Actuarial senescence in a long-lived orchid challenges our current understanding of ageing

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Keywords:

demographic senescence, demography, life history theory, mortality, rate of ageing, survival–reproduction trade-off

Abstract

The dominant evolutionary theory of actuarial senescence – an increase in death rate with advancing age – is based on the concept of a germ cell line that is separated from the somatic cells early in life. However, such a separation is not clear in all organisms. This has been suggested to explain the paucity of evidence for actuarial senescence in plants. We used a 32-year study of *Dactylorhiza lapponica* that replaces its organs each growing season, to test whether individuals of this tuberous orchid senesce. We performed a Bayesian survival trajectory analysis accounting for reproductive investment, for individuals under two types of land-use, in two climatic regions. The mortality trajectory was best-approximated by a Weibull model, showing clear actuarial senescence. Rates of senescence in this model declined with advancing age, but were slightly higher in mown plots and in the more benign climatic region. At older ages, senescence was evident only when accounting for a positive effect of reproductive investment on mortality. Our results demonstrate actuarial senescence as well as a survival-reproduction trade-off in plants, and indicate that environmental context may influence senescence rates. This knowledge is crucial for understanding the evolution of demographic senescence and for models of plant population dynamics.

Introduction

Age trajectories of both mortality and fertility show a remarkable diversity across taxonomic groups and can be positive, negative or constant [1]. Increasing mortality and/or decreasing fertility with age after reproductive maturity, known as demographic senescence, seems to occur in all mammals and birds, which are also the most studied taxa in this regard. Demographic senescence in plants has received less attention, but already 50 years ago J. L. Harper [2] noted that linear survivorship curves (on a semi-log scale), indicating constant mortality risks, were the norm in the handful of plant populations for which long-term observational data was available. Subsequent studies [3,4] have typically supported that actuarial senescence, an increase in mortality with advancing age after maturity, is absent or negligible in perennial plants. Other studies have provided some empirical or theoretical support for the existence of both actuarial senescence [5,6] and "negative senescence" (a decrease in mortality over age in reproductive individuals; [7,8,9]) in the plant kingdom. However, currently there is only scant empirical evidence for either of these mortality trajectories for plants [10], let alone more detailed knowledge of rates of senescence over life courses.

The age trajectory of mortality is a population-level phenomenon that is a composite of individual-level mortality risks. Sources of heterogeneity in mortality among individuals should therefore be accounted for when drawing conclusions about changes in mortality of individuals from mortality trajectories, in particular because a gradual loss of the more "frail" individuals in a population can mask potential increased mortality risks of individuals [11]. One important driver of among-individual variation in mortality is variation in resource allocation among processes affecting mortality and fertility [12,13]. In observational studies in natural populations, life-history trade-offs are notoriously difficult to establish and reported relationships between current reproduction and other aspects of individual fitness range from negative, to zero and even positive [14]. However, individual-based data on mortality and reproductive investment over entire plant life courses may be needed to observe trade-offs in terms of correlations among demographic rates, and previous studies have only rarely been based on such long-term data.

In addition to affecting overall mortality, investments in reproduction may come with costs to somatic maintenance which in turn may affect future mortality and fertility. As first described as part of the “disposable soma theory” [15,16], such a trade-off may be an evolutionary cause of demographic senescence [17,18]. All current evolutionary theories of demographic senescence are based on the premise that any factor that affects mortality and fertility at earlier ages will have a greater impact on individual fitness than one that acts later in life [19]. The disposable soma theory proposes that demographic senescence results from the accumulation of errors in the transcription of macromolecules that constitute the basis of an individual’s phenotypic expression [16,20]. It is further proposed that the resulting deterioration of somatic cells may occur as long as there is no deterioration of the genetic material that will be passed on to offspring. In species with an early-life separation between a germ cell line and somatic cells, it has then been suggested that, after a threshold age has been reached, investment into reproduction will increase fitness more than investment into maintenance, causing demographic senescence [17]. For many plants and other sessile organisms, however, there is no clear separation of germ and somatic cell lines early in life. Therefore, the existence of demographic senescence in such organisms would suggest that more general models, not assuming a germ-soma separation, would be needed to explain its evolution [18].

Environmental conditions influence costs of maintaining low mortality and high fertility [12,14,21,22,23], and therefore presumably affect the optimal balance of maintenance-fertility trade-offs. Environmental variation could thus be expected to also lead to within-species variation in shapes of mortality trajectories. However, current evolutionary theories of demographic senescence predict that variation in resource allocation among individuals may only change mortality and fertility additively or proportionally [24,25,26,27]. That is, it may not have an important effect on the "intrinsic" age-dependent mechanisms of senescence, referred to as the speed of senescence or the ageing rate. This is because age patterns of mortality and fertility are determined by numerous genes of small effect and may be more or less fixed for species [16,20,28]. If environmental conditions still do affect rates of senescence in plants, we would expect rates to be lower in low-resource environments because individuals allocating resources to endure stressful conditions rather than to reproduce should have an advantage [29]. On the other hand, we expect unpredictable high-disturbance environments to cause both high baseline mortality and a stronger increase in mortality with age because these environments would favour allocation to reproduction rather than growth and maintenance. In addition, we hypothesize that damage caused by disturbance could increase rates of senescence, simply by weakening individuals.

We investigated mortality trajectories using 32 years of data for a total of 2184 individuals of the long-lived orchid *Dactylorhiza lapponica*. We know from previous experiments that there are significant costs of reproduction in this species (manifested as increased mortality rates in a given year, and reduced sizes of aerial shoots and tubers the year after reproduction), and that costs depend on the environment [22,23,30,31]. We have also found similar effects in other tuberous orchids from the study areas [22,32,33]. Here we estimated age trajectories of mortality using Bayesian survival trajectory analysis [34]. We quantified rates of actuarial senescence and examined how they vary with age and the effect of environmental conditions (regional climate and mowing regime). In addition, we examined the effect of accounting for individual reproductive investment on mortality trajectories. Specifically we tested the following three hypotheses.

1. Actuarial senescence occurs in *D. lapponica*.
2. Mortality is positively correlated with average life-time reproductive investment, and this affects mortality trajectories of populations.
3. Mowing (by damaging and weakening individuals) and more benign climatic conditions (by favouring high allocation of resources to reproduction) lead to increased rates of senescence.

Our results clearly support the first two hypotheses, and lend weaker support to the third hypothesis.

Methods

Study species

Dactylorhiza lapponica (Laest. ex Hartm.) Soó is a non-clonal, tuberous orchid limited to Fennoscandia, Scotland, and alpine areas in Central Europe [35]. In Fennoscandia, *D. lapponica* is found in species-rich open lawn communities of calcareous fens and surrounding springs in the boreal vegetation zones [36,37]. Vegetative individuals form a leaf rosette that is fully grown by the end of June, while flowering individuals continue to grow during the flowering period, which lasts 3-4 weeks from mid-late June. Flowering individuals produce a single inflorescence with approximately 3-15 flowers without any nectar production. In August, above-ground structures die back and a new replacement tuber grows roots and a belowground shoot bud (30). Fruits are capsules containing minute “dust seeds”. The germination rate is very low, and the underground seedling stage is believed to last at least one year [38]. The first flowering event is estimated to occur at the earliest five years after germination [22]. *Dactylorhiza lapponica* is not a common species in Norway, but occurs in large numbers in some localities, e.g. in the study areas, where it is a useful species for studying aging. It is not considered endangered, like *Nigritella nigra* [32] that has been studied for decades at the inland study area (Sølendet). However, many orchids are endangered [39], and better knowledge of the demography of these species is needed.

Study areas and mowing treatments

In the period 1981-2013, we collected demographic data in two nature reserves in central Norway; the coastal Tågdalen area (63°03’N, 9°05’E) and the inland Sølendet area (62°40’N, 11°50’E), both situated at the transition between the middle and northern boreal zone. The Tågdalen area has an oceanic climate with on average ca. 5 days longer growing season and milder winters with more precipitation (snow) compared to the Sølendet area, which is more continental. Both areas are dominated by species-rich open fens mixed with birch-wooded areas [37], and were used for haymaking until around 1950. Traditionally, fens were scythed every second year. In both study areas, mowing was reintroduced in the mid-1970s in permanent plots (mainly 5 m × 2.5 m). Mown plots are scythed in August every second year. Control plots have been left unmown. In the present study we included data collected in 25 control and 11 mown plots in the inland area and in 12 control and 6 mown plots in the coastal area. In a previous study using the same data up to 2011, we found that asymptotic population growth rate was lower in the inland area, with higher mortality and lower fertility [40]. Mowing was found to typically decrease individual and population growth rates whereas effects on mortality and fertility varied depending on study area and weather, and the results also indicated that mowing may reduce interspecific competition [40].

Data collection

Between 1981 and 2013, all individuals inside the plots were monitored from the year they were first observed to reproduce. This resulted in 2184 individuals for which age since first observed reproduction was known. We used this age since first reproduction rather than total age in our analyses, which is relevant because demographic senescence should occur after reproductive maturity if it is caused by allocation to reproduction. In addition, like other plants, *D. lapponica* becomes reproductive after reaching a certain size [30], so individuals should have been of similar size when included in the study. Because we do not know the entire above-ground age we were not able to investigate possible effects of juvenile growth rate. Indeed, vegetative plants were only divided into three size classes at the censuses, and we did not test any possible trade-offs between growth and mortality or fertility due to the lack of quantitative data on plant sizes. We censused plots in early July each year, marking and counting the flowers, measuring the height of plants flowering for the first time, and noting flowering status, number of flowers, size (height of flowering individuals, rosette size class 1-3 of vegetative individuals) or absence of all above-ground plant parts for each individual marked in previous years. See [40] for further details.

Individuals were sometimes noted to be absent for one or two years and then re-appeared, either due to dormancy or herbivory prior to the yearly census. This should have led to an overestimation of mortality in the last three years since we assumed all absent individuals were dead. In addition, as individuals sometimes do not flower for several years, plants first observed to flower in the first years of the study may have flowered previously. For these reasons we re-fitted mortality models using data where either the last three years, or the first five years were removed. We also estimated ages of individuals using the BaSTA package (see below) rather than specifying them as the first year of observation. Finally, we examined the effect of reducing mortality in 2010, which was a year with extreme (high) mortality rates, to average levels in order to evaluate whether environmental conditions this year were driving the observed patterns. In all cases, mortality trajectories were similar and parameters were not statistically significantly different. Results based on the observed data from all years are presented here.

Data analysis

To understand age-specific mortality patterns of *D. lapponica* in the four treatment – study area combinations, as well as how trajectories are affected by individual level variation in average life-time reproductive investment, we used the R package BaSTA [34]. Bayesian survival trajectory analysis (BaSTA) allows users to explore different functional forms of age-specific mortality when age information is scarce or entirely missing [41]. This package is based on the principles of survival analysis, which require defining a random variable *X* for ages at death, where any given age is represented by *x* and the mortality or hazards rate is defined as

$$\mu\left(x \mid \boldsymbol{\beta}\right) = \lim_{\Delta y \to 0} \frac{\Pr\left(x < X < x + \Delta x \mid X > x, \boldsymbol{\beta}\right)}{\Delta x}, \quad x \geq 0, \tag{1}$$

where **β** is a vector of mortality parameters to be estimated. From Eq. 1 we calculate the cumulative hazards function as

$$H(x \mid \boldsymbol{\beta}) = \int_0^x \mu(t \mid \boldsymbol{\beta}) dt. \tag{2}$$

From Eqs. 1 and 2, a number of demographic functions are derived, particularly the survival function

$$S(x \mid \boldsymbol{\beta}) = \Pr\left(X > x \mid \boldsymbol{\beta}\right) = e^{-H(x \mid \boldsymbol{\beta})}, \tag{3a}$$

the cumulative distribution function (CDF) of ages at death

$$F(x \mid \boldsymbol{\beta}) = \Pr\left(X < x \mid \boldsymbol{\beta}\right) = 1 - S(x \mid \boldsymbol{\beta}), \tag{3b}$$

and the probability density function (PDF) of ages at death

$$f(x \mid \boldsymbol{\beta}) = \frac{d}{dx} F(x \mid \boldsymbol{\beta}) = \mu(x \mid \boldsymbol{\beta}) S(x \mid \boldsymbol{\beta}). \tag{3c}$$

Mortality models tested

We explored four different functional forms for the mortality function in Eq. 1. First a model with constant mortality

$$\mu_0(x \mid \boldsymbol{\beta}) = \beta_0, \tag{4a}$$

where $\beta_0 > 0$, which assumes that mortality does not change with age. Second, the Gompertz mortality model [42], given by

$$\mu_0(x \mid \boldsymbol{\beta}) = \exp(\beta_0 + \beta_1 x), \tag{4b}$$

where $\beta_0 \in \mathbb{R}, \beta_1 > 0$. Here β_0 is the baseline mortality (i.e. when $x = 0$) and mortality increases exponentially with age at a rate determined by parameter β_1. Third, the Weibull mortality model [43], given by

$$\mu_0(x \mid \boldsymbol{\beta}) = \beta_0 \beta_1 (\beta_1 x)^{\beta_0 - 1}, \tag{4c}$$

where $\beta_0, \beta_1 > 0$, where β_0 is the shape parameter and β_1 is the scale parameter. This model assumes that mortality increases (or decreases) as a power function of age. Finally, the logistic mortality model [44,45], given by

$$\mu_0(x \mid \boldsymbol{\beta}) = \frac{\exp(\beta_0 + \beta_1 x)}{1 + \frac{e^{\beta_0}}{\beta_1} \beta_2 \left(e^{\beta_1 x} - 1\right)}, \tag{4d}$$

where $\beta_0 \in \mathbb{R}$, $\beta_1, \beta_2 > 0$. It has been shown that this model is the solution to the Gamma-Gompertz model that incorporates the effect of individual differences in "frailty" on mortality [44]. This frailty is a product of all causes inducing among-individual variation in risk of death (in this study we account for frailty induced by allocating resources to reproduction). In the Gamma-Gompertz model, frailty measures each individual's life-long capacity to survive. Individual frailty is treated as a random variable *Z*, with individual values given by $z > 0$, and it is assumed to follow a Gamma distribution with mean equal to 1 and Var $Z = \beta_2$. Thus, in a population where individuals have the same frailty value (i.e. $z_1 = z_2 = \ldots = 1$), then $\beta_2 = 0$ and the model converges to a simple Gompertz mortality function. As the variability in individual frailty increases, the model develops a mortality plateau at older ages.

We extended the models to account for the effect of what has been commonly described as age-independent mortality, with the addition of a "Makeham term" such that mortality becomes

$$\mu(x \mid \boldsymbol{\beta}, c) = c + \mu_0(x \mid \boldsymbol{\beta}), \qquad 5$$

where $c > 0$, is commonly described as the "age independent" mortality. Finally, we tested "bathtub" or "U-shaped" models that allow declines in early mortality, given by

$$\mu(x \mid \boldsymbol{\beta}, \boldsymbol{\alpha}, c) = \exp\left(\alpha_0 - \alpha_1 x\right) + c + \mu_0(x \mid \boldsymbol{\beta}), \qquad 6$$

where $\alpha_0 \in \mathbb{R}, \alpha_1 > 0$ are the parameters that account for the potential decline in early mortality with age, and parameter $c > 0$ is as described above.

We tested the effect of average lifetime reproductive investment as a continuous covariate under a proportional hazards framework, given by

$$\mu(x, z \mid \boldsymbol{\theta}, \boldsymbol{\gamma}) = \exp(\boldsymbol{\gamma} z)\, \mu_0(x \mid \boldsymbol{\theta}), \ \text{ for } x \geq 0, z \in \mathbb{R}, \qquad 7$$

where covariate *z* is the average lifetime reproduction. In addition, we tested the effect of categorical covariates (i.e. study area and mowing treatments) as multilevel effects on the mortality parameters. For instance, let y_i be an indicator for location such that y_i = 1 if individual *i* belongs to location *A* and y_i = 0 if it belongs to location *B*. Thus, for an individual *i* we have that the Gompertz mortality in Eq. 4b would be

$$\mu(x_i, y_i \mid \boldsymbol{\beta}) = \exp\left[\underbrace{\left(\beta_{0A} y_i + \beta_{0B}(1-y_i)\right)}_{\beta_0} + \underbrace{\left(\beta_{1A} y_i + \beta_{1B}(1-y_i)\right)}_{\beta_1} x_i\right], \text{ for } i = 1, \ldots, n, \qquad 8$$

where $\beta_{0A}, \beta_{0B} \in \mathbb{R}$, $\beta_{1A}, \beta_{1B} > 0$.

The fits of the resulting 24 models were compared based on their DIC (Deviance Information Criterion) [46,47], which is a Bayesian analogue to commonly used information criteria such as the AIC and the BIC. From the selected model we calculated senescence rates as the first derivative of the logarithm of the mortality function, given by $a_x = \frac{d}{dx}\ln\left[\mu(x \mid \cdots)\right]$.

Results

Mortality increased with age since first reproduction in both study areas and in both control and mown plots (figure 1, electronic supplementary material, figure S1). The overall mean age at death across study areas and treatments was 6.4 years after first reproduction (median = 4 years). Two individuals first recorded in 1981 were still alive in 2013. The best fitting mortality model was a Weibull model (table 1), where mortality increases with age albeit decelerating as age progresses, producing declining rates of senescence (the second-best fitting logistic model provided qualitatively similar results; electronic supplementary material, figure S2). In the control plots, mortality doubled between the first and thirtieth year after first reproduction (from 0.07 to 0.14 and 0.04 and 0.08 in the inland and coastal study area, respectively).

Mortality was also affected by reproductive investment (figure 2), with a higher average reproductive investment being associated with higher mortality (figure 3). The mean number of flowers produced per year and individual (average life-time reproductive investment) across all plots was 1.92. The best-fitting model that did not include reproduction had a substantially worse fit (table 1, ΔDIC = 246) than the best model with reproductive investment, and was a logistic model where mortality was predicted to level off after an initial sharp increase (electronic supplementary material, figure S3). Thus, it was necessary to account for life-time reproductive investment in order to identify the increase in mortality with age also for older-aged individuals.

Mortality trajectories differed both among mown and control plots and among study areas. These differences were mainly caused by differences in the scale parameter of the Weibull mortality model (figure 1), causing average mortality to be higher in mown plots and in the inland area (with a harsher climate). Rates of senescence were similar among treatments and study areas (figure 4), but as hypothesized they were slightly higher in mown plots and in the coastal area (with a more benign climate). The logistic mortality model that did not include reproduction suggested large initial differences in rates of senescence between mown and non-mown plots that evened out for older individuals, showing that accounting for reproductive investment affects also predictions of changes in senescence rates (electronic supplementary material, figure S4).

Discussion

This study provides compelling evidence of actuarial senescence in a long-lived, tuberous and non-clonal herb. Only a few detailed studies on short-lived species have previously presented data supporting such patterns [10,48]. The very long time frame over which individuals were followed, and the fact that we accounted for average reproductive investment of individuals when quantifying mortality trajectories constitute the major differences to previous studies. A Weibull function, which can also be interpreted as an accelerated failure time model and where rates of senescence gradually change with age, provided the best-fitting mortality model. In contrast, versions of the Gompertz function with a constant, exponential rate of senescence, are typically used to describe mortality trajectories of animals [49,50]. Whether this reflects a general difference between the animal and plant kingdom remains to be seen after additional careful analyses of mortality trajectories in plants.

We also found support for the existence of a trade-off between processes increasing fertility and decreasing mortality, observing a positive relationship between these two demographic rates that has also been found in recent experimental studies [23,31]. Such life history trade-offs have only rarely been detected in observational studies (but see [51]). This may reflect that reproductive output is constrained within a limited range [14] to ensure that mortality is kept at an optimal level determined by the species' ecological strategy [29]. In fact, several previous studies report a negative correlation between reproductive investment and mortality, indicating that individuals which invest a lot in reproduction are generally fitter, either due to genetics or to past or present environmental conditions [52,53]. In contrast, our results suggest that variation in relative allocation patterns was large relative to variation in overall resource status and that individuals of *D. lapponica* differed in their reproductive strategy, so that some individuals consistently devoted more resources to reproduction at the cost of a higher mortality. Previous studies have rarely been conducted over a comparable time frame, i.e. over the life course of study species, which may also be one cause of why costs of reproduction have typically not been observed without experimental manipulations [54].

A particularly interesting result of this study was that continued senescence was only detected in older individuals when we controlled for average reproductive investment over the individual's life course. The constant population-level rate of mortality at older ages observed in models that excluded reproductive investment may be due to variation in reproductive strategies among individuals that leads to "heterogeneity in frailty" [11]. Heterogeneity in frailty among individuals should lead to within-cohort selection, where more frail individuals with higher mortality are removed from populations, which will curve mortality trajectories downwards. Such a process, not reflecting individual-level patterns but instead driven by among-individual differences, may be one cause of the "mortality plateaus" detected in animals, including humans (cf. [55]). For *D. lapponica*, as for all natural populations, it is likely that sources of such among-individual heterogeneity other than reproductive strategy exist, which would mean that rates of senescence in *D. lapponica* could be higher than that observed here and that a Weibull model with decelerating rates of senescence may no longer provide the best fit if other sources of heterogeneity are accounted for. Because we did find decelerating rates of senescence here, the logistic model (the second best model), which arises as the solution to the gamma-Gompertz frailty model, could also be appropriate ([56]; see Methods). In our case, the qualitative patterns with increasing and decelerating mortality over age were the same in both models. Interestingly, estimates of rates of senescence were much more similar among study areas and treatments when accounting for reproductive investment. These results suggest that one reason many previous studies on plants reported no increase in mortality over age may be that heterogeneity caused by reproductive investment was not accounted for, and we encourage researchers to consider fertility in future studies quantifying mortality trajectories.

Although most of the differences in mortality trajectories among study areas and treatments were explained by variation in age-independent mortality among individuals, our results also suggest small differences in rates of senescence. Baseline, age-independent mortality was higher in mown plots and in the harsher inland area. Rates of senescence, although not statistically significantly different, were slightly higher in mown plots as well, but also in the more benign coastal area. In an unpredictable environment with high baseline mortality caused by disturbance (such as mowing in this case), senescence may be more pronounced if selection has favoured increased reproductive effort early in life [57]. However, only in the harsher inland study area did mowing increase fertility [40], and as individuals in mown plots grow in close proximity to unmown plots it may be unlikely that selection for diverging life histories has produced any evolutionary effect. In a previous study, a consistent effect of mowing across study areas was that individual and population growth rates were lower in mown plots [40]. We did not analyse potential trade-offs with growth here, but the earlier observed negative effect of mowing on growth supports that faster senescence in mown plots is due to phenotypic plasticity caused by a direct weakening of individuals, rather than a higher early-life reproductive investment. We assume such a weakening to be a direct effect of mowing, but effects may also be indirect, in terms of altering strengths of biotic interactions. That rates of senescence were found to be higher in the coastal area with a more benign climate fits with the hypothesis that individuals that maximize reproductive output at the expense of maintenance would have higher fitness in favourable environments. Indeed, flowering was previously found to be more frequent in the coastal area [40].

Conclusions

The presented evidence of senescence occurring in a long-lived plant is important knowledge for developing more comprehensive theories of the evolution of senescence across the tree of life. The disposable soma theory of the evolution of senescence assumes a separation of germ and somatic cell lineages, and that no strong selection against senescence has occurred because DNA in the germ line has been repaired even though cells in the soma have been allowed to deteriorate [15].

In *D. lapponica*, the entire soma, that will also give rise to the germ line, is replaced each year (cf. Methods), so no such separation exists over a plant's life time. The results presented here are, we believe, an interesting first step in understanding mortality trajectories of such species. As a next step we will investigate the age trajectory of fertility in *D. lapponica*, and link mortality and fertility patterns by modelling them simultaneously and accounting for temporal variation in fertility. This should give us more detailed understanding of the mechanisms of how life history trade-offs are related to senescence.

Data, code and materials

Data and R code will be made available at publication.

Competing interests

We have no competing interests.

Authors' contributions

JPD and NS conceived the general study questions, JPD, FC and ORJ conceived the specific study questions, AM and DIØ collected the data, and FC conducted the analysis. JPD wrote the first draft of the manuscript, and all authors contributed substantially to revisions.

Acknowledgements

We thank JW Vaupel for comments on a previous version of the manuscript. A large number of field workers have contributed to the long-term studies; in particular we thank T Arnesen, A Lyngstad and E Moen.

Funding

The field study was conducted with financial support from the Research Council of Norway and the Directorate for Nature Management (to AM and DIØ). JPD, FC and ORJ acknowledge funding by the Max Planck Society and NS by the Swedish Research Council Formas.

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Table 1. The ten best-fitting mortality models for *Dactylorhiza lapponica* (DIC = Deviance Information Criterion).

Model	Shape	Reproductive investment included	DIC
Weibull	Makeham	yes	46461
Logistic	Bathtub	yes	46499
Logistic	Makeham	no	46707
Weibull	bathtub	no	46897
Weibull	bathtub	yes	46915
Weibull	Makeham	no	46949
Logistic	bathtub	no	46985
Logistic	Makeham	yes	47012
Gompertz	bathtub	yes	47116
Gompertz	bathtub	no	47307

Figure legends

Figure 1. Mortality (a) and survivorship (b) over age since first reproduction, and parameter values (c-e) with 95% credible intervals of the best-fitting Weibull mortality model for *Dactylorhiza lapponica* in mown and control plots at the coastal Tågdalen and the inland Sølendet study areas in central Norway. *c* is the Makeham term (c), b_0 is the shape parameter β_0 (d), and b_1 is the scale parameter β_1 (e). Mortality and survivorship are presented from the first reproductive event (age = 0) until the age where survivorship is $S(x) = 0.01$ (when the model predicts that 99% of the cohort is dead). The model also included average life-time reproductive investment as a covariate (cf. figure 2). See figure S1 for a presentation of the underlying data, in terms of nonparametric estimations of survivorship curves (Kaplan-Meier curves).

Figure 2. The posterior density of the average life-time reproductive investment parameter (γ) in the best-fitting Weibull mortality model.

Figure 3. Age trajectories of mortality (right y-axis) of *D. lapponica* in all four treatment – study area combinations, as functions of average life-time reproductive investment (low, mean and high number of flowers: empirical density presented to the left of the trajectory plots). The central lines show the predicted mortality trajectory for a group of individuals with mean value of average life-time reproductive effort. The upper and lower lines show the mortality trajectories for the upper and lower 95% levels in life-time reproductive effort, with higher reproductive efforts leading to higher mortality.

Figure 4. Rates of senescence (ageing rates) of *D. lapponica* in all treatment – study area combinations predicted from the best-fitting Weibull model, with life-time reproductive investment accounted for. These senescence rates correspond to the first derivative of the logarithm of the mortality functions presented in figure 1.

Fig 1.

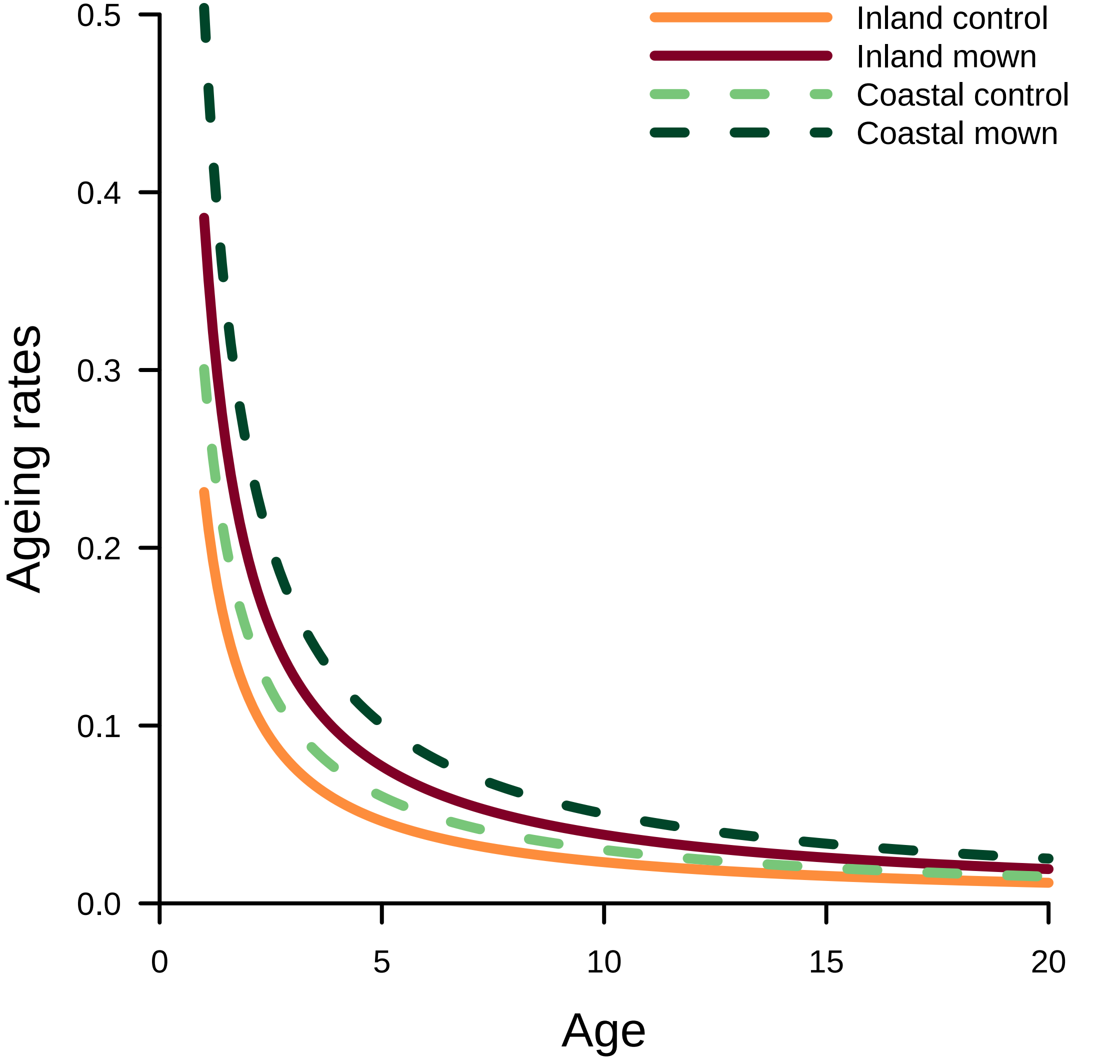

Inland control
Inland mown
Coastal control
Coastal mown
0.5
0.4
0.3
0.2
0.1
0.0
Ageing rates
0
5
10
15
20
Age

Fig. 2

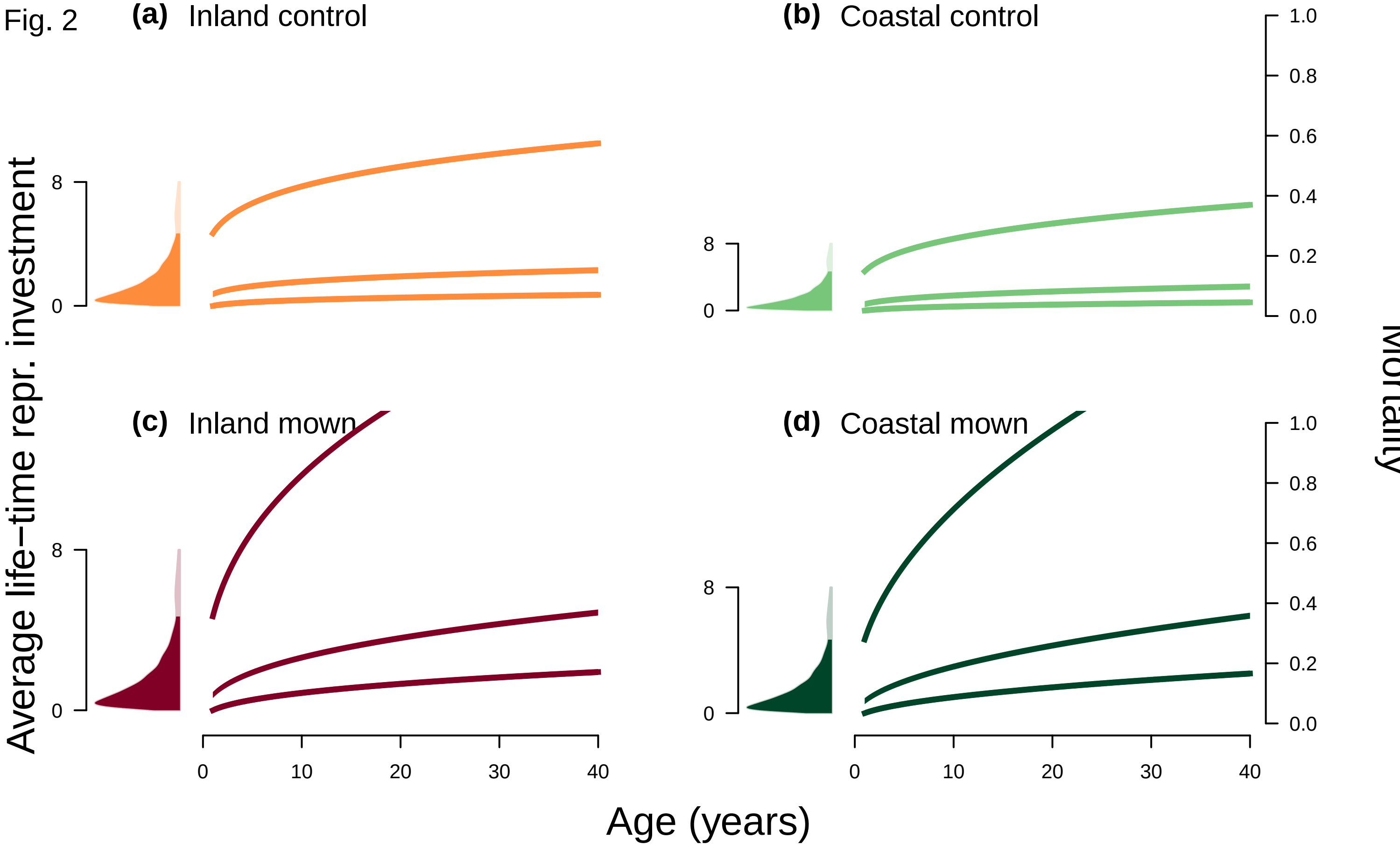

(a) Inland control
(b) Coastal control
(c) Inland mown
(d) Coastal mown
Average life−time repr. investment
Mortality
Age (years)
0
8
0
10
20
30
40
0.0
0.2
0.4
0.6
0.8
1.0

Fig. 3

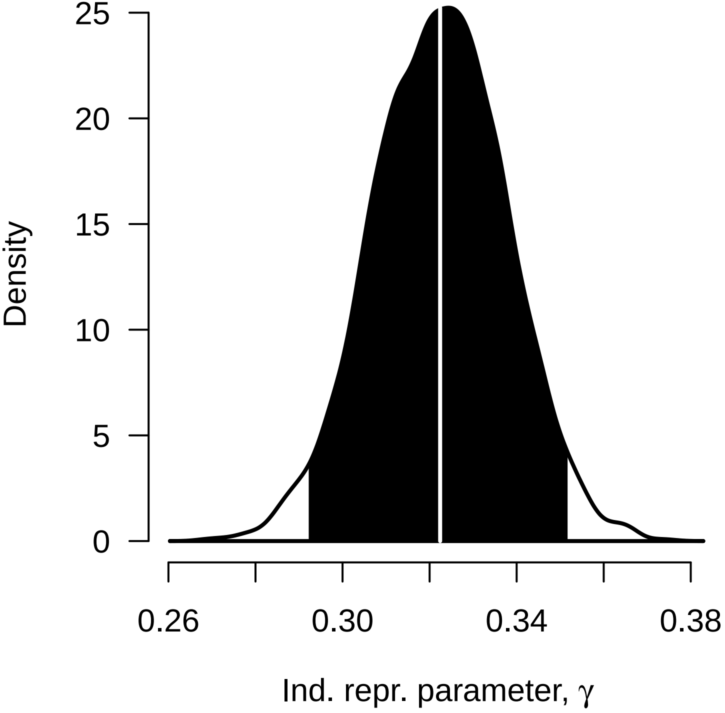
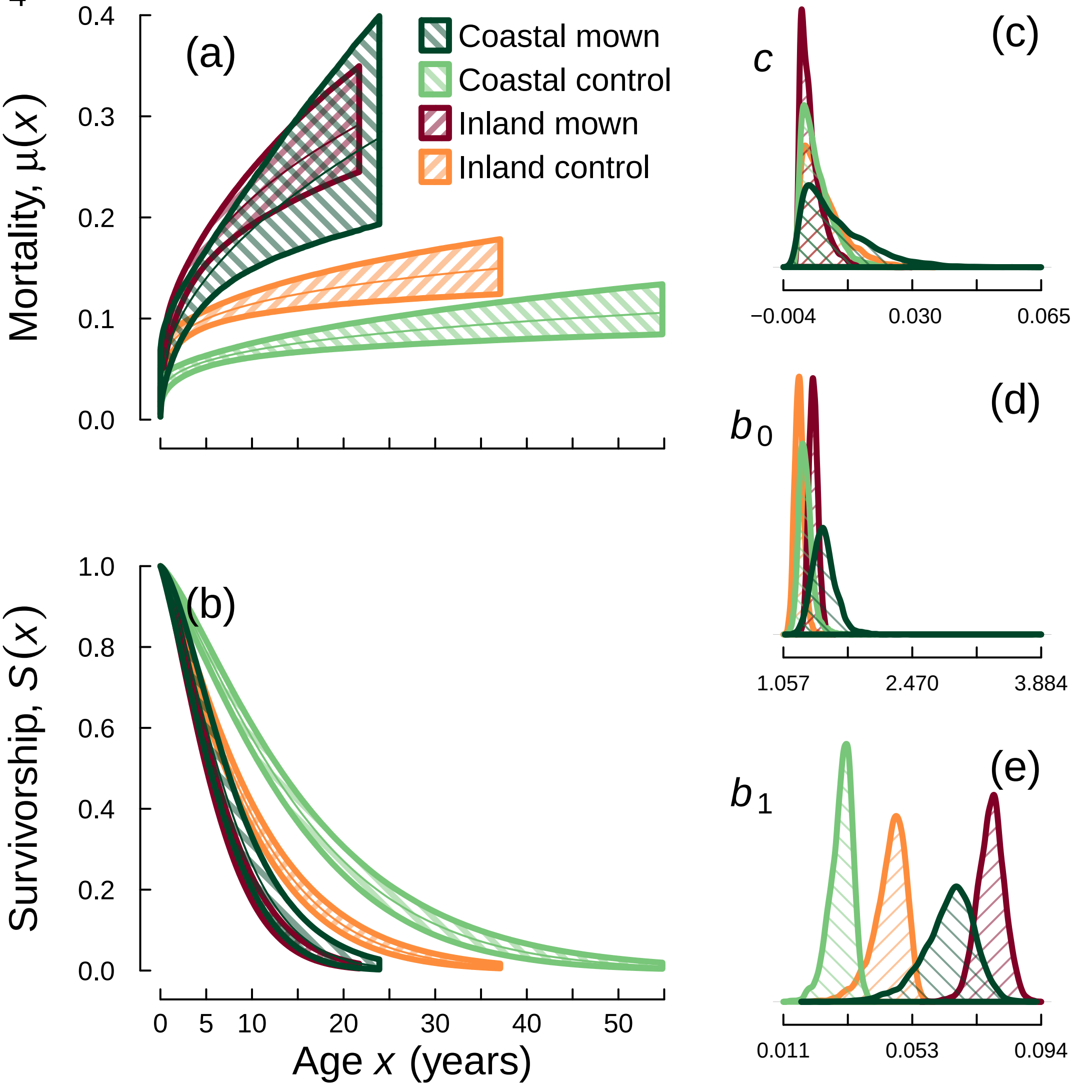


Fig. 4



(a)
(b)
(c)
(d)
(e)
Coastal mown
Coastal control
Inland mown
Inland control
Mortality, μ(x)
Survivorship, S(x)
Age x (years)
0.0
0.1
0.2
0.3
0.4
0.0
0.2
0.4
0.6
0.8
1.0
0
5
10
20
30
40
50
c
b 0
b 1
−0.004
0.030
0.065
1.057
2.470
3.884
0.011
0.053
0.094

Electronic supplementary material

'Actuarial senescence in a long-lived orchid challenges our current understanding of ageing'

Dahlgren *et al*. Submitted to *Proc R Soc B Biol Sci* 2016

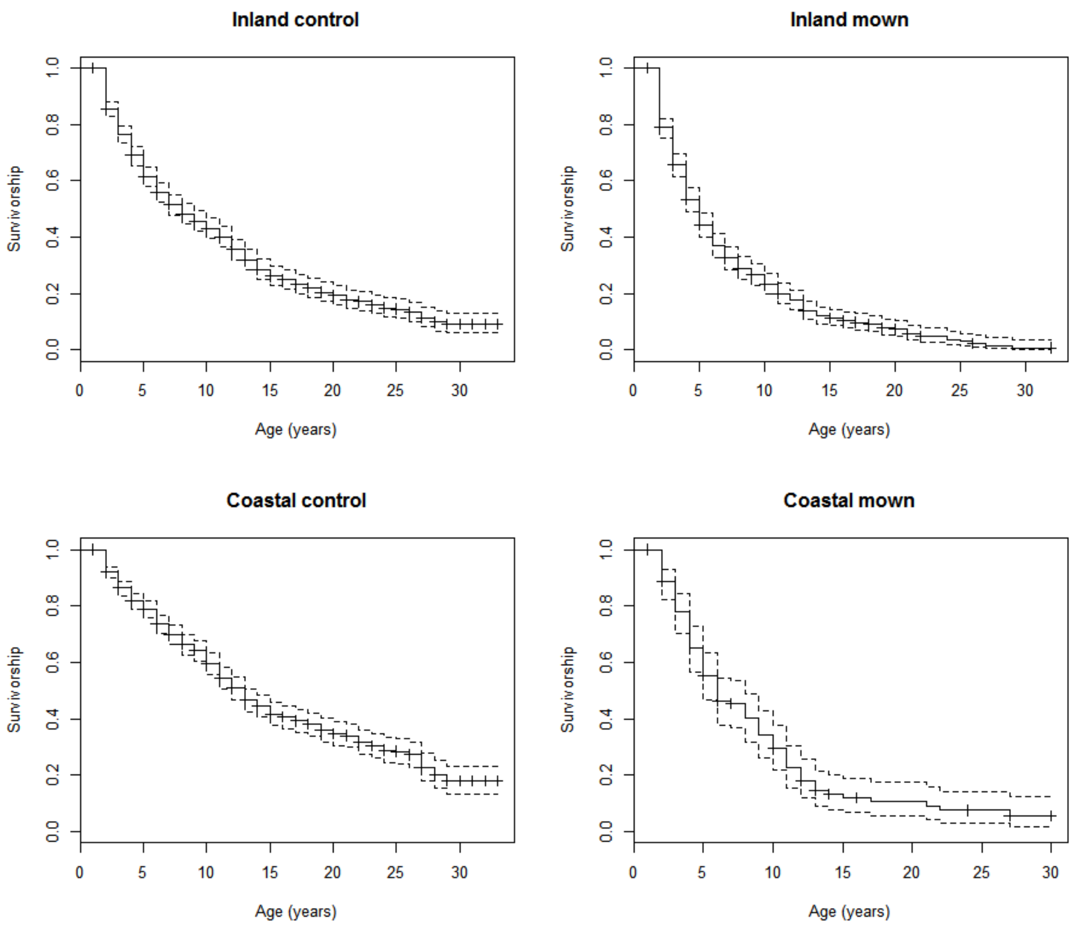


Figure S1. Plots of the Kaplan-Meier estimators of the survivorship curves.

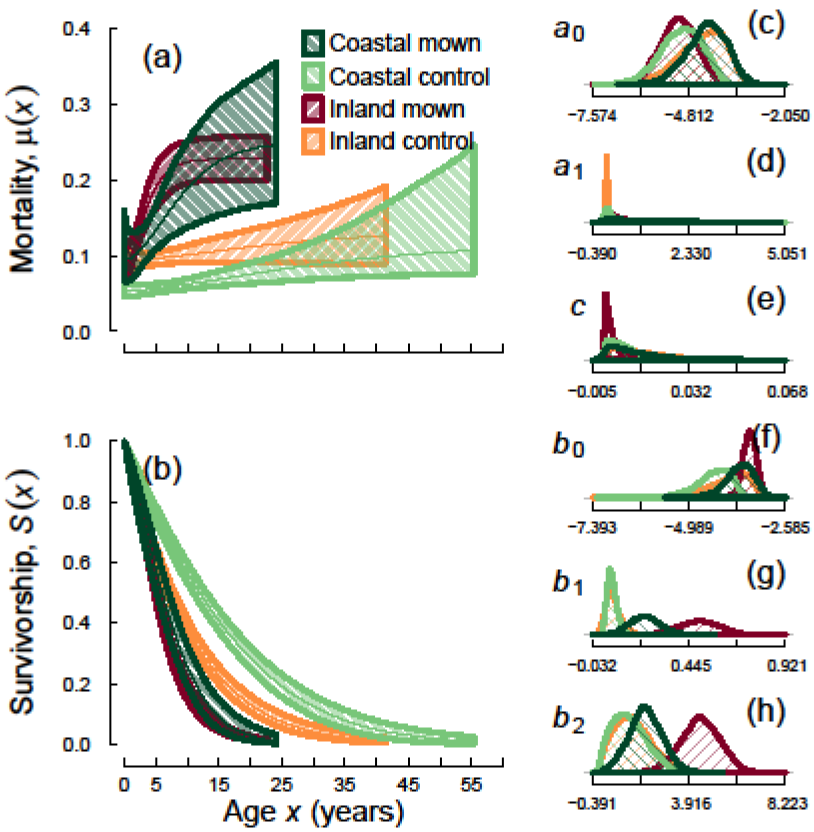


Figure S2. Mortality (a) and survivorship (b) over age since first reproduction, and parameter values (c-h) with 95% credible intervals of the second-best fitting mortality model (logistic including reproductive investment and no Makeham term). Mortality and survivorship are presented from the first reproductive event (age = 0) until the age where survivorship is S(x) = 0.01 (when the model predicts that 99% of the cohort is dead).

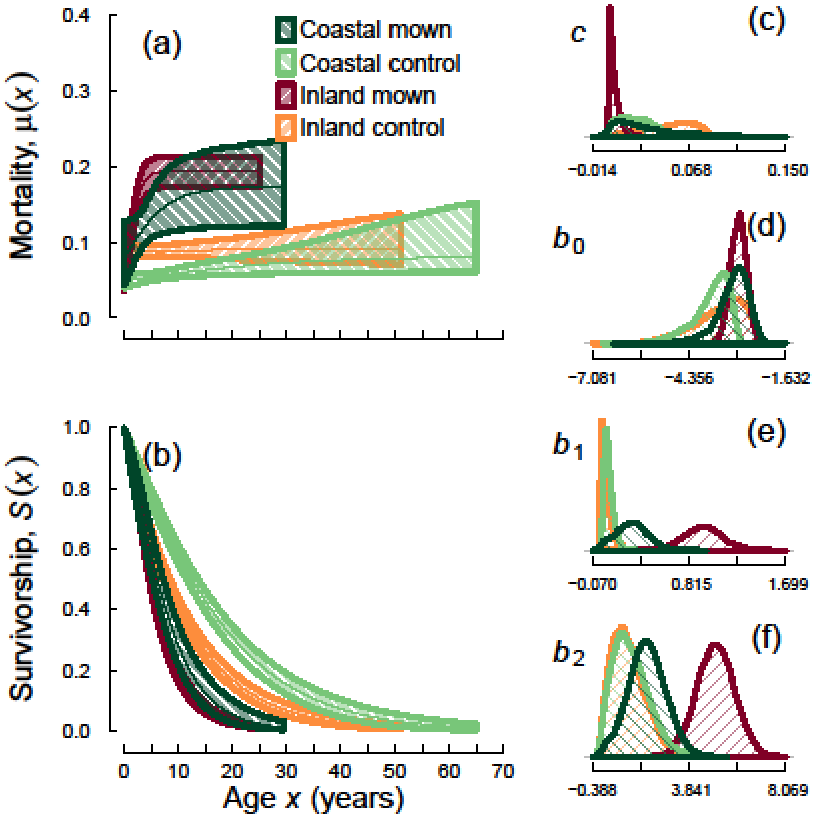


Figure S3. Mortality (a) and survivorship (b) over age since first reproduction, and parameter values (c-f) with 95% credible intervals of the best-fitting mortality model that did not include reproductive investment (the third-best fitting model overall; logistic with a Makeham term). Mortality and survivorship are presented from the first reproductive event (age = 0) until the age where survivorship is S(x) = 0.01 (when the model predicts that 99% of the cohort is dead).

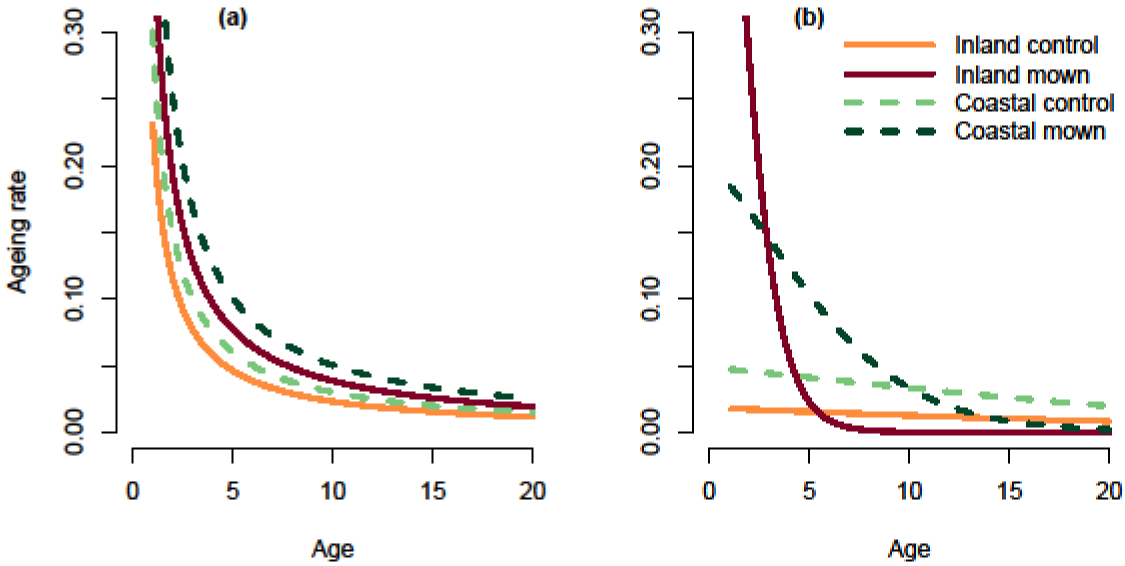


Figure S4. Predictions of senescence rates from the best-fitting Weibull model with reproductive investment accounted for (a; identical to fig. 4 in the manuscript), and predictions from the best-fitting logistic model excluding reproductive investment (b) (see figure S3).