

Songbird genetic diversity is lower in anthropogenically versus naturally fragmented landscapes

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Abstract Natural habitats, and the populations they sustain, are becoming increasingly fragmented by human activities. Parallels between ‘true’ islands and ‘habitat’ islands suggest that standing levels of individual genetic diversity in naturally fragmented populations may predict the genetic fate of their anthropogenically fragmented counterparts, but this hypothesis remains largely untested. We compared neutral-locus genetic diversity of individual song sparrows (*Melospiza melodia*) breeding in a naturally fragmented landscape (small coastal islands) to that of song sparrows in similar-sized ‘urban islands’ separated by roads and housing developments rather than by water. Individuals on coastal islands were more heterozygous and less inbred than those in urban islands. Estimates of population genetic structuring (assessed by pairwise genetic differentiation and Bayesian clustering methods) and contemporary dispersal (based on assignment tests) revealed little structure within either landscape, suggesting that lack of connectivity at the geographic scale we investigated cannot explain the reduced heterozygosity of urban birds. However, within-site genetic similarity was higher in the urban than the coastal landscape. Assuming that historic genetic diversity was similar in these two environments, our findings suggest that anthropogenically fragmented

populations may lose genetic diversity faster than their naturally fragmented counterparts.

Keywords Heterozygosity · Habitat fragmentation · Islands · Effective population size · Songbirds

With human activity increasing the fragmentation of natural habitats and isolating the populations they contain (Balmford et al. 2003), understanding the factors that maintain or erode genetic variation in fragmented populations is key to their conservation. Clear parallels exist between ‘habitat islands’ becoming fragmented by habitat alteration, and physical islands separated by water (MacArthur and Wilson 1967). Both are associated with reduced dispersal among, and reduced genetic diversity within, sites (Frankham 1997; Wilson et al. 2009; Blanchet et al. 2010) relative to landscapes in which habitat is continuous. Thus, naturally fragmented metapopulations such as the inhabitants of small coastal islands may serve as models with which to predict the evolutionary trajectories of populations currently becoming fragmented. In particular, assuming similar sizes of and distances between fragments and similar historic genetic diversity, heterozygosity in longstanding naturally fragmented populations could represent a ‘worst case’ level to which populations currently experiencing anthropogenic fragmentation will eventually decline.

Thanks to a substantial body of theoretical work on the genetics of subdivided populations, most empirical studies addressing the genetic implications of habitat fragmentation have focused on the reduced size and increased isolation of habitat patches (Walker et al. 2008). However, anthropogenic fragmentation also entails an increased ratio of habitat edge to interior, and decreased habitat quality

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(Keyghobadi 2007). The genetic consequences of these latter two aspects of fragmentation have largely been overlooked (Keyghobadi 2007; Walker et al. 2008). The almost exclusive emphasis on area and isolation effects is particularly surprising in relation to studies on birds (e.g. Brown et al. 2004; Croteau et al. 2007; Barnett et al. 2008; Wilson et al. 2009), as most research concerning fragmentation-induced declines in avian species richness and abundance has implicated increased nest predation and brood parasitism, and decreased food availability, i.e. edge and habitat quality effects (Stratford and Robinson 2005). Such demographic processes occurring within habitat fragments may vary across landscapes and affect patterns of genetic diversity. In birds, for example, nest predation causing complete nest failure can remove entire clutches or broods of nestlings from the pool of potential recruits. The resultant increase in reproductive skew can have profound effects on patterns of relatedness within sites (Hatchwell 2009). Differences in colonization history, recent changes in population size (Cornuet and Luikart 1996), and preferential settling of genetically diverse individuals in particular habitats (Porlier et al. 2009) can also affect the way in which genetic variation is distributed across landscapes. Thus, the idea that genetic diversity of anthropogenically fragmented populations should not decline below that of populations inhabiting comparable naturally fragmented habitats remains essentially untested.

Here we compare individual genetic diversity of song sparrows (*Melospiza melodia*) inhabiting either small coastal islands, representing a naturally fragmented landscape, or similar-sized ‘urban islands’ on the adjacent mainland, where habitat fragments of parkland are separated by a matrix of roads and housing developments rather than by open water. These two landscapes are similar in patch (‘island’) size, census sizes and inter-patch distances, but differ dramatically in levels of predator pressure. Coastal-island sites have lower diversity and abundance of predators, and higher nesting success of sparrows, than their urban-island counterparts (Zanette et al. 2003, 2006a, b). We also explore several potential mechanisms that might generate landscape differences in heterozygosity. First, we compare genetic differentiation between sites and estimates of contemporary dispersal in coastal versus urban habitats, to test the hypothesis that these landscapes differ in connectivity. Second, we compare observed heterozygosity in each landscape to that expected under mutation-drift equilibrium, to address the possibility that recent population declines may have transiently elevated heterozygosity in one or both landscapes (Cornuet and Luikart 1996). Third, we compare the genetic similarity of mated pairs (relative to all other potential mates) at coastal- versus urban-island sites, to test the hypothesis that birds in one landscape may be less able to identify and avoid mating with close

relatives. Finally, we compare overall patterns of genetic similarity within coastal- versus urban-island sites, to test the hypothesis that within-patch processes might affect overall genetic similarity differently in different landscapes.

Methods

Study species and sites

Song sparrows are small, socially monogamous songbirds that remain abundant throughout North America. This continent-wide distribution, together with a large number of proposed subspecies, makes them a model species with which to examine patterns of genetic variation (e.g. Zink and Dittmann 1993; Pruett et al. 2008; Wilson et al. 2009). In southwestern British Columbia, song sparrows (subspecies *morphna*) are generally sedentary. Although natal philopatry varies substantially among populations based on local demography, adult philopatry is generally high (Wilson and Arcese 2008). The importance of individual genetic diversity to fitness has been well established for the nearby Mandarte Island population of song sparrows (e.g. Keller et al. 1994; Reid et al. 2007), located just a few kilometers from our coastal-island study sites.

We measured the neutral-locus genetic diversity of 106 adult song sparrows breeding at a total of fourteen study sites (Fig. 1; Table 1). Coastal-island sites ($n = 7$) were small ($<0.2 \text{ km}^2$) tracts of suitable breeding habitat located on small islands within 2 km of the larger ‘mainland’ of Vancouver Island, Canada ($31,284 \text{ km}^2$). These sites are part of the Gulf Islands National Park Reserve and are not inhabited by humans. Urban sites ($n = 7$) were small ($<0.2 \text{ km}^2$) tracts of suitable breeding habitat embedded in the urban matrix of Victoria, Canada, within 20 km of the coastal-island sites. Within each landscape, one or more sites were nested within larger habitat fragments such as an island or conservation area (Fig. 1; Table 1).

The coastal- and urban-island sites studied here do not differ on average in habitat structure (Zanette et al. 2003) or in the breeding density or estimated population size of song sparrows (Clinchy et al. 2004; Table 1). Banded song sparrows have been observed to move between coastal-island sites outside of the breeding season (Smith et al. 1996; Reid et al. 2006). As part of a concurrent study (Zanette et al. 2003), three of seven sites in each landscape received supplementary food throughout the 2002 breeding season. This manipulation had no detectable effect on individual genetic diversity (Supplementary Data). Nor is it likely to have affected dispersal between sites, as food supplementation began only after breeding territories were established and no birds were observed to move between sites during the breeding season.

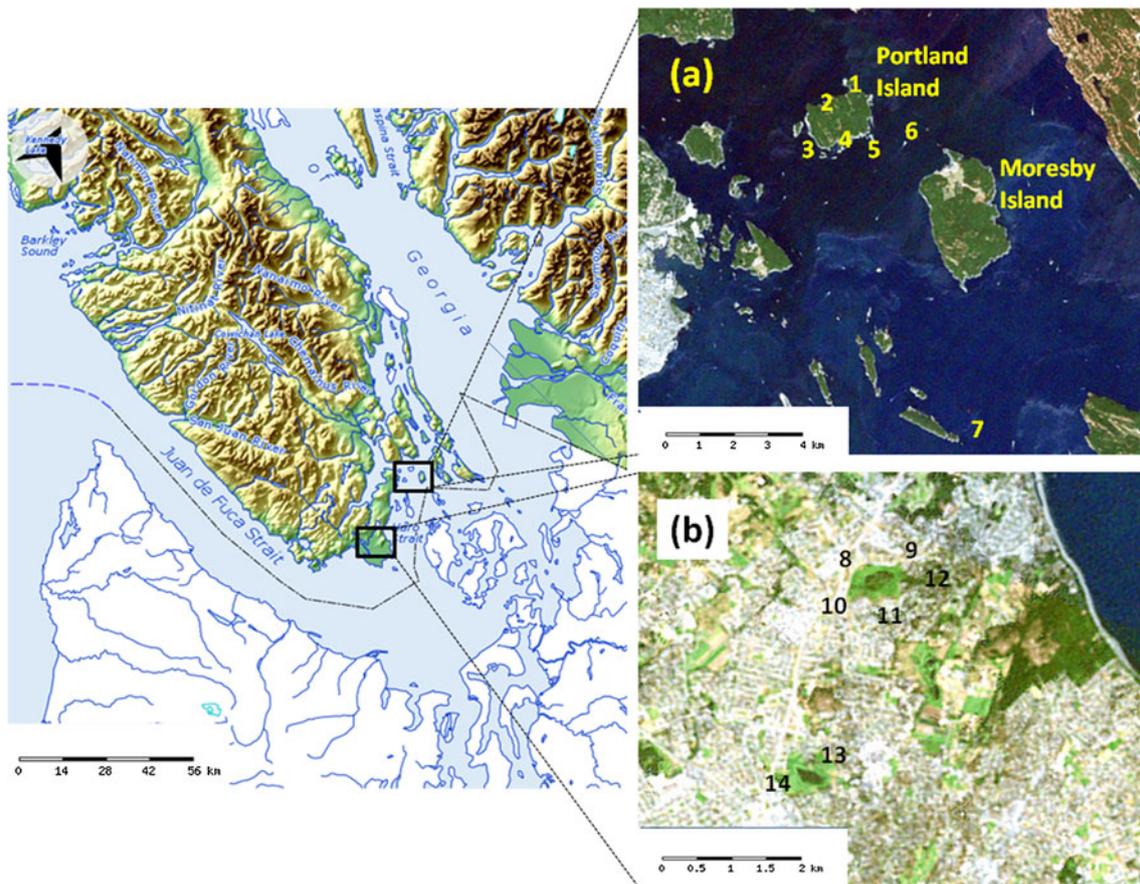


Fig. 1 Map of sampling sites, on Vancouver Island and nearby Gulf Islands, British Columbia, Canada. *Inset (a)* shows coastal island sites (1 Arbutus Point, 2 Royal Cove, 3 Shell Beach, 4 Tortoise Bay, 5

Tortoise Island, 6 Pellow Islets, 7 Rum Island). *Inset (b)* shows urban island sites (8 Chatterton, 9 Dalewood, 10 Emily Carr, 11 Vole Meadow, 12 Fir Tree Glen, 13 Swan Lake East, 14 Swan Lake West)

In spring 2002, we collected blood samples for genetic analysis from 4 to 16 breeding adults per site ($n = 50$ and 56 sparrows, comprising 25 and 28 mated pairs, at coastal and urban-island sites respectively; Table 1). We identified Universal Transverse Mercator (UTM) coordinates in northings and eastings of the geographic centre of each site, using a portable Global Positioning System unit. Pairwise geographic distances between sites were calculated with Geographic Distance Matrix Generator, version 1.2.3 (P. J. Ersts, American Museum of Natural History, Center for Biodiversity and Conservation, http://biodiversityinformatics.amnh.org/open_source/gdmg).

Genotyping methods

We typed all birds at seven microsatellite loci: *Escμ* 1 (Hanotte et al. 1994), *Pdoμ* 5 (Griffith et al. 1999), *Mme* 2 and 7 (Jeffery et al. 2001), and *Sosp* 3, 13 and 14 (L. F. Keller, pers. comm.). Genotyping conditions are detailed elsewhere (Potvin and MacDougall-Shackleton 2009). The microsatellites examined harboured 11–26 alleles per locus, for a total of 129 alleles across all loci. Locus *Mme* 7 is sex-linked

(Jeffery et al. 2001), so females were coded as ‘missing data’ for their second allele at this locus. Thus, estimates of individual genetic diversity (Table 1) were based on seven loci for males and six for females.

We tested for deviations from Hardy–Weinberg equilibrium (HWE) and linkage equilibrium (LE) separately for each landscape, using a Markov chain approach (dememorization = 1,000, 100 batches, 1,000 iterations per batch) in the program GENEPOP 4.0.10 (Rousset 2008) with sequential Bonferroni correction for multiple tests (Rice 1989). We found no evidence for deviations from HWE ($P > 0.10$ for all seven loci) or LE for any pair of loci in either population following Bonferroni correction ($P > 0.03$ for all locus pairs; 42 comparisons).

Individual genetic diversity

For each individual, we calculated multilocus heterozygosity (MLH), defined as the proportion of typed loci at which the individual is heterozygous, using the Excel macro IRmacroN4 (W. Amos; <http://www.zoo.cam.ac.uk/zoostaff/amos/#ComputerPrograms>). Because MLH was

Table 1 List of study sites, including number of song sparrows genotyped (N), estimated number of breeding adults at site (N_{est}), MLH and inbreeding coefficient (f)

Site name	N (male, female)	N_{est}	MLH	f
Coastal-island sites				
Portland Island				
Arbutus Point (AP)	6 (3, 3)	9 (5, 4)	0.95 ± 0.03	0.02 ± 0.01
Royal Cove (RC)	12 (6, 6)	14 (7, 7)	0.89 ± 0.03	0.06 ± 0.03
Shell Beach (SB)	10 (5, 5)	14 (7, 7)	0.83 ± 0.05	0.12 ± 0.05
Tortoise Bay (TB)	4 (2, 2)	18 (9, 9)	0.93 ± 0.05	0.05 ± 0.03
Tortoise Island (TI)	8 (4, 4)	14 (7, 7)	0.88 ± 0.07	0.08 ± 0.05
Pellow Islets (PELL)	4 (2, 2)	8 (4, 4)	0.89 ± 0.07	0.04 ± 0.03
Rum Island				
Rum Island (RUM)	6 (3, 3)	10 (5, 5)	0.90 ± 0.03	0.06 ± 0.02
Urban-island sites				
Rithet's Bog				
Chatterton (CHAT)	16 (8, 8)	20 (10, 10)	0.78 ± 0.05	0.14 ± 0.04
Dalewood (DALE)	8 (4, 4)	16 (8, 8)	0.83 ± 0.05	0.10 ± 0.05
Emily Carr (EC)	6 (3, 3)	17 (9, 8)	0.84 ± 0.06	0.10 ± 0.04
Vole Meadow (VM)	6 (3, 3)	14 (7, 7)	0.85 ± 0.05	0.08 ± 0.03
Fir Tree Glen (FTG)	8 (4, 4)	14 (7, 7)	0.79 ± 0.06	0.14 ± 0.07
Swan Lake				
Swan Lake East (SLE)	6 (3, 3)	13 (7, 6)	0.89 ± 0.05	0.06 ± 0.04
Swan Lake West (SLW)	6 (3, 3)	12 (7, 5)	0.80 ± 0.06	0.12 ± 0.05

MLH and f are reported as means \pm SE

not normally distributed, we used a nonparametric Mann–Whitney U test to compare MLH of birds breeding at coastal- versus urban-island sites. As a complementary analysis, we used the dyadic likelihood method (Milligan 2003) implemented in COANCESTRY 1.0 (Wang 2011) to estimate individual inbreeding coefficients (f) from microsatellite genotypes. We generated 95% confidence intervals for each landscape based on 1,000,000 bootstrap replicates to compare inbreeding coefficients of coastal-versus urban-island birds.

Population genetic structure

To investigate population genetic structuring within and between landscapes, we calculated F_{ST} (Weir and Cockerham 1984) between all fourteen sites. We tested for isolation by distance over the entire study area by comparing all pairwise values of $(F_{\text{ST}}/1 - F_{\text{ST}})$ to the natural log of geographic distance (Rousset 1997), using 10,000 permutations in the subprogram ISOLDE implemented in GENEPOP 4.0 (Rousset 2008). We then assessed the significance of genetic differentiation between all site pairs within a landscape, separately for each landscape, using the exact G -test implemented in GENEPOP. Markov chain parameters included a dememorization number of 1,000, 100 batches, and 1,000 iterations per batch.

We used general linear model regression (GLM) to quantify the effects of geographic distance and landscape type on genetic differentiation $(F_{\text{ST}}/1 - F_{\text{ST}})$ between pairs

of sites within each landscape. Because the farthest-apart coastal island sites were separated by greater geographic distances (about 10 km) than the farthest-apart urban island sites (about 3.7 km), we also conducted a second analysis on the subset of site pairs separated by less than 4 km.

We used Bayesian model-based clustering analysis implemented in STRUCTURE 2.3.3 (Falush et al. 2003) to further compare genetic structuring within the coastal-versus the urban-island landscape. To estimate the optimal number of clusters of individuals (K), we used the admixture and correlated allele frequency models as recommended by the authors. Sampling location was used as prior information to assist clustering and maximize our ability to detect subtle genetic structure (Hubisz et al. 2009). We first tested models $K = 1$ through 14 on the entire dataset to examine genetic differentiation between the two landscapes, then assessed differentiation within each landscape separately by testing $K = 1$ through 7. Running conditions included an initial burn-in period of 50,000 iterations followed by a run length of 100,000. Posterior probability for each value of K was calculated using ln-likelihood scores (Falush et al. 2003). To confirm consistency between trials, we performed five runs for each value of K .

Estimating contemporary dispersal

We used assignment tests to compare levels of contemporary dispersal within coastal versus urban-island

landscapes. This analysis included all birds from all sites, but in light of low structure within landscapes (see below) we pooled all sites within the same habitat fragment (i.e. Portland Island; Rum Island; Rithet's Bog; Swan Lake; Table 1). We used the criterion of Rannala and Mountain (1997) implemented in GeneClass 2.0.h (Piry et al. 2004) to estimate the likelihood of each genotype originating from the habitat fragment in which it was sampled (L_Home; Piry et al. 2004), as some potential source populations likely remained unsampled. We used the algorithm of Paetkau et al. (2004) to conduct Monte Carlo resampling over 1,000 simulated individuals, with a threshold detection probability of $\alpha = 0.05$ to identify first-generation migrants.

Bottleneck analysis

Changes in effective population size can result in a temporary deficit or excess of heterozygosity relative to that predicted from the observed number of alleles, due to the loss of rare alleles in recently bottlenecked populations (Cornuet and Luikart 1996). We used BOTTLENECK 1.2.02 (Cornuet and Luikart 1996) to investigate evidence for recent population declines in each landscape. For this analysis, sites were combined such that each landscape was treated as a single population. We used the two-phase mutation model (TPM; Di Rienzo et al. 1994) to generate distributions expected under mutation-drift equilibrium, with the default settings of 30% multi-step mutations and 1,000 replications. We used the sign test and allele frequency mode shift approaches implemented in BOTTLENECK 1.2.02 to determine whether either landscape showed evidence of recent change in population size.

Genetic similarity between mates and within sites

We calculated Lynch and Ritland's (1999) pairwise coefficient of genetic similarity, r , between all individuals breeding at the same site, using the program MARK (K. Ritland; www.genetics.forestry.ubc.ca/ritland/programs.html). The results reported below were calculated based on locally observed allele frequencies for each site, but findings were qualitatively similar when r was calculated based on allele frequencies across all sites.

To compare patterns of social mate choice with respect to genetic similarity in coastal- versus urban-island landscapes, we compared each individual's coefficient of genetic similarity to its observed mate (r_M) to its mean similarity to all other potential mates (r_P). 'Potential mates' were defined as all adults of the opposite sex, other than the observed mate, breeding at the same site. 'Relative inbreeding' was defined for each individual as $r_M - r_P$; a negative value denotes an individual that was genetically

less similar to its mate than to the set of other potential mates. We examined the effects of landscape type on inbreeding avoidance separately for each sex, using a t -test to compare $r_M - r_P$ for individuals at coastal- versus urban-island sites.

To examine overall genetic similarity within coastal-versus urban-island subpopulations, we used a t -test to compare values of r for all pairwise combinations of individuals (regardless of sex) breeding at the same site. Values of r were normally distributed (Kolmogorov–Smirnov test), as were values derived from r (r_M , r_P , $r_M - r_P$).

Results

Individual genetic diversity

Individual genetic diversity was higher in the coastal-island than the urban-island landscape. Relative to their coastal-island counterparts, sparrows inhabiting urban islands had lower MLH (mean \pm SE = 0.886 ± 0.019 and 0.816 ± 0.022 for coastal- and urban-island birds respectively; Mann–Whitney adjusted $Z_{104} = 2.52$, $P = 0.012$) and higher inbreeding coefficients (mean \pm SE = 0.067 ± 0.037 and 0.114 ± 0.018 for coastal- and urban-island birds respectively). Bootstrap analysis indicated that this difference in f between the two landscapes exceeded 95% confidence limits (97.5% quantile = 0.046, observed difference = 0.047).

Population genetic structure

Mantel testing revealed a positive correlation between geographic (ln distance) and genetic ($F_{ST}/1 - F_{ST}$) distance matrices, suggesting isolation by distance, when all fourteen sites were included in the analysis (one-tailed $P = 0.003$). At a finer spatial scale, genetic differentiation between pairs of sites within the same landscape increased with geographic distance (GLM: $F_{1,39} = 18.24$, $P = 0.0001$) but was not related to landscape type ($F_{1,39} = 0.41$, $P = 0.52$; Fig. 2). These patterns remained consistent when we considered only the subset of pairwise comparisons within 4 km: genetic distance increased with increasing geographic distance ($F_{1,33} = 7.14$, $P = 0.012$), consistent with isolation by distance, but did not vary with landscape type ($F_{1,33} = 0.13$, $P = 0.73$). Exact tests for genetic differentiation between all site pairs within each landscape identified five of 21 pairs of coastal-island sites (Table 2) and one of 21 pairs of urban-island sites (Table 3), as being significantly differentiated. By contrast, testing for differentiation between sites in different landscapes (and thus separated by greater geographic distances)

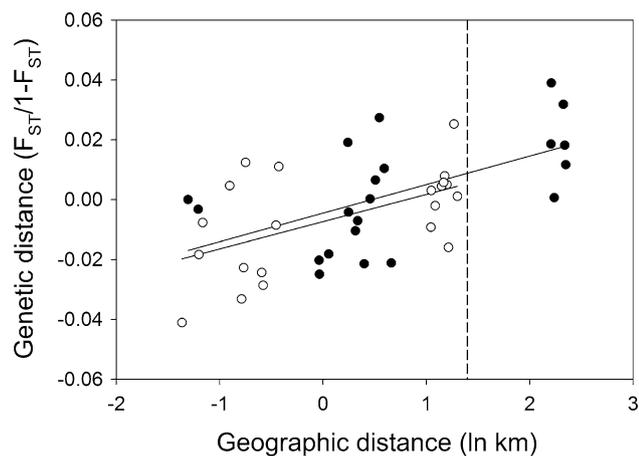


Fig. 2 Genetic distance ($F_{ST}/1 - F_{ST}$) as a function of geographic distance (ln km) between pairs of coastal- and urban-island sites (filled and open circles, respectively). Solid lines indicate simple linear regressions. The subset of points to the left of the dashed vertical line (pairwise geographic distance < 4 km) were also analyzed separately

Table 2 Pairwise matrix of geographic distance (km; above diagonal) and genetic differentiation (F_{ST} ; below diagonal) between coastal-island sites

Site	AP	RC	SB	TB	TI	PELL	RUM
AP	–	0.27	1.81	1.49	1.72	1.27	10.37
RC	0.000	–	1.58	1.40	1.66	1.37	10.47
SB	0.010*	0.000	–	1.06	1.28	1.94	10.22
TB	–0.022	–0.007	–0.018	–	0.30	0.97	9.36
TI	0.027*	0.006	–0.004	–0.003	–	0.96	9.07
PELL	0.019	–0.011	0.010*	–0.026	–0.021	–	9.10
RUM	0.018	0.012	0.031*	0.001	0.018	0.038*	–

Asterisk indicates a significant difference in allele frequencies as determined by an exact G -test

identified 20 of 44 site pairs as being significantly genetically differentiated at $\alpha = 0.05$; pairwise estimates of F_{ST} ranged from 0 to 0.08 for these coastal–urban comparisons.

Bayesian analysis detected significant genetic differentiation between the coastal-island and urban-island landscapes, but not finer scale genetic structuring within either landscape. For the combined dataset, STRUCTURE identified an optimal value of $K = 2$ (posterior probability = 0.82–1), corresponding to our coastal- versus urban-island sites, for four of five runs and $K = 3$ (posterior probability = 0.97) for one run. Within the coastal-island landscape, STRUCTURE grouped all individuals together ($K = 1$) with posterior probabilities ranging from 0.44 to 0.95 for four of five runs, and identified two clusters of individuals (posterior probability = 0.87) for one run. Similarly, within the urban-island landscape, STRUCTURE grouped all individuals together for four runs with a

Table 3 Pairwise matrix of geographic distance (km; above diagonal) and genetic differentiation (F_{ST} ; below diagonal) between urban-island sites

Site	CHAT	DALE	EC	VM	FTG	SLE	SLW
CHAT	–	0.31	0.41	0.47	0.63	3.24	3.55
DALE	–0.008	–	0.56	0.46	0.45	3.32	3.67
EC	0.005	–0.029	–	0.26	0.55	2.84	3.15
VM	0.012	0.012	–0.043	–	0.3	2.85	3.21
FTG	–0.009	–0.034	–0.025	–0.019	–	2.96	3.37
SLE	0.008	0.005	–0.009	0.003	–0.002	–	0.65
SLW	0.025*	0.001	0.004	0.006	–0.016	0.011	–

Asterisk indicates a significant difference in allele frequencies as determined by an exact G -test

posterior probability of 1, and identified two clusters of individuals (posterior probability = 0.73) for one run. Pooling sites within habitat fragments had little effect on our estimates of structuring within each landscape, with STRUCTURE grouping all individuals together ($K = 1$; posterior probabilities for all five runs = 0.67–1 and 1 for coastal-island and urban-island landscapes respectively).

Estimating contemporary dispersal

Assignment tests identified eight of 106 sparrows (7.5%) as originating from a habitat fragment other than the one in which they were sampled. Of these eight apparent migrants, four were sampled in the coastal-island and four in the urban-island habitat.

Bottleneck analysis

We found no evidence of heterozygosity excess that would indicate recent population declines. In each landscape BOTTLENECK identified two of seven loci as heterozygosity-excessive, not significantly different from the values expected under mutation-drift equilibrium (4.10 and 4.22 for coastal- and urban-island birds respectively; sign test: $P = 0.39$ and 0.09 respectively). Similarly, BOTTLENECK identified the L-shaped distribution of allele frequencies expected under mutation-drift equilibrium for each landscape.

Genetic similarity between mates and within sites

Relative inbreeding ($r_M - r_P$) did not differ between coastal- and urban-island sites, for either males (mean \pm SE = 0.026 ± 0.021 and 0.020 ± 0.009 respectively; $t_{51} = 0.315$, $P = 0.75$) or females (mean \pm SE = 0.027 ± 0.022 and 0.018 ± 0.012 respectively; $t_{51} = 0.347$, $P = 0.73$). Relative inbreeding was slightly greater than zero for males (one-sample t -test; $t_{52} = 2.09$, $P = 0.041$),

suggesting some tendency for social mates to be more similar genetically than expected under random mating, but this difference was not significant for females ($t_{52} = 1.86$, $P = 0.069$).

Overall genetic similarity (r ; Lynch and Ritland 1999) of all adults within a site was significantly lower in coastal- than in urban-island sites (mean \pm SE = -0.043 ± 0.004 and -0.028 ± 0.003 respectively; $t_{754} = 3.19$, $P = 0.002$).

Discussion

If naturally fragmented populations provide a baseline estimate of genetic diversity to which populations in a comparatively recently human-altered habitat will eventually decline, then all other things being equal, birds in the urban matrix should be at least as heterozygous as their counterparts in longstanding naturally fragmented (coastal island) populations. Instead, song sparrows on coastal islands were more genetically diverse than were song sparrows at urban islands. The size and physical isolation of islands are known to influence population-level genetic variation for various species, including song sparrows (Wilson et al. 2009). However, we think it unlikely that these factors explain our observed differences between the coastal- and urban-island habitats, because both census sizes (Clinchy et al. 2004; Zanette et al. 2006a, b) and physical distances between sites and habitat fragments (Fig. 1) are comparable between these two landscapes. Instead, the relatively low genetic diversity at urban islands might result from low dispersal among habitat patches in the anthropogenically fragmented landscape, from differences in population history and/or from demographic processes occurring within these patches.

It is possible that some aspect of the urban landscape inhibits dispersal among sites, more so than flight over open water. Highways, for example, are known to restrict dispersal even of large mammals (Riley et al. 2006); although birds are clearly capable of crossing roads, these may still represent a behavioural barrier to dispersal (Lynch and Whigham 1984). However, we found little evidence of genetic differentiation within either landscape, at least at the spatial scale investigated here. Moreover, assignment tests suggested similar levels of contemporary dispersal in the two landscapes. Although our finding of isolation by distance suggests acceptable power to detect population genetic structuring, we emphasize that small sample sizes per site, and relatively low distances among sites and habitat fragments, prevent us from conclusively ruling out landscape differences in connectivity. However, our results suggest that low individual genetic diversity at urban sites cannot easily be attributed to lack of gene flow.

Although census sizes and degree of fragmentation in the coastal- and urban-island landscapes are currently similar, differences in population history may contribute to the reduced individual genetic diversity of urban birds. While we found no evidence for recent genetic bottlenecks in either landscape, it should be noted that our power to detect such events is limited by the number of loci used and their repeat type (Cristecu et al. 2010). Importantly, the matrix of roads and housing developments that now separates urban sites is of recent origin (<200 years), relative to how long oceanic sites have been separated. Thus, song sparrows in the urban matrix may still be in the process of losing heterozygosity and increasing genetic differentiation. In fact, these two effects of habitat fragmentation can proceed at different rates (Keyghobadi et al. 2005). However, differences in the speed of these processes appear unlikely to explain our findings of reduced heterozygosity, without increased differentiation, in the urban landscape. On the contrary, in recently-fragmented landscapes, genetic differentiation among sites tends to occur first, before substantial loss of heterozygosity (Keyghobadi et al. 2005); this is opposite to the pattern we observed. Despite the fact that reductions in genetic diversity are already apparent, urban populations of song sparrows may thus still be in the early stages of responses to fragmentation, suggesting that genetic diversity may continue to erode within this landscape.

In addition to examining potential differences in dispersal through naturally versus anthropogenically fragmented landscapes, we also used patterns of genetic similarity to investigate potential demographic differences occurring within each type of site. Relative to other potential mates within the site, socially mated pairs within the urban landscape were not more genetically similar to one another than social mated pairs within the oceanic landscape. Thus, low individual genetic diversity of urban song sparrows seems not to be explained by differences in mate choice or a reduced ability to detect and avoid mating with relatives, at least when selecting social mating partners. Consistent with this, previous research on a nearby population of coastal-island song sparrows (Mandarte Island) has shown little or no evidence for active inbreeding avoidance in selecting social mates (Keller and Arcese 1998) or in shaping extra-pair mating strategies (Reid et al. 2007). Landscape differences in mating strategies could potentially enhance variance in reproductive success and thereby reduce genetic variation within urban islands. However, in other nearby populations of *M. m. morphna*, extra-pair paternity levels are not lower at coastal- than at urban-island sites (Mandarte Island, 28% of offspring, O'Connor et al. 2006; Seattle, 24% of offspring; Hill et al. 2011). Moreover, estimates of reproductive skew on Mandarte Island are similar regardless of whether

genetic or social mating data are used (O'Connor et al. 2006). Thus, differences between naturally and anthropogenically fragmented populations in either mate choice or mating systems seem unlikely to explain the relatively low genetic diversity we observed in urban birds.

Despite no detectable difference between coastal- and urban-island sparrows in inbreeding avoidance, genetic similarity between all birds within a site (and thus, between potential mates) was higher at urban-island sites. This pattern helps explain why individual genetic diversity is lower at urban-island than coastal-island sites, but in turn, it raises the question of why urban-island sparrows show this pattern of genetic similarity within sites. Recent attention to population differences in demography, and their consequences for generating kin-structured populations (Hatchwell 2009), casts light on this matter. Although the coastal- and urban-island song sparrows studied here have similar census population sizes, and apparently comparable ability to move between habitat fragments, predation on eggs and nestlings is much more frequent in urban- than in coastal-island sites (Zanette et al. 2003, 2006a, b). By removing complete clutches or broods of nestlings from the pool of potential recruits in an all-or none fashion, nest predators increase variation in reproductive success, and can thus influence patterns of genetic variation within sites (Hatchwell 2009). Our findings are consistent with this idea because all-or-nothing patterns of mortality within the urban landscape may increase overall genetic similarity within sites, and thereby erode genetic diversity. Song sparrows do attempt multiple nests each season, but if territories vary in their susceptibility to nest predation, renesting may do little to reduce variation in reproductive success. Moreover, brood parasitism by brown-headed cowbirds *Moluthrus ater* occurs more often in urban- than coastal-island sites (Zanette et al. 2003). Because female nestlings are less able than their brothers to survive in the presence of a cowbird parasite, parasitized broods produce strongly male-biased sex ratios at fledging (Zanette et al. 2005) which could further reduce the effective population sizes of urban-island sparrows.

Other ecological differences between naturally- and anthropogenically-fragmented landscapes may also contribute to the observed difference in individual genetic diversity. For example, heterozygote advantage is less effective in maintaining genetic diversity when most mortality results from factors other than disease (Hoffman et al. 2006) as appears likely at urban-island sites. Competition for preferred habitats can also result in a non-random distribution of individual genetic diversity, if relatively inbred individuals are disproportionately relegated to a particular habitat type (Porlier et al. 2009).

Although naturally- and anthropogenically-fragmented populations differ in individual genetic diversity, it remains

to be seen whether this difference is biologically significant. We observed an average difference in inbreeding coefficient (f) of 0.047 between the coastal- and urban-island habitats, comparable to the difference in f between song sparrows that survived a winter storm and those that did not on nearby Mandarte Island (0.025; Keller et al. 1994). However, that study used pedigree analysis rather than microsatellite genotypes in calculating f , restricting our ability to directly compare effect sizes. Another important caveat is that our study examined only neutral-locus genetic variation. Whether naturally- and anthropogenically-fragmented populations also differ in terms of adaptive genetic variation, and if so, how this affects their evolutionary potential of their inhabitants, remains an open question. Overall, however, our findings suggest that anthropogenically fragmented songbird populations are not analogues of their naturally fragmented counterparts. Despite relatively recent fragmentation of the urban landscape, individuals in this habitat already harbour less genetic variation than do coastal-island birds. More generally, our findings emphasize the importance of demographic processes operating within patches, in addition to movement between patches, in shaping the distribution of genetic variation over landscapes.

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