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Research Article

Silent enemies: can low-virulence macroparasites drive disease-mediated invasions?

Claudia Romeo[1,2*], Elisa Fesce[3*], Lucas A. Wauters[4], Francesca Santicchia[4], Peter W. W. Lurz[5], Andy White[6], Nicola Ferrari[3]

1 *Center for Evolutionary Hologenomics – Globe Institute, University of Copenhagen, Copenhagen, Denmark*
2 *Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna – IZSLER, Brescia, Italy*
3 *Department of Veterinary Medicine and Animal Sciences, Wildlife Health Laboratory, Università degli Studi di Milano, Lodi, Italy*
4 *Environmental Analysis and Management Unit, Guido Tosi Research Group, Department of Theoretical and Applied Sciences, Università degli Studi dell'Insubria, Varese, Italy*
5 *Royal (Dick) School of Veterinary Studies and Roslin Institute, University of Edinburgh, Easter Bush Campus, Roslin, UK*
6 *Department of Mathematics and the Maxwell Institute for Mathematical Sciences, Heriot-Watt University, Edinburgh, UK*

Corresponding author: Elisa Fesce (elisa.fesce@unimi.it)

Abstract

Shared pathogens can alter the interaction between native and alien species resulting in disease-mediated invasions (DMIs). Invasive animals often harbour low-virulence macroparasites, but empirical evidence for macroparasite-driven DMIs is still limited due to their subtile pathological impacts and scarce prominence. Here, we modelled the dynamics of native red squirrels, invasive grey squirrels and their shared nematode *Strongyloides robustus* to assess whether macroparasites can drive DMIs and lead to native species extinction. Our simulations showed that spill-over of the alien parasite can lead to red squirrel extinction, that grey squirrels can amplify the infection in the native host and that the infection can accelerate the replacement of red squirrels compared to direct competition alone, ultimately facilitating invasion by grey squirrels. These results show that low-virulence macroparasites can potentially mediate animal invasions, suggesting that we are overlooking key drivers of native species decline.

Key words: Alien species, helminths, host-parasite dynamics, invasive species, mathematical models, parasite-mediated competition, *Sciurus carolinensis*, *Sciurus vulgaris*

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Introduction

Biological invasions are a major threat to biodiversity worldwide and their impact is predicted to increase in the future (Seebens et al. 2021) and cause habitat alteration and native species extinction at a global scale (Bellard et al. 2016; Pyšek et al. 2020). Once alien species are introduced in a new range, they can interact with native species and the abiotic environment in multiple ways, leading to diversity loss in the native ecosystem (Simberloff 2011).

Beyond direct interactions, such as predation or exploitation competition for resources, alien species can impact native ones indirectly, for instance, by acting as reservoirs for shared pathogens (Bell et al. 2009; Strauss et al. 2012; Bellard et al.

* These authors contributed equally.

2016; Chinchio et al. 2020). Invasive alien species can carry alien pathogens that may spill over to native species or they may acquire local pathogens, possibly amplifying their circulation leading to spill-back (Kelly et al. 2009). Regardless of its origin, when a shared pathogen has a more severe impact on the native species than the alien one, parasite-mediated apparent competition (Hudson and Greenman 1998; Bell et al. 2009; Holt and Bonsall 2017) will favour the latter, possibly resulting in a disease-mediated invasion (DMI, Strauss et al. (2012)). Although field evidence for DMIs in animals is still limited (Prenter et al. 2004; Hatcher et al. 2006, 2012; Strauss et al. 2012), this phenomenon may be more prevalent in nature than has been identified to date, as all organisms harbour complex pathogen communities that are potentially capable of mediating their ecological interactions with competitors (Dobson et al. 2008).

In general, identifying and quantifying the impact of pathogens on wildlife population dynamics can be challenging (Tompkins et al. 2011) and especially so for macroparasites (i.e. helminths and parasitic arthropods). While viral and bacterial infections can be highly virulent, inducing overt clinical symptoms and leading to mortality events that are easier to detect, macroparasitic infections with more subtle pathological effects may often go unnoticed (McCallum and Dobson 1995). The quantification of such sublethal effects on animals in natural conditions is difficult and field experiments that manipulate pathogen transmission to demonstrate causal relationships are often challenging (Shanebeck et al. 2022). However, theoretical studies show that pathogens do not need to be highly prevalent or highly virulent to have regulatory effects on their host populations (Anderson and May 1978). Moreover, in the context of DMIs, less virulent pathogens are more likely to be introduced successfully in the new range compared to more virulent ones, which can be lost at the initial stages of invasion due to high lethality in low-density populations (MacLeod et al. 2010; Strauss et al. 2012; Lymbery et al. 2014). Therefore, the impact of a large number of macroparasitic infections on native-invasive species interactions is likely overlooked, potentially leading to underestimates of the frequency of DMIs and to potentially miss the drivers of native species extinctions.

Mathematical models can integrate field-derived data and system-specific biological processes and, thus, provide a crucial tool for understanding population and epidemiological dynamics that may be hidden in natural systems. In this way, rather than providing context-specific predictions, mathematical models can be used to indicate potential key processes and driving mechanisms by comparing hypothetical scenarios with observed patterns (Restif et al. 2012). A mathematical approach was pivotal in uncovering a well-known, case study example of disease-mediated invasion: the replacement of native red squirrels (*Sciurus vulgaris*) by invasive grey squirrels (*S. carolinensis*) driven by squirrel poxvirus (SQPV) in Great Britain and Ireland (Tompkins et al. 2003). Although red squirrels can be replaced by the alien congener in the absence of disease through exploitation competition for food (reviewed in Wauters et al. (2023)), modelling studies disclosed how SQPV carried by grey squirrels may accelerate the process, ultimately facilitating invasion (Tompkins et al. 2003; Rushton et al. 2005; Howell et al. 2024). In Italy, where grey squirrels were first introduced in 1948 (Martinoli et al. 2010), SQPV appears to be absent (Romeo et al. 2019). However, grey squirrels introduced a parasitic nematode, *Strongyloides robustus*, that can spill over to native red squirrels, reducing their survival (Romeo et al. 2021). The impact of

S. robustus on red squirrels at population scale and whether it affects invasion by grey squirrels is not known.

In this study, we will develop a general mathematical framework of a two host-one macroparasite system that can represent the two squirrel species and *S. robustus* to assess the impact of this low-virulence infection on the system dynamics. We will show that low-virulence macroparasites that induce no overt pathological signs can drive DMIs and lead to native species extinction. Since low-virulence pathogens are likely to be carried by invasive species (MacLeod et al. 2010; Strauss et al. 2012; Lymbery et al. 2014), our work will have important consequences for understanding the potential threat posed by these infections to the conservation of native species.

Materials and methods

Study system

Grey squirrels are native to the eastern part of North America and were introduced repeatedly into Europe, first in Great Britain starting from the second half of the 19th century and, later, in Ireland and Italy (Wauters et al. (2023) and references therein). In most habitats, grey squirrels outcompete the native red squirrel, ultimately driving the species to local extinction (Wauters et al. 2023, but see also Slade et al. (2021)). Replacement of red by grey squirrels occurs mainly through competition for food, leading to a decrease in red squirrels population growth rate via reduced birth and recruitment rates (reviewed in Wauters et al. (2023)). In Great Britain and Ireland, this process is also mediated by SQPV, a shared viral pathogen that has a subclinical impact on grey squirrels (Sainsbury et al. 2000; Tompkins et al. 2002; Chantrey et al. 2019), but is highly pathogenic to red squirrels, usually resulting in death within a few weeks (Tompkins et al. 2002; Chantrey et al. 2014; Fiegna et al. 2016). The role played by grey squirrels as reservoirs of the disease that drives this DMI was indicated by modelling studies (e.g. Tompkins et al. (2003); Rushton et al. (2005)). While SQPV is absent in Italy (Romeo et al. 2019), grey squirrels introduced in the area do carry the gastro-intestinal parasite *S. robustus* (Romeo et al. 2014b). *S. robustus* is a nematode native to North America, where it commonly infects several squirrel species (Chandler 1942; Davidson 1976; O'Brien et al. 2022). Adult female worms inhabit the small intestine of squirrels, where they reproduce asexually, shedding their eggs with host faeces (Bartlett 1995). Eggs hatch in the environment and host exposure to infective larvae likely occurs in dreys and on ground contaminated by faeces. The parasite appears to lack the adult free-living phase typical of other *Strongyloides* species (Bartlett 1995). The prevalence of *S. robustus* in Italian populations of grey squirrels varies between 57% and 81% (Romeo et al. 2014b, a, 2020; Santicchia et al. 2019) and is similar to what is reported in their native range (e.g. Davidson (1976)). *S. robustus* was found to spill over to naïve red squirrels in areas co-inhabited by grey squirrels, with prevalence in infected populations of the native species ranging between 57% and 75% (Romeo et al. 2015, 2021; Santicchia et al. 2020). Recently, two studies have demonstrated that infection by this alien parasite is detrimental to red squirrels at the individual level, decreasing their investment in costly behaviour (Santicchia et al. 2020) and significantly reducing their survival (Romeo et al. 2021).

Model structure

We extend a general, continuous time, deterministic model of a single host-macroparasite system (Greenman and Hudson 2000; Ferrari et al. 2007), shared macroparasite system that can represent the dynamics of red and grey squirrels and *S. robustus*. The model represents the density of red, H_R and grey, H_G, squirrels, the macroparasite abundance in red, P_R and grey, P_G, squirrels and the density of a common pool of free-living infective larvae, L. We modelled parasite abundance (i.e. the mean number of parasites per individual in the whole host population) instead of the whole parasite population size because abundance better reflects field-collected data.

A schematic of the model is shown in Fig. 1 with the model consisting of the following five coupled differential equations:

$$\frac{dH_R}{dt} = (b_R - d_R)\, H_R \left(1 - \frac{H_R}{K_R} - H_G \frac{c_G}{K_R} - \frac{\alpha_R}{d_R - b_R} P_R\right) \quad 1a$$

$$\frac{dH_G}{dt} = (b_G - d_G)\, H_G \left(1 - \frac{H_G}{K_G} - H_R \frac{c_R}{K_G} - \frac{\alpha_G}{d_G - b_G} P_G\right) \quad 1b$$

$$\frac{dP_R}{dt} = \beta_R \Psi L - (\sigma + \alpha_R + b_R) P_R - \frac{\alpha_R}{k_R} P_R^2 \quad 1c$$

$$\frac{dP_G}{dt} = \beta_G \Psi L - (\sigma + \alpha_G + b_G) P_G - \frac{\alpha_G}{k_G} P_G^2 \quad 1d$$

$$\frac{dL}{dt} = h(RH_R + H_G P_G) - \delta L - L(\beta_R H_R + \beta_G H_G) \quad 1e$$

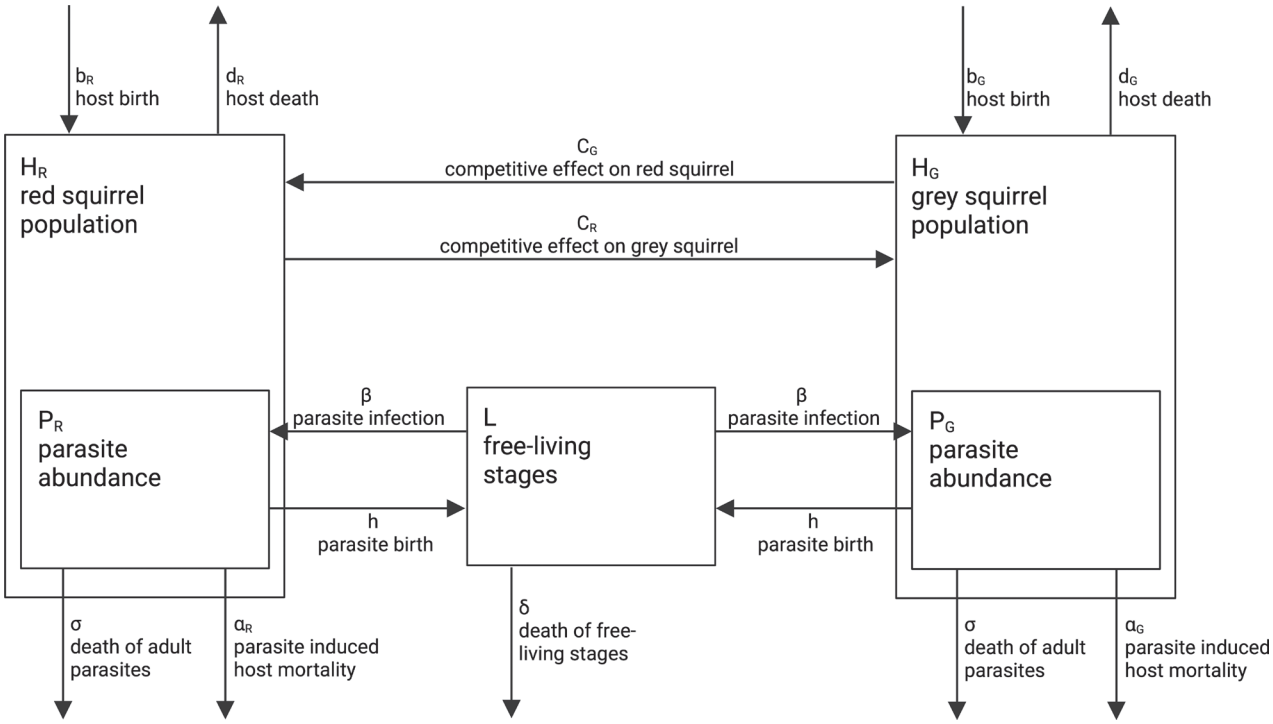

Figure 1. Model scheme. A schematic of the red and grey squirrel and *S. robustus* model system. Created in https://BioRender.com.

For each species (represented by subscript R and G), the host has birth rate *b* and death rate *d*. The demographic growth is offset by intra-specific competition through the carrying capacity K, while inter-specific competitions is accounted for through coefficient *c*. The host population is also reduced by parasite-induced mortality α. We did not include a parasite effect on host fertility, since field investigations did not reveal any effect of *S. robustus* on squirrel reproduction (Romeo et al. 2021). Parasite abundance increases due to encounters of free-living stages with hosts with coefficient β, scaled by the proportion of infecting larvae that develop to adult stages ψ and decreases with the adult parasite death rate σ. The parasite-induced mortality is scaled due to parasite aggregation *k* according to the negative binomial distribution (Anderson and May 1978; Greenman and Hudson 2000; Ferrari et al. 2007). Finally, the free-living larvae population increases with the instantaneous rate of production of infective stages by the two host species *h* and decreases with their death rate δ and through encounters with hosts with coefficient β.

Parameter estimation

We obtained the host demographic parameters from Tompkins et al. (2003), who modelled SQPV dynamics in the same system (see Table 1). This allowed a direct comparison of the effects of a low-virulence macroparasite (our study) with that of a high-virulence microparasite (Tompkins et al. 2003) on the DMI impact on red squirrels. Parasitological parameters, when available, were taken from long-term field studies carried out on this system in Italy or from studies on *S. robustus* in its native range, otherwise they were estimated from studies on other *Strongyloides* species. We derived *S. robustus* adult parasite mortality (σ) from *S. ratti* in rats, which has a lifespan of approximately one month (Wilkes et al. 2004). The death rate of the free-living phase (δ) was estimated from data on *S. robustus* eggs/larvae, whose survival in the environment varies between 12 (Wetzel and Weigl 1994) and 30 days (Bartlett 1995). We set the fertility of a single adult female (*h*) at 64 eggs/day, based on data from *S. robustus* in grey squirrels (Romeo et al. 2014a).

Infection by each *S. robustus* worm reduces the probability of red squirrel survival to the next year by 0.1 in males and by 0.08 in females (Romeo et al. 2021). This corresponds to an average parasite-induced death rate in red squirrels (α_R) of

Table 1. Parameter estimates for the two hosts-shared macroparasite system. R = red squirrel, G = grey squirrel.

Parameter	Value	Description	Reference
K_R, K_G	60, 80	Host population carrying capacity in a 5 km^2 area	Tompkins et al. (2003)
d	0.4 (year^{-1})	Instantaneous death rate of host due to non-parasitic causes	Tompkins et al. (2003)
b_R, b_G	1, 1.2 (year^{-1})	Instantaneous birth rate of host	Tompkins et al. (2003)
c_R, c_G	0.61, 1.65	Competitive effect of one host species on the other	Tompkins et al. (2003)
σ	12 (year^{-1})	Instantaneous death rate of adult parasites	Wilkes et al. (2004)
h	17520 (year^{-1})	Instantaneous rate of production of infective stages	Romeo et al. (2014a)
δ	17 (year^{-1})	Instantaneous death rate of infective free-living stages	Bartlett (1995); Wetzel and Weigl (1994)
ψ	0.7	Proportion of infecting larvae that develop to adult stages	
k	0.05	Aggregation parameter of the Negative Binomial distribution	Romeo et al. (2014a, b)
β	0.0004	Infection transmission coefficient for both species	Romeo et al. (2014b)
α_R, α_G	0.095, 0.0237 (year^{-1})	Instantaneous death rate of host due to parasite for both species	Romeo et al. (2021)

0.095. In the absence of empirical data, we set α_G as a quarter of α_R, based on the assumption of a better adaptation of grey squirrels to *S. robustus* and on theoretical host-parasites dynamics (Anderson and May 1978) where lower α values lead to higher parasite abundance, such as we observed in the alien grey squirrel (Romeo et al. 2014b). Due again to lack of empirical data, we arbitrarily set the proportion of larvae that successfully develop to the adult stage at 0.7 and we *a posteriori* set the encounter rate β in order to obtain the mean parasite abundance of 11 worms/squirrel observed in Italian grey squirrels (Romeo et al. 2014b). Since *S. robustus* is transmitted through environmental contamination and the two squirrels have similar ecology, we considered the same transmission coefficient β for both host species. Finally, based on previous data (Romeo et al. 2014a, b) we considered the aggregation parameter (*k*) of the negative binomial distribution to be on average 0.05.

Simulated scenarios

We simulated five different scenarios by changing the initial conditions of the model. In the one host scenarios 1A) and 1B), we simulated host-parasite dynamics separately in the two squirrel species (setting in turn H_R and H_G at zero). In the two host scenarios, we simulated the population dynamics of the two squirrel species together assuming in turn: 2A) infection by *S. robustus*, but no other forms of interaction between the two hosts (setting c_G and c_R at zero); 2B) infection by *S. robustus* and direct interspecific competition; 2C) direct interspecific competition in the absence of *S. robustus*, in order to quantify the relative contribution of parasite-mediated competition as opposed to the impact of between species competition for resources only. We started all the simulations assuming the introduction of two grey squirrels, while the red squirrel population was always set at its disease-free steady state (except in 1B). In scenarios that included the parasite, the *S. robustus* abundance in grey squirrels was initially set at 10 parasites/host, while for red squirrels, we assumed two parasites/host in scenario 1B.

We carried out a sensitivity analysis for all those parameters lacking empirical estimates, in order to assess their impact on the population and epidemiological dynamics. We performed additional simulations of scenario 2B, varying the values of these parameters within biologically plausible ranges (Suppl. material 1: table S1). All simulations were performed using the package '*deSolve*' (Soetaert et al. 2010) in R software (version 4.3.2, R Core Team 2023).

Results

One-host scenarios

Our simulations showed that the introduction of two grey squirrels infected by *S. robustus* in an area without other competing squirrels (scenario 1A) leads to a sharp increase in the host population size, almost reaching the carrying capacity in 9 years. The subsequent growth of the parasite population (which peaks at 24 parasites/squirrel after 10 years), then brings the host population down to 50 individuals, corresponding to 62.5% of the uninfected carrying capacity. The system reaches its steady state in 15 years, with a parasite abundance of 12.6 parasites/host (Fig. 2A).

The introduction of *S. robustus* into a naïve red squirrel population at carrying capacity and without competing grey squirrels (scenario 1B), leads to a rapid

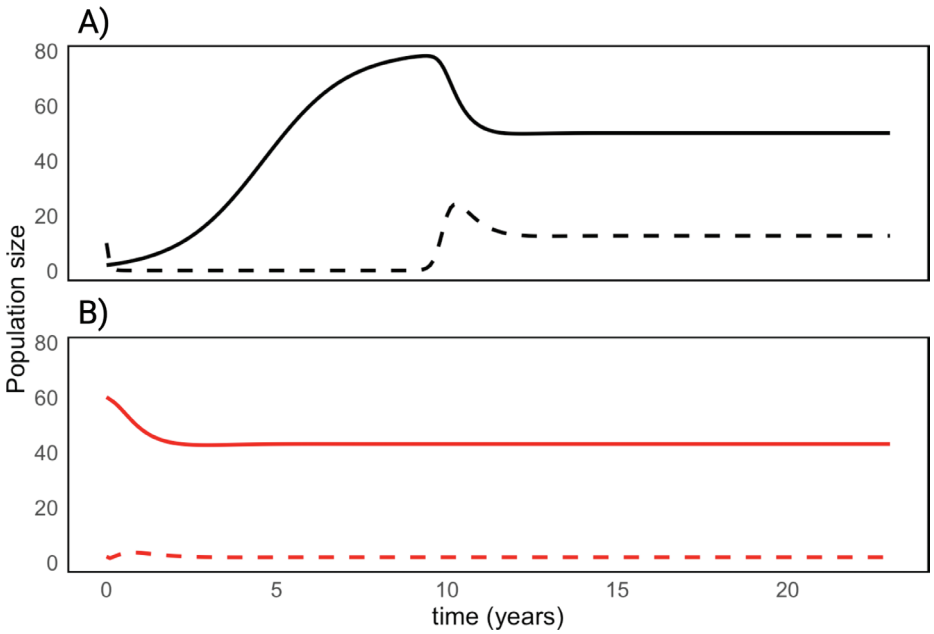

Figure 2. Population and infection dynamics in the one host scenarios. Population dynamics of red and grey squirrels in the one-host scenarios 1A and 1B. **A.** Shows grey squirrel density (black solid line) and *S. robustus* abundance in grey squirrel (black dashed line); **B.** Shows red squirrel density (red solid line) and *S. robustus* abundance in red squirrels (red dashed line).

decrease in the host population size down to 71.6% of the carrying capacity. The steady state of the system is reached in 6 years, with 43.4 red squirrels and a parasite abundance of 1.8 parasites/host (Fig. 2B).

Two-host scenarios

In the hypothetical condition of two infected grey squirrels being introduced into a red squirrel population and no direct competition for food occurring between the two species (scenario 2A), the simulations showed that red squirrels become extinct (i.e. population size < 2) in 15.2 years (Fig. 3A). Just before extinction, parasite abundance in red squirrels increases to 8.5 parasites/host (compared to 1.8 parasites/host in the sole red squirrel population). In this scenario, the grey squirrel population reaches its steady state in 17.2 years, at 50 individuals (62.5% of the uninfected carrying capacity as in the one host-one parasite scenario). When we accounted for both the effect of parasite-mediated competition and of trophic competition between red and grey squirrels (scenario 2B), the red squirrel population became extinct faster, in 10.3 years and grey squirrels reached their steady state at 50 individuals in only 11.7 years (Fig. 3B). For comparison, without parasites and considering only the effect of trophic competition (scenario 2C), red squirrels become extinct in 13.6 years and the grey squirrel population reaches the carrying capacity in 16.9 years (Fig. 3C).

Parameter sensitivity

Within scenario 2B (i.e. simulating the combined effect of parasite-mediated competition and trophic competition), all the tested parameter combinations drove the red squirrel population towards extinction (Suppl. material 1: fig. S1) in a timeframe between 7.5 and 16.5 years (Fig. 4).

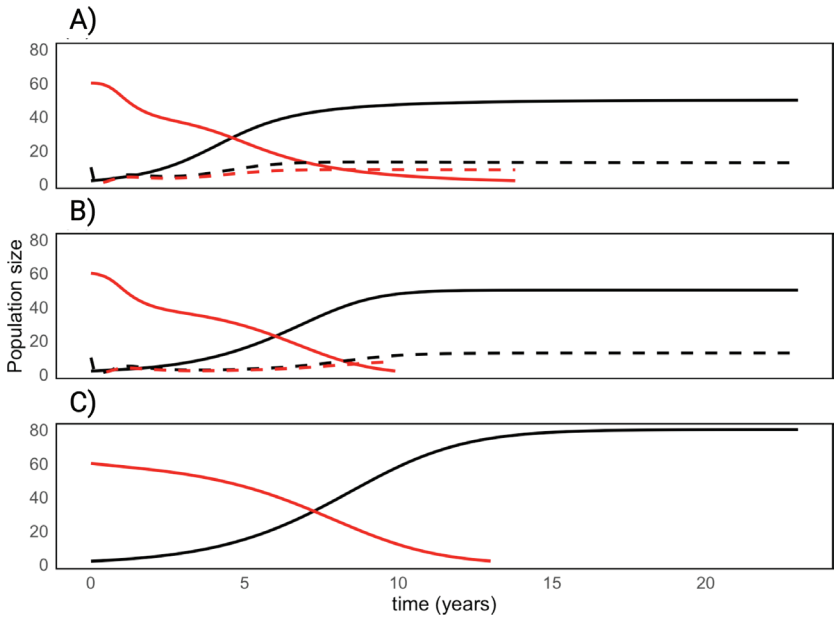

Figure 3. Population and infection dynamics in the two host scenarios. Population density of co-inhabiting red (red solid line) and grey squirrels (black solid line) and the abundance of *S. robustus* in red (red dashed line) and grey squirrels (black dashed line). **A.** Shows the predicted dynamics for scenario 2A (only parasite-mediated competition); **B.** Shows the predicted dynamics for scenario 2B (parasite-mediated competition and competition for food); **C.** Shows the predicted dynamics for scenario 2C (only competition for food).

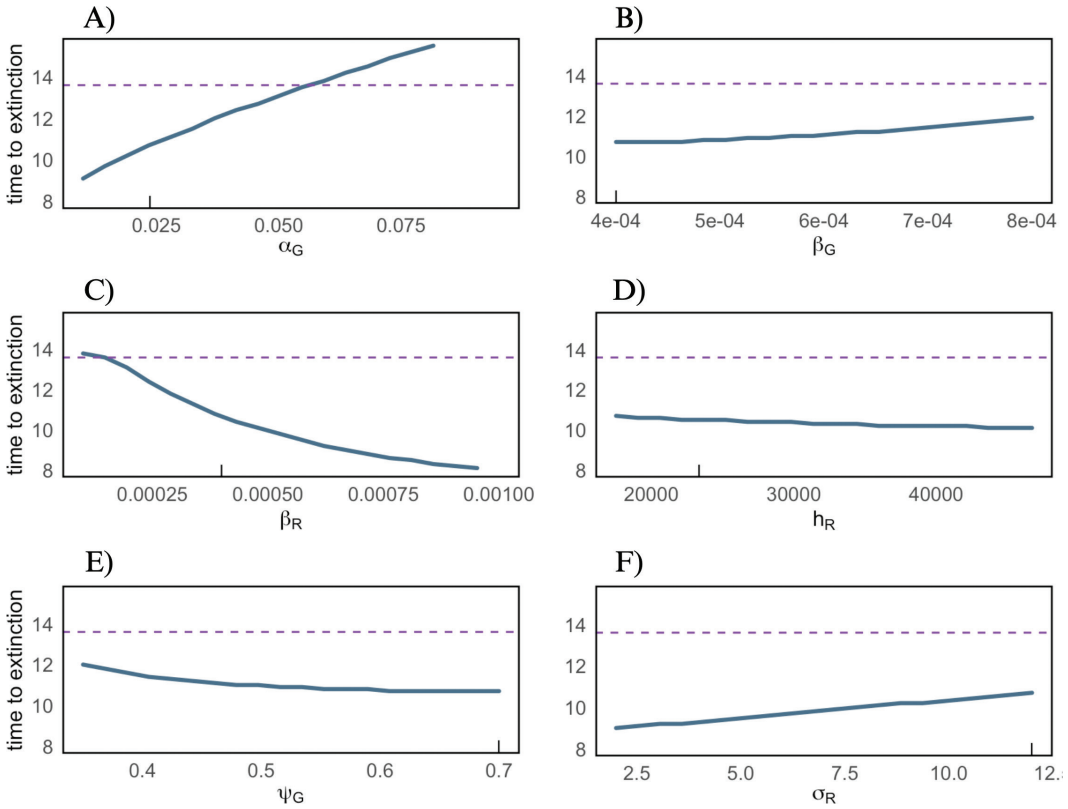

Figure 4. Effect of a change in epidemiological parameters on the time to extinction of red squirrels. **A.** Shows variation in parasite-induced mortality in grey squirrels (α_G); **B.** The infection transmission between grey squirrels and parasite infective stages (β_G); **C.** The infection transmission between red squirrels and parasite infective stages (β_R); **D.** The instantaneous production of infective stages by red squirrels (h_R); **E.** The proportion of infective stages that develop into adult stages in grey squirrels (ψ_G); **F.** The death rate of adult parasites in red squirrels (σ_R). The dashed line represents the time to extinction of red squirrels in the absence of the parasite, when only direct competition occurs. Inner tick marks on the x-axes indicate the value used in the simulated scenarios.

The instantaneous death rate of grey squirrels due to *S. robustus* (α_G, Fig. 4a) was the parameter with the greatest impact on the time to extinction for the red squirrel population, increasing it from 9.1 years needed for a lower α_G of 0.01, to 16.5 years for an α_G of 0.1. An increase in infection transmission between red squirrels and the parasite infective stages (β_R, Fig. 4c) also had a strong effect, reducing the time to red squirrel extinction from 13.8 to 7.9 years. Notably, an increase in infection transmission between grey squirrels and the parasite infective stages (β_G, Fig. 4b) had little effect on red squirrel replacement time, as did changes in h_R, ψ_G and σ_R (Fig. 4d–f).

Discussion

We modelled a two host-shared macroparasite system consisting of competing native red squirrels, alien grey squirrels and their gastro-intestinal nematode *S. robustus*. Our simulations showed that even low-virulence macroparasites, such as helminths, can play a major role in parasite-mediated competition and drive disease-mediated biological invasions.

The negative effect of alien *S. robustus* on individual red squirrels has been highlighted by empirical studies (Santicchia et al. 2020; Romeo et al. 2021); however, it was unclear whether this translated into impacts at the population scale. A key finding from our study is that grey squirrels can amplify *S. robustus* load in red squirrels by a factor of 4.7. As a consequence, parasite spill-over can facilitate invasion by grey squirrels by reducing the time needed for red squirrel extinction by 24% compared to in the absence of the parasite. Furthermore, our hypothetical model scenario that excluded interspecific competition between red and grey squirrels showed that parasite amplification from the invasive species alone may be sufficient to drive the native species to extinction. Spill-over and spill-back DMIs have been shown to occur in a range of plant, invertebrate and vertebrate systems (Strauss et al. 2012) and our study highlights how they can be driven by a shared macroparasitic infection. Our findings further emphasise how subtle, low-virulence infections can play a key role in DMIs, but their lack of prominence in the native system means that they may be overlooked as drivers of native species decline.

Our model findings showed that grey squirrels are not mere carriers of the parasite, but play a pivotal role in the red squirrel-*S. robustus* epidemiological interaction. The model results are supported by field data that indicate how grey squirrels are not essential for *S. robustus* maintenance, as the infection was observed to persist in red squirrel populations several months after grey squirrels' removal (unpublished data). Nevertheless, our results clearly showed the importance of grey squirrels as a source of infection, as their presence detrimentally alters the outcome for red squirrels at the population scale. In the absence of the alien host, we showed that *S. robustus* would have a small impact on the native squirrel population, but when the invasive species is present, the native species is driven to extinction due to increased infection transmission and heightened parasite load. In this context, our work opens some interesting questions on what are the key properties that make a parasite able to mediate the interaction between closely-related species. For the red and grey squirrel system, it has been shown that a highly virulent pathogen (SQPV) can lead to DMI – but our work also indicates that a sub-lethal parasite can play a key role in native species replacement.

We acknowledge limitations with our model study related to the lack of accurate empirical data specific to this host-parasite system. For instance, with the exception of few data gathered from the opportunistic collection of road-kills (Romeo et al. 2013), no empirical estimates of adult parasite abundance in red squirrels were available. To estimate the encounter rate with the parasite, we had, therefore, to rely on the average adult *S. robustus* abundance observed in grey squirrels, which is, instead, based on robust data collected from many individuals culled within control programmes (e.g. Romeo et al. (2014a, b)). Furthermore, while an indirect measure of *S. robustus*-induced mortality was available for red squirrels (Romeo et al. 2021), we lacked an empirical quantification of the parasite's impact in grey squirrels. However, our analyses showed that the time needed for red squirrels to become extinct was not sensitive to changes in most of the epidemiological parameters. Key parameters were the level of disease-induced mortality in grey and red squirrels and the transmission coefficient of the infection. The low virulence of *S. robustus* in grey squirrels leads to a high prevalence of infection and, in turn, to an increase in the density of parasite infective stages which can be transmitted to red squirrels. The time to replacement of red squirrels is, therefore, sensitive to these parameters and can decrease if the virulence in grey squirrels is reduced or transmission of infection to red squirrels is increased. It is worth noting that, if the virulence in grey squirrels is greater than in red squirrels, the time to red squirrel replacement can increase and the invasion can potentially be prevented (see Hilker et al. (2005)).

Our findings support a recent modelling study on red and grey squirrels and SQPV in Great Britain (Howell et al. 2024), which found that infection transmission between red squirrels was a key factor in their replacement by invasive grey squirrels. The authors used recently-gathered epidemiological and immunological evidence to highlight how partial immunity to SQPV in grey squirrels will strongly influence the epidemiological dynamics. There is a lack of information about the immune response against *S. robustus* in the red and grey squirrel system in Italy, but it is known that the response against *Strongyloides* spp. in other mammals is mainly directed at reducing parasitic females' size and fecundity (Viney et al. 2006). A differential immune response in the two host species might, therefore, have relevant implications on the number of eggs shed with faeces and, in turn, on encounter rates with infective stages. Interestingly though, our simulations suggest that the number of eggs shed by red squirrels has a negligible effect on the dynamics of the system.

Although we are aware that the Italian red and grey squirrel system has different population dynamics compared to populations in Great Britain (Gurnell et al. 2004; Wauters et al. 2023), we took host demographic parameters from Tompkins et al. (2003), so we could compare our findings to those that demonstrated SQPV-mediated competition. However, it would be important to test habitat-specific model extensions, as host-parasite dynamics might be strongly affected by environmental characteristics through their influence on target and reservoir host densities (Altizer et al. 2006; Tompkins et al. 2011). Epidemiological theory states that the size of a reservoir population should be above a specific critical threshold for the infection to be maintained and for spill-over to the target population to occur (Haydon et al. 2002; Viana et al. 2014). For instance, modelling studies on SQPV showed that, in conifer forests, where density of squirrels is low and red squirrels seem to have a competitive advantage over the alien species (Slade et al. 2021), the role of the disease in the interaction between the two hosts becomes negligible (Howell et al. 2024). Likewise, predators such as pine martens, who selectively prey

grey over red squirrels (Sheehy et al. 2018), are able to effectively suppress SQPV circulation by reducing the reservoir host density (Slade et al. 2023). Indeed, grey squirrel control has been proven effective in containing SQPV outbreaks in red squirrel populations (Schuchert et al. 2014) because, without continuous spill-over from the reservoir host, highly virulent SQPV can fade out following outbreaks in the red squirrel population (White et al. 2014; Macpherson et al. 2016). Our simulations and field data suggest, however, that the epidemiology of *S. robustus* would be different as it may persist indefinitely in red-only populations even after grey squirrels have been removed. This latter fact requires further investigation as it has potentially serious implications should *S. robustus* ever be introduced to the UK system. Currently, red squirrels have a relative safe-haven in large, conifer-dominated stronghold forests in the UK (Slade et al. 2021). An indefinitely persisting macroparasite infection by *S. robustus* may alter the red-grey-SQPV-system in the UK in such a way that no safe-haven remains for the native squirrel.

Conclusion

Using a theoretical approach and a well-studied model system, we showed that a low-virulence nematode can act as an invasion driver in a similar way to more virulent pathogens. However, we currently lack long data series that would allow us to test whether our mathematical model is able to replicate the dynamics observed in the field. Indeed, our study highlights critical knowledge gaps that should be addressed in field studies to improve our understanding of these systems. In this sense, our work can serve as an example of the model-guided fieldwork framework, in which feedback loops are established between empiricists and modellers for the progression of research (Restif et al. 2012). This study is nonetheless grounded on an extensive body of empirical and theoretical knowledge gathered on this invasive-native vertebrate system, which is probably one of the most well-studied biological invasions in animals (Wauters et al. 2023). Thanks to the research carried out over the last 50 years, we now know that the interaction between these two squirrel species is much more complex than previously thought, with several drivers concurring in mediating the invasion. It is unlikely that this specific system is unique in its complexity; hence, this highlights once again how we are potentially missing several underpinning mechanisms of animal invasions.

Additional information

Conflict of interest

The authors have declared that no competing interests exist.

Ethical statement

No ethical statement was reported.

Use of AI

No use of AI was reported.

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Author contributions

CR: conceptualisation, methodology, validation, writing – original draft; EF: conceptualisation, formal analysis, methodology, software, visualisation, writing – review and editing; LAW: validation, writing – review and editing; FS: validation, writing – review and editing; PWWL: validation, writing – review and editing; AW: validation, supervision, writing – review and editing; NF: conceptualisation, formal analysis, funding acquisition, methodology, project administration, supervision, writing – review and editing.

Author ORCIDs

Claudia Romeo https://orcid.org/0000-0002-5964-2847
Elisa Fesce https://orcid.org/0000-0002-2535-3684
Lucas A. Wauters https://orcid.org/0000-0002-4871-5035
Francesca Santicchia https://orcid.org/0000-0003-4814-9632
Peter W.W. Lurz https://orcid.org/0000-0003-4199-5211
Andy White https://orcid.org/0000-0002-0677-5531
Nicola Ferrari https://orcid.org/0000-0001-8959-3495

Data availability

This type of article is not based on research data and no new data were created or analysed in this study. Modelling is based on previously published data that are referenced in the text. The code for all analyses is available at: https://github.com/elisf02/DMI_SrobustusSquirrel.git.

References

Altizer S, Dobson AP, Hosseini P, Hudson PJ, Pascual M, Rohani P (2006) Seasonality and the dynamics of infectious diseases. Ecology Letters 9: 467–484. https://doi.org/10.1111/j.1461-0248.2005.00879.x

Anderson RM, May RM (1978) Regulation and stability of host-parasite population interactions: I. regulatory processes. Journal of Animal Ecology 47: 219–247. https://doi.org/10.2307/3933

Bartlett C (1995) Morphology, homogonic development, and lack of a free-living generation in *Strongyloides robustus* (Nematoda, Rhabditoidea), a parasite of North American sciurids. Folia Parasitologica 42: 102–114.

Bell SS, White A, Sherratt JA, Boots M (2009) Invading with biological weapons: The role of shared disease in ecological invasion. Theoretical Ecology 2: 53–66. https://doi.org/10.1007/s12080-008-0029-x

Bellard C, Cassey P, Blackburn TM (2016) Alien species as a driver of recent extinctions. Biology Letters 12: e20150623. https://doi.org/10.1098/rsbl.2015.0623

Chandler AC (1942) Helminths of tree squirrels in southeast Texas. The Journal of Parasitology 28: 135–140. https://doi.org/10.2307/3272725

Chantrey J, Dale TD, Read JM, White S, Whitfield F, Jones D, McInnes CJ, Begon M (2014) European red squirrel population dynamics driven by squirrelpox at a gray squirrel invasion interface. Ecology and Evolution 4: 3788–3799. https://doi.org/10.1002/ece3.1216

Chantrey J, Dale T, Jones D, Begon M, Fenton A (2019) The drivers of squirrelpox virus dynamics in its grey squirrel reservoir host. Epidemics 28: e100352. https://doi.org/10.1016/j.epidem.2019.100352

Chinchio E, Crotta M, Romeo C, Drewe JA, Guitian J, Ferrari N (2020) Invasive alien species and disease risk: An open challenge in public and animal health. PLoS Pathogens 16: e1008922. https://doi.org/10.1371/journal.ppat.1008922

Davidson WR (1976) Endoparasites of selected populations of gray squirrels (*Sciurus carolinensis*) in the southeastern United States. Proceedings of the Helminthological Society of Washington 43: 211–217.

Dobson A, Lafferty KD, Kuris AM, Hechinger RF, Jetz W (2008) Homage to Linnaeus: How many parasites? How many hosts? Proceedings of the National Academy of Sciences of the United States of America 105: 11482–11489. https://doi.org/10.1073/pnas.0803232105

Ferrari N, Rosà R, Pugliese A, Hudson PJ (2007) The role of sex in parasite dynamics: Model simulations on transmission of *Heligmosomoides polygyrus* in populations of yellow-necked mice, *Apodemus flavicollis.* International Journal for Parasitology 37: 341–349. https://doi.org/10.1016/j.ijpara.2006.10.015

Fiegna C, Dagleish MP, Coulter L, Milne E, Meredith A, Finlayson J, Di Nardo A, McInnes CJ (2016) Host-pathogen dynamics of squirrelpox virus infection in red squirrels (*Sciurus vulgaris*). Veterinary Microbiology 182: 18–27. https://doi.org/10.1016/j.vetmic.2015.10.012

Greenman JV, Hudson PJ (2000) Parasite-mediated and direct competition in a two-host shared macroparasite system. Theoretical Population Biology 57: 13–34. https://doi.org/10.1006/tpbi.1999.1435

Gurnell J, Wauters LA, Lurz PWW, Tosi G (2004) Alien species and interspecific competition: Effects of introduced eastern grey squirrels on red squirrel population dynamics. Journal of Animal Ecology 73: 26–35. https://doi.org/10.1111/j.1365-2656.2004.00791.x

Hatcher MJ, Dick JTA, Dunn AM (2006) How parasites affect interactions between competitors and predators. Ecology Letters 9: 1253–1271. https://doi.org/10.1111/j.1461-0248.2006.00964.x

Hatcher MJ, Dick JTA, Dunn AM (2012) Disease emergence and invasions. Functional Ecology 26: 1275–1287. https://doi.org/10.1111/j.1365-2435.2012.02031.x

Haydon DT, Cleaveland S, Taylor LH, Laurenson KM (2002) Identifying reservoirs of infection: A conceptual and practical challenge. Emerging Infectious Diseases 12(8): 1468–1473. https://doi.org/10.3201/eid0812.010317

Hilker FM, Lewis MA, Seno H, Langlais M, Malchow H (2005) Pathogens can Slow down or reverse invasion fronts of their hosts. Biological Invasions 7: 817–832. https://doi.org/10.1007/s10530-005-5215-9

Holt RD, Bonsall MB (2017) Apparent competition. Annual Review of Ecology, Evolution, and Systematics 48: 447–471. https://doi.org/10.1146/annurev-ecolsys-110316-022628

Howell E, White A, Lurz PWW, Boots M (2024) Immune interactions and heterogeneity in transmission drives the pathogen-mediated invasion of grey squirrels in the UK. Journal of Animal Ecology 93: 663–675. https://doi.org/10.1111/1365-2656.14074

Hudson P, Greenman J (1998) Competition mediated by parasites: Biological and theoretical progress. Trends in Ecology & Evolution 13: 387–390. https://doi.org/10.1016/S0169-5347(98)01475-X

Kelly DW, Paterson RA, Townsend CR, Poulin R, Tompkins DM (2009) Parasite spillback: A neglected concept in invasion ecology? Ecology 90: 2047–2056. https://doi.org/10.1890/08-1085.1

Lymbery AJ, Morine M, Kanani HG, Beatty SJ, Morgan DL (2014) Co-invaders: The effects of alien parasites on native hosts. International Journal for Parasitology. Parasites and Wildlife 3: 171–177. https://doi.org/10.1016/j.ijppaw.2014.04.002

MacLeod CJ, Paterson AM, Tompkins DM, Duncan RP (2010) Parasites lost – do invaders miss the boat or drown on arrival? Ecology Letters 13: 516–527. https://doi.org/10.1111/j.1461-0248.2010.01446.x

Macpherson MF, Davidson RS, Duncan DB, Lurz PW, Jarrott A, White A (2016) Incorporating habitat distribution in wildlife disease models: Conservation implications for the threat of squirrelpox on the Isle of Arran. Animal Conservation 19: 3–14. https://doi.org/10.1111/acv.12219

Martinoli A, Bertolino S, Preatoni DG, Balduzzi A, Marsan A, Genovesi P, Tosi G, Wauters LA (2010) Headcount 2010: The multiplication of the grey squirrel populations introduced to Italy. Hystrix, the Italian Journal of Mammalogy 21: 127–136.

McCallum H, Dobson A (1995) Detecting disease and parasite threats to endangered species and ecosystems. Trends in Ecology & Evolution 10: 190–194. https://doi.org/10.1016/S0169-5347(00)89050-3

O'Brien PP, Bowman J, Newar SL, Garroway CJ (2022) Testing the parasite-mediated competition hypothesis between sympatric northern and southern flying squirrels. International Journal for Parasitology. Parasites and Wildlife 17: 83–90. https://doi.org/10.1016/j.ijppaw.2021.11.001

Prenter J, MacNeil C, Dick JTA, Dunn AM (2004) Roles of parasites in animal invasions. Trends in Ecology & Evolution 19: 385–390. https://doi.org/10.1016/j.tree.2004.05.002

Pyšek P, Hulme PE, Simberloff D, Bacher S, Blackburn TM, Carlton JT, Dawson W, Essl F, Foxcroft LC, Genovesi P, Jeschke JM, Kühn I, Liebhold AM, Mandrak NE, Meyerson LA, Pauchard A, Pergl J, Roy HE, Seebens H, van Kleunen M, Vilà M, Wingfield MJ, Richardson DM (2020) Scientists' warning on invasive alien species. Biological Reviews of the Cambridge Philosophical Society 95: 1511–1534. https://doi.org/10.1111/brv.12627

R Core Team (2023) R: A language and environment for statistical computing. Austria.

Restif O, Hayman DTS, Pulliam JRC, Plowright RK, George DB, Luis AD, Cunningham AA, Bowen RA, Fooks AR, O'Shea TJ, Wood JLN, Webb CT (2012) Model-guided fieldwork: Practical guidelines for multidisciplinary research on wildlife ecological and epidemiological dynamics. Ecology Letters 15: 1083–1094. https://doi.org/10.1111/j.1461-0248.2012.01836.x

Romeo C, Pisanu B, Ferrari N, Basset F, Tillon L, Wauters LA, Martinoli A, Saino N, Chapuis J-L (2013) Macroparasite community of the Eurasian red squirrel (*Sciurus vulgaris*): Poor species richness and diversity. Parasitology Research 112: 3527–3536. https://doi.org/10.1007/s00436-013-3535-8

Romeo C, Wauters LA, Cauchie S, Martinoli A, Matthysen E, Saino N, Ferrari N (2014a) Faecal egg counts from field experiment reveal density dependence in helminth fecundity: Strongyloides robustus infecting grey squirrels (*Sciurus carolinensis*). Parasitology Research 113: 3403–3408. https://doi.org/10.1007/s00436-014-4005-7

Romeo C, Wauters LA, Ferrari N, Lanfranchi P, Martinoli A, Pisanu B, Preatoni DG, Saino N (2014b) Macroparasite fauna of alien grey squirrels (*Sciurus carolinensis*): Composition, variability and implications for native species. PLoS ONE 9: e88002. https://doi.org/10.1371/journal.pone.0088002

Romeo C, Ferrari N, Lanfranchi P, Saino N, Santicchia F, Martinoli A, Wauters LA (2015) Biodiversity threats from outside to inside: Effects of alien grey squirrel (*Sciurus carolinensis*) on helminth community of native red squirrel (*Sciurus vulgaris*). Parasitology Research 114: 2621–2628. https://doi.org/10.1007/s00436-015-4466-3

Romeo C, McInnes CJ, Dale TD, Shuttleworth C, Bertolino S, Wauters LA, Ferrari N (2019) Disease, invasions and conservation: No evidence of squirrelpox virus in grey squirrels introduced to Italy. Animal Conservation 22: 14–23. https://doi.org/10.1111/acv.12433

Romeo C, Wauters LA, Santicchia F, Dantzer B, Palme R, Martinoli A, Ferrari N (2020) Complex relationships between physiological stress and endoparasite infections in natural populations. Current Zoology 66: 449–457. https://doi.org/10.1093/cz/zoaa029

Romeo C, Piscitelli AP, Santicchia F, Martinoli A, Ferrari N, Wauters LA (2021) Invading parasites: Spillover of an alien nematode reduces survival in a native species. Biological Invasions 23: 3847–3857. https://doi.org/10.1007/s10530-021-02611-7

Rushton SP, Lurz PWW, Gurnell J, Nettleton P, Bruemmer C, Shirley MDF, Sainsbury AW (2005) Disease threats posed by alien species: The role of a poxvirus in the decline of the native red squirrel in Britain. Epidemiology and Infection 134: e521. https://doi.org/10.1017/S0950268805005303

Sainsbury AW, Nettleton P, Gilray J, Gurnell J (2000) Grey squirrels have high seroprevalence to a parapoxvirus associated with deaths in red squirrels. Animal Conservation 3: 229–233. https://doi.org/10.1111/j.1469-1795.2000.tb00107.x

Santicchia F, Romeo C, Ferrari N, Matthysen E, Vanlauwe L, Wauters LA, Martinoli A (2019) The price of being bold? Relationship between personality and endoparasitic infection in a tree squirrel. Mammalian Biology 97: 1–8. https://doi.org/10.1016/j.mambio.2019.04.007

Santicchia F, Wauters LA, Piscitelli AP, Dongen SV, Martinoli A, Preatoni D, Romeo C, Ferrari N (2020) Spillover of an alien parasite reduces expression of costly behaviour in native host species. Journal of Animal Ecology 89: 1559–1569. https://doi.org/10.1111/1365-2656.13219

Schuchert P, Shuttleworth CM, McInnes CJ, Everest DJ, Rushton SP (2014) Landscape scale impacts of culling upon a European grey squirrel population: Can trapping reduce population size and decrease the threat of squirrelpox virus infection for the native red squirrel? Biological Invasions 16: 2381–2391. https://doi.org/10.1007/s10530-014-0671-8

Seebens H, Bacher S, Blackburn TM, Capinha C, Dawson W, Dullinger S, Genovesi P, Hulme PE, van Kleunen M, Kühn I, Jeschke JM, Lenzner B, Liebhold AM, Pattison Z, Pergl J, Pyšek P, Winter M, Essl F (2021) Projecting the continental accumulation of alien species through to 2050. Global Change Biology 27: 970–982. https://doi.org/10.1111/gcb.15333

Shanebeck KM, Besson AA, Lagrue C, Green SJ (2022) The energetic costs of sub-lethal helminth parasites in mammals: A meta-analysis. Biological Reviews of the Cambridge Philosophical Society 97: 1886–1907. https://doi.org/10.1111/brv.12867

Sheehy E, Sutherland C, O'Reilly C, Lambin X (2018) The enemy of my enemy is my friend: Native pine marten recovery reverses the decline of the red squirrel by suppressing grey squirrel populations. Proceedings. Biological Sciences 285: e20172603. https://doi.org/10.1098/rspb.2017.2603

Simberloff D (2011) How common are invasion-induced ecosystem impacts? Biological Invasions 13: 1255–1268. https://doi.org/10.1007/s10530-011-9956-3

Slade A, White A, Kortland K, Lurz PWW (2021) Natural strongholds for red squirrel conservation in Scotland. Nature Conservation 43: 93–108. https://doi.org/10.3897/natureconservation.43.62864

Slade A, White A, Lurz PWW, Shuttleworth C, Tosh DG, Twining JP (2023) Indirect effects of pine marten recovery result in benefits to native prey through suppression of an invasive species and a shared pathogen. Ecological Modelling 476: e110216. https://doi.org/10.1016/j.ecolmodel.2022.110216

Soetaert K, Petzoldt T, Setzer RW (2010) Solving differential equations in R: Package deSolve. Journal of Statistical Software 33: 1–25. https://doi.org/10.18637/jss.v033.i09

Strauss A, White A, Boots M (2012) Invading with biological weapons: The importance of disease-mediated invasions. Functional Ecology 26: 1249–1261. https://doi.org/10.1111/1365-2435.12011

Tompkins DM, Sainsbury AW, Nettleton P, Buxton D, Gurnell J (2002) Parapoxvirus causes a deleterious disease in red squirrels associated with UK population declines. Proceedings of the Royal Society of London. Series B, Biological Sciences 269: 529–533. https://doi.org/10.1098/rspb.2001.1897

Tompkins DM, White AR, Boots M (2003) Ecological replacement of native red squirrels by invasive greys driven by disease. Ecology Letters 6: 189–196. https://doi.org/10.1046/j.1461-0248.2003.00417.x

Tompkins DM, Dunn AM, Smith MJ, Telfer S (2011) Wildlife diseases: From individuals to ecosystems. Journal of Animal Ecology 80: 19–38. https://doi.org/10.1111/j.1365-2656.2010.01742.x

Viana M, Mancy R, Biek R, Cleaveland S, Cross PC, Lloyd-Smith JO, Haydon DT (2014) Assembling evidence for identifying reservoirs of infection. Trends in Ecology & Evolution 29: 270–279. https://doi.org/10.1016/j.tree.2014.03.002

Viney ME, Steer MD, Wilkes CP (2006) The reversibility of constraints on size and fecundity in the parasitic nematode Strongyloides ratti. Parasitology 133: 477–483. https://doi.org/10.1017/S003118200600062X

Wauters LA, Lurz PWW, Santicchia F, Romeo C, Ferrari N, Martinoli A, Gurnell J (2023) Interactions between native and invasive species: A systematic review of the red squirrel-gray squirrel paradigm. Frontiers in Ecology and Evolution 11. https://doi.org/10.3389/fevo.2023.1083008

Wetzel EJ, Weigl PD (1994) Ecological implications for flying squirrels (*Glaucomys* spp.) of effects of temperature on the in vitro development and behavior of *Strongyloides robustus*. American Midland Naturalist 131: 1–43. https://doi.org/10.2307/2426607

White A, Bell SS, Lurz PWW, Boots M (2014) Conservation management within strongholds in the face of disease-mediated invasions: Red and grey squirrels as a case study. Journal of Applied Ecology 51: 1631–1642. https://doi.org/10.1111/1365-2664.12274

Wilkes CP, Thompson FJ, Gardner MP, Paterson S, Viney ME (2004) The effect of the host immune response on the parasitic nematode *Strongyloides ratti*. Parasitology 128: 661–669. https://doi.org/10.1017/S0031182004005062

Supplementary material 1

Supporting information

Authors: Claudia Romeo, Elisa Fesce, Lucas A. Wauters, Francesca Santicchia, Peter W.W. Lurz, Andy White, Nicola Ferrari

Data type: pdf

Explanation note: This supplementary file includes a table and a figure with additional details on the sensitivity analysis. The table presents the parameter ranges, while the figure illustrates the impact of key parameters on grey and red squirrel populations and parasite abundance in grey squirrels.

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