Effects of food abundance on genetic and maternal variation in the growth rate of juvenile red squirrels

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Abstract

Sources of variation in growth in body mass were assessed in natural and experimental conditions of high and low food abundance using reciprocal cross-fostering techniques and long-term data (1987–2002) for a population of North American red squirrels (*Tamiasciurus hudsonicus*). Growth rates were significantly higher in naturally good and food supplemented conditions, than in poor conditions. Mother–offspring resemblance was higher in poor conditions as a result of large increases in both the direct genetic variance and direct-maternal genetic covariance and a smaller increase in the coefficient of maternal variation. Furthermore, the genetic correlation across environments was significantly less than one indicating that sources of heritable variation differed between the two environments. These results are consistent with the hypothesis that selection has eroded heritable variation for growth more in good conditions and indicate the potential for independent adaptation of growth rates in good and poor conditions.

Introduction

The heritable transmission of trait variation from parents to offspring is a necessary condition for evolution by natural selection. As a result, one of the central goals of quantitative genetics has been to determine the degree to which trait variation is determined by genetic and environmental sources. The narrow-sense heritability (h^2), defined as the proportion of total phenotypic variation (σ_Z^2) that is due to additive genetic variation (σ_{AO}^2), provides a useful estimate of the potential for a population to respond to selection. Heritability estimates, however, are characteristic only of the particular circumstances under which they are measured and as such can vary both among populations and over time within a given population as a result of selection history or in response to changes in environmental conditions (Falconer & Mackay, 1996). The inherent temporal and spatial variability in food resources, competitors and predators in natural systems may make heritability estimates from natural populations particularly susceptible to change.

As heritabilities are ratios, differences in heritability estimates between environments can be due to changes in additive genetic variation, environmental variation or both. Several hypotheses have been proposed to explain how sources of trait variation should respond to different environmental conditions (see Hoffmann & Merilä, 1999 for a review). For example, selection is thought to eliminate low fitness alleles more efficiently in common favourable environments, suggesting that less frequent unfavourable conditions should maintain higher levels of genetic variation (Holloway et al., 1990). Falconer (1952) recognized that traits expressed in different environments could be considered separate traits linked by a genetic correlation across environments. In the presence of a genetic correlation across environments, selection in one environment will result in both a correlated response of the trait mean and a corresponding elimination of low fitness alleles in the alternate environment. As a result, the opportunity for both independent adaptation and differences in genetic variance between two environments is influenced by the strength of the genetic correlation across environments. Differences in genetic variation between environments because of differential selection history, therefore, are expected to be associated

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with weak genetic correlations across environments (Hoffmann & Merilä, 1999).

Most of the hypotheses reviewed by Hoffmann & Merilä (1999) concern the effects of environmental conditions on direct genetic and residual variation. However, maternal effects are now widely recognized as important components of total phenotypic variation (Mousseau & Fox, 1998), particularly in mammals and other organism with long periods of maternal dependence (Roff, 1997). Genetically based maternal effects can also influence the evolutionary response to selection (e.g. Wolf et al., 1998), so responses of maternal effect variation to environmental conditions can exacerbate or compensate for differences in direct genetic variation between favourable and unfavourable environments. Some of the same hypotheses that have been proposed regarding the expression of genetic variation in other traits (Hoffmann & Merilä, 1999) can also be applied to maternal effects. However, as maternal effects are sheltered from selection on males, they are expected to maintain higher levels of additive genetic variation in the presence of selection than direct genetic effects (Wade, 1998). Differences in selection history between favourable and unfavourable environments should, therefore, lead to greater differences in direct genetic variation than maternal genetic variation.

Generalizations regarding the effects of environmental quality on heritable variation in natural populations have been slow to emerge. For example, there has been a general trend towards decreased heritability of size-related traits in natural populations of birds experiencing unfavourable conditions (see Merilä, 1997; Hoffmann & Merilä, 1999). This was presumed to be due to decreased additive genetic variation and increased residual variation in stressful conditions although common environmental effects or genetic correlations across environments may have influenced these results (Hoffmann & Merilä, 1999). However, there are many other studies that have demonstrated an increase in genetic variation or no consistent response across traits in unfavourable conditions (e.g. Gebhardt-Henrich & van Noordwijk, 1991; Merilä et al., 1999; Coltman et al., 2001; Kause & Morin, 2001; see also Hoffmann & Merilä, 1999).

North American red squirrels (*Tamiasciurus hudsonicus*) provide a unique mammalian system within which the effects of environmental variability on sources of variation can be examined. In the northern parts of their geographic distribution, red squirrels feed almost exclusively on the seeds of white spruce cones (*Picea glauca*). The production of cones by white spruce trees is spatially synchronous but temporally variable. The abundance of spruce cones provides a relatively simple and repeatable measure of environmental quality that has been found to have important implications for the life history of red squirrels (Berteaux & Boutin, 2000; Humphries & Boutin, 2000; Réale et al., 2003; McAdam & Boutin, 2003).

In this study we used 16 years of data (1987–2002) from a single population of red squirrels to examine the effects of environmental conditions on mother–offspring resemblance in juvenile growth in body mass using mother–offspring regressions. Conditions for growth were defined based on the availability of spruce cones and were confirmed by differences in trait means. We estimated mother–offspring covariances in each of four combinations of good and poor maternal and offspring environments for growth, which allowed us to compare environmental effects on the resemblance of mothers and offspring raised in the same conditions and to estimate the genetic correlation of growth in body mass between good and poor environments.

Mother–offspring covariance estimates, however, are confounded by maternal effects (Lynch & Walsh, 1998), which are known to account for a large proportion of total phenotypic variation in growth in body mass (McAdam et al., 2002). As such, differences in mother–offspring resemblance between good and poor environments may reflect changes in direct additive genetic variance, maternal genetic variance or both. To further investigate changes in these components of variation in response to food availability, we performed cross-fostering experiments in both 1999 and 2000, which represented years of very high and low food abundance, respectively. Finally, we performed a food supplementation experiment in 2000, in which some females were supplied with *ad libitum* access to sunflower seeds to experimentally improve the conditions for growth. Sources of variation including direct genetic, maternal, genotype by environment and residual variation were examined in both the natural environmental conditions as well as the experimental environment to determine the effects of food availability on sources of variation in nestling growth rates.

Materials and methods

A natural population of red squirrels (*T. hudsonicus*) has been monitored in the southwest Yukon (61°N, 138°W) since 1987. This population of squirrels has been extensively studied and details on the population and general live-trapping and census techniques are given elsewhere (Berteaux & Boutin, 2000; Humphries & Boutin, 2000). In this study we were interested in the growth of body mass from soon after birth to approximately 1 month of age, which is just prior to first emergence from the natal nest. Mass measurements at both times periods were made by temporarily removing offspring from their natal nest and weighing them to the nearest gram using a Pesola® spring balance (PESOLA AG, Baar, Switzerland). Over the 16 years of this study there were some cases in which mass measurements were missed or were not taken at the appropriate time. As a result, we restricted the data to include only offspring with initial mass measurements <50 g, final mass measurements <100 g,

and more than 5 days between first and second mass measurements (see also McAdam & Boutin, 2003). Offspring subjected to food or litter size manipulations were excluded from the analysis.

Offspring were classified as having been raised in good or poor conditions for growth based on the availability of spruce cones. The number of spruce cones in the top 3 m (viewed from one side) of each of approximately 250 trees spaced systematically throughout the study areas has been recorded each year since 1986 (see Humphries & Boutin, 2000). The average number of cones counted per tree within a year provides an index (cone index) of the availability of food for that year. Years in which the cone index was >50 were classified as being good years for growth (1992, 1993, 1995, 1996 and 1998), while those with a cone index <50 were classified as poor years (1987, 1988, 1989, 1990, 1991, 1994, 1997, 1999, 2000, 2001 and 2002).

Mature spruce cones are harvested by red squirrels in late summer and early fall and are stored in centrally located caches of cones (middens) for future consumption. Red squirrels depend primarily on stored cones for survival over winter and for reproduction the following spring, but also consume new cones, which might help to meet current reproductive demands late in the breeding season. We briefly investigated the influence of current year's cone production on the growth rates of juvenile red squirrels. The growth in body mass of 1912 juveniles born between 1987 and 2002 were correlated with both the abundance of spruce cones produced in the previous fall and the abundance of cones produced in the current year using multiple regression techniques. In order to determine how the partial correlation coefficients for previous and current year's cone production changed across the breeding season, we repeated this overall analysis with progressively smaller subsets of the data corresponding to offspring born later in the breeding season. Specifically, each repetition of the analysis excluded an additional 10 days of the breeding season starting with the earliest born offspring; those born prior to 22 March.

Mother–offspring covariances were estimated separately for each of the four possible combinations of environmental conditions for mothers (good or poor) and offspring (good or poor). The growth rates of all offspring born to each dam were averaged within each environmental category so that each female and her offspring contributed only one data point to each covariance estimate. The genetic correlation across environments was calculated following Roff (1997; equation 3.12). There were no differences between mother and offspring variances in any of the four environment combinations ($F < 1.6$, ns).

Cross-fostering experiments

Cross-fostering experiments were performed in 1999 and 2000 to partition direct and maternal components of

variation in growth in body mass in both good and poor conditions for growth. In 1998, spruce trees in this area produced more cones than in any of the past 13 years (mean cone index = 286.7 ± 17.2, $n = 248$ trees; see also McAdam and Boutin, 2003). As a result, conditions for growth of juveniles born in the spring of 1999 were very good. Conversely, very few cones were produced in the fall of 1999 (cone index = 27.6 ± 3.9, $n = 248$ trees), so conditions for growth in the spring of 2000 were poor.

All females in each of the study areas (approximately 40 ha each) were monitored for reproductive activity through regular live-trapping starting early in the spring of 1999 and 2000. Changes in female body mass and external morphology allowed us to precisely estimate the timing of parturition. Litters of squirrels were paired as closely as possible based on the timing of parturition and offspring were reciprocally cross-fostered between paired litters such that half the offspring from one litter were exchanged with an equal number of offspring in the paired litter. As a result, natal litter sizes were not changed by the cross-fostering technique. Females appear not to discriminate between genetic and foster offspring and there do not appear to be any differences between offspring transferred to a new nest and offspring raised by their own dam (Humphries & Boutin, 1996; McAdam *et al.*, 2002). All measurements of offspring from the cross-fostering experiments were made on the same day within pairs.

Seven of the 29 pairs of litters cross-fostered in 2000 were supplemented with food in two of the four study areas. The other two areas could not be supplemented with food because of the ongoing long-term study of these populations, but there do not appear to be any differences between these pairs of study areas that would have biased the results of this study. Females within food-supplemented pairs received *ad libitum* access to sunflower seeds (*Helianthus* sp.) from the day on which offspring were cross-fostered until the time when the second mass measurements were collected. Sunflower seeds were provided in a single plastic feeder located on the central midden in each supplemented female's territory. Feeders were checked and replenished with new seed regularly to ensure that supplemented females had *ad libitum* access to seeds.

Growth measurement

In 1999 and 2000, we examined the growth in body mass of red squirrels from soon after birth (3.07 ± 0.07 days) to approximately 25 days of age. Body mass was measured (± 0.1 g) using portable electronic balances. Growth in body mass over this time period is linear. Previous analyses have corrected these linear growth measurements for correlations with the initial mass measurement to eliminate the potential influence of persistent prenatal maternal effects

(McAdam *et al.*, 2002). However, this correction might also result in overly conservative estimates of direct genetic variation if pre- and post-natal growth rates are genetically correlated, so we have not corrected linear growth rates for initial mass measurements in this study.

Statistical analysis

Variation among nestling growth rates was partitioned using separate two-way nested ANOVAS, which were each represented by the linear model

$$Y_{hijk} = \mu + P_h + D_{i(h)} + N_{j(h)} + DN_{ij(h)} + e_{hijk},$$

where Y_{hijk} is the growth rate of the kth squirrel raised by nurse j and born to dam i in the hth pair. μ Is the grand mean growth rate and P_h is the effect of the hth pair. $D_{i(h)}$ and $N_{j(h)}$ are the effects of the ith dam and jth nurse within the hth pair, respectively. $DN_{ij(h)}$ is the interaction effect and e_{hijk} is the residual deviation of the kth squirrel. All effects except μ were assumed to be random independent variables with zero means and variances equal to σ_P^2, σ_D^2, σ_N^2, σ_{DN}^2 and σ_e^2. Variances were estimated from these linear models using restricted maximum likelihood (REML) in the varcomp procedure in S-PLUS (Insightful, 2001a).

Genetic expectations of variance components from the linear model above following Willham (1963) are:

$$\sigma_D^2 = 1/2\sigma_{AO}^2 + 1/4\sigma_{DO}^2$$
$$\sigma_N^2 = \sigma_{AM}^2 + \sigma_{DM}^2 + \sigma_C^2$$
$$\sigma_{DN}^2 = 0$$
$$\sigma_e^2 = 1/2\sigma_{AO}^2 + 3/4\sigma_{DO}^2 + \sigma_E^2.$$

We assumed that offspring within genetic litters were full-sibs (see also McAdam *et al.*, 2002). Twice the dam variance, therefore, estimated the direct genetic variance (σ_{AO}^2), but also included dominance variance ($1/2\sigma_{DO}^2$). We have previously estimated dominance variance to be low for this trait (McAdam *et al.*, 2002) and do not further consider its potential confounding effects here. Maternal variance represented the sum of maternal genetic (σ_{AM}^2), dominance (σ_{DM}^2) and environmental (σ_C^2) variances.

Rutledge *et al.* (1972) initially estimated the direct-maternal genetic covariance [$cov(A_O, A_M)$] from the dam by nurse interaction variance (σ_{DN}^2; see also Lynch & Walsh, 1998; Rauter & Moore, 2002). This expectation assumes that the interaction variance represents the covariance among full sibs raised by their own mother minus both σ_D^2 and σ_N^2 (Lynch & Walsh, 1998). However, in a cross-fostering design offspring raised by an unrelated female experience maternal effects independent of direct genetic effects. As a result, $cov(A_O, A_M)$ is not included in the phenotypic variance of fostered offspring and σ_{DN}^2 has no genetic expectation (see genetic expectations above; Riska *et al.*, 1985). Alternatively, we estimated $cov(A_O, A_M)$

from two separate ANOVA's as the difference between the among-litter variance of offspring that remained in their natal nest and the among-litter variance of cross-fostered offspring (Ahlschwede & Robison, 1971; Riska *et al.*, 1985). In addition, we calculated total phenotypic variance (σ_Z^2) as the total variance within cross-fostered pairs ($\sigma_D^2 + \sigma_N^2 + \sigma_{DN}^2 + \sigma_e^2$) plus $cov(A_O, A_M)$ following Riska *et al.* (1985).

Standard errors for the variance components were estimated by jackknifing at the level of the cross-fostered pair. Variance components were standardized as coefficients of variation (CV) for comparisons across environments to avoid bias because of changes in mean growth rates (CV = SD/mean).

Components of maternal performance for growth

We attempted to decompose maternal effects on offspring growth into the contributions of various maternal characteristics following McAdam *et al.* (2002). We analysed maternal performance data for both 1999 and 2000 in a single general linear model, that included eight potential maternal characteristics (litter size at birth, parturition date, body size, territory size, age, maternal mass at parturition and offspring emergence, and the azimuth of the natal nest) and their interactions with *Year* to test for any differences between good and poor years for growth. Parturition date and litter size were previously found to be important maternal effects on growth (McAdam *et al.*, 2002), so we started with a simple model that included only these two traits and attempted to add additional characteristics and interaction variables. Terms were added sequentially to the model based on changes in the likelihood version of Mallows' *Cp* statistic (Insightful, 2001a).

All statistical analyses were performed using STATIS-TICA (StatSoft, Inc., 2000) and SPLUS 6.1 (Insightful, 2001a,b). Values are presented as mean ± 1 SE.

Results

There was a strong positive correlation between the rate of nestling growth in body mass and the log cone index in the previous fall (slope = 0.261 ± 0.015). The partial correlation between growth and previous year's cone production, however, declined as the season progressed, while the partial correlation between growth rates and the current year's cone production increased. In particular, for offspring born after 30 May, current year's cone production was a better predictor of juvenile growth rates than previous year's cone production. As a result, we considered the environment for growth to be determined by previous year's cone production for those offspring born on or before 30 May (Julian date = 150) and by current year's cone production for those offspring born after 30 May.

Mother–offspring covariances

The average growth rate of juveniles raised in good years (1.91 ± 0.015 g day^{-1}) was significantly higher than the average growth rate of juveniles raised in poor years (1.66 ± 0.017 g day^{-1}; $t_{1910} = 10.87$, $P < 0.0001$; Fig. 1). There was a significant positive covariance between the growth rate of mothers and their offspring in good years [$cov(M_{good}, O_{good}) = 0.061 ± 0.022$, $t_{85} = 2.77$, $P < 0.01$]. The covariance between offspring and maternal growth rates in poor environments [$cov(M_{poor}, O_{poor}) = 0.113 ± 0.074$] was more than 80% greater than in good environments but was not significant as a result of the reduced sample size ($t_{28} = 1.53$, ns). Mother–offspring covariances measured across environments were much lower and were not significantly different from zero [$cov(M_{good}, O_{poor}) = 0.038 ± 0.033$, $t_{37} = 1.15$, ns; $cov(M_{poor}, O_{good}) = -0.014 ± 0.033$, $t_{41} = 0.42$, ns]. As a result, the genetic correlation across environments was calculated as 0.143 ± 0.281 and was not significantly different from zero ($t_{154} = 0.51$, ns), but was significantly lower than one ($t_{154} = 3.05$, $P < 0.01$).

Cross-fostering

The rate of growth in body mass of nestling red squirrels was significantly slower in 2000 (1.66 ± 0.05 g day^{-1}) than 1999 (1.91 ± 0.03 g day^{-1}, $t_{246} = 4.71$, $P < 0.0001$, Fig. 1). Growth rates in 1999 and 2000 were very typical of growth rates recorded in this population over the past 16 years in good (1.91 ± 0.02 g day^{-1}) and poor (1.66 ± 0.02 g day^{-1}) years for growth, respectively (Fig. 1). In addition, survival of offspring from birth to the timing of the second set of measurements was much higher in 1999 than 2000. In 1999, 176 of the 200 (88%) cross-fostered offspring survived to roughly 1 month of age, while in 2000 only 72 of 122 (59%) cross-fostered offspring that did not receive the food supplement were alive at the time of the second set of measurements.

Squirrels that were supplemented with food in 2000 had significantly higher rates of growth in body mass (1.98 ± 0.05 g day^{-1}) than controls within the 2000 season ($t_{102} = 4.06$, $P < 0.0001$) and were not significantly different from growth rates in 1999 ($t_{206} = 1.14$, ns, Fig. 1). Food supplementation also improved juvenile survival in 2000. Thirty-two of the 41 (78%) offspring whose mothers were supplemented with food in 2000 survived to 1 month of age.

Sources of variation in growth

The cross-fostering design revealed significant dam and nurse effects on growth in body mass in 1999 (Table 1). In 2000, there was a significant nurse effect but the dam effect was no longer significant. The dam by nurse interaction was not significant in either 1999 or 2000. The coefficient of total phenotypic variation for growth in body mass was 26% higher in 2000 than in 1999 (Fig. 2). This was because of a large increase in the coefficient of direct additive genetic variation (26%) and a smaller increase in maternal variation and interaction variation, but residual variation actually decreased slightly. In addition the direct-maternal genetic covariance was
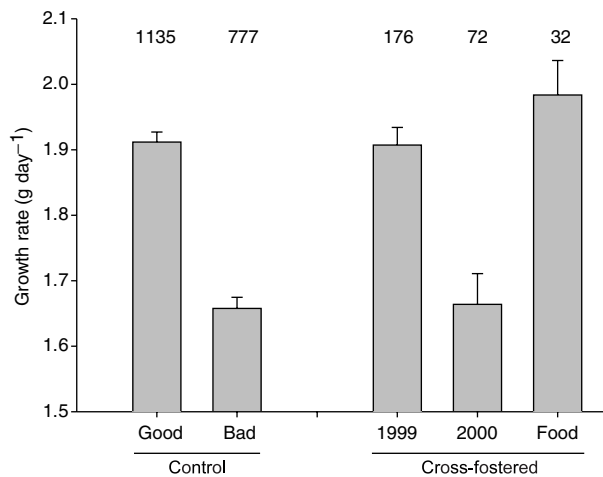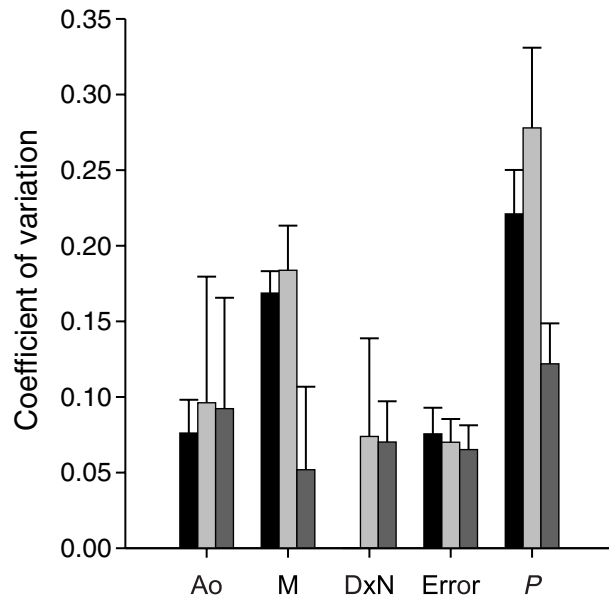

Fig. 1 Rates of growth in body mass (g day^{-1}) of nestling red squirrels (*Tamiasciurus hudsonicus*) in good and poor conditions for growth. Control growth rates were measured on nestling squirrels born in this population over the past 16 years (1987–2002). Growth rates of cross-fostered offspring in 1999 and 2000 correspond to good and poor conditions for growth, respectively. Some cross-fostered offspring received a food supplementation (*Food*) in 2000. Values are presented as means + one standard error. Sample sizes (number of juveniles) are indicated above each bar.

Table 1 Results of model II ANOVA (type III SS) used to partition variation in nestling growth rates of body mass into direct genetic, maternal, interaction and residual components. Separate models are presented for 1999, 2000 and food supplemented red squirrels (*Tamiasciurus hudsonicus*).

Source	1999			2000			Food		
	d.f.	MS	*F*	d.f.	MS	*F*	d.f.	MS	*F*
Pair	32	0.314		19	0.361		5	0.297	
Dam (pair)	33	0.051	2.83**	19	0.054	2.05	6	0.076	1.90
Nurse (pair)	26	0.318	17.67***	8	0.290	10.74**	5	0.061	1.53
Nurse × dam (pair)	26	0.018	0.82	7	0.027	1.93	5	0.040	2.35
Residual	58	0.022		18	0.014		10	0.017	

**$P < 0.01$, ***$P < 0.001$.

Fig. 2 Coefficients of variation for components of growth in body mass of nestlings born in good (black bars), poor (light grey), and experimentally enriched (dark grey) environments. Components of total phenotypic variation (P) include coefficients of direct genetic (Ao), maternal (M), dam by nurse interaction ($D \times N$), and residual (*Error*) variation. Standard errors were calculated by jackknifing.

84% larger in 2000 (0.079 ± 0.060) than 1999 (0.043 ± 0.035), but these were not significantly different.

There were no significant dam or nurse effects on growth rates in food supplemented dyads (Table 1). This was in part because of the small number of cross-fostered dyads that received supplemental food, but the addition of food also greatly reduced the large maternal effects (Fig. 2) and the large, positive direct-maternal genetic covariance (-0.005 ± 0.057) compared with control dyads. Other components of total phenotypic variation in growth in body mass were very similar between food supplemented and control dyads in 2000 (Fig. 2).

Components of maternal performance for growth

The initial model of maternal performance including only litter size and parturition date explained 43% of the total variation in maternal performance for offspring growth ($F_{2,30} = 11.35$, $P < 0.001$). None of the other single maternal characteristics (body size, territory size, age, maternal mass at parturition and offspring emergence, and the azimuth of the natal nest) improved the fit of the model. However, the interaction of litter size and year as well as the interaction of parturition date and year entered into the final model ($n = 33$ pairs, $R^2 = 0.53$, $F_{4,28} = 7.9$, $P < 0.001$). Litter size and parturition date both represented negative maternal effects on offspring growth (litter size: coefficient $\pm$ SE $= -0.29 \pm 0.078$;

parturition date: -0.19 ± 0.047) suggesting that offspring from larger litters born later in the season had lower growth rates. Both the litter size $\times$ year and parturition date $\times$ year terms were positively correlated with maternal performance (litter size $\times$ year: 0.13 ± 0.081; parturition date $\times$ year: 0.09 ± 0.046) indicating that the strength of the negative effect of litter size and parturition date on juvenile growth rates was weaker in 2000 than 1999.

Discussion

Natural populations of red squirrels experience large annual fluctuations in the abundance of spruce cones, which have important implications for many aspects of their life history (Berteaux & Boutin, 2000; Humphries & Boutin, 2000; Réale *et al.*, 2003; McAdam and Boutin, 2003). Our classification in this study of good and poor environments for growth based on the abundance of spruce cones was reflected in large differences between mean phenotypes; offspring born into good conditions had much higher growth rates than offspring born into poor conditions. Cone availability and nestling growth rates in 1999 and 2000, when the cross-fostering experiments took place, were typical of good and poor conditions, respectively. As a result, we are comfortable that the conditions during which the cross-fostering experiments were performed reflected conditions experienced by this population during the last 16 years.

Results of both the cross-fostering experiments and the analysis of long-term data for this same population indicated that sources of resemblance between mothers and offspring were greater during poor conditions for growth. The coefficient of direct additive genetic variance increased by 26%, but there were also increases in coefficient of maternal (nurse) variance, and dam by nurse interaction variance and a decrease in the coefficient of residual variance. Together with an increase in the direct-maternal genetic covariance, these results indicate that there is a greater potential for evolutionary change in response to selection in red squirrels in poor years than in good years for offspring growth.

The documented increase in mother–offspring resemblance in poor conditions in red squirrels is opposite to most previous studies of natural populations of birds, which have suggested that heritabilities tend to decrease in poor conditions (see Hoffmann & Merilä, 1999). Hoffmann & Parsons (1997a) hypothesized that this trend in the bird literature might be due to unique characteristics of nutritional stress. The results of this study, however, indicate that nutritional stress can also result in an increase in additive genetic variance similar to other stress agents (Hoffmann & Parsons, 1997b). Indirect maternal genetic effects can have a large influence on the response of traits to a given amount of selection (Wolf *et al.*, 1998; McAdam *et al.*, 2002) and so maternal effects can contribute to the change in

evolutionary potential between favourable and unfavourable environments. Here, we documented an increase in maternal effects in poor conditions, but this increase was much smaller than the increase in direct genetic effects. Coltman *et al.* (2001) also reported increases in both direct and maternal genetic variation in female Soay sheep (*Ovis aries*) in spring when the energetic demands of reproduction are thought to be stressful (but see Réale *et al.*, 1999). Poor conditions have also been shown to increase maternal variation in great tits (*Parus major*; Gebhardt-Henrich & van Noordwijk, 1991) and prolong the expression of maternal effects in the annual plant *Erigeron annuus* (Stratton, 1989). Clearly additional studies of the effects of environmental quality on direct genetic and maternal variation need to be performed in natural populations before generalizations emerge.

The observed increase in heritable variation in poor conditions in this study is consistent with the hypothesis that stronger and more frequent selection in good years has eroded direct genetic variation for nestling growth rates more than in poor years. In the red squirrel system, years of low cone production are more common than years of high cone production (10 of the last 16 years had low cone production; see Methods for definitions), but many more offspring are produced in years of high cone production, such that 59% of offspring born since 1987 were born in good conditions. In addition, adult over-winter survival is higher following years of high cone production (S. Boutin, unpublished data) so that juvenile squirrels born in years of high food abundance experience intense competition for few vacant territories. More specifically, viability selection on juvenile growth rates from recruitment to breeding age over the past 13 years was positively correlated with the abundance of spruce cones in the previous year, but there was no effect of cones on selection acting during the period of maternal dependence (McAdam & Boutin, 2003). It is likely, however, that most effects of selection history on contemporary variation were largely because of selection history prior to the start of this long-term study.

The observed weak genetic correlation between good and poor environments suggests that selection in one environment will have a limited effect on genetic variation in the opposite environment. The presence of this weak genetic correlation between environments provides the opportunity for differences in selective history to be reflected in differences in genetic variation and suggests the potential for independent adaptation of nestling growth rates to both good and poor environments in this system. Environmentally dependent expression of maternal effects is a possible explanation for the independence of heritable variation across environments and provides a possible mechanism for the maintenance of the observed large positive covariance between direct and maternal genetic effects (McAdam *et al.*, 2002). We found evidence of interactions between year and both litter size and parturition date suggesting that the strength

of these maternal effects differs in good and poor conditions. In both cases the negative maternal effect was stronger in good conditions than poor conditions. These results for litter size, including the increased effect of litter size in good years, are consistent with experimental effects of litter size augmentation on juvenile growth rates (Humphries & Boutin, 2000). There remained, however, a large proportion of unexplained variation in maternal performance that was not correlated with any of the other maternal characters examined here.

The experimental addition of food in 2000 maintained high rates of growth similar to those observed in favourable years. However, the effects of the experimental food addition on sources of variation in nestling growth rates differed from the comparative control results in two important ways. The addition of food greatly reduced both the large maternal effects and the large positive $cov(A_O, A_M)$ for nestling growth rates observed in 1999 and 2000. The experimental addition of food reduced maternal effects by over 70%, suggesting that most of the variation in maternal performance for offspring growth is related to the ability of females to secure food resources for their offspring even in years of very high food abundance. The ability of females to acquire resources for their offspring may frequently represent a strong maternal effect on offspring traits in natural populations where food resources vary in both space and time. Estimates of the strength of maternal effects and $cov(A_O, A_M)$ from animals raised either in the laboratory or captive populations where *ad libitum* food is provided may, therefore, underestimate the importance of maternal effects to variation in offspring traits in natural populations.

Finally, in these analyses we have assumed that offspring born to the same dam were full-sibs. However, female red squirrels mate with multiple males and so there is likely some degree of multiple paternity. An interesting alternative explanation for the increase in variance among dams in unfavourable conditions is that the abundance of food might have influenced the degree of multiple paternity. For example, an increase in the degree of multiple paternity in good conditions would have appeared as a decrease in variance among dams in the present cross-fostering design as σ_D^2 would more appropriately estimate $1/4\sigma_{AO}^2$ rather than $1/2\sigma_{AO}^2$. The decrease in error variance in 2000 and the high CV_{AO} for supplemented squirrels in 2000 are consistent with this hypothesis although standard errors for CV_{AO} are very large. Future studies examining patterns of multiple paternity in this system should address this hypothesis further.

Acknowledgments

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