RESEARCH ARTICLE

Long-distance Eurasian lynx dispersal – a prospect for connecting native and reintroduced populations in Central Europe

Barbora Gajdárová[1,2] · Elisa Belotti[3,4] · Luděk Bufka[3] · Martin Duľa[5,6] · Oddmund Kleven[7] · Miroslav Kutal[5,6] · Jānis Ozoliņš[8] · Carsten Nowak[9] · Tobias E. Reiners[9] · Branislav Tám[10,11] · Josefa Volfová[6] · Jarmila Krojerová-Prokešová[1,12]

Abstract

Dispersal is a key process for the maintenance of intraspecific genetic diversity by ensuring gene flow within and between populations. Despite the ongoing expansion of large carnivores in Europe, lynx populations remain fragmented, isolated, and threatened by inbreeding and loss of genetic diversity. In the course of large carnivore monitoring in the Czech Republic, several biological samples of Eurasian lynx were collected outside the permanent occurrence of this species. Using microsatellite genotyping we identified these as four dispersing lynx males and applied multiple methods (Bayesian clustering in STRUCTURE, Principal Component Analysis (PCA), frequency-based method in GENECLASS2, and machine-learning framework in *assignPOP*) to assign them to possible source populations. For this we used genotypes from five European lynx populations: the Bohemian-Bavarian-Austrian (N = 36), Carpathian (N = 43), Scandinavian (N = 20), Baltic (N = 15), and Harz (N = 23) population. All four dispersers were successfully assigned to different source populations within Europe and each was recorded at a distance of more than 98 km from the edge of the distribution of the source population identified. Such movements are among the longest described for lynx in Central Europe to this point. The findings indicate the ability of lynx males to disperse in human-dominated landscape thus facilitation of these movements via creation and/or protection of potential migratory corridors together with protection of dispersing individuals should be of high importance in conservation of this iconic predator in Central Europe.

Keywords Eurasian lynx · Genetic monitoring · Long-distance dispersal · *Lynx lynx* · Microsatellites

✉ Barbora Gajdárová
gajdarova@ivb.cz

1 Institute of Vertebrate Biology of the Czech Academy of Sciences, Květná 8, 603 65 Brno, Czech Republic

2 Department of Botany and Zoology, Faculty of Science, Masaryk University, Kotlářská 2, 611 37 Brno, Czech Republic

3 The Administration of the Šumava National Park, The Division of Nature Protection, Sušická 399, 341 92 Kašperské Hory, Czech Republic

4 Faculty of Forestry and Wood Sciences, Czech University of Life Sciences Prague, Kamýcká 129, 165 00 Praha-Suchdol, Czech Republic

5 Department of Forest Ecology, Faculty of Forestry and Wood Technology, Mendel University in Brno, Zemědělská 3, 613 00 Brno, Czech Republic

6 Friends of the Earth Czech Republic, Olomouc branch, Dolní náměstí 38, 779 00 Olomouc, Czech Republic

7 Norwegian Institute for Nature Research (NINA), Høgskoleringen 9, NO-7034 Trondheim, Norway

8 The Latvian State Forest Research Institute SILAVA, Rīgas iela 111, Salaspils LV2169, Latvia

9 Center for Wildlife Genetics, Senckenberg Research Institute and Natural History Museum Frankfurt, Clamecystrasse 12, 63571 Gelnhausen, Germany

10 Zoological Department, National Zoological Garden Bojnice, Zámok a okolie 6, 97201 Bojnice, Slovakia

11 Faculty of Agrobiology and Food Resources, Department of Small Animal Science, Slovak University of Agriculture in Nitra, Tr. A. Hlinku 2, 949 76 Nitra, Slovakia

12 Department of Zoology, Fisheries, Hydrobiology and Apiculture, Faculty of AgriSciences, Mendel University in Brno, Zemědělská 1, 613 00 Brno, Czech Republic

Introduction

Dispersal has been recognised as a key process in the dynamics and evolution of natural populations. Through the redistribution of individuals, dispersal is the main factor leading to gene flow within and between populations and thus maintenance of genetic diversity (Bullock et al. 2002; Clobert et al. 2012). However, in human-dominated landscape, populations are often segregated to isolated patches surrounded by an unsuitable matrix, where movement between the patches can be challenging (Fahrig 2003; Seidler et al. 2015; Crooks et al. 2017).

Species with low densities but extensive home-ranges, such as large carnivores, are substantially vulnerable to anthropogenic changes in the landscape. A human-dominated landscape may pose a serious obstacle in particular for long-distance dispersers, reducing the frequency of such dispersal movements. That said, these long-distance dispersal events may often remain undetected, as they are difficult to record, particularly in elusive species (Trakhtenbrot et al. 2005).

Despite the ongoing expansion in numbers and distribution of large carnivores in Europe (Chapron et al. 2014), only a few long-distance movements have been recorded (Bartoń et al. 2019). Grey wolves *Canis lupus*, were recorded to disperse up to 1092 km (Wabakken et al. 2007) and brown bears *Ursus arctos*, up to 467 km (Støen et al. 2006). Compared to bear or wolf, the Eurasian lynx *Lynx lynx,* has been described as a conservative disperser (Zimmermann et al. 2005), although long-distance movements have been recorded, with maximum up to 428 km in Scandinavia (Samelius et al. 2012) and up to 200 km in the Alps (Groff et al. 2009). While the landscapes of the Alps and Scandinavia are comparably remote, very little is known about the ability of lynx to disperse within human-altered landscape (Schmidt 1998; Zimmermann et al. 2005, 2007; Samelius et al. 2012; Herrero et al. 2020). Generally, as many other solitary felids (Janečka et al. 2007; Gour et al. 2013; Fattebert et al. 2015; Wultsch et al. 2016), lynx seem to follow a male-biased dispersal pattern (Samelius et al. 2012; Krojerová-Prokešová et al. 2019), with males dispersing far more widely than females from a given source population, although this has not been unequivocally proven in all areas (Zimmermann et al. 2005, 2007; Herrero et al. 2020).

In Central Europe native populations of lynx occur only in the Western Carpathians and in north-eastern Poland, on the edge of the distribution range of the Baltic population. Besides these native populations, several populations have been established in central, western and southern Europe based on reintroduction of lynx mainly of West-Carpathian origin (Dinaric, Bohemian-Bavarian-Austrian (BBA),

Jura, Alpine, and Vosges-Palatinian populations; Kaczensky et al. 2013; Fig. 1). The population in the Harz Mountains in Germany and that in the Kampinoski National Park in central Poland were both based on the release of lynx of captive origin. Although reintroduction in Central Europe has already started in the 1970s (Kaczensky et al. 2013), established populations are mostly isolated and of small size (fewer than 200 or even less than 100 individuals; Chapron et al. 2014). Further, in each case, the number of released animals was low and in some cases the animals were even closely related. This has inevitably resulted in low levels of genetic diversity and an increased risk of inbreeding depression (Breitenmoser-Würsten and Obexer-Ruff 2003; Sindičić et al. 2013; Bull et al. 2016; Mueller et al. 2020). However, it has been shown that even single dispersing individuals may considerably enhance the genepool and population viability in small populations (Frankham et al. 2002; Bull et al. 2016; Åkesson et al. 2016).

In this study we thus focused on the detection of potential long-distance dispersers of Eurasian lynx within a Central European region, namely the Czech Republic. The country is occupied by the native Carpathian population in the north-east and the reintroduced BBA population in the south-west. The majority of the Czech landscape is highly urbanised with large open areas of agricultural land, dissected by busy motorways. The forested mountain areas, characteristically associated with species such as lynx, are located mainly around the borders. During extensive monitoring of large carnivore populations, biological samples of Eurasian lynx were collected outside the known areas of permanent occurrence of the lynx in the Czech Republic. Knowledge about the origin of these lynx individuals is important for understanding movement patterns within such a fragmented landscape towards reviewing the future conservation strategy for lynx in the entire Central European region as well as for planning for improvements in landscape permeability using mitigation measures like green bridges or underpasses/overpasses. The aim of this study was thus to assign the lynx samples of unknown origin to their possible source populations. We genotyped individuals recorded outside the main distribution range and lynx from potential source populations. We then applied a variety of assignment methods to determine the origin of the focal lynx individuals. Based on our findings, we discuss the conservation implications and potential for long-term natural connectivity of native and reintroduced lynx populations in a single Central European metapopulation.
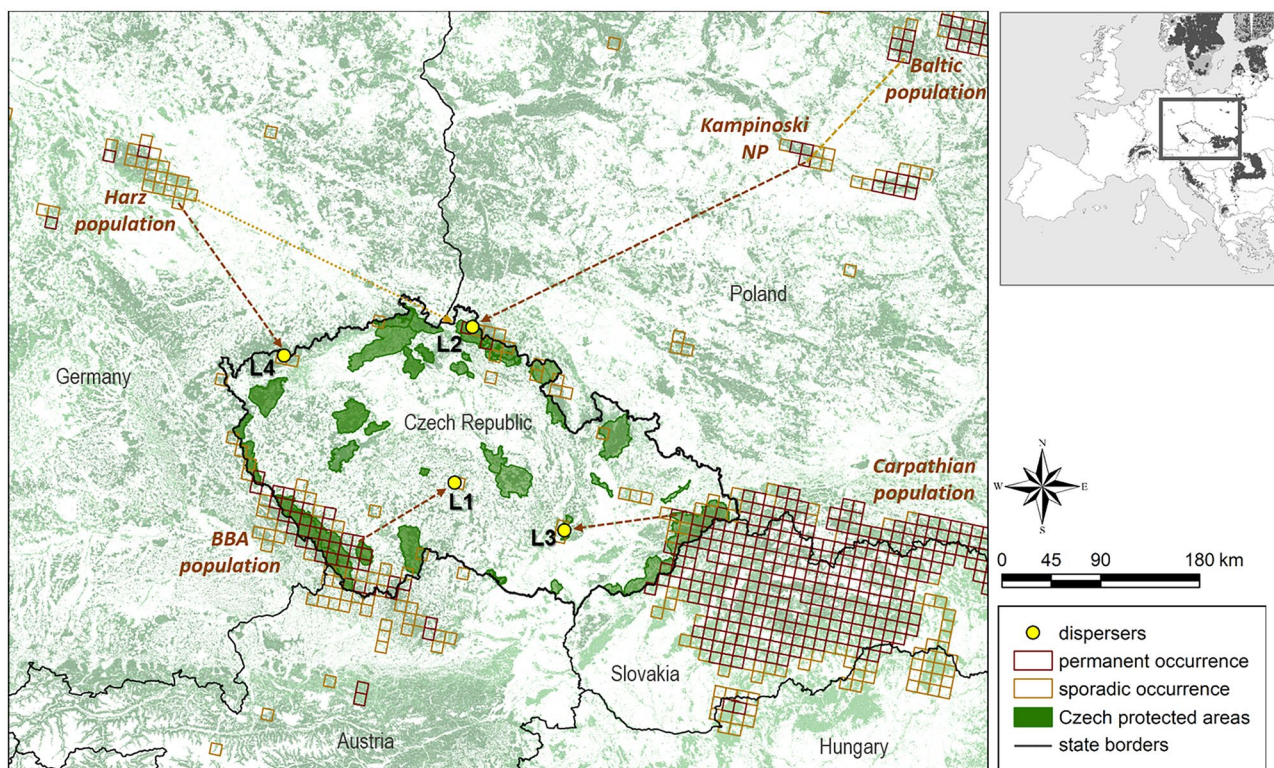
Fig. 1 Sampling sites of four individuals of unknown origin (L1 – D1 motorway, L2 – Jizera Mts, L3 – Moravian Karst, L4 – Ore Mts). Locations of two samples in Jizera Mts as well as locations of three samples in the Moravian Karst are overlapping as the samples were found close to each other. Permanent occurrence (brown squares) and sporadic occurrence (orange squares) of the Eurasian lynx in Central Europe is delineated according to Chapron et al. (2014) and for the Czech Republic updated according to Kutal et al. (2017). Dashed arrows show the possible course of the dispersal based on the results of genetic assignments. As a background we used the forest type cover layer from © Copernicus Land Monitoring Service 2018, European Environment Agency (EEA), European Union

Materials and methods

Sampling and genotyping

Evidence for the presence of lynx individuals of unknown origin ('dispersers') were found outside the permanent distribution range of the lynx in the Czech Republic (Fig. 1; Table 1). Five non-invasive scat samples (two in Jizera Mts, two in the Moravian Karst, one in Ore Mts) were collected during extensive monitoring of large carnivores, especially wolf. One blood sample was collected during immobilisation of lynx in the Moravian Karst and one tissue sample was taken from the carcass found after traffic collision on the D1 motorway. These samples were genotyped as described below.

Tissue, blood, and scat samples were stored in 96% ethanol. DNA from tissue and blood samples was extracted using the Genomic DNA Mini kit Tissue (Geneaid Biotech Ltd., New Taipei City, Taiwan). The QIAamp DNA Stool Mini

Table 1 Information about the four lynx dispersers of unknown origin

ID	Date	Locality	Sample type	Sex	Age (years)	Status
L1	07.10.2013	Czech-Moravian Highlands (D1 motorway)	Tissue	Male	2–3	†
L2	22.03.2016 30.04.2016	Jizera Mountains	Scat Scat	Male	?	?
L3	11.01.2017 12.01.2017 15.06.2017	Moravian Karst	Scat Scat Blood	Male	2–3	?
L4	06.12.2017	Ore Mountains	Scat	Male	?	?

Current status of the animal: † = dead; ? = unknown

Kit (Qiagen GmbH, Hilden, Germany) was used to extract DNA from faecal samples. Dedicated laboratories for DNA extraction and PCR setup were used and we followed strict rules and procedures to prevent contamination. In total, 15 microsatellite loci and the amelogenin marker for sex determination were used. PCR amplification of non-invasive samples was repeated in independent multiple parallels according to Taberlet et al. (1996) and Adams and Waits (2007) with requirements of a minimum of three positive amplifications for homozygotes and two for heterozygotes. We followed the same protocols for isolation, genotyping, PCR conditions and fragment analysis as are given in Krojerová-Prokešová et al. (2019).

To be able to assign the origin of possible dispersers, a reference dataset of 137 unique individual genotypes from five European populations (Fig. 1; Table S1; Table S2) was used. The reference samples were genotyped on the same microsatellite set of loci following the same protocols as described in Krojerová-Prokešová et al. (2019). The reference populations included the BBA (N = 36), Carpathian (the Czech Republic and Slovakia, N = 43), Scandinavian (Norway, N = 20), Baltic (Latvia, N = 15), and Harz (N = 23) population. Tissue samples (77) were taken from individuals found dead, poached, or legally hunted. Non-invasive samples (60; 42 scat, 17 hair, 1 blood samples) were collected during regular monitoring in the Bohemian Forest, the Carpathians, and the Harz Mountains (Table S1).

Analysis of population structure and individual assignment

Differentiation between comparative populations, necessary for correct assignment of dispersers, was quantified by pairwise F_{ST} based on Weir and Cockerham (1984) in the *hierfstat* package (Goudet 2005) in R software (R Core Team 2013). Confidence intervals of F_{ST} values (97.5% CI) were estimated using 999 bootstrap replicates in the same R package.

To infer the structure of populations and origin of dispersers, we used the Bayesian clustering analysis implemented in STRUCTURE v2.3.4 (Pritchard et al. 2000). The program was run with 10 independent runs for each value of K from 1 to 10, with 1 000 000 MCMC iterations and initial burn-in of 100 000. An admixture ancestry model with a correlated allele frequency model, without using sampling locations as prior information, was applied. The most optimal number of genetically distinct clusters was estimated using the ΔK method of Evanno et al. (2005) and evaluated also with the aid of estimators accounting for uneven sampling and hierarchical structure (Puechmaille 2016) in online application StructureSelector (Li and Liu 2018). The same application, integrating Clumpak program (Kopelman et al. 2015), was used to generate graphical representations for specific *K*.

As an alternative approach for investigation of population divergence and assignment of dispersers to potential source populations we performed a Principal Component Analysis (PCA) using package *adegenet v.2.0.0* (Jombart 2008) in R software.

Further, solely for individual assignment we applied the frequency-based method of Paetkau et al. (1995) within the program GENECLASS2 (Piry et al. 2004). Genotypes of all individuals from reference populations were used within one file while genotypes of dispersers were inputted separately. Probability of assignment was performed by simulating 100 000 individuals with Monte Carlo resampling method (Paetkau et al. 2004) and setting the type I errors to 0.05 (Piry et al. 2004).

Finally, to investigate the origin of the dispersers we applied a machine-learning approach in the R package *assignPOP* v1.1.4 (Chen et al. 2018). The program *assignPOP* was developed to overcome issues associated with non-independence and imbalance of datasets (Chen et al. 2018). The *assignPOP* approach included data evaluation, where all individuals from reference populations were randomly divided into training and test sets and the assignment accuracies were estimated via Monte-Carlo cross-validation based on the following parameters: proportion of individuals used in training set: 0.5, 0.7, and 0.9; proportion of loci used in training set: 0.1, 0.25, 0.5, and 1; loci sample method: F_{ST}; iterations: 1000; and model: linear discriminant analysis (LDA). Simulations performed best when all loci and individuals were used (Fig. S1). Therefore, for assignment test we used all individuals from reference populations and all microsatellite loci with chosen model LDA.

Results

Microsatellite genotyping

DNA isolation and subsequent genotyping was successful for all seven samples collected outside the 'permanent' distribution of lynx in the Czech Republic. Within these samples we identified four lynx males (Table 1; Fig. 1).

Structure of populations

The pairwise F_{ST} values confirmed differentiation among all possible source populations sampled (Table 2) with the lowest difference apparent between the BBA and Carpathian populations (F_{ST} = 0.13) and highest value between the BBA and Harz populations (F_{ST} = 0.40).

The Bayesian clustering analysis detected the most probable number of clusters at K = 2 and then at K = 5 (Fig. S2a). The best model for K = 5 was supported also by Puechmaille approach (Fig. S2b). Structure of two

clusters was in concordance with delineation of two lynx subspecies: the Northern lynx (*Lynx lynx lynx*) and the Carpathian lynx (*Lynx lynx carpathicus*). Further clustering at K = 5 corresponded to presumed structure of the five populations sampled (Fig. 2).

PCA analysis likewise clearly distinguished the five possible source populations (Fig. 3), although the differentiation between the Carpathian and the BBA populations (itself founded by lynx from the Carpathians) became clearer when PCA was repeated using only the samples from these two populations (Fig. S3).

Assignment of dispersers

Lynx L3 and L4 were unequivocally assigned to the same probable population of origin by all four methodological approaches – L3 to the Carpathian population (confirmed also by parentage analysis, Appendix 1) and L4 to the Harz population. L1 was classified as of BBA origin except for GENECLASS2 approach, where it had the highest probability to be of Carpathian origin (0.84); an origin from the BBA population however also received relatively high support (0.52). L2 was identified as Baltic lynx by majority

Table 2 Pairwise F_{ST} (97.5% CI) for five lynx populations

	BBA	CAR	SCA	BAL
CAR	0.13 (0.07–0.18)			
SCA	0.39 (0.29–0.50)	0.26 (0.19–0.33)		
BAL	0.35 (0.30–0.42)	0.22 (0.16–0.27)	0.22 (0.14–0.31)	
HAR	0.40 (0.30–0.49)	0.27 (0.19–0.36)	0.33 (0.26–0.41)	0.16 (0.10–0.22)

BBA Bohemian-Bavarian-Austrian population, *CAR* Carpathian population, *SCA* Scandinavian population, *BAL* Baltic population, *HAR* Harz population
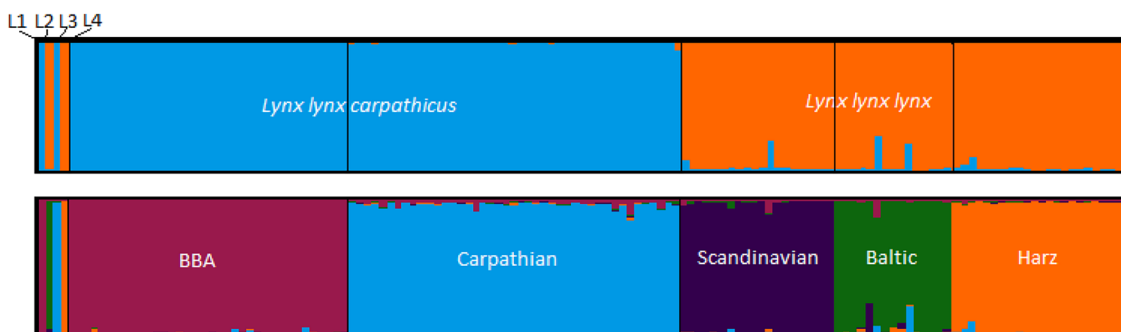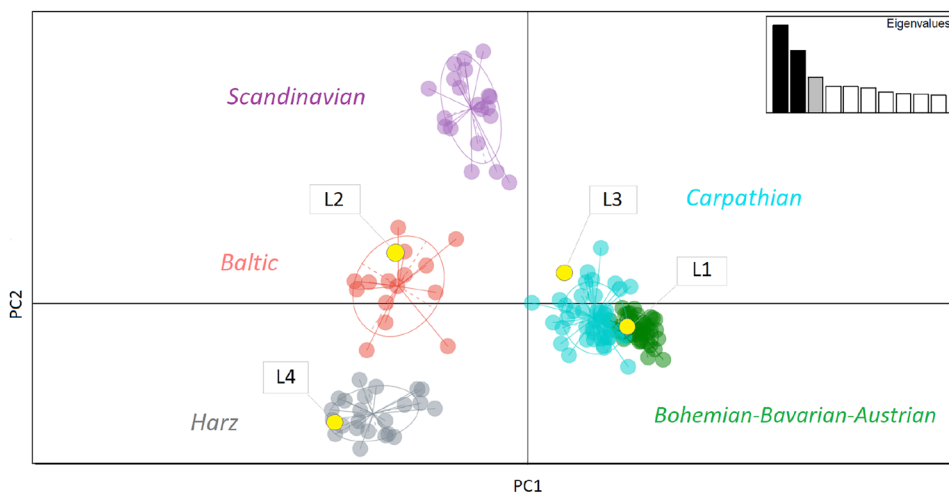

Fig. 2 Proportional membership of lynx dispersers (L1–L4) and lynx from five reference populations (N = 137) to K = 2 and K = 5 genetic clusters

Fig. 3 Principal Component Analysis (PCA) of four dispersers and 137 comparative lynx samples. Principal component axis 1 and axis 2 are shown, explaining 13.34% and 9.54% of the total variance, respectively

of approaches. The only exception was the result obtained in *assignPOP* showing almost equal probability for this individual to be assigned to the Baltic or Harz populations (Figs. 2, 3, 4; Table 3). All individuals were sampled at a distance of more than 98 km (98–456 km) from the border of the distribution range of the identified source population, indicating long-distance dispersal (Table 3).

Discussion

Population origin of dispersers

Except for two cases, all four genetic approaches provided consistent assignment for all dispersers. In the first case,

GENECLASS2 software was unable to distinguish between BBA and Carpathian origin of lynx L1. In the second case, program *assignPOP* assigned individual L2 both to Baltic and Harz population with almost the same probability. Differentiation between these two pairs of populations is in any case the most problematic (Table 2) as the BBA population originated from the Carpathian population and the Harz population was based on the release of captive lynx of mixed origin (von Arx et al. 2009), potentially including also ancestors from the Baltic region.

According to the majority of approaches, male L1, found dead on the D1 motorway in Central Bohemia, originated from the BBA population (103 km straight line distance from the BBA distribution range) from where it is presumed he was unsuccessfully trying to disperse. D1 motorway (the
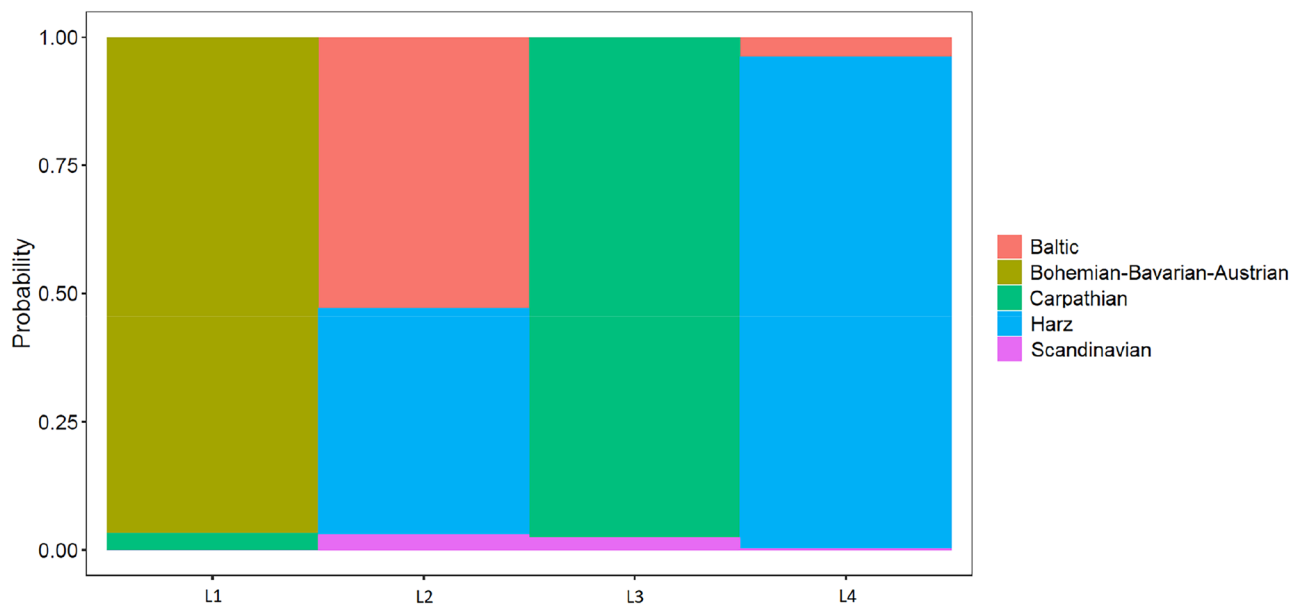

Fig. 4 Probabilities of individual assignment test in *assignPOP* for all four dispersers (L1–L4)

Table 3 Comparison of four approaches for the assignment of dispersers to their putative population of origin

Individual ID	Sampling site	STRUCTURE q ≥ 0.95	PCA	GENECLASS2 (P-value)	*assignPOP* (Pvalue)	The most likely population origin	Estimated minimum dispersal distance* (km)
L1	Czech-Moravian Highlands	BBA	BBA	CAR (0.84) BBA (0.52)	BBA (0.97)	**BBA**	~ 103
L2	Jizera Mountains	BAL	BAL	BAL (0.39)	BAL (0.53) HAR (0.44)	**BAL**	~ 337 (456/283)
L3	Moravian Karst	CAR	CAR	CAR (0.41)	CAR (0.98)	**CAR**	~ 98 (122)
L4	Ore Mountains	HAR	HAR	HAR (0.63)	HAR (0.96)	**HAR**	~ 169

*Estimated minimum distance is the distance between sampling site and the border of the distribution range of the presumed source population measured in ArcGIS 9.3.1 as a straight-line distance to the centroid of the nearest EEA square with continuous lynx occurrence. In the case of L2 a distance of 337 km corresponds to the population in Kampinoski NP, 456 km to the Baltic population and 283 km to the Harz population. For L3, a distance of 122 km corresponds to the distance to the nearest EEA square of its maternal home range (Appendix 1)

main motorway of the Czech Republic) is known to represent an important migration barrier for large mammals (Anděl et al. 2010).

During the seventeenth and eighteenth century lynx were believed to be exterminated in Jizera Mts and in adjacent Krkonoše Mts, but single individuals were spotted there again in 2002 and 2006, respectively (Flousek et al. 2014). Similarly, on the other side of the border with Poland, single lynx have been recorded between 2000 and 2018 (Mysłajek et al. 2019). Due to the mountain wildlife corridor with suitable habitat from the Western Carpathians to this area, it was believed that these lynx are dispersers from the Carpathians (Kratochvíl and Vala 1968). Similarly, lynx L2 sampled in Jizera Mts in the northern part of the Czech Republic was expected to be of Carpathian origin. Surprisingly, our genetic data do not support this presumption as lynx L2 was classified as a Baltic lynx. The edge of the native Baltic population in the north-eastern Poland is 456 km far away from the sampling site of the L2 and to reach the Jizera Mts, the individual would have had to cross highly urbanised central Poland with disrupted migration corridors and many barriers. As an alternative, it is perhaps possible that the smaller population occurring in Kampinoski National Park in central Poland (337 km straight line distance), could have been the possible source of this lynx. This population was founded between 1993 and 2000 (Böer et al. 2000), when 31 individuals from zoos and wildlife parks in Germany, Sweden and Finland were released there. Unfortunately, more detailed information about their ancestry is missing (Böer et al. 2000), thus we can only speculate if lynx of the Baltic origin may have been released there. An origin of L2 from within the Kampinoski National Park may, however, be supported by observations of the high dispersal ability of the original lynx released in that area. Soon after their release some of the lynx dispersed between 10–50 km to adjacent nature reserves (Böer et al. 2000), single individuals even up to 160 km (Reklewski 2006).

There is also a possibility that lynx L2 could come from the Harz population, as was indicated in *assignPOP,* but this is not the most parsimonious explanation and 3/4 of used approaches do not support this hypothesis. The problem with the correct assignment of L2 in *assignPOP* may also be influenced by the reference samples used for the Baltic population. We used 15 samples from Latvia, but recent genetic analyses revealed slight differentiation of the Baltic population in Poland to that in the central part of the distribution range (Ratkiewicz et al. 2014; Lucena-Perez et al. 2020).

Lynx L3 in the Moravian Karst was firstly recorded in September 2016. After that the lynx was captured there, fitted with a GPS collar and radio-tracked from June 2017 till June 2018 (Duľa and Krofel 2020). During autumn 2018, after losing his GPS collar, he disappeared from all monitoring sites established in the whole area, where he was previously regularly observed using camera traps (Duľa, pers. comm.). Our results showed that lynx L3 was of Carpathian origin (minimum dispersal distance 98 km), who, after dispersal, had settled in an area halfway between the Carpathian and BBA populations, suggesting this area might constitute a possible stepping-stone population between these two isolated populations. The results of the genetic assignment were further supported by parentage analysis (Appendix 1), which identified the parent pair of L3 in the Moravian-Silesian Beskids in the Western Carpathians. The pair of lynx identified as the parents is one of a few pairs who gave birth to the majority of juveniles in this area during 2011–2017 (Krojerová-Prokešová et al. 2019) and the centre of its maternal home range was in the straight line distance of 122 km from the site in the Moravian Karst where L3 had been recorded.

There are just a few documented records of lynx since 2017 in Ore Mountains, where the non-invasive sample from L4 was collected. It was initially expected the lynx would have come from the BBA population along the forested borderland between the Czech Republic and Germany, but genetic assignment suggested its origin in the Harz Mts in Germany (169 km distant). This movement shows the potential for gene flow between the BBA and the Harz population, both isolated populations with low (Bull et al. 2016), or declining levels of genetic diversity (Mueller et al. 2020). Admixture between different subspecies could pose a risk because it can lead to low or in an extreme case zero fitness of progeny through loss of local adaptations (Lynch 1991). On the other hand, fitness can be enhanced due to new genetic combinations and increased adaptive potential (i.e., heterosis), especially in inbred populations (Anderson 1949; Abbott et al. 2013; Frankham 2015). In any case, the effect of potential future crossbreeding of the expanding Harz population and neighbouring BBA population, should be closely monitored, since of all the populations included in this study these are the most genetically distinct from one another ($F_{ST} = 0.4$).

Factors affecting dispersal

Assuming the origins of the four lynx individuals sampled in the Czech Republic are accurately identified, the distances travelled from the population of presumed origin (98–456 km) are among the longest described, so far, for lynx dispersal events in Central Europe. For Jura and Alpine populations, the mean dispersal length of subadult lynx is reported at 26–63 km (max. 129 km; Zimmermann et al. 2005). Long-distance dispersals (up to 124 km) are known from the central part of the Baltic population (Latvia, Estonia). The longest dispersal distances, with mean 47–148 km (up to 428 km), have been recorded in Scandinavia, where prey density is lower and home ranges are

larger in comparison to other European populations (Samelius et al. 2012).

All four dispersers were males, which is consistent with the general expectation of male-biased dispersal in polygynous mammals (Greenwood 1980; Dobson 1982) and has previously been reported in the Eurasian lynx (Samelius et al. 2012; Krojerová-Prokešová et al. 2019) as well as in other felids, e.g., leopard (Fattebert et al. 2015), jaguar (Wultsch et al. 2016), bobcat (Janečka et al. 2007), and tiger (Gour et al. 2013). Female lynx seem to disperse less frequently and for shorter distances than males, even though male-biased dispersal was not confirmed in some areas (Zimmermann et al. 2005, 2007; Herrero et al. 2020). Except for Northern Europe (Fennoscandia, Baltic states), where dispersal distances are generally longer (Samelius et al. 2012; Bagrade et al. 2016; Herrero et al. 2020), there seem to be no published records about female dispersal longer than 100 km (see more details given in the supplementary material of the review of Bartoń et al. (2019)). The reluctance of female lynx to disperse over longer distances, particularly in fragmented landscapes, thus reduces the species ability to colonise new areas (Port et al. 2020).

Reported expansion of the distributional range of large carnivores in Europe relates primarily to distribution of wolves and bears (Chapron et al. 2014); by comparison, lynx populations are mostly stable or decreasing and only three populations are slowly increasing in numbers and range (Alpine, Jura and Harz population; Large Carnivore Initiative for Europe 2020). Except for the recent sporadic occurrence of a lynx in the military area Libavá (Kutal et al. 2017), lynx L3 represents the only well-documented expansion of lynx westwards from the Carpathians despite the presence of suitable habitats in neighbouring areas, e.g. mountains along the Czech-Polish border – Jeseníky Mts, the Orlické Mts, Krkonoše Mts and Jizera Mts. Human intervention (traffic mortality, poaching) at the population periphery probably plays a considerable role in limiting the West-Carpathian lynx expansion (Krojerová-Prokešová et al. 2019).

Conservation implications

To facilitate natural movement between lynx populations and to maintain the viability of these populations, there is an urgent need to ensure landscape permeability via creation and/or protection of potential migratory corridors together with protection of dispersing individuals. Establishment of transboundary conservation strategies including appropriate population monitoring focusing on genetic diversity, inbreeding status and demography of the populations will help to form and protect exchange of individuals between the various European lynx populations. Source populations should be large enough and in favourable status to supply enough dispersers to facilitate gene flow; it has been shown, that if there is a contact with a larger population and there are suitable vacant territories and prey availability, the population can successfully expand and retain gene flow even between the areas with potential barriers to movement (Chapron et al. 2014; Bagrade et al. 2016).

The human-mediated translocation of animals between the populations and founding of new small or medium-sized populations in suitable areas in Central Europe, which may act as stepping-stone populations, is another option in reconnecting isolated lynx populations in different areas into one viable metapopulation. Moreover, as suggested by Port et al. (2020), consideration should be given to the translocation of a few females into areas accessible by male dispersers, or regularly visited by them, as a starting point for the development of new stepping-stone populations, as males will be attracted into the area and they will have tendency to stay there. However, reintroduction of lynx populations into new areas should be well-planned (selection of suitable founders with regard to their population origin and genetic status, habitat quality and landscape connectivity in the surroundings) and used very cautiously due to a risk associated with capture and release of animals, high mortality of lynx exploring new areas after release and the conflicts arising with the acceptance of lynx by local key stakeholders (Červený et al. 2019), whose perception highly affects the success of all these conservation efforts.

Data availability Genotype data are given in the Supplementary material, Table S2.

Declarations

Conflict of interest The authors declare that they have no conflict of interest.

Consent to participate The manuscript does not contain any individual person's data – not applicable.

Consent to publish All authors consent to submitting this article to Conservation Genetics.

Research involving human and animal rights Genetic material was collected in accordance with institutional, national, and international guidelines. Permits for animal capture and handling were obtained from the PLA Moravian Karst Administration and the Ministry of Environment of the Czech Republic, permit numbers: SR/0081/JM/2017; 34128/ENV/17-2146/630/17. No animals were killed due to sampling.

References

Abbott R, Albach D, Ansell S, Arntzen JW, Baird SJE, Bierne N, Boughman J et al (2013) Hybridization and speciation. J Evol Biol 26:229–246. https://doi.org/10.1111/j.1420-9101.2012.02599.x

Adams JR, Waits LP (2007) An efficient method for screening faecal DNA genotypes and detecting new individuals and hybrids in the red wolf (*Canis rufus*) experimental population area. Conserv Genet 8:123–131. https://doi.org/10.1007/s10592-006-9154-5

Åkesson M, Liberg O, Sand H, Wabakken P, Bensch S, Flagstad Ø (2016) Genetic rescue in a severely inbred wolf population. Mol Ecol 25(19):4745–4756. https://doi.org/10.1111/mec.13797

Anděl P, Mináriková T, Andreas M (eds) (2010) Ochrana průchodnosti krajiny pro velké savce. Evernia, Liberec, p 137

Anderson E (1949) Introgressive hybridization. John Wiley and Sons, New York. https://doi.org/10.1111/j.1469-185X.1953.tb01379.x

Bagrade G, Ruņģis DE, Ornicāns A, Šuba J, Žunna A, Howlett SJ, Lūkins M, Gailīte A, Stepanova A, Done G, Gaile A, Bitenieks K, Mihailova L, Baumanis J, Ozoliņš J (2016) Status assessment of Eurasian lynx in Latvia linking genetics and demography–a growing population or a source–sink process? Mamm Res 61:337–352. https://doi.org/10.1007/s13364-016-0279-8

Bartoń KA, Zwijacz-Kozica T, Zięba F, Sergiel A, Selva N (2019) Bears without borders: long-distance movement in human-dominated landscapes. Glob Ecol Conserv 1:e00541. https://doi.org/10.1111/j.1365-2907.2011.00192.x

Böer M, Reklewski J, Śmiełowski J, Tyrała P (2000) Reintroduction of the European Lynx to the kampinoski national park/Poland-a field experiment with zooborn individuals. Part III: demographic development of the population from December 1993 until January 2000. Der Zoologische Garten 70(5):304–312

Breitenmoser-Würsten C, Obexer-Ruff G (2003) Population and conservation genetics of two re-introduced lynx (*Lynx lynx*) populations in Switzerland—a molecular evaluation 30 years after translocation. In: Proceedings of the 2nd Conference on the Status and Conservation of the Alpine Lynx Population (SCALP):7–9

Bull J, Heurich M, Saveljev A, Schmidt K, Fickel J, Förster D (2016) The effect of reintroduction on the genetic variability in Eurasian lynx populations: the cases of Bohemian Bavarian and Vosges-Palatinian populations. Conserv Genet 17:1229–1234. https://doi.org/10.1007/s10592-016-0839-0

Bullock JM, Kenward RE, Hails RS (2002) Dispersal ecology. Cambridge University Press, New York, p 480p

Červený J, Krojerová-Prokešová J, Kušta T, Koubek P (2019) The change in the attitudes of Czech hunters towards Eurasian lynx: is poaching restricting lynx population growth? J Nat Conserv 47:28–37. https://doi.org/10.1016/j.jnc.2018.11.002

Chapron G, Kaczensky P, Linnell JD, von Arx M, Huber D, Andrén H et al (2014) Recovery of large carnivores in Europe's modern human-dominated landscapes. Science 346:1517–1519. https://doi.org/10.1126/science.1257553

Chen KY, Marschall EA, Sovic MG, Fries AC, Gibbs HL, Ludsin SA (2018) *assignPOP*: an R package for population assignment using genetic, non-genetic, or integrated data in a machine learning framework. Methods Ecol Evol 9:439–446. https://doi.org/10.1111/2041-210X.12897

Copernicus Land Monitoring Service (2018) European Environment Agency (EEA), European Union. https://land.copernicus.eu/pan-european/high-resolution-layers/forests/forest-type-1/status-maps/forest-type-2018

Clobert J, Baguette M, Benton TG, Bullock JM (eds) (2012) Dispersal ecology and evolution. Oxford University Press. https://doi.org/10.1093/acprof:oso/9780199608898.001.0001

Crooks KR, Burdett CL, Theobald DM, King SR, Di Marco M, Rondinini C, Boitani L (2017) Quantification of habitat fragmentation reveals extinction risk in terrestrial mammals. PNAS 114:7635–7640. https://doi.org/10.1073/pnas.1705769114

Dobson FS (1982) Competition for mates and predominant juvenile male dispersal in mammals. Anim Behav 30:1183–1192. https://doi.org/10.1016/S0003-3472(82)80209-1

Ďuľa M, Krofel M (2020) A cat in paradise: hunting and feeding behaviour of Eurasian lynx among abundant naive prey. Mamm Biol. https://doi.org/10.1007/s42991-020-00070-6

Evanno G, Regnaut S, Goudet J (2005) Detecting the number of clusters of individuals using the software structure: a simulation study. Mol Ecol 14:2611–2620. https://doi.org/10.1111/j.1365-294X.2005.02553.x

Fahrig L (2003) Effects of habitat fragmentation on biodiversity. Annu Rev Ecol Evol Syst 34:487–515. https://doi.org/10.1146/annurev.ecolsys.34.011802.132419

Fattebert J, Balme G, Dickerson T, Slotow R, Hunter L (2015) Density-dependent natal dispersal patterns in a leopard population recovering from over-harvest. PLoS ONE 10(4):e0122355. https://doi.org/10.1371/journal.pone.0122355

Flousek J, Zajac T, Kutal M, Zuczkowski M, Palucki A, Pudil M, Kafka P (2014) Velké šelmy (Carnivora) v Krkonoších, Jizerských horách, Górach Stołowych a na Broumovsku (Česká republika, Polsko) – minulost a přítomnost. Opera Corcontica 51:37–59

Frankham R, Briscoe DA, Ballou JD (2002) Introduction to conservation genetics. Cambridge University Press, pp. 617 ISBN 9780521639859

Frankham R (2015) Genetic rescue of small, inbred populations: meta-analysis reveals large and consistent benefits of gene flow. Mol Ecol 24:2610–2618. https://doi.org/10.1111/mec.13139

Greenwood PJ (1980) Mating systems, philopatry and dispersal in birds and mammals. Anim Behav 28:1140–1162. https://doi.org/10.1016/S0003-3472(80)80103-5

Groff C, Dalpiaz D, Frapporti C, Zanghellini P (eds) (2009) Rapporto Orso 2008 del Servizio Foreste e Fauna della Provincia Autonoma di Trento

Goudet J (2005) Hierfstat, a package for R to compute and test hierarchical F-statistics. Mol Ecol Notes 5:184–186. https://doi.org/10.1111/j.1471-8286.2004.00828.x

Gour DS, Bhagavatula J, Bhavanishankar M, Reddy PA, Gupta JA, Sarkar MS et al (2013) Philopatry and dispersal patterns in tiger

(*Panthera tigris*). PLoS ONE 8(7):e66956. https://doi.org/10.1371/journal.pone.0066956

Herrero A, Heikkinen J, Holmala K (2020) Movement patterns and habitat selection during dispersal in Eurasian lynx. Mamm Res 65:523–533. https://doi.org/10.1007/s13364-020-00499-7

Janečka JE, Blankenship TL, Hirth DH, Kilpatrick CW, Tewes ME, Grassman LIJ (2007) Evidence for male-biased dispersal in bobcats *Lynx rufus* using relatedness analysis. Wildl Biol 13:38–47. https://doi.org/10.2981/0909-6396(2007)13[38:EFMDIB]2.0.CO;2

Jombart T (2008) *adegenet*: a R package for the multivariate analysis of genetic markers. Bioinformatics 24:1403–1405. https://doi.org/10.1093/bioinformatics/btn129

Kaczensky P, Chapron G, Arx M von, Huber D, Andrén H, Linnell J (eds) (2013) Status, management and distribution of large carnivores—bear, lynx, wolf and wolverine—in Europe (Report to the EU Commission, Part 1 and Part 2, 2013)

Kopelman NM, Mayzel J, Jakobsson M, Rosenberg NA, Mayrose I (2015) CLUMPAK: a program for identifying clustering modes and packaging population structure inferences across K. Mol Ecol Res 15(5):1179–1191. https://doi.org/10.1111/1755-0998.12387

Kratochvíl J, Vala F (1968) History of occurrence of the lynx in Bohemia and Moravia. In: Kratochvíl J. (ed.) History of the distribution of the lynx in Europe. Acta Sc Nat Brno 2(4):35–48

Krojerová-Prokešová J, Turbaková B, Jelenčič M, Bojda M, Kutal M, Skrbinšek T, Koubek P, Bryja J (2019) Genetic constraints of population expansion of the Carpathian lynx at the western edge of its native distribution range in Central Europe. Heredity 122(6):785–799. https://doi.org/10.1038/s41437-018-0167-x

Kutal M, Belotti E, Volfová J, Mináriková T, Bufka L, Poledník L, Krojerová J, Bojda M, Váňa M, Kutalová L, Beneš J, Flousek J, Tomášek V, Kafka P, Poledníková K, Pospíšková J, Dekař P, Machciník B, Koubek P, Duľa M (2017) Occurrence of large carnivores – *Lynx lynx*, *Canis lupus*, and *Ursus arctos* – and of *Felis silvestris* in the Czech Republic and western Slovakia in 2012–2016 (Carnivora). Lynx, New Series 48(1):93–107. https://doi.org/10.2478/lynx-2017-0006

Large Carnivore Initiative for Europe, LCIE (2020) Online at https://www.lcie.org/. Accessed on 12. 10. 2020

Li YL, Liu JX (2018) StructureSelector: a web-based software to select and visualize the optimal number of clusters using multiple methods. Mol Ecol Res 18:176–177. https://doi.org/10.1111/1755-0998.12719

Lucena-Perez M, Marmesat E, Kleinman-Ruiz D et al (2020) Genomic patterns in the widespread Eurasian lynx shaped by late quaternary climatic fluctuations and anthropogenic impacts. Mol Ecol 29:812–828. https://doi.org/10.1111/mec.15366

Lynch M (1991) The genetic interpretation of inbreeding depression and outbreeding depression. Evolution 45:622–629. https://doi.org/10.2307/2409915

Mueller SA, Reiners TE, Middelhoff TL, Anders O, Kasperkiewicz A, Nowak C (2020) The rise of a large carnivore population in Central Europe: genetic evaluation of lynx reintroduction in the Harz Mountains. Conserv Genet. https://doi.org/10.1007/s10592-020-01270-w

Mysłajek RW, Kwiatkowska I, Diserens TA, Haidit A, Nowak S (2019) Occurrence of Eurasian lynx in western Poland after two decades of strict protection. Cat News 69:12–13

Paetkau D, Calvert W, Stirling I, Strobeck C (1995) Microsatellite analysis of population structure in Canadian polar bears. Mol Ecol 4:347–354. https://doi.org/10.1111/j.1365-294X.1995.tb00227.x

Paetkau D, Slade R, Burden M, Estoup A (2004) Direct, real-time estimation of migration rate using assignment methods: a simulation-based exploration of accuracy and power. Mol Ecol 13:55–65. https://doi.org/10.1046/j.1365-294X.2004.02008.x

Piry S, Alapetite A, Cornuet J-M, Paetkau D, Baudouin L, Estoup A (2004) GeneClass2: a software for genetic assignment and first-generation migrant detection. J Hered 95:536–539. https://doi.org/10.1093/jhered/esh074

Port M, Henkelmann A, Schröder F, Waltert M, Middelhoff L, Anders O, Jokisch S (2020) Rise and fall of a Eurasian lynx (*Lynx lynx*) stepping-stone population in central Germany. Mamm Res. https://doi.org/10.1007/s13364-020-00527-6

Pritchard JK, Stephens M, Donnelly P (2000) Inference of population structure using multilocus genotype data. Genetics 155:945–959. https://doi.org/10.1111/j.1471-8286.2007.01758.x

Puechmaille SJ (2016) The program structure does not reliably recover the correct population structure when sampling is uneven: subsampling and new estimators alleviate the problem. Mol Ecol Res 16:608–627. https://doi.org/10.1111/1755-0998.12512

R Core Team (2013) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL http://www.R-project.org/.

Ratkiewicz M, Matosiuk M, Saveljev AP, Sidorovich V, Ozolins J, Männil P, Balciauskas L, Kojola I, Okarma H, Kowalczyk R, Schmidt K (2014) Long-range gene flow and the effects of climatic and ecological factors on genetic structuring in a large, solitary carnivore: the Eurasian lynx. PLoS ONE 9(12):e115160. https://doi.org/10.1371/journal.pone.0115160

Reklewski JA (2006) Ryś w Puszczy Kampinoskiej. Kampinoski Park Narodowy, Izabelin, p 68

Samelius G, Andrén H, Liberg O, Linnell JDC, Odden J, Ahlqvist P, Segerström P, Sköld K (2012) Spatial and temporal variation in natal dispersal by Eurasian lynx in Scandinavia. J Zool 286:120–130. https://doi.org/10.1111/j.1469-7998.2011.00857.x

Schmidt K (1998) Maternal behaviour and juvenile dispersal in the Eurasian lynx. Acta Theriol 43:391–408. https://doi.org/10.4098/AT.ARCH.98-37

Seidler RG, Long RA, Berger J, Bergen S, Beckmann JP (2015) Identifying impediments to long-distance mammal migrations. Conserv Biol 29:99–109. https://doi.org/10.1111/cobi.12376

Sindičić M, Polanc P, Gomerčić T, Jelenčič M, Huber Đ, Trontelj P, Skrbinšek T (2013) Genetic data confirm critical status of the reintroduced Dinaric population of Eurasian lynx. Conserv Genet 14:1009–1018. https://doi.org/10.1007/s10592-013-0491-x

Støen O-G, Zedrosser A, Sæbø S, Swenson JE (2006) Inversely density-dependent natal dispersal in brown bears *Ursus arctos*. Oecologia 148:356–364. https://doi.org/10.1007/s00442-006-0384-5

Taberlet P, Griffin S, Goossens B, Questiau S, Manceau V, Escaravage N et al (1996) Reliable genotyping of samples with very low DNA quantities using PCR. Nucleic Acids Res 26:3189–3194. https://doi.org/10.1093/nar/24.16.3189

Trakhtenbrot A, Nathan R, Perry G, Richardson DM (2005) The importance of long-distance dispersal in biodiversity conservation. Divers Distrib 11(2):173–181. https://doi.org/10.1111/j.1366-9516.2005.00156.x

von Arx M, Breitenmoser-Würsten C, Breitenmoser U (2009) Lessons from the reintroduction of the Eurasian lynx in Central and West Europe (Lecciones aprendidas a partir de la reintroducción dellince boreal en Europa Central y Occidental). In Iberian Lynx ex situ conservation: an interdisciplinary approach, pp. 403–409.

Wabakken P, Sand H, Kojola I, Zimmermann B, Arnemo JM, Pedersen HC, Liberg O (2007) Multistage, long-range natal dispersal by a global positioning system-collared Scandinavian wolf. J Wildl Manage 71:1631–1634. https://doi.org/10.2193/2006-222

Weir BS, Cockerham CC (1984) Estimating F-statistics for the analysis of population structure. Evol 38:1358–1370. https://doi.org/10.1111/j.1558-5646.1984.tb05657.x

Wultsch C, Caragiulo A, Dias-Freedman I, Quigley H, Rabinowitz S, Amato G (2016) Genetic diversity and population structure of Mesoamerican Jaguars (*Panthera onca*): implications for conservation and management. PLoS ONE 11(10):e0162377. https://doi.org/10.1371/journal.pone.0162377

Zimmermann F, Breitenmoser-Würsten C, Breitenmoser U (2005) Natal dispersal of Eurasian lynx (*Lynx lynx*) in Switzerland. J Zool 267(4):381–395. https://doi.org/10.1017/S0952836905007545

Zimmermann F, Breitenmoser-Würsten C, Breitenmoser U (2007) Importance of dispersal for the expansion of a Eurasian lynx *Lynx lynx* population in a fragmented landscape. Oryx 41(3):358–368. https://doi.org/10.1017/S0030605307000712

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.