**BALTIC FORESTRY**

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# Genetic Structure of Red Deer (*Cervus elaphus* L.) – A Review of the Population and its Reintroduction in Latvia

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*Abstract*

Today red deer (*Cervus elaphus* L.) are widely distributed, and are an economically important game species in Latvia and Europe. In Latvia, the species went extinct around the 10th century, and was absent for approximately 1000 years, until their reintroduction in the 19th century. This study uses literature and official statistics to describe changes in population size, distribution and harvest from the establishment of the wild population in the beginning of the 20th century until today. 100 red deer individuals from 3 regions, corresponding with the historical distribution of red deer in Latvia, were analyzed using 14 nuclear microsatellite markers.

Red deer population has undergone various changes since initial introductions. They have gone from small, isolated populations in the south and west of Latvia, to covering roughly 90% of all territory, and harvest has increased over time to the current maximum today of approximately 10000 per annum. The primary driver in range expansion was the translocations that took place from the west to the north- east regions between 1963 and 1988.

The analysed individuals were divided into regions based upon the historical information about deer populations in Latvia. We found no evidence of genetic differentiation between the regions, and the patterns of genetic diversity were also similar between the analysed regions. The results presented in this study indicate that currently there do not seem to be reproductive barriers between these populations. However, due to selective hunting, hybridisation and/or growth to infrastructure continued monitoring in the future is imperative. Genetic monitoring will help to detect changes in populations and to assist in the development of management strategies.

**Keywords:** red deer, reintroduction, population distribution, harvest, genetic structure

## Introduction

The red deer (*Cervus elaphus*) has a large distribution in Europe, and can also be found in some parts of Africa, Asia and North America. It is widely but somewhat patchily distributed throughout most of continental Europe, although it is absent from northern Fennoscandia and most of European Russia. It is present on a number of islands including the British Isles and Sardinia. It is the best studied and, together with the roe deer and wild boar, the most widespread European ungulate species (Mitchell-Jones et al. 1999, Lovari et al. 2008).

Genetic studies of red deer have detected three deeply divergent mitochondrial DNA lineages within Europe. The three lineages displayed a phylogeographical pattern dividing individuals into western European, eastern European and Mediterranean (Sardinia, Spain and Africa) groups, suggesting contraction into three separate refuges during the last glaciation (Skog et al. 2008).

In Europe six subspecies are usually listed (e.g. Dolan 1988): Swedish red deer (*C. e. elaphus*), Norwegian red deer (*C. e. atlanticus*), central European red deer (*C. e. hippelaphus*), British red deer (*C. e. scoticus*), Spanish red deer (*C. e. hispanicus*), and Corsican red deer (*C. e. corsicanus*). This is a novel study as it is the first one undertaking genetic analysis of ungulates in Latvia.

During the last 50 years, in Latvia and other parts of Europe red deer populations and their harvest have shown a general pattern of considerable increase regardless of ecological conditions, socio-cultural background or hunting system (Milner et al. 2006). It is a widespread and abundant species across much of its current range, although there is increasing fragmentation of populations

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in central Europe and the species has been lost from some areas due to overhunting, habitat loss (a result of agricultural intensification and urbanization) and deliberate restriction of red deer on forest management grounds (Burbaitė and Csányi 2010).

However, recovery of red deer populations has undoubtedly been facilitated by an immense number of reported translocations, although these themselves probably only reflect a small proportion of the actual translocations undertaken. Red deer, as a prestigious game species, have been translocated throughout Europe for centuries (Hartl et al. 2003). These processes were similar in Latvia to the rest of Europe.

Red deer generate a range of economic, social and environmental benefits. It is an important species for recreational stalking and venison production. Deer management and sporting activities provide employment, which helps to sustain local communities. They are also an important tourism asset, providing enjoyment to many visitors. Red deer have significant ecological value by providing an important source of food (in the form of live prey or carrion) for other animals. In addition, through their browsing, red deer influence vegetation composition and structure (Burbaitė and Csányi 2010). The most widely distributed and abundant prey species, which are the main prey for wolves in many places, are the following three species: red deer, roe deer and wild boar (Zlatanova et al. 2014). Wild ungulates (cervids and the wild boar, *Sus scrofa*) are the main food of wolves and lynxes in Latvia (Žunna 2009, Žunna 2011). They are not only relevant because they are the natural prey of wolves and European lynxes and thus represent an essential prey base in conservation measures directed at encouraging the expansion of these species, but they also represent an alternative to predation on livestock, which is one of the main sources of conflicts between large carnivores and humans (Apollonio et al. 2010).

The widespread distribution and large body size of red deer makes it an important game species (Koubek and Zima 1999). These ungulates represent an immense potential resource both in terms of biodiversity and also in economic terms. More than 5.2 million animals are harvested each year, which represents more than 120000 tons of meat, and a potential hunting revenue of several hundred million euros (Apollonio et al. 2010). At the same time, while they may be exploited in this way as sources of food and recreation, they may also have many negative impacts through damage to forests or agricultural crops, damage through heavy impacts on natural habitats, as vectors of disease, or through collisions with vehicles (Kenward and Putman 2011). Today, red deer are an important hunting resource for Latvia. In the 2009/2010 hunting season 5226 animals were harvested, equating to approximately 445 tons of game meat (Baumanis et al. 2011).

An important challenge for the future management of red deer in Europe is to improve the availability and the quality of population data on which management decisions are based. Despite being one of the largest and most economically important European mammals, with a wide distribution and a probable population size of 1–2 million animals, sound data on red deer populations and their dynamics are patchy, scarce or inconsistent. Against this background, harvesting must balance changing hunting requirements and capabilities with the increasing social and economic impacts of deer and other ungulates (Milner et al. 2006).

While the main anthropogenic impacts relevant to the conservation of genetic diversity of red deer in Europe were translocations and selective hunting in the past, probably the most important one for the future will be habitat fragmentation. This is especially pronounced in Central Europe, where expanding human infrastructure (settlements, roads etc.) causes populations to become more and more isolated from each other. As a consequence, gene flow is reduced, the effective population size decreases and genetic drift and inbreeding in small and discontinuously distributed populations result in the loss of genetic variability (Linnell and Zachos 2011). Ungulates, and red deer especially, have a significant impact on human interests (whether positive or negative), but are also themselves strongly influenced by human actions. By introduction and reintroduction, humans have established a number of exotic species, thereby markedly altering the distribution and abundance of many native European species, and through translocations have had an impact on population genetics (Kenward and Putman 2011).

The red deer is perhaps the species that has undergone the most extensive translocations (Hartl et al. 2003), mostly in attempts to improve trophy quality or establish hunting grounds. In particular in central Europe it is doubtful if there are any truly non-affected indigenous populations left as red deer have great dispersal capacities so that introduced animals or their offspring may disperse into other populations of the region. Whether this eventually leads to ecological problems due to disruption of adapted genotypes is unclear and perhaps not very likely, but the natural genetic pattern of red deer, at least at a regional scale, has been blurred or even destroyed (Linnell and Zachos 2011).

Red and roe deer are today among the most common European ungulates and are arguably, together with wild boar, the most important game species. Nonetheless, they are of conservation concern for a variety of reasons. Firstly, in both species, molecular analyses have identified genetically distinct populations in particular need of protection. Secondly, human influences – in particular selective hunting regimes, translocations and

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habitat fragmentation – have resulted in many challenges for the management and conservation of other local or regional populations. Red deer in particular also offer a well-studied example of hybridisation of an indigenous species with a closely related introduced exotic (in this case, sika deer) (Linnell and Zachos 2011).

In the past, perhaps, management systems for ungulates were rather inflexible (often rather unimaginative), and often directed towards achieving a clear, single objective (Kenward and Putman 2011).

Red deer in Latvia was originally introduced as a new game species as it provides high quality trophies and plenty of game meat. Typically, these were quality animals from Eastern and Central Europe. This was followed by movement of individuals into unoccupied local areas (Skriba 2011). Today, due to captive breeding, red deer is imported into Latvia from a variety of European countries. The origin and quality of these animals is often not known, therefore any animals that make it into the wild may adversely affect local populations. Landscape fragmentation is also becoming an increasingly important issue for Latvia; therefore, it is imperative to obtain detailed information regarding the Latvian deer population status and their genetic characteristics. This information will help guide future management decisions for deer populations.

## Material and Methods

To describe the development of the Latvian deer population we used literature, current data on estimated population size, data from hunted animals as well as the State Forest Service (SFS) official statistics.

Hunting in Latvia is generally organised as a ground leasing system typically for harvesting large game species like moose (*Alces alces*), red deer (*Cervus elaphus*), roe deer (*Capreolus capreolus*) and wild boar (*Sus scrofa*). The average size of a hunting ground is nearly 3,000 ha, since as much as 2,500 ha of woodland are required for the biggest game, moose hunting (Baumanis and Ozoliņš 2010). Control over the hunting and status of game populations is carried out by the State Forest Service (SFS). The SFS evaluates population status both at the country level and locally, i.e. it estimates the density, numbers and trends in population dynamics. For the main game species, the authority predicts the annual quotas and issues the shooting permits. Populations are estimated according to harvest results of previous seasons, while censuses are rarely performed. Quotas are negotiated among the hunters, local game and forestry experts, and land owners. Nevertheless, each individual of moose, deer or wild boar can only be taken by obtaining a distinct shooting permit which is valid only within a certain hunting ground and season.

### DNA analyses

DNA was extracted from deer muscle tissue samples collected from animals legally harvested by hunters for purposes other than this project. Samples were stored at –20 °C until DNA extraction from approximately 30 mg of muscle tissue using the E.Z.N.A Tissue DNA kit (Omega Bio-Tek/VWR).

A total of 12 autosomal microsatellite loci were analysed: BM888, BM4208, BM4513, BM1818, BMC1009 (Bishop et al. 1994), OarFCB193, OarFCB304 (Buchanan and Crawford 1993), OarCP26 (Ede, Pierson and Crawford 1995), MAF35 (Swarbrick, Buchanan and Crawford 1991), INRA11 (Vaiman et al. 1992), NVHRT16, NVHRT21 (Røed and Midthjell 1998), RT1, RT5 (Wilson et al. 1997). Polymerase chain reactions (PCR) were amplified in a volume of 10 microlitres containing 0.5 U HOT FIREPol DNA polymerase (Solis BioDyne), 1 ġL of 10 × concentrated PCR buffer, 2 mM MgCl2, 0.2 mM dNTP, 0.4 ġM final concentrations of each primer and 10-50 ng of DNA. Each forward primer was labeled with a different fluorophore (6-FAM, HEX or TMR) to facilitate visualization using capillary electrophoresis. The PCR profile consisted of an initial denaturation at 95 °C for 20 min followed by 35 cycles of denaturation at 94 °C for 30 s, annealing at 55 °C for 45 s, and extension at 72 °C for 60 s. Final extension of amplified DNA occurred at 72 °C for 10 min. All PCR reactions were carried out in an Eppendorf Mastercycler ep gradient thermal cycler. Amplification fragments were separated on an ABI Prism 3130xl Genetic Analyzer (Applied Biosystems) and visualized with GeneMapper 3.5. Population genetic parameters were calculated using GenAlEx 6.5 (Peakall and Smouse 2006, 2012) and Fstat (Goudet 2001).

The Bayesian clustering approach implemented the software STRUCTURE version 2.1 (Pritchard et al. 2000) to estimate the most likely number of clusters (K) into which the nSSR genotypes were assigned with certain likelihood. The population priors were not used, and a burn-in period of 50,000 iterations followed by 50,000 iterations was used. K was set from 1 to 30, and each run was replicated 20 times. The most likely number of clusters was identified by the delta K criterion with the STRUCTURE HARVESTER Web version 0.6.93 software (Earl and von Holdt 2012).

## Results

At around the 10th century red deer were extinct in Latvia and all neighbouring countries, namely Estonia, Lithuania and the western part of Russia. Red deer remained absent in these regions for the next 1000 years. The nearest countries, where red deer could be found during that time, were the Bialowieza forest in Poland, Carpathian Mountains, and possibly Prussia.

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The first deer parks were established in Latvia at the end of the 18th and the beginning of the 19th century. Animals were sourced from Poland, Germany, Prussia, Galicia, Austria and the Caucasus region. As animals came from these various areas, and there was likely high movement of individuals between parks, the original location that animals came from is not known. While it was generally believed during this time that red deer were only able to survive the Latvian climate within these parks, this theory was refuted as some animals managed to escape. Following the success of these individuals, hunters began to release red deer intentionally to the south-west regions of Latvia, and so the wild population of red deer in Latvia began at the start of the 20th century (Kalniņš 1943).

Before the Second World War in 1940 the number of free-living red deer was approximately 1300 animals; however, they still only remained within the south-west regions of Latvia in Kurzeme and Zemgale. During the war the population was reduced to some 390 individuals, and at the same time the overall territory became smaller and fragmented. Following the end of the war these small, isolated populations began to grow and combine until eventually around 1980 this became one common population.
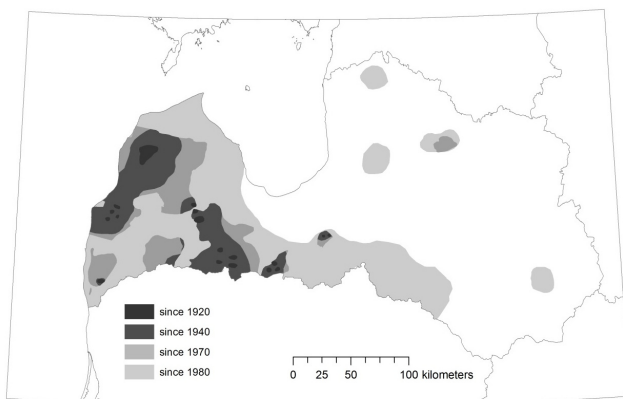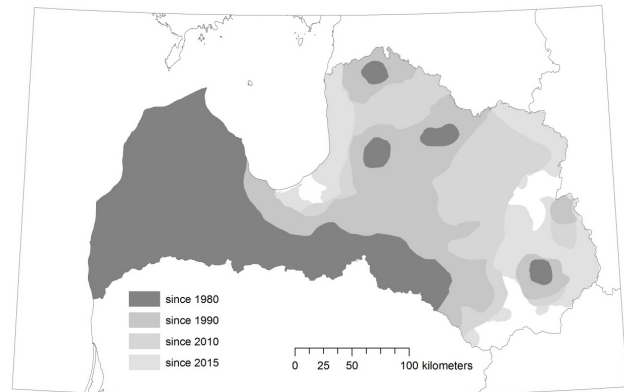


**Figure 1.** Historical spread of deer populations within Latvia between 1920 and 1980 (modified from Skriba 2011)

From 1963 until 1988 red deer were actively introduced to the currently unoccupied areas in the east. A total of 386 animals, consisting of 75 stags, 209 hinds and 102 calves were distributed between 14 separate sites (Skriba 2011). Initially most animals were from the western part of Latvia, with some individuals also being sourced from Bialowieza. As animals began to successfully establish themselves in these areas, animals were typically brought to neighbouring areas to encourage the population to spread.

Summarizing the history of the distribution of red deer in Latvia, it is characterized with having a strong

initial human influence with animals being sourced from various locations with different origins, taking place as two distinct phases roughly 50 years apart. Following the initial redistribution by anthropogenic activities, movement and spread of individuals followed natural processes. Today, the red deer population covers more than 90% of Latvia's territory.



**Figure 2.** Historical prevalence of deer population from 1980 until 2015 (modified from Skriba 2011)

Since their reintroduction into Latvia, red deer numbers have experienced significant change. The first legally hunted individuals were registered in 1910. After World War One the hunting of red deer restarted in 1925, but in 1939 before the Second World War hunting already reached more than 200 individuals per year. During the war, the red deer population suffered significantly both in terms of numbers and range. Therefore, red deer hunting restarted again only in 1953. During the subsequent years red deer numbers increased and reached their maximum in 1993 when there were more than roughly 30,000 individuals, and approximately 7,000 individuals were hunted. In the mid 1990's all even-toed ungulate species in Latvia observed a decrease in numbers, which continued until approximately the year 2000. During this period the estimated red deer population was approximately 20,000 individuals, but the hunting bag decreased to 2,600 animals in 1998. From 2000 until 2015 there was a rapid increase both to the total numbers and hunting bag, with the current population estimated at around 53,000 with an annual harvest of approximately 10,000 individuals. Changes to population level and hunting bag is summarised in Table 1.

A total of 100 individuals were analysed using 14 nuclear microsatellite markers, which were all polymorphic in the analysed red deer samples. The mean number of alleles per marker was 9.4, and the effective number of alleles was 5.4. The mean observed heterozygosity (0.605) was lower than mean expected heterozygosity

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**Table 1.** Changes to red deer population and harvest in Latvia

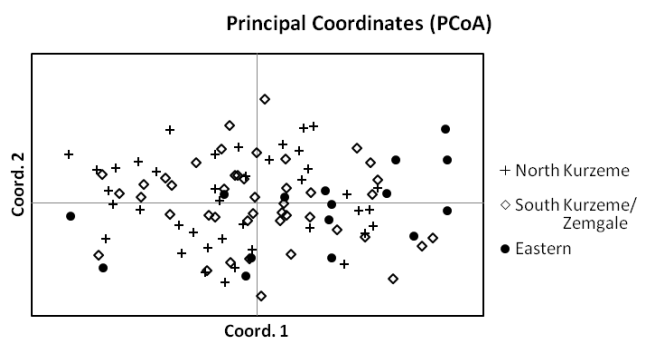| Year | Estimate | Harvest | Year | Estimate | Harvest | Year | Estimate | Harvest |
|------|----------|---------|------|----------|---------|------|----------|---------|
| 1900 | ≈ 30 | - | 1956 | 885 | 3 | 1986 | 16729 | 2959 |
| 1905 | ≈ 60 | - | 1957 | 867 | 5 | 1987 | 18723 | 4218 |
| 1910 | ≈ 200 | ≈ 10 | 1958 | 1032 | 8 | 1988 | 19534 | 5254 |
| 1914 | ≈ 500 | ≈ 10 | 1959 | 1429 | 14 | 1989 | 21577 | 5187 |
| 1923 | 110 | - | 1960 | 1545 | 12 | 1990 | 23180 | 6273 |
| 1924 | 250 | - | 1961 | 1755 | 17 | 1991 | 25311 | 6408 |
| 1925 | 320 | 1 | 1962 | 2010 | 47 | 1992 | 27669 | 6364 |
| 1926 | 415 | 2 | 1963 | 2154 | 47 | 1993 | 30345 | 6997 |
| 1927 | 430 | 3 | 1964 | 2432 | 79 | 1994 | 29195 | 5122 |
| 1928 | 460 | 3 | 1965 | 2791 | 117 | 1995 | 24902 | 4263 |
| 1929 | 495 | 5 | 1966 | 2965 | 142 | 1996 | 21763 | 2789 |
| 1930 | 570 | 1 | 1967 | 3264 | 152 | 1997 | 20462 | 2675 |
| 1931 | 640 | 4 | 1968 | 3622 | 188 | 1998 | 20026 | 2643 |
| 1932 | 760 | 3 | 1969 | 4150 | 211 | 1999 | 20445 | 2915 |
| 1933 | 1040 | 11 | 1970 | 4581 | 248 | 2000 | 21063 | 2942 |
| 1934 | 1050 | 34 | 1971 | 5097 | 288 | 2001 | 22533 | 3071 |
| 1935 | 1220 | 79 | 1972 | 5844 | 356 | 2002 | 22868 | 3282 |
| 1936 | 1470 | 220 | 1973 | 6487 | 512 | 2003 | 24562 | 3277 |
| 1937 | 1590 | 227 | 1974 | 7474 | 532 | 2004 | 26495 | 3467 |
| 1938 | 1560 | 220 | 1975 | 7894 | 882 | 2005 | 28363 | 3815 |
| 1939 | 1470 | 232 | 1976 | 7919 | 1111 | 2006 | 31279 | 3904 |
| 1940 | 1280 | no data | 1977 | 8754 | 1589 | 2007 | 32604 | 3830 |
| 1948 | ≈ 300 | - | 1978 | 9239 | 1852 | 2008 | 34428 | 4354 |
| 1949 | 390 | - | 1979 | 9754 | 1552 | 2009 | 37537 | 5251 |
| 1950 | 468 | - | 1980 | 9979 | 1739 | 2010 | 39817 | 4888 |
| 1951 | 532 | - | 1981 | 11001 | 1927 | 2011 | 43682 | 5606 |
| 1952 | 562 | - | 1982 | 11262 | 1783 | 2012 | 49622 | 7553 |
| 1953 | 630 | 2 | 1983 | 12544 | 1877 | 2013 | 51865 | 8735 |
| 1954 | 732 | 4 | 1984 | 13873 | 2088 | 2014 | 52461 | 9774 |
| 1955 | 903 | 6 | 1985 | 14812 | 2644 | 2015 | 53000 | no data |

The analysed red deer individuals were divided into 3 regional groups as follows: North Kurzeme (NK) (40 individuals), South Kurzeme/Zemgale (SK/Z) (44 individuals), and the Eastern region (E) (16 individuals) (Figure 3).
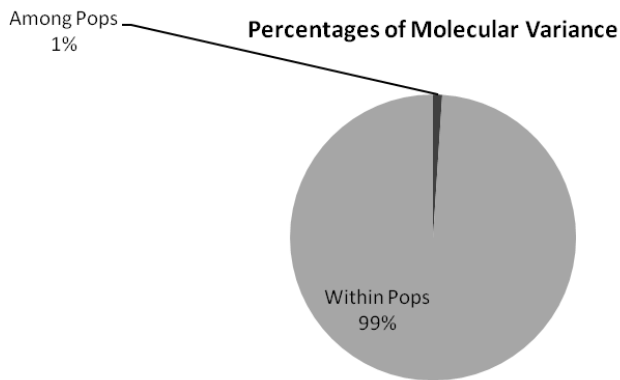


**Figure 3.** Site locations of analysed red deer genetic samples

The mean number of alleles in each regional population was similar (NK – 8.50, SK/Z – 8.29, E – 6.71), as well as the number of alleles with a frequency greater than 5% (NK – 5.86, SK/Z – 5.64, E – 5.50). When the differences in population sizes between the regions was taken into account (as implemented in the software Fstat), there were no significant differences in allelic richness or observed heterozygosity.

The Bayesian clustering approach implemented through the software STRUCTURE did not reveal any population structure or differentiation within or between the analyzed samples and regions. Principal coordinates analysis of the pairwise Nei genetic distances did not indicate any evidence of clustering according to the geographic regions (Figure 4). Analysis of molecular variance (AMOVA) also confirmed the low level of genetic differentiation between the populations, with only 1% of genetic diversity found between populations ($p = 0.001$) (Figure 5).

(0.761). The mean inbreeding coefficient $F$ over all markers was 0.190. However, the inbreeding coefficient $F$ was larger than 0.2 for four markers (BM888, OarCP26, BM4513, NVHRT21) indicating that these markers probably amplified null alleles, thus decreasing the observed heterozygosity. The mean inbreeding coefficient $F$ over the remaining 10 markers was 0.071 (Table 2).

**Table 2.** Genetic diversity parameters of the analysed nuclear microsatellite loci

| Locus | Na | Ne | Ho | He | F |
|-------|------|------|------|------|------|
| **BM888** | 8.000 | 3.547 | 0.394 | 0.718 | 0.451 |
| **BM4208** | 9.000 | 7.828 | 0.880 | 0.872 | -0.009 |
| **OarCP26** | 10.000 | 4.357 | 0.547 | 0.770 | 0.290 |
| **BM1818** | 9.000 | 5.248 | 0.790 | 0.809 | 0.024 |
| **OarFCB304** | 9.000 | 6.400 | 0.840 | 0.844 | 0.004 |
| **OarFCB193** | 10.000 | 4.017 | 0.626 | 0.751 | 0.166 |
| **BM1009** | 11.000 | 6.213 | 0.830 | 0.839 | 0.011 |
| **BM4513** | 17.000 | 9.935 | 0.374 | 0.899 | 0.585 |
| **MAF35** | 11.000 | 6.011 | 0.680 | 0.834 | 0.184 |
| **INRA11** | 10.000 | 6.307 | 0.750 | 0.841 | 0.109 |
| **NVHRT16** | 3.000 | 1.131 | 0.121 | 0.116 | -0.048 |
| **RT1** | 8.000 | 4.798 | 0.690 | 0.792 | 0.128 |
| **RT5** | 7.000 | 3.976 | 0.643 | 0.748 | 0.141 |
| **NVHRT21** | 9.000 | 5.728 | 0.305 | 0.825 | 0.630 |
| **mean** | 9.357 | 5.393 | 0.605 | 0.761 | 0.190 |



### Principal Coordinates (PCoA)

+ North Kurzeme
◇ South Kurzeme/Zemgale
● Eastern

**Figure 4.** Principal coordinates analysis of pairwise Nei genetic distances

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Among Pops
1%

**Percentages of Molecular Variance**

Within Pops
99%

**Figure 5.** Analysis of molecular variance (AMOVA) within and among the analyzed populations (Fst – 0.01, $p = 0.001$)

## Discussion and conclusions

In the last 50 years Latvia has observed increases to the overall distribution and harvest of red deer, a pattern that has been similar for most of Europe (Milner et al. 2006). The primary driver in range expansion was the translocations that took place from the west to the northeast regions between 1963 and 1988. Without these translocations the occupation of these areas through natural means would have been much slower. Today red deer likely occupy more than 90% of Latvia's territory, therefore translocations are no longer necessary and the movement of individuals between regions and to remaining unoccupied areas takes place through natural processes.

Latvia's red deer population is based on individuals that were brought from European countries between the end of the 19th and the beginning of the 20th centuries. In some areas of Europe there are instances of hybridization between red deer and sika deer, and this crossbreeding produces fertile offspring (Linnell and Zachos 2011). It is important to verify both the wild and domestic populations as there have currently been no studies to identify whether these hybrids are present in Latvia. If there are no hybrids currently in Latvia, then it is imperative to monitor and regulate individuals being brought from countries where hybrids may be present. Occasionally, hunters and deer farm owners will release individuals they deem as 'high quality' to improve the trophy quality of the nearby wild population. Taking this into account, the fact that individuals sometimes unintentionally escape, and also the lack of knowledge on genetic purity, there is a risk that these hybrid genes may make it into the wild population.

There are previous studies identifying the negative impacts of selective hunting to population quality (Linnell and Zachos 2011, Kenward and Putman 2011) There has been no evidence that hunting has had a negative impact on trophy quality over the last 50 years in Latvia (Baumanis 2013), however this needs to be monitored for in the future, and genetic analyses can be a useful tool in monitoring the quality of populations. The three main challenges for wildlife managers in Latvia the future are likely importation of hybrid deer, selective hunting, and the development of infrastructure which may contribute to habitat fragmentation.

The analysed individuals were divided into regions based upon the historical information about deer populations in Latvia. There was no evidence of genetic differentiation between the regions, and the patterns of genetic diversity were also similar between the analysed regions. We are unsure whether the results of this study are a result of similar initial populations in northern Kurzeme and south Kurzeme/Zemgale, or whether individuals were highly mixed between sites during the last 50 years. The inbreeding coefficient (Fis) was positive, indicating a lower amount of observed heterozygosity in comparison with the expected values. While this could be due to population structure and restricted geneflow leading to increased levels of inbreeding within the populations, it could also be explained by the amplification of null alleles by the utilised microsatellite markers, four of which showed very high levels of Fis. Some of the markers utilised in this study were overlapping with previous studies of red deer (Zachos 2003, Hmwe 2006, Haanes 2011), which red analysed deer populations throughout Europe. While direct comparisons of genetic polymorphism are difficult due to differences in sampling strategy and populations, overall, the patterns identified in this study are similar to the previous reports. Genetic diversity parameters for each microsatellite marker were presented in only one report (Zachos 2003), which indicated the possibility of null alleles for three markers (BM888, OarCP26, OarFCB304). Two of these markers also had high Fis values in our studied populations – BM888 (0.451) and OarCP26 (0.290). The third marker did not show any evidence for null alleles in our study (OarFCB304 – 0.004). Analysis of our data without the four markers with Fis values over 0.2 indicated that the population levels of inbreeding were not extremely high (0.071). Therefore, caution must be exercised when utilising these markers for genetic analyses in red deer, in particular for pedigree reconstruction, due to the possible presence of null alleles. Additional microsatellite markers should be tested to obtain more precise estimates of inbreeding and other genetic parameters in Latvian red deer populations.

The results obtained in this study did not detect significant population differentiation between the three analysed regions in Latvia. Two of these regions, North Kurzeme and South Kurzeme/Zemgale, have well established red deer populations for approximately 100 years,

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while the Eastern population is much more recent, being established after 1980. While the origin of these populations is not known, the results presented in this study indicate that currently there do not seem to be reproductive barriers between these populations. However, as previously mentioned, additional microsatellite markers should be tested in order to estimate levels of inbreeding more accurately. Analysis of Latvian red deer using mitochondrial markers will also provide additional information about the provenance and relatedness of these populations. In addition, there is molecular evidence about the hybridisation of red deer with sika deer in Western Europe (McDevitt et al. 2009, Smith et al. 2014) and there are also growing concerns about this in Latvia, but no data is currently available for Latvian deer populations. The use of mitochondrial and nuclear DNA markers will enable the assessment of the extent and frequency of inter-species deer hybridisation in Latvia.

This is the first survey of Latvian red deer populations with DNA markers, and these data will provide a baseline for further genetic monitoring in order to detect changes in populations and to assist in the development of management strategies.

## Acknowledgements

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