Animal Behaviour 82 (2011) 131–137

Contents lists available at ScienceDirect

Animal Behaviour

journal homepage: www.elsevier.com/locate/anbehav

Genetic structure reflects natal dispersal movements at different spatial scales in the blue tit, *Cyanistes caeruleus*

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ARTICLE INFO

Article history:
Received 3 January 2011
Initial acceptance 2 March 2011
Final acceptance 8 April 2011
Available online 6 May 2011
MS. number: 11-00005

Keywords:
blue tit
Cyanistes caeruleus
genetic structure
microsatellite
natal dispersal
population fragmentation
sex-biased dispersal

The study of the genetic consequences of dispersal is a central topic in evolutionary, conservation and behavioural research. However, few studies have simultaneously considered dispersal movements from marked individuals and contemporary patterns of gene flow. We analysed the link between dispersal behaviour and gene flow in four populations of blue tits with different degrees of connectivity. For this purpose, we monitored four breeding patches and used genotypic and capture–mark–recapture data to study the genetic consequences of dispersal at different spatial scales. Data on natal dispersal movements revealed that both males and females dispersed less than expected under a random pattern of settlement at the two large spatial scales considered: the whole study area and the two main localities. However, natal dispersal distance was lower than expected under random settlement within natal patches in males whereas an opposite pattern was found for females. Accordingly, microsatellite data revealed limited gene flow between the localities studied and an isolation-by-distance pattern of genetic structure that was particularly strong at the large spatial scale (i.e. considering geographically distant breeding patches). Finally, the strong male philopatry was reflected by a stronger genetic structure and a lower admixed ancestry in this sex. Overall, we found evidence that restricted dispersal and fragmentation may have both contributed to reduce interpopulation gene flow at different spatial scales in a forest species and that there is concordance between genetic studies and those based on capture–mark–recapture.

Dispersal is a life history trait that plays a fundamental role in population dynamics, influencing species distribution and the genetics and structure of populations (Clobert et al. 2001). Restricted dispersal can result in increased population fragmentation and genetic differentiation (Martínez-Cruz et al. 2007; Coulon et al. 2008; Tzika et al. 2008). This population subdivision is expected to reduce effective population sizes, which can ultimately decrease genetic variability and the ability of populations to respond to selection and adapt to novel environmental conditions (Frankham 1996, 2005; Spielman et al. 2004; Willi et al. 2006). On the other hand, restricted dispersal and population fragmentation can also preserve genetic variation from the homogenizing effects of gene flow, favouring the maintenance of local adaptations and promoting intraspecific diversity or speciation (Smith et al. 1997, 2005; Milá et al. 2009). Thus, the study of the genetic consequences of dispersal has important theoretical and applied implications which can be considered major topics in behavioural, evolutionary and conservation research (Saunders et al. 1991; Fahrig 2002).

The advent and application of DNA polymorphic markers has opened up the possibility that researchers can estimate realized gene flow and infer how individuals move across the landscape (Manel et al. 2003). However, few studies have simultaneously considered dispersal movements from marked individuals and contemporary patterns of gene flow (Riley et al. 2006; Boulet et al. 2007; Coulon et al. 2008, 2010; Alcaide et al. 2009). Thus, more empirical information is required to reconcile the patterns of dispersal and genetic structure observed in spatially structured populations (i.e. metapopulations; Ortego et al. 2008; Alcaide et al. 2009; Coulon et al. 2010). Simultaneous analyses of patterns of dispersal and genetic structure considering contrasting scenarios of human-induced or natural population fragmentation and isolation at different spatial scales could help to resolve this problem (Boulet et al. 2007; Coulon et al. 2008, 2010).

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doi:10.1016/j.anbehav.2011.04.007

Table 1
Natal dispersal distances (m) recorded for various blue tit populations

Population	Sex	Mean/SE	Median	*N*	Source
France	Males	463/–	–	35*	Parejo et al. 2007
France	Females	1356/–	–	35*	Parejo et al. 2007
Austria	Combined	328/–	–	65	Foerster et al. 2006
Belgium	Males	–/–	256	70*	Matthysen et al. 2005
Belgium	Females	–/–	936	70*	Matthysen et al. 2005
Germany	Males	–/–	365	218*	Winkel & Frantzen 1991
Germany	Females	–/–	700	218*	Winkel & Frantzen 1991
Spain	Males	331/32	250	77	Present study
Spain	Females	649/112	442	89	Present study

* Sample size is reported for both sexes combined.

In this study, we analysed the link between dispersal behaviour and gene flow in four populations of blue tits with different degrees of connectivity. This forest species can be considered mostly sedentary and disperses over short distances, which would make it particularly prone to population fragmentation (Matthysen et al. 2005; Foerster et al. 2006; Valcu & Kempenaers 2008; Table 1). Furthermore, the short generation time of this species (modal life span = 2 years) can favour the detection of genetic shifts (e.g. changes in allele frequency or genetic diversity) at very short spatiotemporal scales (Hailer et al. 2006; Ortego et al. 2007). In particular, the Mediterranean populations of the blue tit often show a high degree of isolation owing to both natural and human-induced fragmentation of the deciduous forests optimal for this species (Blanco et al. 1997). For these reasons, the Mediterranean populations of blue tits offer an ideal scenario to study the genetic consequences of restricted dispersal and population fragmentation. In this study we monitored four populations (two localities with two patches each) and used detailed capture–mark–recapture information and genotypic data to analyse the genetic consequences of dispersal behaviour at different spatial scales. In particular, we tested the following predictions: (1) because of the small body size, sedentary behaviour and habitat fragmentation, we expected (1.1) the study populations of blue tits would show very restricted natal dispersal movements and (1.2) dispersal distances would be particularly short in males, the less dispersive sex in most bird species (Greenwood 1980); (2) we also expected low gene flow and significant genetic differentiation between patches and localities with limited or no dispersal between them (Coulon et al. 2008); (3) if males are the less dispersive sex, we predicted that this sex would show a more marked genetic structure than females; (4) finally, we expected that genetic similarity between individuals would decrease with geographical distance (i.e. isolation by distance) because of migration–drift equilibrium (Hutchison & Templeton 1999).

METHODS

Study Areas and Population Monitoring

We studied blue tits in two localities of central Spain (Fig. 1): the Cabañeros National Park (Ciudad Real and Toledo provinces, 39°24′N, 3°35′W) and Quintos de Mora (Toledo province, 39°25′N, 4°04′W). Both study localities comprise mainly oak-dominated woodlands with Mediterranean scrublands. In each of these two localities we monitored two breeding patches: El Brezoso and Anchurones in Cabañeros National Park and Valdeyerno and Gil García in Quintos de Mora (Fig. 1). Nestboxes in Cabañeros (Anchurones: 150 nestboxes; El Brezoso: 100 nestboxes) and Quintos de Mora (Gil García: 100 nestboxes; Valdeyerno: 100 nestboxes) were monitored from April to June during 2007–2009. Basic data on reproduction were obtained by frequent checks of nestboxes from nest building until fledging (Sanz & García-Navas 2009). Adult birds were captured when feeding 8-day-old chicks by means of a spring trap shutting the entrance hole as the bird entered the nest. Birds were sexed by the presence/absence of a brood patch (Sanz & García-Navas 2009). Blood samples (≤25 µl) for genetic analyses were obtained by brachial venipuncture and stored in 96% ethanol. All adults and nestlings were individually marked with aluminium rings for further identification (García-Navas et al. 2009; Sanz & García-Navas 2009).

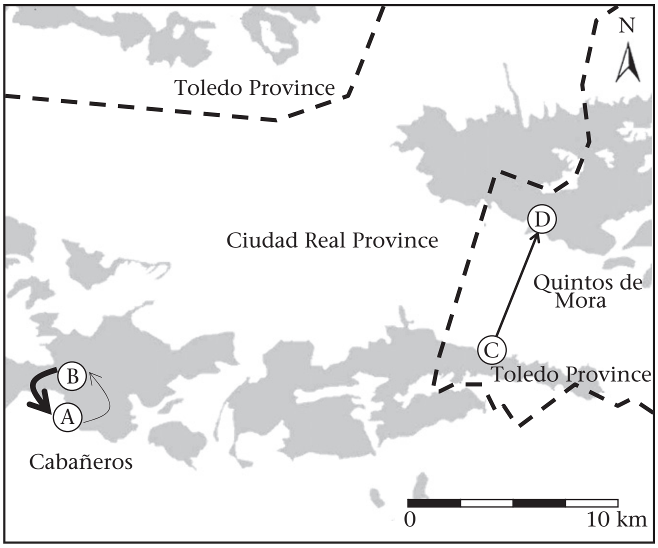

Figure 1. Map of the study area showing the spatial location of the studied blue tit populations at Cabañeros (A: Anchurones; B: Brezoso) and Quintos de Mora (C: Gil García; D: Valdeyerno). Shaded area represents woodlands. Arrows indicate observed dispersal movements and direction. Arrow thickness is proportional to the number of dispersed individuals.

Natal Dispersal Movements

Natal dispersal distance was calculated as the Euclidean distance from the natal to the first-breeding nestbox (Greenwood & Harvey 1982). We analysed differences between the sexes in observed natal dispersal distances using a generalized linear mixed model (GLMM) implemented with the GLIMMIX macro of SAS (SAS Institute 2004). Dispersal distance was analysed using a normal error structure and identity link function. Cohort, locality identity (Cabañeros, Quintos de Mora), breeding patch identity (Anchurones, Brezoso, Gil García or Valdeyerno) nested within locality and brood identity nested within breeding patch were included as random effects.

We also used Monte Carlo simulations to evaluate the possibility that observed natal dispersal patterns have occurred by chance and are merely constrained by the spatial distribution of breeding sites (Manly 1991; Serrano et al. 2008). For this purpose, we considered three different null models: (1) that individuals settle randomly in any nestbox located within the whole study area; (2) that individuals settle randomly in any nestbox located within their natal locality (i.e. Cabañeros or Quintos de Mora); (3) that individuals settle randomly in any nestbox located within their natal patch (i.e. Anchurones, Brezoso, Gil García or Valdeyerno). We performed three different simulations randomly assigning each individual to a nestbox and only constraining its settlement according to any of the three null models considered above. Then we calculated the distance between the natal nestbox for each individual and the randomly assigned nestbox to generate the expected frequency distribution of dispersal distances (i.e. the null model; e.g. Serrano et al. 2008). Our study sites contain a high number of nestboxes and many of them (ca. 40–50%) remain unoccupied every year (see Valcu & Kempenaers 2008). Thus, the three null models considered that natal dispersal decisions are not constrained by intraspecific competition or nest site availability (e.g. Serrano et al. 2008). Simulations of the three null models were repeated 1000 times to obtain the expected frequency distribution of median dispersal distances under the different scenarios. The expected distribution of median dispersal distances was compared with the observed median dispersal distance. Tests of significance were generated by counting the randomized cases that resulted in an equal or larger/smaller value than the observed median dispersal distance and dividing by the total number of randomizations (Manly 1991). Natal dispersal distances are larger in females than in males in the populations studied and so data were analysed separately for each sex (see below).

Microsatellite Genotyping

We genotyped 212 breeding individuals (Anchurones: $N = 33$; Brezoso: $N = 74$; Gil García: $N = 54$; Valdeyerno: $N = 51$) across 14 polymorphic microsatellite markers (García-Navas et al. 2009). We used NucleoSpin Tissue Kits (Machery-Nagel, Düren, Germany) to extract and purify genomic DNA from the blood samples. Approximately 5 ng of template DNA was amplified in 10 μl reaction volumes containing 1× reaction buffer (67 mM Tris–HCL, pH 8.3, 16 mM $(NH_4)_2SO_4$, 0.01% Tween-20, EcoStart Reaction Buffer, Ecogen, Barcelona, Spain), 2 mM $MgCl_2$, 0.2 mM of each dNTP, 0.15 μM of each dye-labelled primer (FAM, PET, VIC or NED) and 0.1 U of *Taq* DNA EcoStart Polymerase (Ecogen). All reactions were carried out on a Mastercycler EpgradientS (Eppendorf) thermal cycler. The PCR programme used was 9 min denaturing at 95 °C followed by 35 cycles of 30 s at 94 °C, 45 s at the annealing temperature (García-Navas et al. 2009) and 45 s at 72 °C, ending with a 5 min final elongation stage at 72 °C. Amplification products were electrophoresed using an ABI 310 Genetic Analyzer (Applied Biosystems, Inc., Foster City, CA, U.S.A.) and genotypes were scored using GeneMapper 3.7 (Applied Biosystems). Microsatellite genotypes were tested for departure from Hardy–Weinberg equilibrium (HWE) at each locus using an exact test (Guo & Thompson 1992) based on 900 000 Markov chain iterations as implemented in the program Arlequin 3.1 (Excoffier et al. 2005). We also used Arlequin 3.1 to test for linkage equilibrium within each pair of loci using a likelihood-ratio statistic, whose distribution was obtained by a permutation procedure (Excoffier et al. 2005).

Spatial Genetic Structure

We investigated genetic structure among the studied blue tit localities and patches. For this purpose we calculated pairwise fixation indexes (F_{ST}) between the different localities and patches and tested their significance with Fisher's exact tests after 10 000 permutations as implemented in Arlequin 3.1 (Excoffier et al. 2005). We also analysed the spatial genetic structure using an individual-based approach as implemented in the program Structure version 2.3.3 (Pritchard et al. 2000; Falush et al. 2003; Hubisz et al. 2009). This program is a Bayesian model-based clustering method, which assigns individuals to populations based on their multilocus genotypes (Pritchard et al. 2000; Falush et al. 2003). For *K* population clusters, the program estimates the probability of the data [$\Pr(X|K)$] and the probability of individual membership in each cluster using a Markov chain Monte Carlo (MCMC) method. We ran the program assuming correlated allele frequencies and admixture (Pritchard et al. 2000; Falush et al. 2003) and using the 'prior population information' option to take sampling locations into account (Hubisz et al. 2009). We conducted five independent runs for each value of *K* to estimate the true number of clusters with 10^6 MCMC cycles, following a burn-in period of 100 000 iterations. The simulated values of *K* ranged from 1 to 4. The number of populations best fitting the data set (*X*) was defined using log probabilities [$\Pr(X|K)$] and ΔK, as described in Evanno et al. (2005). Thus, the estimated number of subpopulations is taken to be the value of *K* at which $\Pr(X|K)$ plateaus. All analyses on spatial genetic structure were performed for all sampled individuals together and considering male and female genotypes separately. The deviation from a pure genotype (admixed ancestry) was calculated for each individual using the assignment scores provided by Structure analyses (e.g. Ortego & Bonal 2010). Then, we used a one-way ANOVA to analyse differences between the sexes in the degree of admixed ancestry.

Isolation by Distance

We explored the occurrence of an isolation-by-distance pattern of spatial genetic structure comparing pairwise relatedness and Euclidean geographical distances between individuals. We used the software Mark (K. Ritland; www.genetics.forestry.ubc.ca/ritland/programs.html) to calculate pairwise relatedness values for all individuals using Queller & Goodnight's (1989) estimator. We also calculated the pairwise geographical distance between all individuals using an Excel spreadsheet. Finally, we used Mantel tests to analyse the association between distance matrixes using ZT software (Bonnet & Van de Peer 2002). The significance of the Mantel test was assessed by 10 000 randomizations of the genetic distance matrix. The test was one tailed as only a positive correlation between geographical and genetic distances is expected (e.g. Gómez et al. 2007; Ortego et al. 2010). This approach was used to analyse all sampled individuals and only considering male–male and female–female comparisons in the whole study area and for Cabañeros and Quintos de Mora localities separately.

Ethical Note

The study was done under licence from the Spanish institutional authorities (Environmental Agency of the Community of Castilla-La Mancha and the Ringing Office of the Ministry of Environment). These institutions provided permits for capturing and ringing blue tits and all these activities were performed following general ethical guidelines for animal welfare and nature conservation. All the breeding adults were captured with spring traps when the chicks were 8 days old (day of hatching = 0). Chicks were banded on day 13 after hatching. The blood-sampling procedure did not have any obvious adverse effects on the birds and our trapping and handling did not cause any harm, nest desertion or any other damage to the individuals or their nests.

RESULTS

Natal Dispersal Movements

We recaptured 166 individuals that were ringed as nestlings, 62 individuals at Cabañeros (Anchurones: 10 males, 10 females; El Brezoso: 21 males, 21 females) and 104 individuals at Quintos de Mora (Gil García: 27 males, 25 females; Valdeyerno: 19 males, 33 females). We did not record any movement between localities (Cabañeros–Quintos de Mora). However, we recorded some movements between close patches: from Gil García to Valdeyerno (two females), from Anchurones to El Brezoso (one male), from El Brezoso to Anchurones (two males, five females; Fig. 1). No individual moved from Valdeyerno to Gil García. After we controlled for

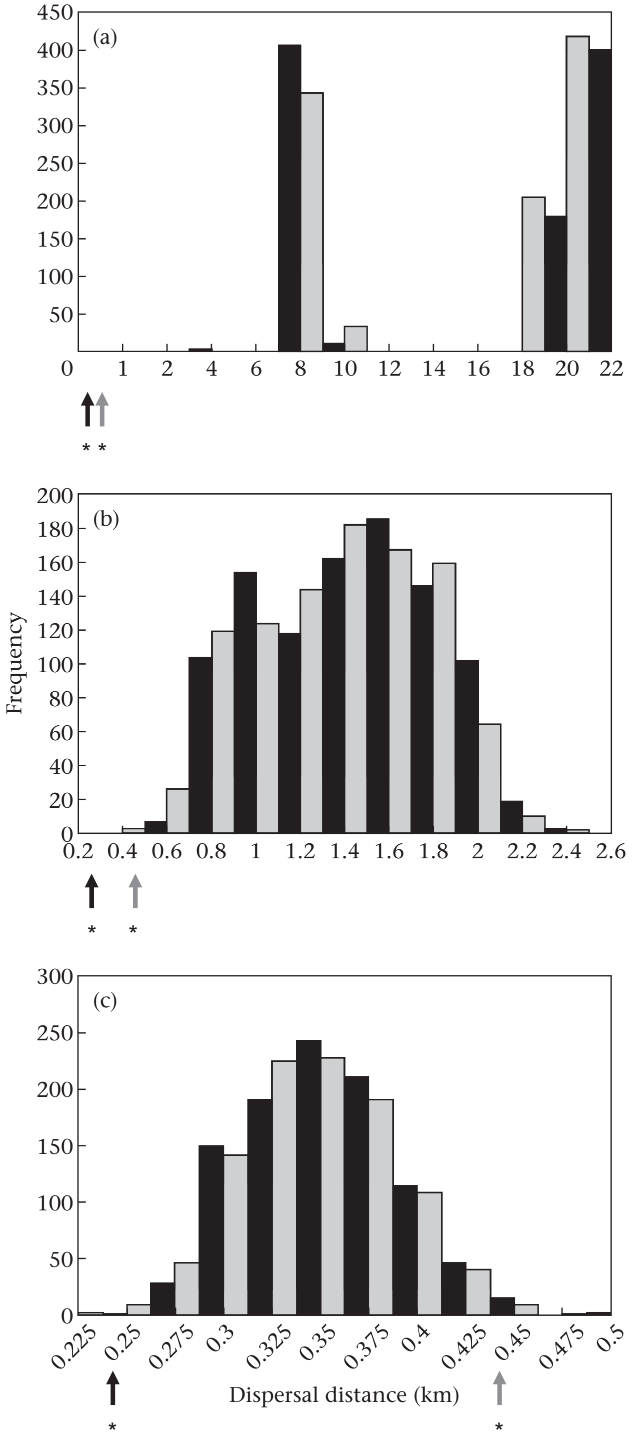

Figure 2. Frequency distribution of median dispersal distances expected for male (black bars) and female (grey bars) blue tits assuming different scenarios of random dispersal: (a) that individuals settle in any nestbox located in the whole study area; (b) that individuals settle in any nestbox located within their natal area (Cabañeros or Quintos de Mora); (c) that individuals settle in any nestbox located within their natal patch (Anchurones, Brezoso, Gil García or Valdeyerno). Arrows indicate the observed value for each sex (male: black arrow; female: grey arrow). $^*P < 0.05$.

random effects, natal dispersal distances were significantly lower in males than in females (GLMM: $F_{1,164} = 6.57$, $P = 0.011$; Table 1).

Monte Carlo simulations revealed that both male and female median dispersal distances were lower than expected under random dispersal assuming that individuals can potentially settle in any nestbox located within the whole study area ($P < 0.001$; Fig. 2a). Similarly, median dispersal distances in both males and females were lower than expected under random dispersal assuming that individuals can potentially settle in any nestbox located within their natal locality (i.e. Cabañeros or Quintos de Mora; $P < 0.001$; Fig. 2b). However, males dispersed less than expected under random dispersal within their natal patch (i.e. Anchurones, Brezoso, Gil García or Valdeyerno; $P = 0.049$; Fig. 2c), but an opposite pattern was found for females ($P = 0.001$; Fig. 2c).

Spatial Genetic Structure

On average, individuals were successfully typed at 13.4 ± 1.3 (SE) microsatellite loci. After applying sequential Bonferroni corrections to compensate for multiple statistical tests, we found that no locus deviated from HWE. There was no evidence of genotypic linkage disequilibrium at any pair of loci (all $P > 0.05$). Pairwise F_{ST} values revealed a significant genetic differentiation between Cabañeros and Quintos de Mora localities ($F_{ST} = 0.003$, $P = 0.006$). This pattern was mainly driven by a marked genetic structure in males ($F_{ST} = 0.011$, $P < 0.001$) that was not significant for females ($F_{ST} = 0.001$, $P = 0.201$). Pooling male and female genotypes, we also found significant pairwise F_{ST} values between different patches located in the two main study areas (Table 2). When we considered each sex separately, we found significant F_{ST} values between some patches located in different localities in males but never in females (Table 2).

Structure analyses considering all individuals revealed a maximum $\Pr(X|K)$ for $K = 2$ (Figs 3a, 4a), indicating the presence of two genetic clusters in the study area. Genetic differentiation was particularly strong between El Brezoso and either of the two patches located in Quintos de Mora locality, although all the analyses revealed high levels of genetic admixture (Fig. 4a). Analyses considering only male genotypes also revealed a maximum $\Pr(X|K)$ for $K = 2$, with a similar pattern of genetic structure to that obtained in the global analyses including all individuals (Figs 3b, 4b). However, analyses for females revealed a maximum $\Pr(X|K)$ for $K = 1$, indicating no genetic subdivision for this sex (Figs 3c, 4c). All these patterns were consistent over different runs for the same K (Fig. 3). The deviation from pure genotypes was significantly lower in males, suggesting a higher admixed ancestry in females (one-way ANOVA: $F_{1,210} = 1241.54$, $P < 0.001$). Overall, these analyses show the presence of two genetic clusters for males but indicate a complete lack of genetic structure in females.

Isolation by Distance

Isolation-by-distance analyses revealed a significant negative correlation between genetic relatedness and geographical distances considering all individuals ($r = -0.0556$, $P < 0.001$) or analysing male ($r = -0.112$, $P < 0.001$) and female ($r = -0.037$, $P = 0.002$) genotypes separately. Considering only data from

Table 2
Pairwise population F_{ST} values between sampling patches considering all individuals (below the diagonal) and male and female genotypes separately (above the diagonal, first male and then female values)

	Anchurones	Brezoso	Gil García	Valdeyerno
Anchurones	–	0.001/0.005	0.007/0.007	0.004/0.007
Brezoso	0.005	–	**0.014**/0.003	**0.014**/−0.002
Gil García	**0.008**	**0.005**	–	0.002/0.002
Valdeyerno	0.006	0.004	0.004	–

Values in bold are statistically significant after sequential Bonferroni correction ($\alpha = 0.05$).

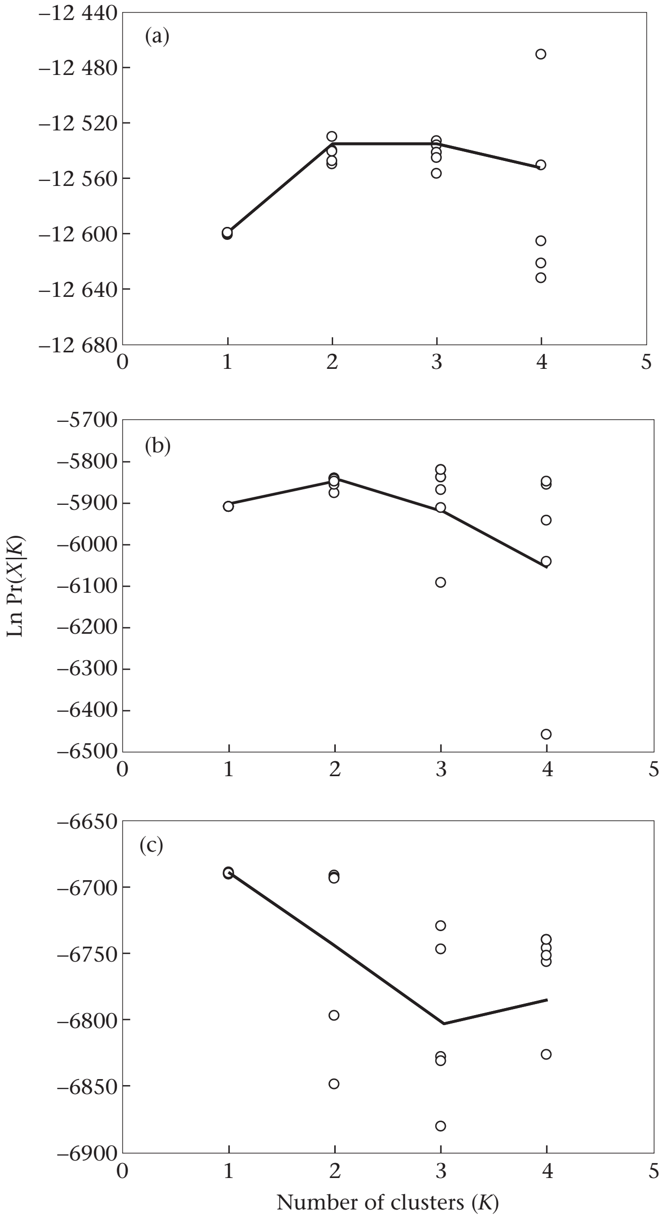

Figure 3. Results of Bayesian clustering analyses considering (a) all individuals and (b) male and (c) female genotypes separately. For each number of population clusters tested (*K*), Pr(*X*|*K*) is the probability of the data.

Cabañeros locality, we found no significant correlation between genetic and geographical distances considering all individuals ($r = 0.024$, $P = 0.962$) or analysing male ($r = 0.003$, $P = 0.543$) and female ($r = -0.029$, $P = 0.119$) genotypes separately. In Quintos de Mora locality we found a marginally significant negative correlation between genetic relatedness and geographical distances ($r = -0.018$, $P = 0.087$) which was mainly driven by male genotypes ($r = -0.068$, $P = 0.009$). By contrast, we found no correlation between genetic relatedness and geographical distances when only female genotypes were considered ($r = 0.024$, $P = 0.829$).

DISCUSSION

We found evidence that restricted dispersal contributes to reducing interpopulation gene flow at different spatial scales. Our results revealed that both males and females disperse shorter distances than expected from null models of random dispersal within both the whole study area and the studied localities (Quintos de Mora and Cabañeros). However, natal dispersal distance was lower than expected under random settlement within natal patches in males, whereas an opposite pattern was found for females. These results suggest that behavioural/physical barriers to dispersal could differ between the sexes and the different spatial scales analysed (Pierson et al. 2010). In males, behavioural constraints are probably the main factor limiting movements within natal patches where suitable habitat shows a continuous distribution in the absence of any physical barrier to dispersal (Greenwood 1980; see below). By contrast, habitat fragmentation could reduce dispersal between habitat patches in females even if this sex is behaviourally predisposed to, or physiologically capable of, dispersing over large distances in continuous habitats (Harris & Reed 2002). As suggested for other forest passerines, females may be reluctant to enter inadequate habitats (agricultural land, scrubland, cleared forest or grassland) owing to increased predation risk and this could result in a fragmentation-derived behavioural inhibition of gene flow (Harris & Reed 2002). Results on dispersal patterns suggest that habitat fragmentation would have a lower impact on male-mediated gene flow because male dispersal is limited even within natal patches showing a continuous distribution of suitable habitats (Pierson et al. 2010). In any case, it should be considered that the consequences of habitat fragmentation on male gene flow are expected to be very dependent on the size of the patch under consideration and smaller patches than those studied here may result in decreased male dispersal rates.

Information on gene flow inferred from genotypic data is mainly concordant with the observed dispersal patterns, although such parallelism depends on the spatial scale analysed (Coulon et al. 2008; Alcaide et al. 2009). Absence of dispersal between localities (Cabañeros, Quintos de Mora; Fig. 1) is reflected by highly significant F_{ST} values and global individual-based clustering analyses. However, although dispersal events were rarely observed between patches within Quintos de Mora, there was a complete absence of genetic structure within this locality, whether considering F_{ST} values or individual-based clustering analyses (Fig. 4). We also found a globally significant correlation between genetic and geographical distances, indicating an isolation-by-distance pattern of genetic structure (Wright 1943; Slatkin 1993; Hutchison & Templeton 1999). The high admixture levels revealed by the Structure analyses suggest that the global pattern of isolation by distance may be compatible with a migration–drift equilibrium scenario, that is, restricted dispersal from nearby populations may have resulted in a significant correlation between genetic and geographical distances (Hutchison & Templeton 1999; Coulon et al. 2008). Alternatively, the observed pattern could have been generated through serial colonization from geographically close populations and subsequent genetic drift as a consequence of persistent founder effects (Ramachandran et al. 2005; Mills et al. 2007; Ortego et al. 2010). Analyses of isolation by distance within the two localities showed a significant correlation between genetic and geographical distances only in Quintos de Mora. In this locality only 1.92% of dispersal events observed involved movements between patches, suggesting that restricted dispersal could have resulted in the observed isolation-by-distance pattern of genetic structure (Hutchison & Templeton 1999; Coulon et al. 2008) which is not reflected by F_{ST} values or individual-based clustering analyses (Van de Casteele et al. 2003; Ortego et al. 2008). Thus, the blue tit population at this locality shows an ‘isolation-by-distance’ model rather than an ‘island’ model of genetic structure (Van de Casteele & Matthysen 2006). By contrast, the absence of correlation between genetic and geographical distances within Cabañeros locality could be the result of the homogenizing effects of migration caused by the

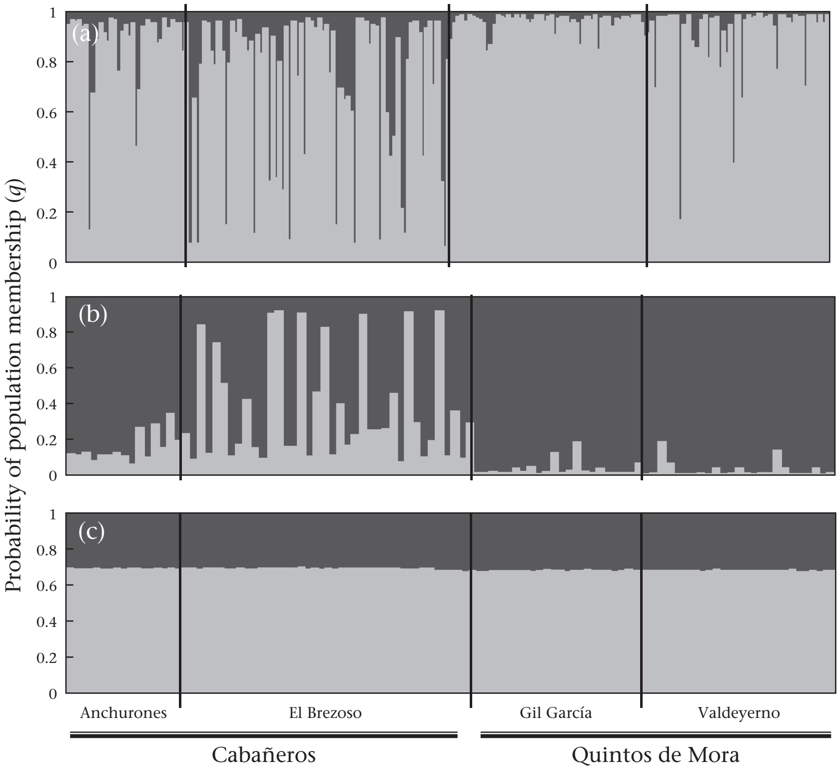

Figure 4. Results of genetic assignment based on the Bayesian method implemented in the program Structure considering (a) all individuals and (b) male and (c) female genotypes separately. Each individual is represented by a thin vertical line, which is partitioned into two coloured segments that represent the individual's probability of belonging to the cluster with that colour.

relatively frequent dispersal events (12.9%) observed between the two patches studied within this locality (Hutchison & Templeton 1999).

Capture–mark–recapture results showed female-biased natal dispersal in the blue tit populations. As suggested for most bird species, the strong male philopatry is probably related to the greater role in territory and resource defence of this sex, which may particularly benefit from being in a familiar locality and settling in natal areas (Greenwood 1980). This female-biased natal dispersal is partially reflected by the observed patterns of genetic structure and gene flow. Despite lower sample sizes in males, some pairwise F_{ST} values were significant in males but none were in females (Table 2). Similarly, individual-based clustering analyses also showed a higher admixed ancestry in females and a significant genetic subdivision between Quintos de Mora and Cabañeros in males that is completely absent in females (Figs 3, 4). Finally, males showed a significant isolation-by-distance pattern of genetic structure within Quintos de Mora locality, which was not present when only female genotypes were considered. Thus, the genotypic results also showed that the observed male sex-biased dispersal is also reflected by stronger genetic structure in the most dispersive sex (Pierson et al. 2010).

Overall, this study shows that there is concordance between dispersal and the observed patterns of genetic structure, indicating that results from capture–mark–recapture studies can provide useful information on the contemporary levels of gene flow present in natural populations (Coulon et al. 2008; Pierson et al. 2010). Future landscape genetic approaches would further contribute to identifying the barriers limiting gene flow in this and other organisms inhabiting highly fragmented landscapes (Smith et al. 1997, 2005; Cushman et al. 2006).

Acknowledgments

We thank Pedro J. Cordero (Instituto de Investigación en Recursos Cinegéticos, IREC) for allowing us to carry out the genetic analyses in his laboratory. Two anonymous referees provided useful discussion and valuable comments on the manuscript. We thank the board of the Cabañeros National Park and Centro Quintos de Mora for the facilities offered during the fieldwork. During this work V.G.N. and J.O. were supported by respective predoctoral FPI (MEC) and postdoctoral JAE-Doc (CSIC) fellowships. This study was funded by the grants 69/2003 (Organismo Autónomo de Parques Nacionales-Ministerio de Medio Ambiente), GCL2007-61395 (Ministerio de Educación y Ciencia) and POIC10-0269-7632 (Junta de Comunidades de Castilla-La Mancha-European Social Fund).

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