Research

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Age-specific variation in survival, reproductive success and offspring quality in red squirrels: evidence of senescence

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Individual performance is expected to decrease with age because of senescence. We analyzed long-term data collected on a North American red squirrel population to assess the influence of age on body mass, survival and reproductive performance, and to study the effects of sex and of environmental conditions during early life on senescence patterns. Mass of males and females did not decrease at the end of life, possibly because body mass mostly reflects overall size in income breeders such as red squirrels. On the other hand, we found evidence of senescence in survival of both sexes and, to a lesser extent, in female reproductive traits. When compared to females, males had both higher survival and delayed decrease in survival, suggesting a weaker senescence in males. The offspring survival from weaning to one year of age also decreased with increasing mother age. This suggests that older females produce juveniles of lower quality, providing evidence of an intergenerational effect of mother's age on juveniles' fitness. Finally, our results indicate that variations in food conditions during early life influenced the reproductive tactics of females in the first years of their life, but not senescence patterns.

The assessment of life history changes with age in animal populations is a key issue in ecology. Most vertebrate populations are strongly age-structured (Charlesworth 1994) so that age-specific life history traits are required to understand both the evolution of life histories (Partridge and Harvey 1988) and population dynamics (Caswell 2001). Life history theory predicts that both survival and reproduction should decrease with age because of senescence, a decrease in functional capacities at old ages (Hamilton 1966, Comfort 1979). The study of senescence in wild populations has proven to be challenging because of the need for long-term monitoring of individuals. Evidence for senescence has been reviewed by Bennett and Owens (2002) for birds and by Gaillard et al. (2003a) for large mammalian herbivores. Both studies concluded that senescence in survival and reproductive success is pervasive in those species. Evidence for senescence in free-ranging populations of small mammals has been more limited (Slade 1995), but some studies reported decreased survival (Broussard et al. 2005) or reproduction (Morris 1996, Broussard et al. 2003) with increasing age.

We have been monitoring the survival and reproductive performance of individual North American red squirrels in the southwestern Yukon for 15 years. Red squirrels are active year round (Humphries et al. 2005), and do not store energy in the form of fat but rather cache conifer cones that are collected in autumn (before they shed their seed) and stored unopened underground. Squirrels rely on this food cache through winter and into the following spring. North American red squirrels are part of a consumer–resource pulse system, with the main food resource (i.e. seeds of white spruce) exhibiting dramatic yearly variations (Boutin et al. 2006). Seed production is highly variable because the conifer trees in the area show mast behaviour and many life history traits in this red squirrel population are strongly affected by seed production (Humphries and Boutin 2000, McAdam and Boutin 2003, Boutin et al. 2006).

Among the theories proposed to explain the occurrence of senescence, the antagonistic pleiotropy theory (Williams 1957) has received the most support in animals (Partridge 2001). In the context of life history evolution, this antagonistic pleiotropy theory involves a tradeoff between a positive effect of genes at a young age and a negative effect of the same genes at old age that should shape age-specific allocation to reproduction. From a life history viewpoint, this has led to the concept of the disposable soma (Kirkwood and Rose 1991). There should therefore be a direct link between senescence and reproductive tactics of individuals (see Nussey et al. 2006 for a recent example in red deer). Environmental conditions in the year of birth have been reported to influence reproductive tactics, so that individuals born in good years, or more generally in good environments, begin to reproduce earlier in life and/or breed more intensively than individuals born in poor years (Langvatn et al. 1996, Forchhammer et al. 2001, Beckerman et al. 2003). These individuals might, however, pay the cost of a good start in terms of lower reproductive values at old ages (Alonso-Alvarez et al. 2006). The dramatic yearly variation in food supply for red squirrels in this population (Boutin et al. 2006) offered us a unique opportunity to investigate the influence of environmental conditions during early life on senescence patterns in a short-lived species.

Given the biology and life history of red squirrels, we made the following predictions. First, given that red squirrels are iteroparous vertebrates with age-structured populations, we expected senescence to affect both reproductive and survival traits (Hamilton 1966, Charlesworth 1994). Also, following Hamilton's (1966) prediction and Jones et al.'s (2008) empirical report of an increase in the strength of senescence with decreasing generation length, we predicted higher effects of senescence on survival and reproduction (i.e. a larger decrease in those traits with increasing age) in red squirrels than in long-lived vertebrates such as large mammals.

Male squirrels are only slightly (5–10%) larger than females (Boutin and Larsen 1993), and have the same duration of reproductive life as females. We therefore predicted that males and females should experience similar rates of senescence (Clutton-Brock and Isvaran 2007).

Reproductive senescence in natural populations is generally assessed through a decrease in reproductive success. However, senescent individuals may raise the same number of offspring, although of lower quality (Kern et al. 2001). When simply considering the number of juveniles weaned by a female to assess reproductive senescence, the rate of senescence may thus be underestimated. We thus measured whether post-weaning survival of juvenile squirrels had any relation to age of their mother, with the assumption that post-weaning survival reflects the quality of juveniles. We predicted that post-weaning survival of juveniles produced by old females should be lower than that of juveniles produced by prime-aged females.

Lastly, we predicted that cohorts born during high food years should have higher senescence rates than cohorts born during low food years because high-food cohorts should reproduce earlier and more intensely than low-food cohorts, and thereby suffer higher reproductive costs (Kirkwood and Rose 1991, Metcalfe and Monaghan 2001).

Methods

Study area and field methods

The study was conducted near Kluane Lake, Yukon, Canada (61°N, 138°W) and began in 1987 (Berteaux and Boutin 2000, McAdam et al. 2007). We report results for the years 1987–2003 for two 40 ha grids separated by the Alaska Highway. All squirrels were marked with numbered ear tags and a unique color combination of wires for identification at a distance. Fates of male and female squirrels were determined through live-trapping, visual observations and/or radio-telemetry from April to August.

Reproductive females were weighed after parturition, and males and females were weighed each time they were trapped (to the nearest gram with a pesola scale). Young of the year were counted, weighed, and sexed soon after birth, tagged and weighed at about 20–25 days of age, and weighed within a week of weaning (around 70 days of age). The average dispersal distance after weaning is less than 100 m (Berteaux and Boutin 2000), so that most young were followed from birth to death. The habitat was boreal forest with white spruce *Picea glauca* as the dominant tree species and spruce seeds husked from cones were the main food resource (McAdam and Boutin 2003). Red squirrels store unopened cones at hoarding sites (middens) that they defend actively throughout the year. Food available for reproduction in a given year is a function of cone production the previous year (McAdam and Boutin 2003). We estimated spruce cone production each year, when cones were fully formed, but harvesting by squirrels had not yet begun (usually at the end of July). We counted the number of new cones in the top three meters of 170 randomly selected trees. Average yearly cone production ranged from about 1 to 283 cones per tree (average 60 ± 20 SE).

Analyses of the influence of age on mass and reproduction

Our data set included repeated measurements of the same individuals at different ages (from 1.3 to 2.4 measures per individual on average, depending on the trait considered; range 1–6). We thus first fitted mixed models (with the MIXED procedure and GLIMMIX macro of SAS ver. 9, Littell et al. 1996) with individual identity included as a random effect to take into account this non-independence in our data set and to avoid pseudo-replication problems (Hurlbert 1984). However, considering that linear models with and without a random effect of individual identity led to the same results, and considering the small number of observations per individual, we also performed linear models without random effect to study age-specific variation in mass (of adult males and females, and of juveniles at weaning), litter size (at birth and at weaning), and number of juveniles recruited, and we only presented these results. We used the GLM and NLIN (to fit piecewise regressions, see below) procedures of SAS ver. 9 for the mass of males and females, mass of juveniles at weaning, litter size at birth and number of recruited juveniles, and the NLMIXED procedure for the number of weaned juveniles. For traits analysed with the GLM and NLIN procedures (models based on a normal error distribution), normality of residuals was assessed with Kolmogorov–Smirnov tests. A normal error distribution was rejected only for litter size (mass of females: $p > 0.15$; mass of males: $p = 0.091$; litter size at birth: $p = 0.011$; average mass of juveniles at weaning: $p > 0.15$; number of recruited juveniles: $p > 0.15$). However, the Kolmogorov–Smirnov test is very conservative (Quinn and Keough 2002) and a graphical inspection of residuals plotted against fitted values of litter size did not reveal any tendency. We thus analyzed variation in litter size at birth with a normal error distribution. For the number of weaned juvenile, we used a Poisson error distribution. The dispersion parameter (c) was close to 1 ($\hat{c} = 1.13$), indicating negligible over-dispersion (Burnham and Anderson 2002).

To assess age-dependence in the life history traits of squirrels, we fitted and compared a set of continuous (linear, quadratic, inverse effect of age) and discrete (two or three age-classes) models (Table 1a). Each model considered (Table 1a) can be seen as a specific hypothesis regarding age-specific variation (Burnham and Anderson 2002) and support for models showing decreasing values with age could be considered as evidence for the existence of senescence (Table 1a). Because there were many more young individuals than old ones in our database, the selection of a significant negative quadratic effect for a given trait could have arisen when the trait increased with age and then levelled of. Therefore, we also considered piecewise regression models that test for the existence of breakpoints, after which traits are declining (Toms and Lesperance 2003). We fitted models with different threshold ages (i.e. breakpoint at age 3, 4, 5 or 6 years of age; Table 1a) with the NLIN and NLMIXED procedure in SAS ver. 9. We also fitted models including a quadratic or a linear effect of age before the breakpoint and a linear or no effect of age after the breakpoint (Table 1a).

Table 1. Relation of mass, survival and reproductive performance to age and food conditions in the year of birth in a population of North American red squirrels, Kluane, Yukon, Canada.
(a) Variables describing age effect used to test the shape of the relationship between life-history traits and age.

Age effect	Notation
Full-age model: 8 classes (1 class per year)	age (8 classes)
linear regression	age
quadratic regression	age+age^2
inverse regression*	1/age
1 juvenile class (1 year) and 1 adult class ($\geq$2 year)	(yearling, adult)
1 prime-age class (1–6 years) and 1 senescent class ($\geq$7 years)	(prime age 1–6, senescent $\geq$7)
1 prime-age class (1–5 years) and 1 senescent class ($\geq$6 years)	(prime age 1–5, senescent $\geq$6)
1 prime-age class (1–4 years) and 1 senescent class ($\geq$5 years)	(prime age 1–4, senescent $\geq$5)
1 juvenile class (1 year), 1 prime-age class (2–6 years), 1 senescent class ($\geq$7 years)	(yearling, prime age 1–6, senescent $\geq$7)
1 juvenile class (1 year), 1 prime-age class (2–5 years), 1 senescent class ($\geq$6 years)	(yearling, prime age 1–5, senescent $\geq$6)
1juvenile class (1 year), 1 prime-age class (2–4 years), 1 senescent class ($\geq$5 years)	(yearling, prime age 1–4, senescent $\geq$5)
Linear regression before and after breakpoint (α)$^\$$	(linear$_{1-\alpha}$ $\times$ linear$_{\alpha-8}$)
Quadratic regression before breakpoint and linear after (α)$^\$$	(quad$_{1-\alpha}$ $\times$ linear$_{\alpha-8}$)
Linear regression before breakpoint and constant after (α)$^\$$	(linear$_{1-\alpha}$ $\times$ constant $_{\alpha-8}$)
Quadratic regression before breakpoint and constant after (α)$^\$$	(quad$_{1-\alpha}$ $\times$ constant$_{\alpha-8}$)

*: this model corresponds to a non linear effect of age (increase in the life history trait at the lowest ages gradually followed by a plateau at the highest ages).

\$: 4 different threshold (α) ages have been considered: 3, 4, 5 and 6 years.

Model selection was based on the Akaike information criterion (AIC), as recommended when several non-nested models have been fitted (Burnham and Anderson 2002). AIC-based model selection has proved to be a very efficient way to test ecological hypotheses and can be seen as a 'multiple working hypotheses' framework (Chamberlin 1890, Anderson et al. 2000). We based our model selection on ΔAICc (difference in AICc, a variant of AIC corrected for small sample sizes, between a given model and the model with lowest AICc) and models' weight (w_i). W_i measures the relative likelihood that a given model is the best among a set of models fitted (Burnham and Anderson 2002).

We first tested for an age effect on the mass of squirrels (used as a proxy of body condition). For females, we considered mass after parturition (taking litter size into account) as these data were available for most females and were standardized for the same breeding period. For males, we used the average mass in May because May was a period of high trapping activity, and most males present on the study area were trapped at least once during this period. We then studied the effect of females' age on litter size at birth (taking mother's mass after parturition into account), on the number of juveniles weaned (whether taking litter size at birth into account or not) and on the juvenile mass at weaning (mean mass per litter after accounting for variation in litter size at birth). Finally, we investigated the effect of mothers' age on the number of juveniles recruited (i.e. number of juveniles weaned surviving to one year of age in a given litter). We only included breeding females for analyzing the age-variation in weaning success and only females who weaned at least one juvenile for analyzing the age variation in recruitment.

It has been found that old females bequeath all or part of their territory to their young more often than younger ones (Berteaux and Boutin 2000, Descamps et al. 2007). This behaviour likely influenced survival of young after weaning. To test the effect of mother's age on recruitment, we thus included in our models a factor called bequeathal that described mother's behaviour at weaning (with three modalities: female kept its territory, female gave part of its territory to one or several of its juveniles, and female gave all its territory to one or several of its juveniles).

No known-age individual >5 years old was present in the study population before 1993 because tagging of squirrels started in 1987. Therefore, to account for potential variations in environmental conditions experienced by different age-classes, we included in our models a discrete variable year.

Survival analyses

Survival analyses were carried out using capture–mark–recapture methods (Lebreton et al. 1992), implemented in the software M-SURGE 1.7 (Choquet et al. 2004, 2005). Goodness-of-fit tests were performed with the software U-CARE (Choquet et al. 2003). As we focused on senescence patterns, we only considered squirrels greater than one year of age. Our dataset consisted of 343 females and 216 males of known age, tagged as juveniles between 1987 and 2001. Male and female data fitted the Cormack–Jolly–Seber model (females: $\chi^2_{26} = 32.57$, $p = 0.18$; males:

Table 1(b). Results of the model selection for survival and mass of males and females, and reproductive parameters of females. n_{obs} indicates the number of measurements (or number of '1' in the matrix of presence–absence used for survival analyses) and n_{ind} the number of individuals measured. Dev indicates the deviance of the model considered; ΔAICc indicates the difference in AICc between the model with lowest AICc and the model considered; w_i indicates the AICc weight of a given model among the whole set of models fitted in Table 1a; R^2 represents the proportion of variance (or deviance) explained by the model. For mass and reproductive parameters, it has been calculated as $1-\frac{\Sigma(y_i - \hat{y}_i)^2}{\Sigma(y_i - \bar{Y})^2}$, where $\bar{y}$ represents the average value for the trait considered, $\hat{y}_i$ the predicted value for individual i and Y_i the observed value for individual i. For survival, it has been calculated as: $\frac{\mathrm{Dev}_i - \mathrm{Dev}_{Null}}{\mathrm{Dev}_{age.year} - \mathrm{Dev}_{Null}}$, where Dev_i represents the deviance of the model of interest, Dev_{Null} the deviance of the null model and $\mathrm{Dev}_{age.year}$ the deviance of the model including the interaction between age (8-modality) and year. Only results for the best model, the full-age dependent model and the model with no age effect are shown (see Supplementary material Appendix 2 for further details on model selection).

Life history trait	Model	np	Dev	ΔAICc	w_i	R^2
Mass of females ($n_{obs}=423$, $n_{ind}=226$)	1/age+litter size+year	17	2494.515	0.000	0.171	0.47
	age (8 classes)+litter size+year	23	2487.745	6.486	0.007	0.48
	litter size+year	16	2522.504	25.818	0.000	0.44
Mass of males ($n_{obs}=483$, $n_{ind}=205$)	1/age+year	16	2674.328	0.000	0.469	0.35
	age (8 classes)+year	23	2671.013	11.923	0.001	0.36
	year	16	2761.002	86.674	0.000	0.23
Survival of females ($n_{obs}=971$, $n_{ind}=343$)	age+age^2+year	18	1269.648	0.000	0.305	0.66
	age (8 classes)+year	23	1268.390	13.400	0.000	0.67
	year	16	1307.952	34.156	0.000	0.38
Survival of males ($n_{obs}=595$, $n_{ind}=216$)	(prime age 1–6, senescent ≥7)	4	880.767	0.000	0.221	0.10
	age (8 classes)	11	874.874	8.492	0.003	0.18
	null model	3	888.151	5.357	0.015	0.00
Litter size ($n_{obs}=580$, $n_{ind}=259$)	(linear_{1-6}; linear_{6-8})+mass+year	18	−226.769	0.000	0.237	0.16
	age (8 classes)+mass+year	23	−235.566	2.277	0.078	0.18
	mass+year	16	−217.939	4.477	0.026	0.14
Number of juveniles weaned ($n_{obs}=580$, $n_{ind}=259$)	(yearling, prime age 2–6, senescent ≥7) +litter size+year	18	1501.222	0.000	0.424	0.24
	age (8 classes)+litter size+year	23	1500.062	9.607	0.003	0.25
	litter size+year	16	1516.859	11.384	0.001	0.20
Average mass of juveniles at weaning ($n_{obs}=258$, $n_{ind}=159$)	(linear_{1-3}; constant_{3-8})+litter size+year	16	1537.311	0.000	0.158	0.19
	age (8 classes)+litter size+year	20	1535.653	7.629	0.003	0.20
	litter size+year	15	1544.995	5.410	0.011	0.17
Number of juveniles recruited ($n_{obs}=286$. $n_{ind}=212$)	(yearling, prime age 2–5, senescent ≥6)+number weaned+ bequeathal+year	20	−306.062	0.000	0.278	0.61
	age (8 classes)+number weaned+bequeathal+year	24	−309.666	5.824	0.015	0.62
	number weaned+bequeathal+year	18	−295.920	5.534	0.017	0.60

χ^2_{33} =28.56, p = 0.69), which was thus used as a starting point for model selection. Because preliminary analyses indicated that recapture probabilities and survival were sex-dependent, we analyzed age-specific variation in survival by considering sexes separately. We used trapping and visual observations from mid-April to mid-June, which corresponded to a period of high trapping intensity (i.e. census of all squirrels in the study area) to identify the individual capture histories. As survival rates were the parameters of interest in our study, we first modelled capture rates to have more statistical power when modeling survival (Lebreton et al. 1992). We compared among models with constant, time-dependent, age-dependent capture rates and capture rates that were a function of capture effort (defined as the average number of times squirrels were captured every year between mid-April and mid-June). For males, the selected model included capture rates that were a function of capture effort, and for females, the selected model included constant capture rates (average capture rates ± SE were 0.88 ± 0.02 and 0.95 ± 0.01 for males and females, respectively; Supplementary material Appendix 1). As for analyses of mass and reproduction, model selection was based on ΔAICc and weights of models (Burnham and Anderson 2002), and notation and biological meaning of models are reported in Table 1a.

Analyses of cohort effects

To investigate how food conditions in the year of birth influenced patterns of age-specific variation, we included in the previously retained models a variable describing food conditions during rearing (i.e. cone production the year before birth, McAdam and Boutin 2003). For each trait, we compared a model with no cohort effect, a model with an additive effect of cohort, and a model with interactive effects between age and cohort, based on AICc. Following McAdam and Boutin (2003), we considered as high-food cohorts squirrels born in years when previous cone production was >50 cones tree^{-1} (range: 73–283 cones tree^{-1}; 5 years out of 17), and as low-food cohorts squirrels born in years when previous cone production was <50 cones tree^{-1} (range: 1–47 cones tree^{-1}; 12 years out of 17).

All results are reported as mean ± SE.

Results

Effects of age on mass

The average mass of females after parturition was 258.7 g ± 1.3 and the average mass of males in May was 257.3 g ± 0.9. After adjusting for yearly variations in environmental conditions (and for litter size for the mass of females), the best age-dependent model for female and male mass included an inverse effect of age (Table 1b; see Supplementary material Appendix 2a–b for complete results of model selection; parameter estimates ± SE for the variable '1/age' were −20.44 ± 3.88 and −25.02 ± 2.61 for females and males, respectively). The mass of both sexes increased until about four years of age and remained constant afterwards (Fig. 1). For males, the model including an inverse effect of age had >7 times more support (i.e. its AIC-weight was >7 times higher) than other models including a decrease in mass after age 3, indicating that a decrease in mass of males at old ages was very unlikely. For females, results were less clear. The model including an inverse effect of age (i.e. a constant mass for females after four years old) was only 1.4 times more supported than the model including a linear positive effect of age from one to five years followed by a linear negative effect afterwards. Thus, our results did not allow rejection of the hypothesis of lower mass for females older than five. Finally, when testing for cohort effects, the best model for females included an additive cohort effect (Table 2). However, the models including either additive or interactive effects of cohort were only about 1.3 times more supported than the model without cohort effects. Therefore, cohort effects on female mass, if any, were small (estimated difference of 3.4 g between females from good and bad cohorts), and our results did not allow us to conclude firmly about the existence of such effects on female mass. For males, the best model included neither additive nor interactive effects of cohort. The model without any cohort effect received at least twice more support than models including additive or

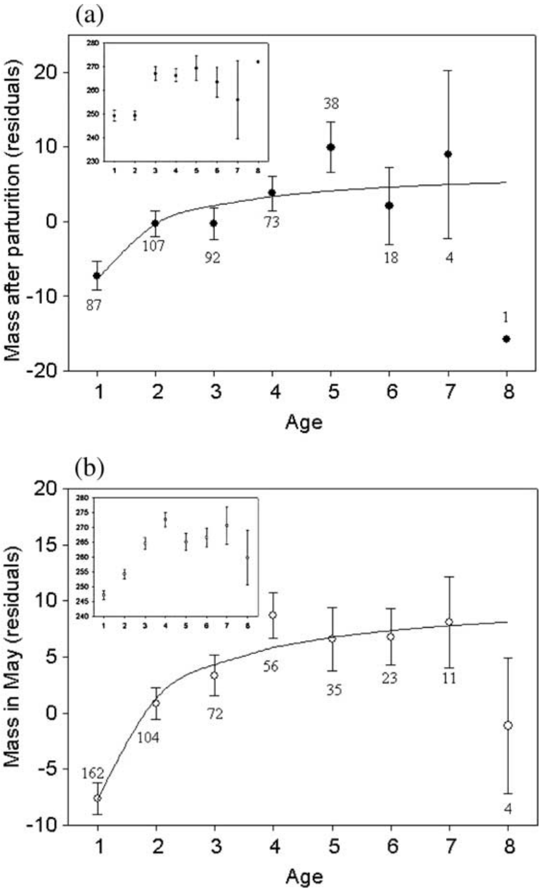

Figure 1. Average mass (±SE) in relation to age in a North American red squirrel population, Kluane, Yukon, Canada: (a) mass of females after parturition (corrected for litter size and year) and (b) mass of males in May (corrected for year). Numbers correspond to sample sizes for each age category. Fitted curves represent an inverse effect of age (1/age). Insets represent the measured values (average ± SE) of female and male mass.

Table 2. Effect of food conditions in the year of birth on mass, survival (of both sexes), and reproductive parameters (of females) in a population of North American red squirrels, Kluane, Yukon. Cohort represents a two-level variable (i.e. high-food and low-food cohorts, based on the food availability during early life). Dev indicates the deviance of the model considered; ΔAICc indicates the difference in AICc between the model with lowest AICc and the model considered; w_i indicates the AICc weight of a given model (calculated considering only a set of three models: the model without any cohort effect, the model with an additive cohort effect, and the model with a cohort effect and an interaction between age and cohort).

Trait	Model	np	Dev	ΔAICc	w_i
Mass of females	1/age+litter size+year	17	2494.515	0.575	0.276
	1/age+litter size+year+cohort	18	2491.758	0.000	0.368
	1/age+litter size+year+cohort+cohort × age	19	2489.627	0.062	0.356
Mass of males	1/age+year	17	2398.222	0.000	0.523
	1/age+year+cohort	18	2397.442	1.378	0.260
	1/age+year+cohort+cohort × age	19	2395.607	1.710	0.217
Survival of females	age+age^2+year	18	1269.648	0.000	0.664
	age+age^2+year+cohort	19	1269.266	1.699	0.284
	age+age^2+year+cohort+cohort × age	21	1268.506	5.114	0.052
Survival of males	(prime age 1–6, senescent ≥7)+year	4	880.767	0.000	0.649
	(prime age 1–6, senescent ≥7)+year+cohort	5	880.766	2.020	0.236
	(prime age 1–6, senescent ≥7) +year+cohort+cohort × age	6	880.196	3.475	0.114
Litter size	(linear$_{1-6}$; linear$_{6-8}$)+mass+year	18	−226.769	3.227	0.157
	(linear$_{1-6}$; linear$_{6-8}$)+mass+year+cohort	19	−226.818	5.370	0.054
	(linear$_{1-6}$; linear$_{6-8}$)+mass+year+cohort+cohort × age	21	−236.606	0.000	0.789
Number of juveniles weaned	(yearling, prime age 2–6, senescent ≥7)+litter size+year	18	1501.222	0.000	0.681
	(yearling, prime age 2–6, senescent ≥7)+litter size+year+cohort	19	1501.039	1.954	0.256
	(yearling, prime age 2–6, senescent ≥7)+litter size+year+cohort+ cohort × age	21	1499.561	4.775	0.063
Average mass of juveniles at weaning	(linear$_{1-3}$; constant$_{3-8}$)+litter size+year	16	1537.311	0.000	0.694
	(linear$_{1-3}$; constant$_{3-8}$)+litter size+year+cohort	17	1537.261	2.242	0.226
	(linear$_{1-3}$; constant$_{3-8}$)+litter size+year+cohort+cohort × age	18	1537.019	4.312	0.080
Number of juveniles recruited	(yearling, prime age 2–5, senescent ≥6)+number weaned+ bequeathal+year	20	−306.062	0.000	0.472
	(yearling, prime age 2–5, senescent ≥6)+number weaned+ bequeathal+year+cohort	21	−308.227	0.165	0.435
	(yearling, prime age 2–5, senescent ≥6)+number weaned+ bequeathal+year+cohort+cohort × age	23	−309.846	3.260	0.093

interactive cohort effects (Table 2), indicating that food conditions in the year of birth did not influence male mass.

Effects of age on survival

Female survival

Female survival showed clear age-variation and decreased from three years of age onwards (Fig. 2). The model controlling for year effects and including a quadratic effect of age (Table 1b; Supplementary material Appendix 2c) had the highest statistical support (AICc weight of 0.305), whereas all models that did not include a decrease in survival with age had no support at all (AICc weights less than 0.001). This strongly supports the existence of survival senescence in female red squirrels. Female survival decreased by about 29% between 4 and 6 years of age, and then by about 70% between 6 and 8 years (Fig. 2). Food conditions in the year of birth did not influence female survival (i.e. models including additive or interactive cohort effects were at least 2.3 times less supported than the model without cohort effect; Table 2).

Male survival

Survival of males decreased at oldest ages (Fig. 2) and did not show significant yearly variations. The model with two age classes (1–7 and ≥7 years old) provided the best fit to the data (Table 1b; Supplementary material Appendix 2d). This model was more than 14 times more supported than any model that did not include a decrease in survival, indicating strong evidence of senescence in male survival. Survival of prime-age males was 47% higher than that of old males (Fig. 2). Food conditions in the year of birth did not influence male survival as models including either additive or interactive cohort effects were at least 2.75 times less supported than the model without any cohort effect (Table 2).

Effects of age on female reproductive traits

Litter size

Litter size at birth varied from one to six, with an average of 3.00 ± 0.04, and was age-dependent. After adjusting for yearly variations in environmental conditions and for the mass of mothers, the best model of litter size variations included a linear increase with mother's age until six years, followed by a linear decrease (Fig. 3a; Table 1b; see Supplementary material Appendix 2e for complete results of model selection; parameter estimates ± SE for the variable 'age' before and after six years old, respectively: 0.071 ± 0.031 and -0.68 ± 0.29). This model received more than four times more support than any model that did not include senescence. However, the observed decrease in litter size at birth after 6 years of age was driven by the very small litter size of one single 8-years old female (Fig. 3a).

When testing for cohort effects, the best model included interactive effects of food conditions in the year of birth and age (Table 2), indicating that females from high-food

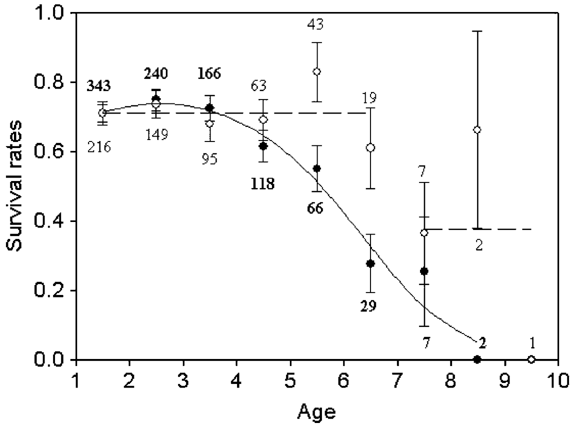

Figure 2. Male (open circles) and female (filled circles) survival (± SE) in relation to age (results from the full-age dependent models) in a North American red squirrel population, Kluane, Yukon, Canada. Numbers correspond to sample sizes for each age and sex category (bold numbers correspond to female sample sizes). Fitted curves represent estimated survival from a model with a quadratic effect of age for females and a model with two-age classes for males (Table 1).

cohorts produced larger litters at the beginning of their life (at one year of age) than females from low-food cohorts, but not later on (Fig. 3b). For high-food cohorts, litter size decreased after six years of age, whereas for low-food cohorts the only female that lived longer than six produced a litter size of five at seven years of age (Fig. 3b). The model including interactive effects of food conditions in the year of birth and age received at least five times more support than models including additive cohort effects or no cohort effect at all (Table 2). It could be that the selection of such a model was mainly driven by what happened the first years of life, where sample sizes were important. However, a model that included an interaction between age and food conditions in the year of birth between 1 and 6 years of age but not afterwards had no support (AICc weight close to 0). Therefore, the interaction between age and food conditions in the year of birth was not only driven by what happened during the first years of life.

Weaning success

Female red squirrels weaned on average 1.11 ± 0.05 juveniles per breeding attempt but this varied with females' age. The best age-dependent model describing the number of juveniles reared successfully to weaning included three age classes (yearlings; prime-aged females 2–6 years old; older females; see Supplementary material Appendix 2f for complete results of model selection). This model was about at least six times more supported than any model that did not include senescence (Supplementary material Appendix 2f). The number of juveniles weaned increased after one year of age, remained constant between 2 and 6 years of age, and then markedly decreased in old females (Fig. 4). When considering the number of weaned juveniles adjusted for litter size at birth (Table 1b), results remained the same and the selected model included the same three age classes (Fig. 4; see Supplementary material Appendix 2g for complete results of model selection). We found no effect of food conditions in the year of birth on the number of juveniles weaned, whether it was adjusted for litter at birth or not, as the models including additive or interactive cohort effects were more than 2.5 times less supported than the model without any cohort effect (Table 2).

Juvenile mass at weaning

The average mass of juveniles at weaning (mean mass per litter) was 155.5g ± 1.4. After adjusting for yearly variations and for litter size, the selected model indicated that the average mass of juveniles at weaning increased with age until three years of age and then remained constant with age (Table 1b; Fig. 5; estimate ± SE for the linear effect before three years of age: 5.09 ± 1.88; see Supplementary material Appendix 2h for complete results of model selection). In addition, we did not find any effect of food conditions in the year of birth on the mass of juveniles at weaning as the models including additive or interactive cohort effects were more than three times less supported than the model without any cohort effect (Table 2).

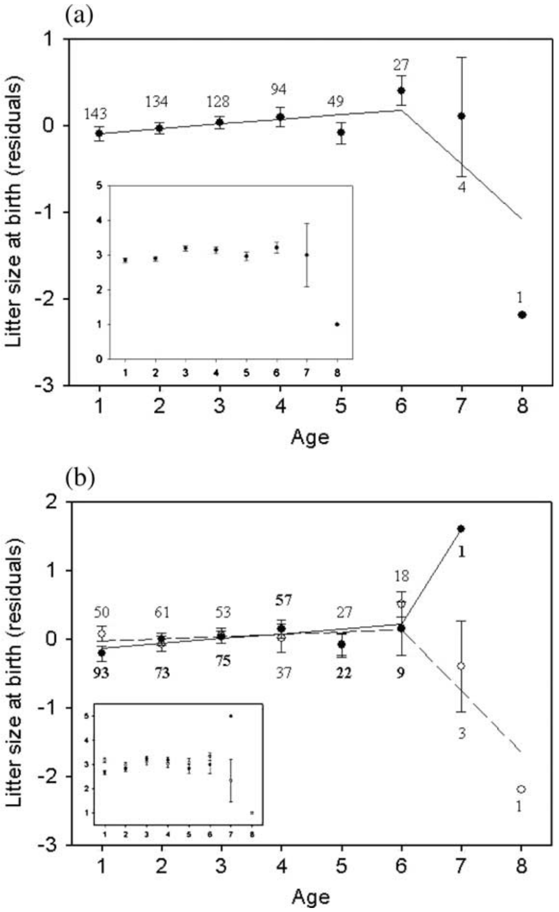

Figure 3. Average litter size at birth (±SE) corrected for mother's mass and year in relation to mother's age (a), and mother's age and food conditions in the year of birth (b) in a North American red squirrel population, Kluane, Yukon, Canada. In (b), high-food (white symbols) and low-food (black symbols) cohorts are represented. Numbers correspond to sample sizes for each age and cohort category (bold numbers correspond to low-food cohorts). Fitted curves represent linear effects of age from piecewise regressions (Table 1). Insets represent the measured values (average ± SE) of litter size.

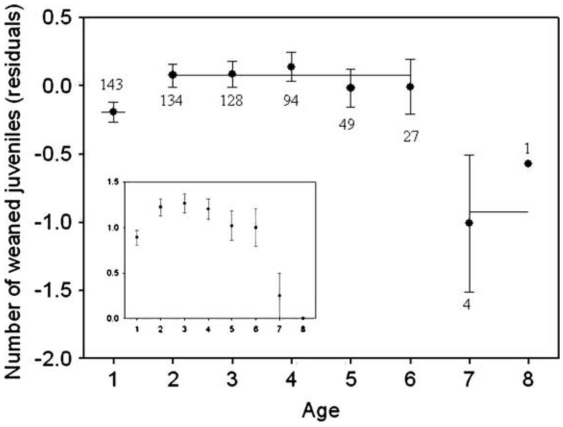

Figure 4. Number of weaned juveniles (±SE) corrected for litter size at birth and year in relation to mother's age in a North American red squirrel population, Kluane, Yukon, Canada. Numbers correspond to sample sizes for each age category. The fitted curve represents the estimated number of weaned juveniles from a model with three age classes (Table 1). The inset represents the measured values (average ± SE) of number of weaned juveniles.

Effects of age on juvenile survival from weaning to one year of age

Survival of weaned juveniles declined for old mothers (Fig. 6). The best model describing the number of juveniles recruited included three age-classes (yearlings, 2–5 years old females, ≥6 years old; see Table 1b and Supplementary material Appendix 2i for complete results of model selection), with the highest number of recruited juveniles for prime-age females (Fig. 6). This model was at least 14 times more supported than any model that did not include senescence (Supplementary material Appendix 2), indicating that a decrease in the number of recruited juveniles at oldest ages was very likely. When testing for an effect of the food conditions in the year of birth, we found that the model including additive effects of cohort and the model without any cohort effect received very similar support (less than 1.1 more support for the model without cohort effect, Table 2). We could thus not reject the hypothesis of a cohort difference in the number of recruited juveniles (females from good cohorts recruited about 0.6 more juveniles on average per reproductive attempt).

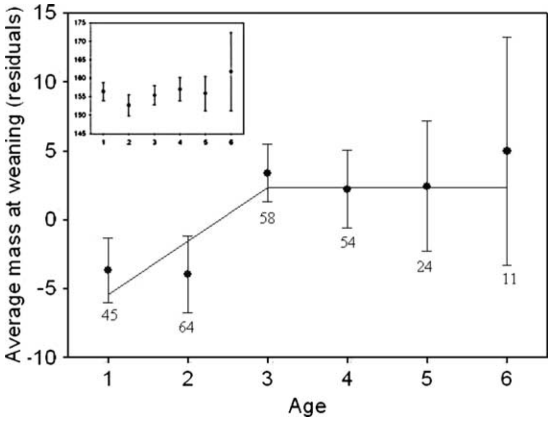

Figure 5. Average mass of juveniles at weaning (±SE) corrected for litter size and year in relation to mother's age in a North American red squirrel population, Kluane, Yukon, Canada. The fitted curve represents linear effects of age from piecewise regressions (Table 1). The inset represents the measured values (average ± SE) of juvenile mass at weaning.

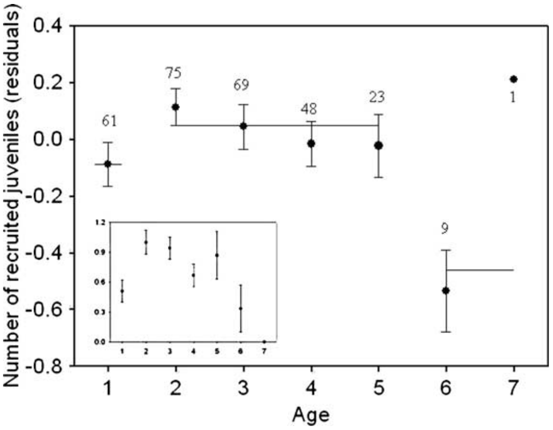

Figure 6. Number of recruited juveniles (±SE) corrected for number of weaned juveniles, bequeathal and year in relation to mother's age in a North American red squirrel population, Kluane, Yukon, Canada. Numbers correspond to sample sizes for each age category. The fitted curve represents the estimated number of recruited juveniles from a model with three age classes (Table 1). The inset represents the measured values (average ± SE) of number of recruited juveniles.

Discussion

Evidence of senescence in red squirrels

In support to our first prediction, we found clear evidence of senescence in both survival and reproductive traits. In our population of North American red squirrels, both females and males showed strong evidence of survival senescence (Fig. 2). Most female reproductive traits also decreased with increasing age but the reproductive senescence was weaker than the survival senescence. Contrary to our second prediction of similar rates of senescence in both sexes the observed decrease in survival was weaker for females than for males. Moreover, old female red squirrels were less efficient at raising young (as indicated by the lower number of juveniles weaned adjusted for litter size at birth; Fig. 4) and the survival of their weaned juveniles (i.e. survival of juveniles after their emancipation) was also lower (Fig. 6). This suggests that, in support to our third prediction, old females produced juveniles of lower quality than prime-aged ones. Finally, we found very little evidence that high food cohorts experienced higher senescence rates that low food cohorts, despite a higher reproductive activity early in life (Fig. 3b).

As increasingly reported from long-term studies of birds and mammals (Bennett and Owens 2002, Gaillard et al. 2003a, Jones et al. 2008), senescence occurred in most fitness components of red squirrels. On the other hand, we did not report a decrease in mass with age for males, and the decrease in female mass, if it occurred, was very limited (Fig. 1). North American red squirrels can be considered as income breeders (sensu Jönsson 1997) because they do not store fat to meet the energetic demands of reproduction.

Therefore, observed variation in body mass of red squirrels might reflect variation in body size rather than in condition. We do expect lesser age variation, and thereby weaker senescence, in size than in condition for mature mammals with determinate growth. Other measures such as immunocompetence (Owens and Wilson 1999, Cichon et al. 2003) would be needed to test for a decrease in physiological performance at the end of life.

Life history variation and magnitude of senescence

It is often argued that senescence will affect few individuals in free-living populations usually characterized by high mortality rates (Hayflick 2000). However, our results clearly show that a substantial proportion of red squirrels are still at risk when senescence occurs. Female survival decreased by 29% at four years of age and by 70% at six years of age, when an average of 35% and 10%, respectively, of one year old females were still alive. From a population dynamics viewpoint, this means that the magnitude of senescence (measured as the product between the effect size, that is the proportion of decrease in a given trait, and the proportion of individuals from a given cohort still present when this decrease occurs) is about 0.10 and 0.07 at 4 and 6 years of age, respectively. The effect size of senescence for red squirrels was thus similar to the one reported in Richardson's ground squirrels, where survival of females decreased by about 40% after four years of age, but its impact on population dynamics was about three times less for ground squirrels (magnitude of about 0.03) because only 7% of one year old females survived until that age (Broussard et al. 2005).

Therefore, the effect size of survival senescence for squirrels was large and appeared to be higher than that reported for large mammalian herbivores. Indeed, the female survival of roe deer only decreased by 7.5% when individuals reached eight years, and by 24% when they reached 12 years (Gaillard et al. 2003b). The higher effect size of senescence in squirrels compared to large mammalian herbivores supports current theories of life history evolution of senescence predicting higher senescence in fast than in slow-living species (Hamilton 1966, Jones et al. 2008). However, as the proportion of one year-olds still at risk when 8 and 12 years of age was higher in roe deer (60% and 37%, respectively) than in squirrels, the impact of senescence on population dynamics was similar in roe deer (magnitude of 0.05 and 0.09 at 8 and 12 years of age, respectively) and in squirrels.

Between-sex differences in the magnitude of senescence

A lower adult survival and thereby a lower lifespan in males often occurs in populations of large herbivores (Gaillard et al. 2000, Clutton-Brock and Isvaran 2007). In these species usually highly polygynous and sexually dimorphic in size, large males have high energy expenditures during the mating period, do not eat during the rut period, and may therefore be subject to greater mortality, especially when environmental conditions deteriorate (Toïgo and Gaillard 2003). On the contrary, male squirrels are about the same size as females (males are 5 to 10% heavier than females, Boutin and Larsen 1993) and have the same reproductive lifetime, so that we predicted similar patterns of senescence in both sexes. We found that males had higher survival than females from 4 years old onwards and that female survival decreased before male survival (from 3 versus 7 years of age onwards, respectively), which suggested an earlier senescence for females than for males. However, further data on age-specific variation in male reproductive success and on age-specific body condition will be needed to assess whether females senesce more than males in red squirrels.

Food conditions in the year of birth and senescence patterns

Senescence is expected to involve a tradeoff between early and late demographic performance (disposable soma theory, Kirkwood and Rose 1991), and thus to depend on the age-specific allocation of energy to reproduction. As expected, female red squirrels from good cohorts reproduced more intensely (i.e. larger litter size) in early life than those born in poor cohorts. Therefore females born in good cohorts could have suffered from higher cumulative reproductive costs than females born in bad cohorts, as reported by Nussey et al. (2006) on red deer in which females that produced more offspring during early life paid the cost of a higher survival senescence. However, we found only weak support for differential intensity of survival or reproductive senescence according to cohort quality, although the litter size decreased after six years of age for females born in high-food cohorts but not for those born in low-food cohorts. As this latter result was based on a small sample size (only one and four females born in low-food and high-food cohorts, respectively, survived after six years of age), further work is needed to assess whether early life conditions influence reproductive senescence in squirrels. Likewise, we did not find any evidence of different senescence patterns among cohorts of contrasted food conditions at birth in the other life history traits we studied. One explanation could be that individuals from good cohorts were of higher average quality than those born in poor cohorts due to long-lasting cohort effects (Lindström 1999, Gaillard et al. 2003c). In such a situation, individuals born from good cohorts could breed more intensely early in life than individuals from poor cohorts without exhibiting stronger or earlier senescence (i.e. they can deal with higher energetic costs without paying any extra fitness costs). In red deer, it has been found that females born in high density years experienced higher rates of senescence despite a later first reproduction, possibly because of a lower phenotypic quality (Nussey et al. 2007). In our red squirrel population, there is some evidence that females from good cohorts were of higher quality than those from poor cohorts (Descamps et al. 2008), which supports the idea that variation in individual quality may have masked any tradeoff between early and late performance. This does not mean that higher breeding activity during early life did not lead to higher rates of senescence, but rather that heterogeneity between cohorts overrode differences in reproductive costs (see Weladji et al. 2008 for similar findings on large herbivores).

Intergenerational effects of senescence: age of mother and quality of juveniles

Weaned juveniles born to old females (≥ six years old) had a lower survival to one year of age than weaned juveniles born to prime-age females, which suggests that old females produced juveniles of lower quality. Such decreases in offspring quality as mothers age have been reported in insects (Kern et al. 2001), rodents studied in laboratory (Wang and vom Saal 2000), cattle (Fuerst-Waltl et al. 2004), deer (DelGuidice et al. 2007), humans (Parsons 1964), and birds (Saino et al. 2002). Nevertheless, evidence of intergenerational effects of mother's age in free-living populations of vertebrates is still scarce despite its evolutionary and demographic importance (Kern et al. 2001). Indeed, when simply considering the number of juveniles weaned by a female to assess reproductive senescence, the overall decrease in reproductive values at oldest ages may be underestimated. For females in our red squirrel population, senescence was expressed in three different ways: a decrease in survival, a decrease in reproductive success and a decrease in the quality of offspring produced.

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Supplementary material (available online as Appendix 016545 at <www.oikos.ekol.lu.se/appendix>) Appendix 1 and 2.