Parasite local maladaptation in the Canarian lizard *Gallotia galloti* (Reptilia: Lacertidae) parasitized by haemogregarian blood parasite

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Abstract

Biologists commonly assume that parasites are locally adapted since they have shorter generation times and higher fecundity than their hosts, and therefore evolve faster in the arms race against the host's defences. As a result, parasites should be better able to infect hosts within their local population than hosts from other allopatric populations. However, recent mathematical modelling has demonstrated that when hosts have higher migration rates than parasites, hosts may diversify their genes faster than parasites and thus parasites may become locally maladapted. This new model was tested on the Canarian endemic lizard and its blood parasite (haemogregarine genus). In this host–parasite system, hosts migrate more than parasites since lizard offspring typically disperse from their natal site soon after hatching and without any contact with their parents who are potential carriers of the intermediate vector of the blood parasite (a mite). Results of cross-infection among three lizard populations showed that parasites were better at infecting individuals from allopatric populations than individuals from their sympatric population. This suggests that, in this host–parasite system, the parasites are locally maladapted to their host.

Introduction

Local adaptation defines a situation in which a population's mean fitness is on average larger in the natal environment than in another environment and when the mean fitness of the native population is greater than the average mean's fitness of a transplanted population (see Gandon *et al.*, 1996, 1998; Gandon & Van Zandt, 1998). Local adaptation can be relatively easily studied within host–parasite systems since cross-infection among different populations allows comparisons of the performances of a parasite in native and novel hosts. Several studies have demonstrated that parasites were more efficient in infecting hosts from their native population, i.e. sympatric hosts, compared with hosts from other populations, i.e. allopatric hosts (Parker, 1985; Lively, 1989; Ballabeni & Ward, 1993; Ebert, 1994; Failloux *et al.*, 1995; Manning *et al.*, 1995; Mopper *et al.*, 1995; Morand *et al.*, 1996). Therefore, it has been commonly assumed that parasites are locally adapted since they have a higher evolutionary potential than their host. Indeed, since parasites normally have larger population sizes and shorter generation times than their hosts, they should evolve faster in the arms race against their hosts. However, a recent model accounting for the migration rates of both hosts and parasites suggests that, in certain circumstances, parasites could be locally maladapted (Gandon *et al.*, 1996). In this model, when parasites migrate more than the hosts, and when host migration rates are not high, the parasites are locally adapted. However, when hosts migrate more than their parasites, and when parasite migration rates are low, the hosts –

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rather than the parasites – should be locally adapted. This result is to be expected, since migration by introducing new genes into a population can be associated with higher evolutionary potential. In the case of isolated populations, where there is no migration of either parasites or the hosts' populations, another mathematical model has shown that local adaptation depends on how well parasites are able to track host genotypes (Morand *et al*., 1996). Usually, parasites migrate more than their hosts because, typically, hosts migrate with numerous parasites either attached to the bodies or within them or because parasites can reach other host populations via intermediate hosts. In species with parental care, vertical parasite transmission is expected and yearling offspring will disperse with their natal parasites. Conversely, in species without parental care (such as lacertid lizards), no vertical parasite transmission is expected and yearlings dispersing soon after hatching should migrate with far fewer natal parasites. As a result, the model predicts these parasites should be locally maladapted. We tested this hypothesis with the Canarian endemic lizard, *Gallotia galloti,* and its blood parasite, a member of the haemogregarine genus. Physiological costs to parasitism by haemogregarines have been demonstrated in another lizard species *Lacerta vivipara* (Oppliger *et al*., 1996; Oppliger & Clobert, 1997). For example, infection with haemogregarines reduces haemoglobin concentration, lowers locomotor speed and reduces the rate of tail regeneration. The ecology of *G. galloti* clearly suggests that juvenile dispersal is high (Vernet and Castanet, personal communication). The prevalence of blood parasites in juvenile lizards is very low compared with adult prevalence (A.O., unpublished observations), meaning that most dispersing juveniles are parasite-free, and become infected only after settlement in new subpopulations. Conversely, the parasite vector (a mite), because of its size (<1 mm), is unable to migrate between host populations by itself and needs a host to migrate. We carried out cross-infection among three lizard populations; two populations belonged to the same island, where migration rate between populations was expected to be higher for hosts than for parasites populations; a third population was from an adjacent island, with no migration between islands expected. We compared the performance of a parasite population in different host populations, and we compared the performance of a parasite population in its native host with parasite populations transplanted from different host populations. Therefore, based on the model predictions, we expected that the performance of a parasite population in its native host would be lower than its performance in novel allopatric hosts and we expected that the performance of a parasite population in its native host would be lower that of the other allopatric transplanted parasites. Cross-infection between islands will be influenced by the host–parasite coevolution patterns on the two islands.

Methods

Parasite and lizard species

Gallotia galloti, is a medium-sized (<130 mm snout–vent length) endemic lizard found in the Canary Islands. On the island of Tenerife it is very abundant, shows spatial heterogeneity (Thorpe & Brown, 1989) and occurs throughout the island. Adults are herbivorous/fruit-eating, whilst yearlings are predominantly insectivorous (Castanet & Baez, 1988). In October 1997, we captured yearlings of undetermined sexes, as well as some adults from two subpopulations of Tenerife island (T1 and T2) and from one population of Gomera island (GO). The two islands are 30 km apart. One hundred and thirty-three individuals from the two populations of Tenerife were collected on the north coast at sea level. Populations were 20 km apart (105 individuals in Valle de Gerra = T1, and 28 individuals in Puerto de la Cruz = T2). Sixteen individuals from Gomera were also collected on the north coast at sea level (Aguro). All lizards were captured in pitfall traps, baited with fruit and tomatoes. At the time of capture, we toe-clipped each individual and collected a drop of blood to make a smear, which was subsequently fixed in methanol and stained with May-Grünwald Giemsa (Colorap de Bioréac, Lausanne, Switzerland). Stained slides were examined using oil-immersion microscopy (500×) and blood parasites were counted. Haemogregarine parasites are naturally widespread in *G. galloti*. This protozoan (phylum Sporozoa) has a complex life cycle which involves a microscopic blood-feeding mite vector (Manwell, 1977). Mites are not very mobile. Parasites are usually intraerythrocytic (Manwell, 1977), but free forms can be observed in severe infections (A.O., personal observation). The genus of Haemogregarine was determined by the morphology of gametocytes found in red cells. No morphological differences between gametocytes were observed between the different study sites and thus we assume that a single species is involved (Manwell, 1977). Parasitaemia was estimated by counting the number of parasites observed per 10 000 red blood cells. A slide was considered negative when, after 5 min (approximately 300 fields of 400 cells per field) of examination, no parasite was observed. At the beginning of the experiment, body mass and snout–vent length (SVL) were measured for each lizard. To prevent horizontal blood parasite transmission (Sorci *et al*., 1997), all mites (the vector of the blood parasite) were removed from the lizard's skin, by scrubbing with ether-soaked cotton. Fifty-six yearlings, free of parasites (30 from T1, 13 from T2 and 13 from GO) and six heavily parasitized adults (three from T1, one from T2 and two from GO) were maintained in large terraria (4–5 individuals from the same population per terraria) for the cross-infection experiment. Terraria were provided with sand, many shelters and water dishes. Lizards were fed *ad libitum* with different fruits and insect larvae (*Tenebrio*

molitor). A heat lamp provided warmth 6 h per day allowing lizards to thermoregulate. The other lizards were released at their capture point.

Cross-infection

Thirteen nonparasitized yearlings from each population were experimentally infected with blood parasites from T1. Parasite-free yearlings were infected in the following manner: we collected about 3 mL of parasitized blood by postorbital puncture of the three heavily parasitized adults from the population T1; this mixed blood was then diluted in 5 mL of physiological saline (NaCl 0.9%); each experimental lizard received 100 μL of this solution by intraperitoneal injection and 100 μL orally. Conversely, eight individuals from T1 were inoculated in the same way with parasites from T2 (blood collected from the heavily parasitized adult from T2) and eight other individuals from T1 were inoculated with parasites from GO (blood collected from the two heavily parasitized adults). Unfortunately, we did not infect GO and T2 individuals with their own parasites because of a lack of terraria.

After 50 days, we again measured body mass and collected a blood sample to estimate the number of infected red blood cells from each lizard. Four lizards died during the initial phase of the experiment (four yearlings from T1, three infected with parasites from GO and one with parasites from T2).

Statistical analysis

The effects of lizard origin or parasite origin on the intensity of parasitaemia following experimental infection were analysed in a nested analysis of variance (ANOVA) in which original treatment was included as a main effect, while the effect of cage was nested within the main effect. To control for the effect of body condition, we also included it as a covariate (GLM procedure, SYSTAT version 6.0 for Windows). Assumptions underlying the models were checked beforehand. Body condition was calculated using regressions of body weight vs. SVL, with residuals from this common regression used as an index of body condition. Parasitaemia data were log-transformed prior to analysis.

Results

Natural prevalence

Natural blood parasite prevalence was not significantly different in the three studied populations; 18% of individuals were parasitized in T1 (19/105), 17.8% in T2 (5/28) and 20% in GO (3/16) (χ^2: 1.809, $P=0.405$).

Intensity of parasite load (percentage of red blood cells parasitized) was also not significantly different among populations (Fig. 1. ANOVA using % of red cells parasitized as the independent factor and populations as factors: $F_{2,24}=0.076$, $P=0.927$).

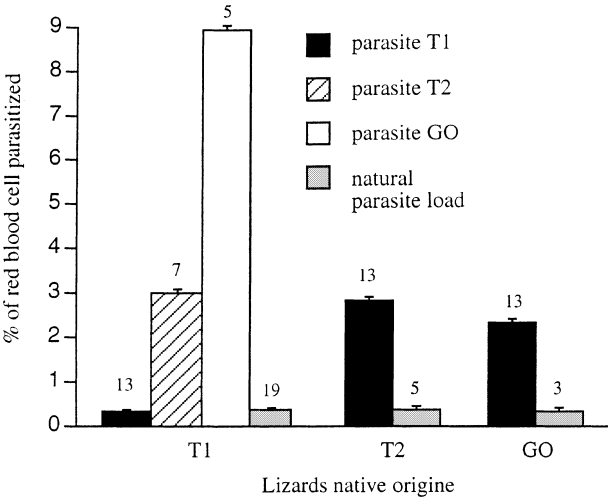

Fig. 1 Mean (+SE) percentage of parasitized red blood cells observed 50 days after experimental infection of captive *Gallotia galloti* with haemogregarines collected from different populations, with regard to different lizard origin (T1 = Tenerife 1, T2 = Tenerife 2, GO = Gomera). Black bars represent parasites from T1, hatched bar represents parasites from T2 and white bar represents parasites from GO. Shaded bars represent the natural parasite load of each population. Number above the bars indicates the sample size.

Parasite performance with regard to lizard origin and parasite origin

Fifty days after experimental infection with parasites from T1, individuals from T2 and GO had many more blood parasites than individuals from T1 (Fig. 1). This difference in infection rate (% of parasitized red cells) with regard to lizard origin was significant (Table 1). An *a posteriori* comparison showed that lizards from GO and T2 had significantly more infected red blood cells than lizards from T1, but there was no significant difference in the percentage of parasitized cells between lizards from GO and from T2 (*post hoc* Fisher's LSD test; GO/T1: $P < 0.02$; T2/T1: $P=0.010$; GO/T2: $P=0.778$). We found no difference in the number of successful infections among the three populations (T1: 77% (10/13); T2: 84% (11/13); GO: 91% (11/12), χ^2: 1.023, $P=0.6$).

Similarly, individuals from T1 had more blood parasites when they were infected with parasites from GO or from T2 than when they were infected with their own parasites (Fig. 1). This difference in infection success (% of parasitized red cells) with regard to parasite origin was significant (Table 2). An *a posteriori* comparison showed that parasites from GO and T2 were better at infecting red blood cells of T1 lizards than parasites from T1. Moreover, parasites from GO infected significantly more

Table 1 ANCOVA: susceptibility of lizards from different origins to haemogregarines coming from Tenerife North. Cages nested within lizard origin and lizard origin were used as factors and body condition as covariate.

Source of variation	d.f.	Mean sum of square	*F*-ratio	*P*
Lizard origin	2	8.444	4.592	0.019
Body condition	1	0.003	0.002	NS
Cage (Lizard origin)	6	1.734	0.943	NS
Error	29	1.839		

Table 2 ANCOVA: effect of parasite origin on the log-transformed percentage of parasitized red cells of *Gallotia galloti* coming from Tenerife North. Cages nested within parasite origin and parasite origin were used as factors and body condition as covariate.

Source of variation	d.f.	Mean sum of square	*F*-ratio	*P*
Parasite origin	2	17.561	24.196	< 0.001
Body condition	1	0.670	0.923	NS
Cage (parasite origin)	4	0.345	0.475	NS
Error	16	0.726		

red cells than parasites from T2 (*post hoc* Fisher's LSD test; GO/T1: $P < 0.001$; T2/T1: $P = 0.001$; GO/T2: $P = 0.038$). We found no difference in the number of successful infections among the three parasite strains (T1: 77% (10/13); T2: 100% (8/8); GO: 100% (5/5), χ^2: 3.191, $P = 0.183$).

Discussion

Our results clearly indicate that haemogregarine blood parasites of *G. galloti* lizards were more readily able to infect lizards coming from distant populations than lizards belonging to a sympatric population. Moreover, when a lizard population was infected with different parasite strains, parasites from distant host populations performed 'better' than local ones. Despite having only one reciprocal control (infection of one population with its own parasite), our results suggest that this outcome is likely to occur in this host–parasite system. Indeed, in this study, prevalence of sympatric parasites and sympatric parasite load per infected lizard did not vary amongst the three studied populations. This suggests that the blood parasite populations have a similar level of virulence with their sympatric host, or/and that lizard populations have a similar level of resistance against their sympatric parasites. Parasite local maladaptation has rarely been documented. Imhoof & Schmid-Hempel (1998) have shown that the trypanosome intestinal parasite *Crithidia bombi* infecting the bumblebee, *Bombus terrestris*, is locally maladapted when cross-infections were carried out over a large geographical range. Kaltz *et al*. (1999) have demonstrated that the fungal pathogen *Microbotryum violaceum* has lower migration rates than its host, *Silene latifolia*, and is locally maladapted for infectiveness.

Migration rates of parasites and hosts on Tenerife island were not investigated, but the ecology of these two species clearly suggests that lizard juvenile dispersal is high (Vernet and Castanet, personal communication) and that parasite vector motility is very low (A. Oppliger, personal observation). Therefore, our results support the prediction of Gandon *et al*. (1996) leading to the situation of host local adaptation. This agreement does not imply that the difference in migration rate between host and parasite is the only factor responsible for this pattern of local adaptation. Other components of the evolutionary potential of parasite and host may act synergistically to produce the same pattern (Michalakis & Gandon, in press). For example, mutation rate, population size and generation time all influence the evolutionary potential of an organism.

The results of the interisland cross-infection experiments are more difficult to interpret. In this case, migration between islands is neither expected for hosts nor for parasites. However, from Thorpe *et al*. (1993, 1994) and the distribution of *G. galloti* on the western Canary Islands, it appears that their colonization is essentially the result of dispersal among islands. Thorpe *et al*. (1994) used several DNA markers and showed that lizards spread from Tenerife in the west to Gomera. Nowadays, if juvenile dispersal occurs between these two islands, it is probably very rare (perhaps by boats, which daily visit both islands). In this case, the scale of migration could be much smaller than the within-island system, but the ratio should still be in favour of the host and thus the results should still be consistent with the prediction of Gandon *et al*. (1996). Otherwise, predictions about local adaptation depend on the coevolutionary process between host and parasite genotypes. For example, in Schistosome–snail systems, local adaptation by the host or by the parasite are both likely to occur (Michelson & DuBois, 1978; Vera *et al*., 1990; Manning *et al*., 1995; Morand *et al*., 1996). In this latter paper (Morand *et al*., 1996), sympatric parasite–host combinations tend to be more compatible (compatibility reflecting both snail susceptibility and schistosome infectiveness); there are exceptions where particular allopatric parasite–host populations are significantly more compatible. In order to explore the variations in geographical patterns of host–parasite compatibility, a mathematical model of the dynamics of host–parasite system has been developed. This model (Morand *et al*., 1996) predicts a dynamic polymorphism where parasite allele frequencies track host allele frequencies but with a lag. Because of this lag it is possible for allopatric combinations to be more compatible than sympatric combination. Such a pattern could occur in our study. However, reciprocal cross-infection among several island populations should be

carried out and genotypic variation in both host and parasite populations should be investigated.

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