

## Pedigree analyses of the Zatorska goose population

M. GRACZYK<sup>1</sup>, K. ANDRES<sup>2</sup>, E. KAPKOWSKA<sup>2</sup>, T. SZWACZKOWSKI<sup>1</sup>

<sup>1</sup>Department of Genetics and Animal Breeding, Poznan University of Life Sciences, Poznan, Poland

<sup>2</sup>Department of Swine and Small Animal Breeding, Institute of Animal Science, University of Agriculture in Cracow, Cracow, Poland

**ABSTRACT:** The structure of the Zatorska breed was estimated in the context of the realized conservation program. The level of genetic diversity and effective population size were estimated as well. The following parameters were evaluated: pedigree completeness index, genetic diversity, inbreeding level, individual increase in inbreeding, generation interval, and parameters connected with general condition of the population. The whole population of the Zatorska breed was housed in an experimental farm of the University of Agriculture in Cracow (Poland). Records were extracted from the studbook. Totally 5514 individuals hatched between 1990–2013 (2835 males and 2679 females) were included in the analysis. The average number of discrete generation equivalents reached 3.76, whereas the maximum discrete generation equivalent was 9.98. The average inbreeding level was low amounting to 1.46% for the whole population and 3.02% for the inbred individuals. The average pedigree completeness index for five generations reached 59.12%, for 10 generations 37.39%, and for all 16 generations it was 23.53%. The average effective population size was estimated from the family size variance and amounted to 67.36 individuals. It can be concluded that the conservation breeding program in the Zatorska goose has been going on well. This is confirmed by the magnitude of obtained estimates of parameters such as a low inbreeding level across generations under satisfactory pedigree completeness. On the other hand, the structure of a small population may be liable to fluctuations. Hence, continuous monitoring of the endangered population (including molecular control) seems to be necessary.

**Keywords:** waterfowl; genetic diversity; local breed; inbreeding level

### INTRODUCTION

Pedigree analysis is an important tool to describe the genetic variability and generation structure. This knowledge is fundamental for verification of the effectiveness of a conservation program (Honda et al. 2006; Korrida et al. 2013). Also, it is an integral part of the strategy of monitoring the structure of populations and mating programs (Pjontek et al. 2012; Borowska and Szwaczkowski 2015).

The breeding program for endangered livestock and poultry breeds defines the principle of conservation of genetic variability, inbreeding level, and reproduction system. Completeness of the pedigree is crucial for performing a pedigree analysis. Most reports concerning the pedigree analysis are conducted for livestock species like cattle,

horses, pigs, and sheep (Honda et al. 2006; Melka and Schenkel 2010; Pjontek et al. 2012; Mokhtari et al. 2014). It should be stressed that pedigree structure in geese is different compared to other poultry species. It is mainly influenced by a longer performance time than in chickens or ducks.

Over the last decades the local breeds have become important from the economic, social, and cultural point of view. They are well adapted to the local environmental conditions and are more resistant to diseases. These factors make local breeds more attractive to organic farms, and this in turn allows to provide consumers with a high quality product. Monitoring the genetic structure of the population, proper management, and promotion on the market are the key factors to preserve the domestic breeds for the future generations (Hodges 1987).

In the 2014 Domestic Animal Diversity Information System (DAD-IS) hosted by FAO (available at <http://dad.fao.org>) 14 851 breeds were collected, whereof 262 are breeds of geese from all over the world. Out of them, 170 breeds are reported for Europe and 19 (three extinct) for Poland (Rischkowsky and Pilling 2007; see also the 2014 DAD-IS report). The Zatorska goose is one of many breeds classified in the World Watch List for Domestic Animal Diversity as endangered-maintained (Scherf 2000). The breed was established in the 1950s. Until 1996 the population size had been 650–700 birds and the flock had belonged to the national pedigree breeding stock. Then the number of birds was reduced to 200–250 and the status of the population was changed to conservation flock included in the country's waterfowl genetic resources (Rabsztyn 2006).

The aim of the study was to estimate the structure of the Zatorska breed in the context of the realized conservation program. The level of genetic diversity and effective population size were estimated as well. Therefore, the following parameters were evaluated: pedigree completeness index, genetic diversity, inbreeding level, individual increase in inbreeding, generation interval, and parameters connected with general condition of the population.

## MATERIAL AND METHODS

**Birds.** Geese were housed in an experimental farm of the University of Agriculture in Cracow (Poland). Records were extracted from the stud-book. The data structure is as follows: ID number, sire number, dam number, sex, and date of hatching. In total, 5514 individuals hatched between 1990–2013 (2835 males and 2679 females) were included into the analysis.

The Zatorska goose is white feathered, its beak and legs are orange. Average body weight at 11 weeks of age is about 5.0 kg for males and 4.5 kg for females. The laying cycle lasts 20 weeks from January/February to May/June. In one cycle geese lay 35–45 eggs. The reproduction program usually involves a compilation of 20 groups consisting of one male and six females. The selection of birds is based on the promise of avoiding relatedness of common ancestors in at least two preceding generations. Production period lasts three years. In the second and third year offspring are obtained (Rabsztyn 2006).

## MATERIAL AND METHODS

The completeness of the pedigree information was examined using discrete generation equivalents ( $g_e$ ) calculated using CFC software tool (Sargolzaei et al. 2006), according to the formula given by Boichard et al. (1997):

$$g_e = \sum_{j=1}^{n_j} \left( \frac{1}{2} \right)^{g_{ij}}$$

where:

$n_j$  = number of known ancestors of the  $j$ -th individual

$g_{ij}$  = number of generations between the  $i$ -th ancestor and the  $j$ -th bird

Additionally, pedigree completeness index (PCI) proposed by MacCluer et al. (1983) was estimated as follows:

$$PCI = \frac{4C_{Sire}C_{Dam}}{C_{Sire}C_{Dam}}$$

where:

$C_{Sire}$ ,  $C_{Dam}$  = contribution of paternal and maternal lines, and  $C$  calculated according to the formula:

$$C = \frac{1}{g \sum_{i=1}^g a_i}$$

where:

$a_i$  = proportion of known ancestors in the  $i$ -th generation

$g$  = number of generations considered

The individual inbreeding coefficient  $F_i$  was estimated by the formula given by Meuwissen and Luo (1992):

$$F_i = A_{ii} - 1, A_{ii} = \sum_{j=1}^j L_{ij}^2 D_{jj}$$

where:

$A_{ii}$  = the  $i$ -th diagonal element of additive relationship matrix **A**

$L_{ij}$  = fraction of alleles derived from an ancestor

$D_{jj}$  = diagonal matrix containing additive genetic variances within the family

( $D_{jj} = 1$ , when both parents are unknown;  $D_{jj} = 0.75 - Fk_j/4$ , when only one parent  $k_j$  of the  $j$ -th individual is known;  $D_{jj} = 0.5 - (Fs_j + Fd_j)/4$  m, when both parents  $s_j$  and  $d_j$  are known.)

Individual increase in the inbreeding coefficient was estimated according to the formula described by Gutierrez and Goyache (2005):

doi: 10.17221/8560-CJAS

$$\Delta F_i = 1 - \sqrt[t-1]{1 - F_i}$$

where:

 $F_i$  = individual inbreeding coefficient $t$  = 'equivalent complete generations'

The above calculations were carried out using the software package ENDOG v4.8 (Gutierrez et al. 2010).

Effective number of founders, founder genome equivalent, and effective number of non-founders were estimated to evaluate genetic diversity in the population studied.

The effective number of founders ( $f_e$ ) (founders' equivalent) is defined as the number of founders which can produce a population with the same diversity of alleles if all founders contribute equally to each descendent generation (Lacy 1989). This parameter was estimated using the formula described by Sargolzaei et al. (2006):

$$f_e = \frac{1}{\sum_{j \in \text{FOUND}} \left( \frac{\sum_{i \in G} t_{ij}}{n_g} \right)^2}$$

where:

 $\text{FOUND}$  = number of founders $G$  = number of analyzed groups $n_g$  = number of individuals in a group

$t_{ij}$  = element of matrix  $\mathbf{T}$  representing the frequency of alleles of the  $i$ -th individual inherited from the  $j$ -th founder

The founder genome equivalent ( $f_{ge}$ ) was estimated as follows (Caballero and Toro 2000):

$$f_{ge} = \frac{1}{2\bar{f}_g}$$

where:

 $\bar{f}_g$  = average relationship in the group analyzed

The effective number of non-founders ( $N_{enf}$ ) showed the effect of genetic drift cumulated in the non-founder generation (Caballero and Toro 2000):

$$N_{enf} = \left( \frac{1}{f_{ge}} - \frac{1}{f_g} \right)^{-1}$$

where:

 $f_{ge}, f_g$  = explained above

The genetic diversity (GD) was estimated using the formula proposed by Lacy (1989, 1995):

$$GD = 1 - \frac{1}{2f_{ge}}, \quad GD' = 1 - \frac{1}{2f_e}, \quad GD' - GD = \frac{1}{2N_{enf}}$$

where:

 $f_e, f_{ge}, N_{enf}$  = explained above

The value of  $1 - GD$  is expressed as the loss of genetic diversity caused by genetic drift and bottleneck effect in the founder generation. The value of  $1 - GD'$  was estimated;  $GD'$  is expressed as the loss of genetic diversity caused by unequal share of alleles of founders in the population (Caballero and Toro 2000). The difference between  $GD'$  and  $GD$  shows a loss of genetic diversity due to genetic drift cumulated in the non-founder generation (Caballero and Toro 2000; Honda et al. 2006). The above parameters were calculated using the software package CFC (Sargolzaei et al. 2006).

Effective population size was estimated from the regression of the year of hatching and based on family size variance fitted to the hatching period of the reproductive individual (Hill 1979; Gutierrez and Goyache 2005):

$$\frac{1}{N_e} = \frac{1}{16ML} \left[ 2 + \sigma_{mm}^2 + 2 \left( \frac{M}{F} \right) \text{cov}(mm, mf) + \left( \frac{M}{F} \right)^2 \sigma_{mf}^2 \right] + \frac{1}{16FL} \left[ 2 + \left( \frac{F}{M} \right)^2 \sigma_{fm}^2 + 2 \left( \frac{F}{M} \right) \text{cov}(fm, ff) + \sigma_{ff}^2 \right]$$

where:

 $M, F$  = number of males and females $L$  = average generation interval $\sigma_{mm}^2, \sigma_{mf}^2$  = variances of the male and female offspring of a sire $\sigma_{fm}^2, \sigma_{ff}^2$  = variances of the male and female offspring of a dam $\text{cov}(mm, mf), \text{cov}(fm, ff)$  = respective covariances

It should be noticed that the family size of a parent consists in its number of sons and daughters kept for reproduction. Additionally, it should be stressed that the period of hatching of the reproductive individual is fitted by the average generation interval.

Generation intervals ( $L$ ) and age at first offspring ( $W$ ) were estimated for four pathways: sire-son ( $L_{ss}$ ), sire-daughter ( $L_{sd}$ ), dam-son ( $L_{ds}$ ), and dam-daughter ( $L_{dd}$ ). The generation interval is defined as the average age of parents at the birth of their progeny kept for reproduction (Honda et al. 2006):

$$L_{or} W = \frac{L_{ss} + L_{sd} + L_{ds} + L_{dd}}{4}$$

The above calculations were carried out using the software package ENDOG v4.8 (Gutierrez et al. 2010).

## RESULTS

The analysis of the population showed that out of 5514 individuals 370 were founders (with both unknown parents) and there were 793 individuals with only one known parent (792 with unknown dam and 1 with unknown sire). Thus, 78.91% of individuals had both parents known. Maximum 16 ancestral paths were traced, but it should be stressed that 7 of them were full generations. The number of animals in each path is presented in Figure 1. The average number of discrete generation equivalents reaches 3.76 whereas the maximum discrete generation equivalents are 9.98. Pedigree completeness index across consecutive generations is given in Figure 1. Please note that the generations are listed from the farthest to the closest generations (sires = 16, grandsires = 15, and so on). The average pedigree completeness index for 5 generations reached 59.12%, for 10 generations 37.39%, and for all 16 generations it was traced to 23.53%.

It should be noted that 2673 individuals had nonzero inbreeding level, which makes up for almost half of the population studied (48.48%). The average inbreeding level was low and for the whole population it was 1.46% whereas for inbred individuals it was 3.02%. In the whole population the average inbreeding coefficient for males amounted to 1.45% and 1.48% for females. In inbred individuals

Table 1. Inbreeding level in the population studied

Inbreeding coefficient (%)	Number of birds	Percentage of birds
0	2841	51.52
0–5	2306	41.82
5–10	289	5.24
10–15	37	0.67
15–20	6	0.11
20–25	25	0.46
25–30	10	0.18
Total	5514	100

these coefficients were 2.98 and 2.96%, respectively. Figure 2 shows the changes of the inbreeding level over the period analyzed. In the population studied the inbreeding level ranged from 0.05 to 28.13% (Table 1). It should be noted that the inbreeding coefficient does not exceed 10% for 47.06% of inbred individuals. The average individual increase in inbreeding was also low and amounted to 0.35% and in the group of inbred birds it was 0.72%. The inbreeding level changed over generations. The inbreeding level and the individual increase in inbreeding in consecutive generations traced are presented in Figure 1. The highest inbreeding level was registered in the 15<sup>th</sup> and 16<sup>th</sup> generation (4.58 and 4.48%, respectively). A similar situation was observed with the other parameter which reached 0.58 and 0.57% for the same generations.

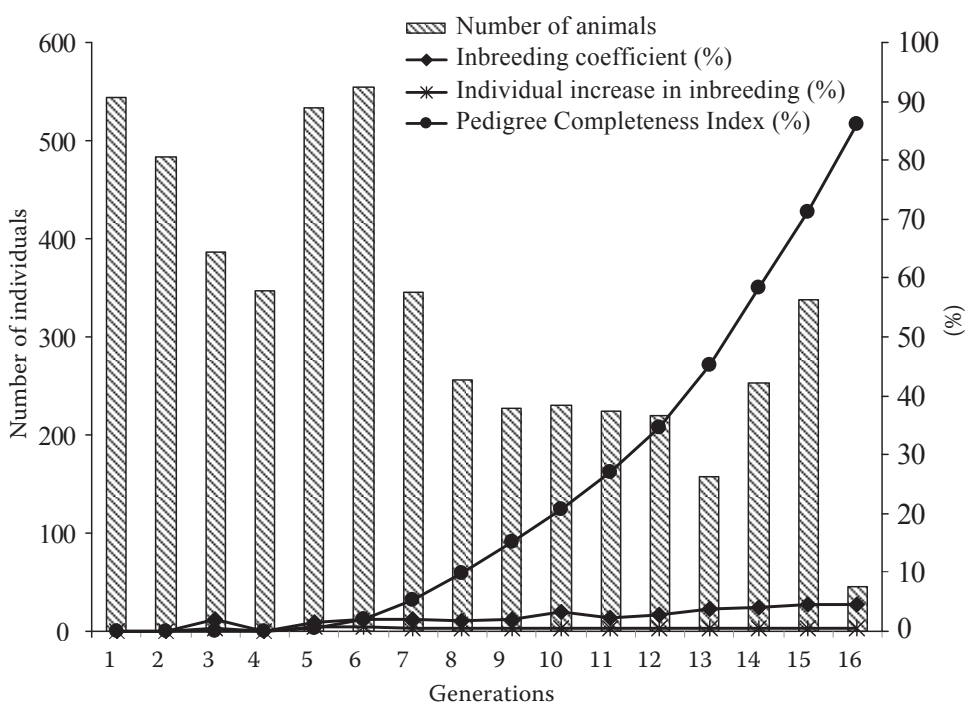
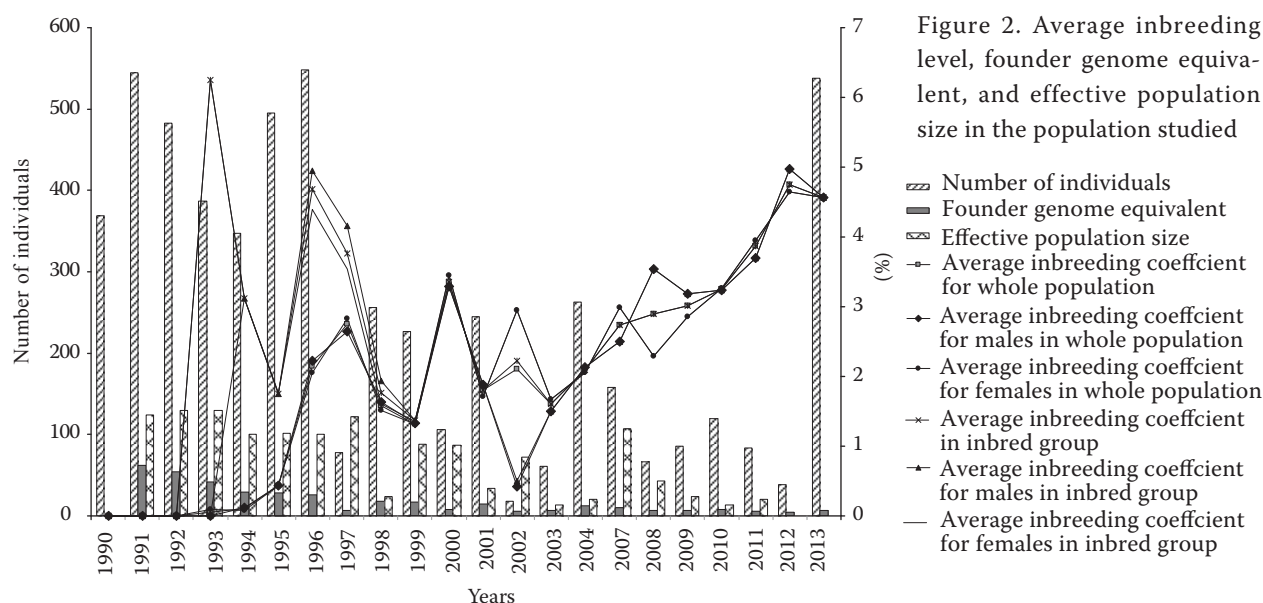


Figure 1. Pedigree completeness index, inbreeding level, and individual increase in inbreeding in 16 generations traced (the generations are listed from the farthest (1) to the closest (16))



doi: 10.17221/8560-CJAS



The estimated founder equivalent was 130.07. This means that this number of founders with the same proportion of alleles can give the same level of genetic diversity in the population analyzed. Figure 2 shows the distribution of founders' genome equivalent over the last 23 years. In the initial period, a high contribution of founders' genome equivalent was observed, but the trend stabilized in the last fifteen years. A loss of genetic diversity caused by genetic drift cumulated in non-founder generation was very low and amounted to 0.0097 (Figure 3). The fluctuation of the loss of genetic diversity caused by genetic drift may be connected with a considerably varied number of individuals in the analyzed period (Figure 2). As already mentioned, the status of the farm has changed

(from pedigree to conservation one). The level of genetic diversity took into account the impact of unequal contribution of founders' alleles and genetic drift. In the analyzed population these parameters were 0.9962 and 0.9860, respectively. Figure 3 shows the changes of genetic diversity affected by genetic drift and unequal contribution of founders' alleles in the mentioned period. Fluctuations of the estimated parameters were observed over time. Genetic diversity is caused by unequal contribution of founders' alleles which could be affected by changing number of individuals in the analyzed period (Figure 2).

In total, 277 sires (with 5143 progeny) and 802 dams (with 4352 progeny) were registered in the goose population, whereas 4435 individuals had no prog-

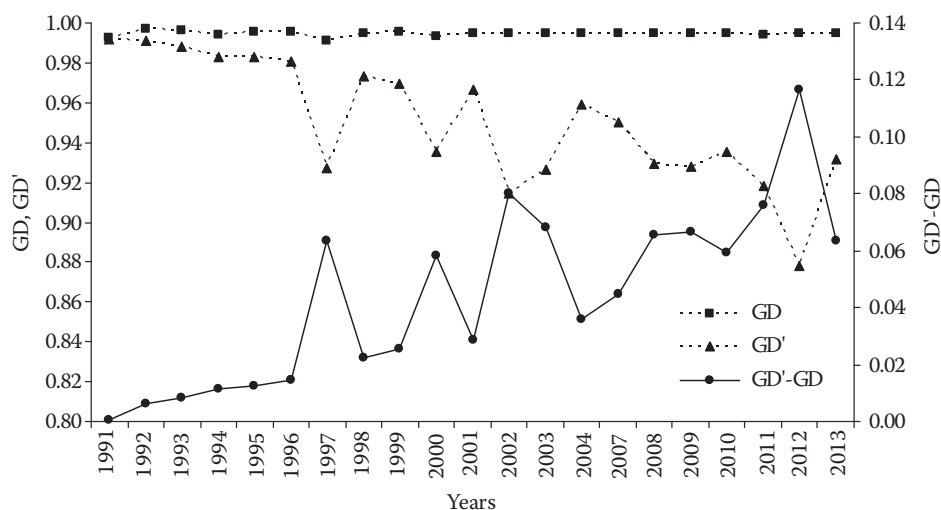


Figure 3. Loss of genetic diversity due to the genetic drift and unequal contribution of founders' genes ( $GD' - GD$ ) and genetic diversity caused by genetic drift ( $1 - GD$ ) and unequal contribution of founders' genes ( $1 - GD'$ ) in the analyzed period

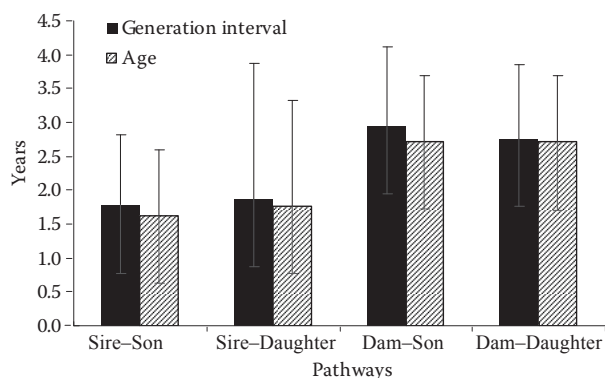


Figure 4. Average age at first offspring and generation interval in four paths with SD

eny. This means that 19.57% of individuals were parents. The number of offspring ranged from 1 to even 81. The highest number of progeny had one sire with 81 progeny and one dam with 23 offspring. From the perspective of the conservation program this fact may be problematic and directly connected with an increasing inbreeding level in the next generations. By the way, it can be noted that 915 full-sib groups existed with an average family size of 4.5 individuals (ranging from 2 to 15).

Figure 2 presents effective population size estimated from family size variance in the fitted period. The average effective population size was estimated from the family size variance and amounted to 67.36 individuals. A downward trend was observed. It may be connected with a decreasing number of birds. The effective population size calculated from the regression of year of hatching amounted to 111.04. The current population includes 471 individuals and is four times larger than the calculated minimum. This fact may slow down the loss of alleles in the native breed.

The average generation interval was 2.34 years with a standard deviation of 1.33 and the average age of parents at the birth of their offspring was 2.20 years (with SD 1.13). Age at first offspring and generation intervals for four pathways are presented in Figure 4. The shortest generation interval was marked on the sire-son path and reached 1.63 years, the longest on the dam-son path and reached 2.73 years.

## DISCUSSION

The discrete generation equivalent is considered as the best way to describe pedigree information

(Siderits et al. 2013). In the population studied completeness of the pedigree was on a satisfactory level. The recorded birds had known ancestors up to three previous generations, which is more than the minimum recommended by the conservation program. According to Fair et al. (2012) pedigree completeness could be connected with the maintaining system, obtaining of hatching eggs, nesting boxes, and system of collecting eggs, e.g. in ostrich population. On the other hand, the goose pedigree completeness is lower in comparison to livestock, for instance German Paint Horses (Siderits et al. 2013) and Iran-Black sheep (Mokhtari et al. 2014).

In endangered breeds, it is crucial to monitor the level of homozygosity. There are just a few reports concerning the evaluation of inbreeding level in geese. However, the mentioned studies were based on molecular markers (Qing-Ping et al. 2009; Li et al. 2012). The evaluation of inbreeding level from pedigree information usually regarded poultry breeds like chicken breeds, less frequently waterfowl. Szwaczkowski et al. (2007) noticed a low inbreeding level (under full pedigree information) in two goose strains from a pedigree farm. Some studies on inbreeding level vs rate have been performed in chicken populations. Lariviere et al. (2011) reported that only in 2 out of 41 native Belgian chicken breeds the inbreeding level per generation exceeded 5%. Low inbreeding level was noticed by Spalona et al. (2007) in 37 indigenous European chicken breeds where the coefficient does not exceed 1%. This indicates that the conservation program is properly managed and there is a minimum loss of genetic variability in the population.

Founders are defined as animals which initiated the population or as animals with both parents unknown. Founders' genome equivalent is always affected by the unequal proportion of contribution of founders and the number of founders' progeny. Founder genome equivalent is usually lower than founder equivalent and increases with a growth in the number of offspring (Lacy 1989). This dependence was clearly observed in the population studied. The dependence between founder genome equivalent and number of offspring was also reported, e.g. rapid decrease of hatched birds contributed to the decrease of founder genome equivalent.

The level of genetic diversity takes into account the impact of unequal contribution of the founder's

doi: 10.17221/8560-CJAS

genes and genetic drift (Lacy 1995). As already mentioned, in the Zatorska goose population genetic diversity was more than 99%. It is reflected in a very low inbreeding level in the whole population. Fluctuation of the inbreeding level was reported, but it should be noted that in the last 15 years (seven generations) it did not exceed 5%. It may slow down the loss of unique alleles in the Zatorska breed.

The results obtained in the present study must be perceived from the perspective of the conservation breeding program. One of the most important parameters estimated for any endangered population is effective population size. According to recommendation by FAO, a minimum effective population size is 50–100 individuals. This number of birds guarantees a continuity of the population. As already mentioned, the effective population size estimated from the family size variance was 67.36 individuals and from regression of the year of hatching for whole population it was 111.04. It could be perceived as a safety status of the population in context of the risk of allele loss and increase of inbreeding level in further generations.

Generation interval is important in endangered populations due to its connection with their inbreeding rate. As already mentioned, the average generation interval was 2.34 years which is correlated with the biology and reproduction system applied in the Zatorska goose. Differences between generation intervals for genders and geese are due to the inclusion of males in the first year of performance and females in the second year. A similar reproductive system is applied in main goose pedigree strains in Poland (Szwaczkowski et al. 2007). The present study provided useful information regarding the population structure and the status of genetic diversity in the local closed population.

## CONCLUSION

From the analyses performed it can be concluded that the conservative breeding program in the Zatorska goose is sufficiently realized. It is confirmed by the magnitude of obtained estimates of parameters such as low inbreeding level across generations under satisfactory pedigree completeness. On the other hand, the structure of a small population may be liable to fluctuations. Hence, continuous monitoring of the endangered population (including molecular control) seems to be necessary.

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Received: 2014–12–20

Accepted after corrections: 2015–06–23

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*Corresponding Author*

Prof. Dr. Tomasz Szwaczkowski, Poznan University of Life Sciences, Department of Genetics and Animal Breeding, Wolynska 33, 60-637 Poznan, Poland  
Phone: +48 61 848 72 49, e-mail: tszwaczkowski@gmail.com

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