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Genetic variability in island populations of two rodent species: bank vole (*Myodes glareolus*) and yellow-necked mouse (*Apodemus flavicollis*)

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We examined changes in genetic diversity in populations of two small mammal species inhabiting islands on a lake. We hypothesized that a less mobile species, such as the bank vole, would lose genetic diversity quicker than the yellow-necked mouse, which can more easily cross habitat barriers. In contrast to this we found that the effects of isolation were much more prominent in the case of the mouse than the vole. In the vole population, on the larger island, genetic diversity remained constant over subsequent years in spite of marked isolation. On the smaller island, we noted an increase in genetic diversity which was probably caused by immigration of a small group of individuals. Genetic diversity in the mouse population decreased markedly over the course of the study. In the bank vole, the preservation of genetic diversity, especially on the larger island, may have been possible due to the specific spatial and social organization of this species, which allows the maintenance of a relatively stable number of individuals. In contrast to the bank vole, the lack of territorial behavior in adult females of yellow-necked mice may lead to frequent dramatic seasonal 'booms and busts' in population size and genetic diversity. This can lead to extremely low numbers of mice, which are likely to lead to the extinction of some island populations.

Introduction

Small island populations are often more likely than others to go extinct due to low levels of genetic diversity (Smith *at al*. 1993, Frankham 1997, Kozakiewicz *et al*. 2009). Low genetic diversity is usually a result of colonization of an island by a small number of individuals (the founder effect) (Ryan *et al*. 1996, Hirota *et al*. 2004, Ratkiewicz & Borkowska 2006), along

with further elimination of alleles due to random processes such as genetic drift. Immigration of a sufficient number of new individuals or mutations could theoretically overcome the loss of genetic diversity, but under natural conditions in island populations immigration of individuals from outside or mutations are likely to be too rare to save the population (Jeanike 1973, Bauchau & le Boulengé 1991, Saunders *et al*. 1991). Isolated but large populations with high reproductive rates are, on the other hand, able to maintain genetic diversity for a long period of time (Soulé 1976, Frankham 1996). In small populations however, even a small decrease in the number of individuals may quickly and permanently limit the genetic pool (Freeland 2005). It is expected that changes in the gene pool should be much faster and deeper in populations inhabiting small remote islands than on islands situated close to the mainland or other islands (Jeanike 1973).

In several studies it has been found that both natural and anthropogenic barriers may lead to the formation of differences in the genetic structure of populations of small rodents (e.g., Gerlach & Muslof 2000, Kozakiewicz *et al*. 2009, Gortat *et al*. 2010, 2013). The following studies, among others, have demonstrated much lower genetic diversity in island populations of small mammals than in mainland ones: Berry and Peters (1997) in the house mouse (*Mus musculus*), Gill (1980) and Aquadro and Kilpatrick (1981) in the deer mouse (*Peromyscus maniculatus*), Schmitt (1978) in the bush rat (*Rattus fuscipes*) and Stewart and Baker (1992) in the masked shrew (*Sorex cinereus*). In some small rodents, over a longer time span, evidence indicates that isolation may lead to microevolution of island populations (Yom-Tov & Moller 1999, Pergams & Ashley 2001).

Durability, intensity and depth of changes in the genetic structure of a population are connected with parameters of the habitat, such as size or degree of isolation, and also with the biology of a given species (Kilpatrick 1981, Lande 1988). It is generally believed that preservation of genetic diversity in a population largely depends on the mobility of individual animals, as well as their ability to cross environmental barriers. In other words, populations of highly mobile animals which are able to cross ecological barriers are more likely to maintain genetic diversity (Jeanike 1973). Accordingly, for species with low mobility, ecological barriers and the resulting isolation of local populations may lead to inbreeding, and to further increase in the genetic differences between isolated populations (Rutkowski *et al*. 2008).

We investigated island populations of the bank vole (*Myodes glaerolus*) and the yellow-necked mouse (*Apodemus flavicollis*). Both species are common in central Europe, usually inhabiting woodland areas and small patches of forest (Pucek 1983). The breeding period in both species lasts from spring to the end of summer or early autumn. The bank vole, however, is less mobile and has smaller home ranges than the larger and wider-ranging yellow-necked mouse (Wolton 1985, Bąkowski & Kozakiewicz 1988, Szacki 1999, Grüm & Bujalska 2000).

A number of papers have been written on the functioning of populations of the yellow-necked mouse and the bank vole in spatially fragmented environments, but only a small number of them are concerned with the underlying genetic structure of these populations (e.g. Aars *et al*. 1998, Redeker *et al*. 2006). Even fewer of those works discuss the genetic structure of island populations of the bank vole and the yellow-necked mouse (e.g. Corbet 1964, Kozakiewicz *et al*. 2009, Gortat *et al*. 2010). In both of these species, seasonal changes in population size may be accompanied by a loss of genetic diversity due to the bottleneck effect. Genetic diversity in such cases can usually only be regained by the dispersal of individuals. Thus, one might expect that in small, highly-isolated populations inhabiting lake islands, genetic structure should be largely dependent on local processes of reproduction and mortality, with immigration and emigration of individuals playing a less important role. However, due to the yellow-necked mouse's higher mobility and greater readiness to cross habitat barriers than the bank vole, we hypothesized that loss of genetic diversity in the former species, resulting from seasonal declines in population numbers, may be more effectively compensated by immigration.

Material and methods

The study area comprised two forested islands located in Lake Mokre in northeastern Poland (21°E, 53°N), within the borders of the Mazury Landscape Park. The larger of the two, Mouse Island (MI), covers approximately 3 ha and is located 750 m and 375 m from the western and eastern lakes hores, respectively (Fig. 1). The smaller Shrew Island (SI), covers 1.5 ha and is located 275 m from the western lake shore and about 500 m from the eastern one. The distance between the islands is 525 m. MI is located a few meters above the water level, whereas SI is periodically partly flooded in spring due to the fluctuating water level. The lake is surrounded by forest.

The study was carried out during the summers of 2003, 2004 and 2008, during the breeding season in the second half of July. In total, three 10-day trapping series were conducted on each island. We used the standard CMR method (Catch–Mark–Release) to trap the rodents. Live traps were situated every 20 m along designated transects. On each island, 60 traps (two per point) were placed at 30 trapping points. Each capture animal was classified to species, and the capture location was recorded. A small fragment of an ear lobe was collected from each captured individual to be used later in genetic analysis. In total we caught 441 bank voles and 102 yellow-necked mice. In 2003 and 2004 on MI we repeatedly captured 9 bank vole individuals and 11 yellow-necked mice individuals. While on SI no bank vole was captured repeatedly in these years. Yellow-necked mice were absent from SI over the course of the entire study period.

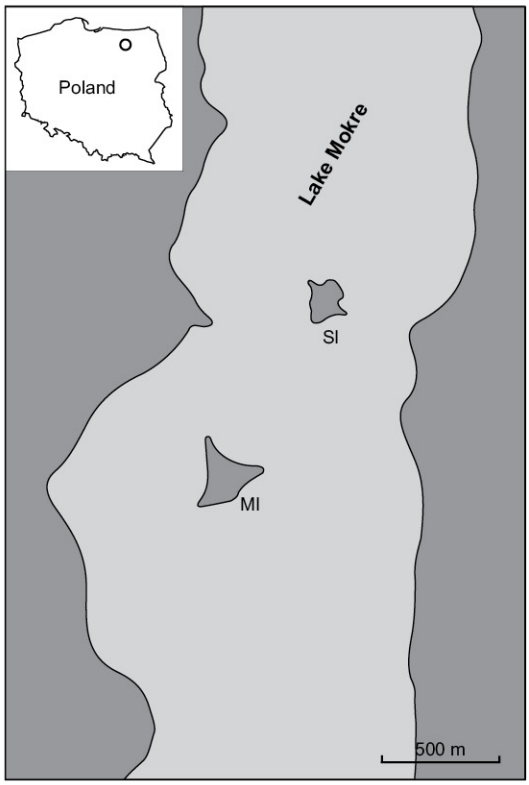

Fig. 1. The study lake. MI and SI are Mouse Island and Shrew Island, respectively.

Genetic analysis

Genomic DNA was successfully extracted from the tissues of ear lobes of 437 bank voles and 100 yellow-necked mice using the DNA Isolation Tissue Kit (A&A Biotechnology), as described in Kozakiewicz *et al*. (2009). The DNA concentration in the isolates was measured using a spectrophotometer (ND-1000, Nanodrop Technologies). The concentrations were on average 50 ng μl^{-1}.

We amplified seven microsatellite loci developed for bank voles: MSCg-4, MSCg-6, MSCg-7, MSCg-9, MSCg-24, LIST3-003, LIST3-007 (Gockel *et al*. 1997, Gerlach & Musolf, 2000, Barker *et al*. 2005) (Appendix 1). For this, we used the multiplex PCR Kit (Qiagen) under the following conditions: initial denaturation: 15 min. at 95 °C; 30 cycles: 30 sec. at 94 °C, 90 sec. at 60 °C and 60 sec. at 72 °C; final elongation: 60 min. at 72 °C. Forward primers were labelled fluorescently on their 5´ ends with NED, FAM or HEX dye. The amplified loci for bank voles were then genotyped with D-filter (ROX as a size standard) using an ABI PRISM 310 automated sequencer (Applied Biosystems). Amplification was scored using a GeneScan Analysis software ver. 3.7.

To genotype the yellow-necked mouse we used five microsatellite loci, described for members of the genus *Apodemus*: GACAD1A, GTTC4A, MSAf-16, MSAA-5, MSAA-6 (Gockel *et al*.

1997, Ohnishi *et al*. 1998, Makova *et al*. 1998) (Appendix 2). As in the case of the bank vole, we used the multiplex PCR method to amplify the markers. The amplification was performed under the following conditions: initial denaturation: 15 min. at 95 °C; 30 cycles: 30 sec. at 94 °C, 3 min. sec. at 57 °C and 60 sec. at 72 °C; final elongation: 15 min. at 68 °C. Forward primers were labelled fluorescently on their 5´ ends as Dye2, Dye3 or Dye4 (Sigma). The length of the amplified fragments was estimated using the CEQ8000 Beckman Coulter automated sequencer. The data were analyzed using Beckman Coulter Fragment Analysis Software.

In all cases, negative PCR controls were included for each set of reactions. Following electrophoresis in agarose gels and analysis in the automatic sequencer, no amplification product was found in any of the negative controls.

Statistical analysis

Relative genetic variation was assessed using allele frequency data. We assessed allelic diversity (A), allelic richness (R) (Petit *et al*. 1998), mean number of private alleles (A_p), private allelic richness (p_R), effective number of alleles (N_e), observed heterozygosity (H_O) and unbiased expected heterozygosity (H_E) (Nei & Roychoudhury 1974) separately for each species, season and island. A fixation index (F_{IS}) was calculated and its significance was tested under a randomization procedure as well as a Bonferroni correction for multiple comparisons. These analyses were performed using GenAlEx ver. 6.0 (Paekal & Smouse 2006), FSTAT ver. 2.9.3.2 (Goudet 2002) and HP-RARE (Kalinowski 2005). Genotypic linkage disequilibrium was evaluated between all pairs of loci, as well as a probability test for deviation from the Hardy-Weinberg equilibrium (HWE), using Genepop on Web ver. 4.0.10 (Raymond & Rousset 1995, Rousset 2008).

In order to investigate the possibility that demographic changes may have affected the levels of genetic diversity, we used BOTTLENECK (Cornuet *et al*. 1999), performing the heterozygosity excess test developed by Cornuet and Luikart (1996). We applied three mutation models (IAM, SMM and TPM — with 80% of SMM and 10% variance) and a permutation test with 1000 iterations to estimate the significance of heterozygosity excess above the degree of heterozygosity assumed for a population in equilibrium. A significant excess of heterozygosity and a shift in allele frequencies are expected to occur in a bottlenecked population.

Genetic differentiation between seasons within populations from both islands, as well as between the islands, was estimated using F_{ST}. Overall F_{ST} (Weir & Cockerham 1984) and pairwise F_{ST} were obtained using FSTAT. The 95% confidence intervals for overall F_{ST} were also estimated using FSTAT. In the case of the bank vole, we applied the hierarchical AMOVA procedure in the Arlequin software (ver. 3.5.1.2) (Excoffier & Lischer 2010) in order to estimate F_{CT} values (the percentage of overall genetic variation explained by differences between groups) and their significance (1000 permutations) for different grouping patterns.

We used the Bayesian-clustering method (STRUCTURE ver. 2; Pritchard *et al*. 2000) to examine how well "populations" from particular seasons corresponded to genetic groups (K). STRUCTURE was run 15 times for each user-defined K (1–6), with an initial burn-in of 50 000, and 100 000 iterations of the total data set. The admixture model of ancestry and the correlated model of allele frequencies were used. Sampling location was not used as prior information. Next, we examined ΔK statistics, which identify the largest change in the estimates of K produced by STRUCTURE, as ΔK may provide a more realistic estimation of K than those based on likelihoods (Evano *et al*. 2005).

To visualize the STRUCTURE results we used STRUCTURE HARVESTER (Earl & vonHoldt 2011). Then, we applied CLUMPP (Jakobsson & Rosenberg 2007) to average the multiple runs given by STRUCTURE and to correct for label switching. The output from CLUMPP was visualized using DISTRUCT ver. 1.1 (Rosenberg 2004) to display the results.

Additionally, an assignment test and detection of first generation migrants were conducted using the Markov Chain Monte Carlo (Patkau *et al*. 2004 simulation algorithm), with 1000 simulated individuals per population and the prob-

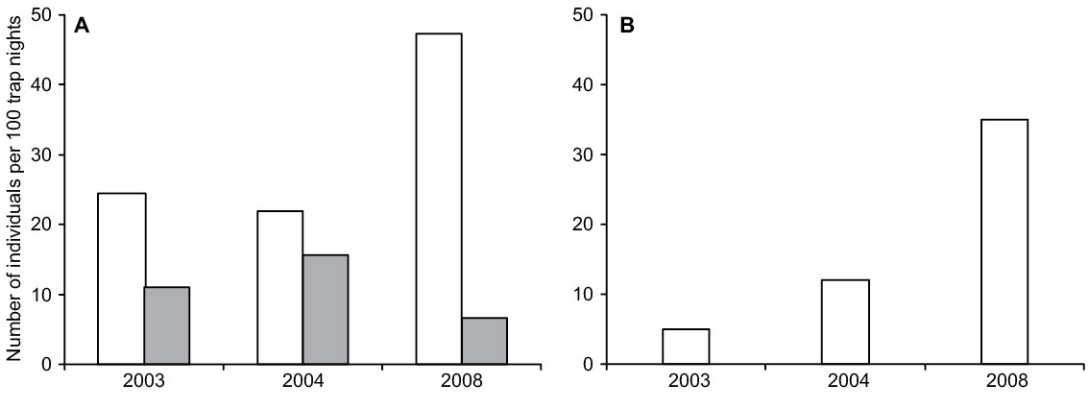

Fig. 2. Densities of bank voles (white bars) and yellow-necked mice (grey bars) during the trapping periods on (**A**) Mouse Island and (**B**) Shrew Island.

ability of the exclusion threshold set to $p < 0.01$ (GENECLASS, Cornuet *et al*. 1999).

In order to characterize the overall genetic pool of each particular island population, each individual was included only once in the analysis. To characterize the genetic pool of seasonal populations, however, all individuals caught in a particular season were included; hence we also analysed the genotypes of re-trapped individuals.

Results

The bank vole

The bank vole was found on both islands during all years of the study. Its numbers were very low on SI in 2003 and 2004; in 2008, however, the species became more abundant. Bank vole densities were higher on MI than on SI in all years, with maximum density in 2008 on SI (Fig. 2).

Genetic variability was higher on MI, both in terms of allelic diversity and heteroztygosity (Table 1). The difference was especially striking in the case of the mean number of private alleles and of private allelic richness, indicating that the majority of the microsatellite alleles identified could be found on MI. On MI the number of alleles was similar across years, while on SI, it increased significantly in 2008. In 2008, we detected alleles which had not been present earlier (in 2003 and 2004) on both islands.

Table 1. Comparison of the mean values of genetic variability indices in the populations of *Myodes glareolus* from Mouse Island and Shrew Island in different years (n = 437). n = sample size, A = allelic diversity, R = allelic richness, A_p = mean number of private alleles, p_R = private allelic richness, N_e = effective number of alleles, H_O = observed heterozygosity, H_E = expected heterozygosity, $p_{(HWE)}$ = p values for HWE exact test for heterozygote deficiency/excess, F_{IS} = fixation index.

	n	A	R	A_p	p_R	N_e	H_O	H_E	$p_{(HWE)}$	F_{IS}
Mouse Island										
2003	73	8.43	7.17	0.14	0.22	5.52	0.787	0.806	< 0.001	0.031
2004	66	8.57	7.30	0.14	0.18	5.38	0.764	0.804	< 0.001	0.057
2008	142	8.14	6.45	0.57	0.18	5.15	0.604	0.791	< 0.001	0.241*
All MI	281	9.29	8.20	2.29	2.06	5.60	0.689	0.810	< 0.001	0.151*
Shrew Island										
2003	15	2.57	2.57	0.00	0.00	1.99	0.524	0.452	0.231	–0.125
2004	36	2.43	2.34	0.14	0.06	1.87	0.448	0.391	0.015	–0.132
2008	105	7.29	5.95	0.29	0.31	4.71	0.668	0.776	< 0.001	0.144*
All SI	156	7.43	7.43	0.43	0.46	4.03	0.603	0.773	< 0.001	0.179*
Total	437	9.71					0.658	0.817	< 0.001	0.195*

* significant F_{IS} value after Bonferroni correction (Bonferroni-corrected p after 840 randomization at α = 0.05 was 0.0012).

Neither island population was at the HWE and both had similar overall F_{IS} values, indicating heterozygote deficiency (Table 1). In the MI population we also found heterozygote deficiency every year. F_{IS} was, however, significantly higher than zero in 2008. On SI, we found heterozygote excess in 2003 and 2004 (but F_{IS} values were not significant), and significant heterozygote deficiency in 2008.

We confirmed the existence of genetic after-effects of demographic bottlenecks on both islands for each year using IAM and TPM models (Wilcoxon test: $p < 0.05$). Similarly, significant heterozygote excess, which also indicated the existence of previous bottlenecks, was also found in the pooled data set of all years within each island, and for all years and both islands using IAM and TPM (Wilcoxon test: $p < 0.05$). No significant heterozygote excess was found in the tests which assumed an SMM model of microsatellite mutation.

In pooled data from the three years, there was small but significant genetic differentiation between the islands ($F_{ST} = 0.085$, $p = 0.05$). In the case of MI, genetic differentiation between years was small, and seemed to be interlinked with a time period separating the compared years (Table 2). On SI, we found a small but significant degree of genetic differentiation between 2003 and 2004, and, when we made pairwise comparisons of both seasons against 2008, about four times the genetic differentiation in 2008 (Table 2). In 2003 and 2004, genetic differentiation between the islands was very high, indicating clear and significant differences in the frequencies of microsatellite alleles. In 2008, however, the microsatellite genetic pool on SI was similar to the MI population (Table 2): the F_{ST} values dropped from 0.23–0.28 to as small as 0.05. This suggested a distinct shift in the frequencies of the microsatellite allele on SI between 2004 and 2008. This result was supported by the STRUCTURE analysis, with which we identified three genetic groups (Fig. 3). MI was dominated by the first of those genetic groups (Fig. 3C: red bars). A portion of the individuals, however, had their highest proportion of ancestry from group two (Fig. 3C: blue bars). On SI in 2003 and 2004, only one genetic group was identified (group three, Fig. 3C: green bars), but in 2008 genetic group two (Fig. 3C: blue bars) clearly prevailed, with a very small portion of individuals from group one or of mixed ancestry. The AMOVA analysis supported this division of genetic variability into three distinct groups: group I: MI2003, MI2004, MI2008; group II: SI2003, SI2004; group III: SI2008 ($F_{CT} = 0.1199, p = 0.008$).

The assignment test indicated that the majority of individuals (79%) were assigned to their proper 'island of origin'. In the case of MI, 4.6% of individuals were assigned to the SI genetic pool from 2008. Within SI, five individuals (3.2%) were assigned to MI. Identification of the first generation migrants indicated that three individuals on MI were classified as migrants from the SI genetic pool of 2008. On SI, three individuals were also identified as first generation migrants from MI (one migrant per year).

Table 2. Genetic differentiation (F_{ST}) of the bank vole populations by years and islands. All values were significant, except for the one shown in boldface. The Bonferroni-corrected p value after 300 randomization at $\alpha = 0.05$ was 0.0033. The overall F_{ST} was 0.099 (95%CI = 0.071–0.133, bootstrapping over loci). F_{ST} between the Mouse Island and Shrew Island (pooled data for all years) was 0.085 and significant ($p = 0.05$). Italics indicates differentiation among years between the two islands.

	Mouse Island		Shrew Island		
	2004	2008	2003	2004	2008
Mouse Island					
2003	**0.0015**	0.0205	*0.2067*	*0.2656*	*0.0549*
2004		0.0205	*0.2204*	*0.2777*	*0.0596*
2008			*0.2318*	*0.2786*	*0.0538*
Shrew Island					
2003				0.0497	0.1650
2004					0.1960

The yellow-necked mouse

During all study years, the yellow-necked mouse was found only on MI, at very low numbers, reaching a minimum in 2008. Over the entire study period, it was found at much lower numbers than the bank vole, which co-occurred on

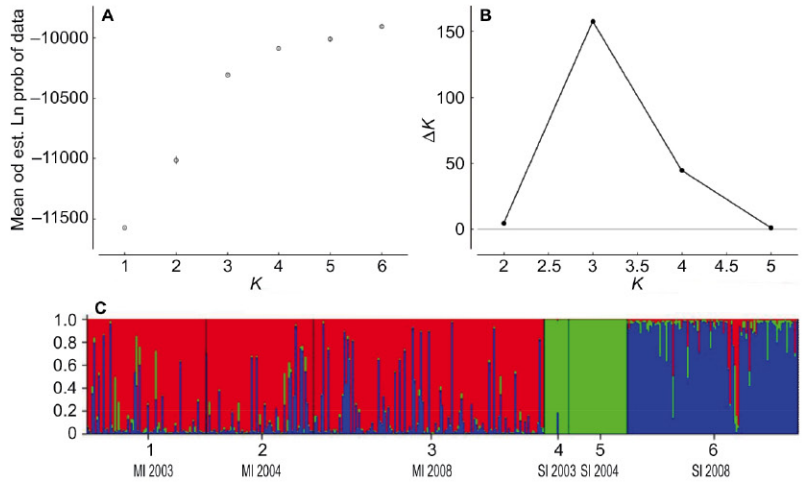

Fig. 3. (**A**) Estimated likelihoods, ln*P*(*D*), of each number of inferred genetic clusters, (**B**) the corresponding Δ*K* curves as a function of *K* for island populations and seasonal populations of *Myodes glareolus*, and (**C**) Bayesian assignment of individuals to three genetic groups indicated by Δ*K*. Each bar represents the estimated posterior probability of each individual belonging to each of the three inferred clusters. Solid black lines define boundaries between the populations and/or year used in the analysis.

MI (Fig. 2). Among the five amplified microsatellite loci, one showed significant heterozygote deficiency, indicated by very high F_{IS} values (0.270–0.660) for each year. This may have been caused by a high frequency of 'null' alleles, although we found that this problem did not bias the results of our genetic analysis. Nonetheless, we analyzed our data for both 'full' and 'reduced' marker sets. Independently on the analyzed marker set, we found that indices of genetic variability decreased between 2004 and 2008 (Table 3). It was most pronounced in the case of heterozygosity levels, which declined almost by half between the 2003 and 2008 seasons. Similarly, independently on the analyzed number of microsatellites, we detected a clear decrease in the number of private alleles in subsequent seasons (A_p and p_R; Table 3).

Due to heterozygote deficiency, the population was not at the HWE during every year of the study, suggesting an increase in the level of inbreeding on MI (Table 3). Following exclusion of the heterozygote deficiency locus, however, a significant heterozygote excess occurred in 2003, a significant heterozygote deficiency in 2004, and a lack of significant deviation from the HWE in 2008. Nonetheless, all F_{IS} values did not differ significantly from zero (Table 3). Hence, we can assume that the results of the heterozygote estimation were strongly biased by homozygote excess at the heterozygote deficiency locus, rather than by a real increase in levels of inbreeding over subsequent years.

For both marker sets, the pooled data for all years indicated significant heterozygote deficiency; this was, however, much less pronounced after the exclusion of one locus (Table 3). This suggested differences in allele frequencies between years. Although we did not find genetic differentiation between 2003 and 2004 (for five microsatellites: $F_{ST} = -0.0039$; for four microsatellites: $F_{ST} = -0.0023$, ns), pairwise comparisons with 2008 indicated significant differences in the frequency of microsatellite alleles (for five microsatellites: $F_{ST} = 0.1855$ and 0.1986 for comparison with 2003 and 2004, respectively; for four microsatellites: $F_{ST} = 0.2193$ and 0.2216 for comparison with 2003 and 2004, respectively, Bonferroni corrected p value after 60 randomizations at $\alpha = 0.05$ was 0.0166). Overall F_{ST} was 0.116 and 0.118 for 'full' and 'reduced' marker sets (95%CI = 0.037–0.207).

In the case of IAM, the BOTLLENECK analysis confirmed the presence of a genetic signature of demographic changes in the pooled data set for all years (Wilcoxon test: $p < 0.05$), but heterozygosity excess was also close to significance in the case of TPM ($p = 0.07$). Analyz-

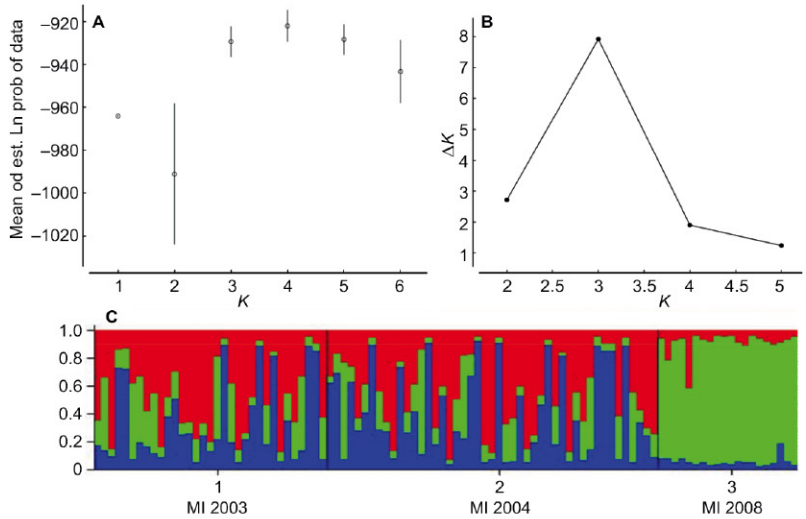

Fig. 4. (**A**) Estimated likelihoods, ln$P(D)$, of each number of inferred genetic clusters, (**B**) the corresponding ΔK curves as a function of K for island populations and seasonal populations of *Apodemus flavicollis*, and (**C**) Bayesian assignment of individuals to three genetic groups, indicated by ΔK. Each bar represents the estimated posterior probability of each individual belonging to each of the three inferred clusters. Solid black lines define boundaries between the populations and/or year used in the analysis.

ing data from separate years, we found bottleneck effects in 2003 and 2004 (IAM, $p < 0.01$, TPM, $p = 0.076$), but not in 2008 (ns heterozygosity excess for both models).

Results of the STRUCTURE analysis suggested the presence of three genetic groups (Fig. 4). Individuals from 2008 had the highest probability of their ancestry originating exclusively from one genetic group (green bars), whereas in 2003 and 2004 we found individuals of mixed origin or belonging to two other genetic groups (blue and red bars).

For the 'full' marker set, 57% of individuals were correctly assigned to the sampling year, mainly because the majority of individuals from years 2003 and 2004 were 'cross-assigned' or had an equally high probability of assignment to both of these years; none of these individuals,

Table 3. Comparison of the mean values of genetic variability indices in the population of *Apodemus flavicollis* from Mouse Island in different years ($n = 100$) for two data sets: (i) 5 microsatellites; (ii) 4 microsatellites, excluding Locus 3. n = sample size, A = allelic diversity, R = allelic richness, A_p = mean number of private alleles, p_R = private allelic richness, N_e = effective number of alleles, H_O = observed heterozygosity, H_E = expected heterozygosity, $p_{(HWE)}$ = p values for HWE exact test for heterozygote deficiency/excess, F_{IS} = fixation index.

	n	A	R	A_p	p_R	N_e	H_O	H_E	$p_{(HWE)}$	F_{IS}
Mouse Island (i)										
2003	33	5.20	4.74	0.40	0.48	2.94	0.630	0.630	< 0.001	0.016
2004	47	5.80	4.97	0.40	0.45	3.17	0.583	0.656	< 0.001	0.123*
2008	20	3.40	3.40	0	0.31	2.07	0.340	0.400	0.008	0.174*
All MI	100	6.20					0.550	0.646	< 0.001	0.189*
Mouse Island (ii)										
2003	33	5.00	4.52	0.50	0.51	2.59	0.644	0.596	0.017	–0.066
2004	47	5.50	4.76	0.25	0.42	2.83	0.617	0.625	< 0.001	0.024
2008	20	3.25	3.25	0	0.38	2.02	0.375	0.357	0.843	–0.024
All MI	100	6.00					0.578	0.616	< 0.001	0.067

* significant F_{IS} value after Bonferroni correction (Bonferroni-corrected p after 300 randomization at $\alpha = 0.05$ was 0.0033).

however, was assigned to year 2008. In the case of the reduced marker set, 61% of individuals were correctly assigned to the year of origin, and a similar admixture of assignments between 2003 and 2004 was found. Only 50% of individuals from 2008 were explicitly assigned to the genetic pool from that year, whereas the other eight individuals had an equally high probability of being assigned to any of the three years. Two individuals from 2008 were assigned to the year 2004.

Discussion

We can be certain that in the past rodents from the mainland settled on the islands which were the focus of our study. In the study area, rodents are probably only able to move between the mainland and the islands during the winter, and only in years when the lake is frozen. As the islands are relatively far from the mainland, it is unlikely that the rodents would be able to swim the several hundred meters necessary to arrive on the islands during summer.

In the bank vole population on MI, the levels of genetic diversity remained constant over subsequent years and showed high levels of heterozygosity, in spite of marked isolation of the island. The high levels of heterozygosity may be the after-effect of a sudden demographic expansion, following a period of low population size (bottleneck effect). Indeed, the genetic signatures of past demographic bottlenecks are clearly observable in the data. It is possible that island populations of rodents fluctuate systematically, as a result for example of periodic floods. Populations of small rodents are known to fluctuate in population sizes (Flowerdew 1985, Alibhai & Gipps 1985, Banach 1987, Kozakiewicz & Kozakiewicz 2008).

The genetic distance (F_{ST}) between the bank vole population in 2008 and previous years was very low, which demonstrates the relative stability of genetic structure of the local population of this species. Other investigations on rodents have shown that their populations may maintain unexpectedly high levels of genetic variability over a long period of time, unless the populations are too small (Vega *et al*. 2007).

On SI, the degree of genetic diversity in the bank vole populations was similarly low in 2003 and 2004, but rose in 2008 (this conclusion was supported by all indices of microsatellite polymorphisms). In 2003 and 2004, the inbreeding coefficient was negative, indicating an excess of heterozygotes in the bank vole population. In 2008, on the other hand, a marked increase was noted in the rate of inbreeding, together with a significant deficiency of heterozygotes, although the island was inhabited by many more individuals than in the previous years. Hence, it may be speculated that a sudden drop in the population size occurred between 2004 and 2008 (the island was flooded in 2006 or in 2007), followed by a recolonization of the island by a group of related individuals, resulting in a founder effect. Hence, the number of alleles and the number of heterozygotic loci in the newly-founded SI population clearly increased in comparison with the previously isolated but relatively stable population, in which a number of alleles had long since been eliminated by genetic drift. We know that during the years 2003–2008 the lake was always frozen in winter months, which could have helped some individuals to reach the islands.

The hyphothesis of a colonization event on SI between 2004 and 2008 may also be supported by the very low genetic distance (F_{ST}) between 2003 and 2004, compared with a very high one between these years and 2008, which shows that there had, during this time, occurred a significant change in the genetic pool of the population. Moreover, the genetic distance between the bank vole populations inhabiting both islands was distinctly lower, indicating that the SI gene pool could have been joined by individuals from MI. This hypothesis is strengthened by the fact that a large number of individuals from the "blue" clade and, in addition, a smaller number of individuals from the "red" clade were found on SI in 2008. These clades prevailed on MI throughout the entire study period, and were totally absent from SI in 2003 and 2004. Absence of the "green" clade in 2008, which had been dominant on SI in 2003 and 2004, and the assignment test and identification of the first generation migrants, also suggest that the variability observed on SI in 2008 is likely the result of the local extinction of the population,

followed by the later immigration of a small group of animals from MI. On the other hand, result of the AMOVA test indicates that the bank vole population in 2008 formed a separate genetic group on SI, which may indicate another source for the colonization of SI. It means that the rodents might have also come to the island from the mainland. This is strongly supported by the presence in 2008 of some alleles which were not present on the islands in 2003 and 2004. To summarize, it is possible that the bank vole population on SI periodically goes extinct and is then replenished by individuals of diverse origins, i.e., migrants from MI or individuals coming to the island from the mainland. On the other hand, the small sizes of the islands and the large distance between them likely indicates that the main source of migrants is the mainland, with only occasional movement of individuals between the islands.

The high likelihood of periodic extinction events for the bank vole population on SI is confirmed by the results of Kozakiewicz *et al.* (2000), which showed that, in an agricultural landscape, the existence of a metapopulation of the species is possible when the mean area of the habitat patches is at least 2 ha, and the mean distance between local populations is 1 km. The degree of isolation of SI is undoubtedly greater than that corresponding to the 1 km distance on the mainland. In addition, the size of the island – max. 1.5 ha, and periodically smaller when part of the island is flooded during high water levels – is too small for the local population to remain constant.

One would expect that a semi-arboreal species such as a mouse would be better able to deal with periodical flooding than a terrestrial bank vole, but unlike the bank vole, the yellow-necked mouse occurred only on MI. This pattern was also observed by Kozakiewicz *et al.* (2009) during their investigation. SI is probably too small to be settled by the yellow-necked mouse, which needs a larger territory in which to live. In addition, according to Kozakiewicz *et al.* (2009), the poor habitat quality on SI does not meet the basic requirements of this species. Contrary to what was the case for the bank vole, and as was confirmed by all of the analyzed indices, genetic diversity of the mouse population decreased markedly between 2004 and 2008. A significant, almost twofold, decrease in the observed heterozygosity and a marked increase in the inbreeding coefficient during that period undoubtedly occurred due to the very low population size of the mouse population and its severe isolation. This was confirmed by the marked rise in genetic distance in 2008 in comparison with the previous years, indicating a very high level of genetic drift in the mouse population. In addition, the STRUCTURE results appear to confirm isolation of the mouse population as well as genetic drift which occurred in it, as the two groups dominating the population in 2003 and 2004 (the "red" and "blue" clades) were by 2008 represented by only a small group of specimens. At this time, the majority of mice belonged to the third genetic group (the "green" clade), which had been marginal in 2003–2004. We realize that a thorough analysis of the sources of gene flow to the island populations of both species should also take into account mainland populations of rodents. Although we have no data on the genetic structure of the mainland population between the years 2005 and 2008, it should be pointed out that an earlier study (Kozakiewicz *et al.* 2009) demonstrated that the mainland populations of both species were characterized by much higher genetic diversity as compared with the island populations. Thus the mainland can be treated as harboring a large source population, which could serve to periodically enrich the genetic pools of the island populations via migration. Migration events of voles and mice between the mainland and the islands in our study, however, are accidental and very rare.

The current study did not confirm our hypothesis that a less mobile species, such as the bank vole, would lose its genetic variability on islands more rapidly than the more active and mobile yellow-necked mouse. Undoubtedly, for both species the water barrier is not sufficient to prevent gene flow from reaching the island populations. Frozen lakes and rivers are crossed very rarely by the bank vole (Dewsbury *et al.* 1982, Aars *et al.* 1998). The yellow-necked mouse, on the other hand, is more likely to cross open areas and is able to travel up to 1 kilometer (Bondrup-Nielsen & Karlsson 1985, Szacki 1999); thus, the mouse would suffer the effects of the isola-

tion less than the vole. We may thus speculate that the preservation of genetic diversity, in particular on the larger island MI, is possible due to the specific social and spatial organization of the local bank vole population, which allows the maintenance of a relatively stable number of individuals. According to Bujalska (1988), Bujalska and Saitoh (2000) and Bujalska and Grüm (2008), the crucial factor is territoriality of adult breeding vole females. Additionally, as shown by Kozakiewicz (1985), in isolated populations of the bank vole, mean home range size decreases with increasing population density. This in turn helps to maintain a constant degree of overlap, irrespective of density, and stabilizes the social and spatial structure, including population size. Due to such a system of social and spatial organization, bank vole populations are able to survive and maintain a constant level of genetic diversity even on a relatively small island (about 3 ha), such as MI. Gliwicz (1980) came to a similar conclusion, i.e., that for island populations of rodents, the arrangement of home ranges allows the population to squeeze in a greater number of individuals while maintaining the lowest possible number of interactions between individuals.

In contrast to the bank vole, in their investigations of an island population of the yellow-necked mouse, Bujalska and Grüm (2005) reported a lack of territorial behavior between adult female mice: all of them breed, not only those who have their own territory. According to the authors, this explains the frequent dramatic seasonal rises and ensuing decreases in population size, even to extremely low numbers. In turn this results in a low winter survival rate leading to very low mouse densities in spring, which, on islands, is likely to cause extinction events (Bujalska & Grüm 2006, 2008). In their long-running investigations of populations of both species on a 4.5 ha lake island, (which is slightly bigger than MI), Bujalska and Grüm (2008) recorded several extinction and re-colonization events in the yellow-necked mouse, while the bank vole population remained constant. Similarly, we found genetic evidence of fluctuating population sizes in the mouse population on MI.

Given that the influx of genes to islands appears to be a random event, it is difficult to predict the future genetic structure of both species on the islands we investigated. A number of scenarios are possible, including periodic extinctions of populations of both species (Alcover *et al.* 1998, Miller *et al.* 2011). Taking into account the results of our work, one may expect temporary rapid decreases in bank vole population sizes on SI and the same for yellow-necked mouse populations on MI, together with resulting periodic decreases of the genetic diversity of those populations. This is in accordance with the so-called 'extinction vortex' mechanism, which was described by Caughley (1994) for small, isolated populations. Consequently, periodic extinction events of these populations are highly probable, followed by re-colonization of the islands. The results of this study also indicate that the bank vole population on MI is large and stable enough to maintain high levels of genetic diversity, unless rapid, unexpected decreases in population size occur (e.g., as caused by predators). In such cases, genetic diversity retained will depend on the final size to which the population will be reduced, and on its reproductive rate (Freeland 2005). Moreover, depending on environmental conditions, selection pressure in that population may favor different genotypes (Gębczyński & Ratkiewicz 1998), which may lead to the elimination of certain alleles in the process of microevolution.

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References

Aars, J., Ims, R. A., Liu, H. P., Mulvey, M. & Smith, M. H. 1998: Bank voles in linear habitats show restricted gene flow as revealed by mitochondrial DNA (mtDNA). — *Molecular Ecology* 7: 1383–1389.

Alcover, J. A., Sans, A. & Palmer, M. 1998: The extent of extinctions of mammals on islands. — *Journal of Biogeography* 25: 913–918.

Alibhai, S. K. & Gipps, J. H. W. 1985: The population

dynamics of bank voles. — *Symposia of the Zoological Society of London* 55: 277–313.

Aquadro, C. F. & Kilpatrick, C. W. 1981: Morphological and biochemical variation and differentiation in insular and mainland deer mice (*Peromyscus maniculatus*). — In: Smith, M. H. & Joule, J. (eds.), *Mammalian population Genetics*: 214–230. University of Georgia Press, Athens, GA.

Banach, A. 1987: Small rodent communities in a complex of forest biotopes. — *Acta Theriologica* 32: 229–244.

Barker, F. S., Helyar, S. J., Kemp, S. J. & Begon, M. 2005: Highly polymorphic microsatellite loci in the bank vole (*Myodes glareolus*). — *Molecular Ecology Notes* 5: 311–313.

Bauchau, V. & le Boulengé, E. 1991: Population biology of woodland rodents in a patchy landscape. — In: le Berre, M. & le Gelte, L. (eds.), *Le rongeur et l'espace:* 275–283. Chabaud, Paris.

Bąkowski, C. & Kozakiewicz, M. 1988: The effect of forest road on bank vole and yellow-necked mouse populations. — *Acta Theriologica* 33: 345–353.

Berry, R. J. & Peters J. 1997: Heterogenous hetrozygosities in *Mus musculus* populations. — *Proceedings of the Royal Society B* 197: 485–503.

Bondrup-Nilsen, S. & Karlsson, F. 1985: Movements and spatial patterns in populations of *Myodes* species. A review. — *Annales Zoologici Fennici* 22: 385–392.

Bujalska, G. 1988: Life history consequences of territoriality in the bank vole. — In: Boyce, M. S. (ed.) *Evolution of life histories of mammals*: 75–79. Yale University Press, New Haven.

Bujalska, G. & Grüm, L. 2005: Reproduction strategy in an island population of yellow-necked mice. — *Population Ecology* 47: 151–154.

Bujalska, G. & Grüm, L. 2006: Winter survival of *Apodemus flavicollis* in Crabapple Island (NE Poland). — *Hystrix Italian Journal of Mammalogy* (n.s.) 17: 173–177.

Bujalska, G. & Grüm, L. 2008: Interaction between populations of the bank vole and the yellow-necked mouse. — *Annales Zoologici Fennici* 45: 248–254.

Bujalska, G. & Saitoh, T. 2000: Territoriality and its consequences. — *Polish Journal of Ecology* 48 (Suppl): 37–49.

Caughley, G. 1994: Directions in conservation biology. — *Journal of Animal Ecology* 63: 215–244.

Corbet, G. B. 1964: Regional variation in the bank vole, *Clethrionomys glareolus*, in the British Isles. — *Proceedings of the Zoological Society of London* 143: 191–219.

Cornuet, J. M. & Luikart, G. 1996: Description and power analysis of two tests for detecting recent population bottlenecks from allele frequency data. — *Genetics* 144: 2001–2014.

Cornuet, J. M., Piry, S., Luikart, G., Estoup, A. & Solignac, M. 1999: New methods employing multilocus genotypes to select or exclude populations as origins of individuals. — *Genetics* 153: 1989–2000.

Dewsbury, D., Baumgardner, D. J., Sawrey, D. K. & Webster, D. G. 1982: The adaptive profile: comparative psychology of red-backed voles. — *Journal of Comparative Physiology and Psychology* 96: 649–660.

Earl, D. A. & von Holdt, B. M. 2011: STRUCTURE HARVESTER: a website and program for visualizing STRUCTURE output and implementing the Evanno method. — *Conservation Genetics Research* 4: 359–361.

Evanno, G., Regnaut, S. & Goudet, J. 2005: Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. — *Molecular Ecology* 14: 2611–2620.

Excoffier, L. & Lischer, H. E. L. 2010: Arlequin suite ver 3.5: a new series of programs to perform population genetics analyses under Linux and Windows. — *Molecular Ecology Resources* 10: 564–567.

Flowerdew, J. R. 1985: The population dynamics of wood mice and yellow-necked mice. — *Symposia of the Zoological Society of London* 55: 315–338.

Frankham, R. 1996: Relationship of genetic variation to population size in wildlife. — *Conservation Biology* 10: 1500–1508.

Frankham, R. 1997: Do island populations have less genetic variation than mainland populations? — *Heredity* 78: 311–327.

Freeland, J. R. 2005: *Molecular ecology*. — John Wiley & Sons, Chichester, England.

Gębczyński, M. & Ratkiewicz, M. 1998: Does biotope diversity promote an increase of genetic variation in the bank vole population? — *Acta Theriologica* 43: 163–173.

Gerlach, G. & Musolf, K. 2000: Fragmentation of landscape as a cause for genetic subdivision in bank voles. — *Conservation Biology* 14: 1066–1074.

Gill, A. E. 1980: Evolutionary genetics of California Island *Peromyscus*. — In: Power, D. M. (ed.), *The California Islands: Proceedings of a Multidisciplinary Symposium:* 719–743. Santa Barbara Museum of Natural History, Santa Barbara, CA.

Gliwicz, J. 1980: Island populations of rodents: their organization and functioning. — *Biological Reviews* 55: 109–138.

Gockel, J., Harr, B., Schlötterer, C., Arnold, W., Gerlach, G. & Tautz, D. 1997: Isolation and characterization of microsatellite loci from *Apodemus flavicollis* (rodentia, muridae) and *Myodes glareolus* (rodentia, cricetidae). — *Molecular Ecology* 6: 597–599.

Gortat, T., Gryczyńska-Siemiątkowska, A., Rutkowski, R., Kozakiewicz, A., Mikoszewski, A. & Kozakiewicz, M. 2010: Landscape pattern and genetic structure of a yellow-necked mouse *Apodemus flavicollis* population in north-eastern Poland. — *Acta Theriologica* 55: 109–121.

Gortat, T., Rutkowski, R., Gryczyńska-Siemiątkowska, A., Kozakiewicz, A. & Kozakiewicz, M. 2013: Genetic structure in urban and rural populations of *Apodemus agrarius* in Poland. — *Mammalian Biology* 78: 171–177.

Grüm, L. & Bujalska, G. 2000: Bank voles and yellow-necked mice: what are interrelations between them? — *Polish Journal of Ecology* 48 (Suppl.): 141–145.

Goudet, J. 2001: *FSTAT V2.9.3, a program to estimate and test gene diversities and fixation indices*. Available at http://www2.unil.ch/popgen/softwares/fstat.htm.

Hirota, T., Hirohata, T., Mashima, H., Satoh, T. & Obara, Y. 2004: Population structure of the large Japanese field

mouse, *Apodemus speciosus* (Rodentia: Muridae), in suburban landscape, based on mitochondrial D-loop sequences. — *Molecular Ecology* 13: 3275–3282.

Jakobsson, M. & Rosenberg, N. A. 2007: CLUMPP: a cluster matching and permutation program for dealing with label switching and multimodality in analysis of population structure. — *Bioinformatics* 23: 1801–1806.

Jeanike, J. R. 1973: A steady state model of genetic poplymorphism on island. — *American Naturalist* 107: 793–795.

Kalinowski, S. T. 2005: A computer program for performing rarefaction on measures of allelic diversity. — *Molecular Ecology Notes* 5: 187–189.

Kilpatrick, C. W. 1981: Genetic structure of insular populations. — In: Smith, M. H. & Joule, J. (eds.), *Mammalian population genetics*: 28–59. University of Georgia Press, Athens, Georgia.

Kozakiewicz, M. 1985: The role of habitat isolation in formation of structure and dynamics of the bank vole population. — *Acta Theriologica* 30: 193–209.

Kozakiewicz, M., Apeldoorn, R. V., Bergers, P., Gortat, T. & Kozakiewicz, A. 2000: Landscape approach to bank vole ecology. — *Polish Journal of Ecology* 48 (Suppl.): 149–161.

Kozakiewicz, M., Gortat, T., Panagiotopoulou, H., Gryczyńska-Siemiątkowska, A., Rutkowski, R., Kozakiewicz, A. & Abramowicz, K. 2009: The spatial genetic structure of bank vole (*Myodes glareolus*) and yellow-necked mouse (*Apodemus flavicollis*) populations: the effect of distance and habitat barriers. — *Animal Biology* 59: 169–187.

Kozakiewicz, M. & Kozakiewicz, A. 2008: Long-term dynamics and biodiversity changes in small mammal communities in a mosaic of agricultural and forest habitats. — *Annales Zoologici Fennici* 45: 263–269.

Lande, R. 1988: Genetics and demography in biological conservation. — *Science* 241: 1455– 1460.

Makova, K. D., Patton, J. C., Krysanov, E. Y. U., Chesser, R. K. & Baker, R. J. 1998: Microsatellite markers in wood mouse and striped field mouse (genus *Apodemus*). — *Molecular Ecology* 7: 247–249.

Miller, M. P., Haig, S. M., Ledig, D. B., Vander Heyden, M. F. & Bennett, G. 2011: Will an "island" population of voles be recolonized if eradicated? Insights from molecular genetic analyses. — *Journal of Wildlife Management* 75: 1812–1818.

Nei, M. & Roychoudhury, A. K. 1974: Sampling variances of heterozygosity and genetic distance. — *Genetics* 76: 379–390.

Ohnishi, N., Ishibashi, Y., Saitoh, T., Abe, S. & Yoshida, M. C. 1998: Polymorphic microsatellite DNA markers in the Japanese wood mouse *Apodemus argenteus*. — *Molecular Ecology* 7: 1431–1432.

Paekal, R. & Smouse, P. E. 2006: GenAlEx 6: genetic analysis in Excel. Population genetic software for teaching and research. — *Molecular Ecology* 6: 288–295.

Patkau, D., Slade, R., Burden, M. & Estoup, A. 2004: Genetic assignment methods for the direct, real-time estimation of migration rate: a simulation-based exploration of accuracy and power. — *Molecular Ecology* 13: 55–65.

Pergams, O. R. W. & Ashley, M. V. 2001: Microevolution in island rodents. — *Genetica* 112–113: 245–256.

Petit, R. J., el Mousadik, A. & Pons, O. 1998: Identifying populations for conservation on the basis of genetic markers. — *Conservation Biology* 12: 844–855.

Pritchard, J. K., Stephens, M. & Donnelly, P. 2000: Inference of population structure using multilocus genotype data. — *Genetics* 155: 945–959.

Pucek, M. 1983: Habitat preference. — *Acta Theriologica* 28 (Suppl. 1): 31–40.

Ratkiewicz, M. & Borkowska, A. 2006: Genetic structure is influenced by environmental barriers: empirical evidence from the common vole *Microtus arvalis* populations. — *Acta Theriologica* 51: 337–344.

Raymond, M. & Rousset, F. 1995: GENEPOP (version 1.2): population genetics software for exact tests and ecumenicism. — *Journal of Heredity* 86: 248–249.

Rosenberg, N. A. 2004: DISTRUCT, a program for the graphical display of population structure. — *Molecular Ecology Notes* 4: 137–138.

Rousset, F. 2008: Genepop'007: a complete reimplementation of the Genepop software for Windows and Linux. — *Molecular Ecology Resources* 8: 103–106.

Redeker, S., Andersen, L. W., Pertoldi, C., Madsen, A. B., Jensen, T. S. & Jørgensen, J. M. 2006: Genetic structure, habitat fragmentation and bottlenecks in Danish bank voles (*Myodes glareolus*). — *Mammalian Biology* 71: 144–158.

Rutkowski, R., Jagołkowska, P., Mazgajski, M. D. & Rejt, Ł. 2008: Mitochondrial marker for studying European woodpeckers on various taxonomic levels. — *Annales Zoologici* 58: 907–913.

Ryan, A., Duke, E. & Fairley, J. S. 1996: Mitochondrial DNA in bank voles (*Myodes glareolus*) in Ireland: evidence for a small founder population and localized founder effects. — *Acta Theriologica* 41: 45–50.

Saunders, D. A., Hobbs, R. J. & Margules, C. R. 1991: Biological consequences of ecosystem fragmentation: a review. — *Conservation Biology* 5: 18–32.

Schmitt, L. H. 1978: Genetic variation in isolated populations of the Australian bush-rat, *Rattus fuscipes*. — *Evolution* 32: 1–14.

Smith, F. D. M., May, R. M., Pellew, R., Johnson, T. H. & Walter, K. R. 1993: How much do we know about the current extinction rate? — *Trends in Ecology & Evolution* 8: 375–378.

Soulé, M. E. 1976: Allozyme variation, its determinants in space and time. — In: Ayala, F. J. (ed.) *Molecular evolution*: 66–77. Sinauer associates, Sunderland, Massachusetts.

Stewart, D. T. & Baker, A. J. 1992: Genetic differentiation and biogeography of the masked shrew in Atlantic Canada. — *Canadian Journal of Zoology* 70: 106–14.

Szacki, J. 1999: Spatially structured populations: how much do they match the classic metapopulation concept? — *Landscape Ecology* 14: 369–379.

Vega, R., Vázquez-Domínguez, E., Mejía-Puente, A. & Cuarón A. D. 2007: Unexpected high levels of genetic variability and the population structure of an island endemic rodent (*Oryzomys couesi cozumelae*). — *Bio-*

logical Conservation 137: 210–222.
Weir, B. S. & Cockerham, C. C. 1984: Estimating *F*-statistics for the analysis of population structure. — *Evolution* 38: 1358–1370.
Wolton, R. J. 1985: Spatial distribution and movements of wood mice, yellow-necked mice and bank voles. — *Symposia of the Zoological Society of London* 55: 249–275.
Yom-Tom, Y. S. & Moller, H. 1999: Competition, coexistence, and adaptation amongst rodent invaders to Pacific and New Zealand islands. — *Biogeography* 26: 947–958.

Appendix 1. Frequency of microsatellite alleles in the *Myodes glaerolus* population from Shrew Island and Mouse Island over three years. Alleles are presented as an approximate length (in base pairs) of PCR product indicated by sequencer.

Locus	Allele	Mouse Island			Shrew Island		
		2003	2004	2008	2003	2004	2008
MSCg-4	109	0.103	0.167	0.187	0.000	0.000	0.000
	111	0.055	0.015	0.014	0.333	0.153	0.024
	113	0.000	0.000	0.000	0.000	0.014	0.000
	115	0.068	0.023	0.000	0.000	0.000	0.000
	117	0.007	0.000	0.000	0.000	0.000	0.000
	119	0.096	0.083	0.011	0.333	0.125	0.067
	121	0.158	0.174	0.215	0.233	0.444	0.257
	123	0.007	0.038	0.004	0.000	0.000	0.043
	125	0.226	0.250	0.327	0.000	0.000	0.167
	127	0.103	0.083	0.165	0.000	0.000	0.143
	131	0.014	0.015	0.004	0.000	0.000	0.238
	133	0.034	0.045	0.000	0.000	0.000	0.000
	135	0.130	0.106	0.074	0.100	0.264	0.062
MSCg-6	94	0.199	0.205	0.102	0.333	0.389	0.171
	98	0.137	0.136	0.113	0.000	0.000	0.219
	100	0.411	0.417	0.419	0.667	0.611	0.381
	102	0.253	0.242	0.366	0.000	0.000	0.229
MSCg-7	81	0.041	0.076	0.035	0.000	0.000	0.005
	85	0.014	0.061	0.081	0.000	0.000	0.010
	87	0.034	0.045	0.085	0.100	0.167	0.200
	89	0.000	0.008	0.000	0.000	0.000	0.033
	91	0.116	0.068	0.007	0.000	0.000	0.000
	95	0.021	0.023	0.070	0.000	0.000	0.010
	97	0.048	0.083	0.042	0.000	0.000	0.038
	101	0.089	0.023	0.000	0.000	0.000	0.095
	103	0.288	0.174	0.176	0.900	0.833	0.171
	105	0.274	0.341	0.296	0.000	0.000	0.352
	107	0.075	0.098	0.187	0.000	0.000	0.086
	109	0.000	0.000	0.021	0.000	0.000	0.000
MSCg-9	159	0.000	0.015	0.000	0.000	0.000	0.000
	161	0.089	0.121	0.106	0.000	0.000	0.010
	165	0.027	0.030	0.088	0.067	0.000	0.167
	167	0.075	0.106	0.194	0.267	0.000	0.090
	169	0.288	0.212	0.384	0.000	0.000	0.314
	173	0.014	0.045	0.004	0.667	1.000	0.352
	175	0.068	0.045	0.000	0.000	0.000	0.000
	177	0.027	0.038	0.056	0.000	0.000	0.000
	179	0.062	0.023	0.004	0.000	0.000	0.000
	180	0.000	0.000	0.004	0.000	0.000	0.000
	181	0.349	0.364	0.162	0.000	0.000	0.062
	183	0.000	0.000	0.000	0.000	0.000	0.005

continued

Appendix 1. Continued.

Locus	Allele	Mouse Island			Shrew Island		
		2003	2004	2008	2003	2004	2008
MSCg-24	92	0.144	0.091	0.141	0.000	0.000	0.290
	94	0.171	0.174	0.201	0.000	0.000	0.157
	96	0.068	0.083	0.158	0.000	0.000	0.000
	98	0.089	0.121	0.127	0.767	0.861	0.357
	100	0.137	0.053	0.032	0.233	0.139	0.005
	102	0.048	0.098	0.088	0.000	0.000	0.095
	104	0.082	0.045	0.049	0.000	0.000	0.000
	106	0.068	0.121	0.092	0.000	0.000	0.090
	108	0.192	0.212	0.102	0.000	0.000	0.005
	110	0.000	0.000	0.004	0.000	0.000	0.000
	114	0.000	0.000	0.007	0.000	0.000	0.000
LIST3-003	218	0.240	0.250	0.095	0.667	0.528	0.190
	224	0.082	0.023	0.204	0.000	0.000	0.138
	230	0.027	0.015	0.000	0.200	0.389	0.086
	234	0.151	0.144	0.106	0.000	0.000	0.114
	236	0.000	0.000	0.000	0.000	0.000	0.171
	238	0.171	0.182	0.120	0.133	0.083	0.119
	242	0.260	0.288	0.310	0.000	0.000	0.119
	246	0.041	0.053	0.102	0.000	0.000	0.000
	250	0.027	0.045	0.063	0.000	0.000	0.062
LIST3-007	220	0.144	0.053	0.183	0.567	0.569	0.357
	222	0.075	0.083	0.018	0.000	0.000	0.005
	224	0.021	0.076	0.042	0.433	0.431	0.167
	226	0.110	0.144	0.183	0.000	0.000	0.062
	228	0.123	0.091	0.201	0.000	0.000	0.019
	230	0.295	0.227	0.201	0.000	0.000	0.252
	232	0.233	0.326	0.173	0.000	0.000	0.138

Appendix 2. Frequency of microsatellite alleles in the *Apodemus flavicollis* population from Mouse Island over three years. Alleles are presented as an approximate length (in base pairs) of PCR product indicated by sequencer.

Locus	Allele	2003	2004	2008	Locus	Allele	2003	2004	2008
GACAD1A	128	0.030	0.011	0.000	MSAf-16	198	0.212	0.277	0.000
	130	0.030	0.074	0.000		228	0.061	0.064	0.025
	132	0.485	0.436	0.450		230	0.167	0.170	0.300
	134	0.030	0.021	0.050		234	0.318	0.223	0.575
	136	0.242	0.213	0.475		236	0.227	0.245	0.100
	138	0.136	0.213	0.025		238	0.015	0.011	0.000
	140	0.030	0.032	0.000		256	0.000	0.011	0.000
	150	0.015	0.000	0.000	MSAA-5	122	0.424	0.383	0.000
GTTC4A	188	0.000	0.021	0.025		140	0.348	0.394	0.000
	190	0.439	0.457	0.125		143	0.197	0.149	1.000
	196	0.000	0.011	0.100		146	0.015	0.000	0.000
	198	0.455	0.309	0.400		149	0.015	0.032	0.000
	200	0.000	0.011	0.050		152	0.000	0.043	0.000
	202	0.106	0.191	0.300	MSAA-6	212	0.712	0.713	0.925
						214	0.045	0.074	0.000
						216	0.197	0.160	0.075
						218	0.045	0.053	0.000