

## LETTER

# Indirect effects on fitness between individuals that have never met via an extended phenotype

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### Abstract

Interactions between organisms are ubiquitous and have important consequences for phenotypes and fitness. Individuals can even influence those they never meet, if they have extended phenotypes that alter the environments others experience. North American red squirrels (*Tamiasciurus hudsonicus*) guard food hoards, an extended phenotype that typically outlives the individual and is usually subsequently acquired by non-relatives. Hoarding by previous owners can, therefore, influence subsequent owners. We found that red squirrels breed earlier and had higher lifetime fitness if the previous hoard owner was a male. This was driven by hoarding behaviour, as males and mid-aged squirrels had the largest hoards, and these effects persisted across owners, such that if the previous owner was male or died in mid-age, subsequent occupants had larger hoards. Individuals can, therefore, influence each other's resource-dependent traits and fitness without ever meeting, such that the past can influence contemporary population dynamics through extended phenotypes.

### Keywords

Extended phenotype, indirect effect, inheritance, KRSP, quantitative genetics, resource hoarding, *Tamiasciurus*, Territory.

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## INTRODUCTION

Organisms socially interact when they mate, fight and compete for resources. Social interactions mean individuals will influence the traits and fitness of other individuals, both positively through cooperation, but also negatively through competition for limited resources, while other interactions may have neutral consequences (Scott 1977; Moore *et al.* 1997). It is often implicitly assumed that organisms need to meet to influence each other, but this need not always be the case. If organisms possess extended phenotypes (traits of the individual that exist outside their physical body; Dawkins 1978, 1982) then they can alter the environment that others experience. 'Ecosystem engineers' (Jones *et al.* 1994) such as earthworms (Thompson *et al.* 1993) and beavers (*Castor spp.*; Naiman *et al.* 1986; Rosell *et al.* 2005) are well known to alter the environment for their own benefit (see also the separate but related concept of: 'niche construction', Odling-Smee *et al.* 2003; Scott-Phillips *et al.* 2014), and so will influence any organisms that use the modified environment. By altering the environment that another individual might experience, organisms can influence the plastic traits of others, or perhaps even their fitness, despite never actually meeting (Laird 2012).

Beyond these classic examples of 'ecosystem engineers' it is not yet clear how broadly applicable such effects may be. Yet, many organisms alter their environment by building dens, nests or burrows, which can influence organisms that subsequently choose to use or live in these structures (Bailey 2012). Furthermore, organisms may leave imprints on the environment such as scent cues that can influence those that encounter them, without direct physical contact (Cisterne *et al.* 2014; Dewan *et al.* 2018). Aspects of organisms' extended phenotypes are potentially extensive and varied (Dawkins 1978, 1982), facilitating myriad potential consequences for others.

It is, therefore, necessary to test how extended phenotypes can cause individuals to influence the phenotypes and fitness of those they never meet and assess the consequences for ecological and evolutionary processes. To do this, we studied a population of North American red squirrels (*Tamiasciurus hudsonicus*; hereafter 'red squirrels') at a study site in the Yukon Territory of Canada for 30 years. Red squirrels defend territories of *c.* 0.34 ha (LaMontagne *et al.* 2013), centred on a 'midden', a pile of white spruce (*Picea glauca*) cone scales, within which red squirrels cache food they require to survive over winter (Smith 1968b). Stored food typically consists of white spruce cones (the red squirrels' main food source in this population) harvested that autumn ('new cones') as well as

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cones stored from previous years' crops ('old cones'). The presence of these food stores is also associated with increased local mammal and bird abundance (Posthumus *et al.* 2015). Middens vary in the number of cones of each type they contain, with larger hoards leading to improved overwinter survival of the occupant (Larivée *et al.* 2010; LaMontagne *et al.* 2013), earlier breeding in the spring for females and increased reproductive success for males (Haines 2017). Therefore, they are analogous to fat stores, which can then be mobilised to fuel survival and reproduction. The difference is that red squirrels store their resources in the ground, not on their bodies, and hence these stores represent an *extended* phenotype.

Along with being an important contributor to the current occupant's survival and fitness, middens last well beyond the lifespan of a single individual. Any given midden may be defended over time by a sequence of many individuals that are typically not related and do not overlap in tenure (Hatt 1929; Smith 1968b, 1981), although in some cases a female squirrel will 'bequeath' her territory to one of her offspring, and leave to find another territory (Price & Boutin 1993; Berteaux & Boutin 2000; Boutin *et al.* 2000; Lane *et al.* 2015). Spruce cones cached in a midden can be consumed at least for 4 years post-caching (Donald & Boutin 2011; S. Boutin, unpublished data), so cones cached by previous owners can be used by the next squirrel that acquires the territory. Therefore, the number of cones cached in a squirrel's midden is an extended phenotype, and furthermore may well be influenced by both the current and the previous occupant of the territory. This then creates a mechanism through which the previous owner can influence the resource-dependent traits, and so possibly fitness, of the current occupant.

We identified how the previous occupant of the territory influences the traits (cone hoard sizes, as well as the date a female gives birth in spring; 'parturition date'), and the fitness (estimated as lifetime reproductive success; LRS) of the current owner. Since cones are placed in the ground in autumn when we measured hoard size, we expected no influence of the previous owner on the size of new cone hoards, but we did expect the previous owner to influence the size of old cone hoards. Females give birth earlier when they have access to increased food resources (Réale *et al.* 2003; Kerr *et al.* 2007; as this increases the chance their pups will acquire a territory and so recruit into the population; Williams *et al.* 2014; Fisher *et al.* 2017), so we expected that traits of the previous occupant that were associated with larger cone hoards would be the traits that cause the subsequent hoard owner to breed earlier. We made the same prediction for LRS; traits of the previous owner that are associated with larger hoard sizes should confer higher fitness on the current occupant of the territory.

## METHODS

### Data collection

We followed individually marked squirrels as part of the Kluane Red Squirrel Project (KRSP) in the Yukon Territory of Canada from 1987 through 2017. We used different subsets of this entire data set for our analyses here. In each year, we monitored

marked females in two unmanipulated 40-ha. study areas ('Kloo' and 'Sulphur') for signs of pregnancy and to tag their pups. We also tagged any immigrants and, therefore, tracked all squirrels resident in the population for their entire lifetime (see: McAdam *et al.* 2007 for further details on the study system). We enumerated the entire population in spring and autumn censuses to determine ownership of territories. These territories are exclusive and are based around large piles of discarded cone scales (middens). Middens are semi-permanent, so the same midden can be owned sequentially by multiple different squirrels across an extended period of time. For example, one midden remained active throughout the entire study period (between 1987 and 2017) and was owned by 13 different squirrels across this 31-year period. We assigned ownership of a midden based on territorial vocalizations called 'rattles' (Smith 1968a, 1978), and only included squirrels owning a midden in our analysis, i.e. those that had recruited into the population. We focus here on territory ownership in autumn, as this is when spruce cones are available to be stored, and so when we measured the size of cone hoards in middens. We recognised previous territory owners when the identity of the territory owner in one autumn was different to that in the previous autumn. We only included the first instance of a squirrel holding a given territory in the analysis, so there were no multiple records if a squirrel held the same territory for several years, but we did include a squirrel multiple times if it was observed holding different territories in different years (although this is not that common as red squirrels do not 'trade-up' their territory when vacancies are available; Larsen & Boutin 1995). Each squirrel-midden combination, therefore, only had a single previous owner, and since middens left undefended through the death of the owner are typically taken over by new individuals quickly (Price *et al.* 1986; Siracusa *et al.* 2017a,b), the previous owner typically held the territory a single year ago. If the midden was undefended for > 1 year, the previous owner would be more than 1 year in the past, which we accounted for by fitting an interaction between the traits of the previous owner and the number of years between them and the current owner (see Data analysis).

From 2012 through 2017, we estimated the number of cones stored in the primary middens of all squirrels defending territories. We did this in late September after most caching was completed. Some squirrels owned more than one midden in autumn, but we did not consider secondary middens as they are not held by the majority of squirrels and are not used extensively for resource storage when they are held. For each midden, we identified its perimeter as the location where the cone bracts gave way to typical forest floor, measured the longest axis and the perpendicular width at the midpoint of the long axis. Assuming an elliptical shape (following: Larivée *et al.* 2010), we estimated the area as follows:

$$\frac{\text{length}}{2} \times \frac{\text{width}}{2} \times \pi$$

Within each quarter of the midden, we placed two 30 × 30 cm square quadrats at *c.* ¼ and ¾ of the distance from the centre of the midden towards the perimeter, such that each quarter of the midden had two quadrats placed wholly within it, giving eight in total (method adapted from Larivée *et al.*

2010). If the midden was very small, such that a quarter could not fit two quadrats, then only a single quadrat was placed. We then excavated all quadrats to a depth of 10 cm from the surface of the bract pile and counted the number of old and new cones found within. If an excavated tunnel was found, new and old cones inside were counted to a depth of 30 cm, but not out of the area of the quadrat. If cones are not buried in middens they will open, releasing their seeds. This makes them useless as food to red squirrels, so we did not count opened cones even if they were within our quadrats. We calculated the number of old and new cones per cm<sup>2</sup> of midden surface sampled (accounting for the number of quadrats), multiplied by the area of the midden and then rounded to the nearest whole number, to give an estimate of the size of the old and new cone hoard in the midden (old and new cones indexes). We were able to separately count old and new cones, as new cones are purple and green, and were often sticky with sap, while old cones were dark brown and never sticky.

For the 6-year period (2012–2017), we were able to assess the effect of previous owners on old and new cone hoard sizes. Across a much-extended period of data collection, we were also able to estimate effects of previous owners on female parturition dates (springs of 1992–2017) and the LRS of males and females (squirrels born from 1991 to 2009; although LRS of males was not available until 2003, see below). Due to maternities identified at birth since 1987, and paternities identified based on genetic analysis of ear biopsies since 2003, we have very good information on the number of offspring each individual has and their survival, and also a well-resolved pedigree (Gunn *et al.* 2005; Lane *et al.* 2007; McFarlane *et al.* 2011). We estimated LRS as the total number of pups an individual dammed or sired that survived to 200 days. As we do not typically monitor individuals over winter, this requires individuals to have survived their first winter and recruited into the population as adults in the spring when we conduct a population census. We excluded individuals for which we had missed one or more breeding events, individuals that died of unnatural causes such as dying in a trap, individuals that were born after 2009 (as their LRS would be underestimated), as well as males pre-2003 for which we were not able to assess siring success. These restrictions gave us the following sample sizes: cone hoards, 323 measures, 148 unique females and 125 unique males; parturition date, 525 measures across 402 unique females; and LRS, 393 measures across 274 unique females and 119 unique males. As mentioned above, female red squirrels may bequeath their territory to an offspring, which tends to be a daughter and the fastest growing pup in the litter (Berteaux & Boutin 2000; Robertson *et al.*, *in prep*). We repeated all analyses with all instances of the previous occupant being the mother excluded ( $n = 30, 69$  and  $65$  for both types of cone, parturition dates and LRS, respectively). Those results are qualitatively similar to the ones we present here, and so we present them in the supplementary materials (Tables S1-4).

### Data analysis

We used mixed-effect models to determine the effects of the previous occupant on current cone hoard estimates, parturition

dates and LRS. This involved four linear models, either with the old cone hoard size, new cone hoard size, parturition date or LRS as the response variable, all fitted in R using the package ‘MCMCglmm’ (Hadfield 2010).

For the models of both types of cone hoard and parturition dates, we included the random effect of squirrel identity, since some squirrels had multiple measurements of hoard sizes and parturition dates. Given we also included a term to estimate the additive genetic variance (which was negligible for cone hoard size, see supplementary materials), this allowed us to also estimate permanent environment effects (where individuals are consistently different from one-another, but for reasons not due to additive genetic differences or other variables included in the model). In every model, we also fitted the identity of the previous owner as an additive genetic effect, which allowed us to estimate variance in indirect genetic effects from previous midden owners. These were always very close to zero and are discussed in the supplementary materials. We also used the pedigree to calculate the genetic relatedness between the territory owner and the previous occupant ( $r$ ). We included in all models the random effect of year, to account for year-to-year variations in the traits at the population level.

We added a different set of fixed effects to each model to control for contemporary factors (traits of the current occupant and environmental influences) that might influence the traits. To the models of old and new cone hoard sizes, we included the current occupant’s sex, age, study area, and the linear effect of year. Within an individual’s lifetime, cone hoards have been shown to increase and then decrease (Haines 2017). We therefore estimated a different relationship between hoard sizes and age before and after the approximate age of peak cone hoard sizes by including a term for whether the squirrel was older than 3 years or not (approximately the age of peak cone hoards), and the interaction between this term and age. Fitting two relationships like this is preferable to using age<sup>2</sup> to detect initial increases followed by decreases (Simonsohn 2018; e.g. due to senescence) although the results for models in which age was fitted as a quadratic term instead led to similar conclusions (not shown). For parturition date, we included the occupant’s age in years as a categorical variable (with 7 and 8 year olds, the oldest, grouped together), to account for nonlinear trends (typically late parturition dates as a yearling breeder and potential senescent delays in old age), the individual’s study area, and a linear effect of year. For LRS, we included the individual’s study grid, a linear effect of year, and whether the individual experienced a ‘mast year’ (when spruce trees produce a super abundance of cones; Kelly 1994; LaMontagne & Boutin 2007) or not in their lifetime, as this has been shown to greatly increase LRS (Descamps *et al.* 2008; Hämäläinen *et al.* 2017).

To investigate whether the previous occupant influenced the traits of the current owner, in all models, we included the sex and lifespan of the previous owner as fixed effects, with separate relationships for lifespan of the previous owner below and above 3 years of age. We included sex of the previous owner because males have been found to cache more and have larger hoards (Donald & Boutin 2011; Archibald *et al.* 2013), which could influence the next owner of the territory.

We also included the interaction between these traits of the previous owner and the time in years between the current and previous owners, because we predicted that previous occupants farther in the past should affect the current occupant less. We also included the main effect of this lapsed time, which represented the length of time the midden was unoccupied before the current occupant. As unoccupied middens will have their stored cones gradually removed by other red squirrels, we expected larger values of this lapsed time to be associated with smaller old cone hoards, later parturition dates, and lower LRS estimates.

For cone hoards and LRS, we used a Poisson error-structure with a log-link function, an inverse Gamma prior for each random effect ( $V = 1$ ,  $v = 0.002$ , chosen to be non-informative), and 500 000 iterations, with the first 10 000 discarded, and 1/40 of the remaining iterations kept, to form the posterior distributions. For parturition date, we used the same specifications, except a normal error structure and link function. For all models, we standardised each continuous predictor by subtracting the mean and dividing by the standard deviation, which improves model convergence and interpretability of regression coefficients (Schielzeth 2010). We also scaled parturition dates in this way, but not cone hoard sizes or LRS, as this transformation gives negatives and non-integers, unsuitable for Poisson models. Importance of terms was judged by the distance of the mode of the posterior distribution from zero, and the spread of the 95% credible intervals. Successful convergence was assessed with Heidelberger and Welch's convergence diagnostic (Heidelberger & Welch 1983), while we performed three runs of each model to ensure different chains reached the same qualitative result.

## RESULTS

### **Males have larger hoards and leave more cones for the next territory owner, thereby influencing resource-dependent traits of the next occupant**

The sex of the previous midden owner had important consequences for the current midden owner, which acted through the extended phenotype of the size of the hoard. Males hoarded more new cones, (coefficient plots for all models are given in Fig. 1). This effect on hoard size carried over to the next owner of the midden. If the previous midden owner was a male, the current occupant had a larger old cone hoard (Figs 1a and 2a), an earlier parturition date (Figs 1c and 2b, although this effect marginally overlapped zero), and a higher LRS (Figs 1d and 2c). For example, if the previous owner was male rather than female, the current occupant is expected to acquire a midden with an extra 1318 old cones. If each cone contains an average of 2.53 kJ (Fletcher *et al.* 2010), this equates to an additional 3337 kJ. Furthermore, a female that acquired her midden from a male would give birth 2.26 days earlier on average, and have 0.59 more offspring survive to 200 days over her lifetime than a female that acquired her midden from another female. As expected, this effect of the sex of the previous occupant on the size of the old cone hoard weakened with increasing number of years between the

current and previous owner (Fig. 1a). Surprisingly, the effect of whether the previous owner was a male on LRS was enhanced with increasing time between the previous and current owner (although this effect marginally overlapped zero; Fig. 1d).

### **Mid-aged squirrels have larger hoards and leave more cones for the next territory owner**

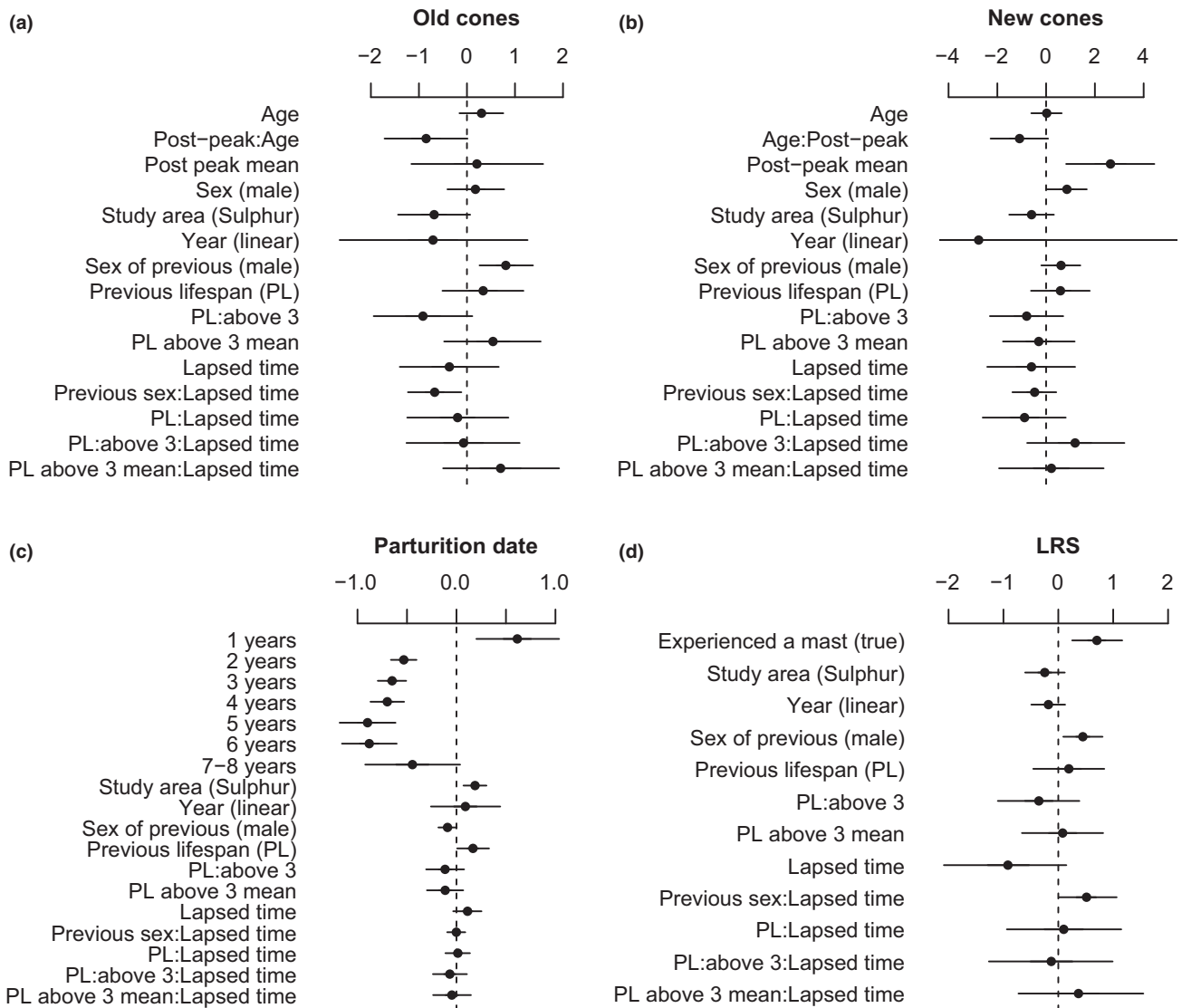
We found that the lifespan of the previous midden owner influenced the current midden owner through hoard size. Within the lifetime of current owners, old and new cone hoard sizes both showed no initial increase before 3 years of age, and then declined with age for squirrels that were older than 3 years (Fig. 3a and b). Concurrent with this late-life decline in hoard size, red squirrels that lived longer than 3 years of age left fewer old cones to the next occupant of the territory the longer they lived (Fig. 3c). We also found that parturition dates were later if the previous occupant lived longer, with no change in this relationship pre- and post-3 years of age (Fig. 3d). Note, however, that each of these effects slightly overlapped zero, suggesting they should be interpreted with caution. Parturition dates were also latest in yearlings, earliest at ages 5 and 6, and tended to be later at ages 7 and 8. LRS was unaffected by the age of the previous owner.

While not of direct interest to this study, contemporary effects on traits remained. Individuals had a higher LRS if they experienced a mast year in their lifetime. Old but not new cone hoards were smaller and parturition dates later on one study area (Sulphur) compared to the other (Kloo), but LRS was not different. No trait showed a consistent change across years, while LRS was lower the longer the midden had been unoccupied (although this effect marginally overlapped zero). The model intercept and estimates of the variance components are given in Table 1.

Repeating these analyses after removing instances in which the previous owner was the squirrel's mother, did not qualitatively change the results (see Supplementary materials, Tables S1-4). Estimates of direct genetic variance for cone hoard sizes were negligible, and so it is not surprising that we did not detect any indirect genetic effects from the previous owner for any trait (see Supplementary materials, Table S5). Including cases where the previous owner was the mother, the relatedness between the previous and current midden owner was 0.092, 0.110 and 0.085 for both types of cone, parturition dates and LRS respectively.

## DISCUSSION

Indirect effects have been suggested to play a 'special role' in evolution (Bailey *et al.* 2018), as they cause individuals to influence each other's phenotypes and fitness (Griffing 1967; Scott 1977; Moore *et al.* 1997). Here, we have documented that indirect effects can occur between organisms that often never meet and may not even have been alive at the same time, facilitated through an extended phenotype. Extended phenotypes have received much theoretical interest since Dawkins (Dawkins 1978, 2004; Jones *et al.* 1994, 1997; Sterelny



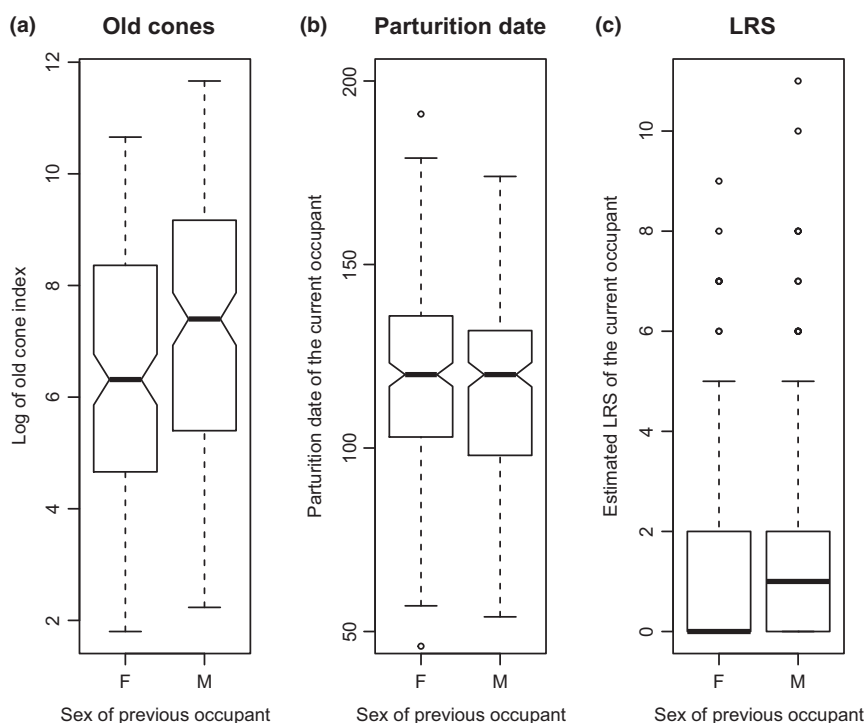
**Figure 1** Coefficient plots displaying estimated means and 95% credible intervals (CRIs) of the models for (a) old cone hoards, (b) new cone hoards, (c) parturition dates (note negative values indicate earlier dates) & d) lifetime reproductive success. As the study area 'Kloo' was the default, the effect of study area is how the trait mean differs on 'Sulphur'. 'Age' refers to the effect of age of the current occupant on the trait, while 'Post-peak:Age' refers to the additional effect of age when the individual was older than 3 years. 'Post-peak mean' indicates whether traits in individuals older than 3 had different means. 'Previous sex' indicates the additional effect of the previous owner being a male, with it being a female the default. 'Previous lifespan (PL)' refers to the previous occupant's lifespan, while 'PL:above 3' refers to the additional effect of this lifespan if it was above 3 years. 'PL above 3 mean' indicates whether traits in individuals where the previous territory occupant lived longer than 3 years had different means. 'Lapsed time' is the time period between the previous and current occupant; it is interacted with each trait of the previous occupant (e.g. 'Previous sex: lapsed time'). For new cone hoards, the x-axis has been truncated to display the effects nearer zero; the CRIs for the effect of year were  $-13.8$  to  $9.3$ .

*et al.* 1996; Jablonka 2004; Turner 2004; Hunter 2009; Bailey 2012), but extending empirical work to a range of systems has lagged behind. Individuals of many species hold territories that they may modify, for instance by digging out burrows (Weber *et al.* 2013; Metz *et al.* 2017), building nests (Perna & Theraulaz 2017) or caching resources within them (Larivière *et al.* 2010). Additionally, organisms may alter the environment by constructing some structure in it, such as a spider's web (Blamires *et al.* 2017a,b), or leave scent cues to be detected by others (Cisterne *et al.* 2014). Therefore, there is the potential for effects mediated by these extended phenotypes to be widespread throughout the natural world, yet our

study is the first to quantify influences on life-history traits and fitness of other individuals mediated through an extended phenotype.

#### Sex of previous owner influences cone hoards, parturition dates and fitness

If the previous owner of the territory was a male, current occupants had earlier parturition dates and higher LRSs, due to acquiring larger hoards of old cones. We expect this is applicable more broadly, hence any trait which is related to how much an organism alters its environment might be



**Figure 2** (a) Old cone hoards were larger when the previous occupant was a male. Old cone hoards have been ln-transformed to improve viewability. (b) Parturition dates (days since start of year, with 1st January as 1) tended to be earlier when the previous owner was a male. (c) Estimated LRS was higher when the previous owner was a male. Notches in a & b indicate 95% confidence intervals of the median (the thick black bar in all plots).

expected to predict how much it influences other organisms indirectly. Often, only one sex of a species controls a territory, builds a nest, or engages in some form of environmental manipulation. It is therefore only this sex which is expected to exert indirect effects through these environmental changes, and anything which alters the survival and so relative prevalence of this sex, or its distribution in the environment, will alter the influence of these indirect effects.

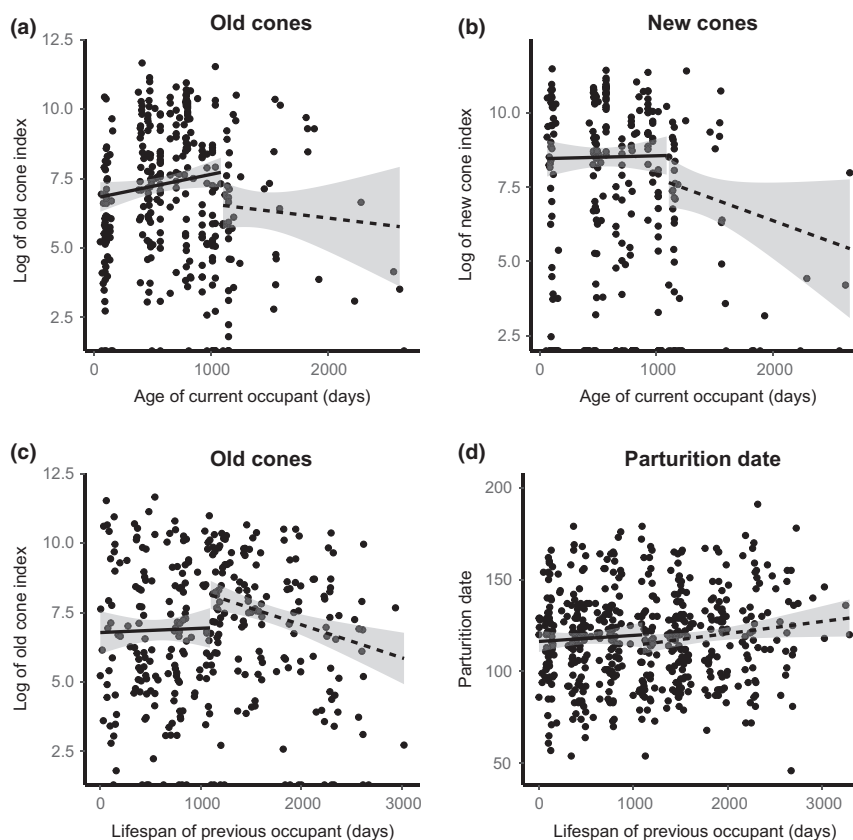
Finding more cones in middens previously owned by males appears to suggest that males hoard more cones than females, then fail to use them, as they are still present for the next occupant. Male energy expenditure during the mating season is approximately as high as female energy expenditure during lactation (Lane *et al.* 2010), so it is unlikely that males simply do not need to use these resources to fuel reproduction. Instead, males may hoard more as hoard size is related to the number of offspring they sire in the spring, which is not true for females (Haines 2017). Over-winter mortality would prevent males from consuming these additional cones during the mating season (males are more likely to die over winter than females; LaMontagne *et al.* 2013; Fisher *et al.* 2017), leaving the larger hoard for the next owner. We also found that males did not have larger old cone stores, which might seem to contradict the general findings that they have larger cone hoards. However, this can be explained by the fact that we used only the first instance of holding a territory for each squirrel, so males likely have not yet had time to enlarge their hoard of old cones.

If male red squirrels leave behind more resources, it makes it beneficial for a juvenile squirrel to settle in a territory that

had previously been held by a male to take advantage of the extra resources. This is generally true for any case of environmental manipulation; it would be advantageous for other organisms to detect and exploit any changes made by others. In the case of squirrels, we do not believe there is a choice where a juvenile squirrel settles because the availability of territories is often very limited, with competition being intense. (Price & Boutin 1993; Larsen & Boutin 1994), especially in years of high density (Williams *et al.* 2014; Fisher *et al.* 2017). Vacant territories tend to be filled quickly (Boutin & Schweiger 1988), but we do not know which traits of juveniles predict success in competing for them (Robertson *et al. in prep*). The previous occupant of a territory might, therefore, be stochastic, although this may not be true in other systems. Ultimately, this effect of the previous owner will still have consequences for the fitness of the current occupant, and so a portion of the variance in reproductive success of a population can be attributed to the caching behaviour of now-dead individuals. The effects of the previous owner on fitness, therefore, have the potential to have interesting effects on the distribution of fitness in a population (discussed below).

#### Largest hoards passed on by previous owners of intermediate age

We found that cone hoard sizes decreased beyond 3 years of age. This suggests that the ability to find or cache cones declines in old age and is perhaps an example of a senescent decline. Phenotypic senescence is increasingly commonly found in natural populations (Nussey *et al.* 2013), and has been detected in this study system before (McAdam *et al.* 2007;



**Figure 3** (a) Old cone hoards declined with age in squirrels older than 3 years. (b) New cone hoards declined with age in squirrels older than 3 years. (c) Cone hoards were largest if the territory was acquired from an individual of intermediate lifespan and declined if the previous owner lived beyond 3 years. (d) Parturition dates (days since start of year, with 1st January as 1) were later the long the previous individual lived. For all plots, cone hoards have been logged to improve viewability.

Descamps *et al.* 2008), including for cone hoard size (Haines 2017). This senescence in the stored resources an individual has access to is analogous to senescence in fat reserves in organisms that store energy in their bodies ('capital breeders' Jönsson 1997). This demonstrates senescence is a general phenomenon that even extends beyond the commonly considered case of 'performance' traits such as body mass or clutch size, to include an extended phenotype, and so should be considered in other situations where individuals alter their environment.

Additionally, the late-life decline in off-body resources has, unlike fat stores, consequences for genetically unrelated individuals that did not physically interact with the focal individual: subsequent owners. The size of the old cone hoard of the current owner tended to be lower if they acquired a territory from a long-lived squirrel. This demonstrates that it would be best to acquire a territory from an individual that died in the prime of their life, but this is also the age when red squirrels are least likely to die (Descamps *et al.* 2007). For the same reasons as described above for sex, we think red squirrel juveniles have limited ability to choose the territory they first settle on, and so would not be able to seek-out middens of prime-aged squirrels. Still, any factor that shortens individuals' lives, and so causes individuals to die nearer the age of peak hoard size, should increase the number of cones passed on to subsequent owners, the implications of which remain to be explored.

**Table 1** Model intercept and variance component estimates for each model

Trait	Model intercept	Permanent environmental	Year	Residual
Old cones	6.91	0.007 ( $< 0.001$ –1.068)	2.078 (0.442–16.85)	3.230 (1.967–4.912)
New cones	1.82	0.007 ( $< 0.001$ –2.365)	43.13 (9.974–713.7)	6.153 (3.026–9.155)
Parturition date	0.609	$< 0.001$ ( $< 0.001$ –0.032)	0.714 (0.386–1.408)	0.199 (0.158–0.252)
LRS	–0.494	NA	0.263 (0.074–0.824)	0.004 ( $< 0.001$ –1.265)

For variance components, we give the posterior distribution mode, with the 95% credible intervals in parentheses. 'NA' indicates the term was not estimated in the model. Note the 'Year' term in the LRS model is birth year, while in the other models it is the year the trait was expressed.

#### Phenotypic effects not underpinned by genotypic effects

We estimated very low direct additive genetic variance in both old and new cone hoard sizes (see Supplementary materials). Since individuals did not possess genetic variance for hoard size, it was not surprising that we did not find any indirect genetic variance in their effects on the parturition date or LRS of the next owner of the territory. There was, however, a modest amount of variance for the indirect genetic effect on old cone

hoard size, although the credible intervals were very close to zero. These indirect genetic effects, might, therefore, provide a mechanism by which hoard size could evolve despite the absence of direct genetic effects of hoard size. The broad confidence intervals in our indirect genetic variance estimate, however, suggest caution in this interpretation. Further investigation of indirect genetic effects acting through extended phenotypes is needed to determine whether they provide a general route through which traits lacking direct genetic variance could evolve.

### Whose phenotype is it anyway?

The indirect effects from previous owners that we have identified have the potential to influence evolutionary processes. While we estimated that indirect genetic effects acting through the previous midden owner were very small (see Supplementary materials) for resource-dependent traits and fitness, we still estimated phenotypic effects of previous owners on these traits. This means that evolutionary change will conform less well to models of evolution that only consider contemporary effects, as past individuals can influence current traits and fitness. Since stored resources in this system depend on the masting of spruce trees, past environments can influence contemporary resource-dependent traits, requiring us to possibly model these ecological ‘memory’ effects to understand trait distributions (Filotas *et al.* 2014). Maternal effect models of evolution might be useful here, as such models incorporate a lag in evolutionary change (Kirkpatrick & Lande 1989), something which may be occurring here. Quantifying how past environments, past individuals and historical selection can influence contemporary traits, and the consequences of this for ecological and evolutionary processes is an exciting next step for this line of research to take. In general, a given individual’s phenotype, and even its fitness, might only partly be under direct control, and some aspects of its phenotype may be influenced by individuals with whom it has not directly interacted through extended phenotypes.

### CONCLUSIONS

Due to an extended phenotype, the food hoard size, the date of spring breeding and the lifetime reproductive success of a red squirrel currently occupying a territory are influenced by the previous owner of the territory. This means that key traits, and even the fitness of an individual, are not under its direct influence but are influenced by previous individuals, and so possibly previous environments. A greater appreciation of what can be considered extended phenotypes, including the phenotypes of individuals when they interact with others, and their quantitative genetic underpinnings, will give us a greater appreciation of how these traits influence the ecology and evolution of populations.

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### AUTHORSHIP

DNF and AGM conceived the research question. SB initiated the long-term study and all authors contributed to field logistics, data collection and the writing of the manuscript. DNF drafted the manuscript and conducted the data analysis, with guidance from AGM. All authors approved of the final manuscript for submission.

### DATA ACCESSIBILITY STATEMENT

Data available from the Figshare Repository: <https://doi.org/10.6084/m9.figshare.7588628.v1>.

### REFERENCES

- Archibald, D.W., Fletcher, Q.E., Boutin, S., McAdam, A.G., Speakman, J.R. & Humphries, M.M. (2013). Sex-specific hoarding behavior in North American red squirrels (*Tamiasciurus hudsonicus*). *J. Mammal.*, 94, 761–770.
- Bailey, N.W. (2012). Evolutionary models of extended phenotypes. *Trends Ecol. Evol.*, 27, 561–569.
- Bailey, N.W., Marie-Orleach, L., Moore, A.J. & Simmons, L. (2018). Indirect genetic effects in behavioral ecology: does behavior play a special role in evolution? *Behav. Ecol.*, 29, 1–11.
- Berteaux, D. & Boutin, S. (2000). Breeding dispersal in female North American red squirrels. *Ecology*, 81, 1311–1326.
- Blamires, S.J., Hasemore, M., Martens, P.J. & Kasumovic, M.M. (2017a). Diet-induced co-variation between architectural and physicochemical plasticity in an extended phenotype. *J. Exp. Biol.*, 220, 876–884.
- Blamires, S.J., Martens, P.J. & Kasumovic, M.M. (2017b). Fitness consequences of plasticity in an extended phenotype. *J. Exp. Biol.*, 221.
- Boutin, S. & Schweiger, S. (1988). Manipulation of intruder pressure in red squirrels (*Tamiasciurus hudsonicus*): effects on territory size and acquisition. *Can. J. Zool.*, 66, 2270–2274.
- Boutin, S., Larsen, K.W. & Berteaux, D. (2000). Anticipatory parental care: acquiring resources for offspring prior to conception. *Proc. R. Soc. B Biol. Sci.*, 267, 2081–2085.
- Cisterne, A., Vanderduys, E.P., Pike, D.A. & Schwarzkopf, L. (2014). Wary invaders and clever natives: sympatric house geckos show disparate responses to predator scent. *Behav. Ecol.*, 25, 604–611.
- Dawkins, R. (1978). Replicator selection and the extended phenotype. *Z. Tierpsychol.*, 47, 61–76.
- Dawkins, R. (1982). *The Extended Phenotype*. Oxford University Press, Oxford.
- Dawkins, R. (2004). Extended phenotype - But not too extended. A reply to Laland, Turner and Jablonka. *Biol. Philos.*, 19, 377–396.
- Descamps, S., Boutin, S., Berteaux, D. & Gaillard, J.M. (2007). Female red squirrels fit Williams’ hypothesis of increasing reproductive effort with increasing age. *J. Anim. Ecol.*, 76, 1192–1201.
- Descamps, S., Boutin, S., Berteaux, D. & Gaillard, J.-M. (2008). Age-specific variation in survival, reproductive success and offspring quality in red squirrels: evidence of senescence. *Oikos*, 117, 1406–1416.
- Dewan, I., Garland, T., Hiramatsu, L. & Careau, V. (2018). I smell a mouse: Indirect genetic effects on voluntary wheel-running distance, duration and speed. *Behav. Genet.*, 49, 1–11.



- Donald, J.L. & Boutin, S. (2011). Intraspecific cache pilferage by larder-hoarding red squirrels (*Tamiasciurus hudsonicus*). *J. Mammal.*, 92, 1013–1020.
- Filotas, E., Parrott, L., Burton, P.J., Chazdon, R.L., Coates, K.D., Coll, L. *et al.* (2014). Viewing forests through the lens of complex systems science. *Ecosphere*, 5, 1–23.
- Fisher, D.N., Boutin, S., Dantzer, B., Humphries, M.M., Lane, J.E. & McAdam, A.G. (2017). Multilevel and sex-specific selection on competitive traits in North American red squirrels. *Evolution (N. Y.)*, 71, 1841–1854.
- Fletcher, Q.E., Boutin, S., Lane, J.E., LaMontagne, J.M., McAdam, A.G., Krebs, C.J. *et al.* (2010). The functional response of a hoarding seed predator to mast seeding. *Ecology*, 91, 2673–2683.
- Griffing, B. (1967). Selection in reference to biological groups. I. Individual and group selection applied to populations of unordered groups. *Aust. J. Biol. Sci.*, 20, 127–139.
- Gunn, M.R., Dawson, D.A., Leviston, A., Hartnup, K., Davis, C.S., Strobeck, C. *et al.* (2005). Isolation of 18 polymorphic microsatellite loci from the North American red squirrel, *Tamiasciurus hudsonicus* (Sciuridae, Rodentia), and their cross-utility in other species. *Mol. Ecol. Notes*, 5, 650–653.
- Hadfield, J.D. (2010). MCMC methods for multi-response generalized linear mixed models: the MCMCglmm R package. *J. Stat. Softw.*, 33, 1–22.
- Haines, J.A. (2017). *Resources and reproductive trade-offs affect fitness, life history traits, and sexual selection in red squirrels*. University of Alberta, Edmonton.
- Hämäläinen, A., McAdam, A.G., Dantzer, B., Lane, J.E., Haines, J.A., Humphries, M.M. *et al.* (2017). Fitness consequences of peak reproductive effort in a resource pulse system. *Sci. Rep.*, 7, 9335.
- Hatt, R.T. (1929). The red squirrel: its life history and habits, with species reference to the Adirondacks of New York and the Harvard Forest. *Roosevelt Wildl. Ann.*, 2, 11–146.
- Heidelberger, P. & Welch, P.D. (1983). Simulation run length control in the presence of an initial transient. *Oper. Res.*, 31, 1109–1144.
- Hunter, P. (2009). Extended phenotype redux. How far can the reach of genes extend in manipulating the environment of an organism? *EMBO Rep.*, 10, 212–215.
- Jablonka, E. (2004). From replicators to heritably varying phenotypic traits: the extended phenotype revisited. *Biol. Philos.*, 19, 353–375.
- Jones, C.G., Lawton, J.H. & Shachak, M. (1994). Organisms as ecosystem engineers. *Ecosystem Management*. Springer, New York, NY, pp. 130–147.
- Jones, C.G., Lawton, J.H. & Shachak, M. (1997). Positive and negative effects of organisms as physical ecosystem engineers. *Ecology*, 78, 1946–1957.
- Jönsson, K.I. (1997). Capital and income breeding as alternative tactics of resource use in reproduction. *Oikos*, 78, 57.
- Kelly, D. (1994). The evolutionary ecology of mast seeding. *Trends Ecol. Evol.*, 9, 465–470.
- Kerr, T.D., Boutin, S., LaMontagne, J.M., McAdam, A.G. & Humphries, M.M. (2007). Persistent maternal effects on juvenile survival in North American red squirrels. *Biol. Lett.*, 3, 289–291.
- Kirkpatrick, M. & Lande, R. (1989). The evolution of maternal characters. *Evolution (N. Y.)*, 43, 485–503.
- Laidre, M.E. (2012). Niche construction drives social dependence in hermit crabs. *Curr. Biol.*, 22, R861–R863.
- LaMontagne, J.M. & Boutin, S. (2007). Local-scale synchrony and variability in mast seed production patterns of *Picea glauca*. *J. Ecol.*, 95, 991–1000.
- LaMontagne, J.M., Williams, C.T., Donald, J.L., Humphries, M.M., McAdam, A.G. & Boutin, S. (2013). Linking intraspecific variation in territory size, cone supply, and survival of North American red squirrels. *J. Mammal.*, 94, 1048–1058.
- Lane, J.E., Boutin, S., Gunn, M.R., Slate, J. & Coltman, D.W. (2007). Genetic relatedness of mates does not predict patterns of parentage in North American red squirrels. *Anim. Behav.*, 74, 611–619.
- Lane, J.E., Boutin, S., Speakman, J.R. & Humphries, M.M. (2010). Energetic costs of male reproduction in a scramble competition mating system. *J. Anim. Ecol.*, 79, 27–34.
- Lane, J.E., McAdam, A.G., Charmantier, A., Humphries, M.M., Coltman, D.W., Fletcher, Q. *et al.* (2015). Post-weaning parental care increases fitness but is not heritable in North American red squirrels. *J. Evol. Biol.*, 28, 1203–1212.
- Larivée, M.L., Boutin, S., Speakman, J.R., McAdam, A.G. & Humphries, M.M. (2010). Associations between over-winter survival and resting metabolic rate in juvenile North American red squirrels. *Funct. Ecol.*, 24, 597–607.
- Larsen, K.W. & Boutin, S. (1994). Movements, survival, and settlement of red squirrel (*Tamiasciurus hudsonicus*) offspring. *Ecology*, 75, 214–223.
- Larsen, K.W. & Boutin, S. (1995). Exploring territory quality in the North American red squirrel through removal experiments. *Can. J. Zool.*, 73, 1115–1122.
- McAdam, A.G., Boutin, S., Sykes, A.K. & Humphries, M.M. (2007). Life histories of female red squirrels and their contributions to population growth and lifetime fitness. *Ecoscience*, 14, 362.
- McFarlane, S.E., Lane, J.E., Taylor, R.W., Gorrell, J.C., Coltman, D.W., Humphries, M.M. *et al.* (2011). The heritability of multiple male mating in a promiscuous mammal. *Biol. Lett.*, 7, 368–371.
- Metz, H.C., Bedford, N.L., Pan, Y.L. & Hoekstra, H.E. (2017). Evolution and genetics of precocious burrowing behavior in *Peromyscus* mice. *Curr. Biol.*, 27, 3837–3845.e3.
- Moore, A.J., Brodie, E.D.I. & Wolf, J.B. (1997). Interacting phenotypes and the evolutionary process: i. Direct and indirect genetic effects of social interactions. *Evolution (N. Y.)*, 51, 1352–1362.
- Naiman, R.J., Melillo, J.M. & Hobbie, J.E. (1986). Ecosystem alteration of boreal forest streams by beaver (*Castor canadensis*). *Ecology*, 67, 1254–1269.
- Nussey, D.H., Froy, H., Lemaitre, J.-F., Gaillard, J.-M. & Austad, S.N. (2013). Senescence in natural populations of animals: widespread evidence and its implications for bio-gerontology. *Ageing Res. Rev.*, 12, 214–225.
- Odling-Smee, F.J., Laland, K.N. & Feldman, M.W. (2003). *Niche Construction: The Neglected Process in Evolution*. Princeton University Press, Princeton.
- Perna, A. & Theraulaz, G. (2017). When social behaviour is moulded in clay: on growth and form of social insect nests. *J. Exp. Biol.*, 220, 83–91.
- Posthumus, E.E., Koprowski, J.L. & Steidl, R.J. (2015). Red squirrel middens influence abundance but not diversity of other vertebrates. *PLoS ONE*, 10, e0123633.
- Price, K. & Boutin, S. (1993). Territorial bequeathal by red squirrel mothers. *Behav. Ecol.*, 4, 144–155.
- Price, K., Broughton, K., Boutin, S. & Sinclair, A.R.E. (1986). Territory size and ownership in red squirrels: response to removals. *Can. J. Zool.*, 64, 1144–1147.
- Réale, D., McAdam, A.G., Boutin, S. & Berteaux, D. (2003). Genetic and plastic responses of a northern mammal to climate change. *Proc. Biol. Sci.*, 270, 591–596.
- Rosell, F., Bozsér, O., Collen, P. & Parker, H. (2005). Ecological impact of beavers *Castor fiber* and *Castor canadensis* and their ability to modify ecosystems. *Mamm. Rev.*, 35, 248–276.
- Schielzeth, H. (2010). Simple means to improve the interpretability of regression coefficients. *Methods Ecol. Evol.*, 1, 103–113.
- Scott, J.P. (1977). Social genetics. *Behav. Genet.*, 7, 327–346.
- Scott-Phillips, T.C., Laland, K.N., Shuker, D.M., Dickins, T.E. & West, S.A. (2014). The niche construction perspective: a critical appraisal. *Evolution*, 68, 1231–1243.
- Simonsohn, U. (2018). Two-Lines: a valid alternative to the invalid testing of U-shaped relationships with quadratic regressions. *Adv. Methods Pract. Psychol. Sci.* 1, 538–555.
- Siracusa, E., Boutin, S., Gunn, M.R., Gorrell, J.C., Coltman, D.W., Dantzer, B. *et al.* (2017a). Familiarity with neighbours affects intrusion risk in territorial red squirrels. *Anim. Behav.*, 133, 11–20.

- Siracusa, E., Morandini, M., Boutin, S., Humphries, M.M., Dantzer, B., Lane, J.E. *et al.* (2017b). Red squirrel territorial vocalizations deter intrusions by conspecific rivals. *Behaviour*, 154, 1259–1273.
- Smith, C.C. (1968a). The adaptive nature of social organization in the genus of three squirrels *Tamiasciurus*. *Ecol. Monogr.*, 38, 31–64.
- Smith, M.C. (1968b). Red squirrel responses to spruce cone failure in Interior Alaska. *J. Wildl. Manage.*, 32, 305–317.
- Smith, C.C. (1978). Structure and function of the vocalizations of tree squirrels (*Tamiasciurus*). *J. Mammal.*, 59, 793–808.
- Smith, C.C. (1981). The indivisible niche of *Tamiasciurus*: an example of nonpartitioning of resources. *Ecol. Monogr.*, 51, 343–363.
- Sterelny, K., Smith, K.C. & Dickison, M. (1996). The extended replicator. *Biol. Philos.*, 11, 377–403.
- Thompson, L., Thomas, C.D., Radley, J.M.A., Williamson, S. & Lawton, J.H. (1993). The effect of earthworms and snails in a simple plant community. *Oecologia*, 95, 171–178.
- Turner, J.S. (2004). Extended phenotypes and extended organisms. *Biol. Philos.*, 19, 327–352.
- Weber, J.N., Peterson, B.K. & Hoekstra, H.E. (2013). Discrete genetic modules are responsible for complex burrow evolution in *Peromyscus* mice. *Nature*, 493, 402–405.
- Williams, C.T., Lane, J.E., Humphries, M.M., McAdam, A.G. & Boutin, S. (2014). Reproductive phenology of a food-hoarding mast-seed consumer: resource- and density-dependent benefits of early breeding in red squirrels. *Oecologia*, 174, 777–788.

#### SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

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