Heredity (2010) 104, 168–173

www.nature.com/hdy

ORIGINAL ARTICLE

Fine-scale genetic structure and dispersal distance in the harvester ant *Pogonomyrmex barbatus*

SS Suni[1,2] and DM Gordon[1]
[1]*Department of Biological Sciences, Stanford University, Stanford, CA, USA and* [2]*University of Arizona, Tucson AZ, USA*

Dispersal has important genetic and evolutionary consequences. It is notoriously difficult to study in some ant species, because reproductives fly from parent nests to mating aggregations and then to new nest sites. We used genetic techniques to measure dispersal distance and characterize patterns of genetic variation in a population of the harvester ant *Pogonomyrmex barbatus.* This population consists of two interdependent yet genetically distinct mitochondrial lineages, each associated with specific alleles at nuclear loci. We found moderate levels of genetic structure for both lineages and a significant pattern of isolation by distance when individual colonies were the operational unit of study. Dispersal distances calculated from the slope of the regression of genetic on geographic distance were 65.3 m for J1 and 85.8 m for J2. These results are consistent with previous observations of many mating aggregations over small geographic areas. In dependent-lineage populations like our study population, females must mate with males of the opposite lineage to produce workers, and with males of the same lineage to produce female reproductives. Because lineage ratios differ from 1:1 throughout the southwestern United States, restricted dispersal between sites with different lineage ratios could have important effects on dependent-lineage population dynamics. Our results suggest that it is unlikely that many individuals disperse from areas dominated by one lineage to areas dominated by another. Short dispersal distances lead to low gene flow, giving local populations evolutionary independence.
Heredity (2010) **104,** 168–173; doi:10.1038/hdy.2009.124; published online 30 September 2009

Keywords: dispersal; social insect; genetic structure; *Pogonomyrmex barbatus*

Introduction

Dispersal processes influence the extent to which genetic diversity is maintained in populations (Chesser and Ryman, 1986; Crespi and Taylor, 1990; Clobert *et al*., 2001), the level of gene flow between populations (Bohonak, 1999), and the ability of species to expand their ranges (Holt, 2003). The distances that organisms disperse and their ability to colonize areas directly influence population dynamics and evolution of populations (Clobert *et al*., 2001). Restricted dispersal can lead to decreased gene flow and increased levels of inbreeding among groups of individuals. In ants, dispersal behavior and level of gene flow are associated with social structure. Ants with one queen per nest (monogyne) are thought to disperse over large distances and show little genetic structure whereas dispersal is thought to be much more restricted in ants with multiple queens per nest (Pamilo *et a1*., 1997; Ross *et al*., 1997; Chapuisat and Keller, 1999; DeHeer *et al*., 1999; Rüppell *et al*., 1999; Liautard and Keller, 2001; Seppä *et al*., 2005; but see Sundström *et al*., 2003).

Harvester ants (genus *Pogonomyrmex*) are a group of monogynous species whose behavior and ecology have been studied extensively. Our understanding of the evolution of harvester ant populations is constrained by a lack of studies on dispersal and fine-scale genetic structure. Dispersal distance has been notoriously difficult to measure using standard mark-recapture techniques because reproductives fly first to large mating aggregations and then to new nest sites. Here, we used genetic techniques to study dispersal in a dependent-lineage population of about 300 colonies of the widespread harvester ant *Pogonomyrmex barbatus*, lineages J1 and J2 from Helms Cahan and Keller (2003). This population has been the subject of long-term ecological, behavioral, and demographic studies (Gordon and Kulig, 1996).

Certain populations of *P. barbatus* operate under a unique dependent-lineage system, in which reproductive status is associated with genotype (Helms Cahan *et al*., 2002; Julian *et al*., 2002; Volny and Gordon, 2002a; Helms Cahan and Keller, 2003). These populations are thought to have arisen from ancient hybridization between *P. barbatus* and another harvester ant species *P. rugosus* (Schwander *et al*., 2007) or from introgression of *P. barbatus* alleles into *P. rugosus* (Anderson *et al*., 2006). Dependent-lineage populations contain two interbreeding but genetically distinct mitochondrial lineages that are each associated with specific alleles at nuclear loci (Helms Cahan and Keller, 2003). Between-lineage matings result in the production of workers, and within-lineage matings result in the production of reproductives, so females must mate with males of both lineages (Helms Cahan *et al*., 2004). Males are haploid and develop from unfertilized eggs. By analogy with Fisher's sex ratio theory (Fisher, 1939), the optimal lineage ratio for maintenance of the dependent-lineage

Correspondence: Dr S Suni, Department of Biological Sciences, Stanford University, Stanford, CA 94305, USA.
E-mail: sssuni@stanford.edu
Received 30 September 2009; revised 19 May 2009; accepted 24 August 2009; published online 30 September 2009

system is 1:1 (Linksvayer *et al*., 2006). However, the lineage ratio differs significantly from 1:1 at several sites in southeastern Arizona (Schwander *et al*., 2006; Suni *et al*., 2007). Asymmetrical lineage ratios exacerbate constraints put on colony establishment and reproduction by the dependent-lineage system. In areas with highly asymmetrical lineage ratios, queens of the more frequent lineage may have a hard time finding and mating with males of the opposite lineage.

The level of dispersal between sites with different lineage ratios could have important effects on the dynamics of dependent-lineage populations. Restricted dispersal may decrease the chances that queens find and mate with males of the opposite lineage, and found colonies. The goals of this study were to determine the population genetic structure and scale of isolation by distance (IBD) in the geographic area around the long-term study site, and to use measures of genetic differentiation between pairs of individual queens to infer the average distance between parent and offspring colonies.

Materials and methods

Sampling

Pogonomyrmex barbatus colonies occur throughout the southwestern United States. Colonies have one queen, and can reach a mature size of more than 10 000 female workers (Gordon, 1992). Queens can live for over 20 years (Gordon, 1991). Male and female reproductives are produced in the early summer, then fly to a mating aggregation after the summer rains. After mating, males die, and the newly mated females fly or walk away, dig nests, and find new colonies. Each queen attends only one mating flight and produces all the ants in the colony for the next 20 or more years, using the sperm garnered at the original mating flight. When the queen dies, no more workers are produced, and eventually the colony dies; colonies do not adopt new queens.

In late July of 2006, we collected seven male ants from 28–37 colonies at four locations around our long-term study site in southeastern Arizona (Figure 1; Gordon and Kulig, 1996), for a total of 910 individuals. We recorded the location of each colony in UTM with a GPS device. The total area surveyed was about 5.6 km^2, measured as the area of the smallest polygon that encompasses all sampling locations. There are no apparent physical barriers to dispersal in the area. In each location, colonies were sampled in an area of approximately 0.5 km^2. We stored all samples in 95% ethanol until DNA extraction and analysis. All sampling locations were within the geographic range of dependent-lineage *P. barbatus* (Anderson *et al*., 2006; Schwander *et al*., 2007).

Identification of queen genotypes

We used queen genotypes in all population genetic analyses because they are the reproductive individuals in the population and are thus the most appropriate for population genetic studies. Queens produce all males in *P. barbatus* (Suni *et al*., 2007), so we inferred each queen genotype from the genotypes of seven of her sons. This is straightforward because males are haploid: each son has one of the queen's two alleles at each locus. The probability of incorrectly identifying the queen's genotype when seven of her sons are sampled is $(0.5)^7$, or 0.0078 for each locus.

Genetic analyses

DNA was extracted from males by boiling heads in 250 µl of a 5% Chelex (Bio-Rad, Hercules, CA, USA) solution at 95 °C for 20 min. Samples were centrifuged and the supernatant was used as the template for PCR amplification. Individuals were genotyped at eight microsatellite loci using eight primer sets: L-18 (Foitzik *et al*., 1997), Myrt3 (Bourke *et al*., 1997), PO8 (Wiernasz *et al*., 2004), Pb5, Pb7, Pb8, Pb10 (Volny and Gordon, 2002b) and PR-1 (Gadau *et al*., 2003), according to the procedure described in Helms Cahan and Keller (2003). Myrt3 and PR-1 are diagnostic for lineage. PCR products were run on an ABI 3100 automated DNA sequencer (Applied Biosystems, Foster City, CA, USA) using fluorescent

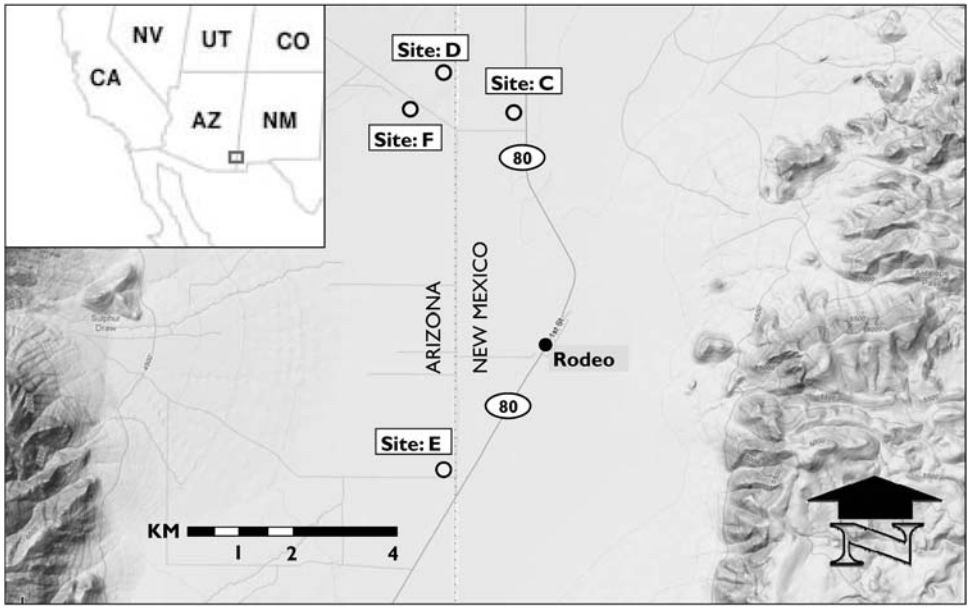

Figure 1 Our four sampling locations in southeast Arizona and southwest New Mexico.

dyes, and analyzed using GENOTYPER software (Applied Biosystems).

Population genetic structure

Using a traditional population genetics approach, we characterized patterns of genetic variation within and between the four sampling locations. All population genetic analyses were conducted separately for each lineage because there is virtually no current gene flow between lineages (Helms Cahan and Keller, 2003). We used queen genotypes derived from male genotypes in all analyses. We used the program F_{STAT} (Goudet, 1995) to test for departure from Hardy–Weinberg equilibrium within locations, and to test for linkage disequilibrium, using 10 000 permutations. We used the program Genepop (Raymond and Rousset, 1995) to calculate Wright's F_{ST} between locations and to perform Mantel tests to test the correlation between genetic distances $F_{ST}/(1\text{-}F_{ST})$ and geographic distances. Geographic distances among individuals were calculated using UTM coordinates of sampling locations. We used the program SPAGeDi (Hardy and Vekemans, 2002) to test for significance of pairwise F_{ST} values. We also used this program to calculate and test for significance of the inbreeding coefficient, Fis. We used the maximum likelihood program STRUCTURE (Pritchard *et al*., 2000) to understand how genetically similar individuals are distributed among the four sampling locations without any previous assumptions of where population subdivision occurs. This program assumes that individuals come from a specified number of populations and computes the likelihood of the observed frequency distribution of genotypes. It then computes the probability that each individual comes from each of the designated groups. We ran the program under the admixture model with a burn in of 25 000 and run lengths of 50 000, and allowed the number of populations to vary from 1 to 4 to determine how many distinct genetic groups are present in the geographic area sampled.

Dispersal distance

To estimate dispersal distance for each lineage, we used a landscape genetics approach based on genotypes of individual queens pooled from all sampling locations. In landscape genetics, individuals are the operational unit of study so breaks in gene flow across geographic areas can be identified without defining populations in advance (Manel *et al*., 2003). If limited dispersal causes a decrease in genetic similarity between individuals at increasing geographic distances, Wright's (1943) IBD model and genetic neighborhood (1946) can then be used to estimate the dispersal distance (for example, Broquet *et al*., 2006; Watts *et al*., 2007). Wright defines a genetic neighborhood as $4D\pi\,\sigma^2$, where D is density and σ^2 is the mean squared axial parent–offspring dispersal distance (Wright, 1946). Assuming IBD within populations, σ^2 can be calculated as $\sigma^2 = 1/(b\,4D\pi)$, where b is the slope of the regression of genetic distance (a) on the logarithm of geographic distance (r) (Rousset, 2000). Rousset's a is a measure of genetic difference between individuals analogous to $F_{ST}/(1\text{-}F_{ST})$ (Rousset, 2000).

We calculated Rousset's a from genotypic data and estimated the scale of IBD for individuals in the geographic area around the long-term study site using program SPAGeDi (Hardy and Vekemans, 2002). This program characterizes the spatial genetic structure of mapped individuals using genotypic data, by computing linear regressions of pairwise statistics on geographical distances. We used permutation tests (10 000 permutations) to test that the slope of the regression of genetic distance a on the logarithm of geographic distances was significantly different from zero.

Using a landscape genetic analysis to estimate dispersal distance was appropriate here because we found a positive relationship between pairwise F_{ST} values of sampling sites and their geographic distances for one lineage, and found significant IBD from the approach based on individuals for both lineages (see Results). We calculated density separately for each lineage, as the number of colonies of each lineage on our long-term study site divided by the area of the long-term study site. Our estimates of density were 0.0017 colonies per square meter for J1 and 0.0023 colonies per square meter for J2. To compare the strength of IBD between lineages we estimated and compared confidence intervals around the slopes by jack-knifing on the sampled populations.

Results

Genetic diversity

We obtained estimates of genetic structure and dispersal distance from the genotypes of 130 queens at eight microsatellite loci. All loci were unlinked for both lineages. For J1, one locus deviated slightly from Hardy–Weinberg equilibrium at sites D and E ($P<0.05$). For J2, two loci deviated slightly from Hardy–Weinberg equilibrium at two sites: Pb5 and Pb8 at sites C and D ($P<0.05$). Nevertheless, we retained all loci for analyses because all these loci were in Hardy–Weinberg equilibrium at the other sites. Number of alleles and gene diversity (Nei, 1978) are shown in Table 1. We classified queens as J1 or J2 by their genotypes at the diagnostic loci Myrt3 (Bourke *et al*., 1997) and PR-1 (Gadau *et al*., 2003). Our sample included 59 J1 colonies and 71 J2 colonies. The number of colonies of each lineage did not differ significantly at any site (χ^2; 1 d.f.; $P>0.05$). Frequencies of J1 ranged from 0.36 to 0.57 (Table 2).

Population genetic structure

The population genetic analyses based on the four sampling locations revealed moderate genetic structure at small spatial scales for both lineages; pairwise F_{ST} values ranged from 0.03 to 0.12 for J1 and from 0.013 to

Table 1 Allele number and gene diversity corrected for sample size (He; Nei, 1978) at 8 microsatellite loci for each lineage

Locus	*J1*		*J2*	
	Alleles	*He*	*Alleles*	*He*
L-18	17	0.88	10	0.82
Myrt 3	5	0.65	2	0.07
PO-8	5	0.32	7	0.55
Pb7	10	0.56	20	0.82
Pb5	11	0.82	13	0.84
Pb8	8	0.73	14	0.87
Pr-1	3	0.083	3	0.22
Pb-10	4	0.26	5	0.46

Table 2 Number of colonies of each lineage at each of the sampling sites in an area of about 5.6 km^2

Site	*Colonies sampled*	*No. J1*	*No. J2*
C	28	11	17
D	32	15	17
E	33	12	21
F	37	21	16

Table 3 Pairwise F_{ST} values between sampling sites for J1 (top right) and J2 (bottom left). Bold numbers represent values significant at $P<0.05$

	C	*D*	*E*	*F*
C		**0.076**	0.035	0.03
D	**0.032**		**0.048**	**0.12**
E	**0.052**	**0.048**		**0.068**
F	0.07	**0.1**	**0.013**	

0.1 for J2, and most were significant (Table 3). We found a significant correlation between genetic distance $F_{ST}/(1\text{-}F_{ST})$ and geographic distance for pairs of sampling sites ($P=0.042$) for J1 and no significant correlation between genetic distance and geographic distance for pairs of sampling sites for J2 ($P=0.29$). The correlation between the F_{ST} values of the lineages was not significant ($P=0.78$). The reduction in heterozygosity within sites because of inbreeding, Fis, was 0.34 for J1 (s.e. = 0.03; $P<0.001$) and 0.26 for J2 (s.e. = 0.034; $P<0.001$). Results from STRUCTURE provided the strongest support for three distinct genetic groups among the individuals sampled. The pattern was the same for both lineages for which analyses were conducted separately. The groups corresponded to the sampling locations; sites B, D and E were dominated primarily by one genetic group. Most individuals (70% for J1 and 66% for J2) could be assigned with high probability (>0.80) to one of the three groups. The rest of the individuals had genomes that were assigned more equally to more than one population, indicating that there is gene flow between some of the sampling locations. Site C had the highest amount of genetic admixture for both lineages; only 53% of the individuals could be assigned with high probability to one of the three genetic groups for J1 and only 40% for J2.

Dispersal distance

We found significant isolation by distance for both lineages from the analysis conducted at the individual level (Figure 2; lineage 1 shown). The slope of the regression of genetic on geographic distance was 0.011 for J1 (s.e. = 0.002; $P=0.018$) and 0.004 for J2 (s.e. = 0.0008; $P=0.037$). The 95% confidence intervals (CI) were [0.0069–0.015] for J1 and [0.0023–0.0056] for J2. A *z*-test for difference between slopes of regression lines revealed that these estimates are significantly different ($P=0.036$). For J1, the mean squared axial parent–offspring dispersal distance is 4264 m^2 and the average dispersal distance is 65.3 m (95% CI, 51–79.4). For J2, the mean squared axial parent–offspring dispersal distance is 7365 m^2 and the average dispersal distance is 85.8 m (95% CI, 68.6–103).

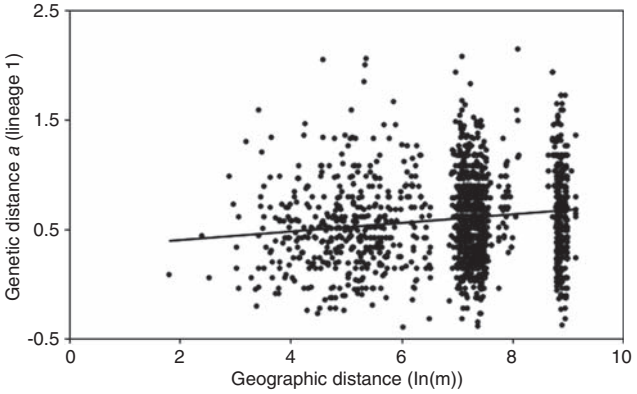

Figure 2 Regression of genetic distance *a* among individuals on the logarithm of geographic distance (m) for lineage 1.

Discussion

Dispersal in *P. barbatus*

We found short average dispersal distances of 65.3 m for J1 and 85.8 m for J2 in dependent-lineage *P. barbatus*. Both the comparison among sampling locations and the comparison among individuals revealed similar patterns of isolation by distance for J1 indicating that gene flow is restricted across the 5.6 km^2 geographic area sampled. However, only the comparison among individuals revealed significant isolation by distance for J2, indicating that J2 may disperse farther than J1. This is further supported by the finding that the slopes of the regressions of genetic on geographic distance for individuals were significantly different for the two lineages.

What conditions favored the evolution of such short dispersal distances in dependent-lineage *P. barbatus*? Long-distance dispersal is the favored strategy in temporally or spatially varying environments (see Johnson and Gaines, 1990 for a review). There are no apparent barriers to dispersal in the geographic area sampled, and the temporal conditions are consistent throughout the region. The lack of habitat heterogeneity may have contributed to the evolution or maintenance of short dispersal distances in *P. barbatus*. Also, dispersal in many *Pogonomyrmex* species occurs during hot, dry weather and the risk of desiccation may favor a dispersal strategy that minimizes the time that newly mated queens spend above ground.

Accuracy of our estimates of dispersal distance

Our estimates of dispersal distance rely on the assumptions that individuals are sampled at an appropriate geographic scale and that density is constant throughout the landscape, but the estimates are robust to departures from the ideal sampling scheme (Leblois *et al*., 2003) or spatial variation in density (Leblois *et al*., 2004). Rousset (2000) defines the ideal sampling scale as $10\sigma \times 10\sigma$, about 0.43 km^2 for J1 and 0.74 km^2 for J2. Ours was just over 5.6 km^2, and thus exceeds the ideal for both lineages. Comparisons between indirect (genetic) and direct (mark-recapture) estimates of dispersal distance have revealed that indirect approaches may underestimate the true dispersal distance by about a factor of 2 (Sumner *et al*., 2001; Broquet *et al*., 2006; Watts *et al*., 2007). Twice our estimate of dispersal distance is 131 m

for J1, and 172 m for J2. These distances are consistent with observations that distances between mating aggregations in the closely related ant species *P. rugosus* were 400 m apart (Hölldobler, 1976), and our own observations that there are one or more mating aggregations of *P. barbatus* each year on a site that is about 400 m across (Gordon DM, personal observations). Assuming a uniform distribution of distances traveled to the mating aggregation, individuals would travel less than 400 m to aggregations. Our results suggest when newly mated queens leave the aggregation, to find new colonies, they do not fly much farther than the distances they traveled from their parent nest.

The results from STRUCTURE further support the finding that the level of gene flow between most sampling locations is low. For both lineages, site C has the highest amount of genetic admixture, perhaps because it is centrally located with respect to the other sites. This corresponds to the low F_{ST} values we found between site C and the other sites, and indicates that though the average dispersal distance is small, some individuals probably disperse much farther than the average.

Our results are consistent with studies of other species within the genus *Pogonomyrmex* in terms of inbreeding and genetic structure. Cole and Wiernasz (1997) found inbreeding in another harvester ant species and suggested that it may be common within the genus. The high levels of inbreeding that we find in dependent-lineage *P. barbatus* are likely a consequence of restricted dispersal. Another contributing factor to inbreeding could be that on our long-term study site, about 20% of the colonies above reproductive age do not produce reproductives at all, and of those that do, about 25% produce only male alates (Gordon and Wagner, 1997). The pool of reproductives at the mating aggregation likely is representative of only some colonies. Also, females of some species of harvester ants mate more often with larger males (Wiernasz *et al*., 2005). If this kind of non-random mating occurs also in *P. barbatus*, it could increase inbreeding by limiting the number of colonies whose male reproductives are successful at mating.

The F_{ST} values we found among sites separated by 1–5 m are similar to those found in other studies on harvester ants at much larger geographic distances. Schwander *et al*. (2007) found F_{ST} values of 0.01–0.15 over hundreds of kilometers. Strehl and Gadau (2004) found a positive correlation between genetic and geographic distances, also over hundreds of kilometers. These are much larger geographic areas than our study, indicating that restricted dispersal and high population subdivision over small geographic areas may be a unique feature of dependent-lineage *P. barbatus*.

Evolution and maintenance of the dependent-lineage system

The dependent-lineage system may have arisen from hybridization between populations of *P. barbatus* and *P. rugosus* (Helms Cahan and Keller, 2003; Schwander *et al*., 2007), or may have evolved in *P. barbatus* and introgressed into *P. rugosus* (Anderson *et al*., 2006). The lineages may have become fixed for incompatible alleles at different nuclear loci (Helms Cahan and Keller, 2003) or evolved incompatible nuclear-mitochondrial combinations (Linksvayer *et al*., 2006). In any case, restricted dispersal might have helped to prevent the influx of alleles from the parental species, facilitating evolution of the system. In a related harvester ant species that also operates under the dependent-lineage system, matings between dependent-lineage queens and males of *P. rugosus* produce viable reproductive offspring, although such matings occur rarely because of pre-zygotic mechanisms of reproductive isolation (Schwander *et al*., 2008). It is possible that when the system was in its infancy, such mechanisms of reproductive isolation did not exist. Reduced gene flow between the hybrid population and the parental species may have contributed to the persistence of the hybrid population and development of the dependent-lineage system.

Once the system evolved, its persistence depended on the maintenance of both lineages in populations. We hypothesized that if lineage ratios differ among sites, dispersal between sites with different lineage ratios could contribute to the maintenance of the system. We found that lineage ratios did not differ significantly from 1:1 at any sampling location. Infrequent dispersal between sampling locations probably does not destabilize the dependent-lineage system because there are an adequate number of colonies of both lineages at each site to maintain the system.

To our knowledge, this is the first study that uses genetic techniques to estimate dispersal distance in an ant species. We found moderate genetic structure and restricted dispersal in a monogynous species of the harvester ant. This contrasts with what is generally thought to be true for monogynous ant species, and indicates that more studies are needed to clarify the relationship between social structure and genetic structure in ants. It is possible that dependent-lineage populations are unique among monogynous ants in terms of dispersal distance. Small dispersal distances may have promoted the evolution of the dependent-lineage system.

Acknowledgements

We thank T Brookhart for creating a figure, the Southwestern Research Station for their facilities, the American Museum of Natural history for its support, and three anonymous reviewers for their helpful comments.

References

Anderson KE, Gadau J, Mott B, Johnson RA, Altamirano A, Streho C *et al.* (2006). Distribution and evolution of genetic caste determination in *Pogonomyrmex* seed harvester ants. *Ecology* **87**: 2171–2184.

Bohonak AJ (1999). Dispersal, gene flow, and population structure. *Q Rev Biol* **74**: 21–45.

Bourke AFG, Green HAA, Bruford MW (1997). Parentage, reproductive skew and queen turnover in a multiple-queen ant analyzed with microsatellites. *Proc R Soc Lond B Biol Sci* **264**: 277–283.

Broquet T, Johnson CA, Petit E, Thompson I, Burel F, Fryxell JM (2006). Dispersal and genetic structure in the American marten, *Martes americana*. *Mol Ecol* **15**: 1689–1697.

Chapuisat M, Lekker L (1999). Extended family structure in the ant *Formica paralugubris*: the role of breeding system. *Behav Ecol Sociobiol* **46**: 405–412.

Chesser RK, Ryman N (1986). Inbreeding as a strategy in subdivided populations. *Evolution* **40**: 616–624.

Clobert J, Danchin E, Dhondt AA, Nichols JD (2001). *Dispersal*. Oxford University Press, Oxford, UK.
Cole BJ, Wiernasz DC (1997). Inbreeding in a lek-mating ant species, *Pogonomyrmex occidentalis*. *Behav Ecol Sociobiol* **40**: 79–86.
Crespi BJ, Taylor PD (1990). Dispersal rates under variable patch density. *Am Nat* **135**: 48–62.
DeHeer CJ, Goodisman MAD, Ross KG (1999). Queen dispersal strategies in the multiple-queen form of the fire ant *Solonopsis invicta*. *Am Nat* **153**: 660–675.
Fisher RA (1939). State of enumeration as a factor influencing the variance of the number of progeny, frequency of mutants, and related quantities. *Ann Eugen* **9**: 406–408.
Foitzik S, Haberl M, Gadau J, Heinze J (1997). Mating frequency of *Leptothorax nylanderi* ant queens determined by microsatellite analysis. *Insectes Soc* **44**: 219–227.
Gadau J, Strehl CP, Oettler J, Hölldobler B (2003). Determinants of intracolonial relatedness in *Pogonomyrmex rugosus* (Hymenoptera; Formicidae): mating frequency and brood raids. *Mol Ecol* **12**: 1931–1938.
Gordon DM (1991). Behavioral flexibility and the foraging ecology of seed-eating ants. *Am Nat* **138**: 379–411.
Gordon DM (1992). How colony growth affects forager intrusion between neighboring harvester ant colonies. *Behav Ecol Sociobiol* **31**: 417–427.
Gordon DM, Kulig AW (1996). Founding, foraging, and fighting: colony size and the spatial distribution of harvester ant nests. *Ecology* **77**: 2293–2409.
Gordon DM, Wagner D (1997). Neighborhood density and reproductive potential in harvester ants. *Oecologia* **109**: 556–560.
Goudet J (1995). FSTAT Version 1.2: a computer program to calculate F statistics. *J Hered* **86**: 485–486.
Hardy OJ, Vekemans X (2002). SPAGeDi : a versatile computer program to analyze spatial genetic structure at the individual or population levels. *Mol Ecol Notes* **2**: 618–620.
Helms Cahan SH, Parker JD, Rissing SW, Johnson RA, Polony TS, Weiser MD *et al.* (2002). Extreme genetic differences between queens and workers in hybridizing *Pogonomyrmex* harvester ants. *Proc R Soc Lond B Biol Sci* **269**: 1871–1877.
Helms Cahan SH, Keller L (2003). Complex hybrid origin of genetic caste determination in harvester ants. *Nature* **424**: 306–309.
Helms Cahan SH, Julian GE, Rissing SW, Schwander T, Parker JD, Keller L (2004). Loss of phenotypic plasticity generates genotype-caste association in harvester ants. *Curr Biol* **14**: 2277–2282.
Hölldobler B (1976). The behavioral ecology of mating in harvester ants. *Behav Ecol Sociobiol* **1**: 405–423.
Holt RD (2003). On the evolutionary ecology of species' ranges. *Evol Ecol Res* **5**: 159.
Johnson ML, Gaines MS (1990). Evolution of dispersal: theoretical models and empirical tests using birds and mammals. *Ann Rev Ecol Syst* **21**: 449–480.
Julian GE, Fewell JH, Gadau J, Johnson RA, Larrabee D (2002). Genetic determination of the queen caste in an ant hybrid zone. *Proc Natl Acad Sci USA* **99**: 8157–8160.
Leblois R, Estoup A, Rousset F (2003). Influence of mutational and sampling factors on the estimation of demographic parameters in a 'continuous' population under isolation by distance. *Mol Biol Evol* **20**: 491–502.
Leblois R, Rousset F, Estoup A (2004). Influence of spatial and temporal heterogeneities on the estimation of demographic parameters in a continuous population using individual microsatellite data. *Genetics* **166**: 1081–1092.
Liautard C, Keller L (2001). Restricted effective queen dispersal at a microgeographic scale in polygynous populations of the ant Formica exseta. *Evolution* **55**: 2484–2492.
Linksvayer T, Wade M, Gordon DM (2006). Genetic caste determination in harvester ants: possible origin and maintenance by cyto-nuclear epistasis. *Ecology* **87**: 2185–2193.
Manel S, Schwartz MK, Luikart G, Taberlet P (2003). Landscape genetics: combining landscape ecology and population genetics. *Trends Ecol Evol* **18**: 189–197.
Nei M (1978). Estimation of average heterozygosity and genetic distance from a small number of individuals. *Genetics* **89**: 583–590.
Pamilo P, Gertsch P, Thoren P, Seppä P (1997). Population genetics of social insects. *Ann Rev Ecol Syst* **28**: 1–25.
Pritchard J, Stephens M, Donnelly P (2000). Inference of population structure using multilocus genotype data. *Genetics* **155**: 945–959.
Raymond M, Rousset R (1995). GENEPOP version 1.2: population genetics software for exact tests and ecumenicism. *J Hered* **86**: 248–249.
Ross KG, Krieger MJB, Shoemaker DD, Vargo EL, Keller L (1997). Hierarchical analysis of genetic structure in native fire ant populations: results from three classes of molecular markers. *Genetics* **147**: 643–655.
Rousset F (2000). Genetic differentiation between individuals. *J Evol Biol* **13**: 58–62.
Rüppell O, Heinze J, Hölldobler B (1999). Genetic and social structure of the queen size dimorphic ant *Leptothorax cf. andrei*. *Ecol Entomol* **26**: 76–82.
Schwander T, Helms Cahan S, Keller L (2006). Genetic caste determination imposes costs during colony founding. *J Evol Biol* **19**: 402–409.
Schwander TS, Helms Cahan S, Keller L (2007). Characterization and distribution of *Pogonomyrme*x harvester ant lineages with genetic caste determination. *Mol Ecol* **16**: 367–387.
Schwander TS, Suni SS, Helms Cahan S, Keller L (2008). Mechanisms of reproductive isolation between an ant species of hybrid origin and one of its parents. *Evolution* **62**: 1635–1643.
Seppä P, Gyllenstrand N, Corander J, Pamilo P (2004). Coexistence of the social types: genetic population structure in the ant *Formica exsecta*. *Evolution* **58**: 2462–2471.
Strehl CP, Gadau J (2004). Cladistic analysis of Paleo-island populations of the Florida harvester ant Hymenoptera: Formicidae. based upon divergence of mitochondrial DNA sequences. *Florida Entomol* **87**: 576–581.
Sumner J, Rousset F, Estoup A, Moritz C (2001). Neighborhood size, dispersal and density estimates in the prickly forest skink *Gnypetoscincus queenslandiae*. using individual genetic and demographic methods. *Mol Ecol* **10**: 1917–1927.
Sundström L, Keller L, Chapuisat M (2003). Inbreeding and sex-biased gene flow in the ant *Formica exsecta*. *Evolution* **57**: 1552–1561.
Suni SS, Gignoux C, Gordon DM (2007). Male parentage in dependent-lineage populations of the harvester ant *Pogonomyrmex barbatus*. *Mol Ecol* **16**: 5149–5155.
Volny V, Gordon DM (2002a). Genetic basis for queen-worker dimorphism in a social insect. *Proc Natl Acad Sci USA* **99**: 6108–6111.
Volny V, Gordon DM (2002b). Characterization of polymorphic microsatellite loci in the red harvester ant *Pogonomyrmex barbatus*. *Mol Ecol Notes* **2**: 302–303.
Watts PC, Rousset F, Saccheri IJ, Leblois R, Kemp SJ, Thompson DG (2007). Compatible genetic and ecological estimates of dispersal rates in insect *Coenagrion mercuriale:* Zygoptera. populations: an analysis of 'neighborhood size' using a more precise estimator. *Mol Ecol* **16**: 737–751.
Wiersasz DC, Yencharis J, Cole BJ (1995). Size and mating success in males of the western harvester ant, *Pogonomyrmex occidentalis* (Hymenoptera: Formicidae). *J Insect Behav* **8**: 523–531.
Wiernasz D, Perroni C, Cole B (2004). Polyandry and fitness in the western harvester ant, *Pogonomyrmex occidentalis*. *Mol Ecol* **13**: 1601–1606.
Wright S (1943). Isolation by distance. *Genetics* **28**: 114–138.
Wright S (1946). Isolation by distance under diverse systems of mating. *Genetics* **31**: 39–59.