

Original Article

# Spatial variation in food supply, mating behavior, and sexually transmitted disease epidemics

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Mating behavior helps to shape the epidemiology of sexually transmitted infections (STIs) by determining the rate at which infectious and susceptible individuals make contact. To the extent that mating behavior shows spatial variation, it is expected that STI epidemics will also show spatial heterogeneity (provided that dispersal is limited). However, empirical data are lacking for natural systems. Here, therefore, we investigate the association between local mating behavior and STI epidemiology in natural populations of the two-spot ladybird, *Adalia bipunctata*, and dissect the causes of this variation. Variation in mating behavior was observed over local and regional spatial scales and high mating rate led to stronger local epidemics of the STI. Initial ecological observations suggested that reduced mating rate was associated largely with low food supply (and possibly to a lesser extent with local population density) and not with differences in thermal environment. We tested the potential role of food supply in fuelling STI epidemics in laboratory populations and confirmed that high food provision led to greater STI transmission. We conclude that food supply is strongly associated with mating rate in the field and that this produces heterogeneity in STI epidemiology. A consequence is that STI-induced sterility will reduce reproduction in habitat patches of high food quality. *Key words*: disease, environment, nutrition, pathogen, sexual transmission, spatial heterogeneity. [*Behav Ecol*]

## INTRODUCTION

The importance of parasites for driving various aspects of animal reproductive behavior has been widely discussed. Individual mating decisions may be biased against mates infected with parasites due to the fitness costs of infection (Johnstone 1995). Mating systems may also evolve in response to the presence of sexually transmitted infections (STIs), promoting the coexistence of promiscuity and monogamy within populations (Boots and Knell 2002; Kokko et al. 2002). Parasites may even drive the evolution of sexual reproduction itself (Jaenike 1978; Hamilton 1980; Hamilton et al. 1990). Conversely, reproductive behavior may affect parasitism. Such effects are perhaps most obvious for STIs, where, for instance, variation in host mating behavior will influence both the rate at which infectious and susceptible individuals make contact (Antonovics et al. 1995) and equilibrium disease levels (Thrall et al. 2000).

Study of the influence of host mating behavior on STI dynamics has focused largely on humans. There, the importance of between-individual variation in mating rate in determining the spread of infection is well understood, with  $R_0$  for an STI being driven by the most promiscuous individuals

(Yorke et al. 1978; Hyman et al. 1999). Variation in mating rate can also occur spatially, and if mating rate is high compared with rates of movement, this heterogeneity may be reflected in spatially varying disease epidemics, with areas of high mating rate being hotspots for STI spread. However, although theoretical work has considered the impact of spatial heterogeneity on STI epidemiology in humans (Anderson and May 1991; Auvert et al. 2000; Castillo-Chavez and Li 2008), empirical data on natural systems are lacking.

In this study, we examined the pattern and causes of spatial heterogeneity in STI epidemics in an empirical model invertebrate system. Central and eastern European populations of the two-spot ladybird, *Adalia bipunctata* L., are infected with a virulent sterilizing ectoparasitic mite *Coccipolipus hippodamiae* whose larvae transmit between partners during host copulation (Hurst et al. 1995). Adult *A. bipunctata* emerge from overwintering with moderate infection prevalence, but sexual activity during the spring initiates epidemic spread of infection. The epidemics are frequently severe enough that nearly all of the surviving members of the overwintered cohort become infected (Webberley et al. 2004; Webberley, Buszko, et al. 2006; Webberley, Tinsley, et al. 2006). Sexual transmission is sufficient to explain the epidemiology in the field, with sensitivity analyses of model simulations showing that mating rate is the key determinant of transmission (Webberley, Buszko, et al. 2006). Thus, this disease represents a canonical STI. Surprisingly, STI presence does not affect the mating

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success of individual hosts (Webberley et al. 2002), despite strong sterility and mortality effects of infection (Hurst et al. 1995; Ryder et al. 2007).

Laboratory study has revealed a variety of ecological factors that shape mating rate in *A. bipunctata*. In particular, the likelihood of mating is in part mediated by contact rate, which is affected by both thermal environment and density (Brakefield 1984; Ryder et al. 2005). Willingness to mate on contact is additionally affected by food supply, with poorly fed females rejecting male suitors (Perry et al. 2009). If such parameters vary spatially in natural populations, then local variation in mating rate and STI epidemiology would be expected to follow, provided that dispersal is limited.

We assessed the degree of heterogeneity in mating rate in *A. bipunctata* at local and regional spatial scales in 2 European cities where *C. hippodamiae* undergoes annual epidemics. We then reconciled observations of local mating rate to STI epidemiology and examined whether thermal environment, density, and/or food supply were associated with elevated mating rate. We observed striking heterogeneity in mating behavior at both local (500 m) and regional (12 km) scales. In both cases, mating rate heterogeneity fed through to STI dynamics and in both cases our initial field observations indicated an underlying association with food supply, with higher mating rates occurring in sites where food was abundant. Laboratory microcosm study confirmed the potential for food to impact on STI epidemiology. Our results provide a mechanistic flow from spatial variation in food, which generates spatial variation in mating behavior, and finally feeds into disease epidemiology.

## MATERIALS AND METHODS

### Field sites

Between 1 May ("day 1") and 28 July 2009 ("day 89"), we monitored both mating rate and *C. hippodamiae* infection prevalence on *A. bipunctata* in Toruń, Poland. We focused on a suburban location (hereafter "Toruń suburb") in which stands of mature unmanaged lime trees (*Tilia* spp.) and rose bushes (*Rosa* spp.) were interspersed within the same 0.5 km<sup>2</sup> patch of land and represented the 2 main *A. bipunctata* habitats present (grid reference 53°1'6.24"N, 18°34'11.56"E). A total of 9 lime tree stands and 4 stands of rose bushes were sampled, comprising all those containing ladybird beetles in the site. To extend the spatial scale over which we could monitor epidemic spread, we also obtained regular prevalence samples during the same period for a second site in Toruń, a district about 1 km from the suburban site and adjacent to the town center (the "Toruń town center" site). This site was dominated by managed lime trees on pavements (also approximately 0.5 km<sup>2</sup>; grid reference 53°0'43.28"N, 18°34'49.32"E).

The data we collected in 2009 indicated that epidemic heterogeneity could occur over small spatial scales in *A. bipunctata*, even within the same habitat type (e.g., suburban vs. town center lime trees). The following year, therefore, between May and August 2010, we focused on the lime tree habitat exclusively to compare suburban and town center dynamics in more detail. This second field season was based in Stockholm, Sweden. It was necessary to move the study from Toruń because, along with many other European cities, Toruń subsequently lost much of its *A. bipunctata* population after the invasion of *Harmonia axyridis* Pallas.

In Stockholm, in 2010, our suburban site was formed from two 0.5-km linear sampling areas, 1 in each of 2 adjacent suburbs to the north of the city center—Kista (grid reference 59°24'14.17"N, 17°56'32.38"E) and Akalla (grid

reference 59°24'52.48"N, 17°54'55.31"E) (hereafter collectively referred to as "Stockholm suburb 2010"). The city center site (approximately 12 km distant) was again comprised of two 0.5 km linear sampling areas, 1 in each of 2 adjacent parts of the city center—Strandvägen, Östermalm (grid reference 59°19'53.76"N, 18°5'3.78"E) and Ringvagen, Södermalm (grid reference 59°18'31.01"N, 18°4'52.61"E) (hereafter collectively referred to as "Stockholm city center"). Sampling began on 17 May 2010 and ended on 15 August 2010.

### Monitoring infection prevalence and mating rate

For both Toruń and Stockholm, our analysis of mite epidemiology concentrates on the overwintered cohort, in which the epidemic can be observed readily from one year to the next. All beetles collected until the end of June are of the overwintered cohort. Thereafter, new and overwintered beetles can be differentiated by elytral color, new cohort individuals being orange red in contrast to the deep red of overwintered individuals.

To observe changes in infection prevalence and mating rate, we carried out weekly population sampling across the spring–summer season, when *A. bipunctata* emerge from their overwintering period and begin mating, initiating the epidemic. Beetles were collected from lime trees using a beating tray. We concentrated on the lower branches, rather using a ladder to access the higher branches, as this allowed us to cover a greater area of the habitat per unit time. Beating trays cannot be used readily on rose bushes, but it is easy to collect beetles by eye from this habitat. In each case, freshly collected beetles were placed individually into Eppendorf tubes and returned within the hour to the laboratory. Beetles found in copula at the point of collection were transferred to a single Eppendorf without separation. Pairs collected in this way continued to copulate, so we were able to calculate the fraction of individuals that were copulating in each sample. The infection status of all beetles was scored under a dissecting microscope in the laboratory using a methodology we have described previously (Hurst et al. 1995). Beetles were also scored as to cohort (overwintered vs. emerged in current season) using elytral color depth following Webberley, Buszko, et al. (2006). After scoring, beetles were returned to the same stand of trees or bushes from which they were collected, usually on the same day (occasionally the following morning if sampling continued into the evening, in which case they were maintained overnight at 4 °C).

In Toruń, we collected approximately 100 beetles weekly for the town center site and for each of the 2 habitat types within the suburban site (= approximately 300 beetles sampled per week). In both cases, stands of suitable habitat that had been identified in advance were sampled systematically, with subsamples of between 5 and 25 beetles being collected per stand, to ensure that the whole sampling area was covered each week. In Stockholm in 2010, population samples were collected using the same subsampling approach we adopted in Toruń, again minimizing bias toward any one part of the site. A minimum sample of 75 beetles was recovered from each of the 2 paired suburb sites and from each of the 2 paired city center sites (i.e., minimum weekly  $n = 150$  suburb, 150 city center).

### Supplementary observations of mating rate and its ecological determinants

We supplemented pair data gathered during the sampling above with in situ behavioral observations within our habitats. In Toruń, we focused our observations on a subsample of 6 of the 13 suburban lime tree/rose bush stands from which

beetles were sampled for prevalence estimation. To facilitate the comparison of lime tree and rose bush habitats, we restricted our comparison to the suburb site (i.e., comparing lime trees and rose bushes within the site). In Stockholm, we selected 1 of the suburb sites (Akalla) and 1 of the city center sites (Södermalm) for our observations. Mating propensity may vary through the day, so we undertook 3 daily observation sessions, at 9 AM, 1.30 PM, and 6 PM, each of exactly 1-h duration. There were 3 observation days every week in Toruń and either 3 or 4 per week in Stockholm. Tree (or bush) foliage was searched exhaustively by hand during each session, with observers working through the same length of habitat in a consistent manner from one session to the next. When beetles were found, we recorded whether they were single or in a mating pair. We used 2 observers to extend the area over which we could make observations. To exclude potential observer bias, both observers worked simultaneously on each site during every observation session.

We derived an approximate index of local population density from the number of beetles recovered per standardized observation session. In addition, we examined the association between mating and thermal ecology, in terms of ambient and body temperatures. Ambient temperatures were recorded continuously using data loggers positioned in the tree or bush foliage, and an infrared thermometer was used to measure the body temperatures of beetles in situ. As this analysis indicated that thermal environment (and body temperatures) varied very little between study sites during the course of the epidemics, either in Poland or in Sweden, we give details of relevant methodology and results in [Supplementary material](#). Aphid abundance was observed anecdotally in both Poland and Sweden during in situ observations and the initiation of ladybird oviposition was noted as a further indicator of food availability. *A. bipunctata* females will oviposit without repeated mating, even beyond the point where sperm supplies are exhausted ([de Jong et al. 1993](#)). Given a substantial fraction of the population will have mated at least once in all populations (as testified by the presence of the STI in all places), the presence of *A. bipunctata* eggs within a habitat is a useful supplementary indicator that there is sufficient food available to sustain reproduction (note the STI affects egg fertility not egg production). These initial field observations were then supplemented with a laboratory microcosm experiment designed to test the hypothesis that food availability could have been a key factor associated with mating rate throughout our field sites (see following section).

#### Laboratory investigation of food–STI epidemiology link

To examine whether diet affects the dynamics of the STI on *A. bipunctata*, 12 replicate microcosms were established, 6 under conditions of low food and 6 under conditions of high food, and the transmission of the mite was monitored for each.

To this end, *A. bipunctata* were collected in Stockholm (Sweden) between 22 and 25 May 2012 and then scored for sex and mite presence. The microcosms were constituted from these field collected beetles, each comprising 1 infectious male, 1 infectious female, 5 uninfected females, and 5 uninfected males, and maintained in incubators at 20 °C and a 20:4 light:dark regime. Infectious individuals harbored a minimum of 5 infective mite larvae, but were not “heavily” infected (which can be judged from the presence or absence of mite larvae from the outer surface of the elytra). Infectious and uninfected beetles were maintained separately for 7 days and acclimated to their food regime. Ladybirds on the “high” food regime were given 5–6 large *A. pisum* aphids per female or 2 per male and ad libitum access to an artificial food

source containing water, sugar, and yeast. This level of aphid provisioning corresponds to a normal level of consumption for adult *A. bipunctata* based on laboratory observations under the same conditions. Ladybirds on “low” food were maintained on the artificial diet alone, which maintains normal longevity but is insufficient to support oogenesis.

After 7 days, infectious and uninfected beetles were mixed in a population cage (20 cm diameter, 30 cm high) and fed daily according to their food treatment. The following measures were then made over the following 15 days—1) *mating rate*: the number of copulating pairs of ladybirds was recorded at the start of each day, to provide an index of sexual activity; 2) *oviposition activity*: presence of eggs was noted daily, to provide an index of female reproductive capacity; and 3) *mite infection*: all beetles were removed and scored for mite infection on days 4, 7, 10, 14, and 15 (daily scoring was avoided to minimize the impact of repeated scoring on behavior and transmission). A small reserve population of beetles was maintained under the appropriate food regime, and these were used to replace any individuals that died during the experiment. Dead individuals were scored for infection status and replaced with individuals of identical infection status.

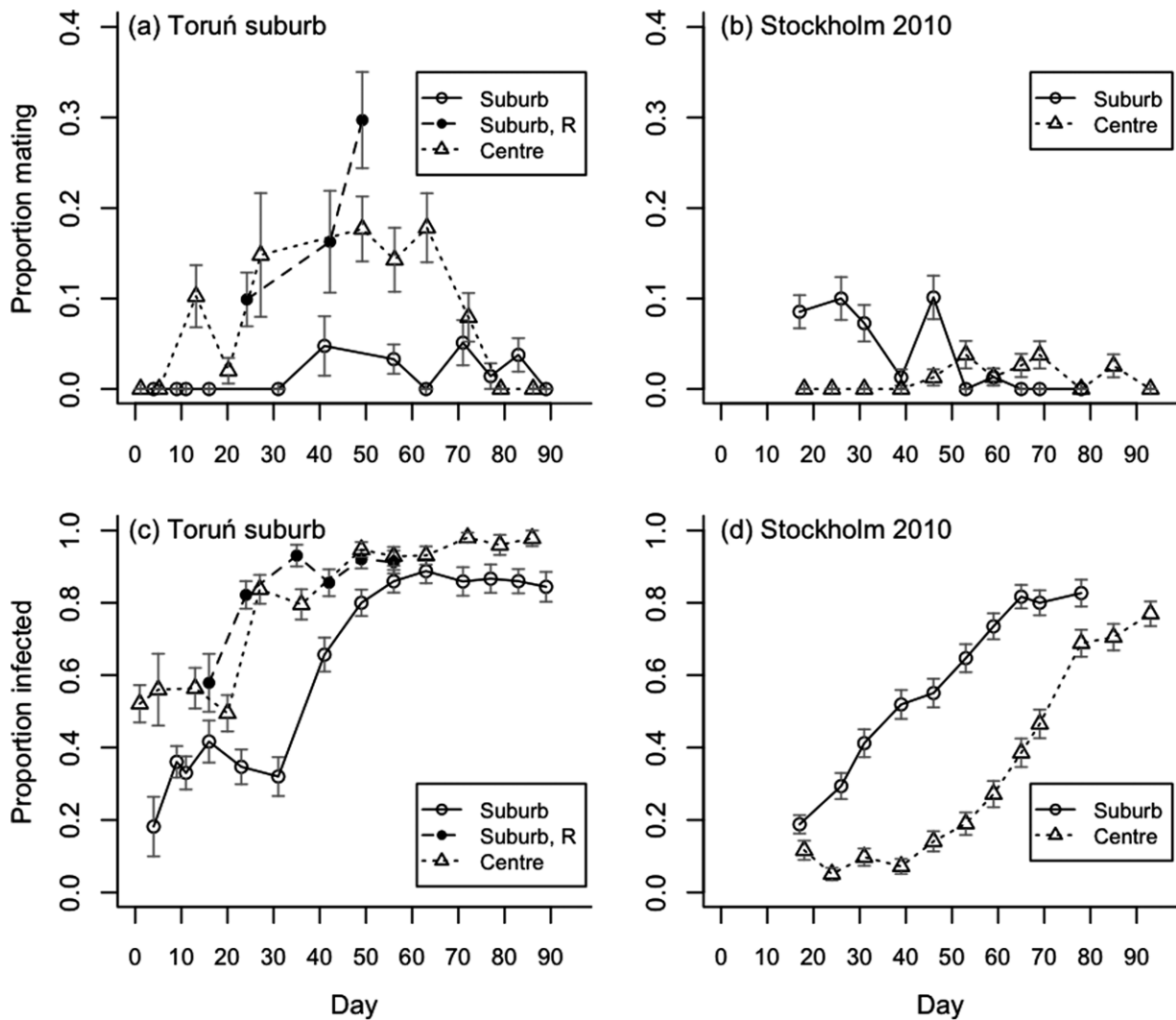
## RESULTS

### Mating behavior and STI epidemiology in Toruń

Mating rates derived from the weekly collections revealed differences in the onset and frequency of mating between habitats. On the lime trees in the suburban site, we did not observe any copulating pairs until day 41. In contrast, we recovered mating pairs as early as day 24 in our samples on the adjacent roses ([Figure 1a](#)). The proportion of ladybirds sampled in copula across the season on the roses was also significantly higher than on the adjacent trees (mean proportion in copula: trees = 0.013, roses = 0.183; generalized linear model with quasibinomial errors, residual deviance = 38.4, df = 14,  $P < 0.0001$ ). This finding was supported by the equivalent data obtained from our in situ observation sessions, with the proportion of ladybirds observed in copula being significantly higher on the roses than the adjacent lime trees (generalized linear model with quasibinomial errors, residual deviance = 122.1, df = 64,  $P < 0.0001$ ).

We also recovered mating pairs earlier from the town center lime trees compared with the suburban limes (day 13 on the town center limes vs. day 41 at the suburban site; [Figure 1a](#)) and the proportion of ladybirds found in copula across the season was significantly higher on the town center limes (mean proportion in copula: suburb limes = 0.013, town center limes = 0.078; generalized linear model with quasibinomial errors, residual deviance = 114.9, df = 21,  $P < 0.01$ ).

Our in situ observation data indicated that the suburban rose bushes had considerably higher beetle density than the adjacent limes (we infer this from differences in the mean number of ladybirds observed per hour of hand searching [rose bushes vs. lime trees, morning observations:  $58.3 \pm 38.0$  vs.  $15.2 \pm 3.14$ ; afternoon:  $55.7 \pm 31.95$  vs.  $15.1 \pm 3.72$ ; evening:  $46.7 \pm 21.17$  vs.  $11.5 \pm 3.53$ ; means are shown  $\pm$  95% confidence intervals]). In addition, it was notable that although aphids were abundant on the suburban roses, they were rarely seen on the suburban lime trees. Similarly, although oviposition and other indicators of recruitment (larval development, appearance of new adults) were commonly observed on the suburban roses, we only detected minute levels on the suburban lime trees in nearly 3 months of exhaustive searching. Thus, density, food supply, and recruitment (a useful indicator that adequate food is available for reproduction) were all higher on the suburban roses than on



**Figure 1**

Mating and infection prevalence data for Toruń (Poland) in 2009 and Stockholm (Sweden) in 2010: (a) proportion of *Adalia bipunctata* found in copula in the prevalence samples from the rose bush (R) and lime tree habitats in the Toruń suburb and town center sites; (b) proportion of *A. bipunctata* found in copula in the prevalence samples in the Stockholm suburb and city center sites (2010); (c) proportion of beetles infected with *Coccipolipus hippodamiae* in the Toruń suburb and town center sites; (d) proportion of beetles infected with *C. hippodamiae* in the Stockholm suburb and city center sites (2010). See insets for keys; error bars show  $\pm 1$  standard error.

the adjacent lime trees. The town center lime trees showed a similar contrast with the suburban lime trees, showing both an early, abundant aphid supply and early recruitment.

Our mating rate data were reflected in separate epidemics of the STI. In Toruń, we observed discrete epidemics over 2 spatial scales. In the suburban site, we observed a relatively late epidemic on lime trees (where aphids were rare) compared with the adjacent roses (where they were abundant), despite the close physical proximity of the 2 habitats (Figure 1c). On the suburban trees, prevalence was relatively stable between day 1 (1 May) and day 31 at around 30% of individuals infected. After then, we recorded a steep increase to a plateau of approximately 85% by around day 56. In contrast, prevalence on the rose bushes was already 58% and rising by day 16 and had reached a plateau of approximately 90% as early as day 35.

A similar contrast was observed between the trees in the suburban site and the trees in our town center site. In the town center (where aphids were again abundant, as on the suburban rose bushes), we again observed a relatively stable

phase before the epidemic started, as we did in the suburb (Figure 1c). However, prevalence remained at around 50% during this phase in the center, compared with approximately 30% in the suburb. The epidemic increase occurred earlier in the town center, remaining stable only until around day 20 and increasing to a plateau of about 95% by day 49. In both cases, the onset of the epidemic occurred about 1 week after the onset of mating in these locations.

#### Mating behavior and STI epidemiology in Stockholm

In Stockholm in 2010, we observed temporally distinct patterns of mating between city center and suburban sites (both lime trees only). We did not recover pairs in copula in the city center site until around day 30, whereas the very first samples (i.e., day 1) at the suburban sites yielded mating pairs (Figure 1b). The proportion of ladybirds sampled in copula across the season was also significantly higher in the suburban site compared with the city center (mean proportion in copula: suburb = 0.041, city center = 0.013; generalized linear

model with quasibinomial errors, residual deviance = 135.2,  $df = 26$ ,  $P < 0.05$ ).

The lack of mating in the city center seems to have occurred despite the fact that density was higher in these sites than in the suburban lime tree sites (inferred from the mean number of ladybirds observed per hour of hand searching in our in situ observation sessions [suburban vs. city center sites, morning observations:  $18.1 \pm 4.5$  vs.  $32.7 \pm 5.89$ ; afternoon:  $18.6 \pm 4.1$  vs.  $37.2 \pm 9.0$ ; evening:  $16.5 \pm 5.0$  vs.  $28.5 \pm 6.3$ ; values are shown  $\pm 95\%$  confidence intervals]). However, although density was higher in the city center, the lower mating rates recorded there were associated with a striking absence of aphids relative to the suburb, where they were more common. Evidence of oviposition was also lacking in the city center and there was effectively no recruitment of a new generation (egg clutches, new larvae, and pupae are conspicuous during in situ observation sessions when they are present). In the suburb, by contrast, new egg clutches were readily observed, as was the appearance of larvae, pupae, and new adults.

The observed variation in mating rate was again reflected in variation in STI epidemics between our suburban and city center sites. In the suburban sites, prevalence had already begun to increase by the time sampling started, rising from about 20% to a plateau at just over 80% by day 50 (Figure 1d). In contrast, we did not detect an increase in prevalence in the city center sites until around day 30, with the proportion infected only reaching a plateau, at around 70% prevalence, by day 62. At this point, the overwintered cohort was in its last stages, and we can thus conclude that the late onset of mating, and the lower mating rate, allowed some individuals to escape infection entirely in this location.

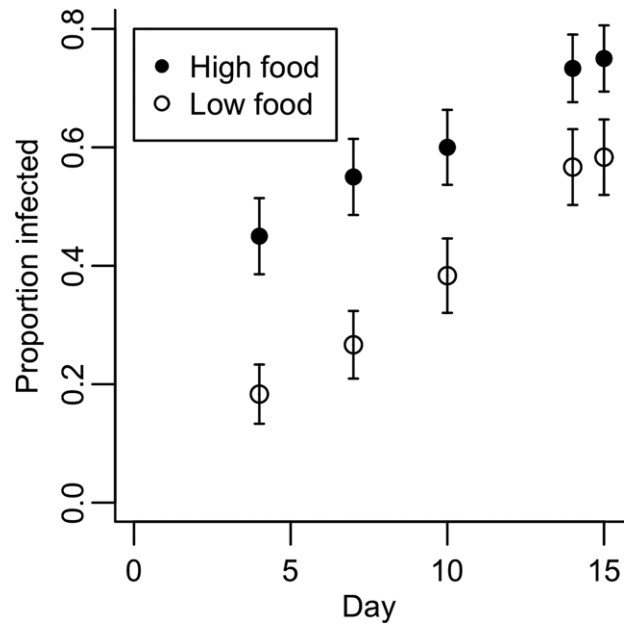
#### Laboratory microcosms: food supply and STI transmission

The low food treatment was associated with significant reductions in the amount of copulation recorded (mean number of copulating pairs observed each day, high food:  $1.05 \pm 0.26$ ; low food:  $0.14 \pm 0.13$ ;  $t$ -test with unequal variances:  $t = 6.21$ ,  $df = 7.22$ ,  $P = 0.0004$ ) and in the amount of oviposition in the microcosms (mean number of eggs per day, high food:  $21.3 \pm 6.4$ ; low food:  $1.2 \pm 1.6$ ;  $t$ -test with unequal variances:  $t = 5.95$ ,  $df = 5.64$ ,  $P = 0.0013$ ).

The impact of reduced food on mating and oviposition was correlated with a clear effect on the amount of *C. hippodamiae* transmission. In both the high and the low food treatments, the proportion of beetles newly infected with *C. hippodamiae* rose steadily following the introduction of infectious beetles (Figure 2). However, there was a significant and rapid divergence between the 2 treatments, with the high food microcosms showing a faster initial increase and greater overall prevalence (generalized additive model with binomial errors, likelihood ratio test: deviance =  $-31.72$ ,  $df = -1$ ,  $P < 0.0001$ ).

#### DISCUSSION

Theoretical work has considered the impact of spatial heterogeneity in mating behavior on STI epidemiology (Auvert et al. 2000; Castillo-Chavez and Li 2008), but empirical data on natural systems have been lacking. In this study, we documented spatial heterogeneity in mating rate in *A. bipunctata* over a local (50–500 m) and regional (12 km) scale that led to the emergence of distinct epidemics of the STI across habitat patches in the first case and across the regional landscape in the second case. These results make it clear that spatial variation in mating biology is an important consideration in understanding STI epidemiology in natural animal populations. Perhaps the most surprising observation



**Figure 2**  
Proportion of *Adalia bipunctata* newly infected with *Coccipolipus hippodamiae* in the laboratory microcosm experiment under the high and low food treatments. See insets for keys; error bars show  $\pm 1$  standard error.

is that very different *A. bipunctata* mating rates can be observed on adjacent habitat patches, leading to distinct STI epidemics, despite the potential homogenizing force of dispersal between the patches. Thus, understanding of STI epidemiology requires analysis of mating system variation over both local and regional scales.

In most natural systems, the rate at which infection spreads during disease epidemics probably varies spatially (Hochberg and Lawton 1990; Grenfell and Bolker 1998) and previous studies have recorded heterogeneity in infection dynamics over small spatial scales in invertebrates. Trematode infections in intertidal bivalves, for instance, can show marked heterogeneity over small distances due to variation in habitat properties within sampling sites (Thielges and Reise 2007). Spatial variation in spirochete infections in ticks (*Borrelia burgdorferi*, which causes Lyme disease) is also associated with habitat factors (Van Buskirk and Ostfeld 1998); and within-habitat clustering of parasite genotypes has even been recorded for leaf miners infecting particular oak trees, due to the fidelity of leaf miners to individual trees that may vary in age and phenology (Mopper et al. 2000). However, spatial heterogeneity in epidemic dynamics is not commonly reported for natural animal populations, and not at all (to the best of our knowledge) for animal STIs. That it should occur over such fine spatial scales is surprising because male and female *A. bipunctata* are highly mobile and could easily migrate between contiguous habitats (e.g., dispersal of the adults away from patches of habitat that have become saturated with *A. bipunctata* eggs and larvae as reproduction proceeds in the spring regularly occurs en masse later in the season). Mixing of dispersing adults between the 2 habitats might therefore have been expected to cause infection prevalence to “even out” over such small distances. That such mixing did not occur may indicate that the overwintered cohort tend to wait for their food supply to develop in situ once they have emerged from nearby overwintering substrates and moved into a particular habitat (i.e., rose bushes or lime trees, in our study). Indeed, some works suggest that although *A. bipunctata* dispersal may occur within

a patch to areas of high aphid density, there may be little discrimination between patches on the basis of aphid abundance (Schellhorn and Andow 2005).

It is also interesting to note that theoretical studies point toward the existence of risky and safe mating strategies in populations affected by STIs (Boots and Knell 2002; Kokko et al. 2002). In one sense, it could be argued that we see a similar pattern in the *A. bipunctata*–*C. hippodamiae* system, with some individuals opting for a more promiscuous strategy that carries greater risk of being infected with the STI but greater rewards in terms of genetic and direct benefits. Whether these theoretical expectations are borne out in our study is unclear, because the fitness costs of infection are severe in habitat patches that sustain higher mating rates (i.e., greater promiscuity) and are incurred by most individuals in the patch. Individuals playing the “safe” strategy would also have the additional cost in our system of perhaps not reproducing at all (as well as missing out on genetic and direct benefits).

Our study indicates that ladybird nutrition is the most likely cause of mating rate variation and spatial heterogeneity in STI epidemiology. Thermal environment can be discounted as a key cause of local heterogeneity in our study, with any differences in either beetle or environmental temperature at this spatial range being small (see [Supplementary material](#)). This is not to say that thermal environment does not affect mating propensity, but that variation in thermal environment was in our study trivial at these spatial scales. Density may explain part of the variation in mating rate, because high density and high mating rate were associated in Poland (although meaningful comparison of density on different habitat types proved difficult), but measures of density in Stockholm (where all observations were on lime trees) indicate that mating rate was low in areas where density was notably higher. Thus, although we know from laboratory study that density can affect mating rate (as may have been the case in Toruń; [Ryder et al. 2005](#)), it is unlikely to have been the principle cause of variation in our study. In contrast, judging from our initial observations of aphid abundance in the field, food supply was always positively related to mating rate. Higher aphid abundance was noted in all sites with elevated mating rates. The presence of an adequate food supply can also be inferred from oviposition activity (only well-fed ladybirds oviposit) and other indicators of reproduction and recruitment (new larvae, pupae, and adults), all of which were higher in sites with increased mating rates. Our laboratory experiment lend support to this inference, with microcosm populations maintained under high food supply showing elevated mating rates and greater transmission.

We note that, ecologically, our results suggest that populations with high food, which are most able to oviposit, will also be the most affected by the sterilizing pathogen. Overall, therefore, we predict that the observed effect of nutrition on STI epidemiology will reduce *A. bipunctata* recruitment. Although the mite does not affect ladybird copulation rate, it does render female hosts infertile within 10–14 days ([Hurst et al. 1995](#)). Infected ladybirds continue to oviposit, but lay eggs that shrivel and fail to hatch. The association of high food areas with profound STI epidemics in this system will therefore have the effect of reducing female fertility in the very habitat patches that are most suitable for reproduction. The STI will thus reduce the extent to which high food patches as a source of recruitment.

Nutrition and condition have been found to be important in determining disease epidemiology in a number of other circumstances. For instance, mast years for acorns are associated with a rise in rodent density and these high density populations have higher disease prevalence ([Pedersen and Greives 2008](#)). Increased prevalence probably results from

combined effects on individual condition and transmission opportunities. In our study, the influence of nutrition is mediated by behavior, with low food supply decreasing the likelihood of mating. In ladybirds, this may occur mainly through modulation of female refractoriness to mating, rather than changes in activity and encounter rate. Our work indicates that studies focusing on the interactions between nutrition, behavior, and disease transmission would be timely.

## SUPPLEMENTARY MATERIAL

Supplementary material can be found at <http://www.behco.oxfordjournals.org/>

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## REFERENCES

- Anderson RM, May RM. 1991. Infectious diseases of humans: dynamics and control. New York: Oxford University Press.
- Antonovics J, Iwasa Y, Hassell MP. 1995. A generalized-model of parasitoid, venereal, and vector-based transmission processes. *Am Nat*. 145:661–675.
- Auvert B, Buonamico G, Lagarde E, Williams B. 2000. Sexual behavior, heterosexual transmission, and the spread of HIV in sub-Saharan Africa: a simulation study. *Comput Biomed Res*. 33:84–96.
- Boots M, Knell RJ. 2002. The evolution of risky behaviour in the presence of a sexually transmitted disease. *Proc R Soc Lond B Biol Sci*. 269:585–589.
- Brakefield PM. 1984. Ecological studies on the polymorphic ladybird *Adalia bipunctata* in the Netherlands. 2. Population dynamics, differential timing of reproduction and thermal melanism. *J Anim Ecol*. 53:775–790.
- Castillo-Chavez C, Li BT. 2008. Spatial spread of sexually transmitted diseases within susceptible populations at demographic steady state. *Math Biosci Eng*. 5:713–727.
- Grenfell BT, Bolker BM. 1998. Cities and villages: infection hierarchies in a measles metapopulation. *Ecol Lett*. 1:63–70.
- Hamilton WD. 1980. Sex versus non-sex versus parasite. *Oikos*. 35:282–290.
- Hamilton WD, Axelrod R, Tanese R. 1990. Sexual reproduction as an adaptation to resist parasites (a review). *Proc Natl Acad Sci USA*. 87:3566–3573.
- Hochberg ME, Lawton JH. 1990. Spatial heterogeneities in parasitism and population-dynamics. *Oikos*. 59:9–14.
- Hurst GDD, Sharpe RG, Broomfield AH, Walker LE, Majerus TMO, Zakharov IA, Majerus MEN. 1995. Sexually-transmitted disease in a promiscuous insect, *Adalia bipunctata*. *Ecol Entomol*. 20:230–236.
- Hyman JM, Li J, Stanley EA. 1999. The differential infectivity and staged progression models for the transmission of HIV. *Math Biosci*. 155:77–109.
- Jaenike J. 1978. A hypothesis to account for the maintenance of sex within populations. *Evol Theory*. 3:191–194.
- Johnstone RA. 1995. Sexual selection, honest advertisement and the handicap principle—reviewing the evidence. *Biol Rev Camb Philos Soc*. 70:1–65.
- de Jong PW, Verhoog MD, Brakefield PM. 1993. Sperm competition and melanin polymorphism in the 2-spot ladybird, *Adalia bipunctata* (Coleoptera, Coccinellidae). *Heredity*. 70:172–178.

- Kokko H, Ranta E, Ruxton G, Lundberg P. 2002. Sexually transmitted disease and the evolution of mating systems. *Evolution*. 56:1091–1100.
- Mopper S, Stiling P, Landau K, Simberloff D, Van Zandt P. 2000. Spatiotemporal variation in leafminer population structure and adaptation to individual oak trees. *Ecology*. 81:1577–1587.
- Pedersen AB, Greives TJ. 2008. The interaction of parasites and resources cause crashes in a wild mouse population. *J Anim Ecol*. 77:370–377.
- Perry JC, Sharpe DMT, Rowe L. 2009. Condition-dependent female remating resistance generates sexual selection on male size in a ladybird beetle. *Anim Behav*. 77:743–748.
- Ryder JJ, Hathway J, Knell RJ. 2007. Constraints on parasite fecundity and transmission in an insect-STD system. *Oikos*. 116:578–584.
- Ryder JJ, Webberley KM, Boots M, Knell RJ. 2005. Measuring the transmission dynamics of a sexually transmitted disease. *Proc Natl Acad Sci USA*. 102:15140–15143.
- Schellhorn NA, Andow DA. 2005. Response of coccinellids to their aphid prey at different spatial scales. *Popul Ecol*. 47:71–76.
- Thieltges DW, Reise K. 2007. Spatial heterogeneity in parasite infections at different spatial scales in an intertidal bivalve. *Oecologia*. 150:569–581.
- Thrall PH, Antonovics J, Dobson AP. 2000. Sexually transmitted diseases in polygynous mating systems: prevalence and impact on reproductive success. *Proc R Soc Lond B Biol Sci*. 267:1555–1563.
- Van Buskirk J, Ostfeld RS. 1998. Habitat heterogeneity, dispersal, and local risk of exposure to Lyme disease. *Ecol Appl*. 8:365–378.
- Webberley KM, Buszko J, Isham V, Hurst GDD. 2006. Sexually transmitted disease epidemics in a natural insect population. *J Anim Ecol*. 75:33–43.
- Webberley KM, Hurst GDD, Buszko J, Majerus MEN. 2002. Lack of parasite-mediated sexual selection in a ladybird/sexually transmitted disease system. *Anim Behav*. 63:131–141.
- Webberley KM, Hurst GDD, Husband RW, Schulenburg J, Sloggett JJ, Isham V, Buszko J, Majerus MEN. 2004. Host reproduction and a sexually transmitted disease: causes and consequences of *Coccipolipus hippodamiae* distribution on coccinellid beetles. *J Anim Ecol*. 73:1–10.
- Webberley KM, Tinsley MC, Sloggett JJ, Majerus MEN, Hurst GDD. 2006. Spatial variation in the incidence of a sexually transmitted parasite of the ladybird beetle *Adalia bipunctata* (Coleoptera: Coccinellidae). *Eur J Entomol*. 103:793–797.
- Yorke JA, Hethcote HW, Nold A. 1978. Dynamics and control of transmission of gonorrhoea. *Sex Transm Dis*. 5:51–56.