
1100
Median lifespan (days)
500
700
900
1100
0
-10
-20
ln(A)
r = -0.60
p < 0.0001

Figure S4

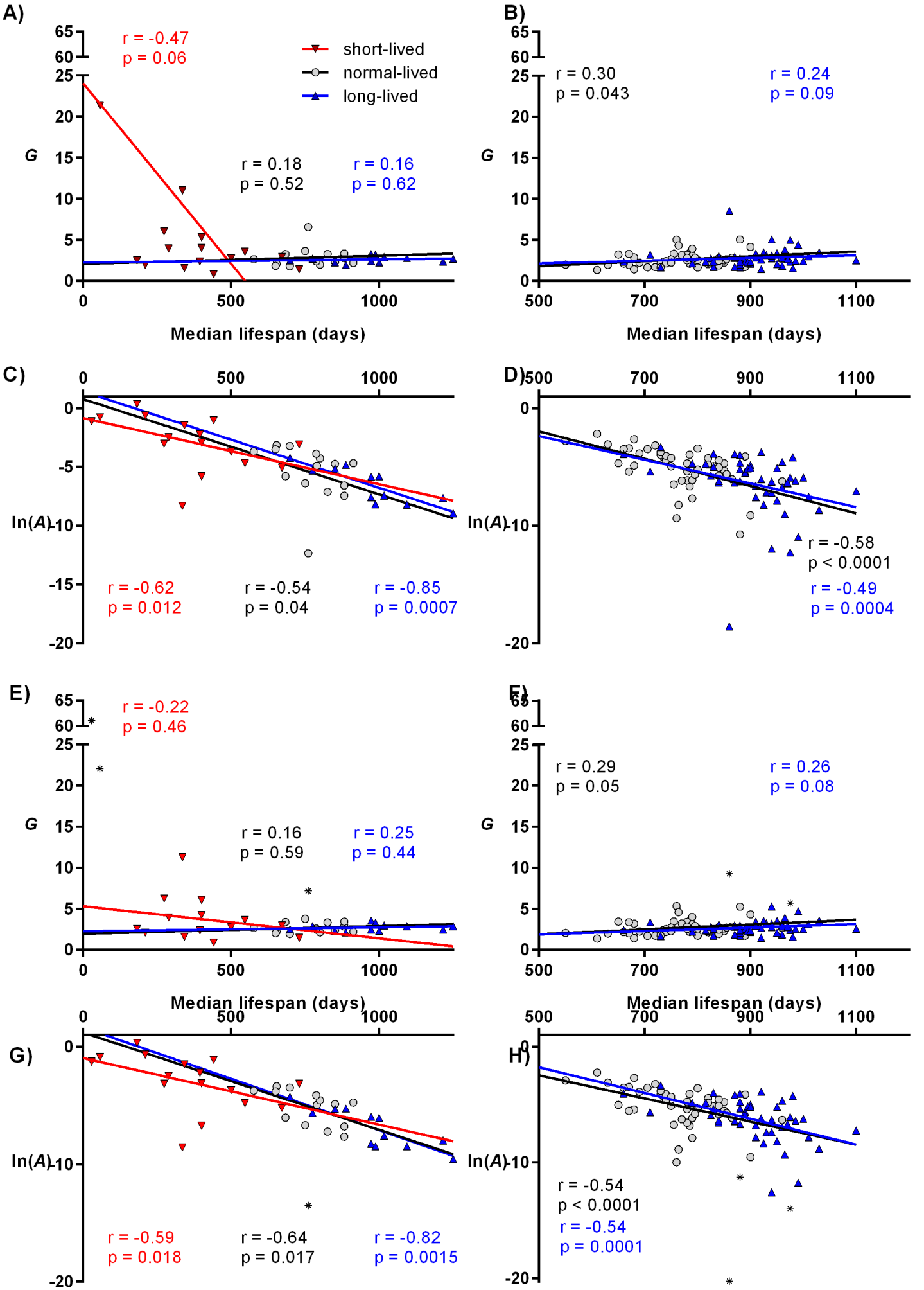
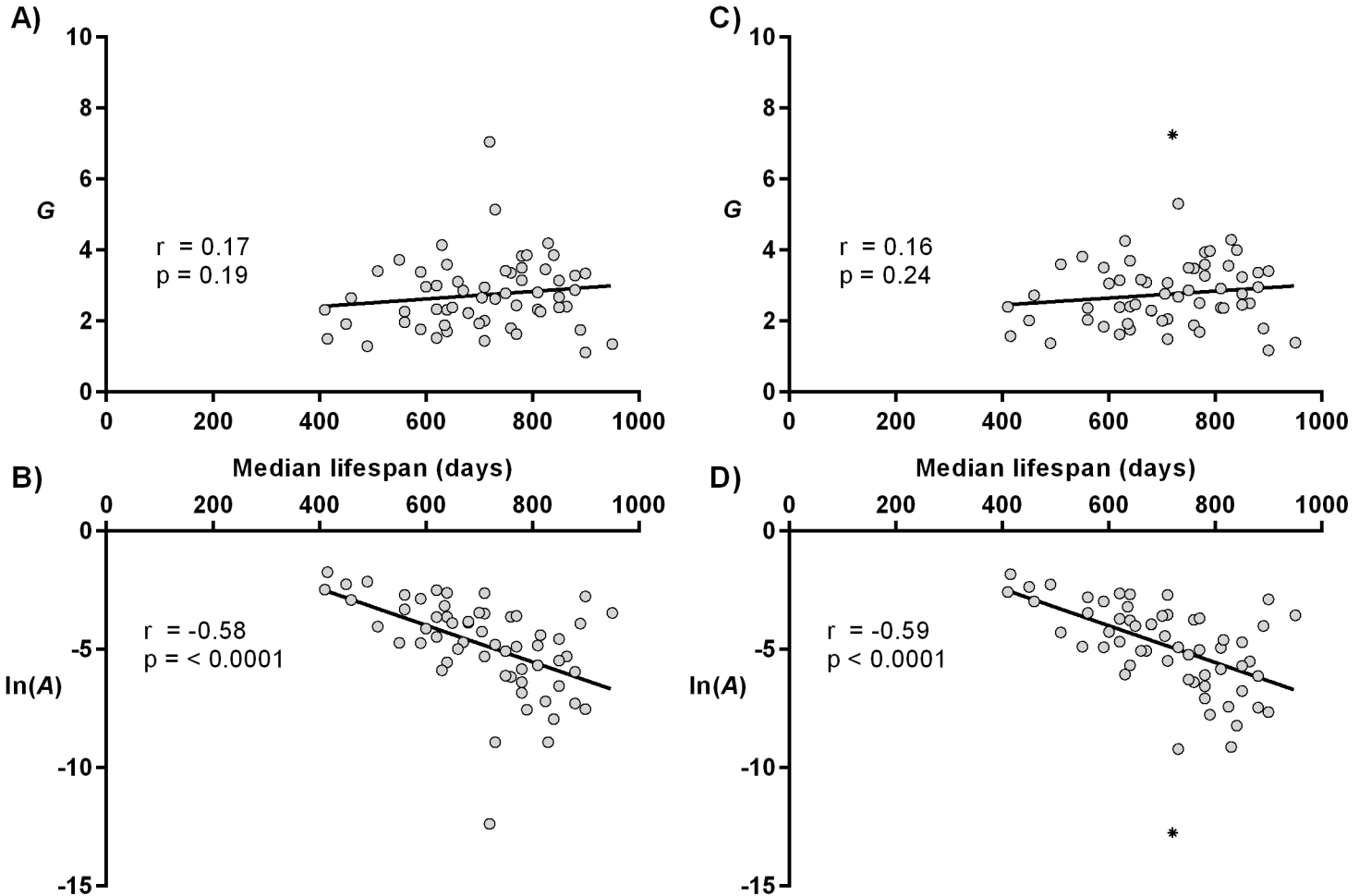
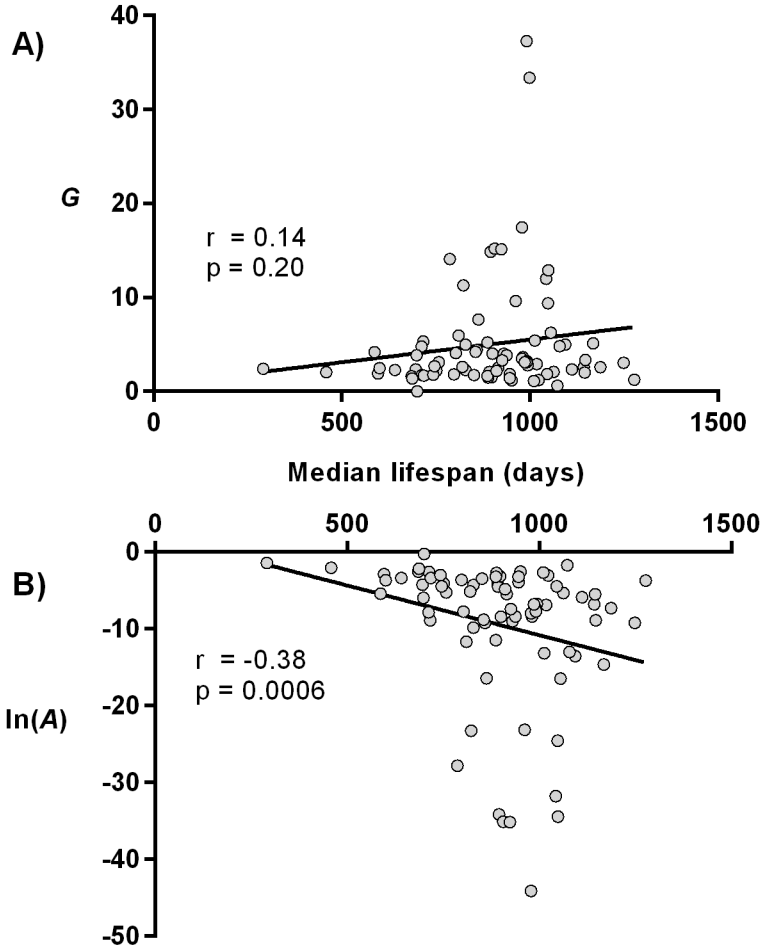
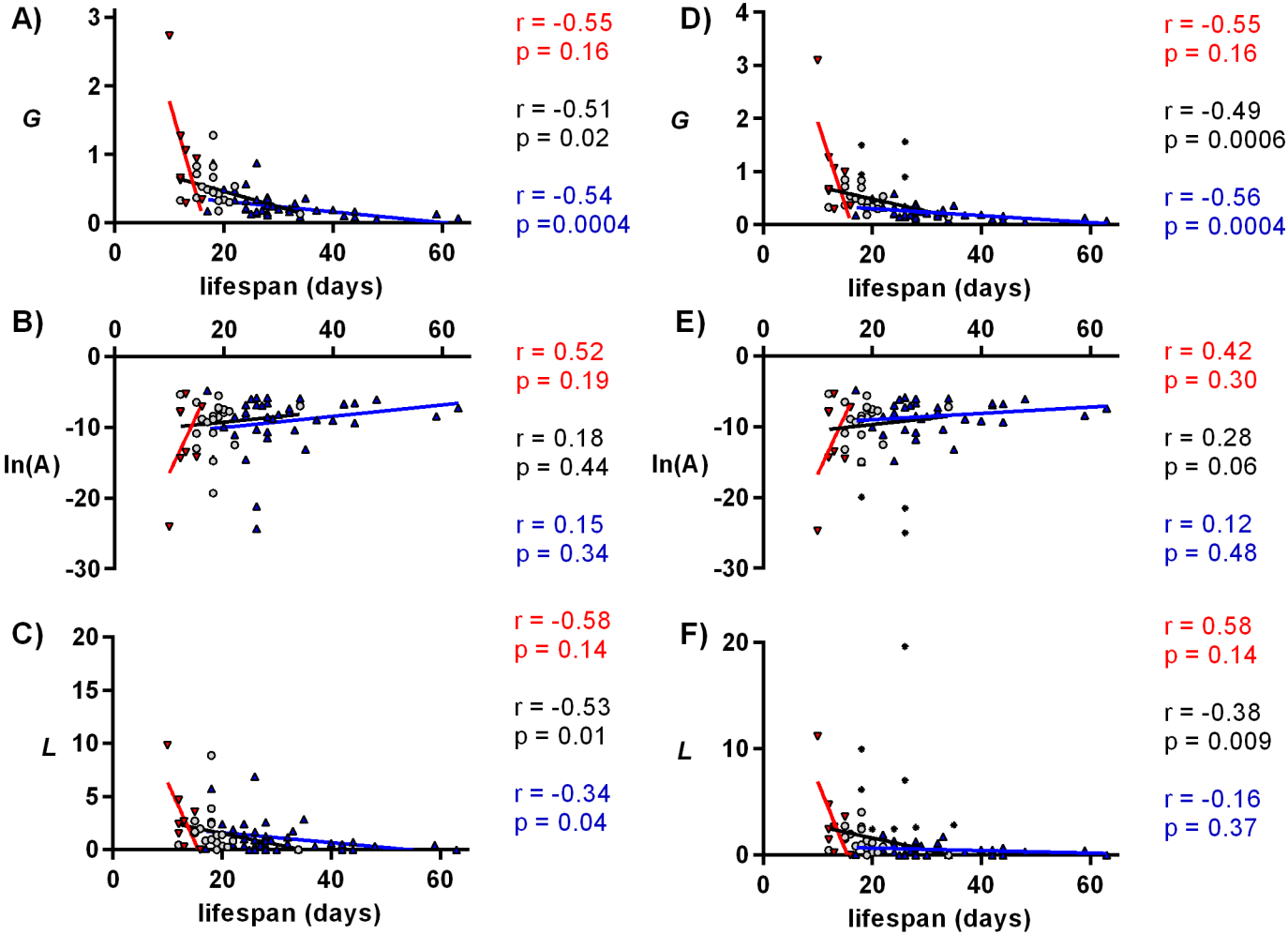

A)
B)
C)
D)
E)
F)
G)
H)
short-lived
normal-lived
long-lived
G
ln(A)
Median lifespan (days)
r = -0.47
p = 0.06
r = 0.18
p = 0.52
r = 0.16
p = 0.62
r = 0.30
p = 0.043
r = 0.24
p = 0.09
r = -0.62
p = 0.012
r = -0.54
p = 0.04
r = -0.85
p = 0.0007
r = -0.58
p < 0.0001
r = -0.49
p = 0.0004
r = -0.22
p = 0.46
r = 0.16
p = 0.59
r = 0.25
p = 0.44
r = 0.29
p = 0.05
r = 0.26
p = 0.08
r = -0.59
p = 0.018
r = -0.64
p = 0.017
r = -0.82
p = 0.0015
r = -0.54
p < 0.0001
r = -0.54
p = 0.0001

Figure S5


A)
C)
B)
D)
G
ln(A)
r = 0.17
p = 0.19
r = 0.16
p = 0.24
r = -0.58
p = < 0.0001
r = -0.59
p < 0.0001
Median lifespan (days)
0
200
400
600
800
1000
2
4
6
8
10
-5
-10
-15


A)
G
r = 0.14
p = 0.20
Median lifespan (days)
B)
ln(A)
r = -0.38
p = 0.0006


A)
G
lifespan (days)
r = -0.55
p = 0.16
r = -0.51
p = 0.02
r = -0.54
p =0.0004
B)
ln(A)
r = 0.52
p = 0.19
r = 0.18
p = 0.44
r = 0.15
p = 0.34
C)
L
lifespan (days)
r = -0.58
p = 0.14
r = -0.53
p = 0.01
r = -0.34
p = 0.04
D)
G
lifespan (days)
r = -0.55
p = 0.16
r = -0.49
p = 0.0006
r = -0.56
p = 0.0004
E)
ln(A)
r = 0.42
p = 0.30
r = 0.28
p = 0.06
r = 0.12
p = 0.48
F)
L
lifespan (days)
r = 0.58
p = 0.14
r = -0.38
p = 0.009
r = -0.16
p = 0.37

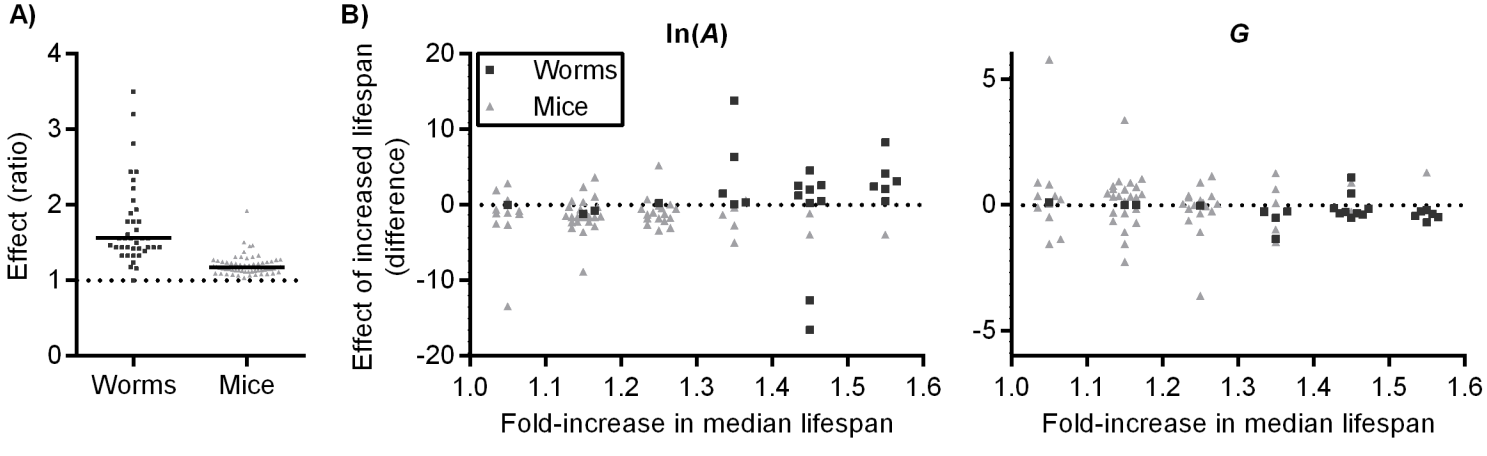

A)
Effect (ratio)
4
3
2
1
0
Worms
Mice
B)
ln(A)
Worms
Mice
Effect of increased lifespan (difference)
20
10
0
-10
-20
1.0
1.1
1.2
1.3
1.4
1.5
1.6
Fold-increase in median lifespan
G
5
0
-5
1.0
1.1
1.2
1.3
1.4
1.5
1.6
Fold-increase in median lifespan

Table S1. Previously published (Yen *et al.* 2008) maximum likelihood estimations of the Gompertz parameter values obtained from published survival curves (Set 1).

Gene and allele or strain	Background	Control				Intervention				
		n	Median lifespan (days)	ln(*A*)	*G*	n	Median lifespan (days)	ln(*A*)	*G*	
Atm $^{-/-}$, *Terc* $^{-/-}$	C57BL/6×WWG		-	-	-	51	343 ↓	-1.5 ↑	1.6 ↓	(Wong *et al.* 2003)
NZB/W	NZW×NZB(F1)		-	-	-	22	274 ↓	-3.16	6.25 ↑	(Conde *et al.* 1998)
Bub1b $^{H/H}$	129/Sv×FVB		-	-	-	212	182 ↓	0.32 ↑	2.55	(Baker *et al.* 2004)
C/ebp $^{b/b}$	C57BL/6J	30	684 *	-6.02	3.4	30	851 ↑	-5.29	2.33	(Chiu *et al.* 2004)
Lep $^{ob/ob}$	C57BL/6J					40	547 ↓	-4.81	3.65	
Cat TG	B6(B6C3F1) ×C57BL/6J	58	790	-4.95	2.44	42	973 ↑	-5.95	2.48	(Schriner *et al.* 2005)
GH TG	C56BL/6×SJL	16	800	-4.56	2.14	9	400 ↓	-6.73	6.06 ↑	(Bartke 2003)
Ghr $^{-/-}$	OLA-BALB/cJ	15	698	-3.48	1.95	11	888 ↑	-5.25	2.14	(Coschigano *et al.* 2003)
Ghrhr $^{lit/lit}$	C57BL/6	31	882	-6.77	2.86	35	1094 ↑	-8.47	2.9	(Flurkey *et al.* 2001)
Igf1r $^{+/-}$	129/J	17	654	-3.36	2.25	20	775 ↑	-5.62	2.88	(Holzenberger *et al.* 2003)
Insr $^{-/-}$ (Firko)	FVB×129S4	67	913	-4.75	2.22	60	1017 ↑	-7.56 ↓	2.98	(Blüher *et al.* 2003)
Klotho $^{kl/kl}$	C57BL/6J + C3H/J		-	-	-	29	57 ↓	-0.88	22.06 ↑	(Kuro-O *et al.* 1997)
Lmna $^{L530P/L530P}$	C57BL/6 + 129S1/Sv		-	-	-	25	29 ↓	-1.27	61.06 ↑	(Mounkes *et al.* 2003)
MsrA $^{-/-}$	C57BL/6 + 129/SvJ	14	788	-4.13	2.58	17	400 ↓	-3.12	4.23	(Moskovitz *et al.* 2001)
p53 $^{+/-}$	129/SV + C57BL/6	56	826 *	-7.22	3.34	217	500 ↓	-3.7 ↑	2.75	(Tyner *et al.* 2002)
p53 $^{+/m}$	129/SV + C57BL/6					35	672 ↓	-5.2	2.97	
p66 $^{shc\ -/-}$	129/Sv	14	761	-13.5	7.17	15	973 ↑	-8.25	3.57	(Migliaccio *et al.* 1999)
Pit1 $^{dw/dw}$	C3H/HeJ + DW/J	34	882	-7.66	3.44	25	1216 ↑	-7.96	2.47	(Flurkey *et al.* 2001)
Plau TG (α-MUPA)	FVB/N	33	851	-4.87	2.38	33	988 ↑	-8.47 ↓	3.32	(Miskin and Masos 1997)
PolgA $^{mut/mut}$	129 + C57BL/6		-	-	-	38	336 ↓	-8.57 ↓	11.27 ↑	(Trifunovic *et al.* 2004)
Prdx1 $^{-/-}$	B6×129SvEv		-	-	-	34	730 ↓	-3.15	1.46 ↓	(Neumann *et al.* 2003)
Prop1 $^{df/df}$	Ames Stock	13	650	-3.8	2.04	16	1250 ↑	-9.57 ↓	2.89	(Brown-Borg *et al.* 1996)
Prop1 $^{df/df}$	Ames Stock	26	750	-6.68	3.8	24	1000 ↑	-6.05	2.33 ↓	(Bartke *et al.* 2001)
Senescence Accelerated Mice (SAM)	AKR	377	395 **	-2.22	2.36	493	289 ↓	-2.48	3.94 ↑	(Takeda *et al.* 1981)
Top3B $^{-/-}$	C57BL/6J + 129/svEv		-	-	-	30	441 ↓	-1.1 ↑	0.89 ↓	(Kwan and Wang 2001)
Trx TG	C57BL/6	82	577	-3.73	2.67	94	699 ↑	-4.26	2.52	(Mitsui *et al.* 2002)
Wrn $^{-/-}$, *Terc* $^{-/-}$	C57BL/6 + 129Sv + BALB/c + SLJ		-	-	-	39	210 ↓	-0.67 ↑	2.08	(Chang *et al.* 2004)

Gompertz parameter values, their statistical significance and genetic backgrounds are as published previously (Adapted from TABLE 1, Yen *et al.* 2008). Statistically-significant changes relative to controls (as determined by Yen et al.) are indicated by up (increased) or down (decreased) arrows. When controls were not reported, statistical comparisons were made against generic, representative control values (*G* = 2.93 and ln(*A*) = -5.43). Tg: transgene. Median lifespans were obtained from the original cited paper.

* *C/ebp* and *lep* mutants shared the same control group, as did p53 $^{+/-}$ and $^{+/m}$ mice

** Controls for Senescence-Accelerated Mice ("senescence-resistant" mice) were considered to be short-lived for the purposes of this analysis

Supplemental References

Baker, D. J., K. B. Jeganathan, J. D. Cameron, M. Thompson, S. Juneja *et al.*, 2004 BubR1 insufficiency causes early onset of aging-associated phenotypes and infertility in mice. Nature genetics **36:** 744-749.

Bartke, A., J. C. Wright, J. A. Mattison, D. K. Ingram, R. A. Miller *et al.*, 2001 Longevity: Extending the lifespan of long-lived mice. Nature **414:** 412-412.

Bartke, A., 2003 Can growth hormone (GH) accelerate aging? Evidence from GH-transgenic mice. Neuroendocrinology **78:** 210-216.

Blüher, M., B. B. Kahn and C. R. Kahn, 2003 Extended Longevity in Mice Lacking the Insulin Receptor in Adipose Tissue. Science **299:** 572-574.

Brown-Borg, H. M., K. E. Borg, C. J. Meliska and A. Bartke, 1996 Dwarf mice and the ageing process. Nature **384:** 33-33.

Chang, S., A. S. Multani, N. G. Cabrera, M. L. Naylor, P. Laud *et al.*, 2004 Essential role of limiting telomeres in the pathogenesis of Werner syndrome. Nat Genet **36:** 877-882.

Chiu, C.-H., W.-D. Lin, S.-Y. Huang and Y.-H. Lee, 2004 Effect of a C/EBP gene replacement on mitochondrial biogenesis in fat cells. Genes Dev **18:** 1970-1975.

Conde, C., S. Weller, S. Gilfillan, L. Marcellin, T. Martin *et al.*, 1998 Terminal Deoxynucleotidyl Transferase Deficiency Reduces the Incidence of Autoimmune Nephritis in (New Zealand Black x New Zealand White)F1 Mice. The Journal of Immunology **161:** 7023-7030.

Coschigano, K. T., A. N. Holland, M. E. Riders, E. O. List, A. Flyvbjerg *et al.*, 2003 Deletion, But Not Antagonism, of the Mouse Growth Hormone Receptor Results in Severely Decreased Body Weights, Insulin, and Insulin-Like Growth Factor I Levels and Increased Life Span. Endocrinology **144:** 3799-3810.

Flurkey, K., J. Papaconstantinou, R. A. Miller and D. E. Harrison, 2001 Lifespan extension and delayed immune and collagen aging in mutant mice with defects in growth hormone production. Proceedings of the National Academy of Sciences **98:** 6736-6741.

Holzenberger, M., J. Dupont, B. Ducos, P. Leneuve, A. Geloen *et al.*, 2003 IGF-1 receptor regulates lifespan and resistance to oxidative stress in mice. Nature **421:** 182-187.

Kuro-O, M., Y. Matsumura, H. Aizawa, H. Kawaguchi, T. Suga *et al.*, 1997 Mutation of the mouse klotho gene leads to a syndrome resembling ageing. Nature **390:** 45-51.

Kwan, K. Y., and J. C. Wang, 2001 Mice lacking DNA topoisomerase IIIβ develop to maturity but show a reduced mean lifespan. Proceedings of the National Academy of Sciences **98:** 5717-5721.

Migliaccio, E., M. Giorgio, S. Mele, G. Pelicci, P. Reboldi *et al.*, 1999 The p66shc adaptor protein controls oxidative stress response and life span in mammals. Nature **402:** 309-313.

Miskin, R., and T. Masos, 1997 Transgenic Mice Overexpressing Urokinase-Type Plasminogen Activator in the Brain Exhibit Reduced Food Consumption, Body Weight and Size, and Increased Longevity. The Journals of Gerontology Series A: Biological Sciences and Medical Sciences **52A:** B118-B124.

Mitsui, A., J. Hamuro, H. Nakamura, N. Kondo, Y. Hirabayashi *et al.*, 2002 Overexpression of human thioredoxin in transgenic mice controls oxidative stress and life span. Antioxid Redox Signal **4:** 693-696.

Moskovitz, J., S. Bar-Noy, W. M. Williams, J. Requena, B. S. Berlett *et al.*, 2001 Methionine sulfoxide reductase (MsrA) is a regulator of antioxidant defense and lifespan in mammals. Proceedings of the National Academy of Sciences **98:** 12920-12925.

Mounkes, L. C., S. Kozlov, L. Hernandez, T. Sullivan and C. L. Stewart, 2003 A progeroid syndrome in mice is caused by defects in A-type lamins. Nature **423:** 298-301.

Neumann, C. A., D. S. Krause, C. V. Carman, S. Das, D. P. Dubey *et al.*, 2003 Essential role for the peroxiredoxin Prdx1 in erythrocyte antioxidant defence and tumour suppression. Nature **424:** 561-565.

Schriner, S. E., N. J. Linford, G. M. Martin, P. Treuting, C. E. Ogburn *et al.*, 2005 Extension of murine life span by overexpression of catalase targeted to mitochondria. Science **308:** 1909-1911.

Takeda, T., M. Hosokawa, S. Takeshita, M. Irino, K. Higuchi *et al.*, 1981 A new murine model of accelerated senescence. Mechanisms of Ageing and Development **17:** 183-194.

Trifunovic, A., A. Wredenberg, M. Falkenberg, J. N. Spelbrink, A. T. Rovio *et al.*, 2004 Premature ageing in mice expressing defective mitochondrial DNA polymerase. Nature **429:** 417-423.

Tyner, S. D., S. Venkatachalam, J. Choi, S. Jones, N. Ghebranious *et al.*, 2002 p53 mutant mice that display early ageing-associated phenotypes. Nature **415:** 45-53.

Wong, K.-K., R. S. Maser, R. M. Bachoo, J. Menon, D. R. Carrasco *et al.*, 2003 Telomere dysfunction and Atm deficiency compromises organ homeostasis and accelerates ageing. Nature **421:** 643-648.

Yen, K., D. Steinsaltz and C. V. Mobbs, 2008 Validated analysis of mortality rates demonstrates distinct genetic mechanisms that influence lifespan. Experimental Gerontology **43:** 1044-1051.

Table S2. Tabular data for lines of inbred mice described in Figure 4.

	Female				Male			
Strain	Median lifespan (days)	n	ln(A)	G	Median lifespan (days)	n	ln(A)	G
129S1/SvImJ	770	32	-5.01	2.50	880	32	-7.45	3.36
A/J	640	32	-5.67	3.69	620	30	-4.67	3.15
BALB/cByJ	760	32	-6.38	3.48	710	32	-3.53	2.05
BTBR T+ tf/J	600	32	-4.26	3.05	560	32	-3.46	2.36
BUB/BnJ	620	24	-2.63	1.62	490	25	-2.26	1.37
C3H/HeJ	780	29	-7.07	3.94	710	32	-5.48	3.07
C57BL/10J	890	32	-4.00	1.79	780	27	-6.08	3.27
C57BL/6J	865	29	-5.50	2.50	900	32	-7.64	3.40
C57BLKS/J	850	32	-4.70	2.45	825	32	-7.42	3.55
C57BR/cdJ	880	32	-6.12	2.96	850	32	-6.76	3.24
C57L/J	720	32	-12.75	7.24	730	32	-9.21	5.30
CAST/EiJ	670	17	-5.06	3.09				
CBA/J	640	30	-3.77	2.40	650	32	-4.00	2.46
DBA/2J	640	32	-2.67	1.76	705	32	-4.43	2.76
FVB/NJ	760	29	-3.75	1.88	590	25	-2.97	1.84
KK/HlJ	590	32	-4.91	3.51	620	32	-3.71	2.38
LP/J	810	32	-4.93	2.37	810	32	-5.84	2.91
MOLF	680	32	-3.94	2.28	630	32	-6.05	4.26
MRL/MpJ	550	31	-4.89	3.81				
NOD.B10Sn-H2 /J	660	32	-5.06	3.16	700	31	-3.58	2.01
NON/ShiLtJ	750	31	-6.27	3.50	840	32	-8.22	3.99
NZO/HlLtJ	560	32	-2.79	2.03	415	31	-1.81	1.57
NZW/LacJ	730	32	-4.91	2.68	770	32	-3.68	1.69
P/J	680	32	-3.94	2.29				
PL/J	410	32	-2.57	2.40	460	32	-2.97	2.72
PWD/PhJ	850	32	-5.70	2.77	815	27	-4.59	2.36
RIIIS/J	790	32	-7.75	3.97	830	32	-9.12	4.29
SJL/J	450	30	-2.36	2.01	510	16	-4.28	3.60
SM/J	750	32	-5.22	2.86	780	31	-6.56	3.59
SWR/J	635	31	-3.20	1.92	710	32	-2.70	1.49
WSB/EiJ	950	32	-3.56	1.39	900	32	-2.89	1.17

Table S3. Tabular data for ad libitum-fed recombinant inbred mice described in Figure S3.

	Male				Female			
Line Number	Median lifespan (days)	n	ln(A)	G	Median lifespan (days)	n	ln(A)	G
3	700	5	-0.26	0.00	1045	5	-4.47	1.86
7	1048	5	-24.58	9.39	1143	5	-6.81	2.58
13	696	5	-4.26	2.36	863	5	-16.45	7.65
14	946	5	-3.92	1.86				
16	758	6	-5.28	3.12	687	5	-2.18	1.37
19	981	5	-8.38	3.66	938	5	-8.41	3.87
22	1168	5	-14.64	5.11				
23	1063	11	-5.35	2.07	991	15	-7.74	3.13
24	829	5	-4.31	2.30	1023	5	-3.05	1.22
25	979	5	-7.99	3.47	979	5	-44.12	17.45
26	1043	5	-31.75	11.98	1049	5	-34.42	12.90
28					716	5	-8.90	5.31
41	787	5	-27.82	14.08	986	5	-6.83	3.10
46					1147	5	-8.91	3.36
48	751	5	-4.10	2.22	898	5	-3.20	1.51
49	892	4	-4.24	2.08	712	5	-7.85	4.79
50	459	5	-2.07	2.04	291	5	-1.42	2.41
51	1018	5	-6.89	2.91	820	5	-5.14	2.63
52	713	5	-2.59	1.74				
56	1277	5	-3.72	1.26	1145	5	-5.52	2.02
60					1073	5	-1.73	0.62
62	1055	5	-16.48	6.26				
66	685	5	-2.56	1.60	743	5	-3.01	1.79
79	1094	5	-13.54	4.98				
80	803	5	-7.75	4.09	887	5	-11.46	5.22
84	900	5	-8.43	4.01	810	5	-11.70	5.96
86	1111	5	-5.91	2.34	926	4	-7.43	3.33
89	951	5	-2.55	1.19	889	5	-2.74	1.48
90	829	5	-9.86	4.99	906	5	-35.11	15.19
92	994	5	-6.79	2.79	798	5	-3.66	1.82
94	718	5	-3.41	1.69	699	5	-5.99	3.84
97	991	5		37.24	999	5		33.35
98	1187	5	-7.32	2.58	1249	5	-9.22	3.07
99	930	5	-9.01	4.00	887	5	-3.25	1.63
100	895	5	-34.14	14.85	924	5	-35.15	15.11
103	823	5	-23.27	11.28	855	5	-8.81	4.23
107	1079	5	-13.01	4.83	596	5	-2.88	1.93
110	587	5	-5.45	4.20	851	5	-3.47	1.76
112	946.5	4	-3.24	1.42	600	5	-3.70	2.49
114	1013	5	-13.17	5.42	911	5	-4.85	2.20
115	992	5	-7.43	3.22	746	5	-4.49	2.70
117	893	5	-4.53	2.07	641	5	-3.41	2.30
122	859	5	-9.22	4.42	1010	5	-2.68	1.12
123	915	5	-5.46	2.51	962	5	-23.16	9.62

Line numbers correspond to those from source publication (Liao *et al.* 2010).

Table S4. Source publications and tabular data for *daf-2*, *isp-1* and *eat-2* mutant lines described in Figure 7.

Source Publication	n		Median Lifespan (days)		Fractional contribution to lifespan increase		
	N2	mutant	N2	mutant	*A*	*G*	*S*
daf-2 (e1370)							
(Kaeberlein *et al.* 2006)	58	41	19	37.5	0.40123	0.49956	0.09921
(Tullet *et al.* 2008)	90	88	21	39	0.436832	0.552616	-0.01055
(Troemel *et al.* 2006)	90	88	16	39	-0.10334	0.896655	0
(Yang and Hekimi 2010a)	400	150	18	44	0.121681	0.812151	-0.06617
(Wang *et al.* 2008)	80	80	19	34	-0.23511	0.75992	0.004969
(Mehta *et al.* 2009)	40	40	22	29	-0.241	0.577434	0.181565
(Tissenbaum and Guarente 2001)	142	293	18	45	-0.02147	0.906075	-0.07245
(Hsu *et al.* 2003)	48	41	20	45	-0.20419	0.790542	-0.00526
(Lin *et al.* 2001)	348	140	19	46	-0.1727	0.821857	-0.00544
(Garsin *et al.* 2003)	75	75	16	31	-0.05878	0.941221	0
(Huang *et al.* 2004)	180	114	16	36	-0.191	0.775917	-0.03308
(Dorman *et al.* 1995)	48	37	17	38.5	-0.06963	0.905061	0.025306
(Apfeld and Kenyon 1999)	100	100	18	53	-0.19923	0.787241	-0.01353
(Berdichevsky *et al.* 2006)	70	100	19	40	0.74393	0.23844	0.01763
(Kenyon *et al.* 1993)	49	42	19.5	44	-0.14888	0.764857	-0.08626
(Oh *et al.* 2005)	125	46	16	42	0.256288	0.608954	-0.13476
(Van Raamsdonk *et al.* 2010)	100	100	22	38	-0.07721	0.814284	0.108506
(Van Raamsdonk and Hekimi 2009)	309	168	19	66	-0.15918	0.814043	-0.02678
isp-1 (qm150)							
(Feng *et al.* 2001)	380	283	22	34	-0.25783	0.733226	0.008946
(Curtis *et al.* 2006)	157	45	21	28	0.310793	-0.16912	0.520091
(Van Raamsdonk and Hekimi 2009)	309	114	19	43	-0.20072	0.778142	-0.02114
(Dingley *et al.* 2009)	38	37	14	21	-0.40716	0.586179	-0.00666
(Yang and Hekimi 2010b)	150	150	22	35	-0.33977	0.638542	-0.02169
(Lee *et al.* 2010)	107	96	15	27	0.867935	0.098988	-0.03308
(Yang and Hekimi 2010a)	400	200	21	35	-0.17697	0.816204	-0.00682
(Baruah *et al.* 2014)	112	117	22	32	-0.39118	0.608112	-0.00071
(Yee *et al.* 2014)	250	200	18	34	-0.22033	0.732193	-0.04748
(Hsu *et al.* 2003)	48	35	20	27	-0.28045	0.707574	-0.01197
(Torgovnick *et al.* 2010)	238	162	16	25	0.330464	0.589386	-0.08015
(Bennett *et al.* 2014)	365	206	20	26	0.245278	0.189918	0.564804
(Mouchiroud *et al.* 2011)	55	53	13	20	-0.17964	0.27395	0.546407

eat-2 (ad1116)							
(Panowski *et al.* 2007)	70	48	18	26	-0.27081	0.634363	-0.09483
(Park *et al.* 2010)	60	60	20	26	-0.29919	0.593557	-0.10725
(Lakowski and Hekimi 1998)	50	36	20	29	-0.05582	0.623448	0.320736
(Van Raamsdonk and Hekimi 2009)	240	240	18	35	0.475685	0.361144	0.163172
(Carrano *et al.* 2009)	67	74	23	30	-0.27415	0.725852	0
(Seo *et al.* 2015)	75	75	17	24.5	0.896784	-0.0909	0.012314
(Hsu *et al.* 2003)	48	41	21	29	0.967482	-0.03252	0
(Greer and Brunet 2009)	86	84	23	29	-0.19653	0.748508	-0.05496
(Schreiber *et al.* 2010)	100	71	18	20	-0.32004	0.599089	0.080875
(Ching *et al.* 2011)	63	70	15	23	-0.31604	0.676153	-0.00781
(Schleit *et al.* 2011)	181	139	18	24	0.753534	-0.07513	-0.17133
(Yuan *et al.* 2012)	65	60	20.5	32	-0.01853	0.981473	0
(Gaglia *et al.* 2012)	75	94	16	24	-0.27517	0.703776	-0.02105
(Rousakis *et al.* 2013)	110	160	19	22	0.318461	0.597187	-0.08435
(Thondamal *et al.* 2014)	102	108	18	22	0.404702	-0.24621	0.349087
(Yee *et al.* 2014)	150	150	19	31	-0.19329	0.748434	0.058274
(Zimmerman and Kim 2014)	61	83	17.5	20	-0.1679	0.570557	-0.26155
(Chin *et al.* 2014)	100	59	15	22	-0.24939	0.721649	-0.02897
(Asthana *et al.* 2015)	217	189	18	22.5	-0.35872	0.641277	0
(Hine *et al.* 2015)	100	100	20	29	0.589966	0.244302	0.165732
(Seo *et al.* 2016)	101	81	19.5	31	0.574392	0.385386	-0.04022
(Singh *et al.* 2016)	93	88	21	32	0.154243	0.733848	-0.11191
(Merkwirth *et al.* 2016)	101	63	19	26	-0.26723	0.693701	-0.03907

Supplemental References

Apfeld, J., and C. Kenyon, 1999 Regulation of lifespan by sensory perception in Caenorhabditis elegans. Nature **402:** 804-809.

Asthana, J., D. Yadav, A. Pant, A. K. Yadav, M. M. Gupta *et al.*, 2015 Acacetin 7-O-α-l-rhamnopyranosyl (1–2) β-D-xylopyranoside Elicits Life-span Extension and Stress Resistance in Caenorhabditis elegans. The Journals of Gerontology Series A: Biological Sciences and Medical Sciences.

Baruah, A., H. Chang, M. Hall, J. Yuan, S. Gordon *et al.*, 2014 CEP-1, the Caenorhabditis elegans p53 Homolog, Mediates Opposing Longevity Outcomes in Mitochondrial Electron Transport Chain Mutants. PLoS Genet **10:** e1004097.

Bennett, C. F., H. Vander Wende, M. Simko, S. Klum, S. Barfield *et al.*, 2014 Activation of the mitochondrial unfolded protein response does not predict longevity in Caenorhabditis elegans. Nat Commun **5**.

Berdichevsky, A., M. Viswanathan, H. R. Horvitz and L. Guarente, 2006 C. elegans SIR-2.1 Interacts with 14-3-3 Proteins to Activate DAF-16 and Extend Life Span. Cell **125:** 1165-1177.

Carrano, A. C., Z. Liu, A. Dillin and T. Hunter, 2009 A conserved ubiquitination pathway determines longevity in response to diet restriction. Nature **460:** 396-399.

Chin, R. M., X. Fu, M. Y. Pai, L. Vergnes, H. Hwang *et al.*, 2014 The metabolite α-ketoglutarate extends lifespan by inhibiting ATP synthase and TOR. Nature **510:** 397-401.

Ching, T.-T., W.-C. Chiang, C.-S. Chen and A.-L. Hsu, 2011 Celecoxib extends C. elegans lifespan via inhibition of insulin-like signaling but not cyclooxygenase-2 activity. Aging Cell **10:** 506-519.

Curtis, R., G. O'connor and P. S. Distefano, 2006 Aging networks in Caenorhabditis elegans: AMP-activated protein kinase (aak-2) links multiple aging and metabolism pathways. Aging Cell **5:** 119-126.

Dingley, S., E. Polyak, R. Lightfoot, J. Ostrovsky, M. Rao *et al.*, 2009 Mitochondrial respiratory chain dysfunction variably increases oxidant stress in Caenorhabditis elegans. Mitochondrion.

Dorman, J. B., B. Albinder, T. Shroyer and C. Kenyon, 1995 The age-1 and daf-2 genes function in a common pathway to control the lifespan of Caenorhabditis elegans. Genetics **141:** 1399-1406.

Feng, J. L., F. Bussiere and S. Hekimi, 2001 Mitochondrial electron transport is a key determinant of life span in Caenorhabditis elegans. Developmental Cell **1:** 633-644.

Gaglia, M. M., D.-E. Jeong, E.-A. Ryu, D. Lee, C. Kenyon *et al.*, 2012 Genes That Act Downstream of Sensory Neurons to Influence Longevity, Dauer Formation, and Pathogen Responses in *Caenorhabditis elegans*. PLoS Genet **8:** e1003133.

Garsin, D. A., J. M. Villanueva, J. Begun, D. H. Kim, C. D. Sifri *et al.*, 2003 Long-lived C. elegans daf-2 mutants are resistant to bacterial pathogens. Science **300:** 1921.

Greer, E. L., and A. Brunet, 2009 Different dietary restriction regimens extend lifespan by both independent and overlapping genetic pathways in C. elegans. Aging Cell **8:** 113-127.

Hine, C., E. Harputlugil, Y. Zhang, C. Ruckenstuhl, Byung c. Lee *et al.*, 2015 Endogenous Hydrogen Sulfide Production Is Essential for Dietary Restriction Benefits. Cell **160:** 132-144.

Hsu, A.-L., C. T. Murphy and C. Kenyon, 2003 Regulation of Aging and Age-Related Disease by DAF-16 and Heat-Shock Factor. Science **300:** 1142-1145.

Huang, C., C. J. Xiong and K. Kornfeld, 2004 Measurements of age-related changes of physiological processes that predict lifespan of Caenorhabditis elegans. Proceedings of the National Academy of Sciences of the United States of America **101:** 8084-8089.

Kaeberlein, T. L., E. D. Smith, M. Tsuchiya, K. L. Welton, J. H. Thomas *et al.*, 2006 Lifespan extension in Caenorhabditis elegans by complete removal of food. Aging Cell **5:** 487-494.

Kenyon, C., J. Chang, E. Gensch, A. Rudner and R. Tabtiang, 1993 A C. elegans mutant that lives twice as long as wild type. Nature **366:** 461-464.

Lakowski, B., and S. Hekimi, 1998 The genetics of caloric restriction in Caenorhabditis elegans. Proc Natl Acad Sci U S A **95:** 13091-13096.

Lee, S. J., A. B. Hwang and C. Kenyon, 2010 Inhibition of Respiration Extends C. elegans Life Span via Reactive Oxygen Species that Increase HIF-1 Activity. Curr Biol **20:** 2131-2136.

Lin, K., H. Hsin, N. Libina and C. Kenyon, 2001 Regulation of the Caenorhabditis elegans longevity protein DAF-16 by insulin/IGF-1 and germline signaling. Nat Genet **28:** 139-145.

Mehta, R., K. A. Steinkraus, G. L. Sutphin, F. J. Ramos, L. S. Shamieh *et al.*, 2009 Proteasomal Regulation of the Hypoxic Response Modulates Aging in C. elegans. Science **324:** 1196-1198.

Merkwirth, C., V. Jovaisaite, J. Durieux, O. Matilainen, Sabine d. Jordan *et al.*, 2016 Two Conserved Histone Demethylases Regulate Mitochondrial Stress-Induced Longevity. Cell **165:** 1209-1223.

Mouchiroud, L., L. Molin, P. Kasturi, M. N. Triba, M. E. Dumas *et al.*, 2011 Pyruvate imbalance mediates metabolic reprogramming and mimics lifespan extension by dietary restriction in Caenorhabditis elegans. Aging Cell **10:** 39-54.

Oh, S. W., A. Mukhopadhyay, N. Svrzikapa, F. Jiang, R. J. Davis *et al.*, 2005 JNK regulates lifespan in Caenorhabditis elegans by modulating nuclear translocation of forkhead transcription factor/DAF-16. Proceedings of the National Academy of Sciences of the United States of America **102:** 4494-4499.

Panowski, S. H., S. Wolff, H. Aguilaniu, J. Durieux and A. Dillin, 2007 PHA-4/Foxa mediates diet-restriction-induced longevity of C. elegans. Nature **447:** 550-555.

Park, S.-K., C. D. Link and T. E. Johnson, 2010 Life-span extension by dietary restriction is mediated by NLP-7 signaling and coelomocyte endocytosis in C. elegans. The FASEB Journal **24:** 383-392.

Rousakis, A., A. Vlassis, A. Vlanti, S. Patera, G. Thireos *et al.*, 2013 The general control nonderepressible-2 kinase mediates stress response and longevity induced by target of rapamycin inactivation in Caenorhabditis elegans. Aging Cell **12:** 742-751.

Schleit, J., V. Z. Wall, M. Simko and M. Kaeberlein, 2011 The MDT-15 Subunit of Mediator Interacts with Dietary Restriction to Modulate Longevity and Fluoranthene Toxicity in *Caenorhabditis elegans*. PLoS ONE **6:** e28036.

Schreiber, M. A., J. T. Pierce-Shimomura, S. Chan, D. Parry and S. L. Mcintire, 2010 Manipulation of Behavioral Decline in *Caenorhabditis elegans* with the Rag GTPase *raga-1*. PLoS Genet **6:** e1000972.

Seo, M., K. Seo, W. Hwang, H. J. Koo, J.-H. Hahm *et al.*, 2015 RNA helicase HEL-1 promotes longevity by specifically activating DAF-16/FOXO transcription factor signaling in Caenorhabditis elegans. Proceedings of the National Academy of Sciences **112:** E4246-E4255.

Seo, M., S. Park, H. G. Nam and S.-J. V. Lee, 2016 RNA helicase SACY-1 is required for the longevity caused by various genetic perturbations in Caenorhabditis elegans. Cell Cycle**:** 00-00.

Singh, A., N. Kumar, L. Matai, V. Jain, A. Garg *et al.*, 2016 A chromatin modifier integrates insulin/IGF-1 signalling and dietary restriction to regulate longevity. Aging Cell **15:** 694-705.

Thondamal, M., M. Witting, P. Schmitt-Kopplin and H. Aguilaniu, 2014 Steroid hormone signalling links reproduction to lifespan in dietary-restricted Caenorhabditis elegans. Nat Commun **5**.

Tissenbaum, H. A., and L. Guarente, 2001 Increased dosage of a sir-2 gene extends lifespan in Caenorhabditis elegans. Nature **410:** 227-230.

Torgovnick, A., A. Schiavi, R. Testi and N. Ventura, 2010 A role for p53 in mitochondrial stress response control of longevity in C. elegans. Experimental Gerontology **45:** 550-557.

Troemel, E. R., S. W. Chu, V. Reinke, S. S. Lee, F. M. Ausubel *et al.*, 2006 p38 MAPK Regulates Expression of Immune Response Genes and Contributes to Longevity in *C. elegans*. PLoS Genet **2:** e183.

Tullet, J. M. A., M. Hertweck, J. H. An, J. Baker, J. Y. Hwang *et al.*, 2008 Direct Inhibition of the Longevity-Promoting Factor SKN-1 by Insulin-like Signaling in C. elegans. Cell **132:** 1025-1038.

Van Raamsdonk, J. M., and S. Hekimi, 2009 Deletion of the mitochondrial superoxide dismutase sod-2 extends lifespan in Caenorhabditis elegans. PLoS Genet **5:** e1000361.

Van Raamsdonk, J. M., Y. Meng, D. Camp, W. Yang, X. Jia *et al.*, 2010 Decreased energy metabolism extends life span in Caenorhabditis elegans without reducing oxidative damage. Genetics **185:** 559-571.

Wang, M. C., E. J. O'rourke and G. Ruvkun, 2008 Fat Metabolism Links Germline Stem Cells and Longevity in C. elegans. Science **322:** 957-960.

Yang, W., and S. Hekimi, 2010a A mitochondrial superoxide signal triggers increased longevity in Caenorhabditis elegans. PLoS Biol **8:** e1000556.

Yang, W., and S. Hekimi, 2010b Two modes of mitochondrial dysfunction lead independently to lifespan extension in Caenorhabditis elegans. Aging Cell **9:** 433-447.

Yee, C., W. Yang and S. Hekimi, 2014 The Intrinsic Apoptosis Pathway Mediates the Pro-Longevity Response to Mitochondrial ROS in C. elegans. Cell **157:** 897-909.

Yuan, Y., C. S. Kadiyala, T.-T. Ching, P. Hakimi, S. Saha *et al.*, 2012 Enhanced Energy Metabolism Contributes to the Extended Life Span of Calorie-restricted Caenorhabditis elegans. Journal of Biological Chemistry **287:** 31414-31426.

Zimmerman, S. M., and S. K. Kim, 2014 The GATA transcription factor/MTA-1 homolog egr-1 promotes longevity and stress resistance in Caenorhabditis elegans. Aging Cell **13:** 329-339.

SUPPORTING INFORMATION

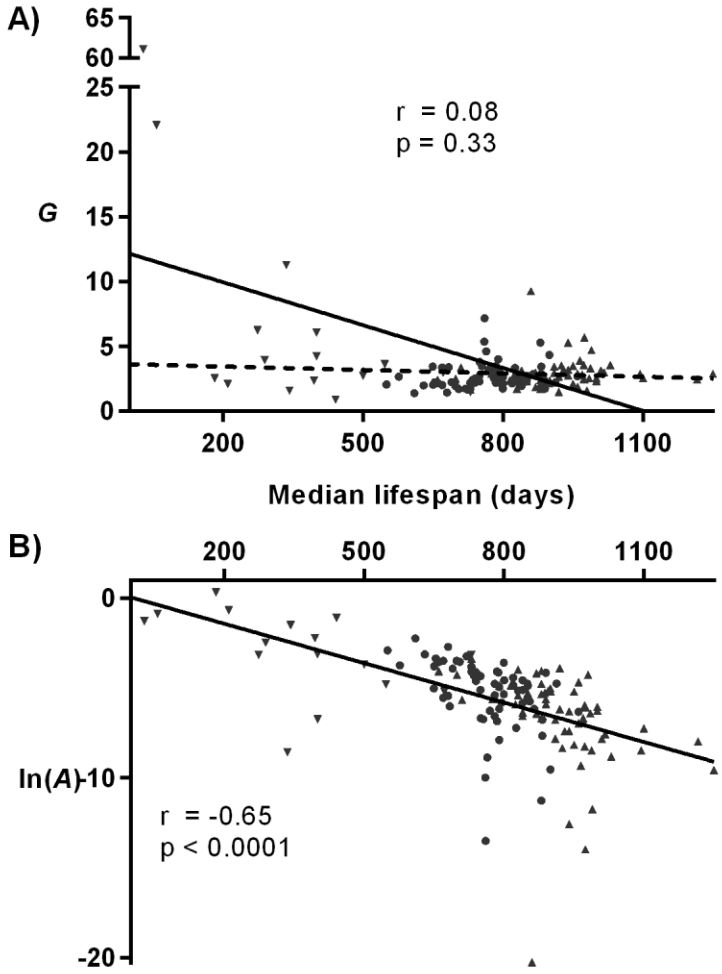


Figure S1. Maximum likelihood (MLE) estimations of the Gompertz parameter **(A)** '*G*' and **(B)** '*A*', expressed as its natural logarithm, plotted against median lifespan for lines of mice with varying lifespans, including both studies 1 and 2. Lines of best fit, Spearman correlation coefficients (r) and associated p values are shown. Note that the line of best fit (determined by linear regression) is shown as an aid for the reader, and the p-values shown were determined separately by non-parametric methods. Thus, the slope of the line of best fit in panel A is unduly influenced by the presence of a small number of abnormally high parameter values for the

shortest-lived strains of mice. In contrast, the rank-based non-parametric Spearman correlation coefficient is less sensitive to outliers, and hence better reflects the orientation of the majority of points. The dashed line in panel (A) shows the line of best fit with the two highest '*G*' values excluded.

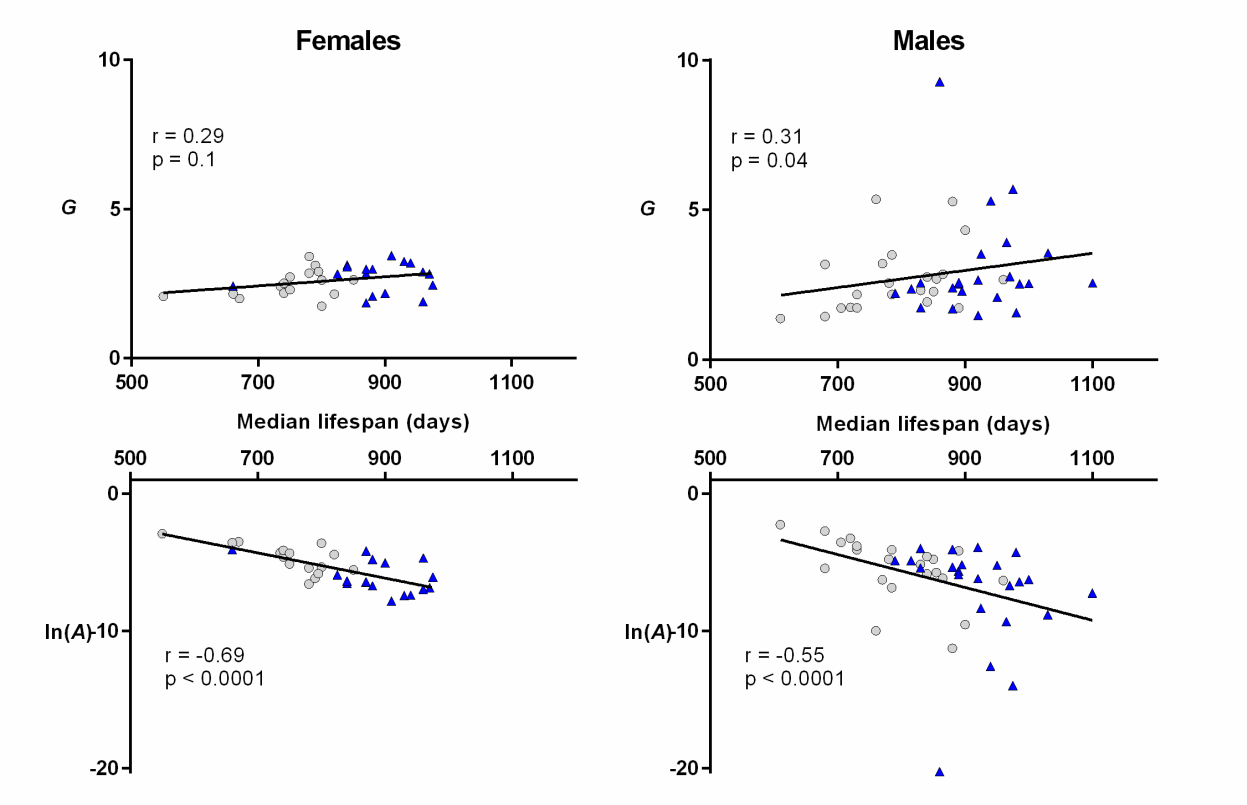


Figure S2. Maximum likelihood (MLE) estimations of the Gompertz parameters '*G*' and '*A*', plotted against median lifespan for lines of mice with varying lifespans, separated by sex (Set 2). Long-lived strains are represented by blue triangles and normal-lived (control) strains by grey circles. Lines of best fit, Spearman correlation coefficients (r) and associated p values are shown. Note that the line of best fit (determined by linear regression) is shown as an aid for the reader, and the p-values shown were determined separately by non-parametric methods.

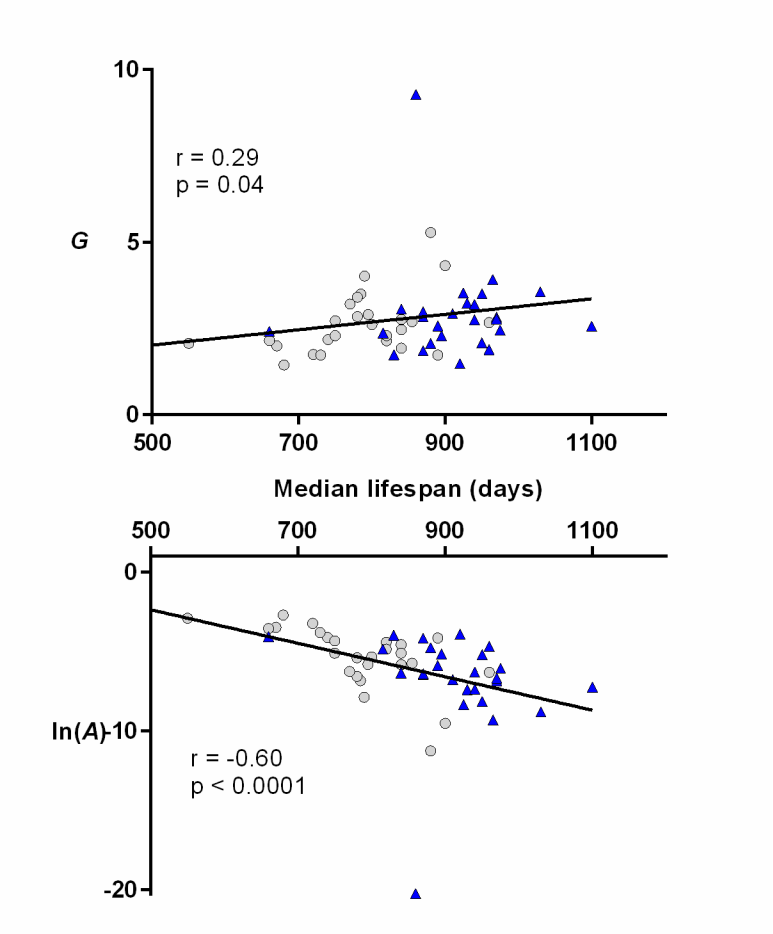


Figure S3. Maximum likelihood (MLE) estimations of the Gompertz parameters '*G*' and '*A*', plotted against median lifespan for lines of mice with varying lifespans, for mice in the C57BL6/J genetic background only (from Set 2). Long-lived strains are represented by blue triangles and normal-lived (control) strains by grey circles. Lines of best fit, Spearman correlation coefficients (r) and associated p values are shown. Note that the line of best fit (determined by linear regression) is shown as an aid for the reader, and the p-values shown were determined separately by non-parametric methods.

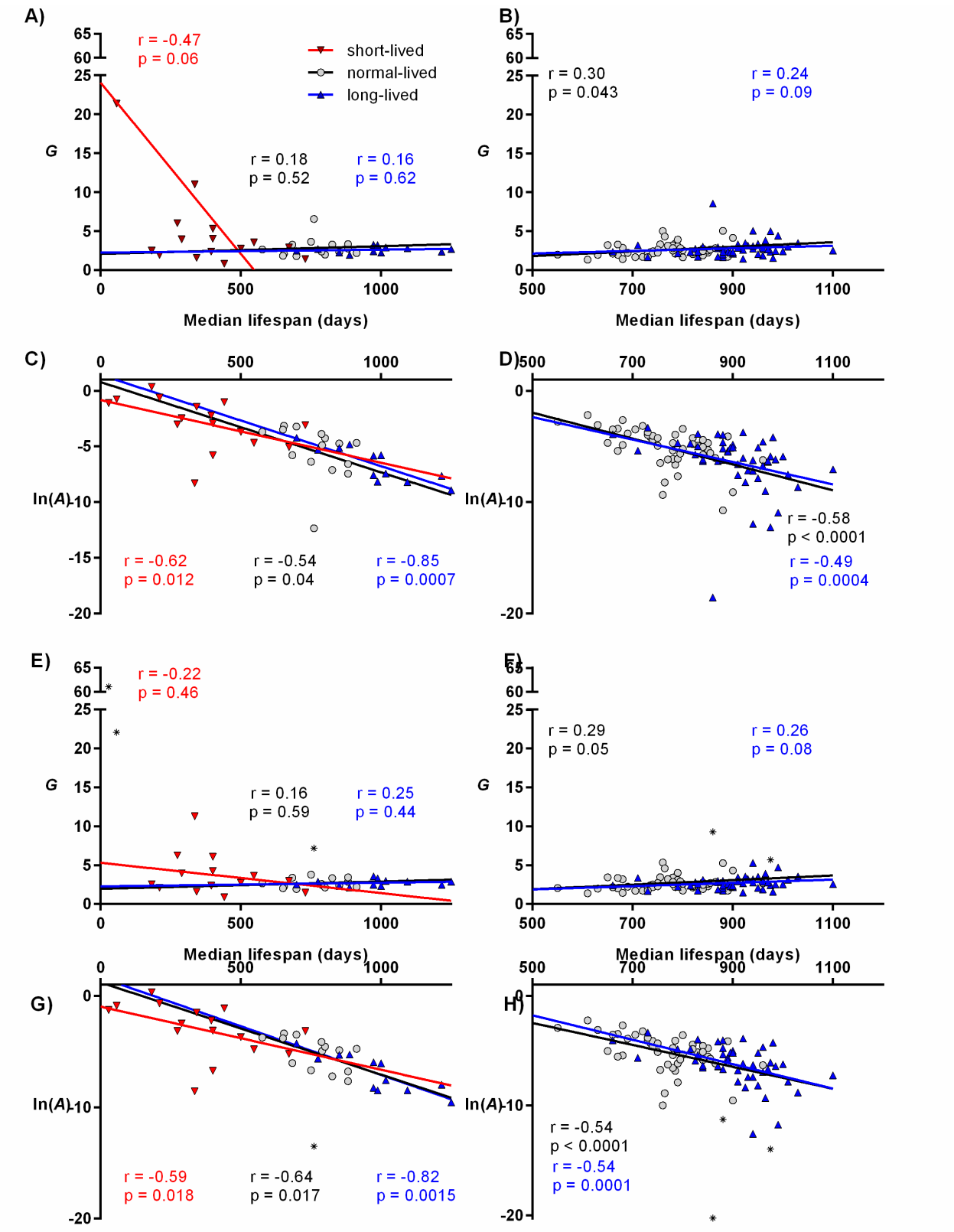

A)
r = -0.47
p = 0.06
short-lived
normal-lived
long-lived
r = 0.18
p = 0.52
r = 0.16
p = 0.62
G
Median lifespan (days)
B)
r = 0.30
p = 0.043
r = 0.24
p = 0.09
G
Median lifespan (days)
C)
ln(A)
r = -0.62
p = 0.012
r = -0.54
p = 0.04
r = -0.85
p = 0.0007
D)
ln(A)
r = -0.58
p < 0.0001
r = -0.49
p = 0.0004
E)
r = -0.22
p = 0.46
G
r = 0.16
p = 0.59
r = 0.25
p = 0.44
Median lifespan (days)
F)
r = 0.29
p = 0.05
r = 0.26
p = 0.08
G
Median lifespan (days)
G)
ln(A)
r = -0.59
p = 0.018
r = -0.64
p = 0.017
r = -0.82
p = 0.0015
H)
ln(A)
r = -0.54
p < 0.0001
r = -0.54
p = 0.0001

Figure S4 Correction for maximum likelihood parameter estimation error and possible bias due to outliers, corresponding to Figure 2. Panels **(A)** through **(D)** show relationship between Gompertz parameters and median lifespan after correction for estimation error (as described below). As previously described (Promislow *et al.* 1999), MLE consistently overestimated *'G'* and underestimated ln(*A*). However, the magnitude of these estimation errors (as median (interquartile range), 0.087 (0.050 to 0.16), -0.16 (-0.29 to -0.094) for *'G'* and ln(*A*) respectively) did not appreciably alter the parameter-lifespan relationship. Panels **(E)** through **(H)** show relationship between Gompertz parameters and median lifespan in the absence of potential outliers (shown as asterisks, and excluded from statistical analysis). Outliers were identified using the robust regression and outlier removal (ROUT) method (with Q set to the recommended value of 1%, corresponding to a 1% false discovery rate) in Prism (Motulsky and Brown 2006).

We determined parameter estimation errors via a resampling engine that we wrote in Visual Basic for Applications 6.5 (Microsoft). This produced lifespans from a simulated cohort of a theoretical population with a Gompertz distribution of known *'G'* and *'A'* values.

The inverse of the Gompertz cumulative hazard function (Conover *et al.* 2010) in the form of the following function was used to generate survival times for populations adhering to the Gompertz model:

(1/G)*ln((G/A)*-ln(rand())+1)

The effectiveness of this algorithm was tested by modeling a hypothetical Gompertzian population with an *'A'* of 0.001 (ln(*A*) of -6.908) and a *'G'* of 0.1, with a sample size of 10000. This generated data in a Gompertz distribution yielding MLE parameter estimates of $A = 0.00098$, $G = 0.10044$.

In order to account for potential bias in parameter estimation we predicted estimation error for each survival experiment included in our analysis, and used this to determine corrected values. First, we generated subsamples from a Gompertzian population with the same parameters as for the original survival experiment. For each experiment, 1000 subpopulations were generated, each consisting of a number of subsamples corresponding to the original sample size. Parameter values were estimated from these using MLE, and the median difference between these subsample estimates and the estimates from the survival experiment were used to determine estimation error. This was then subtracted from the original parameter values to produce corrected values. Note that, while this accounts for systematic estimation error due to sample size and location in the parameter space, it cannot account for random variation due to sample size alone.

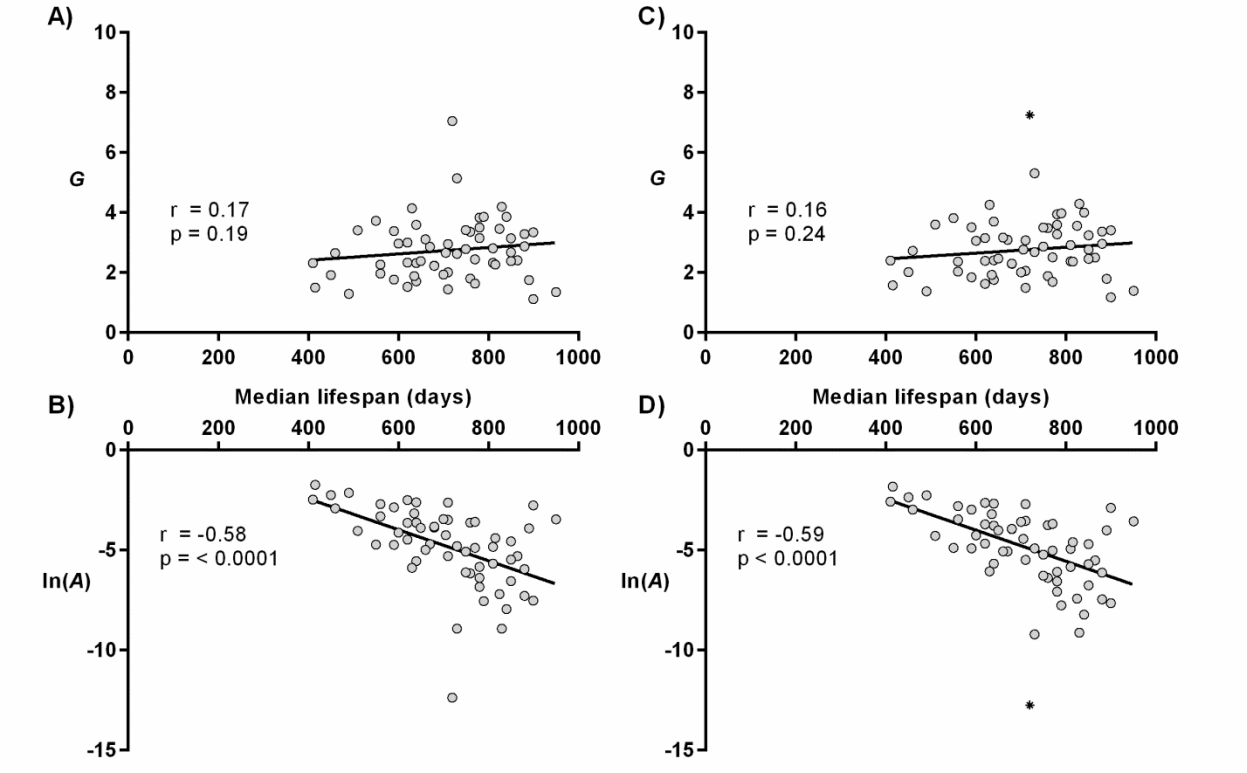


Figure S5 Correction for maximum likelihood parameter estimation error and possible bias due to outliers, corresponding to Figure 4 (inbred mice from Jackson labs Mouse Phenome Database). Panels **(A)** and **(B)** show relationship between Gompertz parameters and median lifespan after correction for estimation error as described for Figure S1. MLE consistently overestimated *'G'* and underestimated ln(*A*). However, the magnitude of these estimation errors (as median (interquartile range), 0.086 (0.064 to 0.11), -0.14 (-0.20 to -0.10) for *'G'* and ln(*A*) respectively) did not appreciably alter the parameter-lifespan relationship. Panels **(C)** and **(D)** show relationship between Gompertz parameters and median lifespan in the absence of potential outliers (shown as asterisks, and excluded from statistical analysis). Outliers were identified using the robust regression and outlier removal (ROUT) method (with Q set to the recommended value of 1%, corresponding to a 1% false discovery rate) in Prism (Motulsky and Brown 2006).

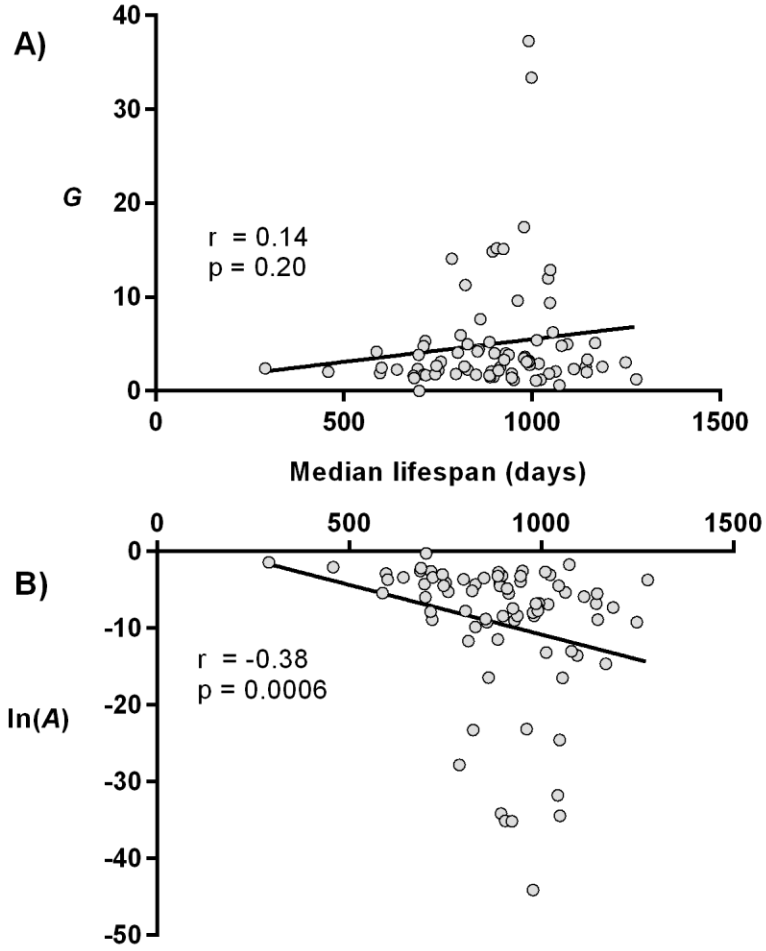


Figure S6 Nonlinear regression estimations of the Gompertz parameter values plotted against median lifespan for ad libitum-fed recombinant inbred strains (Table S3) (Liao *et al.* 2010). Lines of best fit, Spearman correlation coefficients (r) and associated p values are shown. **(A)** Gompertz parameter *'G'*, acceleration of mortality rates with age. **(B)** Natural logarithm of *'A'*, baseline mortality. Note that while the line of best fit (determined by linear regression) is shown as an aid for the reader, the p-values shown were determined by non-parametric methods.

Nonlinear regression was used rather than MLE as we found it to be more robust for small sample sizes, for which MLE often failed to converge on a parameter estimate. To confirm that non-linear regression could reliably estimate parameters from a sample size of five we used

the resampling tool described in the legend for Figure S2 to generate 250 groups of 5 simulated animals for a range of *'G'* and *'A'* parameter values encompassing the typical parameter space for mice (ln(*A*), *'G'* values of -2,1; -3, 1.5; -4, 2; -5, 2.75; -6, 3.5; -7, 4; -8,4.5). We estimated Gompertz parameters from these simulated groups via non-linear regression in Prism to estimate parameter values. Estimates for *'A'* and *'G'* were strongly correlated with their starting values ($r \geq 0.98$, $p < 0.0001$ for both parameters). Thus a sample size of 5 is sufficient to obtain meaningful estimates of the Gompertz parameters by NLR.

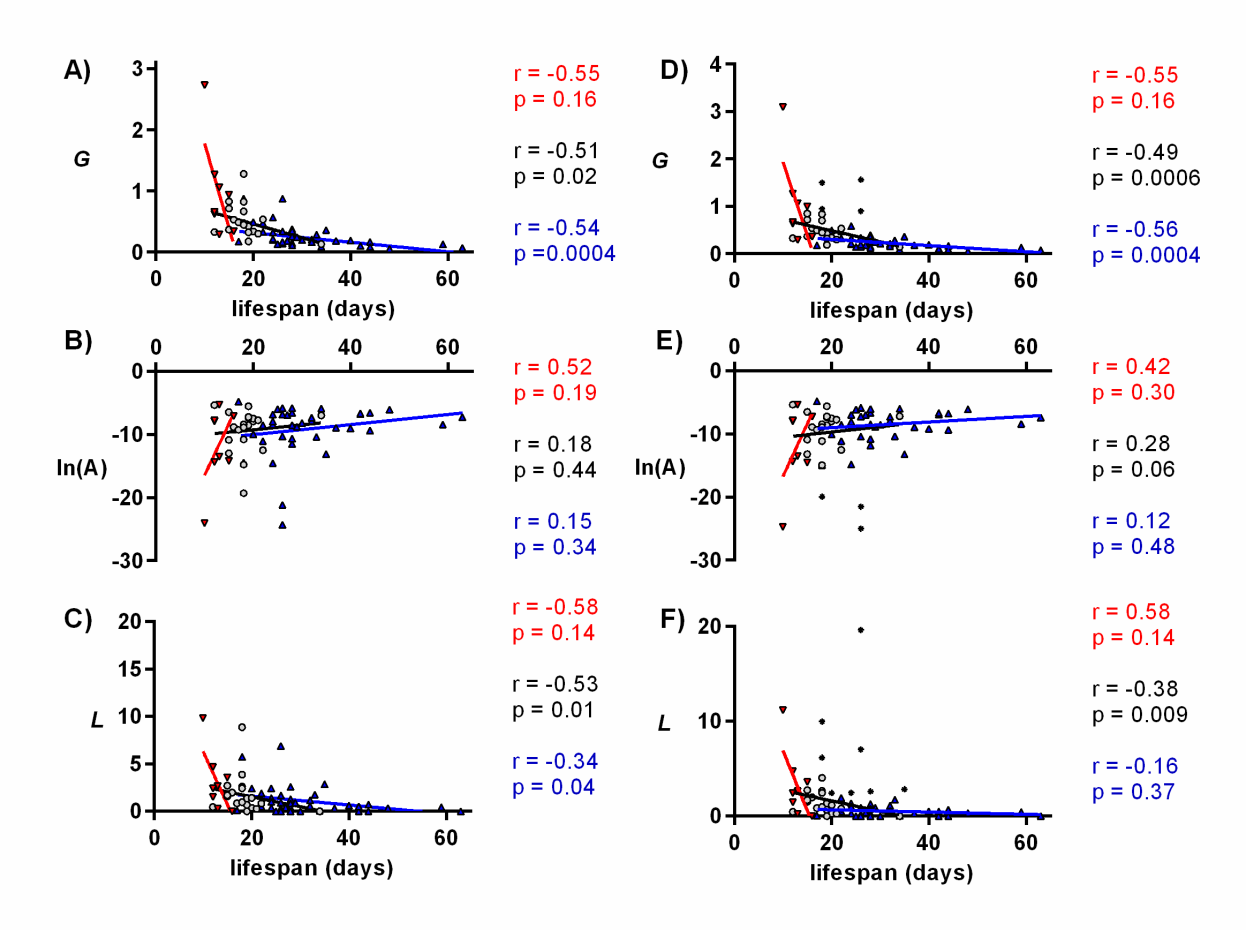


Figure S7 Correction for maximum likelihood parameter estimation error and possible bias due to outliers, corresponding to Figure 4. Panels **(A)** through **(C)** show relationship between logistic parameters and median lifespan after correction for estimation error as described in the legend for Figure S2 and below. MLE tended to overestimate *'G'* and underestimate ln(*A*) and *'L'*. However, the magnitude of these estimation errors (as median (interquartile range), 0.0021 (-0.00014 to 0.0073), -0.044 (-0.11 to -0.015), -0.015 (-0.035 to 0.0012) for *'G'*, ln(*A*) and *'L'* respectively) did not appreciably alter the parameter-lifespan relationship. Panels **(D)** through **(F)** show relationship between Gompertz parameters and median lifespan in the absence of potential outliers (shown as asterisks, and excluded from statistical analysis). Outliers were identified

using the robust regression and outlier removal (ROUT) method (with Q set to the recommended value of 1%, corresponding to a 1% false discovery rate) in Prism (Motulsky and Brown 2006).

The following function representing the inverse of the logistic cumulative hazard function (Conover *et al.* 2010) was used to simulate populations following the logistic model:

(ln(-(G*(1-(L*A)/G-(1-(1-rand())^(-L)))/(L*A)))/G

To confirm that this yielded samples following a logistic distribution, we modelled a hypothetical logistic population with an *'A'* of 0.000045 (ln(*A*) of -10), *'G'* of 0.5 and *'L'* of 1. This yielded MLE parameter estimations of 0.0000448, 0.4999 and 1.008425 respectively.

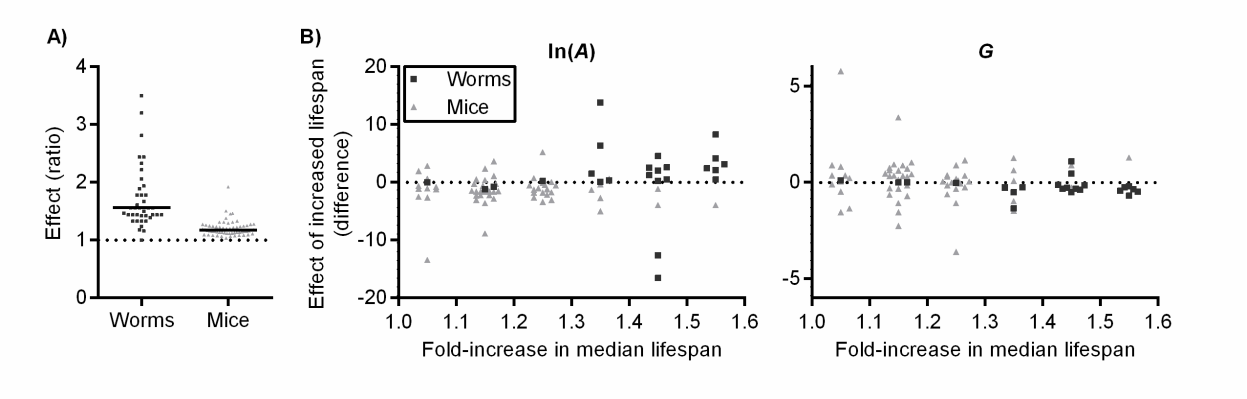


Figure S8. The relationship between changes to Gompertz parameters and magnitude of lifespan extension in *C. elegans* vs. mice. **(A)** Magnitude of lifespan extension (as a ratio between long-lived and control strains) for *C. elegans* and mice. **(B)** Effect of different magnitudes of lifespan extension on Gompertz parameters, expressed as a difference for ln(*A*) and '*G*' between long-lived and control strains. In order to facilitate comparisons between lifespan extensions of similar magnitude, long-lived strains are binned along the x-axis according to the degree of lifespan extension.

Table S1. Previously published (Yen *et al.* 2008) maximum likelihood estimations of the Gompertz parameter values obtained from published survival curves (Set 1).

		Control				Intervention				
Gene and allele or strain	Background	n	Median lifespan (days)	ln(*A*)	*G*	n	Median lifespan (days)	ln(*A*)	*G*	
Atm $^{-/-}$, *Terc* $^{-/-}$	C57BL/6×WWG		-	-	-	51	343 ↓	-1.5 ↑	1.6 ↓	(Wong *et al.* 2003)
NZB/W	NZW×NZB(F1)		-	-	-	22	274 ↓	-3.16	6.25 ↑	(Conde *et al.* 1998)
Bub1b $^{H/H}$	129/Sv×FVB		-	-	-	212	182 ↓	0.32 ↑	2.55	(Baker *et al.* 2004)
C/ebp $^{b/b}$	C57BL/6J	30	684 *	-6.02	3.4	30	851 ↑	-5.29	2.33	(Chiu *et al.* 2004)
Lep $^{ob/ob}$	C57BL/6J					40	547 ↓	-4.81	3.65	
Cat TG	B6(B6C3F1) ×C57BL/6J	58	790	-4.95	2.44	42	973 ↑	-5.95	2.48	(Schriner *et al.* 2005)
GH TG	C56BL/6×SJL	16	800	-4.56	2.14	9	400 ↓	-6.73	6.06 ↑	(Bartke 2003)
Ghr $^{-/-}$	OLA-BALB/cJ	15	698	-3.48	1.95	11	888 ↑	-5.25	2.14	(Coschigano *et al.* 2003)
Ghrhr $^{lit/lit}$	C57BL/6	31	882	-6.77	2.86	35	1094 ↑	-8.47	2.9	(Flurkey *et al.* 2001)
Igf1r $^{+/-}$	129/J	17	654	-3.36	2.25	20	775 ↑	-5.62	2.88	(Holzenberger *et al.* 2003)
Insr $^{-/-}$ (Firko)	FVB×129S4	67	913	-4.75	2.22	60	1017 ↑	-7.56 ↓	2.98	(Blüher *et al.* 2003)
Klotho $^{kl/kl}$	C57BL/6J + C3H/J		-	-	-	29	57 ↓	-0.88	22.06 ↑	(Kuro-O *et al.* 1997)
Lmna $^{L530P/L530P}$	C57BL/6 + 129S1/Sv		-	-	-	25	29 ↓	-1.27	61.06 ↑	(Mounkes *et al.* 2003)
MsrA $^{-/-}$	C57BL/6 + 129/SvJ	14	788	-4.13	2.58	17	400 ↓	-3.12	4.23	(Moskovitz *et al.* 2001)
p53 $^{+/-}$	129/SV + C57BL/6	56	826 *	-7.22	3.34	217	500 ↓	-3.7 ↑	2.75	(Tyner *et al.* 2002)
p53 $^{+/m}$	129/SV + C57BL/6					35	672 ↓	-5.2	2.97	
p66 $^{shc\ -/-}$	129/Sv	14	761	-13.5	7.17	15	973 ↑	-8.25	3.57	(Migliaccio *et al.* 1999)
Pit1 $^{dw/dw}$	C3H/HeJ + DW/J	34	882	-7.66	3.44	25	1216 ↑	-7.96	2.47	(Flurkey *et al.* 2001)
Plau TG (α-MUPA)	FVB/N	33	851	-4.87	2.38	33	988 ↑	-8.47 ↓	3.32	(Miskin and Masos 1997)
PolgA $^{mut/mut}$	129 + C57BL/6		-	-	-	38	336 ↓	-8.57 ↓	11.27 ↑	(Trifunovic *et al.* 2004)
Prdx1 $^{-/-}$	B6×129SvEv		-	-	-	34	730 ↓	-3.15	1.46 ↓	(Neumann *et al.* 2003)
Prop1 $^{df/df}$	Ames Stock	13	650	-3.8	2.04	16	1250 ↑	-9.57 ↓	2.89	(Brown-Borg *et al.* 1996)
Prop1 $^{df/df}$	Ames Stock	26	750	-6.68	3.8	24	1000 ↑	-6.05	2.33 ↓	(Bartke *et al.* 2001)
Senescence Accelerated Mice (SAM)	AKR	377	395 **	-2.22	2.36	493	289 ↓	-2.48	3.94 ↑	(Takeda *et al.* 1981)
Top3B $^{-/-}$	C57BL/6J + 129/svEv		-	-	-	30	441 ↓	-1.1 ↑	0.89 ↓	(Kwan and Wang 2001)
Trx TG	C57BL/6	82	577	-3.73	2.67	94	699 ↑	-4.26	2.52	(Mitsui *et al.* 2002)
Wrn $^{-/-}$, *Terc* $^{-/-}$	C57BL/6 + 129Sv + BALB/c + SLJ		-	-	-	39	210 ↓	-0.67 ↑	2.08	(Chang *et al.* 2004)

Gompertz parameter values, their statistical significance and genetic backgrounds are as published previously (Adapted from TABLE 1, Yen *et al.* 2008). Statistically-significant changes relative to controls (as determined by Yen et al.) are indicated by up (increased) or down (decreased) arrows. When controls were not reported, statistical comparisons were made against generic, representative control values (*G* = 2.93 and ln(*A*) = -5.43). Tg: transgene. Median lifespans were obtained from the original cited paper.

* *C/ebp* and *lep* mutants shared the same control group, as did p53 $^{+/-}$ and $^{+/m}$ mice

** Controls for Senescence-Accelerated Mice ("senescence-resistant" mice) were considered to be short-lived for the purposes of this analysis

Table S2. Tabular data for lines of inbred mice described in Figure 4.

	Female				Male			
Strain	Median lifespan (days)	n	ln(A)	G	Median lifespan (days)	n	ln(A)	G
129S1/SvImJ	770	32	-5.01	2.50	880	32	-7.45	3.36
A/J	640	32	-5.67	3.69	620	30	-4.67	3.15
BALB/cByJ	760	32	-6.38	3.48	710	32	-3.53	2.05
BTBR T+ tf/J	600	32	-4.26	3.05	560	32	-3.46	2.36
BUB/BnJ	620	24	-2.63	1.62	490	25	-2.26	1.37
C3H/HeJ	780	29	-7.07	3.94	710	32	-5.48	3.07
C57BL/10J	890	32	-4.00	1.79	780	27	-6.08	3.27
C57BL/6J	865	29	-5.50	2.50	900	32	-7.64	3.40
C57BLKS/J	850	32	-4.70	2.45	825	32	-7.42	3.55
C57BR/cdJ	880	32	-6.12	2.96	850	32	-6.76	3.24
C57L/J	720	32	-12.75	7.24	730	32	-9.21	5.30
CAST/EiJ	670	17	-5.06	3.09				
CBA/J	640	30	-3.77	2.40	650	32	-4.00	2.46
DBA/2J	640	32	-2.67	1.76	705	32	-4.43	2.76
FVB/NJ	760	29	-3.75	1.88	590	25	-2.97	1.84
KK/HlJ	590	32	-4.91	3.51	620	32	-3.71	2.38
LP/J	810	32	-4.93	2.37	810	32	-5.84	2.91
MOLF	680	32	-3.94	2.28	630	32	-6.05	4.26
MRL/MpJ	550	31	-4.89	3.81				
NOD.B10Sn-H2 /J	660	32	-5.06	3.16	700	31	-3.58	2.01
NON/ShiLtJ	750	31	-6.27	3.50	840	32	-8.22	3.99
NZO/HlLtJ	560	32	-2.79	2.03	415	31	-1.81	1.57
NZW/LacJ	730	32	-4.91	2.68	770	32	-3.68	1.69
P/J	680	32	-3.94	2.29				
PL/J	410	32	-2.57	2.40	460	32	-2.97	2.72
PWD/PhJ	850	32	-5.70	2.77	815	27	-4.59	2.36
RIIIS/J	790	32	-7.75	3.97	830	32	-9.12	4.29
SJL/J	450	30	-2.36	2.01	510	16	-4.28	3.60
SM/J	750	32	-5.22	2.86	780	31	-6.56	3.59
SWR/J	635	31	-3.20	1.92	710	32	-2.70	1.49
WSB/EiJ	950	32	-3.56	1.39	900	32	-2.89	1.17

Table S3. Tabular data for lines of ad libitum-fed recombinant inbred mice described in Figure S3.

	Male				Female			
Line Number	Median lifespan (days)	n	ln(A)	G	Median lifespan (days)	n	ln(A)	G
3	700	5	-0.26	0.00	1045	5	-4.47	1.86
7	1048	5	-24.58	9.39	1143	5	-6.81	2.58
13	696	5	-4.26	2.36	863	5	-16.45	7.65
14	946	5	-3.92	1.86				
16	758	6	-5.28	3.12	687	5	-2.18	1.37
19	981	5	-8.38	3.66	938	5	-8.41	3.87
22	1168	5	-14.64	5.11				
23	1063	11	-5.35	2.07	991	15	-7.74	3.13
24	829	5	-4.31	2.30	1023	5	-3.05	1.22
25	979	5	-7.99	3.47	979	5	-44.12	17.45
26	1043	5	-31.75	11.98	1049	5	-34.42	12.90
28					716	5	-8.90	5.31
41	787	5	-27.82	14.08	986	5	-6.83	3.10
46					1147	5	-8.91	3.36
48	751	5	-4.10	2.22	898	5	-3.20	1.51
49	892	4	-4.24	2.08	712	5	-7.85	4.79
50	459	5	-2.07	2.04	291	5	-1.42	2.41
51	1018	5	-6.89	2.91	820	5	-5.14	2.63
52	713	5	-2.59	1.74				
56	1277	5	-3.72	1.26	1145	5	-5.52	2.02
60					1073	5	-1.73	0.62
62	1055	5	-16.48	6.26				
66	685	5	-2.56	1.60	743	5	-3.01	1.79
79	1094	5	-13.54	4.98				
80	803	5	-7.75	4.09	887	5	-11.46	5.22
84	900	5	-8.43	4.01	810	5	-11.70	5.96
86	1111	5	-5.91	2.34	926	4	-7.43	3.33
89	951	5	-2.55	1.19	889	5	-2.74	1.48
90	829	5	-9.86	4.99	906	5	-35.11	15.19
92	994	5	-6.79	2.79	798	5	-3.66	1.82
94	718	5	-3.41	1.69	699	5	-5.99	3.84
97	991	5		37.24	999	5		33.35
98	1187	5	-7.32	2.58	1249	5	-9.22	3.07
99	930	5	-9.01	4.00	887	5	-3.25	1.63
100	895	5	-34.14	14.85	924	5	-35.15	15.11
103	823	5	-23.27	11.28	855	5	-8.81	4.23
107	1079	5	-13.01	4.83	596	5	-2.88	1.93
110	587	5	-5.45	4.20	851	5	-3.47	1.76
112	946.5	4	-3.24	1.42	600	5	-3.70	2.49
114	1013	5	-13.17	5.42	911	5	-4.85	2.20
115	992	5	-7.43	3.22	746	5	-4.49	2.70
117	893	5	-4.53	2.07	641	5	-3.41	2.30
122	859	5	-9.22	4.42	1010	5	-2.68	1.12
123	915	5	-5.46	2.51	962	5	-23.16	9.62

Line numbers correspond to those from source publication (Liao *et al.* 2010)

Table S4. Source publications and tabular data for *daf-2*, *isp-1* and *eat-2* mutant lines described in Figure 7.

Source Publication	n		Median Lifespan (days)		Fractional contribution to lifespan increase		
	N2	mutant	N2	mutant	*A*	*G*	*S*
daf-2 (e1370)							
(Kaeberlein *et al.* 2006)	58	41	19	37.5	0.40123	0.49956	0.09921
(Tullet *et al.* 2008)	90	88	21	39	0.436832	0.552616	-0.01055
(Troemel *et al.* 2006)	90	88	16	39	-0.10334	0.896655	0
(Yang and Hekimi 2010b)	400	150	18	44	0.121681	0.812151	-0.06617
(Wang *et al.* 2008)	80	80	19	34	-0.23511	0.75992	0.004969
(Mehta *et al.* 2009)	40	40	22	29	-0.241	0.577434	0.181565
(Tissenbaum and Guarente 2001)	142	293	18	45	-0.02147	0.906075	-0.07245
(Hsu *et al.* 2003)	48	41	20	45	-0.20419	0.790542	-0.00526
(Lin *et al.* 2001)	348	140	19	46	-0.1727	0.821857	-0.00544
(Garsin *et al.* 2003)	75	75	16	31	-0.05878	0.941221	0
(Huang *et al.* 2004)	180	114	16	36	-0.191	0.775917	-0.03308
(Dorman *et al.* 1995)	48	37	17	38.5	-0.06963	0.905061	0.025306
(Apfeld and Kenyon 1999)	100	100	18	53	-0.19923	0.787241	-0.01353
(Berdichevsky *et al.* 2006)	70	100	19	40	0.74393	0.23844	0.01763
(Kenyon *et al.* 1993)	49	42	19.5	44	-0.14888	0.764857	-0.08626
(Oh *et al.* 2005)	125	46	16	42	0.256288	0.608954	-0.13476
(Van Raamsdonk *et al.* 2010)	100	100	22	38	-0.07721	0.814284	0.108506
(Van Raamsdonk and Hekimi 2009)	309	168	19	66	-0.15918	0.814043	-0.02678
isp-1 (qm150)							
(Feng *et al.* 2001)	380	283	22	34	-0.25783	0.733226	0.008946
(Curtis *et al.* 2006)	157	45	21	28	0.310793	-0.16912	0.520091
(Van Raamsdonk and Hekimi 2009)	309	114	19	43	-0.20072	0.778142	-0.02114
(Dingley *et al.* 2009)	38	37	14	21	-0.40716	0.586179	-0.00666
(Yang and Hekimi 2010a)	150	150	22	35	-0.33977	0.638542	-0.02169
(Lee *et al.* 2010)	107	96	15	27	0.867935	0.098988	-0.03308
(Yang and Hekimi 2010b)	400	200	21	35	-0.17697	0.816204	-0.00682
(Baruah *et al.* 2014)	112	117	22	32	-0.39118	0.608112	-0.00071
(Yee *et al.* 2014)	250	200	18	34	-0.22033	0.732193	-0.04748
(Hsu *et al.* 2003)	48	35	20	27	-0.28045	0.707574	-0.01197
(Torgovnick *et al.* 2010)	238	162	16	25	0.330464	0.589386	-0.08015
(Bennett *et al.* 2014)	365	206	20	26	0.245278	0.189918	0.564804
(Mouchiroud *et al.* 2011)	55	53	13	20	-0.17964	0.27395	0.546407

eat-2 (ad1116)							
(Panowski *et al.* 2007)	70	48	18	26	-0.27081	0.634363	-0.09483
(Park *et al.* 2010)	60	60	20	26	-0.29919	0.593557	-0.10725
(Lakowski and Hekimi 1998)	50	36	20	29	-0.05582	0.623448	0.320736
(Van Raamsdonk and Hekimi 2009)	240	240	18	35	0.475685	0.361144	0.163172
(Carrano *et al.* 2009)	67	74	23	30	-0.27415	0.725852	0
(Seo *et al.* 2015)	75	75	17	24.5	0.896784	-0.0909	0.012314
(Hsu *et al.* 2003)	48	41	21	29	0.967482	-0.03252	0
(Greer and Brunet 2009)	86	84	23	29	-0.19653	0.748508	-0.05496
(Schreiber *et al.* 2010)	100	71	18	20	-0.32004	0.599089	0.080875
(Ching *et al.* 2011)	63	70	15	23	-0.31604	0.676153	-0.00781
(Schleit *et al.* 2011)	181	139	18	24	0.753534	-0.07513	-0.17133
(Yuan *et al.* 2012)	65	60	20.5	32	-0.01853	0.981473	0
(Gaglia *et al.* 2012)	75	94	16	24	-0.27517	0.703776	-0.02105
(Rousakis *et al.* 2013)	110	160	19	22	0.318461	0.597187	-0.08435
(Thondamal *et al.* 2014)	102	108	18	22	0.404702	-0.24621	0.349087
(Yee *et al.* 2014)	150	150	19	31	-0.19329	0.748434	0.058274
(Zimmerman and Kim 2014)	61	83	17.5	20	-0.1679	0.570557	-0.26155
(Chin *et al.* 2014)	100	59	15	22	-0.24939	0.721649	-0.02897
(Asthana *et al.* 2015)	217	189	18	22.5	-0.35872	0.641277	0
(Hine *et al.* 2015)	100	100	20	29	0.589966	0.244302	0.165732
(Seo *et al.* 2016)	101	81	19.5	31	0.574392	0.385386	-0.04022
(Singh *et al.* 2016)	93	88	21	32	0.154243	0.733848	-0.11191
(Merkwirth *et al.* 2016)	101	63	19	26	-0.26723	0.693701	-0.03907

Supplemental References

Apfeld, J., and C. Kenyon, 1999 Regulation of lifespan by sensory perception in Caenorhabditis elegans. Nature **402:** 804-809.

Asthana, J., D. Yadav, A. Pant, A. K. Yadav, M. M. Gupta *et al.*, 2015 Acacetin 7-O-α-l-rhamnopyranosyl (1–2) β-D-xylopyranoside Elicits Life-span Extension and Stress Resistance in Caenorhabditis elegans. The Journals of Gerontology Series A: Biological Sciences and Medical Sciences.

Baker, D. J., K. B. Jeganathan, J. D. Cameron, M. Thompson, S. Juneja *et al.*, 2004 BubR1 insufficiency causes early onset of aging-associated phenotypes and infertility in mice. Nature genetics **36:** 744-749.

Bartke, A., J. C. Wright, J. A. Mattison, D. K. Ingram, R. A. Miller *et al.*, 2001 Longevity: Extending the lifespan of long-lived mice. Nature **414:** 412-412.

Bartke, A., 2003 Can growth hormone (GH) accelerate aging? Evidence from GH-transgenic mice. Neuroendocrinology **78:** 210-216.

Baruah, A., H. Chang, M. Hall, J. Yuan, S. Gordon *et al.*, 2014 CEP-1, the Caenorhabditis elegans p53 Homolog, Mediates Opposing Longevity Outcomes in Mitochondrial Electron Transport Chain Mutants. PLoS Genet **10:** e1004097.

Bennett, C. F., H. Vander Wende, M. Simko, S. Klum, S. Barfield *et al.*, 2014 Activation of the mitochondrial unfolded protein response does not predict longevity in Caenorhabditis elegans. Nat Commun **5**.

Berdichevsky, A., M. Viswanathan, H. R. Horvitz and L. Guarente, 2006 C. elegans SIR-2.1 Interacts with 14-3-3 Proteins to Activate DAF-16 and Extend Life Span. Cell **125:** 1165-1177.

Blüher, M., B. B. Kahn and C. R. Kahn, 2003 Extended Longevity in Mice Lacking the Insulin Receptor in Adipose Tissue. Science **299:** 572-574.

Brown-Borg, H. M., K. E. Borg, C. J. Meliska and A. Bartke, 1996 Dwarf mice and the ageing process. Nature **384:** 33-33.

Carrano, A. C., Z. Liu, A. Dillin and T. Hunter, 2009 A conserved ubiquitination pathway determines longevity in response to diet restriction. Nature **460:** 396-399.

Chang, S., A. S. Multani, N. G. Cabrera, M. L. Naylor, P. Laud *et al.*, 2004 Essential role of limiting telomeres in the pathogenesis of Werner syndrome. Nat Genet **36:** 877-882.

Chin, R. M., X. Fu, M. Y. Pai, L. Vergnes, H. Hwang *et al.*, 2014 The metabolite α-ketoglutarate extends lifespan by inhibiting ATP synthase and TOR. Nature **510:** 397-401.

Ching, T.-T., W.-C. Chiang, C.-S. Chen and A.-L. Hsu, 2011 Celecoxib extends C. elegans lifespan via inhibition of insulin-like signaling but not cyclooxygenase-2 activity. Aging Cell **10:** 506-519.

Chiu, C.-H., W.-D. Lin, S.-Y. Huang and Y.-H. Lee, 2004 Effect of a C/EBP gene replacement on mitochondrial biogenesis in fat cells. Genes Dev **18:** 1970-1975.

Conde, C., S. Weller, S. Gilfillan, L. Marcellin, T. Martin *et al.*, 1998 Terminal Deoxynucleotidyl Transferase Deficiency Reduces the Incidence of Autoimmune Nephritis in (New Zealand Black x New Zealand White)F1 Mice. The Journal of Immunology **161:** 7023-7030.

Conover, C. A., L. K. Bale, J. R. Mader, M. A. Mason, K. P. Keenan *et al.*, 2010 Longevity and Age-Related Pathology of Mice Deficient in Pregnancy-Associated Plasma Protein-A. The Journals of Gerontology Series A: Biological Sciences and Medical Sciences **65A:** 590-599.

Coschigano, K. T., A. N. Holland, M. E. Riders, E. O. List, A. Flyvbjerg *et al.*, 2003 Deletion, But Not Antagonism, of the Mouse Growth Hormone Receptor Results in Severely Decreased Body Weights, Insulin, and Insulin-Like Growth Factor I Levels and Increased Life Span. Endocrinology **144:** 3799-3810.

Curtis, R., G. O'connor and P. S. Distefano, 2006 Aging networks in Caenorhabditis elegans: AMP-activated protein kinase (aak-2) links multiple aging and metabolism pathways. Aging Cell **5:** 119-126.

Dingley, S., E. Polyak, R. Lightfoot, J. Ostrovsky, M. Rao *et al.*, 2009 Mitochondrial respiratory chain dysfunction variably increases oxidant stress in Caenorhabditis elegans. Mitochondrion.

Dorman, J. B., B. Albinder, T. Shroyer and C. Kenyon, 1995 The age-1 and daf-2 genes function in a common pathway to control the lifespan of Caenorhabditis elegans. Genetics **141:** 1399-1406.

Feng, J. L., F. Bussiere and S. Hekimi, 2001 Mitochondrial electron transport is a key determinant of life span in Caenorhabditis elegans. Developmental Cell **1:** 633-644.

Flurkey, K., J. Papaconstantinou, R. A. Miller and D. E. Harrison, 2001 Lifespan extension and delayed immune and collagen aging in mutant mice with defects in growth hormone production. Proceedings of the National Academy of Sciences **98:** 6736-6741.

Gaglia, M. M., D.-E. Jeong, E.-A. Ryu, D. Lee, C. Kenyon *et al.*, 2012 Genes That Act Downstream of Sensory Neurons to Influence Longevity, Dauer Formation, and Pathogen Responses in *Caenorhabditis elegans*. PLoS Genet **8:** e1003133.

Garsin, D. A., J. M. Villanueva, J. Begun, D. H. Kim, C. D. Sifri *et al.*, 2003 Long-lived C. elegans daf-2 mutants are resistant to bacterial pathogens. Science **300:** 1921.

Greer, E. L., and A. Brunet, 2009 Different dietary restriction regimens extend lifespan by both independent and overlapping genetic pathways in C. elegans. Aging Cell **8:** 113-127.

Hine, C., E. Harputlugil, Y. Zhang, C. Ruckenstuhl, Byung c. Lee *et al.*, 2015 Endogenous Hydrogen Sulfide Production Is Essential for Dietary Restriction Benefits. Cell **160:** 132-144.

Holzenberger, M., J. Dupont, B. Ducos, P. Leneuve, A. Geloen *et al.*, 2003 IGF-1 receptor regulates lifespan and resistance to oxidative stress in mice. Nature **421:** 182-187.

Hsu, A.-L., C. T. Murphy and C. Kenyon, 2003 Regulation of Aging and Age-Related Disease by DAF-16 and Heat-Shock Factor. Science **300:** 1142-1145.

Huang, C., C. J. Xiong and K. Kornfeld, 2004 Measurements of age-related changes of physiological processes that predict lifespan of Caenorhabditis elegans. Proceedings of the National Academy of Sciences of the United States of America **101:** 8084-8089.

Kaeberlein, T. L., E. D. Smith, M. Tsuchiya, K. L. Welton, J. H. Thomas *et al.*, 2006 Lifespan extension in Caenorhabditis elegans by complete removal of food. Aging Cell **5:** 487-494.

Kenyon, C., J. Chang, E. Gensch, A. Rudner and R. Tabtiang, 1993 A C. elegans mutant that lives twice as long as wild type. Nature **366:** 461-464.

Kuro-O, M., Y. Matsumura, H. Aizawa, H. Kawaguchi, T. Suga *et al.*, 1997 Mutation of the mouse klotho gene leads to a syndrome resembling ageing. Nature **390:** 45-51.

Kwan, K. Y., and J. C. Wang, 2001 Mice lacking DNA topoisomerase IIIβ develop to maturity but show a reduced mean lifespan. Proceedings of the National Academy of Sciences **98:** 5717-5721.

Lakowski, B., and S. Hekimi, 1998 The genetics of caloric restriction in Caenorhabditis elegans. Proc Natl Acad Sci U S A **95:** 13091-13096.

Lee, S. J., A. B. Hwang and C. Kenyon, 2010 Inhibition of Respiration Extends C. elegans Life Span via Reactive Oxygen Species that Increase HIF-1 Activity. Curr Biol **20:** 2131-2136.

Liao, C. Y., B. A. Rikke, T. E. Johnson, V. Diaz and J. F. Nelson, 2010 Genetic variation in the murine lifespan response to dietary restriction: from life extension to life shortening. Aging Cell **9:** 92-95.

Lin, K., H. Hsin, N. Libina and C. Kenyon, 2001 Regulation of the Caenorhabditis elegans longevity protein DAF-16 by insulin/IGF-1 and germline signaling. Nat Genet **28:** 139-145.

Mehta, R., K. A. Steinkraus, G. L. Sutphin, F. J. Ramos, L. S. Shamieh *et al.*, 2009 Proteasomal Regulation of the Hypoxic Response Modulates Aging in C. elegans. Science **324:** 1196-1198.

Merkwirth, C., V. Jovaisaite, J. Durieux, O. Matilainen, Sabine d. Jordan *et al.*, 2016 Two Conserved Histone Demethylases Regulate Mitochondrial Stress-Induced Longevity. Cell **165:** 1209-1223.

Migliaccio, E., M. Giorgio, S. Mele, G. Pelicci, P. Reboldi *et al.*, 1999 The p66shc adaptor protein controls oxidative stress response and life span in mammals. Nature **402:** 309-313.

Miskin, R., and T. Masos, 1997 Transgenic Mice Overexpressing Urokinase-Type Plasminogen Activator in the Brain Exhibit Reduced Food Consumption, Body Weight and Size, and Increased Longevity. The Journals of Gerontology Series A: Biological Sciences and Medical Sciences **52A:** B118-B124.

Mitsui, A., J. Hamuro, H. Nakamura, N. Kondo, Y. Hirabayashi *et al.*, 2002 Overexpression of human thioredoxin in transgenic mice controls oxidative stress and life span. Antioxid Redox Signal **4:** 693-696.

Moskovitz, J., S. Bar-Noy, W. M. Williams, J. Requena, B. S. Berlett *et al.*, 2001 Methionine sulfoxide reductase (MsrA) is a regulator of antioxidant defense and lifespan in mammals. Proceedings of the National Academy of Sciences **98:** 12920-12925.

Motulsky, H., and R. Brown, 2006 Detecting outliers when fitting data with nonlinear regression - a new method based on robust nonlinear regression and the false discovery rate. BMC Bioinformatics **7:** 123.

Mouchiroud, L., L. Molin, P. Kasturi, M. N. Triba, M. E. Dumas *et al.*, 2011 Pyruvate imbalance mediates metabolic reprogramming and mimics lifespan extension by dietary restriction in Caenorhabditis elegans. Aging Cell **10:** 39-54.

Mounkes, L. C., S. Kozlov, L. Hernandez, T. Sullivan and C. L. Stewart, 2003 A progeroid syndrome in mice is caused by defects in A-type lamins. Nature **423:** 298-301.

Neumann, C. A., D. S. Krause, C. V. Carman, S. Das, D. P. Dubey *et al.*, 2003 Essential role for the peroxiredoxin Prdx1 in erythrocyte antioxidant defence and tumour suppression. Nature **424:** 561-565.

Oh, S. W., A. Mukhopadhyay, N. Svrzikapa, F. Jiang, R. J. Davis *et al.*, 2005 JNK regulates lifespan in Caenorhabditis elegans by modulating nuclear translocation of forkhead transcription factor/DAF-16. Proceedings of the National Academy of Sciences of the United States of America **102:** 4494-4499.

Panowski, S. H., S. Wolff, H. Aguilaniu, J. Durieux and A. Dillin, 2007 PHA-4/Foxa mediates diet-restriction-induced longevity of C. elegans. Nature **447:** 550-555.

Park, S.-K., C. D. Link and T. E. Johnson, 2010 Life-span extension by dietary restriction is mediated by NLP-7 signaling and coelomocyte endocytosis in C. elegans. The FASEB Journal **24:** 383-392.

Promislow, Tatar, Pletcher and Carey, 1999 Below-threshold mortality: implications for studies in evolution, ecology and demography. Journal of Evolutionary Biology **12:** 314-328.

Rousakis, A., A. Vlassis, A. Vlanti, S. Patera, G. Thireos *et al.*, 2013 The general control nonderepressible-2 kinase mediates stress response and longevity induced by target of rapamycin inactivation in Caenorhabditis elegans. Aging Cell **12:** 742-751.

Schleit, J., V. Z. Wall, M. Simko and M. Kaeberlein, 2011 The MDT-15 Subunit of Mediator Interacts with Dietary Restriction to Modulate Longevity and Fluoranthene Toxicity in *Caenorhabditis elegans*. PLoS ONE **6:** e28036.

Schreiber, M. A., J. T. Pierce-Shimomura, S. Chan, D. Parry and S. L. Mcintire, 2010 Manipulation of Behavioral Decline in *Caenorhabditis elegans* with the Rag GTPase *raga-1*. PLoS Genet **6:** e1000972.

Schriner, S. E., N. J. Linford, G. M. Martin, P. Treuting, C. E. Ogburn *et al.*, 2005 Extension of murine life span by overexpression of catalase targeted to mitochondria. Science **308:** 1909-1911.

Seo, M., K. Seo, W. Hwang, H. J. Koo, J.-H. Hahm *et al.*, 2015 RNA helicase HEL-1 promotes longevity by specifically activating DAF-16/FOXO transcription factor signaling in Caenorhabditis elegans. Proceedings of the National Academy of Sciences **112:** E4246-E4255.

Seo, M., S. Park, H. G. Nam and S.-J. V. Lee, 2016 RNA helicase SACY-1 is required for the longevity caused by various genetic perturbations in Caenorhabditis elegans. Cell Cycle**:** 00-00.

Singh, A., N. Kumar, L. Matai, V. Jain, A. Garg *et al.*, 2016 A chromatin modifier integrates insulin/IGF-1 signalling and dietary restriction to regulate longevity. Aging Cell **15:** 694-705.

Takeda, T., M. Hosokawa, S. Takeshita, M. Irino, K. Higuchi *et al.*, 1981 A new murine model of accelerated senescence. Mechanisms of Ageing and Development **17:** 183-194.

Thondamal, M., M. Witting, P. Schmitt-Kopplin and H. Aguilaniu, 2014 Steroid hormone signalling links reproduction to lifespan in dietary-restricted Caenorhabditis elegans. Nat Commun **5**.

Tissenbaum, H. A., and L. Guarente, 2001 Increased dosage of a sir-2 gene extends lifespan in Caenorhabditis elegans. Nature **410:** 227-230.

Torgovnick, A., A. Schiavi, R. Testi and N. Ventura, 2010 A role for p53 in mitochondrial stress response control of longevity in C. elegans. Experimental Gerontology **45:** 550-557.

Trifunovic, A., A. Wredenberg, M. Falkenberg, J. N. Spelbrink, A. T. Rovio *et al.*, 2004 Premature ageing in mice expressing defective mitochondrial DNA polymerase. Nature **429:** 417-423.

Troemel, E. R., S. W. Chu, V. Reinke, S. S. Lee, F. M. Ausubel *et al.*, 2006 p38 MAPK Regulates Expression of Immune Response Genes and Contributes to Longevity in *C. elegans*. PLoS Genet **2:** e183.

Tullet, J. M. A., M. Hertweck, J. H. An, J. Baker, J. Y. Hwang *et al.*, 2008 Direct Inhibition of the Longevity-Promoting Factor SKN-1 by Insulin-like Signaling in C. elegans. Cell **132:** 1025-1038.

Tyner, S. D., S. Venkatachalam, J. Choi, S. Jones, N. Ghebranious *et al.*, 2002 p53 mutant mice that display early ageing-associated phenotypes. Nature **415:** 45-53.

Van Raamsdonk, J. M., and S. Hekimi, 2009 Deletion of the mitochondrial superoxide dismutase sod-2 extends lifespan in Caenorhabditis elegans. PLoS Genet **5:** e1000361.

Van Raamsdonk, J. M., Y. Meng, D. Camp, W. Yang, X. Jia *et al.*, 2010 Decreased energy metabolism extends life span in Caenorhabditis elegans without reducing oxidative damage. Genetics **185:** 559-571.

Wang, M. C., E. J. O'rourke and G. Ruvkun, 2008 Fat Metabolism Links Germline Stem Cells and Longevity in C. elegans. Science **322:** 957-960.

Wong, K.-K., R. S. Maser, R. M. Bachoo, J. Menon, D. R. Carrasco *et al.*, 2003 Telomere dysfunction and Atm deficiency compromises organ homeostasis and accelerates ageing. Nature **421:** 643-648.

Yang, W., and S. Hekimi, 2010a Two modes of mitochondrial dysfunction lead independently to lifespan extension in Caenorhabditis elegans. Aging Cell **9:** 433-447.

Yang, W., and S. Hekimi, 2010b A mitochondrial superoxide signal triggers increased longevity in Caenorhabditis elegans. PLoS Biol **8:** e1000556.

Yee, C., W. Yang and S. Hekimi, 2014 The Intrinsic Apoptosis Pathway Mediates the Pro-Longevity Response to Mitochondrial ROS in C. elegans. Cell **157:** 897-909.

Yen, K., D. Steinsaltz and C. V. Mobbs, 2008 Validated analysis of mortality rates demonstrates distinct genetic mechanisms that influence lifespan. Experimental Gerontology **43:** 1044-1051.

Yuan, Y., C. S. Kadiyala, T.-T. Ching, P. Hakimi, S. Saha *et al.*, 2012 Enhanced Energy Metabolism Contributes to the Extended Life Span of Calorie-restricted Caenorhabditis elegans. Journal of Biological Chemistry **287:** 31414-31426.

Zimmerman, S. M., and S. K. Kim, 2014 The GATA transcription factor/MTA-1 homolog egr-1 promotes longevity and stress resistance in Caenorhabditis elegans. Aging Cell **13:** 329-339.

File S2. Sample generator. (.zip, 21 KB)

www.genetics.org/lookup/suppl/doi:10.1534/genetics.116.192369/-/DC1/FileS2.xlsm