ELSEVIER

Biological Conservation 87 (1999) 249–254

BIOLOGICAL CONSERVATION

Genetic variability and differentiation of two bearded vulture *Gypaetus barbatus* populations and implications for reintroduction projects

Juan José Negro [a,*], María José Torres [b]

[a] *Estación Biológica de Doñana, Apdo. 1056. 41080 Sevilla, Spain*
[b] *Departamento de Microbiología, Facultad de Medicina, C/ Dr. Fedriani s/n, 41013 Sevilla, Spain*

Received 31 October 1997; received in revised form 20 March 1998; accepted 9 April 1998

Abstract

Reintroduced bearded vultures *Gypaetus barbatus* in the Alps descend mainly from birds collected in the former Soviet Union. A second project has been initiated in southern Spain using the same bloodlines. We used DNA fingerprinting to assess levels of genetic variability, and to determine genetic differences, among birds from the Pyrenees and birds from the Alps reintroduction project. The degree of fingerprint band-sharing was high within both populations, which points to low levels of genetic variation in each population. On the other hand, common fingerprint fragments occurred at markedly different proportions in each population, suggesting some degree of population differentiation. Interbreeding would increase genetic variability in the resulting populations. We thus recommend the incorporation of wild Pyrenean birds to both the Alps and the Spanish breeding projects.

Keywords: Bearded vulture; DNA fingerprinting; Genetic variability; Captive breeding; Reintroduction

1. Introduction

The bearded vulture, or lammergeier, *Gypaetus barbatus*, is a large scavenger inhabiting mountain ranges in Eurasia and Africa (Brown and Amadon, 1968). In Western Europe, the species has suffered a dramatic population decline in the 20th century and it is listed as endangered (Tucker and Heath, 1994). Extant European populations are confined to the Pyrenees, the Balkans, and the islands of Corsica and Crete (Hiraldo et al., 1979).

Captive-bred individuals are being released in the Alps, where bearded vultures disappeared at the turn of the century (Bustamante, 1996; Mingozzi and Estève, 1997). This is a multinational effort, with release sites in Austria, France, Italy and Switzerland, and participating breeding centres in these countries and in Germany, The Netherlands and Israel. More recently, a second reintroduction program has been initiated in the Cazorla mountains in Andalusia, southern Spain. This project is sponsored by the Andalusian Autonomous Government and it will work in close collaboration with the Alps project. Bearded vultures last reproduced in Cazorla in the mid-1980s.

The World Conservation Union (see Kleiman et al., 1994) prescribes that, in reintroduction programs, source populations should be the most closely related genetically to the original native stock. The rationale for this is that reintroduced individuals will adapt better to local conditions. Additionally, if interbreeding with the nearest native populations occurred, the risk of outbreeding depression would be lower (Griffiths et al., 1996a; Bullock and Hodder, 1997).

The westernmost wild population of bearded vultures in continental Europe inhabits the Pyrenees, with *c.* 70 breeding pairs (Heredia, 1995; Razin, 1995). This population is located halfway between the Alps and Cazorla, and about 1000 km away from each location. For the Cazorla project, current plans are that birds for releases will be produced in a breeding centre already built on site. These breeding facilities will rely on individuals transferred from the Alps reintroduction project, and also on wild birds that may enter rehabilitation centres in Spain. The Alps captive population originated from 30 founder birds, most of which had been collected

* Corresponding author. Tel.: 00-34-95-4232340; fax: 00-34-95-4621125; e-mail: negro@ebd03.ebd.csic.es

0006-3207/98/$—see front matter
PII: S0006-3207(98)00056-1

in locations within the former Soviet Union (Frey et al., 1995), thus several thousand km away from the Iberian Peninsula.

Clinal variation in size and plumage pattern has been reported for bearded vultures (Hiraldo et al., 1979). The largest individuals are from Central Asia, and the smallest from Africa. Asian birds show more stripes and dots in different parts of the plumage than African birds; those from the Iberian Peninsula show an intermediate pattern. Based on morphological characters, Hiraldo et al. (1984) proposed two subspecies: *Gypaetus barbatus barbatus* in Eurasia and Northern Africa, and *G. b. meridionalis* in Eastern and Southern Africa. However, bearded vulture populations have never been genetically profiled, and genetic distinction between the Iberian birds and the Eastern birds in the captive stock has not been stablished.

Our aim was to use multilocus DNA fingerprinting to assess levels of genetic variability within and between these two groups. Ultimately, we want to have a tool for genetic management practices of the captive stock and to choose the birds destined to be released (see, e.g. Rave, 1995). Multilocus DNA fingerprinting finds its main application in the field of parentage determination, but it is now being used widely to assess genetic variation (Rave et al., 1994; Rave, 1995), and to address population questions (Lynch, 1991; Degnan, 1993; Freeman-Gallant, 1996).

2. Materials and methods

We obtained blood samples from 26 captive bearded vultures from the Alps reintroduction project kept at the Vienna Breeding Unit in 1995. Eleven of those birds had been wild-caught (founder birds), while the rest were captive-bred. There were two siblings, and the rest of the birds were, to our knowledge, unrelated. All birds but two, one from the Pyrenees and the other from Greece, were caught or descended from birds collected in the former Soviet Union; for the work we report here, we used 22 of the Eastern birds and the one from the Pyrenees. We also obtained samples from 15 Pyrenean birds in 1994 and 1995. The blood was preserved in lysis buffer (Seutin et al., 1991) and stored at 4°C until processing.

Genomic DNA was extracted using a standard phenol-chloroform procedure (Negro et al., 1996). Aliquots of 5 g of DNA were digested overnight at 37°C with 20 units of the restriction enzyme *Hinf*I and electrophoresed on 20 cm long 0.7% agarose gels at 29 V for 36 h, in 0.5x TBE buffer. Lambda *Hind*III was run once on each gel as a molecular size marker. The DNA fragments were southern-blotted to a positively charged nylon membrane and crosslinked at 80°C for 2 h. Membranes were hybridized with the microsatellite probe $(GGAT)_4$ labelled with Digoxigenine, overnight at 42°C in a hybridization oven (Negro et al., 1996). The $(GGAT)_4$ probe was selected because it had been used in previous parentage studies of raptors and always showed patterns inherited in a Mendelian fashion (Wolfes et al., 1991; Negro et al., 1996). For the detection, we used the chemiluminiscent substrate CDP-Star from Boeringher–Mannheim. Hybridization and detection were carried out according to the manufacturer's protocol. Hybridized blots were exposed to Boeringher–Mannheim X-film for 15 min to 4 h at room temperature.

The fingerprints were analysed manually placing the autoradiograms on a light box. Comparison of banding patterns were confined to lanes on the same gel, which held up to 15 samples. All fragments in the range between 3.5 and 15 kilobase pairs were scored and assumed to be unlinked. A search was made for bands that could be population-specific.

To estimate genetic similarity within and between populations, we calculated band-sharing coefficients between paired combinations of birds in each gel. These coefficients are calculated as $2(N_{AB})/(N_A + N_B)$, where N_{AB} is the number of bands shared by birds A and B, and N_A and N_B are the number of bands scored in birds A and B, respectively (Wetton et al., 1987). We ran four different gels. In the first three gels we placed 7–8 consecutive samples from birds in the Alps project, followed by 5–8 samples from Pyrenean birds. In the fourth gel we took alternate samples from the two populations. Most individual samples were run just in one gel, except nine birds from the Pyrenees which were run in two different gels. Because the accuracy of band comparisons declines with increased distance between lanes (Parker et al., 1994), we determined band-sharing scores for blocks of up to eight birds (*c.* half of the membrane).

For each population and also for between-population comparisons, we obtained a mean band-sharing coefficient and standard error averaging the similarities of all possible pairwise combinations. However, individuals were used in multiple comparisons and thus data were interdependent (Lynch, 1990). To cope with this problem, corrected standard errors of the means were also estimated with a procedure similar to the subsampling routine proposed by Danforth and Freeman-Gallant (1996). These authors randomly sampled n truly independent values of similiarity from the overall data set, calculated the mean and then repeated the process 1000 times. Instead of using the iteration approach, we obtained all possible combinations of independent dyads in each group.

For the Pyrenees, we obtained a maximum of seven independent dyads, 1575 times. For the Alps, we got 10 independent comparisons 23,625 times (n was dependent on sample size and the organization of the

individuals on each gel). For band-sharing between one Pyrenean bird and one from the Alps, we utilized 12 independent dyads 165,888 times. We calculated the mean band-sharing and standard deviation of each string of independent dyads and then calculated the mean of the means, as well as the mean of the standard deviations for each group of comparisons.

Differences among means were tested by two-tailed *t*-tests using both corrected and uncorrected standard errors.

3. Results

A mean ± SD of 6.7 ± 1.2 ($n = 22$) and 7.4 ± 1.0 ($n = 15$) scorable bands was analysed in the fingerprints of the Eastern and Pyrenean birds, respectively. The difference was not significant (Kolmogorov–Smirnov test, $p > 0.05$).

There were three well defined regions showing restriction fragments. The upper bands, 1–4 per individual, were in the range between 10 and 15 kb (kilobases). There was also a middle range (6–9 kb), with 1–3 bands per individual. The lower bands, 1–5 per individual, were in the range between 3.5–5 kb. We identified some commonly occurring fragments present in widely different proportions in the two populations (Table 1). Those differences point to genetic differentiation between the two populations, even though it would not be possible to assign an individual of unknown origin to one or the other of these populations based on presence/absence of common bands.

The within- and between-population mean band-sharing coefficients (Table 2) show that corrected and uncorrected estimations were almost equal, as well as their standard errors. Differences were significant for uncorrected means, but become non-significant for corrected means (except one case, see Table 2), possibly due to reduced samples sizes for independent values.

4. Discussion

The level of genetic variation detected with the microsatellite probe (GGAT) was considerable, as it allowed distinction between all individuals tested. However, the degree of band-sharing among unrelated bearded vultures, in both the Pyrenean and Eastern populations, was > 0.50, and this points to a low level of genetic variation. This observation contrasts with data from other raptor studies which utilized the same probe (band-sharing varies for different probes, even when used with the same set of individuals, see, e.g. Degnan, 1993). Unrelated pairs of lesser kestrels *Falco naumanni*, a raptor species with much larger populations than the bearded vulture, showed a mean band-sharing of 0.21 (Negro et al., 1996). Unrelated eagle owls *Bubo bubo*, another relatively common species, showed a mean band-sharing of 0.19 (Wolfes et al., 1991). In fact, 'background' levels of band-sharing for outbred avian populations typically average less than 0.30 (Rave, 1995 and references therein).

Higher band-sharing values, and presumably lower levels of genetic variation compared to the bearded vulture, have been reported for the red kite *Milvus milvus*, a patchily distributed and threatened European raptor (May et al., 1993). They have also been shown in endangered harbour seals *Phoca vitulina* in the Dutch Wadden Sea (Kappe et al., 1995), the rare Hawaiian goose, or nene, *Branta sandvicensis* (Rave, 1995), and also isolated small populations of the California Channel Island fox *Urocyon littoralis*. For the latter, all individuals in some islands showed complete absence of variability in restriction–fragment profiles (Gilbert et al., 1990).

The population of bearded vultures in the Pyrenees is small, with *c*. 70 pairs counted in 1995 (Heredia, 1995; Razin, 1995), but it was even smaller, with *c*. 50 pairs, in the 1970s (Hiraldo et al., 1979). The nearest extant population is in the island of Corsica, numbering less than a dozen breeding pairs. There is no evidence of

Table 1
Occurrence of commonly observed restriction bands in two populations of bearded vultures

	Proportion of individuals			
Range:	10–15 kb	6–9 kb		3.5–5 kb
Band:	A	B	C	D
Pyrenees ($n = 15$)	0.46	0.54	1.00	0.13
Eastern region ($n = 22$)	0.00	1.00	0.36	0.82

Table 2
Mean ± SE band sharing coefficients within and between two populations of bearded vultures (sample sizes in parentheses)

	Pyrenees		Eastern region	p [a]
Uncorrected mean ± SE				
Within populations	0.54 ± 0.11 (55)		0.59 ± 0.12 (64)	0.01
Between populations		0.47 ± 0.13 (67)		
p [a]	0.003		0.00001	
Corrected mean ± SE				
Within populations	0.53 ± 0.11 (7)		0.60 ± 0.12 (10)	0.23
Between populations		0.44 ± 0.12 (12)		
p [a]	0.13		0.005	

[a] *t*-test for the difference between each within-population means and the between population mean.

exchange between these two populations. Bearded vultures seem to be poor dispersers, as young marked in either the Pyrenees or the Alps are only resighted in the mountain range where they fledged (or were released) (Heredia and Heredia, 1991).

Population genetics theory predicts low genetic variability in small populations and thus the results for the Pyrenees can be explained as a loss of variation due to inbreeding. Some authors (see, e.g. Heredia, 1989; Donázar, 1991) have in fact warned that inbreeding depression may be of concern for the conservation of the Pyrenean population. Deleterious effects of inbreeding are suspected, for example, in the small and isolated red kite population of Wales. Their breeding success was significantly lower (Cordero et al., 1997), and age at first breeding delayed (Evans et al., 1998), compared with more heterozygous birds from the larger populations of Spain and Germany, which were released in Southern England as part of a re-establishment project.

Surprisingly, mean band-sharing values were not lower for Eastern bearded vultures compared to the Pyrenean population (they were in fact slightly higher, although not significantly so for corrected values). Further analyses with larger sample sizes would be needed to confirm whether the difference is genuine. In any case, the low variability of the birds in the Alps project could be interpreted in two ways: (1) the bearded vulture population in the former Soviet Union was small when wild birds were collected and sold to European zoos in the 1960s and 1970s. (2) The birds were collected in only a few locations and may be interrelated. Although it is only a speculation, we agree with the managers of the Alps project (Frey et al., 1995) in favouring the second hypothesis. There are no reliable estimations of population numbers in the former Soviet Union, but the strongholds of the species, and the larger contiguous distribution area, lie precisely in the extense mountain ranges of southern Russia, central Asia and eastern China (Brown and Amadon, 1968). In addition, it is logical to suppose that only a reduced number of collectors worked for the European zoos and that the same few breeding territories were visited year after year. Given the long life and stability of breeding pairs in the species, siblings or close relatives may have been part of the captive stock of the Alps project.

Two results indicate that population differentiation may exist. First, the between-population mean band-sharing seemed to be lower than within-population means. The fact that corrected measures were not significantly different may cast some doubt on this result, but it has to be noted that the correction procedure requires very large data sets (Danforth and Freeman-Gallant, 1996), and this is an almost impossible task working with the rare bearded vulture. Second, we have shown that some common fragments occurred at markedly different proportions in each population. Even though we cannot assign an individual with complete certainty to either population from its fingerprint profile, the frequency of some alleles was clearly different.

4.1. Implications for conservation

In reintroduction programs, an effort has to be made to maintain the highest possible levels of genetic variation (Sarrazin and Barbault, 1996). This can be achieved by pairing individuals that show high heterozygosity or come from geographically distant populations. However, in the latter case the risk of outbreeding depression may be a concern.

In the reintroduction project of bearded vultures in southern Spain, the IUCN's recommendation of using locally adapted individuals suggests the breeding and release of Pyrenean birds, as the only remaining birds native to the Iberian Peninsula. However, several captive individuals have already been transferred from the Alps project (A. Llopis, pers. com.), and further transfer of individuals is likely (Frey et al., 1995).

Both the wild Pyrenean population and the captive population in the Alps project seem to be inbred, and this may outweigh the risk of outbreeding depression. In addition, our results pointing to genetic differentiation between Pyrenean and Eastern birds does not mean a large genetic distance (which has not been measured), nor reproductive incompatibility.

If band-sharing was genuinely lower between than within populations, genetic variability would be increased quickly by mating Pyrenean birds with Eastern birds. Therefore, the main recommendation we can make at this point is for managers of both the Cazorla and the Alps reintroduction projects to take the necessary steps to obtain Pyrenean birds, and not rely almost exclusively on individuals originating from a presumably small area in the former Soviet Union. Such an approach to restore genetic variability with individuals of different provenances has already been put to practice with endangered populations of other raptorial species, such as the red kite in Great Britain (Cordero et al., 1997), or the peregrine falcon *Falco peregrinus* in North America (Cade et al., 1988).

About 25% of all territories in the Pyrenees are currently occupied by trios comprising two males and one female. Several factors leading to such polyandrous trio formation have been suggested (Donázar, 1991) : biased sex ratios, low food availability, high breeding density and genetic relatedness between males.

About 20 nestlings fledge each year in the Spanish Pyrenees, plus about 8–10 in the French Pyrenees (Heredia and Heredia, 1991). We have already sexed all bearded vultures included in this study using PCR-based techniques (Griffiths et al., 1996b). It would be

possible to climb to Pyrenean nests to take blood samples of nestlings for molecular sexing. Selected nests could be revisited to remove male nestlings to be paired with females originating from the captive stock in the Alps project. Removal of wild birds should be done sequentially, at a rate of at most two males in about 10 years, and rotating among nests to include the genetic component of as many pairs as possible.

The proposed measure of sequential extraction of males in the Pyrenees would have little effect on the demographic parameters of the wild population, which is even expanding, but would increase the genetic variability of birds to be released in both Cazorla and the Alps. In the particular case of Cazorla, IUCN's recommendations would partly be met, as Iberian bloodlines would be well represented in the reintroduced birds.

If it was judged desirable to increase the genetic variability of the wild Pyrenean population, the extracted wild birds could be substituted with captive-bred nestlings with different genetic profiles. Further investigations are needed, however, to determine whether current genetic variability is lower than in the past, by using e.g. museum skins, or else, is an intrinsic characteristic of some long-lived raptor species (Negro and Hiraldo, 1994).

Acknowledgements

The study was funded by the 'Consejería de Medio Ambiente' of the Andalusian Autonomous Government (Spain). We thank H. Frey (Foundation for the Conservation of the Bearded Vulture) and J. Guiral (Autonomous Government of Aragón, Spain) for providing blood samples, and the staff of the Microbiology Department in the University of Sevilla, particularly Drs. J.C. Palomares, J. Aznar and E. Perea, for giving permission to use their laboratories. Parts of the analyses were carried out at the Laboratorio de Ecología Molecular (Estación Biológica de Doñana). E. Collado provided unvaluable computer assistance. F. Hiraldo and J. Bustamante critically reviewed the manuscript. JJN was supported by the Spanish Council for Research (CSIC), and MJT by the Spanish Ministry of Education and Culture.

References

Brown, L., Amadon, D., 1968. Eagles, Hawks and Falcons of the World. Country Life, London.

Bullock, J.M., Hodder, K.H., 1997. Reintroductions: challenges and lessons for basic ecology. Trends in Ecology and Evolution 12, 68–69.

Bustamante, J., 1996. Population viability analysis of captive and released bearded vulture populations. Conservation Biology 10, 822–831.

Cade, T., Enderson, J.H., Thelander, C.G., White, C.M., (Eds.), 1988. Peregrine falcon populations. Their management and recovery. The Peregrine Fund, Boise, ID.

Cordero, P.J., Evans, I.M., Parkin, D.T., Galbraith, C.A., 1997. Studies of the genetics of a naturalised population of red kites *Milvus milvus* in England established by translocation. In: Tew, T.E., Crawford, T.J., Spencer, J.W., Stevens, D.P., Usher, M.B., Warren, J. (Eds.), The role of genetics in conserving small populations, JNCC, Peterborough, UK. pp. 89–96.

Danforth, B.N., Freeman-Gallant, C.R., 1996. DNA fingerprinting data and the problem of non-independence among pairwise comparisons. Molecular Ecology 5, 221–227.

Degnan, S.M., 1993. Genetic variability and population differentiation inferred from DNA fingerprinting in silvereyes (Aves: Zosteropidae). Evolution 47, 1105–1117.

Donázar, J.A., 1991. Unidades reproductoras inusuales: tríos poliándricos. In: Heredia, R., Heredia, B. (Eds.), El Quebrantahuesos en los Pirineos. Colección Técnica. ICONA, Madrid, pp. 39–46.

Evans, I.M., Cordero, P.J., Parkin, D.T., 1998. Successful breeding at one year of age by red kites *Milvus milvus* in southern England. Ibis, 140, 53–57.

Freeman-Gallant, C.R., 1996. Microgeographic patterns of genetic and morphological variation in savannah sparrows (*Passerculus sandwichensis*). Evolution 50, 1631–1637.

Frey, H., Knotzinger, O., Llopis, A., 1995. The breeding network. An analysis of the period 1978 to 1995. In: Frey, H., Kurzweil, J., Bijleveld, M. (Eds.), Bearded Vulture: Reintroduction into the Alps. Annual Report 1995. Foundation for the Conservation of the Bearded Vulture, Wassenaar, The Netherlands. pp. 13–38.

Gilbert, D.A., Lehman, N., O'Brien, S., Wayne, R., 1990. Genetic fingerprinting reflects population differentiation in the California Channel Island fox. Nature 344, 764–767.

Griffiths, H.I., Davison, A., Birks, J., 1996a. Species reintroductions. Conservation Biology 10, 923.

Griffiths, R., Daan, S., Dijkstra, C., 1996b. Sex identification in birds using two CHD genes. Proc. R. Soc. London B. 263, 1251–1256.

Heredia, B., 1989. Plan para la conservación del quebrantahuesos en los Pirineos. Quercus 40, 4–14.

Heredia, R., 1995. Bearded vultures in the Spanish Pyrenees. Reproduction in 1995. In: Frey, H., Kurzweil, J., Bijleveld, M. (Eds.), Bearded Vulture: Reintroduction into the Alps. Annual Report 1995. Foundation for the Conservation of the Bearded Vulture, Wassenaar, The Netherlands. pp. 54–55.

Heredia, R., Heredia, B., 1991. El quebrantahuesos (*Gypaetus barbatus*) en los Pirineos. Colección Técnica. ICONA, Madrid.

Hiraldo, F., Delibes, M., Calderón, J., 1979. El Quebrantahuesos *Gypaetus barbatus* L. Monografía 22. ICONA, Madrid.

Hiraldo, F., Delibes, M., Calderón, J., 1984. Comments on the taxonomy of the bearded vulture *Gypaetus barbatus* (Linnaeus 1758). Bonn Zool. Beitr. 35, 91–95.

Kappe, A.L., Van de Zande, L., Vedder, E.J., Bijlsma, R., Van Delden, W., 1995. Genetic variation in *Phoca vitulina* (the harbour seal) revealed by DNA fingerprinting and RAPDs. Heredity 74, 647–653.

Kleiman, D.G., Stanley Price, M.R., Beck, B.B., 1994. Criteria for reintroductions. In: Olney, P.J.S., Mace, G.M., Feitsner, A.T.C. (Eds.), Creative conservation: interactive management of wild and captive animals. Chapman and Hall, London, pp. 287–303.

Lynch, M., 1990. The similarity index and DNA fingerprinting. Mol. Biol. Evol. 7, 478–484.

Lynch, M., 1991. Analysis of population genetic structure by DNA fingerprinting. In: Burke, T., Dolf, G., Jeffreys, A.J., Wolff, R. (Eds.), DNA fingerprinting: approaches and applications. Birkhauser Verlag, Basel, Switzerland, pp. 113–126.

May, C., Wetton, J.H., Davis, P.E., Brookfield, J.F.Y., Parkin, D.T., 1993. Single-locus profiling reveals loss of variation in inbred populations of the red kite (*Milvus milvus*). Proc. R. Soc. Lond. B 251, 165–170.

Mingozzi, T., Estève, R., 1997. Analysis of a historical extirpation of the bearded vulture *Gypaetus barbatus* (L). in the Western Alps (France–Italy): former distribution and causes of extirpation. Biological Conservation 79, 155–171.

Negro, J.J., Hiraldo, F., 1994. Lack of allozyme variation in the Spanish Imperial Eagle (*Aquila adalberti*). Ibis 136, 87–90.

Negro, J.J., Villarroel, M., Tella, J.L., Kuhnlein, U., Hiraldo, F., Donazar, J.A., Bird, D.M., 1996. DNA fingerprinting reveals a low incidence of extra-pair fertilizations in the lesser kestrel. Animal Behaviour 51, 935–943.

Parker, P.G., Waite, T.A., Heinrich, B., Marzluff, J.M., 1994. Do common ravens share ephemeral food resources with kin? DNA fingerprinting evidence. Animal Behaviour 48, 1085–1093.

Rave, E.H., 1995. Genetic analyses of wild populations of Hawaian geese using DNA fingerprinting. The Condor 97, 82–90.

Rave, E.H., Fleischer, R.C., Duvall, F., Black, J.M., 1994. Genetic analyses through DNA fingerprinting of captive populations of Hawaiian Geese. Conservation Biology 8, 744–751.

Razin, M., 1995. A short report on the bearded vultures in the French Pyrenees in 1995. In: Frey, H., Kurzweil, J., Bijleveld, M. (Eds.), Bearded Vulture: Reintroduction into the Alps. Annual Report 1995. Foundation for the Conservation of the Bearded Vulture, Wassenaar, The Netherlands. p. 53.

Sarrazin, F., Barbault, R., 1996. Reintroduction: challenges and lessons for basic ecology. Trends in Ecology and Evolution 11, 474–478.

Seutin, G., White, B.N., Boag, P.T., 1991. Preservation of avian blood and tissue samples for DNA analyses. Can. J. Zool. 69, 82–90.

Tucker, G.H., Heath, M.F., 1994. Birds in Europe. Their Conservation Status. BirdLife International, Cambridge, UK.

Wetton, J.H., Carter, R.E., Parkin, D.T., Walters, D., 1987. Demographic study of a wild house sparrow population by DNA fingerprinting. Nature 327, 147–149.

Wolfes, R., Mathe, J., Seitz, A., 1991. Forensics of birds of prey by DNA fingerprinting with ^{32}P-labeled oligonucleotide probes. Electrophoresis 12, 175–180.