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Genetic variation and effective population size in an isolated population of the common hamster, *Cricetus cricetus*

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Abstract. An isolated population of the common hamster, forming the western border of the species range in Poland was analysed by the use of 16 microsatellite loci and the mtDNA control region in two consecutive generations. The genetic diversity and the effective population size in this population are low. We found the evidence for ancient bottleneck in this population, but the results of tests for recent reduction of Ne were ambiguous. However, population functions properly i.e. it is in HW equilibrium, Fis and relatedness coefficients do not indicate inbreeding. It indicates that even isolated and small populations of the common hamster have good chance of survival on the condition of the protection and restoration of the habitats. Moreover, the sex-related differences in dispersal in the common hamster were demonstrated through the relatedness analysis.

Key words: endangered species, inbreeding, relatedness coefficients, sex-related gene flow

Introduction

During the second half of the XX century the intensification of agriculture in Europe caused drastic changes in agriculture landscape and decline of biodiversity in the fields. High productivity in the intensive agriculture requires high input of fertilizers and pesticides, increased mechanization and the farm size and simplified crop rotation with low proportion of permanent crops. On the other hand, many small farms became hardly productive and were in consequence abandoned, which resulted in increased area of the fallow land (Henle et al. 2008). The changes in agriculture practice severely affected the inhabitants of the fields. Many plant and animal species connected with agriculture landscape became endangered or even extinct (Stoate et al. 2001). The destruction of habitats with traditional agriculture created the systems of fragmented, isolated populations. Small, isolated populations are confined to the remnant patches of habitat and very often they are hardly connected with any other populations by the gene flow. In result such populations have high probability of extinction (Reed & Frankham 2003).

The common hamster (*Cricetus cricetus* L.) is one of the species endangered by the agricultural changes. The hamsters in Europe are totally agriculture dependent (Nechay 2000). The common hamster is the only hamster species with wide European range reaching to Western Europe (Niethammer 1982) and it was the agriculture development that made such range expansion possible. The species was very successful in the fields with populations of high density. As a result it was considered a serious pest and was extirpated using rodenticides and hamster trappers (Nechay 2000). The breakdown of the population numbers was noticed first in Western Europe in the second half of the XX century and since that time the reduction in the distribution and fragmentation of the range were observed. In Poland, the species range decreased by 75 % (Ziomek & Banaszek 2007). The hamsters inhabited central and southern part of the country, while currently they can be found mostly in

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the Małopolska Upland and the Lublin Upland with Roztocze (Surdacki 1971, Ziomek & Banaszek 2007) (Fig. 1). The remaining parts of the range comprise isolated, small populations with very little probability of any gene flow (Banaszek et al. 2011a). Such populations were found in the Sandomierz Basin, Kraków-Częstochowa Upland and Upper Silesia and they form current western and southern range border in Poland (Ziomek & Banaszek 2007).

All the small, isolated populations belong to Pannonia lineage, one of two phylogeographic groups of the common hamster present in Poland (Banaszek et al. 2010), (Fig. 1). The protection of these populations is important as with the loss of them, the chances of survival of Pannonia lineage in Poland might be lowered. Whereas, the phylogeographic lineages in Poland should be treated as ESUs (evolutionary significant units) and MUs (management units) with significant genetic differentiation in mitochondrial and nuclear DNA and the evidence for the morphological differentiation (Banaszek et al. 2010, 2011a, b). The genetic diversity of the isolated populations that form the current border of the species range in Poland is very low. However, none of these populations showed clear effects of the recent bottleneck (Banaszek et al. 2011a).

As we aimed to study the viability of a small, isolated hamster population we selected one, from the Upper Silesia, located in Jaworzno city, which was most probably completely isolated, as the genetic differentiation (Fst values) with all the other analyzed hamster populations was never lower than the treshold 0.20, indicating total disruption of the gene flow (Banaszek et al. 2011a). The particular aims of the presented work were: 1) to assess the level of genetic variability of the population 2) to test for ancient and recent reduction in the population size 3) to estimate the effective population size 4) to estimate the inbreeding and relatedness coefficients.

Material and Methods

Sample collection

The Jaworzno site is situated in the Upper Silesia (Fig. 1). The hamster population was located there in 2004 and at that time it was the only one in this area and seemed to be the only one remaining in the Upper Silesia (Ziomek & Banaszek 2007). The population occurs on 10 ha area of a mosaic of narrow strips of fields and fallow land, which is enclosed from the west by a road and from the north by buildings of the Jeziorki settlement. However it is not isolated from other agrocenoses surrounding the plot. One hundred

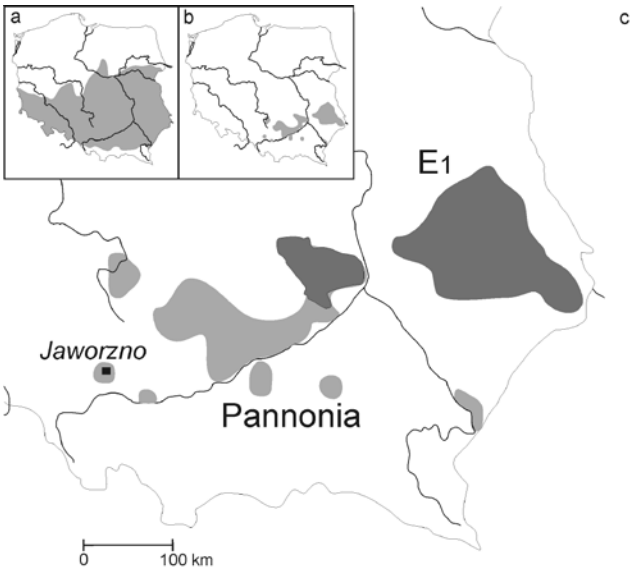

Fig. 1. *The common hamster (*Cricetus cricetus *L.) distribution in Poland. The species distribution before population crash after Surdacki (1971) (a) the present species range after Ziomek & Banaszek (2007) (b) the localization of the Jaworzno site (c) with the ranges of the phylogeographic lineages, Pannonia and E1 indicated (Banaszek et al. 2010).*

twelve active burrows were found throughout the 2007 year in this area.

A total of 66 hamsters were sampled during 2005-2007. In 2005 six hamsters were collected, one hamster was captured in 2006 and 59 hamsters in 2007. The sample of 2007 was collected throughout the season of hamsters activity (April-September) during field inspections every two weeks. Nine of the hamsters of the 2007 sample were adults collected in the spring or later in the season. In the spring of 2007 in the whole study area just ten active burrows were found. Of all the observed overwintering animals just one female avoided capture. Some of the individuals of 2005-2006 sample were observed or captured on the site during 2007 season. Summing up, the hamsters of 2005-2006 and the adults of 2007 were considered as the parental generation and the immatures of 2007 as the next generation. As we collected almost all overwintering animals in 2007 year, they are most probably the offspring of the captured parental generation. However, it is also possible that they are immigrants and descendants of other animals.

The hamsters were captured in live traps set near the hamsters' burrows. They were put under anaesthesia and the ear tips were clipped. All the animals were released. The capture and handling of animals in the field was conducted under the permissions of the Minister of the Environment DOPog.-402-02-54/04/

aj and DLOPiK-op/ogiz-4200/IV-3/815/08/aj Warsaw, Poland and the Local Ethics Committee on Animal Research in Białystok 2003/53 and 2007/69 Białystok, Poland.

DNA extraction and genotyping

The ear tips were frozen and the total genomic DNA was extracted from frozen tissues following the standard protocol supplied with the GenomicMini kit (A & A Biotechnology).

All the individuals were sexed in the field, however all the immatures were also sexed by the amplification of the *Sry* gene – successful amplification denotes males (Bryja & Konečný 2003). As positive control of PCR amplification the mtDNA *cytb* sequence was co-amplified (details in Banaszek et al. 2010). Seventeen microsatellite loci available for the common hamster: Ccrμ3, 4, 10, 11, 12, 13, 15, 17, 19 and 20 (Neumann & Jansmann 2004, AJ532553-AJ532554, AJ532556-AJ532563) and CriCriIPK-01, 03, 05, 06, 07, 09 and 12 (Jacob & Mammen 2006, AM167541, AM167543-AM167548) were used. Partial sequences of the mitochondrial control region (*ctr*) were obtained from all collected hamsters. The PCR profiles for microsatellite and *ctr* amplification, the method of microsatellite analysis and sequencing reactions for *ctr* were described in detail in Banaszek et al. (2009, 2010, 2011a).

Data analysis

A part of this material (n = 38) was genotyped and used to describe basic measures of diversity in Jaworzno population (Banaszek et al. 2011a). However, the amplification of all microsatellite loci in these individuals was repeated. In such case we were able to check for the genotyping errors (simple mistakes in genotyping) in the sample of 38 individuals and found none. The value of 0.01 % of genotyping errors was further used in maternity analysis in CERVUS 3.0.3 (Kalinowski et al. 2007). To resolve the problem of more complicated kinds of genotyping errors, we used MICRO-CHECKER (van Oosterhaut et al. 2004). This program discriminates between Hardy-Weinberg deviations caused by lack of panmixia and the effects of genotyping errors, such as large allele dropout, the presence of stutter peaks or null alleles. We found that CriCriIPK-07 locus showed the evidence of the general excess of homozygotes for most allele size classes which might indicate the presence of null allele. When the smaller sample was tested no null alleles were found (Banaszek et al. 2011a). It is possible that the presence of sibling groups in the larger sample gave the false evidence for null allele. However, as the parental and relatedness analyses are especially vulnerable to such kind of error, we decided to exclude the CriCriIPK-07 locus from further analyses. No evidence for scoring errors was found in all the other loci.

Basic measures of genetic diversity for Jaworzno population (mean number of alleles, observed and expected heterozygosity, inbreeding coefficient and HW equilibrium) were reported (Banaszek et al. 2011a). Here, the sample was increased to 66 individuals and the analysis followed the generations. The measures of diversity are given for two generations separately. The genetic variability indices (mean number of alleles and heterozygosities) and tests for departures from the Hardy-Weinberg equilibrium for the total population and generations were computed in ARLEQUIN 2.0.9 (Schneider et al. 2000). Moreover, the allelic richness, i.e. number of alleles corrected for different sample size in the generations, was calculated in FSTAT 2.9.3 (Goudet 1995). For multiple comparisons we used Bonferroni correction wherever applicable. The genetic diversity of the population was also described by Pemberton's d2, the individual-specific internal distance measure (Pemberton et al. 1998). The d2 is calculated as the squared distance in repeat units between the two alleles the individual has in the microsatellite locus, averaged over all loci the individual was scored. The d2 distance is a measure of a genetic distance between the gametes that formed the individual. The allele lenghts contain historical information about the time of the allele coalescence. As the microsatellites mutate mostly by the step-wise mutation model i.e. most mutations consist of an increase or decrease of one repeat, the greater distance (in repeat units) between the alleles, the less inbred is the individual (Pemberton et al. 1998). The mean d2 was calculated for generations to check for the changes in the level of the genetic diversity and also for the sexes to check for the contribution of the sexes to the total diversity.

The program BOTTLENECK 1.2.02 (Cornuet & Luikart 1996) was used to test for recent reductions in population size. We used both methods implemented in the program: 1) heterozygosity excess under the infinite allele model (IAM) and two phase mutation model (TPM) and (2) the distribution of alleles frequency. In the TPM we checked two options with different proportion (90 % and 70 %) of alleles attributed to stepwise mutation model (SMM). The statistical significance of the heterozygosity excess was tested by the Wilcoxon test. Moreover, the Garza

& Williamson (2001) M ratio was calculated in M P Val software. This ratio relates the number of alleles in locus in a given sample with the allelic range and is sensitive to reduction in population size. Moreover, lowered M value may indicate stronger or more ancient reductions in size than the BOTTLENECK software (Garza & Williamson 2001). The parameters of the two-phase mutation model for the M value calculation were the following: the proportion of one-step mutations p_s = 90 % and the average size of non one-step mutations $\Delta g = 3.5$, as advised by Garza & Williamson (2001) on the basis of simulations. Theta ($\Theta = 4Ne\mu$, where Ne – effective population size and μ – mutation rate) was set at 10, which indicates large pre-bottleneck population.

The effective population size was estimated by the TempoFs program under the temporal method of allele frequency shifts (Jorde & Ryman 2007). We used sampling plan I i.e. the hamsters were collected nondestructively and subsequently returned to the population. The sample interval was one generation.

We used CERVUS 3.0.3 for maternity analysis just to get some pairs of potential mothers and offspring to check the reliability of the relatedness analysis. The maternity analysis was performed using the likelihood-based approach in CERVUS. Six females were considered potential mothers of 50 immatures collected in the site in 2007. The simulations were used to estimate the critical difference in log-likelihood scores (DLOD) between the most likely and the second most likely candidate mother at a 95 % confidence level. Critical DLOD scores were generated from 100000 maternity simulations in which we assumed that 80 % of the females were caught in a population and that 100 % of the loci were typed, with a 0.01 % typing error. Both DLOD scores and number of mismatches between mother-offspring were considered in assigning parentage.

SPAGeDi-1.3 software (Hardy & Vekemans 2002) was used to estimate the pairwise coefficients of relatedness R of Queller & Goodnight (1989). To ensure that 16 microsatellite loci gave R close to expected values, the mean pairwise R was estimated for six mothers and their offspring assigned by CERVUS at a 95 % confidence level (expected R = 0.5). Then, estimates of pairwise relatedness were determined between all the hamsters collected in the site, between the hamsters belonging to separate generations and between sexes. The standard errors and 95 % confidence intervals for the relatedness estimates were calculated and the R values were judged to differ significantly from zero or 0.5, if the 95 % CIs did not overlap the values. The Levene's test was used to test the differences in the variance of the relatedness coefficients and the Mann-Whitney test to test the differences between the means of the relatedness coefficients between the sexes.

Results

Three hundred and sixty six base pairs of the mitochondrial control region were sequenced for 66 hamsters and no variability was found. All individuals possessed the Po2 haplotype (GenBank accession no EU 016107) (Banaszek et al. 2009).

Sixteen microsatellite loci were analyzed for 16 hamsters of parental generation and 50 hamsters of the subsequent generation of immatures. The frequencies of alleles for both generations were given in Appendix. Mean number of alleles was about three in both generations and it was slightly higher in offspring generation, which might indicate new mutations and/or migration of individuals (Table 1). We calculated also the allelic richness, which reflects differences in sample size and it was slightly lower in the immature generation than in adults (Table 1). Observed and expected heterozygosities did not differ significantly from each other in both generations. Both values are slightly lower in offspring than in adults (Table 1). In the parental generation there were no loci out of HWE and the global probability test for the HWE was not significant (Table 1). For the offspring generation the global test gave highly significant result, however, after Bonferroni correction, there was only one locus, CriCriIPK-06, out of HWE. The locus had significant excess of heterozygotes ($p = 0.0004$). However, most of heterozygotes were the youngsters, which could be siblings, as they were collected during three days near two neighbouring burrows. Without these individuals ($n = 15$), the offspring generation was found to be in HWE ($p = 0.36$). Fis values for both generations and for the total population indicated that there was no inbreeding (Table 1). Fis value calculated for the immatures with the group of siblings excluded, did not change significantly and equaled –0.105.

All the hamsters were genetically sexed by the amplification of the *Sry* gene. The results for adult individuals were always in agreement with sexing in the field showing the reliability of the molecular method. On the other hand, six immatures, which constitute over 10 % of the youngs, were incorrectly sexed in the field. This result shows that for the analyses where the sex of individual is important, the subadult hamsters should always be sexed by genetic methods. The sex proportion in the immatures was

shifted towards males and the shift was significant (1.5 : 1, $\chi^2 = 18.53$, df = 1, $p < 0.01$).

The mean d2 value did not differ significantly between generations (t = 1.402, df = 64, p = 0.16) and was slightly lower in immatures than in parental generation (Table 1). Immature males and females had similar mean d2 value, for adults the difference in mean d2 between sexes was greater and marginally significant (t = 1.923, df = 14, p = 0.075; Fig. 2).

no bottleneck was found when the percent of SMM in the TPM model was changed to 90 (p = 0.14). The Garza & Williamson (2001) M ratio was low in the Jaworzno population and equaled 0.635 (p = 0.0004). CERVUS assigned 10 immatures (20 %) to candidate mothers at the 95 % confidence level. The mean R value calculated in SPAGeDi for the mother-offspring pairs found by CERVUS was 0.396. For the material with very low allelic diversity and in effect of low

Table 1. *Genetic diversity measures for two generations in Jaworzno population of the common hamster. A – mean number of alleles, A_R – allelic richness, Ho and He – observed and expected heterozygosities, Fis – inbreeding coefficient, HWE – probability value for the Hardy-Weinberg equilibrium, d2 – the individual-specific internal distance measure of Pemberton et al. (1998).*

Generation	n	A	A_R	Ho	He	Fis	HWE	d2
Parental	16	3.06	3.01	0.56	0.52	–0.08	0.54	49.84
Offspring	50	3.25	2.81	0.48	0.43	–0.13	0.36*	39.78
Total	66	3.38	2.83	0.50	0.46	–0.12	0.42*	54.31

** For the offspring and total population calculated with the group of siblings excluded.*

The effective population size estimated from the temporal allele frequency shifts was Ne = 10 with 95 % CIs = 6-24. The ratio between Ne/N could then be estimated for 0.15.

The tests for recent population bottlenecks were performed for total population with immatures from two neighbouring burrows, which were the reason for the HW disequilibrium, excluded. The allele frequency distribution was L-shaped, as expected for non bottlenecked population. Under IAM (p = 0.005) and under TPM with 70 % of SMM (p = 0.01) we found that the population was bottlenecked. However,

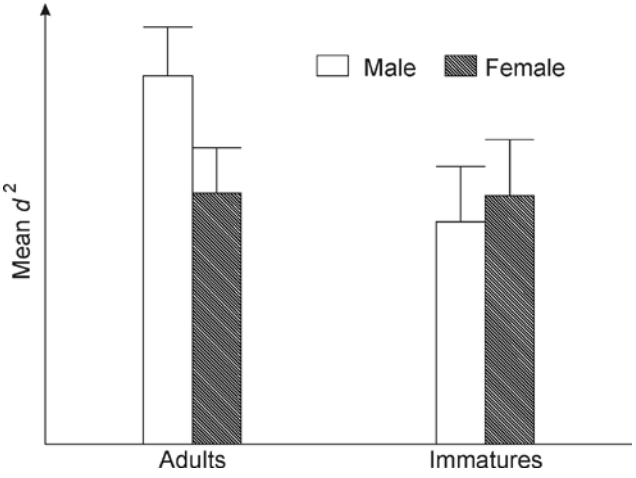

Fig. 2. *The difference between males and females in mean d2 value (± SE) in* Cricetus cricetus *Jaworzno population in two generations: parental and offspring.*

resolution power, we suppose that this value is close enough to the expected 0.5 relatedness coefficient between mothers and their offspring, especially that the 95 % CIs overlap 0.5. The mean relatedness value for total population was low (Table 2) and indicated that the hamsters were not related to each other. The relatedness values for males and females were very similar and the SE value indicated that there was great variation in the sample considering relatedness of individuals. The hamsters in parental generation showed negative value of the relatedness coefficient, which was significantly different from zero. Such result might indicate that the individuals are less related than two randomly chosen ones selected from the randomly mating population. The difference between adult females and males in the mean R value was not statistically significant (Mann-Whitney test, p = 0.22), however there was significant difference in the variance of R ($F_{1, 58} = 9.64$, p = 0.003), with the males showing greater differentiation in relatedness than the adult females. The immatures were, as expected, more related than parental generation and it were young males that showed positive value of R coefficient, while young females did not show closer relatedness to one another. All the calculations were performed with the immatures from two neighbouring burrows excluded. These youngsters were highly related (Table 2), most of them were siblings, although it is interesting that they were all males.

Table 2. *Mean (± SE) relatedness coefficients and their 95 % confidence intervals estimated for mothers and offspring found by CERVUS, among all individuals and among males and females in two generations of the common hamsters in Jaworzno population.*

Relatedness category	N	Number of pairs	R ± SE	95 % CIs
Mother – offspring		10	0.396 ± 0.196	0.270-0.509
Total	51	1 275	–0.017 ± 0.002	–0.004-0.001
Females	26	325	–0.023 ± 0.022	–0.058-0.015
Males	25	300	–0.022 ± 0.022	–0.065-0.018
Parental generation	16	120	–0.066 ± 0.002	–0.114 - –0.015
Females Ad	6	15	0.004 ± 0.064	–0.074-0.089
Males Ad	10	45	–0.079 ± 0.023	–0.180-0.009
Immatures	35	595	0.036 ± 0.026	0.013-0.064
Females juv	20	190	0.008 ± 0.056	–0.042-0.059
Males juv	15	105	0.089 ± 0.050	0.043-0.157
Immatures (two neighbouring burrows)	15	105	0.246 ± 0.066	0.184-0.314

Discussion

The common hamster population in Jaworzno is the example of very low genetic diversity and low effective population size. Non endangered species in Cricetidae typically have high numbers of alleles as for example *Microtus arvalis* with 12 up to 34 alleles in microsatellite loci (Borkowska & Ratkiewicz 2010). *Tscherskia triton*, which is not endangered and fairly abundant hamster species in China, has mean number of alleles typically higher than 10 (Song et al. 2005). Even some populations of *Cricetus cricetus*, although the species is constantly declining in Europe, show fairly high levels of genetic diversity, as for example the populations from Southern Moravia, Czech Republic, where mean number of 10 alleles was found (Neumann et al. 2004). In Jaworzno, we did not find the locus with more than five alleles, the mean number of alleles was three and the expected heterozygosity slightly lower than 0.5. The diversity measures for Jaworzno population are within the range of values characteristic for populations on the West European species boundary. The mean number of alleles for West European populations is from 1.6 to 5.4 and heterozygosity (He) is 0.11-0.50 (Neumann et al. 2004). The effective population size of Jaworzno is low and the proportion of the Ne/N is at maximum 0.15, if for N the sampled individuals (n = 66) were substituted. As the population is naturally more numerous than captured individuals, as indicated by the number of active burrows (n = 112), the Ne/N ratio is most probably lower. It is worth to mention that in microtine rodents, as for example the bank vole, the effective population sizes might be that high, that the Ne/N ratio is close to one (Borkowska & Ratkiewicz 2004). Very low levels of diversity were also visible in the mean d2 values. In *Tscherskia triton* the mean d2 value for total population was over 100 and for the males it was close to 200 (Song et al. 2005), while in the common hamsters from Jaworzno the mean d2 value for total population was lower than 50 and only for adult males it was somewhat higher (58.6).

The low genetic diversity in Jaworzno population is most probably the result of the long-term historical bottlenecks. The Pannonian populations in southern Poland originated from the area of Moravia and inhabited southern Poland as long ago as in the second stadial of Vistulian glaciation, which started 53.35 ka BP (Banaszek et al. 2010). However, as the habitat and climatic conditions during glaciation were harsh, the Pannonian populations in Poland most probably never reached very high numbers characteristic for hamsters. No demographic expansion was shown for Polish Pannonia in contrast to the populations of Pannonia in the Carpathian Basin (Neumann et al. 2005, Banaszek et al. 2010). Such scenario is supported by the detection of ancient bottleneck in our study based on M ratio, which was 0.635. This value is in the range of 0.6-0.65 M values characteristic for reduced populations, given as examples by Garza & Williamson (2001). Low M value for the common hamster population may be the result of repeated bottlenecks or long term low population size. The diversity levels for Polish Pannonia populations after northward migration from the Moravian region were most probably very low and for the mtDNA diversity potentially close to zero. The Jaworzno population shows a single haplotype for the mtDNA control region. This haplotype is characteristic for larger area of southern Poland, in which only one additional haplotype was found in one, out of four studied populations (Banaszek et al. 2009). On the other hand, quickly evolving microsatellites should

rebuild the higher diversity levels and most probably in some extent they did, as the Pannonian populations in Poland show high levels of interpopulation diversity (Banaszek et al. 2011a). Before the range of this lineage in Poland was fragmented, this diversity could be of intrapopulation kind. In contrast to ancient reductions in numbers, the recent genetic bottleneck cannot be clearly demonstrated for the Jaworzno population. The results of the BOTTLENECK tests are ambivalent. However, such results seem to be typical for the common hamster populations. The current bottlenecks are not revealed, even in populations on the western European border of the species range, which are presently critically endangered or became recently extinct as in the Netherlands (Neumann et al. 2004). The demographic bottleneck in the common hamster populations is evident all over Europe (Nechay 2000). The Jaworzno population forms current western range border in Poland, in the geographic region were most populations are already extinct (Surdacki 1971, Ziomek & Banaszek 2007). However, it is still possible that the demographic bottleneck did not cause the genetic bottleneck in some populations which survived.

Apart from generally low levels of genetic diversity, the Jaworzno population seems to function quite properly. It is in Hardy-Weinberg equilibrium, the Fis indices even indicate a trend towards outbred crossings, the relatedness coefficients for the whole population indicate that the individuals are not closely related and in consequence not inbred. The relatedness coefficient for the offspring generation is higher than for adults, however, it is natural, as part of individuals was sampled before they had a chance to disperse. The clear difference is visible between mean relatedness of young females, which are not related and young males, which showed positive mean R value. Potentially, the onset of dispersal for the sexes is different in the common hamster, with the young males dispersing later than females. Such difference was found in the ground squirrel, with young males dispersing two weeks later than females (Hanski & Selonen 2009), although in this species it was connected with longer distances of female dispersal, which is rather not the case for the common hamster. The relatedness in parental generation in Jaworzno is lower and the kinship between female pairs is closer than between males, although the difference is not statistically significant. However, the males are more differentiated than females in the pairwise relatedness coefficients, which indicates that part of them were migrants. This result shows that in the common hamster the males are the dispersing sex and females are more resident. It is in agreement with the information on the home ranges and dispersal in the common hamster acquired from the observations of the marked or radio tagged animals (Karaseva 1962, Kayser & Stubbe 2003). The different dispersal may be also connected with the male-biased sex ratio. According to the local resource competition hypothesis, bias in sex ratio tends towards dispersing sex in poor habitats (Cockburn et al. 2002), which would be in agreement with currently deteriorating habitats available for hamsters. The sex related male-biased gene flow was described in *Tscherskia triton* (Song et al. 2005). In *T. triton*, besides the difference in relatedness coefficients, the sex related gene flow was shown by differences in mean d2 value, i.e. the mean d2 value for males was higher within population than for females. In Jaworzno population the difference between the sexes was not statistically significant which might be in consequence of generally low mean d2 values. Summing up, regardless of low diversity levels there is no clear evidence of inbreeding. Only the lowered, in comparison with *T. triton*, values of the mean d2 may suggest some level of inbreeding, as the low d2 values demonstrate short coalescence times for alleles (Pemberton et al. 1998).

In conclusion, the Jaworzno population appears to be viable as the inbreeding was not detected in the population, although the effective population size and genetic diversity are low. The observational data confirm that in some years when the climatic conditions are favourable the population has vigorous reproduction, good recruitment and grows in numbers (Ziomek J., observations during 2004-2010). Although we described one local population in Poland, the conclusions are relevant to the whole European range of the common hamster, as in most parts it became heavily fragmented and consists of small populations (Weinhold 2008). Currently, the hamsters still can be saved by the protection of their habitats. To avoid further loss of the genetic variability, the possibility of gene flow, brought by connection of habitats is necessary. In the analysis of microsatellite diversity in the Polish part of the common hamster range we showed that the Pannonian populations were isolated from one another (Banaszek et al. 2011a). However, the recent, detailed inventarization in the Upper Silesia revealed the presence of some more local populations of the common hamster (Skowrońska et al. 2011). It is possible, that in microscale some natural gene flow is still present, as indicated by slightly negative Fis indices in the Jaworzno

population. Summing up, protection and restoration of available habitats, detailed field inventarizations with recognizing possible migration corridors and adjusting agriculture management to the demands of the species is necessary for the protection of hamster populations.

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Appendix. *The allele frequencies in 16 microsatellite loci analyzed in two consecutive generations (Gen1 n = 16, Gen2 n = 50) of the common hamster, Jaworzno population.*

Locus	Generation	Frequencies				
		Allele 1	Allele 2	Allele 3	Allele 4	Allele 5
Ccrμ3	Gen1	0.28	0.09	0.62		
	Gen2	0.09	0.06	0.85		
Ccrμ4	Gen1	0	0	0.03	0.31	0.66
	Gen2	0.01	0.01	0.01	0.15	0.82
Ccrμ10	Gen1	0.66	0.34			
	Gen2	0.64	0.36			
Ccrμ11	Gen1	0.37	0.22	0.41		
	Gen2	0.46	0.28	0.26		
Ccrμ12	Gen1	0.12	0.59	0.28		
	Gen2	0.11	0.75	0.14		
Ccrμ13	Gen1	0.41	0.44	0.15		
	Gen2	0.60	0.35	0.05		
Ccrμ15	Gen1	0.09	0.75	0.16		
	Gen2	0.06	0.93	0.01		

Ccrμ17	Gen1	0.03	0.66	0.31		
	Gen2	0	0.85	0.15		
Ccrμ19	Gen1	0.59	0.41			
	Gen2	0.73	0.27			
Ccrμ20	Gen1	0	0.10	0.34	0.06	0.50
	Gen2	0.01	0.03	0.41	0.05	0.50
CriCriIPK-01	Gen1	0.03	0.84	0.13		
	Gen2	0.04	0.78	0.18		
CriCriIPK-03	Gen1	0	0.22	0.78		
	Gen2	0.01	0.38	0.61		
CriCriIPK-05	Gen1	0.13	0.06	0.28	0.22	0.31
	Gen2	0.08	0.04	0.26	0.22	0.40
CriCriIPK-06	Gen1	0.50	0.03	0.41	0.06	
	Gen2	0.51	0.03	0.45	0.01	
CriCriIPK-09	Gen1	0.34	0.28	0.38	0	
	Gen2	0.15	0.47	0.37	0.01	
CriCriIPK-12	Gen1	0.13	0.84	0.03		
	Gen2	0.06	0.94	0		