

Proceedings of the
III Iberian lynx Conservation Seminar
November 17-19, 2008



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III Iberian Lynx Conservation Seminar



November 17th-19th 2008, Huelva (Spain)

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Block A: “Setting the frame”

History, current status and existing conservation strategies



Current status of the Iberian lynx in Andalusia

Miguel Ángel Simón¹, Rafael Cadenas², José María Gil-Sánchez³, Marcos López-Parra⁴, José García⁵, Leonardo Fernández⁵, Gema Ruiz⁴ and Guillermo López⁶

1. Iberian lynx LIFE conservation project. Consejería de Medio Ambiente, Junta de Andalucía. c/ Fuente del Serbo, 3. 23071. Jaén. miguelangel.simon@juntadeandalucia.es
2. Instituto Andaluz de la Caza. Consejería de Medio Ambiente, Junta de Andalucía. Avd. Manuel Siurot, 50. 41071. Sevilla.
3. Iberian lynx LIFE conservation project. EGMASA. Avd. Andalucía, 104 Esc 3, 1º. 23006. Jaén.
4. Iberian lynx LIFE conservation project. EGMASA. c/ Johan Gutenberg, 1. 41092. Sevilla.
5. Iberian lynx LIFE conservation project. EGMASA. C/ Plus Ultra, nº8, 7ª Planta. 21001. Huelva.
6. Iberian lynx LIFE conservation project. EGMASA. c/ Pepe Espaliú, 2. 14004. Córdoba.

Abstract

The conservation status of the Iberian lynx was found to be extremely critical by the end of the 20th Century, when only 150 individuals remained in the wild secluded into two isolated populations, Doñana and eastern Sierra Morena, both located in Andalusia. The Andalusian Government, together with a number of partners, adopted different conservation measures that received support from the European Union in the form of two LIFE-Nature conservation projects. These projects have proved essential to avoid the potential extinction of the Iberian lynx in the wild and to stabilize both free-ranging populations. Presently, in situ conservation efforts are focused on the following objectives: (1) Maintaining and expanding the two existing populations; (2) Recovering extinct population nuclei following IUCN reintroduction criteria; (3) Maximizing genetic diversity by "genetically connecting" the two existing populations; (4) Continuing to promote local, national and international support to ensure the recovery of this highly endangered species. Nowadays, the Sierra Morena population continues to grow –both numerically and in surface area– at an annual basis. In fact, the number of lynxes has increased from 38 adults (individuals of more than one-year-of-age) registered in 2001 to 95 in 2008. The Doñana population remains stable and a translocation program, with the ultimate goal of genetic and demographic reinforcement, is currently taking place. Also, in situ efforts are presently focused on the recovery of historical population nuclei through reintroduction programs. Preparation works are being carried out since 2005 and the first releases are scheduled to begin in 2009.

Introduction

The Iberian lynx (*Lynx pardinus*), a species endemic to the Iberian Peninsula (Ferrer & Negro, 2004), is considered the most endangered felid in the world, as it has been catalogued as "critically endangered" by the IUCN (2003). Legal protection for the species is guaranteed by the Bern Convention, and the protection of its habitat is a priority by the European Commission (Habitat Directive 92/43). The Iberian lynx's distribution area suffered a 98% decline during the second half of the last century (Rodríguez & Delibes, 1992; Guzmán et al. 2004) and, by 2002, about 160 individuals remained, distributed into two isolated breeding populations: the Doñana Natural Space and its surroundings, and the Andújar-Cardena mountains in Eastern Sierra Morena, both in Andalusia (Guzmán et al. 2004). The Iberian lynx is a specialized predator, with more

than 90% of its diet comprised of wild rabbits (Gil-Sánchez et al. 2006). Mediterranean scrubland areas represent the optimal habitat for this species (Palomares, 2001). Current threats to the species include (1) scarcity of rabbits due to mixomatosis and rabbit haemorrhagic disease (RHD), (2) habitat destruction, and (3) human-caused mortality (road kills, leghold traps, poaching, etc.) (Guzmán et al. 2004). Given the critical situation of the species, the Andalusian Regional Ministry of Environment began working in early 2000 on measures to avoid the Iberian lynx extinction. These included: (1) to attain an accurate knowledge of the actual distribution range of the species, (2) to carry out a monitoring program of both rabbit and lynx populations, (3) to sign collaborative agreements with land owners, (4) to recover rabbit populations, (5) to

control non-natural causes of mortality, (6) to establish a captive population to help preserve current genetic diversity and (7) to ensure support from the local people that inhabit near both wild populations. Initial results from this Andalusian program allowed to determine what the actual distribution range of the Iberian lynx was and helped develop accurate population monitoring methods. By 2002, a more ambitious and participative LIFE-Nature conservation project was granted. The project, entitled "Recovery of the Iberian lynx populations in Andalusia" (LIFENAT 02/E/8609) lasted four years (2002-2006) and involved the participation of the Andalusian Regional Ministry of the Environment, the three main Andalusian hunter associations (Asociación de Titulares de Cotos de Andalucía -ATECA-, Federación Andaluza de Caza -FAC- and Asociación de Productores de Caza -APROCA-) and two conservation organizations (Ecologistas en Acción Andalucía -EEA- and CBD-Hábitat Foundation, belonging to the Spanish Environment Ministry). The main objectives of this project (which included both short- and medium-term conservation measures) were (1) to augment lynx population size, (2) to maximize the connection between isolated populations, (3) to promote habitat conservation and resources availability, (4) to improve public perception of the species, (5) to contribute to the maintenance of a genetic "pull" representative of the species, (6) to create an effective lynx population monitoring system and (6) to decrease threats for the lynx. Scientific oversight was provided by the Superior Council for Scientific Research (CSIC), Doñana's Biological Station. The main goal of this LIFE project was to stop and revert the rate of decline that had been observed during the previous decades. This included avoiding potential losses in both populations (through controlling threats and stabilizing resources availability), while increasing their carrying capacity to allow an increase of the population size. These specific goals were successfully attained, since both population nuclei could be stabilized and Sierra Morena one began increasing (LIFE project, unpublished data). Such results helped establish the basis for requesting a second LIFE-Nature project. This project, entitled "Conservation and Reintroduction of the Iberian Lynx in Andalusia" (LIFENAT 06/E/209), was granted in 2006 and will be ongoing until 2011. This new LIFE project focuses on the

following objectives: (1) Maintaining and expanding the two existing populations; (2) Recovering extinct population nuclei -following IUCN reintroduction criteria-, (3) Maximizing genetic diversity by "genetically connecting" the two existing populations; (4) Continuing to promote local, national and international support to ensure the recovery of this highly endangered species. The project counts with the participation of the Andalusian Regional Ministries of Environment, Public Works and Agriculture, the Extremadura Regional Ministry of Environment, the three main Andalusian hunter associations (ATECA, FAC and APROCA) and four conservation organizations (EEA, CBD-Hábitat Foundation, the Spanish Society for the Conservation and Study of Mammals (SECEM), and World Wildlife Fund (WWF)/Spain). The project was structured based on the "Recovery Strategy for the Iberian Lynx", presented in the II International Seminar on the Conservation of the Iberian Lynx, which was held in Córdoba in December 2004 (Olszanska & Breitenmoser, 2004). Presently, we are into the third year of this LIFE project and preliminary results indicate that the Sierra Morena population continues to increase, and also Doñana is beginning to do it, as it is currently being genetically and demographically enhanced via translocations of selected Sierra Morena individuals. In addition, firsts releases of the reintroduction of the species into areas of historical occupancy is scheduled to take place at the end of 2009.

Starting point

During 2001-2002, the actual distribution range of the Iberian lynx was studied in Andalusia. Transects searching for lynx signs (mainly scats and tracks) were performed in 366 5x5 km UTM squares, covering the known distribution area in 1988 (Rodríguez & Delibes, 1992). Lynx scats were confirmed by molecular methods (J.A. Godoy, unpublished data). Only 55 out of the 366 squares were positive, corresponding with the two previously identified nuclei: Doñana and the Andújar-Cardena mountains, in the Sierra Morena range. About 80% of the distribution area was composed of private hunting lands. Afterwards, a quantification of rabbit abundance was performed through the lynx distribution area by means of transects. Rabbit abundance ranged between 0,6 and 4 rabbits/ha.

Agreements with land owners

The establishment of agreements with land owners and with hunter societies allowed the conservation projects to perform management measures throughout the lynx distribution. A total of 125,088 Has have currently being protected via collaborative agreements with landowners: 106,359 Has signed with the Andalusian Regional Ministry of the Environment, 13,729 Has with CBD-Habitat Foundation, and 5,000 Has with WWF-Spain. In addition, the Iberian lynx presently occupies 60,000 Has of public lands.

Conservation Strategy

Both Iberian lynx LIFE conservation projects have been based on a general strategy Focused on:

- **Stabilization of existing territories:** This has been carried out focusing on the following points: (1) stabilising and increasing availability of resources through habitat improvement, and (2) decreasing actual threats for the lynx. Habitat improvement management is focused on getting optimal vegetation structure both for rabbits and lynxes (Palomares, 2001). It is mainly based on the improvement of meadows (small scrub clearances and cereal plantations) and increasing shelters for rabbits (artificial burrows, hiding areas, etc). Also, a reduction of hunting pressure on rabbits is being carried out in critical areas. Rabbit releases are being performed in areas where rabbit abundance is under 1 rabbit/Ha in autumn, which is considered the minimum necessary to sustain Iberian lynx territories (Palomares, 2001). Rabbit releases are mainly being conducted using enclosures of about 5 Has with a predator exclusion fence (that is permeable to lynxes but not to other carnivores), although soft releases in the field without enclosure have also been performed. The last tool to increase rabbit availability is the establishment of supplementary feeding stations, aiming at maintaining specific lynxes in a determined area. These stations are baited with domestic rabbits and allow managing the population in case of potential rabbit populations crashes.
- **Decreasing causes of mortality.** Threats affecting Iberian lynx are mostly human-caused, although other causes can also affect the species' population dynamic. The main human-related threats affecting Iberian lynx include poaching, road kills and drowning in wells (Rodríguez & Delibes 1992; Guzmán et al. 2004), whereas the main natural threat is infectious

diseases (Meli et al. 2009; López et al. 2009). Several measures have been implemented to control these threats. Specific surveillance against illegal hunting methods is being carried out by LIFE project staff in areas of lynx occurrence. Also, control of road kills, mainly in the Doñana population, is one of the most important issues being addressed through the current LIFE project. Part of this control is being address by the construction of wildlife underpasses and ecoducts (Figure 2), placing wildlife exclusion fences on problem roads, installing dissuasive road night lights (catadioptrics), advertising road signals and speed bumps for traffic, etc. Moreover, specific brochures and booklets have been developed to increase awareness among drivers in the Doñana area. In addition, to address the problem of lynxes drowning in wells, all known wells in areas of lynx occupancy have been covered with metallic nets. Finally, to control the risk of infectious diseases a surveillance program is being carried out throughout both Iberian lynx populations. This program includes thorough health screenings of all handled animals, and also an indirect surveillance through scat analysis.

- **Settlement of new territories:** This is performed by increasing carrying capacity and decreasing threats for the lynx. The recovery of new territories has been made using territory recovery units (TRUs) as the main methodology. TRUs are composed of different habitat improvement measures performed in about 500 Ha (mean surface area of an Iberian lynx home range). Wild rabbit releases were included in TRUs only when rabbit abundance is under 1 rabbit/Ha.

- **"Compression" of occupied areas:** This strategy works under the same premises of the abovementioned ones; i.e., by increasing carrying capacity it is possible to reduce home range surfaces, which in turn will allow a higher number of individuals to inhabit a specific area.

- **Public awareness campaigns:** These campaigns have been taking place at a regular basis since 2002, and they are developed by all partners that participate in the conservation of this species. The incorporation of the hunter societies to the communication group has been one of the most effective ways to gain access to the hunters' opinion and support. Conservation associations have also been important to bring national and local awareness regarding the situation of the species.

- **Reintroduction:** This entails a medium- to long-term aspect of the conservation strategy. The first macro-habitat analysis for the suitability of areas for potential Iberian lynx reintroduction in Andalusia was performed in 1999 (F. Palomares, unpublished data). This habitat model became the basis for further work, although resource availability was not considered in it. Given that this factor is thought to be the most important one to take into account when developing a reintroduction strategy, a more exhaustive work was needed. Initial work towards Iberian lynx reintroduction began under the first LIFE conservation project, in 2002, and it has always been based on the IUCN guidelines for reintroduction (IUCN, 1998). The detection of potential reintroduction areas in Andalusia was performed by a multi-criteria analysis (MCA). The MCA is a decision-making tool developed for complex problems that follows a logical, well-structured, decision-making process where multiple criteria are involved. The steps followed in this analysis were (1) the identification of factors determining the suitability of areas that could host Iberian lynx, (2) compilation of information on everyone of the identified factors, (3) integration of all information in a Geographical Information System (GIS) and (4) evaluation of the resulting models. The identified factors that were important in determining lynx presence included habitat features (vegetation structure, altitude and slope), resource availability (rabbit abundance and drinking points availability) and threats (proximity to urban nuclei and to communication routes). Afterwards, a Weighted Linear Combination (WLC) analysis was performed (Barredo, 1996). With this method, the suitability of a pixel is obtained as the sum of partial suitability values of every factor in that area. The final potentiality map for Andalusia took also into account the historical distribution of the species and the degree of protection degree of each area. The result was the identification of three optimal nuclei where to carry out initial Iberian lynx reintroductions in Andalusia: Guarrizas Valley, Guadalmellato Valley and Hornachuelos Natural Park. Once this map was generated, all three areas were analysed in a fine-scale comparison through hierarchical analysis of 35 variables grouped into seven groups: (1) Social attitude, (2) Possibility of integration in the current metapopulation, (3) Carrying capacity, (4) Habitat structure, (5) Possibilities of expansion to other a-priori

suitable areas, (6) Illegal persecution pressure and (7) other threats. Among the three selected areas, the Guadalmellato Valley area was selected as the best one and, thus, the first reintroduction -scheduled for late 2009- will take place at this specific site. After signing agreements with the owners, habitat management measures are already being made in the area.

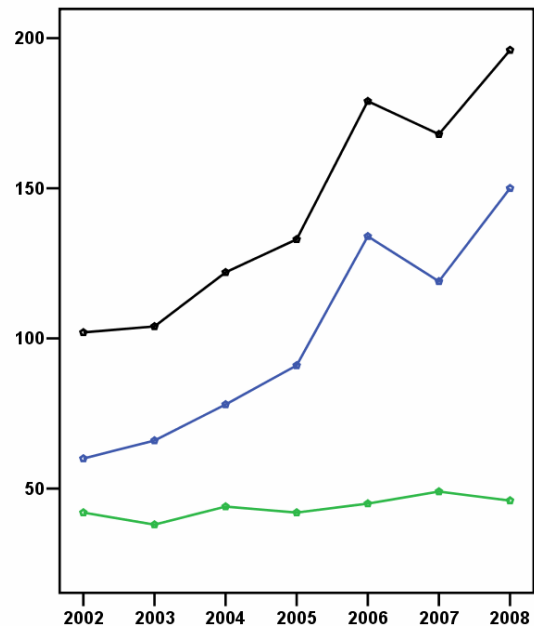


Figure 1. Annual-based evolution of the estimated total number of Iberian lynxes in Andalusia between 2002 and 2008 (black line), by subpopulations (Blue line: Sierra Morena; Green line: Doñana).

Lynx and rabbit monitoring programs

A monitoring program of both rabbits and lynxes is being carried out by the Andalusian Regional Ministry of the Environment since 2001. Rabbit abundance is continuously monitored - via transects and latrine censuses- in areas with lynx presence. A disease surveillance program (focusing mostly on RHD) is also carried out every year during critical time-periods. Iberian lynx monitoring is carried out by means of photo-trapping, radio-telemetry, direct observations and search for lynx signs (tracks, scats, etc.). Evaluation of actions undertaken at the population scale has been performed using information on inter-annual rabbit latrine survey in 2.5 km UTM squares, since its surface (625 Ha) is about the size of a breeding female home range (Ferrerías et al. 1997; Palomares, 2001). In Sierra Morena, treated squares have been compared to control ones (without management actions).

Genetic reinforcement

The low effective size has, in fact, reduced the genetic diversity of both Iberian lynx wild populations and, data shows that genetic drift has taken place in Doñana and Sierra Morena. In addition, Doñana's genetic diversity is 30% lower than that of Sierra Morena (Godoy et al. unpublished data). The most effective way to increase genetic diversity in the Doñana Iberian lynx population is via translocations of individuals from Sierra Morena. To date, two individuals from Sierra Morena have been released in Doñana, one in 2008 and the second one in 2009. Both were soft-released, being kept between 2-4 weeks in large (3-8 Ha) acclimatization pens placed within the most protected environs of the Doñana area. Translocations will continue to take place until at least four individuals from Sierra Morena will settle in Doñana.

Results

Conservation strategy

- Stabilization and compression of territories, and settlement of new ones: When comparing treated squares (squares where TRUs have been performed into) with control ones, the former increase overall rabbit abundance in 6 years, has been about 100%, whereas untreated squares have not shown significant differences in rabbit abundance during the same period of time. The most effective conservation measures have been the supplementary feeding stations and the rabbit release enclosures. Yet, the most important aspect that affect rabbit population dynamics in our study area has been the RHD (Figure 5; see Calvete, this book). In Doñana, rabbit abundance has slightly increased since 2002, although the effect of the carried out actions is not clearly related to this increase. Due to landscape features, the evaluation of actions over the rabbit population, however, is much more difficult to be performed in Doñana than in Sierra Morena. A TRU has been considered successful when the rabbit increase has allowed the settlement and reproduction of an adult female, while we have considered them partially successful if settlement has been recorded but without reproduction. Most TRUs both in Sierra Morena and in Doñana have been considered successful. In Sierra Morena, eight out of the 13 established TRUs can be considered successful and two partially successful. A total of 12 new Iberian lynx territories have been established in Sierra

Morena since 2001, where the population has increased from 38 adults (considering animals over 1 year-of-age; $n=60$ individuals including young-of-the year) in 2002 to 95 adults ($n=150$ individuals including young-of the year) in 2008 (Figure 6). In Doñana, nine new female Iberian lynx territories have been established since 2002, although the effect of the habitat management units is difficult to determine. The number of individuals in the Doñana population has remained stable at approximately 31-32 adults, yet the number of territorial females has increased from 10 to 19 between 2002 and 2008 (Figure 7), while the total number of males has decreased. This is largely due to the loss of males after the 2007 Feline leukaemia (FeLV) outbreak in the Coto del Rey nuclei (López et al. 2009; Meli et al. 2009).

- Decreasing causes of mortality: Measures to control road mortality seem to be working efficiently up to now. Although road kills increased during 2003-2006, they have decreased during 2006-2009 (Figure 8; Fernández et al. this book). Control of illegal hunting methods has helped decrease the number of leghold traps and snares to almost zero. In 2008, the epidemiological surveillance program allowed to detect the first case of poisoning in an Iberian lynx. Fortunately, the perpetrator could be identified and he is currently down by law. The epidemiological surveillance program is also working effectively. Thanks to this program, which is led by staff of the Andalusian Government, the *Ex-situ* Conservation program and the University Zürich, an outbreak of feline leukemia virus that occurred in Doñana population in 2007 could be rapidly detected and controlled (López et al. 2009; Meli et al. 2009;).

- Public awareness campaigns: The outreach campaigns have made the population aware of the situation of the Iberian lynx and supportive of the proposed reintroduction in this area. Altogether, outreach efforts have largely focused on the most critical groups: hunters, farmers, local populations of Doñana, Sierra Morena and the proposed release site, and children. As part of these communication efforts, the LIFE conservation project publishes a monthly bulletin and maintains an updated webpage (www.lifeline.org). Public information is proving to be a very effective tool for local awareness and support.

- Reintroduction: Habitat management measures are presently being carried out in the Guadalmellato Valley, where the first Iberian

lynx reintroduction is scheduled to take place. Six wild-born individuals will be released during the second half of 2009. The selected method includes a soft-release technique in on-site acclimatization enclosures of approximately 4 Ha each. Most landowners in the area have already signed collaborative agreements with the Andalusian Regional Ministry of the Environment within the frame of the Iberian lynx LIFE-Nature conservation project..

Genetic reinforcement

The genetic and demographic reinforcement performed in Doñana via translocations of Sierra Morena males is still incipient. The first translocated individual was released in Doñana National Park in January 2008. It was a three-year-old male named "Baya" that belonged to the Cardena subpopulation (Ruiz et al. this book). It was released in Coto del Rey subpopulation, (a high rabbit density place) where there were three adult females and no adult males due to the FeLV outbreak that took place in 2007. Baya immediately settled in the area and copulated with the three territorial females. All three became pregnant and gave birth to a total of eight young, of which three were still alive by the end of 2008. The introduction of Sierra Morena genes into the Doñana population is considered essential for the long-term genetic health of this wild population. In January 2009, "Caribú", a three-year-old male that originated from the Andújar subpopulation, was soft-released in Doñana's Biological Reserve, one of the best protected areas within the National Park. This place is a low-density rabbit area where one adult female and one adult male inhabit. In contrast with the first experience, this male has not shown any interest on attempting to settle within the vicinity of the release site, and it is currently dispersing northwards out of the Doñana population in a moving considered as "homing behaviour". Experiences with other lynx species (unpublished data) point out the possibility that this individual returns to the Doñana population by itself after several months.

Safeguarding genetic diversity in captivity

Both LIFE conservation projects have helped provide all the Iberian lynx founders that presently conform the Iberian Lynx Conservation Breeding Program (Vargas et al. this book). Since 2002, 36 individuals have been brought into captivity (6 from Doñana and 30 from Sierra Morena; Table 1). From these, 24

lynxes were specifically extracted for the captive breeding program, whereas the other 10 lynxes were brought into captivity because they had problems that compromised their survival in the wild. The extraction rate was based on a model developed in order to avoid potential impacts on the population dynamics of this species (Palomares et al. 2002).

Discussion

The conservation strategy developed for the Iberian lynx in Andalusia seems to be advancing in the right way. The Sierra Morena population has experienced a substantial increase since 2001, when active conservation measures began to be implemented. The number of lynxes in this population has increased at a much higher rate than the surface area they are occupying; i.e., territories are being "compacted" and there seems to be current evidence of population saturation in Sierra Morena (LIFE project, unpublished data). The productivity is following an every-other-year cycle, probably due to the stratification of one-year-old individuals inside the population that hinder the survival of cubs born the following year. Moreover, about 70% of the juveniles are lost within their first year of life, and there have been several recorded cases of intraspecific aggression, which seems to be one of the major causes of this loss. Moreover, juveniles are currently settling in areas with very low rabbit densities or with very poor habitat structure. Population expansion seems difficult due to lack of resources in all areas surrounding the current distribution area. Nevertheless, there are suitable areas for the establishment of Iberian lynx within 15-45 km from the current Andújar-Cardena population. In fact, all available information points towards asserting that the rapidly growing Yeguas River subpopulation has motivated the foundation of a new population nucleus in Southern Castille-La Mancha Community, 15 km away from the Andalusian Sierra Morena population (Castille-La Mancha Government, unpublished data). Iberian lynx conservation efforts in Doñana have promoted the establishment of an important number of new territories outside the National Park during the period 2001-2009, which have made up for the dramatic loss of territories suffered inside the national park between 2001-2003. The key to this change of trend outside the National Park has been the awareness of the local population and the decrease in road mortality. The

number of territorial females (19) and wildborn cubs (24) recorded in Doñana in 2008 have been the maximum ever recorded in this population. We believe that all the abovementioned measures have prevented the extinction of this small population. However, more active management measures are needed to ensure the long-term conservation of this population. Reintroductions are considered to be key in moving forward towards the recovery of the endangered Iberian lynx. Once the protection and effective conservation of the currently existing populations are granted, the next step is the establishment of new free-ranging lynx populations in areas of historical occupancy. The Iberian lynx is critically endangered, and can only be recovered through hard work and solid partnerships between national and international stakeholders. The effort of politicians, biologists, scientists, veterinarians, hunters, and the whole society is sorely needed to help recover the lynx populations that roamed in the past throughout the Mediterranean forests and scrublands of the Iberian Peninsula.

Acknowledgements

F. Coves, J. Guirado, C. Gañán, M. Delibes, F. Palomares, N. Fernández, A. Vargas, F. Martínez, M. Navarro, M. Briones, and N. Guzmán give support to the all the Iberian lynx conservation programs carried out in Andalusia. The other members of the LIFE project field team (G. Valenzuela, G. Garrote, M.A. Díaz-Portero, A. Leiva, E. Rojas, R. Arenas, J. Rodríguez, J. Bueno, S. Lillo, J. Pérez, M.I. García, M. Moral, J.A. Báñez, R. Sanabria, R.B. Millán, D. Palacios, J.M. Martín, CBD Hábitat Foundation team and WWF/Adena team), together with the rest of the LIFE project team (S. Saldaña, M.P. Delgado, R. García, I. Tenorio, M. Vara, Ecologistas en Acción, SECEM, ATECA, APROCA and FAC) are making all this work possible.

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The Iberian lynx Conservation Breeding Programme

Astrid Vargas^{1*}, Íñigo Sánchez², Fernando Martínez¹, Antonio Rivas¹, José Antonio Godoy³, Eduardo Roldán⁴, Miguel Ángel Simón⁵, Rodrigo Serra⁶, M^a José Pérez⁷, Alex Sliwa⁸, Miguel Delibes³, Miguel Aymerich⁹ and Urs Breitenmoser¹⁰.

1. Programa de Conservación *Ex-situ* del linco ibérico. Centro de cría "El Acebuche". Parque Nacional de Doñana. 21760. Matalascañas, Huelva. *lynxessitu@lynxessitu.es
2. Zoobotánico de Jerez. c/ Taxdirt s/n. 11404. Jerez de la Frontera, Cádiz.
3. Estación Biológica de Doñana/CSIC. c/ Américo Vespucio, s/n, 41092, Sevilla.
4. Grupo de Ecología y Biología de la Reproducción. Museo Nacional de Ciencias Naturales. c/ José Gutiérrez Abascal, 2. 28006. Madrid.
5. Iberian lynx LIFE conservation project. Consejería de Medio Ambiente, Junta de Andalucía. c/ Fuente del Serbo, 3. 23071. Jaén.
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10. IUCN Cat Specialist Group. Institute of Veterinary Virology. University of Bern. Laenggassstrasse, 122. 3012. Bern.

Abstract

The Iberian Lynx Conservation Breeding Program follows a multidisciplinary approach, integrated within the National Strategy for the Conservation of the Iberian lynx, which is carried out in cooperation with national, regional, and international institutions. The main goals of the *ex-situ* conservation program are to: (1) maintain a genetically and demographically-managed captive population; (2) create new Iberian lynx free-ranging populations through re-introduction. To achieve the first goal, the Program aims at maintaining 85% of the genetic diversity presently found in the wild for the next 30 years. This requires managing 60(30.30) Iberian lynx as breeding stock. Growth projections indicate that the *ex-situ* program should achieve such population target by the year 2010. Once this goal is reached, re-introduction efforts could begin. Thus, current *ex-situ* efforts focus on producing physiologically and behaviorally sound captive-born individuals. To achieve this goal, we use management and research techniques that rely on multidisciplinary input and knowledge generated on the species life history, behavior, nutrition, veterinary and health aspects, genetics, reproductive physiology, endocrinology and ecology. Particularly important is adapting our husbandry schemes based on research data to promote natural behaviors in captivity (hunting, territoriality, social interactions) and a stress-free environment that is conducive to natural reproduction. The aim of this communication is to provide an overview of how scientific knowledge from various disciplines is proving essential to shape management efforts within the Iberian lynx breeding program. The communication is divided into 6 sections: Genetic and Demographic Management, Captive Husbandry, Health and Veterinary Aspects, Reproductive Physiology, Reintroduction, & Outreach/Capacity Building.

Introduction

Owing to the precarious situation of the Iberian lynx (*Lynx pardinus*) in the wild (Calzada et al. 2009; Simón et al. 2009), conservation measures need to be implemented effectively and efficiently, integrating efforts and working

tools. Iberian lynx conservation could be conceived as a puzzle whose pieces should fit together adequately. One of such pieces involves *ex-situ* conservation, which includes - among other activities- captive breeding,

genetic and demographic management of the captive population, management of a Biological Resources Bank (BRB), preparing captive-born animals for release, as well as capacity building, education, and outreach efforts (Vargas et al. 2005b).

Current Iberian lynx conservation breeding efforts focus on producing physiologically and behaviorally sound captive-born individuals (Figure 1) that are suitable for future reintroduction efforts. For this purpose, we use management and research techniques that rely on multidisciplinary input generated on the species' life history, behavior, nutrition, veterinary and health aspects, genetics, reproductive physiology, endocrinology, and ecology. The program stresses the importance of adapting our husbandry schemes based on research data to promote natural behaviors in captivity (hunting, territoriality, social interactions) and a stress-free environment that is conducive to natural reproduction. Some relevant research areas include: determining fecal hormone profiles for adult and subadult lynx (Pelican et al. 2009; Dehnhardt et al. 2009) studying reproductive behavior and cub development (Antonevich et al. 2009), determining reproductive health of male and female breeders (Roldán et al. 2009; Goeritz et al. 2009), developing a non-invasive pregnancy test (Jewgenow et al. 2009), establishing sound bio-security and biomedical protocols (Martínez et al. 2009), establishing reference values for blood parameters (Pastor et al. 2009; García et al. 2009), genotyping all founders and making pairing recommendations based on genetic distance between breeders (Godoy et al. 2009; Leus & Lacy, 2009). Within this scheme, one of our goals is to minimize the use of potentially invasive methods while simultaneously enhancing the trust between the animals and their keepers to assist in securing information on animal weight and gestational status. Over the past four years, a total of fifteen pregnancies have resulted in the birth of 31 live offspring, of which 24 survive to date (Vargas et al. 2008a). This text describes various organizational aspects of the Iberian lynx Conservation Breeding Program, while emphasizing how results from multidisciplinary life science research can be integrated into an adaptive management approach to help recover the world's most endangered felid species.

The Iberian Lynx *Ex-situ* Conservation Program

The Iberian lynx *ex-situ* conservation program is integrated within the National Strategy for the Conservation of the Iberian lynx, officially endorsed by the Spanish National Commission for the Protection of Nature (MARM, 2008; Calzada et al. 2009). National, regional and international institutions collaborate with the program, which is currently implemented through a "multilateral commission" that includes the central governments of Spain and Portugal, together with the autonomous governments of Andalusia, Extremadura and Castille-La Mancha, Spain. Portugal, where no Iberian lynx populations were detected during the last 2002–2003 census, has developed its own *ex-situ* conservation action plan in coordination with the Spanish program (Serra et al. 2005) and it is presently building a captive breeding facility while beginning to work on improving habitat for future re-establishment of lynx populations (Sarmiento et al. 2009).

The route map of the *ex-situ* Program is the Iberian Lynx Captive Breeding Action Plan, an adaptive management document reviewed annually by an advisory captive breeding committee. The Plan's most recent update, which emphasizes habitat conservation using *ex-situ* efforts as a working tool, has been recently endorsed by the highest Spanish authorities at the Sectorial Conference, in May 2008 (Vargas et al. 2008b). The main *Ex-situ* Program goals are twofold: (1) To maintain a genetically and demographically managed captive population that serves as a "safety net" for the species and (2) To help establish new Iberian lynx free-ranging populations through re-introduction programs. To accomplish these goals, the Iberian lynx conservation breeding program encompasses management and applied research strategies in the following six areas.

1. Genetic and demographic management of the captive population.

Iberian lynx genetic goals for the *ex-situ* program were established based on recommendations provided by the IUCN/SSC Conservation Breeding Specialist Group in collaboration with the Iberian lynx *in situ* conservation managers. Population modelling using the programme PM2000 (2004) revealed that it would be impossible to maintain 90% of existing gene diversity for 100 years because

the wild population could not withstand extraction of the required 12 founders per year for 5 years (Lacy and Vargas, 2004; Leus and Lacy, 2009). At the time of this first analysis in spring 2004 there were six wild-caught lynxes already at El Acebuche captive breeding center, a facility that was constructed in 1991 in Doñana National Park, Southeastern Spain. At the same time, modelling suggested the feasibility of maintaining 85% of the existing genetic diversity for the next 30 years. The outcome eventually would be the *ex-situ* management of 60 individuals (30 males, 30 females) as breeding stock (Lacy and Vargas, 2004; Godoy et al. 2009; Leus and Lacy, 2009). This goal could be attained by adding four founders (mostly cubs or juveniles) per year for 5 years as well as one extra founder every 2 years (from the handicapped lynxes that normally enter rescue centres) for the entire duration of the programme. This level of extraction rate would have a minor impact on the viability of the wild populations according to the model designed by Palomares et al.

(2002). Following this scheme, the programme could achieve its population target of 60 individuals by 2009 and reintroduction of captive-raised lynxes could begin in 2010, provided that adequate habitat was prepared. Originally, modelling predicted the availability of 12 to 13 captive-born lynxes annually from 2011 through 2019 (Lacy and Vargas, 2004). A recent update of the projections, using data from actual captive reproduction over the past 5 years, indicates that the annual number of animals available for release every year will oscillate between 20 and 40 Iberian lynxes (Godoy et al. 2009).

Between 2002 and 2008, the Andalusian government has culled 26 wild-born Iberian lynxes for the captive breeding program; in addition, 10 more lynxes were brought into captivity because they had problems that compromised their survival in the field (Simón et al. 2009) although not all of these lynxes can be considered as potential founders of the captive population. The year 2008 marked the population. The year 2008 marked the end of



Figure 1. Iberian lynx female with captive born cubs..

the planned extractions of wild lynxes for incorporation into the breeding population; still, those found injured or handicapped -with compromised probabilities of survival in the wild- will continue to trickle into the breeding program. At the time of writing this communication, there are 58 lynxes in the breeding program (Figure 2), of which 42 (28 founders and 14 captive-born) and 6 (all founders) have pure Sierra Morena and Doñana ancestry, respectively. Ten more lynxes born in captivity descend from crossings between Doñana and Sierra Morena founders. Thus, the program is presently one year ahead of growth projections (Godoy et al. 2009) and can already begin to provide captive-born lynxes for reintroduction efforts.

To provide the captive space needed to achieve the program's genetic goals, construction of two new Iberian lynx breeding centers -Granadilla (Extremadura) y Odelouca (Silves, Portugal)- is under way. An additional center in Cabañeros (Castille-La Mancha) is also scheduled for future construction. All breeding centers are strategically placed so they can be co-managed by administrations that commit to habitat preparation for future reintroductions. Breeding centers will function as a network, following EEP standards, each managing approximately eight Iberian lynx breeding pairs. All centers considered, the captive population will be managed as a metapopulation, a genetic counterpart to the Sierra Morena and Doñana free-ranging populations (Godoy et al. 2009). Construction of breeding centers is tightly linked --via Memorandums of Understanding-- to a regional compromise to prepare habitat for future reintroduction efforts. In this fashion, in-situ and *ex-situ* efforts work together towards a common goal.

2. Captive husbandry and behavior

Captive husbandry is based on multidisciplinary input from a variety of animal-care fields, such as nutrition, behavior, genetics, physiology and veterinary medicine, together with the systematic use of the scientific method. One of the Program's key husbandry challenges is to strike a balance between fostering natural behaviors in captivity (hunting, territoriality, social interactions, etc.) and creating a stress-free environment where animals are more prone to mate. For this reason, the breeding program favours naturalistic enclosures and

promotes natural behaviors via environmental enrichment (Manteca, 2009; Martos, 2009). In order to get important information about the animals (such as their body mass or determining whether or not the females are pregnant), certain training techniques are being used. Some of these include obtaining regular weights by having the lynx step on a measuring scale. Such techniques are designed to avoid using invasive methods, which would stress the animals, and they also serve as a way to strengthen the trust between the animals and their keepers. Special care is taken to avoid domestication of captive lynxes, although this becomes a greater challenge in the case of hand-reared, abandoned cubs (Rivas et al. 2009).

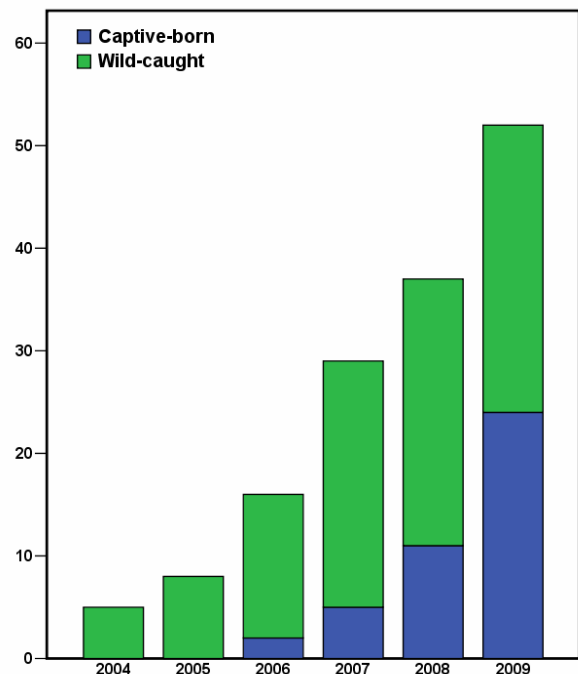


Figure 2. Population growth of the Iberian lynx captive breeding program from Jan 2004 to Jan 2009 (Blue bars = captive-born cubs; green bars = wild-caught individuals).

Lynx behavior and activity patterns are also being carefully observed by a round-the-clock video surveillance system (see description in Vargas, 2005a) (Figure 3), which provides a great deal of information on conducts that could not easily be learned through observations in the wild. For instance, we have learned that mating behavior in the Iberian lynx follows a similar pattern as that of other felids (Figure 4) (Lanier & Dewsbury, 1976; Rodríguez-Llanes, 2006). Breeding season in captivity takes place throughout January and February, with most



Figure 3. Round-the-clock video surveillance system.

births occurring in March and April, as it is the case in the wild (Palomares et al. 2005; Palomares et al. 2009). The actual mating period lasts between two and three days during which lynxes copulate an average of 28 times (range: 13-65; $n=460$ bouts in 21 pairs; Table 1). Gestation period, counted from the time of the first copulation, ranges from 63-66 days ($n = 12$). All parturitions that have occurred before 61 gestational days ($n=4$) have resulted in the birth of either dead or weak and not completely developed young, which were considered premature. Although variation between individuals is very high, most females are very consistent regarding their own timing to enter estrous and regarding the number of days they are gravid (Table 1). Labour during whelping (regarded as the time lapsed between the first visible contractions until the delivery of the last offspring) varies widely between females, with some of them delivering each young within 10-15 min intervals while others taking up to 9 hours between the delivery of each offspring (Table 1). Primiparous females have a higher rate of failure to raise their young than multiparous ones. Out of the eight

females that have whelped at El Acebuche center, only two first-time mothers –Saliega and Aura– managed to nurse all their young during the first post-natal month. Two other dams –Esperanza and Boj– kept only one of their cubs and abandoned the rest of the offspring in their first litter, while three other females –Adelfa, Aliaga and Brisa – aborted one of their young and delivered the rest of the offspring prematurely during their first gestation. One female, Artemisa, aborted two undeveloped fetuses at 42 gestational days. Our video-surveillance system permits early detection of behavioral problems, which has resulted in the survival of 7 Iberian lynx cubs that were hand-raised after being abandoned by their first-time mothers (Rivas et al. 2009).

This unintrusive surveillance system has also allowed us to identify the existence of a sensitive period when Iberian lynx cubs become highly competitive to the point of siblicide (Vargas et al. 2005a; Antonevich et al. 2009). Spontaneous aggression erupted at 44 days of age in the first Iberian lynx litter born in captivity. The largest cub (a female) in a litter

of three was killed by a brother who delivered lethal bites to the larynx and skull. Agonistic behavior has been observed in nine of eleven subsequent Iberian lynx litters of two or more cubs, with the most intensive fighting occurring around the 7th post-natal week (Antonevich et al. 2009). This same phenomenon has been observed in the Eurasian lynx by Russian scientists at the Tchernogolovka facility who recorded aggressive behavior- in 16 of 31 litters, with deaths occurring in four cases (Naidenko & Antonevich, 2009). The authors indicate that the highest prevalence of agonistic behavior in Eurasian lynx cubs occurred at 36 to 64 days of age, with the greatest frequency during the 7th post-natal week. Although siblicide in Iberian lynx has not been directly observed in nature, a 1-month-old cub was found in the wild in 2003

with severe injuries compatible with bites from another cub. It may also be relevant that free-ranging Iberian lynxes generally give birth to three cubs, but 70% of the females are usually observed with only two young after approximately three months post-parturition (Palomares et al. 2005). Sibling aggression in Iberian lynx has influenced programme husbandry as staff must be vigilant and prepared to break up aggressive bouts during the sensitive period.

3. Health and veterinary aspects.

The health considerations involved in captive breeding, re-introduction and translocation programs are a source of great concern to conservation biologists, since captive-bred animals could potentially transmit infectious disease to wild populations (e.g., tuberculosis in

Year	Female	Male	Date 1st mating	Number of matings	Gestation (days)	Labour	Cubs born	Cubs weaned
2005	Saliega	Garfio	23/01/2005	> 5 (unk)	64	?	3	2
2006		Garfio	19/01/2006	43	63	20'	2	2
2007		Jub	18/01/2007	43	64	1 h	2	2
2008		Jub	18/01/2008	65	64	32'	3	2
2005	Aura	Jub	28/01/2005	25	NP	N/A	N/A	N/A
2006		Garfio	30/01/2006	26	NP	N/A	N/A	N/A
2007		Garfio	15/01/2007	21	65	6 h	3	3
2008		Garfio	07/02/2008	21	65	5h 55'	2	2
2005	Esperanza	Garfio	11/02/2005	20	NP	N/A	N/A	N/A
2006		Jub	09/02/2006	25	65	>6h	2	2**
2007		Jub	03/01/2007	24	66	4h 40'	2	1
2008		Jub	06/02/2008	33	66	2h 43'	1	1
2006	Aliaga	Cromo	15/02/2006	13	56*	50'	2	0
2007		Cromo	22/01/2007	23	NP	N/A	N/A	N/A
2008		Cromo	07/02/2008	33	64	5h 22'	3	3***
2006	Adelfa	Cromo	19/01/2006	19	NP	N/A	N/A	N/A
2007		Cromo	27/01/2007	33	61*	1 h	3	0
2008		Garfio	01/02/2008	32	63	3h 25'	2	2
2007	Artemisa	Almoradux	02/01/2007	23	42*	N/A	2	0
2008		X	X	X	X	X	X	X
2007	Brisa	Arcex	20/01/2007	15	NP	N/A	N/A	N/A
2008	Brisa	Arcex	19/01/2008	25	59*	9h	2	1***
2008	Boj	Arcex	02/02/2008	31	65	6h	3	2***

Table 1. Mating, gestation, parturition and cubs weaned (3-months-of age) of captive Iberian lynx between 2005 and 2008 (NP= Non pregnant; N/A = Not applicable; * = whelping before due time; ** = one cub hand-raised; *** = all cubs hand-raised).

reintroduced Arabian oryx, *Oryx leucoryx*; Viggers et al. 1993) and vice-versa (e.g., wild-caught black-footed ferrets transmitted canine distemper to potentially uninfected, captive individuals; Williams et al. 1988). It is generally felt that most of these conservation programs were lacking: (1) sufficient information on disease distribution and risk in captive populations; (2) sufficient information on disease incidence, distribution and risk in wild populations; (3) quarantine systems to prevent disease transmission; (4) a system to adequately track and detect pathogens.

Because relatively little was known about the diseases affecting the Iberian lynx, actions to improve our knowledge of the main diseases affecting the species was imperative. The Iberian Lynx Conservation Breeding Program established a Veterinary Advisory Team (GAAS) dedicated to address diverse aspects of veterinary and research management, as well as protocol development. To improve the understanding of the various diseases that could potentially affect the species, the Program's main lines of action involve the establishment of preventive disease protocols for the captive population, capacity building of veterinary staff working with in-situ and *ex-situ* populations, and conducting research on general veterinary science (Martínez et al. 2009).

Research projects have helped determine the incidence and prevalence of infectious pathogens in captive and wild lynx populations (Millán, 2006; Meli et al. 2009; López et al. 2009), determination of normal vs pathological blood values (Pastor et al. 2009; García et al. 2009), and establishing parameters that point towards a potential renal disfunction in wild and captive lynxes (Jiménez et al. 2008; Jiménez et al. 2009). The results of research, protocol development, and standardization efforts, coupled with dissemination and sharing of knowledge and experience among veterinarians working in the program are all contributing to more consistent diagnosis and treatment.

4. Reproductive physiology.

Reproductive physiology studies and associated technologies increase the success rate of any captive-breeding program and are important in helping with the conservation of wild felids in captivity. Reproductive technologies are available for three major purposes: (1) assessing fertility and monitoring reproductive status; (2)

assisting in breeding and maintenance of gene diversity; and (3) learning more about reproductive mechanisms of the endangered Iberian lynx.

Consistent with the need for more basic species information, our husbandry and breeding efforts have established/reconfirmed normative data on sexual maturity (males, 3 years; females, 3 years; Roldán et al. 2009; Goeritz et al. 2009, respectively), oestrus length (3-7 days), copulations per pairing (28) and gestation interval (63-66 days) (see above). Faecal hormone monitoring has demonstrated that females experience ovarian cycles from January through May whereas males maintain testosterone year-round (Pelican et al. 2009; Dehnhardt et al. 2009). But species peculiarities also have been revealed, for example, oestrogen metabolite concentrations during pregnancy are 10-fold greater than in other felid species (Brown, 2006; Brown, 2009; Pelican et al. 2009; Dehnhardt et al. 2009). Additionally, progesterin excretion profiles are unusually lengthy, largely because ovarian corpora lutea (sites of earlier ovulations) remain active much longer than in most other felids (Goeritz et al. 2009). Prolonged non-pregnant luteal activity has also been described in the closely related Eurasian lynx (*Lynx lynx*); (Jewgenow et al. 2006; Dehnhardt, 2009) and in the Canada lynx (Fanson, 2009) suggesting an idiosyncratically-conserved mechanism for lynx in general.

Regarding male reproductive physiology, sperm traits seen in the Iberian lynx are lower than those reported for some other felid species, yet higher than those reported for the Eurasian lynx and the bobcat (Gañán et al. in press; Roldán, 2009). It has also been found that thawed-out Iberian lynx spermatozoa are capable of fertilising viable in vitro matured domestic cat oocytes, thus opening up the possibility of examining functional capacity of spermatozoa from this species under laboratory conditions (Gañán et al. in press).

The *ex-situ* population is also being used to explore a novel means of diagnosing pregnancy. Because all ovulating lynxes produce rising progesterone (regardless of conception), conventional hormone monitoring is not useful for identifying a gestating female (Pelican et al. 2009). However, increasing concentrations of the hormone relaxin (in blood or urine) are indicative of

pregnancy in felids (Van Dorsser et al. 2007). A unique non-invasive means of collecting blood has been developed using blood sucking Triatomine bugs (*Rhodnius prolixus* or *Dipetalogaster maxima*) placed in specially-drilled hiding holes in the lynx's cork nestbox (Voigt et al. 2007; Jewgenow et al. 2009). As much as 3 ml of blood can be extracted per bug, more than adequate for the assay. Urine is captured using special collection devices distributed throughout the animal's enclosure. Pregnant females express a positive relaxin signal from 32 to 50 day post-copulation of a 65-day gestation (Jewgenow et al. 2009), thereby allowing managers to prepare for an impending parturition.

In order to assist breeding and maintaining genetic diversity, the Iberian Lynx *Ex-situ* Program collaborates with the maintenance of Biological Resources Banks (BRBs) for conservation of biomaterial gathered from wild and captive Iberian Lynx populations. Biomaterials are presently being safeguarded at two locations: the National Museum of Natural Sciences in Madrid (Roldán et al. 2009) and the Miguel Hernández University in Elche, Alicante (León et al. 2009). Although the Museum of Natural Sciences places special emphasis on reproductive samples and the MH University focuses on multipotential somatic cells, both banks preserve tissue, blood, serum, and other biological materials. The conservation of gametes will allow the Breeding Program to extend future options without the limitation of space or the risk of disease transmission, while opening the opportunity of prolonging the possibilities of reproduction for individual animals after their death (Roldán et al. 2009). The storage of samples at both BRBs provides a safety net for these valuable materials and ensures that biosamples will be available for additional analysis whenever needed, which is a crucial resource for potential retrospective studies.

5. Re-introduction.

The small size of current Iberian lynx free-ranging populations renders them highly vulnerable to stochastic events. Thus, it is imperative to create, as soon as possible, new wild populations, while simultaneously increasing numbers in the existing ones. Prior to any re-introduction/translocation a detailed viability study is required (IUCN Guidelines for Re-introductions) (IUCN, 1998). It is important to

determine if the cause or causes that brought the species to extinction in the specific area have been eradicated and, if so, if there is administrative and local population support for the program and if the habitat is prepared to support a viable population of the species (Simón et al. 2009). All re-introductions and translocations must be performed using scientific support and the Iberian lynx should be no exception (Calzada et al. 2009). Such conservation techniques require an interdisciplinary approach, with input from experts in ecology, veterinary medicine, genetics, physiology and behavioral sciences, as well as support from socio-political and information sciences. All stages of programme development and implementation must have well-defined protocols that document objectives, methodology, responsibilities, as well as the accountability of the organizations and individuals involved (MARM, 2008).

The Andalusian government, by means of the current LIFE-Nature Project for the Reintroduction of the Iberian lynx in Andalusia, has evaluated different potential areas for lynx reintroduction in this Spanish region and has selected two areas that comply with IUCN criteria (Simón et al. 2009). The first reintroduction is scheduled to take place in 2009 using wild-born individuals from the Sierra Morena population (Simón et al. 2009). Reintroduction of captive-born lynxes is scheduled for 2010. Captive candidates will be chosen based on genetic and behavioural criteria. Preparation for release will include maintaining animals in large preconditioning enclosures with minimal human contact and exposure to natural stimuli, including live prey. Lessons from other reintroduction programs will be essential for the planning and implementation of the first Iberian lynx reintroduction.

6. Capacity Building and Outreach Efforts.

Awareness, education, and scientific training are essential to all conservation breeding programs. Education and awareness efforts should be focused on changing prevalent attitudes that contribute to habitat destruction and species extinction. One advantage enjoyed by conservation breeding programs is their ability to gain public attention, particularly if the animal in question is charismatic and attractive to the broader public. The Iberian lynx is one such case, and raising public

awareness for the need for habitat conservation to guarantee survival of the species in the wild is one of the Program's objectives. It is important to emphasize that breeding and keeping lynxes in captivity, with no hope of ever returning them to the wild or of recovering the natural populations would be a pointless exercise. The breeding program encourages, cooperates with, and supports media interest in the Iberian lynx, while taking every opportunity to remind the public of the primary importance of habitat conservation.

One way of providing an open window to the Breeding Program is by sharing on-line information via a frequently updated Iberian lynx web page (<http://www.lynxexsitu.es>), featuring monthly newsletters, pictures and videos of all captive lynxes, along with general interest and scientific articles, plus information on the Program's protocols and working methods. An English language version is currently in production to further expand the scope of communication and awareness efforts. As part of its training efforts, El Acebuche Center organizes on-site internships for recent college graduates interested in acquiring first hand knowledge on the Iberian lynx conservation program.

Conclusions

To date, the Iberian lynx conservation breeding program includes 58 (30.28) lynxes and is ahead of Action Plan forecasts (Lacy and Vargas, 2004). Between 2005 and 2008, fifteen litters have been produced (three prematurely) with 24 surviving young (Figure 2). The Program's management team works closely with in situ conservation authorities, managers and researchers. For example, skills and resources are being shared to address the increase in disease concern for lynxes and prey, and projects are coordinated to monitor genetic status of the wild and captive populations. Most importantly, our multidisciplinary research is generating new insights into the unique biology of this species while our intensive management is on course to meet the overall recovery goal of attempting reintroduction of captive-raised lynxes by 2010. Finally, the Program carries out a capacity-building plan aimed at preparing professionals for working on endangered species conservation as well as sensitizing the general public and decision-makers about the importance of conserving habitat for the

recovery of this charismatic felid. In this fashion, the guidelines and experiences offered in 2009 illustrate how an *ex-situ* breeding programme can be integrated to compliment the challenge of sustaining wild Iberian lynxes in nature, which is always the highest priority.

Acknowledgements

The authors would like to thank all the institutions (and associated staff) that collaborate with the Iberian Lynx Conservation Breeding Program, including the Spanish Ministry of the Environment, the governments of Andalusia, Extremadura, Castilla-La Mancha and Portugal, the Life-Nature Project for Iberian Lynx Conservation in Andalusia, the Species Survival Commission of the International Union for the Conservation of Nature (Cat Specialist Group and Conservation Breeding Specialist Group), the National Scientific Research Council (Doñana Biological Station, the National Museum of Natural Sciences, and Almeria Arid Zones Station), the Seville Diagnostic Analysis Center, the Autonomous University of Barcelona, the Miguel Hernández University, the Complutense University of Madrid, the University of Huelva, the University of Cordoba, the University of Zurich - Vet Clinical Laboratory (Switzerland), KORA (Switzerland), the Smithsonian Institution (USA), the Jerez Zoo, the Terra Natura Foundation, the Fuengirola Zoo, the Barcelona Zoo, the European Association of Zoos and Aquariums, the Iberian Association of Zoos and Aquariums, the International Society for Endangered Cats, PolePosition, The BBVA Foundation, the Andalusian School of Biologists, and the SEO/Birdlife and Doñana National Park volunteer programs.

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Block B: “The wider picture”

Guidance, strategic approaches and conservation tools



Goals and approaches for the Iberian lynx

Urs Breitenmoser

IUCN Cat Specialist Group. Institute of Veterinary Virology. University of Bern.

u.breitenmoser@kora.ch

Laenggassstrasse, 122. 3012. Bern.

Abstract

Since the Iberian lynx has been listed as Critically Endangered in the IUCN Red List in 2002, it has been in the focus of the international conservation community. In Spain and Portugal, the struggle for the survival of the world's most threatened cat species goes back to the years 1973 and 1974, respectively, when the Iberian lynx was protected by law. However, the continued decline of the populations in both countries demonstrated how futile legal protection alone is: Although intelligence to effectively conserve the Iberian lynx was generated, it took almost another thirty years until the society as a whole was ready to take up the assignment. But still today, responsibilities and competences more often challenged than shared among key players. Conservation of the Iberian lynx –facing virtually all possible extrinsic and intrinsic threats to a medium-sized cat in a human-dominated landscape– is a highly complex endeavour that must involve all parts of the civil society, not only experts and institutions engaged in nature conservation. All conservation measures implemented bare the risk of failure. As long as responsibility is not shared and risks are mutually borne, important decisions are delayed and opportunities might be missed. The first two goals of the Iberian lynx recovery plan – to halt the further decline of the remnant populations and to establish a captive population – are largely fulfilled. The next step is to enhance the distribution of the lynx. Under the present conditions, this is best achieved through reintroduction. To create new (independent) populations is also important to reduce the risk that any catastrophic event affecting the two remnant populations could eradicate the Iberian lynx in the wild. Reintroduction involves new key players and stakeholders and bears new risks. Several approaches and procedures are possible, all involving certain risks and difficult decisions. A plan for the start of the reintroduction must be discussed in this seminar to achieve a broad consensus among key players. Only carefully planned, effectively implemented and rigorously monitored actions can secure the creation of new populations and hence the survival of the Iberian lynx. "Laissez fair" has brought the Iberian lynx to the verge of extinction. With only two small and extremely vulnerable population nuclei left, an active and courageous conservation approach is indispensable.



Strategy for the conservation of the Iberian lynx in Spain

Javier Calzada

Dpto. Biología Ambiental y Salud Pública. Universidad de Huelva.

javier.calzada@dbasp.uhu.es

Avda. de las Fuerzas Armadas s/n. 21071. Huelva.

Abstract

A new Strategy for the Conservation of the Iberian Lynx (*Lynx pardinus*) has recently been approved by the Spain's maximum authorities in Environmental Policy in the Sectorial Conference of Environment. The new Strategy has been developed in a different working framework from the one that led to the first Strategy for the Conservation of the Iberian Lynx in 1999. The demographic situation of the Iberian lynx has never been worse. However, there have never been so many human and financial resources available, and the species has never been the focus of so much public attention and concern. The ultimate goal of the Strategy is to make the Iberian lynx a functional part of the Mediterranean scrubland habitat again. To this end, the recovery of the species involves both successfully managing the remaining populations and choosing and restoring areas to carry out reintroduction projects that will lead to the establishment of new wild populations. The new Strategy has set a roadmap for the conservation and recovery of the Iberian lynx, as well as specific numerical targets that must be met in a given period of time. They are the following: (1) Stabilize the populations by combating the causes of threat to the species, (2) Increase the number of individuals in the wild populations so that the Iberian lynx can be downlisted from CR, Critically Endangered to EN, Endangered by 2011, and (3) Increase the number of wild populations, so that the species can be downlisted from EN, Endangered to VU, Vulnerable by 2020. First target should be achieved through the Regional Recovery Plans that must adopt the guidelines set in the Strategy and develop them fully and efficiently. Achieving the second goal implies to increase the number of individuals in the lynx populations until at least one of them has more than 50 mature individuals – which must not amount to more than 90% of all the wild mature individuals. If necessary, restocking and population exchange projects should be done in order to increase the abundance of the species in the existing populations. To achieve the third target, the species must reach to the 250 mature individuals and not show signs of decline, and this will not be possible if not carried out habitat restoration and reintroduction projects in all the autonomous communities of Spain where the Iberian lynx occurs or occurred until recently.

The genetics of Iberian lynx populations: Implications for management

José Antonio Godoy¹, Laura Soriano, Severine Roques and Mireia Casas-Marcé.

Estación Biológica de Doñana/CSIC 1. godoy@ebd.csic.es

c/ Américo Vespucio, s/n, 41092, Sevilla.

Abstract

Molecular markers can be used to describe genetic patterns found within species (diversity, inbreeding, structure, etc.) and, when combined with proper models, to gain insights into the evolutionary and demographic processes that generated them (selection, genetic drift, bottlenecks, gene flow, etc.). Only with this information can conservation units be defined, short- and long-term viability evaluated and appropriate management strategies designed. We have analysed contemporary genetic variation in Iberian lynx (*Lynx pardinus*) for mitochondrial and microsatellite markers. A high level of genetic differentiation is observed between the two extant Doñana and Sierra Morena populations. Mitochondrial diversity is extremely low with only two haplotypes observed which differ in one single position. Nuclear microsatellite diversity levels are globally low when compared to other felid species, and is 33% lower in Doñana than in Sierra Morena, both for expected heterozygosity and allelic diversity. Reduced diversity is also found at MHC (Major Histocompatibility Complex) loci, with only three alleles observed at two loci, the three being present in Sierra Morena but only two in Doñana. A signal for a recent demographic bottleneck is present in the data for both populations. Observed genetic patterns are consistent with the predominance of genetic drift in recent times affecting both populations, but more intensely Doñana, where the magnitude and rate of accumulation of inbreeding could be severely affecting population viability. Increased risk of inbreeding depression and steep rate of loss of genetic diversity, in combination with recent historical connection and absence of evidence for adaptive divergence between populations, calls for the genetic management of the species as a single unit. The Iberian lynx *ex-situ* program is consequently managed as a single mixed population and already harbours several captive-born interpopulational litters. More recently, the first wild progeny of interpopulational crosses were born in Doñana in spring 2008 after the release of a single Sierra Morena male. Molecular markers are also being used to monitor the genetic composition of the *ex-situ* conservation program and will eventually be used to support its genetic management through the estimation of the coancestry between founders. Finally, the combined use of hypervariable molecular markers and non-invasively collected samples is being evaluated as an alternative/complementary approach to the monitoring of wild and reintroduced populations.

Introduction

In the last few decades, the prominent and important role that genetics can have for the conservation of endangered species has become apparent. On the one hand, extensive theoretical, experimental and empirical evidence supports the view that genetic factors might importantly contribute to extinction risks under scenarios that are typical of recently declining and fragmented populations (Frankham, 2005). Under these circumstances, genetic drift might become the predominant evolutionary force, leading to the loss of genetic diversity and the accumulation of inbreeding over time. These processes might in turn compromise population viability through the concomitant loss of adaptive potential and the negative impact of inbreeding on survival and reproduction (i.e.:

inbreeding depression) (O'Grady et al. 2006; Willi & Hoffmann, 2009). Going beyond early discussions on the relative importance of genetic vs. ecological in extinction, and once the interactions between these two are recognized, genetic factors are best viewed as key ingredients of the extinction vortex recipe, which thus needs to be evaluated for an integral approach to species conservation (Gilpin & Soulé, 1986). On the other hand, genetics can very significantly contribute to the conservation efforts to preserve endangered species, and must become part of a multidisciplinary approach to conservation. Firstly, a genetic analysis of endangered species and populations with molecular markers provides a description of patterns of genetic variation (diversity, inbreeding, structure, etc.),

which can be used to gain insights into the evolutionary and demographic processes that generated them (selection, genetic drift, bottlenecks, gene flow, etc.). Only with this information can conservation units be defined, short- and long-term viability evaluated and appropriate management strategies designed. Secondly, molecular techniques provide efficient tools for species, populations, gender and individual identification, which, especially when combined with non-invasively collected samples, can be effectively used for the monitoring of endangered and reintroduced populations.

The genetics of Iberian lynx wild populations

The Iberian lynx (*Lynx pardinus*) has suffered a steep decline and fragmentation during the last century which has resulted in its distribution being currently restricted to two extant populations: Doñana and Sierra Morena. Doñana is a peripheral population that became effectively isolated from the rest of the species distribution probably more than five decades ago, and has remained isolated and at a census size of around fifty since then. On the contrary, Sierra Morena population has remained connected to surrounding populations till recently, and has been continuously shrinking in size until recent active conservation measures have first stabilized and then slowly increased its census size from its low at 150. Under this demographic history, genetic drift may have resulted in a pattern of low genetic diversity and high inbreeding within, and high genetic differentiation between these two remnant populations, although the magnitude of these changes are difficult to predict without precise estimates of demographic parameters over time.

We have analysed contemporary genetic variation in Iberian lynx for mitochondrial and microsatellite markers. This study substantially extends our previous work, both in terms of sample sizes and number of markers (Johnson et al. 2004). As hypothesized, a high level of genetic differentiation is observed between Doñana and Sierra Morena populations. Mitochondrial diversity is extremely low with only two haplotypes observed which differ in one single position. These two haplotypes seem to be alternatively fixed in each population. Our previous study detected the presence of the Doñana haplotype in a museum specimen

from Sierra Morena, and the occurrence of a third now lost haplotype in Western Sierra Morena (Huelva province) (Johnson et al. 2004). These results with mitochondrial DNA provide direct evidence of genetic diversity losses in recent times due to drift in the small remnant populations and to the extinctions of previously differentiated populations.

Drift acts more intensely on the mitochondrial than in the nuclear genome due to its four times lower effective population size, an outcome of its haploidy and its uniparental transmission. To contrast the mtDNA patterns observed, we have also analysed nuclear diversity with a set of 36 microsatellite markers. Iberian lynx nuclear microsatellite diversity levels are globally low, both in terms of heterozygosity and allelic diversity. Iberian lynx has lower diversity than most felid species analysed to date, to the exception of some extreme examples of small isolated populations like the Gir forest lions and the Florida panthers (prior to the successful translocation of pumas from Texas). Furthermore, microsatellite heterozygosity and allelic diversity are 33% lower in Doñana than in Sierra Morena, what seems consistent with the smaller size and longer isolation of the former. At the same time, private alleles - i.e. "alleles that are found in one population but not in the other"- are up to four times more abundant in Sierra Morena than in Doñana, indicating that the allelic diversity found in Doñana may be mostly -but not totally- a subset of that found in Sierra Morena.

We are also analysing Iberian lynx genetic variation at class I and class II loci of the Major Histocompatibility Complex (MHC), a set of genes whose variation influences, among other things, the range of pathogens the immune system is able to respond to (Piertney & Oliver, 2006). Preliminary data show reduced diversity in Iberian lynx when compared to that reported for other species, indicating the loss of diversity could also be affecting adaptive variation at functionally important genes.

Effective populations sizes estimated from microsatellite data based on linkage disequilibrium are on the order of nine for Doñana and twelve for Sierra Morena. Diversity projections using simple genetic drift models and these estimated effective sizes predict a further reduction of heterozygosity to 50% of

current in only 12,5 (60 years, assuming a generation length of five years) and 16,6 generations (83 years) for Doñana and Sierra Morena, respectively. Furthermore, an effective size like that estimated for Doñana could retrospectively explain the loss of 33% heterozygosity from an ancestral diversity similar to that currently present in Sierra Morena in less than ten generations (50 years).

Implications for management

As expected from the known demographic history of the species, genetic patterns found in Iberian lynx population have probably been shaped by the predominant action of genetic drift in recent times, according to size and time since isolation in each population, and must not be interpreted as the consequence of a long history of independent evolution. The high levels of differentiation found between Doñana and Sierra Morena populations are most likely the result of random allele frequency fluctuations occurring in each population since their isolation from each other. Moreover, evidence for adaptive divergence between these two populations is lacking, and slight contemporary differences in morphometry may also be attributed to random genetic drift (Pertoldi et al. 2006). A similar argument can be made for contemporary differences in pelage pattern frequencies: the small-spot pattern that is currently predominant in Sierra Morena disappeared from the Doñana population a few decades ago (Beltrán & Delibes, 1993). Finally, these two remnant populations were probably interconnected by gene flow prior to their final isolation resulting from fragmentation of the species distribution by the mid-twentieth century, what would have hindered the development of strong adaptive divergences.

On the other hand, low current genetic diversity is probably the consequence of a steep and rapid loss in recent times, especially in Doñana where it might have reached over 33% in magnitude. Under this scenario, diversity losses may be equated to inbreeding accumulation, so that such a rate reflects inbreeding values that are often associated to a high frequency of genetic disorders, low fertility and other negative effects, as illustrated by the notorious case of the Florida panther (Roelke et al. 1993). No hard evidence of inbreeding depression in the Iberian lynx exists so far, but this possibility must be

seriously considered in view of the observed genetic patterns and known demographic history.

When we juxtapose the two aspects discussed above -i.e. "the lack of adaptive divergences and the accumulation of inbreeding"- it becomes clear that the risks of inbreeding depression largely outweighs risks of outbreeding depression in the Iberian lynx. The species *ex-situ* program is consequently managed as a single mixed population and already harbours several captive-born interpopulational litters. Moreover, the first wild progeny of interpopulational crosses was born in Doñana in spring 2008 after the release of a single Sierra Morena male, what constitutes the first successful example of translocation of Iberian lynxes.

The genetics of the captive population

A first objective of the *ex-situ* program is to capture the highest possible proportion of the diversity still present in the wild. To achieve this, a sufficient number of wild individuals must enter the captive population as founders. Simple drift models predict that a random selection of ten individuals will capture 95% of the existing diversity, and this proportion goes up to 98% when twenty individual are sampled. By mid 2008, the number of potential founders (individuals that have been captured but which have not necessarily reproduced) in the Iberian lynx *ex-situ* population was four from Doñana and 24 from Sierra Morena. These founders harbour 96% of Sierra Morena's heterozygosity, but only 77% of Doñana's, indicating the need to increase Doñana representation in the captive stock. Furthermore, in a structured species like the Iberian lynx, not only absolute numbers counts, but also the relative representation of each population becomes important. Optimal contribution of each population to the mixed captive stock was determined as 36% and 64% for Doñana and Sierra Morena, respectively, from our empirical estimates of each population's diversity and their degree of differentiation (Toro & Caballero, 2005). When these proportions are met the maximum potential heterozygosity of the *ex-situ* program could reach 54%. The current relative representation of the two Iberian lynx populations in the captive stock (14% from Doñana, 86% from Sierra Morena) is, however, still far from this optimum. The expected heterozygosity for the current (mid

2008) set of founders is 50%, a little below its theoretical maximum of 54%, due both to an insufficient representation of Doñana genetic diversity and a biased composition towards Sierra Morena.

The second goal of the *ex-situ* program will be to preserve as much heterozygosity as possible over time. The genetic goals of the program were initially established at preserving 85% of the initial heterozygosity in 30 years (Lacy & Vargas, 2004). Although a rather modest goal when compared to the more usual target of 95% in 100 years, this target is considering both the minimization of the impact to the wild populations and the short-term aspiration of the program. Retaining the maximum heterozygosity through time will require some genetic management that is better postponed until the carrying capacity of the population is reached, in order to favour rapid growth at the initial phases. Genetic management usually invokes the "minimum kinship" strategy, by which priority in reproduction is given in each breeding season to individuals showing the minimum average kinship to the rest of the population. Simulation studies suggest that this strategy is effective at preserving both heterozygosity and allelic diversity and at minimizing inbreeding (Fernández & Caballero, 2001; Fernández et al. 2003; Fernández et al. 2004). For the implementation of this strategy, kinship is usually estimated from the captive pedigree assuming totally unrelated founders. Under most realistic scenarios, certainly including the Iberian lynx, this assumption is not met, due to population structure and autocorrelated captures. A simple approach to this strategy is to favor matings between individuals from different populations, which would be minimally related. Molecular markers can assist the genetic management of the captive population by providing estimates of founder relatedness to be incorporated in the implementation of a minimum kinship strategy (Russello & Amato, 2004).

Prospects for Iberian lynx management

The complexity of the Iberian lynx management will soon increase with the incorporation of new captive breeding centres and with the establishment of reintroduced populations. These multiple captive centres need to be co-ordinately managed to maximize global diversity and to minimize local inbreeding. Tools have been recently

developed to achieve this while maintaining a decided level of genetic structure and limiting the number of migrants as desired (Fernández et al. 2008). The genetic issues discussed for the captive population applies as well to the reintroduced populations: diversity should be maximized and inbreeding minimized. In this case, however, only the number and origin of the reintroduced individuals can be adjusted. In the early phases of reintroduction, captive born individuals with low inbreeding, low value for the captive stock and low relatedness with previously released individuals should be selected. Reintroduced populations will need to be closely monitored demographically and genetically, and the genetic characterization of non-invasive samples (i.e., genetic individualization of faeces) could be a useful tool for both purposes (Schwartz et al. 2007). Eventually, captive, wild and reintroduced subpopulations could be managed co-ordinately as a global metapopulation through captures, reinforcements, translocations and reintroductions. Management will become less relevant as the final goal of establishing a viable and genetically healthy metapopulation of Iberian lynxes is approached.

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Selection process for Iberian lynx reintroduction areas in Andalusia

José M. Gil-Sánchez^{1*}, José García^{1‡}, Rafael Arenas², Javier Rodríguez¹, Manuel Moral², Joaquín Pérez¹, José M. Martín¹, Eva Rojas², Maribel García-Tardío¹ and Miguel A. Simón³.

1. Iberian lynx LIFE conservation project. EGMASA. Avd. Andalucía, 104 Esc 3, 1º. 23006. Jaén.

* jmgilsanchez@yahoo.es; ‡ tenerife3@vitaldent.com

2. Iberian lynx LIFE conservation project. EGMASA.
c/ Pepe Espaliú, 2. 14004. Córdoba.

3. Iberian lynx LIFE conservation project. Consejería de Medio Ambiente, Junta de Andalucía.
c/ Fuente del Serbo, 3. 23071. Jaén.

Abstract

To carry the first Iberian lynx reintroduction out, we began with desk works in early 2005. GIS models were performed in order to identify the most suitable areas for the reintroduction of the species in Andalusia. Following a multicriteria analysis, three potential areas for the reintroduction of the Iberian lynx were pre-selected based on habitat features, antropic pressure, protection degree and historic distribution range. These areas were: river Guadalquivir valley, river Guadarrama valley and Hornachuelos Natural Park, all three areas located in Sierra Morena. Afterwards, we made a detailed evaluation and comparative analysis of the three areas through a preliminary study on the causes of extinction and their current impact. Causes of extinction were described through 21 key environmental variables. Non-natural mortality was probably the main cause of extinction in all three areas, especially due to the massive use of legholds for rabbits. Nowadays, this threat has disappeared. Hornachuelos was discarded due to both its low potential for its integration in the Iberian lynx meta-population in the short term, and its smaller carrying capacity. Conditions in Guadalquivir and Guadarrama remain well balanced, although the first zone showed a better habitat structure. The environmental situation in eastern Sierra Morena shows a remarkable carrying capacity, allowing an estimated meta-population of about 140 Iberian lynx territories.

Introduction

The election of a suitable place where perform the reintroduction of a species is thought to be essential to its successful outgoing. A thorough process to find out the best potential areas for an Iberian lynx reintroduction in Andalusia has been performed within the framework of the two last Iberian lynx conservation LIFE-Nature projects (02/E/8609 and 06/E/209). This selection was made following the IUCN reintroduction guidelines (IUCN, 1998). First works on this matter began in 2005 by means of the performance of a GIS model that gave an aprioristic approximation which allowed pre-selecting the potential best areas for reintroducing the Iberian lynx in Andalusia. Between September 2006 and December 2007, the necessary information for a fine-scale selection of the best area out of those pre-selected was completed. Habitat quality and availability were evaluated relying on data of the maximum possible quality, through the

study of the key factors for survival and reproduction of released individuals (Griffith et al. 1989; Wolf et al. 1996, 1998; Saavedra & Sargatal 1998). These parameters are known to determine the success of a program with the establishment of a self-sustaining population of the species (Rodríguez et al. 2003). The objectives of the evaluation process and final selection were: (1) To study causes of extinction (Short et al. 1992; IUCN, 1998), (2) To quantify and evaluate the key elements of the existing habitat, not only biological but also the social attitude of the human environment due to its significant impact on reintroduction programs (Breitenmoser, 1998; Byrne & Sheik 1999; Breitenmoser et al. 2001), (3) To prepare an analysis of carrying capacity, and (4) To compare the set of parameters generated in the three pre-selected areas in order to choose those showing the best conditions.

Methods

Preliminary gross selection

The preliminary gross selection of potential Iberian lynx reintroduction areas in Andalusia was performed through a multicriteria analysis (MCA) (see García et al. 2004). The MCA is a decision-making tool developed for complex problems in which multiple criteria are involved in order to avoid confusion, since with this method members of a team do not have to agree on the relative importance of the Criteria or the rankings of the alternatives. After performing the MCA, the model was displayed with a Geographic Information System (GIS) using Biomapper software, which gave a predictive map of the areas of maximum suitability for the Iberian lynx occurrence in Andalusia. The conditioning variables taken into account were divided into (1) macro-habitat and anthropic impact variables (which led to the construction of a suitability-impact model), (2) surface, (3) historical distribution, (4) protection degree of the area and (5) trophic determinants.

Suitability-impact model: Macro-habitat and anthropic impact variables were analysed by means of a suitability-impact model, which resulted in a predictive map of optimal habitats for the occurrence of the Iberian lynx. The identification of the factors determining the potential of a place for hosting Iberian lynxes was based both on bibliography and on information of the current Iberian lynx population. Six factors were identified as determinant for the presence of the species: (1) altitude, (2) sloping, (3) vegetation structure, (4) abundance of water sources, (5) abundance of human population nuclei and (6) presence of anthropic communication structures. Firstly, the maximum altitude recorded in the Iberian lynx distribution of the last 30 years (Rodríguez & Delibes, 1990) was 1300 masl, which was included in the model as a restrictive value. Similarly, given that 80% of the current Iberian lynx population occurs in areas with less than 30° sloping, this value was included as a restriction in the model. To define the importance of the vegetation structure over the presence of the Iberian lynx, a model of habitat selection was previously performed. This model explored macro-habitat preferences in relation to availability in the whole range of the current Iberian lynx distribution. In agreement with previous data (Palomares et al. 1999) we found a positive selection of dense

scrubland areas, which were considered a restrictive factor within the model. Palomares et al. (1999) found that Iberian lynx territories present a significantly higher percentage of water sources than those non-occupied ones. Data of the current Iberian lynx population also show a decreasing rate of lynx occurrence when getting further from water resources. Thus, distance to water sources was coded in the model. Lastly, we found that Iberian lynx occurrence in current populations is inversely related to the proximity with both municipal townships and great communication structures (main roads and highways). Given this, both distance to municipal townships and distance to principal roads were coded in the model. Once determined, the specific weight of every factor was calculated through an analytical hierarchies method in a pair comparison procedure using IDRISI software (Bosque et al. 1994; Saaty, 2008). A poll was performed to several experts in the Iberian lynx (belonging to the department of applied biology of the Doñana Biological Station and to the Iberian lynx conservation LIFE project) to define the comparison matrix of all groups of variables. Consistency of all assigned weights was evaluated by a consistence ratio. Afterwards, a Weighted Linear Combination analysis was performed (Barredo, 1996). With this method, the suitability of a pixel is obtained as the sum of partial suitability values of every factor in that area. Partial suitability values are expressed as the result of the multiplication of each value by its weight in a concrete area. Cartography used in the study was composed of 50 m side pixels. Finally, four approaches were made to carry the multicriteria analysis out, which allowed identifying those areas selected by all four approaches and so, doing a minimum error risk selection. The different approaches were:

a) Equal weights: Facing the possibility that weights could sub- or over-estimate some of the factors, the analysis was performed giving identical weight to every considered factor.

b) Different weights: Three different models were performed: (1) weight pattern generated by the poll data of the Doñana Biological Station staff, (2) weight pattern generated by the poll data of the Iberian lynx conservation LIFE project staff and (3) weight pattern generated by both.

Weight assigned to every factor was calculated using the WEIGHT module of the IDRISI program (Eastman, 1997). Once the first evaluation of data consistence was done, weights were re-calculated to reach acceptable values for every factor. It consisted mainly in sub-consider extreme values of the polls. All information layers were created using ARCVIEW 3.1 (ESRI, 1996). Afterwards, all layers were integrated using ARC-GIS (ESRI, 1996) and four different model were created. Since each model represented different quantitative values, "suitable areas for the lynx" were considered those with a value equal to the mean plus once the standard deviation of the values of each model. Models were evaluated checking the percentage of lynx locations correctly classified. All models were acceptable, since they correctly classify 75% to 85% of lynx locations. A convergence model by Boolean intersection of the four models was made, and only those areas which were suitable for the lynx in all four models were further considered in the convergence model.

Variable	Selection criterion
Habitat & antropic impact	Suitability-impact model
Surface	Larger than 2000 Ha
Historical distribution	Distribution in 1990
Protection degree	Inside SIC limits
Trophic determinants	High rabbit density

Table 1. Variables determining the occurrence of the Iberian lynx in an area and their limits to be considered in the preliminary gross selection of the potential Iberian lynx reintroduction areas in Andalusia.

Final model: Once the converged suitability-impact model pointed out the aprioristic best areas for hosting Iberian lynxes in Andalusia, the rest of restrictions were applied to obtain the final model of the preliminary gross selection of potential Iberian lynx reintroduction areas in Andalusia. Based on bibliography on Iberian lynx ecology and taking IUCN reintroduction guidelines (IUCN, 1998) into account, we fixed several restrictions to the model attending the rest of variables (see table 1). The final map showed southern Hornachuelos Natural Park, Guadalmellato river valley and Guarrizas river valley as the best

aprioristic areas to perform the Iberian lynx reintroduction into (figure 1).

Description of the pre-selected areas

The three pre-selected areas belong to the Sierra Morena large eco-regional unit, specifically to the eastern half right of the border with the Countryside in the River Guadalquivir (Figs. 1 and 2). Their surfaces are: 17,578 ha in Guadalmellato, 14,226 ha and 13,436 ha in Guarrizas Hornachuelos. The average annual temperature varies between 16 and 18 ° C and average annual rainfall between 500 and 700 mm. The altitude ranges between 400 and 800 m. Local rivers are typically Mediterranean regime, with a strong dry season that interrupt the flow, but there remains enough water so it seems not being a limiting factor for wildlife (Ruiz-Olmo, 1998; Doadrio, 2001;). Vegetation of all three areas consists of sclerophyllous meso-Mediterranean scrubs. Human population density is low, and it is basically limited to game wardens and farming employees. Because the land is mostly private, access is fairly limited. Main uses are hunting (red deers and wild boars) and shooting (rabbits and red partridges).

Fine-scale selection

In order to determine (1) the causes that provoked the extinction of the Iberian lynx in each area, (2) the long-term viability of a reintroduced Iberian lynx population in each area and (3) the integration possibilities of each reintroduced population into the current existing one, we defined the following two groups of variables:

1. Variables related to the causes of extinction:

Knowing the causes that led the species to the extinction in each area was considered crucial because if these causes continue to act and are not mitigated the project is doomed to failure from its inception (IUCN, 1998). Three key factors have been cited as responsible for the regressive dynamics suffered throughout the twentieth century: (1) rabbit scarcity, (2) habitat destruction and (3) human persecution. However, the relative impact of each factor has not been studied in detail (Rodríguez & Delibes 2002, 2003, 2004). In the three pre-selected areas, the first two factors cannot be considered as directors of Iberian lynx local extinction, since both have maintained optimal conditions during the last decades. Thus, we only took human persecution into account

(Valverde, 1963; García-Perea & Gisbert 1986; Ferreras et al. 2001, Rodríguez & Delibes, 2004). To this end, we collected both unpubl. (oral communications and official reports) and published information (ICONA, 1973; Rodríguez & Delibes 1990) about lynx mortality and the responsible causes, some of which could be quantified indirectly. Legholds used for killing rabbits could be quantified through the amount of extracted rabbits (Rodríguez & Delibes 1990). The final variables included in the model were (1) number of lynxes killed by direct persecution during the Twentieth Century (We used the value as a whole and also for each one of these causes of death distinguishing four categories: legholds, snares, shots and others) and (2) number of rabbits hunted with legholds (standardized specimens / ha and year).

2. Variables related to current limiting factors: These variables were classified into four different groups:

2.A. Potential causes of mortality: To evaluate this factor, several variables were recorded: (1) **Official proceedings against the owner because of illegal hunting methods** per year and area. For associated statistical analysis, it was assumed that the effort had been uniform for each zone. The period considered was from 1997 to 2006. (2) **Number of found illegal hunting methods per effort unit.** Refers to the illegal gear (stocks and bonds) located during field sampling by the staff of Life program between 2006 and 2007. Each journey was regarded as a sampling unit, which corresponds to two hours of search by a person on foot. Each set of traps found was considered a separate event. (3) **Total number of carnivores killed in roads and railways.** We evaluated the roads and railway lines within a 5 km radius. We considered as priority routes those located within the optimal habitat (Palomares et al. 2000). In order to both obtain a quantitative index of risk and identify high-risk tranches for the Iberian lynx, carnivores were sampled due to their eto-ecological similarities with the lynx. In the case of roads, selected sections were sampled using weekly transects by car, at half the maximum speed allowed on the road. We also walked along the railways in order to detect high-risk stretches. (4) **Number of carnivores killed per effort unit.** Each tour vehicle was considered a sampling unit. (5) **Identified high-risk**

stretches. Consideration was given to those sections of the communication channels that were found particularly dangerous because they crossed areas of good habitat, high rabbit abundance and killed carnivores. Resulting data of variables 1, 2 and 5 were compared with reference values in current Sierra Morena Iberian lynx population.

2.B. Habitat structure: Landscape structure of all three areas has not undergone significant changes over the past 20 years, thus conditions existing prior to the extinction of the lynx persist. This could be *a priori* considered as optimal (Palomares, 1999; García et al. 2004; Breitenmoser et al. 2006). Despite this, several structural variables (at least locally important for the species), such as protection and refuge (Palomares, 2001) were defined through a stratified sampling based on a transect per 2.5 x 2.5 km grid. We scored coverage and height of the three structural vegetation types in a 25 m diameter circle in a total of nine equidistant points per transect (with a separation of 500 m.). These variables were quantified by visual estimations (Palomares, 2001). We used both data from Doñana population (Palomares, 2001) and from a lynx high-density area of the Jándula river valley (see table 2) were used as reference values. Vegetation in Hornachuelos was not assessed because this area was ruled out early in the study (see results). (1) **Percentage of tree cover.** Dominant species: *Q. ilex*, *Q. suber*, *Q. faginea*, *O. europaea* var. *silvestris*, *P. pinaster*, *P. pinea*. (2) **Percentage of coverage of shrub scrub.** Dominant

VARIABLE	DOÑANA		ANDUJAR	
	MEAN	CI95%	MEAN	CI95%
% Tree coverage	14.2	± 4.45	5.9	± 3.68
% shrub scrub coverage	20.4	± 3.52	8.8	± 6.29
% serial scrub coverage	34.4	± 9.48	18.8	± 1.00
Mean tree height (m.)	6.8	± 0.88	5.1	± 0.92
Mean shrub scrub height (cm.)	250.0	± 140.0	134.0	± 21.3
Mean serial scrub height (cm.)	100.0	± 60.0	57.3	± 14.4

Table 2. Reference values of the vegetal structure occurring in areas where Iberian lynx inhabits: Doñana (Palomares et al. 2000) and Sierra Morena (LIFE project unpubl. data).

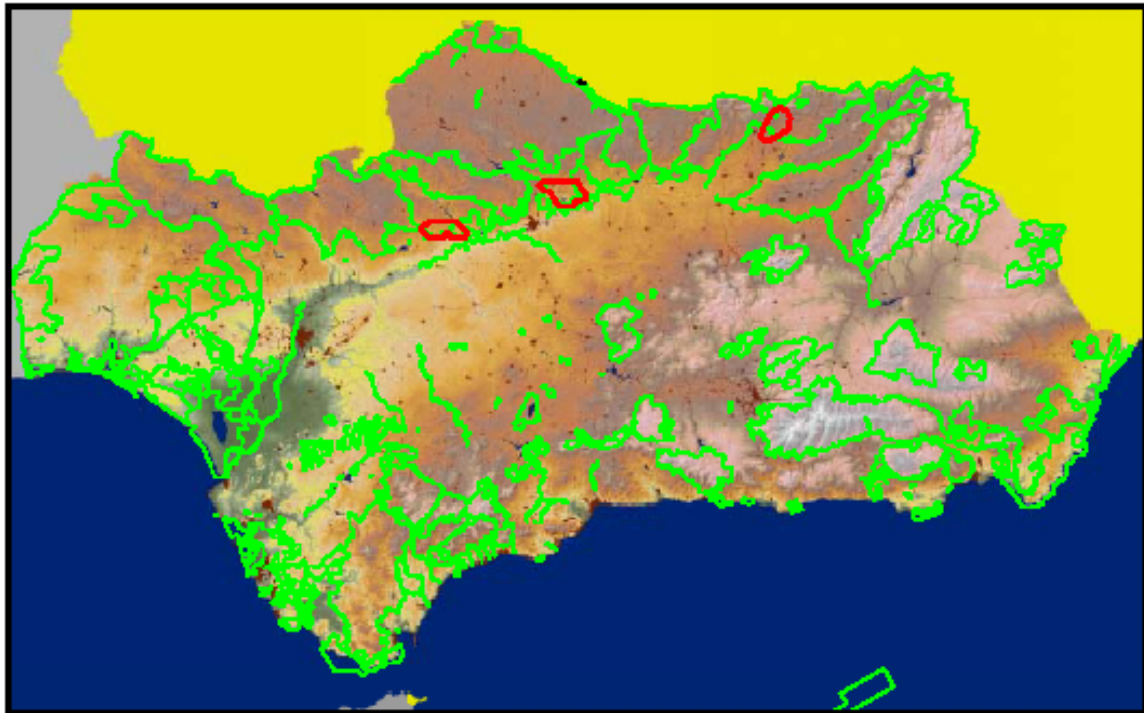


Figure 1. SIC areas in Andalusia (in green) in relation to the three pre-selected Iberian lynx reintroduction areas.

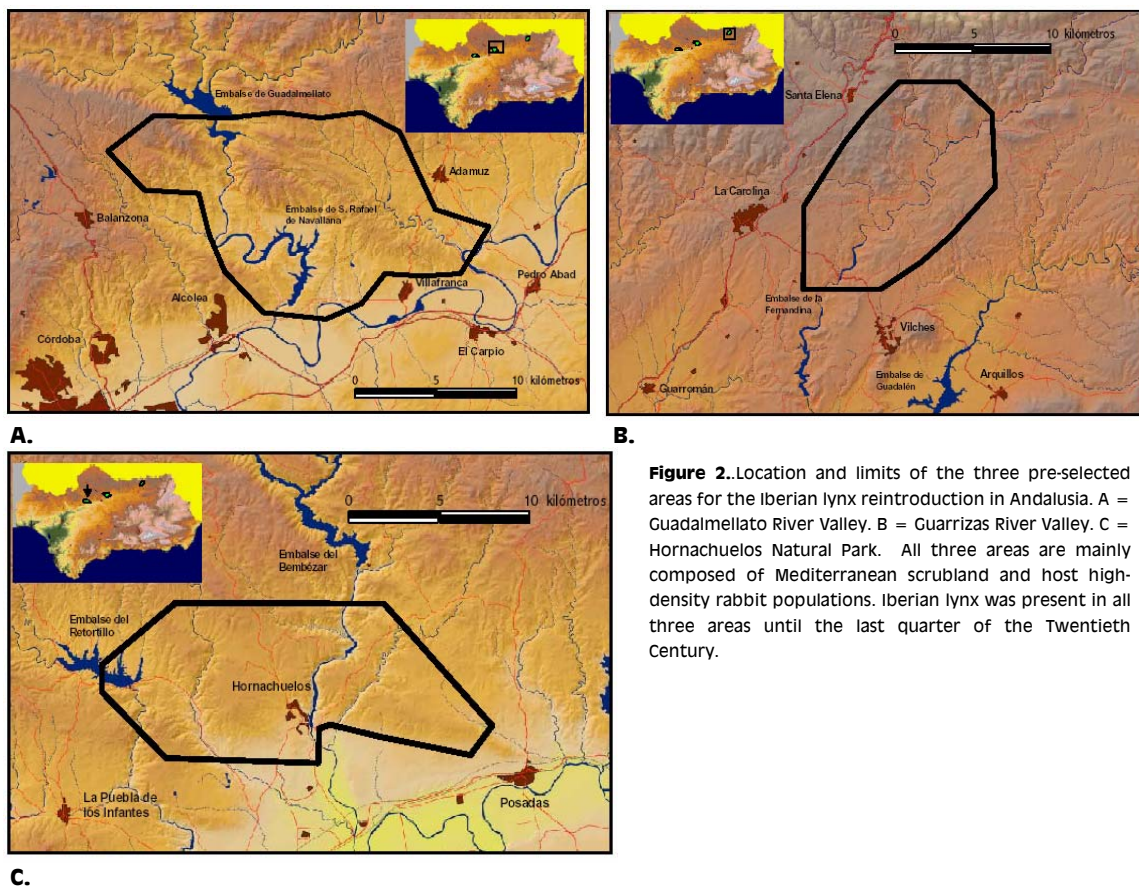


Figure 2. Location and limits of the three pre-selected areas for the Iberian lynx reintroduction in Andalusia. A = Guadalmellato River Valley. B = Guarrizas River Valley. C = Hornachuelos Natural Park. All three areas are mainly composed of Mediterranean scrubland and host high-density rabbit populations. Iberian lynx was present in all three areas until the last quarter of the Twentieth Century.

species: *P. lentiscus*, *Phyllirea angustifolia*, *Q. coccifera*, *A. unedo*. (3) **Percentage of coverage of serial scrub.** Dominant species: *C. ladanifer*, *C. albidus*, *C. monpeliensis*, *R. offincinalis*, *Ulex* sp. (4) **Average height trees.** (5) **Average height shrub scrub.** (6) **Average height serial scrub.** (7) **Availability of micro-habitat suitable for reproduction.** Breeding habitat may be a first-order limiting factor for species with high dependence on a particular and scarce type (Newton, 1979). The Iberian lynx is known to be able to use both small cavities (in rocky grounds, in big trees and in piles of tree stumps derived from selviculture) and dense bushes as dens (Valverde, 1963, Palomares & Fernández 2000, LIFE project unpubl. data). It was assumed that this factor is not a limiting parameter, since all three areas hosted lynx populations until recent years.

2.C. Resources availability: Similarly to the case of the Canadian lynx (*Lynx canadensis*) and the mountain hare (*Lepus timidus*) (Ruggiero et al. 1999) the Iberian lynx is totally dependent from the wild rabbit (Gil-Sánchez et al. 2006). A study on the feasibility of the Canadian lynx reintroduction programs revealed that the main director variable for success was the abundance of mountain hares (Stuery & Murray, 2004). High availability of hares could compensate even moderate levels of non-natural mortality, through compensatory mechanisms related to the improvement of the breeding success (Stuery & Murray, 2004). A preliminary rabbit abundance survey was performed as part of the abovementioned preliminary gross selection of the best areas for reintroduction. That information was updated in 2007 through applying the standard methodology used by the monitoring team of the LIFE project (LIFE project unpubl. data): rabbit latrine counts and kilometric abundance indexes (KAIs) in a basis of a 2.5 km grid. The relative results in rabbit latrines per Km were converted into absolute rabbit abundance using a formula derived from the Andújar-Cardena population ($\text{rabbits / ha} = 0.1163 \times \text{latrines / km} - 0.2816$, $R = 0.95$, $P < 0.001$). Sampling was conducted during the summer, so data correspond to the period of maximum rabbit abundance. These works were not carried out in Hornachuelos since the region was ruled out early in the study (see results). The variables obtained were: (1) **mean rabbit abundance**, (2) **seasonal abundance of rabbits (KAI)** and (3) **number of 2.5 km UTM**

squares exceeding the threshold of rabbit abundance that allows the establishment of territorial lynxes (1 rabbit / ha). This value was stated in the Andújar population with the same assessment methodology (LIFE project unpubl. data). The results were compared with data from Andújar-Cardena as a benchmark.. These data allowed performing a map of both distribution and abundance of rabbits in the two prospected areas.

2.D. Possibilities of connection with other populations and/or of expansion to other potential areas: The connectivity between potentially suitable areas has been shown to be a highly limiting factor for Eurasian lynx (*Lynx lynx*) reintroduction programs (Kramer-Schadt et al. 2004). Breitenmoser et al. (2004) highlighted the importance of creating a strong metapopulation within Sierra Morena through reintroduction programs. Moreover, the selection process is essential to study both the potential exchange of individuals between populations and the possibilities of expanding into other patches of suitable habitat. The connection between populations or recolonization of other areas would depend on the availability of dispersant individuals and their possibilities of movement between different areas (Ferrerías, 2001). The variables related to the possibilities of inter-population connection were: (1) **Distance to the nearest Iberian lynx population through suitable habitats for dispersal** (that is *effective distance*; Ferrerías, 2001). The nearest population is the Andújar-Cardena's in all cases. Mediterranean oak and scrub formations, riparian forests and forestall plantations of pines and eucalyptus were considered as optimal habitats (Palomares et al. 2000; Palomares, 2001). (2) **Number and surface of nuclei with high-density of rabbits that are potentially reachable by dispersants.** Data come from own sources for Andalusia's part and from Guzmán et al. (2004) for Castilla-La Mancha's part of the surroundings of the reintroduction area. Information generated in the Iberian lynx Doñana population was used as reference value to evaluate the possibility of contact between the three pre-selected areas, the current population and the potentially good-conditions reachable nuclei. Thus, it was assumed that the contact through dispersing individuals was possible when the effective distance between areas was no longer than 42 km by permeable habitat (Ferrerías et al. 2004).

2.E. Social attitude: The support of the local human population has been described as one of the major factors limiting reintroduction projects success (see review by Rodríguez et al. 2003). The Institute for Advanced Social Studies of Andalusia (IESA/CSIC) conducted a sociological study to assess the attitude of the population towards the Iberian lynx reintroduction in the three pre-selected areas (explained in detail in this seminar). The variables taken into account in our analysis were (1) **Percentage of local population in agreement with the reintroduction in their area**, (2) **percentage of local hunters in agreement with the reintroduction in their area**. Data generated by this poll were used for comparison among the three pre-selected areas.

After its measurement, each variable was classified into one of these three categories of suitability for the species: (1) **suitable**, if the value of the variable was equal to or better than the reference ones, (2) **unsuitable**, if the value was worse than the reference and not subjected to improvement and (3) **conditioned suitability**, if the value was worse than the reference ones but could be improved through the management by this project. Given that the sample sizes were reduced in some variables, the non parametric Mann Whitney U-test was used to perform the mean contrast. In the case of percentages, the G-statistic applied on contingency tables with continuity correction was selected.

We used the total number of territorial females an area can host as the carrying capacity of a determined place. The number of potentially breeding female territories is a good indicator of the survival of populations of medium-size and big felines, such as the Iberian lynx (Kenney et al. 1995, Gaona et al. 1998; Palomares et al. 2001; Ferreras et al. 2002; Fernández et al. 2006). Carrying capacity has been established based on rabbit availability once suitability of habitat had been previously verified. Rabbit availability was expressed as 2.5 Km UTM squares above the threshold of Iberian lynx occurrence (1 rabbit / ha). To estimate number of territories an area can host, we used the average home range surface of resident females in Doñana population (4.1 km², 95% CI = 2.6 - 5.8, n = 14; Fernandez et al. 2006). Total number of lynx territories an area can hold was calculated using the average number of lynx / area

observed in Sierra Morena population between 2003 and 2006 (mean = 4.78, 95% CI = 3.76 - 5.76; LIFE project, unpubl. data) The carrying capacity was considered as suitable when the lower estimated limit was not smaller than the minimum number of adult female territories registered in the two currently existing populations (10 territories, registered in Doñana population in 2006) (LIFE project, unpubl. data).

In order to make the final selection, we performed a process that followed ordered steps. The first step was the identification of non-suitable variables. In case these variables were limiting and non-improvable in the context of the present LIFE project, the area was automatically dismissed. The second step was a comparative analysis which considered the areas which passed the first filter. The aim of this was to select the best area, and two approaches were followed:

1. Hierarchized comparison. An order of hierarchized comparison was stated according to the relative importance of seven parameters summarized by the 20 variables previously described: (1) local social attitude, (2) possibility of integration in the current metapopulation, (3) Carrying capacity, (4) habitat structure, (5) possibilities for expansion to other potential areas, (6) amplitude of illegal hunting methods for carnivores and (7) risk of being knocked down. It is assumed that each parameter limits those placed in a lower position. Thus, it suffices to keep a better position in the first parameter to be classified as better. While being equal, comparison is made through the following parameter and so on.

2. Equitable comparison. It assumes that each one of the seven parameters has the same limiting relative importance. Thus, the comparison is made through the percentage of occurrences of each area in the best position for each parameter.

Results

1. *Variables related to causes of extinction*: We could compile information about 3, 6 and 2 Iberian lynxes extracted from the field in Guadalmellato, Guarrizas and Hornachuelos, respectively, during the last quarter of the last century. The most common cause of extraction was legholds for rabbits, which were well recognized as responsible for the death of

many more unrecorded lynxes in Guadalmellato area during that period. Moreover, Rodríguez and Delibes (1990) recorded a larger number of dead lynxes during the same period in all three areas. Their data point out legholds as responsible for about 50% of the deaths, whereas direct shooting was responsible for about a 25%. Thus, all available data suggest that the extinction of all three populations was caused by non-natural deaths, being legholds for rabbits the main cause of death. We could not obtain quantitative information on the number of rabbits taken in legholds in Guadalmellato and Guarrizas, although some land owners and guards in Guadalmellato revealed a number of 200 legholds per land during 2 months. In Guarrizas, we found quantitative information on the number of rabbits taken in legholds in the town of Vilches, where, in 1988, 8.18 individuals per hectare were captured (Rodríguez & Delibes 1990).

2. Variables related to current limiting factors: Table 3 provides an integrated view of both the values of the 21 variables describing the quality of the evaluated areas and those of current lynx populations. Most of the variables did not differ from the reference values. In Guadalmellato, rate of official proceedings against lands was statistically higher ($U = 7.5$, $Z = -3.2$, $p = 0.0013$) than that found in the Andújar-Cardena lynx population during the same period (Table 1). Shrub scrub cover ($G = 5.3$, $p = 0.02$, $df = 1$) and serial scrub cover ($G = 0.008$, $p = 0.92$, $df = 1$) were found to be significantly higher in Guadalmellato than in Doñana, although these differences have a positive contribution to the species. In Guarrizas, serial scrub coverage is significantly lower than that presented in Andújar ($G = 3.89$, $p = 0.07$, $df = 1$). Finally, separation between Hornachuelos and the nearest population is over the known dispersive potential of the species.

Final selection

Despite its difference with the reference value, the rate of official proceedings against lands in Guadalmellato can probably be considered as "very low". In fact, specific field sampling gave data which were very similar to those found in the reference population. In conclusion, the potential mortality by direct persecution can probably be regarded as compatible with the presence of a stable population of the species, and therefore it can be currently assessed as a

suitable parameter. Anyway, it is a parameter we can manage in the context of this reintroduction project. The coverage of serial scrub has a rather indirect contribution to the species, since it improves the availability of refuge for the rabbits (Palomares et al. 2001). In this way, the significant difference found in Guarrizas regarding serial scrub cover would be minor because of the abundance of these lagomorphs, so that this variable was considered as suitable. So the only area holding a totally unsuitable variable is Hornachuelos, (the potential for metapopulation integration) (variable ref. 15, Table 3). Therefore, Hornachuelos was excluded from the selection process. Furthermore, this key limitation is compounded by the fact that the carrying capacity in this area is significantly lower than that recorded for the other two pre-selected areas (Table 2).

Comparative analysis between both suitable areas: No significant difference were found between Guarrizas and Guadalmellato regarding the social attitude of the local population (both showed a high value of about 90%) ($G = 0.00503$, ns, $df = 1$). Attitude of hunters was worse in Guarrizas than in Guadalmellato ($G = 15.02$, $p = 0.0015$, $Df = 1$). However, the net difference is not too large (13.3 percentage points), since at least three quarters of the hunters supported the lynx reintroduction in both areas. Guadalmellato showed slightly better connectivity with other population nuclei than Guarrizas (table 2, fig. 3). However, the connection probability between a source and a receiving area also depends on the size of the first one, as this will determine the amount of dispersing individuals (Ferrerías, 2001). Guarrizas is located closer to the sub-population of the Jándula valley (Fig. 5), which is significantly higher than the Yeguas subpopulation, which is the nearest one to Guadalmellato. Givdedn this, both areas were considered equally suitable. Carrying capacity showed tyo be slightly higher in Guadalmellato than in Guarrizas (26 and 23 territories, respectively). AS the difference was small, equal suitability was assumed in both populations. In fact, mean rabbit abundance in time of maximum did not show significant differences between both areas ($U = 126.5$, $Z = -0.78$, $p = 0.43$). Guadalmellato showed better scrub coverage than Guarrizas, what is thought to determine a higher quality habitat in terms of refuge availability. On the other hand, Guarrizas

VARIABLE	Guadalme.	Guarrizas	Hornach.	Reference
1.- Number of proceedings against lands / year and Km2.	0.18 ± 0.04	0.096 ± 0.10	0.083 ± 0.06	0.03 ± 0.03
2.- Number of located illegal hunting methods / sampling unit	0 ± 0.00	0 ± 0.00	0 ± 0.00	0.019 ± 0.01
3.- Number of carnivores killed in roads	17	7	-	-
4.- N carnivores killed in roads / 10 Km. And sampling	0.14 ± 0.11	2.37 ± 2.92	-	-
5.- Km of high-risk stretches	1.6	13	-	55.6
6.- % tree coverage	30.1 ± 3.9	26.4 ± 5.5	-	5.9 ± 3.6
7.- % shrub scrub coverage	34.9 ± 4.8	17.2 ± 7.0	-	8.8 ± 6.2
8.- % serial scrub coverage	33.8 ± 5.4	9.2 ± 4.0	-	18.8 ± 1.0
9.- Mean tree height (m.)	5.6 ± 0.3	4.8 ± 1.1	-	5.1 ± 0.9
10.- Mean shrub scrub height (cm.)	208.2 ± 19.4	87.5 ± 20.4	-	134.0 ± 21.3
11.- Mean serial scrub height (cm.)	62.8 ± 5.2	44.6 ± 19.0	-	57.3 ± 14.4
12.- Rabbit abundance in spring (indiv./ha)	3.1 ± 0.80	3.6 ± 0.8	1.8 ± 0.8	1.6 ± 0.4
13.- Rabbit abundance in fall (KAI: indiv./Km.)	(0.75 - 3.4)	(1.1 - 1.3)	-	(0.25 - 1.18)*
14.- N 2.5 Km squares with > 1 rabbit/ ha spring	18	20	8	(16 – 27)*
15.- Distance to the nearest lynx population (Km)	33.1	45.7	90	42
16.- N of high-rabbit density nuclei reachable by dispersants	2	1	2	-
17.- Surface of high-rabbit density nuclei reachable by dispersants (Km.²)	167	460	156	-
18.- % local population in agreement with reintroduction	91.2	91.5	85.8	-
19.- % local hunters in agreement with reintroduction	89.8	76.5	88.6	-
20.- Carrying capacity (territories)	26 ± 11.5	23 ± 10	12 ± 5	10**
21.- Carrying capacity (individuals)	124 ± 24	110 ± 23	57 ± 12	35**

Table 3. Values of the different variables studied both in the potential reintroduction areas and in the reference populations (data from Andújar-Cardena, except those indicated in italics, which are Doñana's). Blue data indicates statistically differences with reference values. Variable values are shown as mean and confidence interval of 95%, unless further another descriptive measure is indicated.

was found to have better conditions for natural expansion because there is a vast area of high rabbit density and good habitat structure 7 km northern the pre-selected area (Fig. 5). Areas with high rabbit density close to Guadalmellato are much smaller than that in close to Guarrizas (Fig. 3).

Guadalmellato showed a rate of official proceedings against lands very close to the critical level of statistical significance ($U = 25.5$, $Z = -1.85$, $p = 0.053$). The rate of found illegal hunting methods in the field, however, was well balanced. Guarrizas recorded the highest rate of animals knocked down in roads ($U = 371.0$, $Z = -2.38$, $p = 0.016$), primarily due to the contribution of the railway. It also showed a greater amount of high-risk stretches in the roads. This study has assumed a negligible impact of the high speed railway crossing Guadalmellato area, since it is fenced off. Anyway, this assumption could be incorrect.

According to the hierarchized comparison, Guadalmellato has priority over Guarrizas due to the better vegetation structure of the first one. According to the equitable comparison

both areas get the same value selection (Tables 3 and 4).

Discussion

A detailed establishment of the causes that drove Iberian lynx populations to their local extinction in each of the three pre-selected areas is very difficult to achieve. The lack of quality information forced us to perform a retrospective approximation based on speculation. Available data clearly point the anthropogenic sources of mortality out as the cause of the Iberian lynx extinction in all three areas. Thus, the effect of non-compensated mortality must be inferred in an indirect way. The Iberian lynx is currently strictly protected by law and non-selective hunting methods are completely illegal in Andalusia. Given this, the scenario has become particularly favourable for the species, although collected data show that there is still some residual activity of the use of illegal hunting methods. Anyway, reference data show that the survival of a lynx population is compatible with these levels of trapping activity. Nevertheless, there is no doubt that the ultimate goal should be the eradication of those illegal hunting methods.

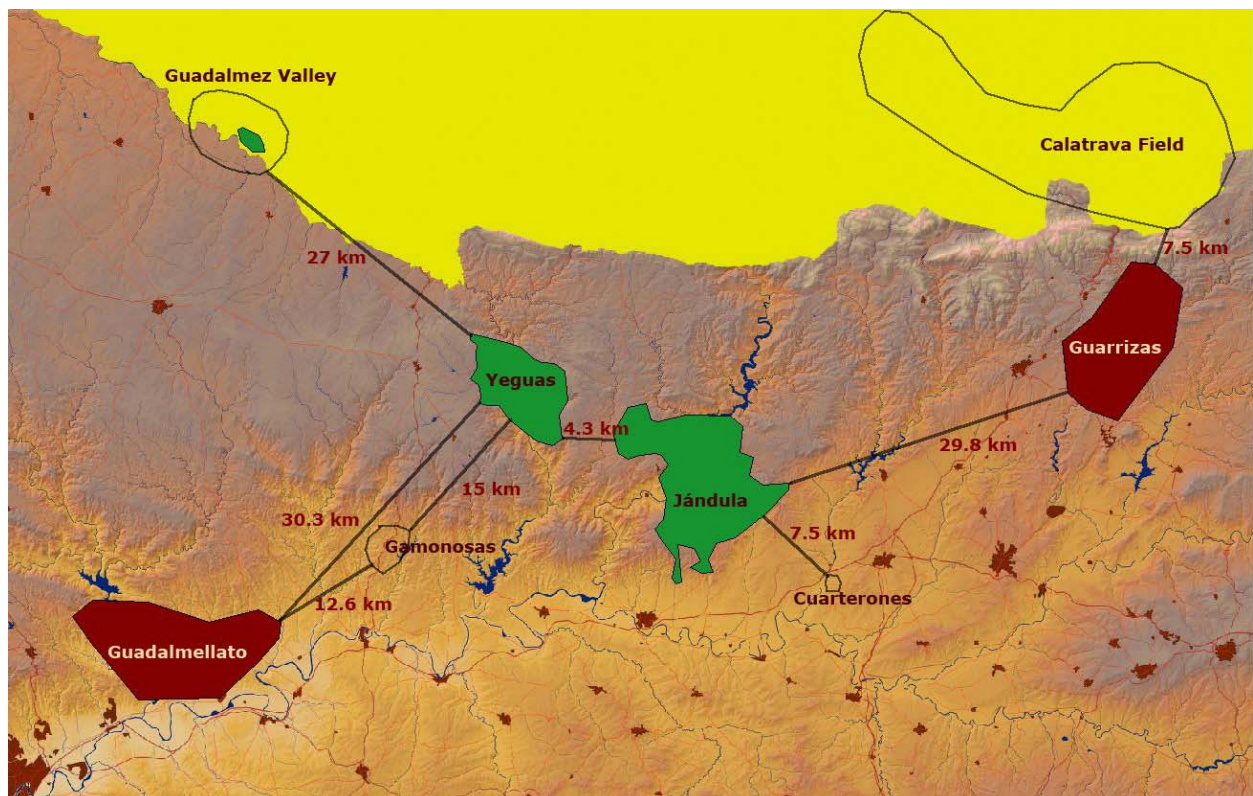


Figure 3. High-rabbit density nuclei conforming the potential Iberian lynx metapopulation of Western Sierra Morena. Mean distance among nucleus is indicated.

After the analysis, Hornachuelos was discarded due to both its long separation from the Andújar-Cardena Iberian lynx population and its low carrying capacity. However, the recolonization of Hornachuelos Chain is essential for the arrival of the species to high-rabbit-density areas of Southern Seville's Sierra Morena. The establishment of the Iberian lynx in those areas (Villanueva del Río y Minas and Vía River Canyon) would be the first step for the connection of Doñana and Sierra Morena populations. This would be possible between these areas and Doñana through the river Guadiamar green corridor. The priority must currently be to recover a large metapopulation to reach an effective population size that ensures the long-term viability of the species. Current conditions in Eastern Sierra Morena are excellent: seven nuclei with optimal rabbit density, summarizing approximately 841 km² (Fig. 5). This area holds, only in Andalusia, a carrying capacity of about 140 territories females with about 560 individuals. Rodríguez and Delibes (1990) suggested that the minimum necessary effective size to avoid problems related to inbreeding should be of 50 reproductive individuals. Although Doñana population has persisted with a lower effective size for at least 50 years (Valverde, 1963, Gaona et al. 1998), it is very likely that the minimum effective size for this species is significantly greater than 50 (Beissinger & McCullough 2002). The estimates for the potential Eastern Sierra Morena meta-population offer much better guarantees to maintain the long-term of the Iberian lynx population.

Conditions in Guadalmellato and Guarrizas are well balanced. Just the habitat structure is of higher quality at the first nucleus while the natural expansion possibilities are better in the second one. Given the scenario in which Guadalmellato is a priority, it is really difficult to dismiss Guarrizas, especially when considering that the Iberian lynx conservation is a race against the clock. The main limiting factor in the reintroduction processes will be the availability of individuals, since habitat requires feasible measures in this context. Moreover, the logistics of the current LIFE conservation project could afford working on both areas. In any case, if selecting only one of those areas, the works in the other one should not stop thinking about a sub-sequent reintroduction project (based, of course, in the

results on the first one and in the availability of individuals).

Acknowledgements

F. Coves, J. Guirado, C. Gañán, M. Delibes, F. Palomares, N. Fernández, A. Vargas, F. Martínez, M. Navarro, M. Briones, and N. Guzmán give support to the all the Iberian lynx conservation programs carried out in Andalusia. The other members of the LIFE project team (G. Valenzuela, G. Garrote, G. López, M.A. Díaz-Portero, A. Leiva, E. Rojas, R. Arenas, J. Bueno, S. Lillo, J. Pérez, M.I. García, M. Moral, J.A. Báñez, R. Sanabria, R.B. Millán, D. Palacios, CBD Hábitat Foundation team, WWF/Adena team, S. Saldaña, M.P. Delgado, R. García, I. Tenorio, M. Vara, Ecologistas en Acción, SECEM, ATECA, APROCA and FAC) are making all this work possible.

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Block C: “Reintroduction: landscapes and animals”

Case studies, release protocols and post-release monitoring



Breeding European wildcats in species-specific enclosures for reintroduction

Marianne Hartmann

Wildcat project. University of Zürich. mhart@access.uzh.ch

Tierstation Bockengut. CH-8810. Horgen.

Communication

In the course of a long-term experimental study in Switzerland, a species-specific enclosure for European Wildcats was developed in which animals encounter all the structures and stimuli relevant to their behaviours as regards all functional cycles.

The natural habitat of a species is commonly regarded as the environment where the animals' needs are best met. Since the species has adapted to the demands of this environment in the course of its evolution, this is where the animals are able to perform their natural behaviours. Consequently, the behaviour of animals living in the wild can be used as a basis for deducing zoo environments. However, most animals are difficult to observe in the wild. A large, complex and richly structured enclosure can be a substitute for the natural habitat if the animals are provided with the essential structures and stimuli necessary for performing their natural behaviours. An enclosure can be used as a substitute for the natural habitat and therefore as a reference for other housing systems if the animals in this enclosure are free of behavioural disturbances and if their behaviour does not deviate from that of their conspecifics in the wild, which is the case in my wildcat enclosures. The cats show a rhythm of activity similar to the one of their conspecifics in the wild, and an electronic feeding device, specifically tailored to wildcats, enables them to express very nearly the whole range of their natural hunting behaviour. The essential structures must be available in the right arrangement within the enclosure, with the keeper's appropriate behaviour as the second and equally important factor. The results of this study as well as the feeding technique have been applied in several wildlife parks in Switzerland and Germany.

Since 1993 the offspring of our cats have been provided for the wildcat reintroduction project in Bavaria. This project by the Bund Naturschutz in Bayern has been underway since 1984 under the direction of Guenther Worel. Between 1984 and 2008, 580 wildcats were released in three different regions in Bavaria. In order to avoid endangering autochthonous populations by removing individuals, captive-bred cats were used exclusively for reintroduction. Some of them were bred at the Bund Naturschutz wildcat station of Wiesenfelden, in enclosures offering naturalistic structures. Others were donated to the reintroduction project by more than 30 different zoos and wildlife parks in Europe. Prior to release all these animals passed a training of several months in the Wiesenfelden facilities. To a large extent they were trained by feeding live prey animals. Only the cats originating from Tierstation Bockengut, Switzerland, were released without any further training, because they had received extensive training in their parents' enclosures by the electronic feeder.

For several years there was no way of getting any information as to the fate of the Swiss cats after their release, because no direct monitoring was being done at that time. After the one realized in the very first years of the reintroduction, no further radio-tracking study had been planned by the Bund Naturschutz in Bayern.

In 1999 we could radio-track eleven wildcats in the course of a pilot study. Eight of these cats had been bred in our species-specific enclosures in Switzerland, whereas three cats came from zoos. All cats equipped with radio-collars were released in the Spessart, a hilly and wooded region in Bavaria. Besides testing the tracking equipment our interest was focused on the behaviour and survival of the cats.

The eleven wildcats were released from two sites in different seasons. Five cats bred in our enclosures in Switzerland were released in June. After leaving their cage in the forest the cats roved about for three to twenty days before they settled down. All of them survived until at least the middle of August and there was proof for two of them of being alive in Mai 2000, eleven months after their release. In addition, we have obtained evidence in the field indicating that they had no problems with catching enough prey.

At the end of September, six additional animals marked by collar transmitters were released of which three were killed crossing roads during the first two weeks after their release. We

assume that mainly seasonal effects led to this high mortality. With dusk setting in at an earlier time in autumn, we registered dislocations of the cats already in the earlier evening when traffic is much heavier, and, therefore, the danger for a cat to get killed is much higher than at later hours in summer.

Our three-years' study planned to follow the pilot phase has not been realized up to now. Extensive data on the behaviour of the animals after their release, on population density and on the survival and reproduction of part of the population should have been collected. These data would allow us to measure the success, which is indispensable for a reintroduction project.



Veterinary aspects in lynx translocation and reintroduction

Marie-Pierre Ryser-Degiorgis

Centre for Fish and Wildlife Health. Vetsuisse Faculty. University of Berne
marie-pierre.ryser@itpa.unibe.ch

Box 8466, CH-3001 Berne.

Abstract

Appropriate veterinary measures are required to quantify and minimise the veterinary risks attending the translocations of lynx as other animals, and to ensure health of released stock throughout the programme. Veterinary considerations should be performed both at the individual level (to ensure the survival of each single individual to be translocated) and at the ecosystem level (to prevent the movement of pathogens through the movement of animals).

Health risk assessment consists of evaluating whether or not important health-related risks are associated with the translocation of lynx. Independently of this risk, emphasis should be put on extensive sampling and information collection. However, while setting protocols, it is essential to differentiate between the analysis that are needed to decide whether an animal can be translocated or not, and the investigations that are pure scientific documentation. Furthermore, protocols should be regularly re-evaluated during the project implementation, in order to improve them if appropriate.

For a complete, long-term evaluation of the health situation in particular, and of the success of the translocation project in general, a post-release veterinary monitoring is also necessary. Furthermore, it is essential to analyze and publish available data in order to be able to learn from and to share experiences that could be valuable for other projects.



Sanitary control plan for the Iberian lynx reintroduction in Andalusia

Guillermo López¹, Fernando Martínez², Isabel Molina³, Irene Zorrilla⁴, Cristina Martínez⁵, Astrid Vargas² and Miguel A. Simón⁶

1. Iberian lynx LIFE conservation project. EGMASA. c/ Pepe Espaliú, 2. 14004. Córdoba. guiloza@gmail.com
2. Programa de Conservación *Ex-situ* del lince ibérico. Centro de cría "El Acebuche". Parque Nacional de Doñana. 21760. Matalascañas, Huelva.
3. Red Andaluza de CREAs. EGMASA. c/ Johan Gutenberg, 1. 41092. Sevilla.
4. EGMASA. Centro de Análisis y Diagnóstico de la Fauna Silvestre de Andalucía. Avd. Lope de Vega, 9. 29010. Málaga.
5. Parque Nacional de Doñana, El Acebuche. 21760. Matalascañas, Huelva.
6. Iberian lynx LIFE conservation project. Consejería de Medio Ambiente, Junta de Andalucía. c/ Fuente del Serbo, 3. 23071. Jaén.

Abstract

Sanitary aspects are of great importance in the reintroduction of species. Firstly, an optimum sanitary status of the released individuals is essential to the successful ongoing of the project, since only the best prepared individuals will be able to afford the stress cost that reintroduction itself (capture, transport, quarantine, release in an unknown area, contact with unknown individuals, etc) entails. Secondly, the interspecific contact of individuals that arrive to a new area is known to be much higher than that they have in their original territories, and so a previous knowledge of the sanitary status of the fauna living in the release area is crucial for evaluating the epidemiological risks and to elaborate preventive protocols. Thirdly, as far as reintroduction are usually performed through the movement of small groups of individuals, stochastic factors such as diseases are known to have a specially high relevance in the population dynamics of the reintroduced population. Thus, outbreaks can sometimes be devastating for a reintroduction project. Given these aspects, and due to the scarcity of the critically endangered Iberian lynx, a sanitary program is considered essential to perform reintroductions in the species. A draft sanitary plan for the reintroduction of the Iberian lynx has been performed by the Iberian lynx LIFE conservation project in collaboration with the advisory group of sanitary aspects of the Iberian lynx, which is presented here for its evaluation of discussion. The main actuation lines of this plan proposal are: (1) sanitary evaluation of the fauna living in the release area (mainly carnivores, but also ungulates and rabbits), (2) preventive protocol in domestic carnivores, (3) pre-release sanitary protocol of handled individuals and (4) post-release sanitary monitoring of the released individuals.

Introduction

As the most endangered felid in the world, the Iberian lynx is currently facing a situation that entails several different conservation challenges. Once the management of the species has allowed both the stabilization of its two only remaining populations (Simón et al. 2009) and the creation of a self-sustaining captive stock of individuals (Vargas et al. 2008), the following steps are being mainly focused on the decrease of the loss of genetic diversity and on the foundation of new population nuclei (Simón et al. 2009). Reintroduction of the Iberian lynx in areas of recent extinction is part of the Spanish Conservation Strategy of the species (Calzada et al. 2009). As explained in previous presentations, the first Iberian lynx

reintroduction is going to take place within the framework of the current Iberian lynx LIFE conservation project in Andalusia (LIFENAT 06/E/209) (Simón et al. 2009).

Diseases are relevant factors influencing the outgoing of reintroduction projects (Viggers et al. 1993; Wolff & Seal 1993), and so the design and implementation of preventive and sanitary monitoring protocols is considered essential in such projects (Cunningham, 1995; Teixeira et al. 2007). The sanitary status of the Iberian lynx population has been studied for more than 15 years (Roelke et al. 2007; Meli et al. 2009), and an intensive sanitary monitoring program is being carried out since 2003 (Meli et al. 2009). Given

this, there is currently an essential knowledge about the main agents capable of affecting population dynamics of the species. Some pathogens, such as the Feline Leukaemia Virus or the Canine Distemper Virus, are known to be able to cause devastating effects if they reach an Iberian lynx population (Meli et al. 2009; López et al. 2009), so the prevention of its incorporation into the reintroduced lynx population seems to be crucial for the success of the project. Given the high genetic importance of each Iberian lynx individual, every step in the reintroduction of the species is being carefully considered and evaluated, and so the draft sanitary control plan for the Iberian lynx reintroduction (DSCP) has been developed relying on the experience and knowledge of all veterinarians and biologists working in one way or another in relation to the sanitary aspects of the Iberian lynx conservation. In order to cover the higher number of fronts of work, this plan has been divided into four principal actuation lines: (1) sanitary evaluation of the fauna living in the release area, (2) preventive protocol in domestic carnivores, (3) pre-release sanitary protocol of individuals to be released in the area and (4) post-release sanitary monitoring of the released individuals.

Sanitary evaluation of associated fauna

Firstly, we considered the potential sanitary risk that the preys entail to the Iberian lynx.

The Iberian lynx is a specialist predator whose diet is mainly composed of rabbits (*Oryctolagus cuniculus*), while rodents and birds are considered minor preys (Gil-Sánchez et al. 2006). Feeding on wild ungulates (red, fellow and roe deers, and wild boars) has also been recorded (Gil-Sánchez et al. 2006). Rabbits and small minor preys have never shown to represent a sanitary risk for the Iberian lynx, but wild ungulates living in areas of occupancy of the species have shown very high Tuberculosis (TBC) prevalence, what does represent a real risk for the Iberian lynx (Briones et al. 2000; Aranaz, 2004; Gortázar et al. 2008). Moreover, Aujeszky's Disease (AD) prevalence have shown to be high in wild boars in Spain (Ruiz-Fons et al. 2006; Ruiz-Fons et al. 2008). Although it has never been detected in the Iberian lynx, the pathogenicity and the biological cycle of this agent is thought to represent a potential risk for the lynx and other carnivores. Under this scenario, an evaluation of TBC in all ungulate species and of AD only in wild boars is included in the DSCP. An annual sampling during the hunting period should be practiced, collecting lung samples of all ungulates plus tonsil samples of wild boars to run PCRs of TBC and AD respectively (see table 1).

Secondly, we considered the sanitary risk that other carnivores (both domestic and wild) represent to the Iberian lynx in the reintroduction areas. Contact with and killing of

Group	Species	TBC	AD	FeLV	Cytauzoon	FIV	CDV	FPV	FCoV	Sampling
Ungulates	Red deer	PCR lung								Fall-Winter
	Fellow deer	PCR lung								Fall-Winter
	Wild boar	PCR lung	PCR Tonsils							Fall-Winter
Carnivores	dog, fox, wolf	PCR lung					ELISA blood; PCR swab & faeces	PCR blood & faeces	PCR blood & faeces	One week/month
	domestic cat, wildcat	PCR lung		ELISA blood PCR bone marrow	PCR blood	ELISA blood PCR blood	ELISA blood; PCR swab & faeces	PCR blood & faeces	PCR blood & faeces	One week/month
	Other carnivores	PCR lung					ELISA blood; PCR swab & faeces	PCR blood & Faeces	PCR blood & Faeces	One week/month

Table 1. Infectious agents to analyse in the sanitary evaluation of the associated fauna, both through direct capture and through carcase findings (TBC = Tuberculosis; AD = Aujeszky Disease; FeLV = Feline Leukaemia Virus; FIV = Feline Immunodeficiency Virus; CDV = Canine Distemper Virus; FPV = Feline Parvovirus; FCoV = Feline Coronavirus).

other carnivore species has been widely documented for the Iberian lynx (see Palomares & Caro, 1999; figure 1). Moreover, intraspecific contact and aggressions are known to be more common than normal in carnivore reintroductions, since the reintroduced species must colonize a niche that is already occupied by another one. These factors, together with the fact that the most dangerous agents for the lynx are carried by other carnivores make consider this group as the main sanitary threat for the Iberian lynx in the reintroduction project. The DSCP includes both sanitary evaluations of carnivores before releasing lynxes and sanitary monitoring of the carnivore population once lynxes have been released. The

scope of these actions is all carnivore species living in the area that are known to be able to directly contact with the Iberian lynx (see table 1). The pre-release sampling would be performed in a period enough to reach a significant number of samples of all species. The continued monitoring surveillance would be performed at least one week per month since the release of the first individuals. Also, all carcasses of dead carnivores found during the usual work of the monitoring team will be taken, sampled and analysed. Agents to analyse are those which have shown to represent a greater risk for the Iberian lynx, and the techniques to perform the analysis are mainly PCRs and ELISAs (see table 1).



Figure 1. European genet (*Genetta genetta*) killed by a female Iberian lynx in Doñana. Iberian lynxes use to kill other carnivores biting their neck, and after that they abandon the body in the field. This is considered a risky behaviour from a sanitary point of view.

Preventive protocol in domestic carnivores

Domestic carnivores are known to be a source of disease for the Iberian lynx (Meli et al. 2009), and so this group is considered of high sanitary risk in the reintroduction project. Spanish and Andalusian laws bind to properly identify through microchip and vaccinate against rabies all domestic carnivores (dogs, cats and ferrets). This law is, however, broken by many people, mainly in rural areas. The first objective of the DSCP is to achieve that inhabitants of

reintroduction areas living with domestic carnivores obey the current law. Although an important first step, this point is considered insufficient, and a further preventive protocol for the domestic carnivores is included in the DSCP. The objectives of this protocol are (1) to decrease the domestic carnivore pressure, (2) to decrease both their aggressive behaviour and their mobility, and (3) to improve the sanitary status of the domestic carnivores living in potential contact with the lynx population.

The lines of the preventive protocol are (1) voluntary free-cost castrations, (2) voluntary free-cost vaccinations and (3) control of feral dogs and cats.

Voluntary free-cost castrations: It will be offered to the owners of dogs and cats inside the reintroduction area the possibility of perform castrations or sterilizations to their pets with no charge for them.

Voluntary free-cost vaccinations: The owners of domestic carnivores living inside the reintroduction area will have the possibility of vaccinating their pets with no charge, once they have been sampled in the sanitary evaluations phase. Cats would be vaccinated with the pentavalent PureVax® RCPch FeLV vaccine, dogs with the heptavalent EURICAN® MHPPI2L vaccine and the ferrets with a Ferret Distemper Virus vaccine.

Control of feral dogs and cats: All feral dogs and cats captured in the trapping for sanitary evaluation that lack of an owner will be carried to an animal protection society to wait for an owner.

Pre-release sanitary protocol

Iberian lynxes to be released in the field must be sanitarilly evaluated and quarantined in order to (1) ensure they are in enough good condition to afford the cost of reintroduction handling with chances to adequately integrate in the new population, (2) ensure that they are healthy and free of pathogens that could affect the dynamics of the population, and (3) collect data about their behaviour to adapt, as much as possible, individually their future management. Those individuals captured in the field to be translocated to reintroduction areas must follow the official *Sanitary Protocol for Iberian Lynxes Translocated Between two Different Populations*. This protocol consists of a quarantine of a minimum of 15 days and two sanitary evaluations: one at the beginning and

another one at the end of the quarantine. Quarantine enclosures should be those located in the captive breeding centres, although suitable enclosures of rescue centres can also be used. Infectious agents to be evaluated are listed in table 2. Animals positive to blood or swab PCR of Feline Coronavirus (FCoV), Feline Calicivirus (FCV), Feline Leukaemia Virus (FeLV), Feline Immunodeficiency Virus (FIV), Canine Distemper Virus (CDV), Feline Herpesvirus (FHV) or Feline Parvovirus (FPV) will be ruled out for translocations. Individuals in quarantine must be always deparasited and, when necessary, they may also be vaccinated. Handling of these individuals must be elusive, and their natural behaviour must be stimulated. Individuals coming from the training enclosures of the captive breeding program must follow the same protocol, although their own enclosures can be used as quarantine enclosures as long as they have not contact with other individuals.

Post-release sanitary monitoring

The sanitary monitoring of the reintroduced Iberian lynx population will be the same as the one practiced over the currently exiting populations. Similarly to current distribution areas, exclusive installations to perform medical exams to the Iberian lynxes should be habilitated in the area. These installations must count with all the material listed in the Manual for the Sanitary Management of the Iberian Lynx (internal document not published; see figure 2). Annual checkups of all individuals will be maid during the first three years. As of the fourth year, a sample of the population will be evaluated annually. Non-intrusive monitoring via analysis of scats will be developed when it is considered necessary. Captures and sanitary evaluations will be preferably made during the autumn and early winter, in order to not disturb the breeding season. Standard Iberian lynx techniques and procedures will be employed in all sanitary evaluations of the species.

	FCV	FCoV	FHV	FPV	FeLV	FIV	CDV	FPV	A.p.	M.h.	C.h.	M.t.	C.f.
Serology	Inf.	Inf.	Inf.	Inf.		Inf.	Inf.						
PCR	Det.	Det.	Det.	Det.	Det.	Det.	Det.	Inf.	Inf.	Inf.	Inf.	Inf.	Inf.

Table 1. Infectious agents to analyse in the pre-release sanitary evaluations of the Iberian lynx, both deterministic (which positive excludes the animal for a translocation), and informative (which do not). (FCV = Feline Calicivirus; FCoV = Feline Coronavirus; Feline Herpesvirus; Feline Parvovirus; FeLV = Feline Leukaemia Virus; FIV = Feline Immunodeficiency Virus; CDV = Canine Distemper Virus; FPV = Feline Parvovirus; A.p. = Anaplasma phagocytophilum; M.h. = Mycoplasma haemofelis; C.h. = Candidatus M. haemominutum; M.t. = Mycoplasma turicensis; C.f. = Cytauxzoon felis).

Conclusion

The draft sanitary plan presented here is thought to be an important management tool to prevent Iberian lynx reintroduction failure due to infectious diseases. Thus, the redactor group of this proposal believes that the implementation of the plan might be essential to control, as much as possible, the sanitary variables in the Iberian lynx reintroduction project (mainly at its first stages), since these sanitary variables have shown to be of capital importance in the population dynamics of the Iberian lynx and other carnivores. Given the current status of the species, efforts must be

put on all possible variables to have more chances to succeed.

Acknowledgements

The whole LIFE project team (J.M. Gil, M. López, L. Fernández, G. Valenzuela, G. Garrote, G. Ruiz, M.A. Díaz-Portero, A. Leiva, E. Rojas, R. Arenas, J. Rodríguez, J. Bueno, S. Lillo, J. Pérez, M.I. García, M. Moral, J.A. Báñez, R. Sanabria, R.B. Millán, D. Palacios, B. Torralba, S. Saldaña, J.M. Martín, M.P. Delgado, R. García, I. Tenorio and M. Vara) and the rest of GAAS members are making all this work possible.



Figure 2. View of the field clinical installations and equipment currently used to perform sanitary evaluations in Doñana free-ranging Iberian lynxes. Installations should not be used to evaluate any other species, and prophylaxis measures must be strict. The same check-up material and equipment should not be used in different populations.

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Genetic introgression in the Florida panther population

Dave Onorato

Fish and Wildlife Research Institute, Florida Fish and Wildlife Conservation Commission dave.onorato@myfwc.com

566 Commercial Boulevard, Naples, FL 34104, USA.

Communication

The worldwide decline in wild felid populations is often associated with varied anthropogenic factors that perpetually lead to endangerment or extinction. The Florida panther (*Puma concolor coryi*) provides a perfect example of the plight faced by many populations of wild felids in the 21st century. This population of puma was severely reduced during the 19th and 20th centuries due to a variety of reasons, most notably the loss of habitat and conflicts with an expanding human population in the southeastern United States. This decline led to the listing of the Florida panther as endangered by the Federal Government in 1967, with additional protection afforded under the Endangered Species Act in 1973. By the early 1980s, the panther population was

restricted to the southern tip of peninsular Florida and encompassed < 20 individuals. To obtain a more complete understanding of the general biology of the panther, the Florida Fish and Wildlife Conservation Commission initiated research on this population in 1981. Early findings revealed that panthers were being negatively impacted by a variety of factors including low levels of genetic variation associated with inbreeding depression. Federal and state officials initially attempted to avert the continued decline of panthers via the initiation of a captive breeding program in 1991. This project involved the removal of 10 kittens from the wild population from 1991-1992. Attempts at captive breeding were suspended in 1992 due to increasing fears that

Period	N	H _o	H _e	Mean # alleles/locus
1969-1985	33	0.473 (0.18)	0.483 (0.17)	3.8
1986-1990	30	0.384 (0.21)	0.410 (0.18)	2.9
1991-1995	38	0.402 (0.22)	0.380 (0.18)	3.4
1996-2000	85	0.471 (0.18)	0.484 (0.17)	4.7
2001-2007	195	0.496 (0.17)	0.514 (0.18)	5.6
Western NA	14	0.585 (0.18)	0.631 (0.18)	4.6

Figure 1. Changes in the levels of genetic variation (H_o and H_e, observed and expected heterozygosity, respectively) in cohorts of Florida panthers born prior to genetic restoration (<1996) and subsequent to introgression (highlighted in orange). Samples from cougars in western North America are included for comparisons. Analyses were completed on genotypes derived from 18 microsatellite loci that were determined to be variable in Florida panther. Results are preliminary.

the wild population was continuing to experience deterioration in genetic viability and survival of the population was deemed questionable. Subsequent discussions among state and federal officials led to the approval of a genetic restoration program in 1995 that involved the introgression of 8 breeding-aged female Texas pumas (*Puma concolor stanleyana*) into the remaining wild population of panthers in South Florida. Five of these 8 females bred successfully and produced a minimum of 20 offspring.

Preliminary results from this genetic restoration project based on more than a decade of research involving the efforts of multiple state and federal agencies appear to indicate that it has been successful. Genotypic data from 18 microsatellite loci were collected from > 380 panthers sampled between 1940 and 2007. These genetic data were combined with field observations and biomedical records to assess changes in genetic variation, population structuring, ancestry and demographic variables. Additionally, temporal trends associated with reproductive abnormalities and congenital defects were delineated within the panther population during the pre- and post-introgression periods. Subsequent to genetic introgression, there has been an increase in heterozygosity in new cohorts of panthers. Conversely, there has been an observed decrease in the prevalence

of atrial septal defects and cryptorchidism. The ancestral composition of post-introgression cohorts is significantly more heterogeneous in comparison to panthers born prior to the release of the Texas pumas. A resulting expansion of the panther population has occurred since 1995, when surveys indicated the presence of only 25 panthers. Surveys during 2007 detected the presence of 117 adult and subadult panthers. This expansion has resulted in the recolonization of undeveloped former range in southwestern Florida.

Recovery of the Florida panther, as defined by the U.S. Fish and Wildlife Service, will depend upon the establishment of 3 populations consisting of at least 240 panthers each, therein highlighting the challenges faced by managers and researchers. While inbreeding and genetic variation remain issues of concern, our findings associated with the genetic restoration project should assist in developing effective panther conservation strategies. Additionally, this assessment of genetic introgression in a wild population of felids should be applicable to the conservation of other imperiled large carnivores worldwide, such as the Iberian lynx (*Lynx pardinus*). Ultimately though, as with most endangered large mammals, recovery will continue to hinge largely on the preservation of usable-space and improving prospects for recolonization..



Figure 2. View of a Florida panther

The Colorado Canada Lynx Reintroduction Program

Tanya M. Shenk¹ and Rick H. Kahn

Colorado Division of Wildlife 1. tanya.shenk@state.co.us

317 West Prospect Road, Fort Collins. Colorado, 80526, USA.

Communication

The Canada lynx (*Lynx canadensis*) occurs throughout the boreal forests of northern North America. While Canada and Alaska support healthy populations of the species, the lynx is currently listed as threatened under the Endangered Species Act of 1973, as amended (16 U. S. C. 1531 et. seq.) (U. S. Fish and Wildlife Service, 2000) in the coterminous United States. Colorado represents the southern-most historical distribution of naturally occurring lynx, where the species occupied the higher elevation, montane forests in the state (U. S. Fish and Wildlife Service 2000). Thus, Colorado is included in the federal listing as lynx habitat. Lynx were extirpated or reduced to a few animals in Colorado, however, by the late 1970's (U. S. Fish and Wildlife Service 2000) due, most likely due to multiple human-associated factors, including predator control efforts such as poisoning and trapping (Meaney, 2002). Given the isolation of Colorado to the nearest northern populations, the Colorado Division of Wildlife considered reintroduction as the only option to attempt to reestablish the species in the state.

Therefore, a reintroduction effort was begun in 1997, with the first lynx released in Colorado in 1999. To date, 218 wild-caught lynx from Alaska and Canada have been released in southwestern Colorado. The goal of the Colorado lynx reintroduction program is to establish a self-sustaining, viable population of lynx in this state. Evaluation of incremental achievements necessary for establishing viable populations is an interim method of assessing if the reintroduction effort is progressing towards success. There were 7 critical criteria to be met before concluding a viable population had been established: 1) development of release protocols that lead to a high initial post-release survival of reintroduced animals, 2) long-term survival of lynx in Colorado, 3) development of site fidelity by the lynx to

areas supporting good habitat in densities sufficient to breed, 4) reintroduced lynx must breed, 5) breeding must lead to reproduction of surviving kittens 6) lynx born in Colorado must reach breeding age and reproduce successfully, and 7) recruitment must equal to or be greater than mortality over an extended (~ 10 year) period of time (Shenk, 2006).

The fundamental approach taken to evaluate the status of each of these criteria was to PIT-tag and place telemetry collars on every lynx released and as many Colorado-born kittens surviving to adulthood as possible, followed by intensive monitoring of these animals through satellite, aerial and ground-tracking. All establishment criteria, except (7) have been achieved. Lynx populations in Canada and Alaska have long been known to cycle in response to the 10-year snowshoe hare (*Lepus americanus*) cycle (Elton & Nicholson 1942). Northern populations of lynx respond to snowshoe hare lows first through a decline in reproduction followed by an increase in adult mortality; when snowshoe hare populations increase, lynx respond with increased survival and reproduction (O'Donoghue et al. 2001). Therefore, annual survival and reproduction are highly variable but must be sufficient, overall, to result in long-term persistence of the population. It is not known if snowshoe hare populations in Colorado cycle and if they do, where in the approximate 10-year cycle we are currently. Given this uncertainty, documenting persistence of lynx in Colorado for a period of at least 10-15 years would provide support that a viable population of lynx can be sustained in Colorado even in the event snowshoe hares do cycle in Colorado.

Survival, movement patterns, reproduction, and landscape habitat-use were documented through aerial (n = 9496) and satellite (n = 23,791) tracking. Most lynx remained near the

core release area in southwestern Colorado. From 1999-August 2008, there were 112 mortalities of released adult lynx. The primary known causes of death included 30.4% human-induced deaths which were confirmed or probably caused by collisions with vehicles or gunshot. Malnutrition and disease/illness accounted for 18.8% of the deaths. An additional 36.6% of known mortalities were from unknown causes. The monthly mortality rate was lower inside the core research area than outside, and slightly higher for male than for female lynx, although 95% confidence intervals for sexes overlapped (Devineau et al. 2009). Mortality was higher immediately after release (first month = 0.0368 [SE = 0.0140] inside the study area, and 0.1012 [SE = 0.0359] outside the study area), and then decreased according to a quadratic trend over time.

Reproduction is critical to achieving a self-sustaining viable population of lynx in Colorado. Reproduction was first documented from the 2003 reproduction season and again in 2004, 2005 and 2006. Lower reproduction occurred in 2006 but did include a Colorado-born female giving birth to 2 kittens, documenting the first recruitment of Colorado-born lynx into the Colorado breeding population. No reproduction was documented in 2007 or 2008. The cause of the decreased reproduction from 2006-2008 is unknown. One possible explanation would be a decrease in prey abundance. Additional reproduction is likely to have occurred in all years from females we were no longer tracking, and from Colorado-born lynx that have not been collared. The dens we find are more representative of the minimum number of litters and kittens in a reproduction season. To achieve a viable population of lynx, enough kittens need to be recruited into the population to offset the mortality that occurs in that year and hopefully even exceed the mortality rate to achieve an increasing population.

From snow-tracking, the primary winter prey species ($n = 548$ kills) were snowshoe hare (annual = 74.9%, SE = 4.6, $n = 9$) and red squirrel (*Tamiasciurus hudsonicus*, annual = 16.5%, SE = 4.1, $n = 9$); other mammals and birds formed a minor part of the winter diet. The annual percent of snowshoe hare kills ranged from a low of 55.56% in 1999 to a high of 90.77% in winter 2002-2003. Lynx appear to

remain faithful to an area during winter months, and exhibit more extensive movements away from these areas in the summer. Reproductive females had the smallest 90% utilization distribution home ranges ($\bar{x} = 75.2$ km², SE = 15.9 km²), followed by attending males ($\bar{x} = 102.5$ km², SE = 39.7 km²) and non-reproductive animals ($\bar{x} = 653.8$ km², SE = 145.4 km²). Most lynx currently being tracked are within the core research area. During the summer months, lynx were documented to make extensive movements away from their winter use areas. Extensive summer movements away from areas used throughout the rest of the year have been documented in native lynx in Wyoming and Montana.

Lynx use-density surfaces were generated to illustrate relative use of areas throughout Colorado and areas of use in New Mexico, Utah and Wyoming. Within the areas of high use in southwestern Colorado, site-scale habitat use, documented through snow-tracking, supports mature Engelmann spruce (*Picea engelmannii*)-subalpine fir (*Abies lasiocarpa*) forest stands with 42-65% canopy cover and 15-20% conifer understory cover as the most commonly used areas in southwestern Colorado. Little difference in aspect (slight preference for north-facing slopes), slope ($\bar{x} = 15.7^\circ$) or elevation ($\bar{x} = 3173$ m) were detected for long beds, travel and kill sites ($n = 1841$). A total of 37 dens have been found from 2003-2006. All of the dens except one have been scattered throughout the high elevation areas of southern Colorado. In 2004, 1 den was found in southeastern Wyoming, near the Colorado border. Dens were located on steep (slope = 30° , SE=20), north-facing, high elevation ($\bar{x} = 3354$ m, SE = 31 m) slopes. The dens were typically in Engelmann spruce/subalpine fir forests in areas of extensive downfall of coarse woody debris. All dens were located within the winter use areas used by the females.

The program also investigates the ecology of snowshoe hare, the primary prey of Canada lynx, in Colorado. A study comparing snowshoe hare densities among mature stands of Engelmann spruce/subalpine fir, lodgepole pine (*Pinus contorta*) and Ponderosa pine (*Pinus ponderosa*) was completed in 2004 with highest hare densities found in Engelmann spruce/subalpine fir stands and no hares found in Ponderosa pine stands (Zahratka & Shenk

2008). A study to evaluate the importance of young, regenerating lodgepole pine and mature Engelmann spruce/subalpine fir stands in Colorado by examining density and demography of snowshoe hares that reside in each was initiated in 2005 and will continue through 2009 (Ivan, 2005).

From results to date it can be concluded that the Colorado Division of Wildlife developed release protocols that ensure high initial post-release survival of lynx, and on an individual level, lynx demonstrated they can survive long-term in areas of Colorado. We also documented that reintroduced lynx exhibited site fidelity, engaged in breeding behavior and produced kittens that were recruited into the Colorado breeding population. What is yet to be demonstrated is whether current conditions in Colorado can support the recruitment necessary to offset annual mortality in order to sustain the population. Monitoring of reintroduced lynx will continue in an effort to document such viability.

Acknowledgements

The Colorado lynx reintroduction program involves the efforts of many people across North America, in Canada and the USA, all of whom deserve thanks. These people include state, federal and provincial biologists, researchers, technicians, pilots, managers, veterinarians and administrators.

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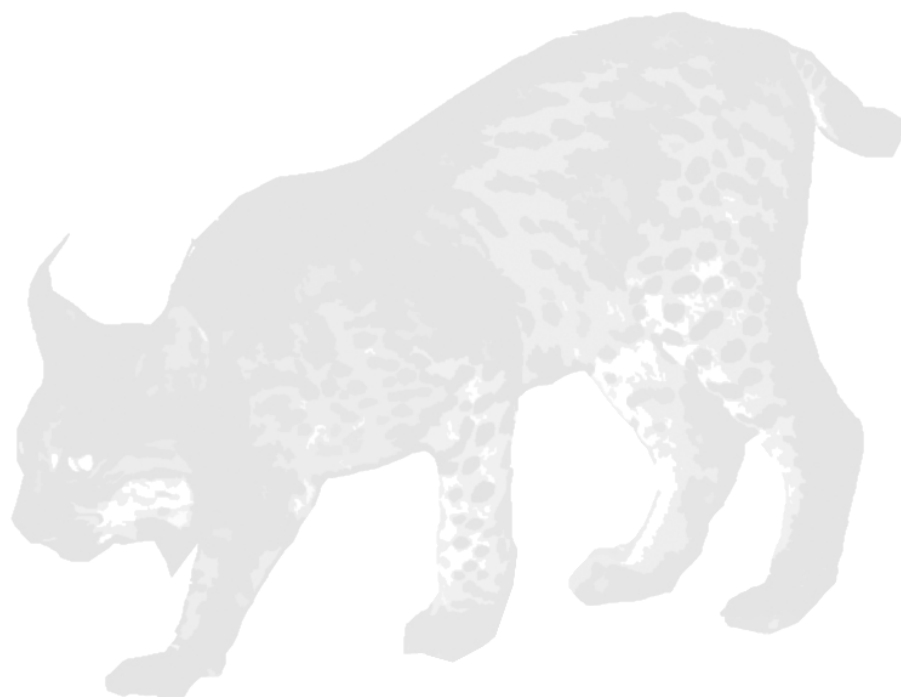
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Translocation and post-release monitoring in Eurasian lynx reintroduction

Andreas Ryser¹ Kuno von Wattenwyl¹, Christian Willisch¹, Fridolin Zimmermann¹, Marie-Pierre Ryser-Degiorgis¹ and Urs Breitenmoser¹.
Koordinierte Forschungsprojekte zur Erhaltung und zum Management der Raubtiere in der Schweiz, KORA. 1. a.ryser@kora.ch
Thunstrasse 31. CH-3074. Muri.

Abstract

To establish a new population of Eurasian lynx (*Lynx lynx*) in eastern Switzerland, we caught 12 free ranging lynx in western Switzerland and translocated them to the eastern Swiss Alps from 2001–2008. The animals underwent veterinary checkups and were brought to a quarantine station for 2-4 weeks before being released. We followed all animals closely by means of radio-telemetry. Post release monitoring included post-release migration, home-range establishment, hunting and prey choice, social behaviour and reproduction, observed by means of radio-telemetry, camera trapping and snow tracking. Four lynx showed far ranging movements before eventually establishing home-ranges near the release sites. In one case, a male lynx leaving the Alps was recaptured and brought back into the release area, where he has remained since. Two lynx were found dead (road kill and heart disease, respectively); the contact to five animals was lost for unknown reasons. Of 13 kittens born (2002-2007), at least five reached independency at the age of one year. One subadult dispersed 200 km to the southern edge of the Alps. All lynx fed on the expected main prey: roe deer (*Capreolus capreolus*) and chamois (*Rupicapra rupicapra*). The population is still small and it is too early to assess the success of the reintroduction project. Some aspects can however be addressed as positive or negative points already now. The decision about the translocation was based on a broad political consultation between the Federal institutions and several cantons and involved stakeholders. While this consultation was indispensable for the acceptance of the project, it resulted in some political compromises, such as limiting the number of animals to be released to 12 lynx. The project was however planned as an adaptive process, allowing reacting with further releases over several years according to the monitoring results. Animals from two source populations (north-western Alps and Jura) were brought together to enhance genetic diversity. Protocols for captures, transport, quarantine and veterinary supervision were developed in close cooperation between wildlife biologists, veterinarians and wildlife management staff, and proved to be effective. Monitoring of the released animals was intensive, but due to financial and political constraints, offspring could not be radio-tagged. The further monitoring of the developing population is done by means of camera-trapping, a method with limited validity in such a small nucleus. Information of stakeholders and the broad public was comprehensive, what however also boosted the continued and often controversial discussion about the return of this large predator.

Genetic re-stocking in the Doñana Iberian lynx population

Gema Ruiz¹, Juan A. Franco², Marcos López², Leonardo Fernández²
Guillermo López³ and Miguel A. Simón⁴

1. Iberian lynx LIFE conservation project. EGMASA. c/ Johan Gutenberg, 1. 41092. Sevilla.

2. Iberian lynx LIFE conservation project. EGMASA. C/ Plus Ultra, nº8, 7ª Planta. 21001. Huelva.

3. Iberian lynx LIFE conservation project. EGMASA.
c/ Pepe Espaliú, 2. 14004. Córdoba.

4. Iberian lynx LIFE conservation project. Consejería de Medio Ambiente, Junta de Andalucía.
c/ Fuente del Serbo, 3. 23071. Jaén.

Introduction

The main goal of the first Iberian lynx translocation, which took place in late 2007 under the framework of the Iberian lynx LIFE conservation Project (LIFE06NAT/E/209), was to achieve a genetic reinforcement of the inbred Doñana population, although also a demographic reinforcement was pursued. Even though the original restocking project was scheduled to take place in a different part of the Protected Area, a crisis management led to reconsider the initial release site. The initially selected place was the Biological Reserve of Doñana, where the protection level is the highest in all Doñana and where there is enough suitable non-occupied surface of good habitat to host a new territory. But given that the Feline Leukaemia Virus outbreak that hit Doñana in 2007 wiped out all males living in *Coto del Rey* subpopulation (5 adult breeding and 3 subadult males) (López et al. 2009; Meli et al. 2009), the final choice was to release a Sierra Morena-coming male in such nucleus of the Doñana National Park. Although its prior objective was to increase the genetic diversity of the Doñana population, this specific management also pursued to decrease the potential risk of short-term extinction of the Doñana Iberian lynx population. Moreover, the translocation of a male had the partial objective of ensuring all three resident females living in southern *Coto del Rey* did not leave the nucleus in search of a mate during the imminent breeding season.

The importance that this nuclei had have in the dynamics of the entire Doñana Iberian lynx population was analyzed by means of a population model, which results predicted that the probability of extinction of the population

would increase in 10% in case one single territory was lost in *Coto del Rey* subpopulation (Revilla et al. 2007). The most probable scenario would have been the settlement of a juvenile male, with lower reproductive potential than that of an adult. In fact, this has been proven to be the case, since only one subadult male has naturally re-colonized the area one year after the FeLV outbreak took place.

The situation found after the loss of all territorial males in southern *Coto del Rey* left three breeding females available in Doñana's main "population source" which, paradoxically, was a situation that would allow for a more rapid gene flow of Sierra Morena genes through the Doñana population.

Acclimatization pen

The first translocation carried out in Doñana Iberian lynx population was decided to be performed by means of soft-release techniques aiming to allow a flexible and gradual acclimatization of the released individual (figure 2). This experience would allow to test the facilities and to learn as much as possible from this release in order to improve techniques for future translocation and reintroduction management. Thus, a two-hectare enclosure 3.85 m high and an additional 0.5 m buried underground was built. Inside the enclosure there was a supplementary feeding station (SFS), as well as a den site. To create points of attraction for the resident females, faeces of the male handled were distributed along the paths surrounding the enclosure. More of these scats were placed inside the enclosure both to promote exploratory movements of the male in the

acclimatization pen and to maximize the utilization of the available surface. Actual placement of the acclimatization pen was carefully chosen after evaluating the home-range overlap of the resident females (using a 90% kernel) of the three territorial females (see figure 1). Finally, all movements would be monitored with a surveillance system based on an infrared vision video-camera (365° movement) operated by LIFE project personnel 24 hours per day. One LIFE project technician at a time stayed in a hide placed out of the enclosure to monitor both the reaction of the females –they were all radio-tagged- and the behaviour of the male, waiting for the best moment to proceed with the release.

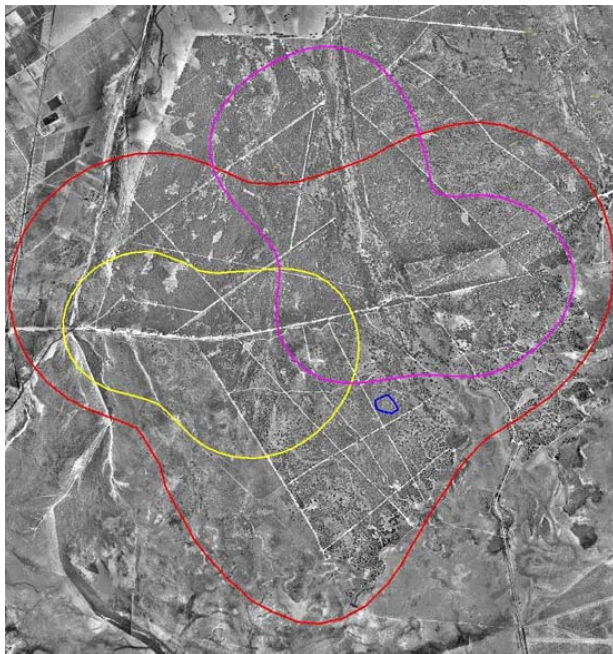


Figure 1. Location of the acclimatization pen (blue line) in relation to the 90% Kernel-based home range of the three resident females (Rayela = red line; Viciosa = Yellow line; Wari = purple line).

Selection of the male

The male selected for translocation, *Baya*, belonged to the 2005 cohort. One of the selection criteria included that the animal would be negative to *Cytauxzoon felis*, since this parasite has never been detected in the Doñana free-ranging population (see Millán et al. 2007) and the effects of its incorporation in there are unpredictable. *Baya* was going to be plenty reproductively adult in 2007-2008 season, and so the translocation of an individual in its age would grant the longer possible in Doñana, maximizing the

introgression of new genes in the population. In addition, the spermatic quality of *Baya* was checked through electro-ejaculation. *Baya* showed the best sperm quality out of the males in its age. To avoid the introduction of any foreign disease to the receptor area, *Baya* was kept in quarantine for 19 weeks by the *Ex-situ* Conservation Program and, after that, it was transferred to the acclimatization enclosure.

Pre-release Considerations

The final decision regarding the moment of actual release was determined taking the three following factors into account: (1) Positive and continuous interactions through the wire mesh between *Baya* and the resident females, (2) estimation of the timing of oestrous of the females based on their birthing periods in previous years (that was during the first days of January for the most precocious female), and (3) establishing a maximum limit for holding the male in the pen if no interactions with females had been observed (it was decided that January 10th would be the maximum waiting period, always considering the observed behaviours during the acclimatization phase. Despite this, the decision would change if necessary, because a learning adaptive management was searched. In this way, it was considered important to check hunting abilities in and out the SFS, as well as the physical appearance and the lack of stress-derived behaviours.

Acclimatization process

Baya's translocation took place on December 21st, 2007. Resident females visited the perimeter of the enclosure from the very first day. It was possible to observe differences in behaviour and in time spent in the enclosure in every female through the time (their interest and the time spent in the enclosure increased while oestrous date got closer).

Baya was released on January 1st, 2008, when LIFE Project personnel estimated that the animal was properly acclimated to the site, and considering that there have been a large number of positive interactions through the mesh with the three territorial females present in the area. During the time spent inside the enclosure *Baya* used the supplementary feeding station and also hunted wild rabbits. It moved for the whole available space of the



Figure 2. Baya, the first Sierra Morena-coming male released in Doñana as a population and genetic reinforcement, just after being released in the acclimatization soft-release enclosure. (Photo: Eduardo Abad/EFE)

enclosure and neither stress-derived behaviours nor stereotypical movements were ever detected. The actual release took place after one of the females, called *Wari*, spent more than a day in the vicinity of the enclosure, vocalizing towards the male and showing signs of oestrous.

Post-release Monitoring

Intensive post-release monitoring efforts were conducted by the LIFE project's specialized staff in order to learn as much as possible from this release. These efforts continue to date but in a less intensive way.

Radiotelemetry. Just after the release, at least two daily locations were obtained from *Baya*. The animal was followed for a while if it presented activity and, at the same time, all three territorial females were also daily located. *Baya* and *Wari* could be watched together showing good interactions on January 6th, 7th and 8th (see figure 3). The male never moved beyond 4 km from the release point. After two months, and given that *Baya* had settled in the area, it was included in the regular radio-monitoring protocol of Doñana population, which consists of at least 2-3 locations per week.

Photo-trapping and tracking. The doors of the SFS were equipped with photo-trapping

cameras, which allowed for a precise control of the animals and the use they made of the enclosures. Altogether, a total of 212 lynx photographs have been obtained from the 6 photo-trapping stations during a 6-month period (from January to June). *Baya* and two of the territorial females used the acclimatization pen to move and hunt into.



Figure 3. Baya and Wari, during their interactions in early January 2007.

Monitoring gene flow. This genetic aspect of the project is monitored through two kinds of efforts: sampling of mixed-origin cubs for paternity tests, and identifying individual

lynxes via scat collection. The latter method could become an effective and non-invasive way of studying gene flow throughout the Doñana population.

Conclusions

Six months after its release, *Baya* was considered successfully settled in *Coto del Rey* since it has never dispersed out this nucleus. During the 2008 breeding season, *Baya* mated with the three adult females of the nucleus, although it did not with a 2-year-old subadult one. Three out of the eight crossbred cubs (J.A. Godoy, unpublished data.) born in 2008 have already reached dispersal age (see figure 4). We consider that this first stage of the re-stocking project, which involved ensuring the settlement of a Sierra Morena male in Doñana, has been successful. More time is needed to assess the genetic impact of this management action in Doñana at a population level.

Acknowledgements

J. M. Gil made the selection of the Sierra Morena lynx and proceeded with captures together with R. Arenas. N. Viqueira and M. J. Pérez, performed the pre-release sanitary evaluations. The *Ex-situ* Conservation Program team gave ideas about management and took care of *Baya* during the quarantine. The Doñana's Iberian lynx monitoring LIFE project team (D. Palacios, J.M. Martín, J.A. Báñez, R. Sanabria and R.B. Millán) helped with the surveillance and post-release monitoring. The rest of the LIFE project team (E. Rojas, G. Valenzuela, G. Garrote, M.A. Díaz-Portero, A. Leiva, J. Rodríguez, J. Bueno, S. Lillo, J. Pérez, M.I. García, M. Moral, B. Torralba, S. Saldaña, M.P. Delgado, A. Álvarez, I. Martín, R. García, I. Tenorio and M. Vara) is making all these work possible.

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Figure 4. One of the three wildborn crossbred cubs (male from Sierra Morena x female from Doñana) that reached the dispersing age in Doñana during 2008. (Photo: Gema Ruiz).

Block D: “Reintroduction: People and organizations”

Participatory planning, public involvement and communication



Human attitude survey in potential Iberian lynx reintroduction areas in Sierra Morena

Regina Lafuente

Instituto de Estudios Sociales Avanzados/CSIC. rlafuente@iesa.csic.es

c/ Campo Santo de los Mártires, 7.14004. Córdoba

Introduction

The success of the Iberian lynx reintroduction in areas historically inhabited by this species depends, to a great extent, on the collaboration of the people living in those areas. A reintroduction project must avoid, or at least preview, the possible conflicts of interests that can stem from the practice of certain conservation measures. The study "Public Opinion about the Iberian Lynx in the Possible Reintroduction Areas in Andalusia" was set in motion with this criterion. This study was developed by the Institute for Advanced Social Studies of Andalusia (IESA/CSIC) by request of the Environment Regional Ministry of the *Junta de Andalucía*, and it is part of the preparatory actions of the 06NAT/00209 LIFE project "Conservation and Reintroduction of the Iberian lynx in Andalusia".

The selection of the three studied areas (*Guarrizas* in *Jaén* and *Guadalmellato* and *Hornachuelos* in *Córdoba*), as well as the localities included in each one, was developed by the Andalusian Environment Regional Ministry through the action A.1 of the abovementioned LIFE project: "Selection of the reintroduction area", attending to the distance of these localities to the areas where reintroduction will be carried out.

The main goal of this research was to evaluate the social support for this project in every one of the pre-selected areas. Both, the support of the general population and that one coming from the hunting sector were independently evaluated. In the first part of the study, social perceptions, attitudes, values and opinions about the Iberian lynx reintroduction project were analyzed. In the second part, the results of the opinion poll conducted on the hunters living in the three pre-selected areas were presented. The Iberian lynx LIFE conservation project is especially interested in collaborating with the hunters, since both the right use of

the hunting lands and the right hunting practices may contribute to the improvement and the conservation of the habitat of the endangered species. In the third stage, once ascertained the large social support for the reintroduction of the Iberian lynx, the research was into the opinions of those social collectives that could be affected, in one way or another, by the Iberian lynx reintroduction project.

Methods

The design of this research considered the development of two different methodologies to analyse the information provided by the different social sectors: (1) quantitative method (two first stages) and (2) qualitative method (third stage).

1st stage (general public survey): The sample size is 1200 people older than 16 years-old (400 individuals in each region, according to sex and age quotas). The mean length of every telephonic interview is 15 minutes. The error margin is ± 2.89 % for the global data, and ± 5 % for the regions, with a confidence level of 95.5 %.

2nd stage (hunters survey): The sample size is 600 people with firearm license (200 individuals in each region). The mean length of every telephonic interview is 20 minutes. The error margin is ± 3.87 % for the global data, and ± 6.86 % for the regions, with a confidence level of 95.5 %.

3rd stage (representatives of social collectives survey): Hunting collectives, farmers, fishers, tourist sector, educative community, ecologists, etc. were selected. The mean length of every telephonic interview is one hour.

Most relevant results

1. General public: The attitude to the Iberian lynx is very positive (see figure 1). The great

majority considers that its conservation is important (1) to future generations, (2) to preserve the nature, (3) to control populations of other carnivores, and (4) as a tourist attraction (see table 1). In general, it is not believed that the reintroduction of the Iberian lynx is detrimental to the hunting nor to the rabbit populations. Nine out of every ten polled would support a future reintroduction of the Iberian lynx in their region. The main reasons put forward by them are related to the value of the region's natural patrimony ("it is an autochthonous animal", it has existed in the region historically", etc), to the intrinsic value of the species ("it is a beautiful animal", it is a bequest for future generations", etc), and to the ecologic value of the conservation of the species in general.

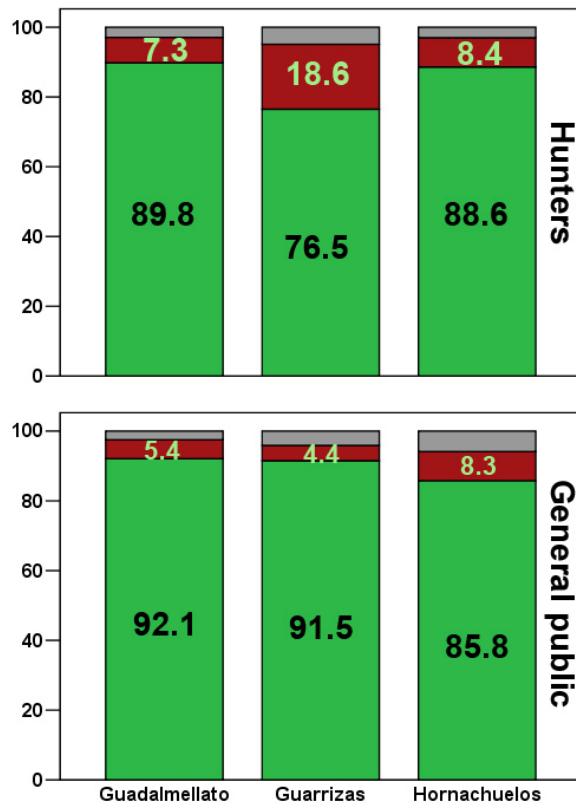


Figure 1. Percentage of agreement with the Iberian lynx reintroduction project in the three pre-selected areas both in the hunting sector and in the general public (agree = green; disagree = dark red; no response = grey).

The reintroduction of the Iberian lynx in areas of historic distribution is considered as "very important" by two out of every three polled. The majority of those polled attached more importance to the economic value that the

recovery of animal species may grant Andalusia (mainly tourist attraction) than to its possible negative effects (hunting or farming limitations). The surveyed population clearly perceives that the Iberian lynx is a much endangered species, and there is a generalized optimistic attitude to the avoidance of its extinction in all three pre-selected areas. Most of the surveyed people are in favour of the execution of all the efforts that are necessary to protect the Iberian lynx, even though it is expensive and it entails to have fewer resources for other actuations. To the majority of the polled, deforestation and lack of water are the main threats for the conservation of the animal species in general. Roads, illegal hunting methods and hunting were the following aspects, but they were far from the two first ones. These aspects are less highlighted when the surveyed people refer to their own town reality. According to the polled in all three areas, care of the country was the most important measure to protect the Iberian lynx, followed by the prohibition of the use of leg holds and foot snares. Public awareness and information campaigns, as well as wild rabbit restocking, road adaptations and reduction of hunting periods are also considered very important by a very high percentage of the population. The majority of polled said they do not know that there were lynxes in their region in the past. Also, a great majority of people polled in all three areas considered necessary to increase the number of protected natural spaces in Andalusia.

2. Hunters: Most hunters would agree with the reintroduction of the Iberian lynx in their region (88.6% hunters living in *Hornachuelos*, 89.9% of those living in *Guadalmellato* and 76.5% of those living in *Guarrizas*) (see Figure 1).

The percentage of hunters polled in *Guadalmellato* and *Hornachuelos* that have pointed out at least one benefit for the hunting derived from the Iberian lynx reintroduction project is higher than the percentage of those who pointed any trouble out, whereas in *Guarrizas*, the percentage of hunters in both groups is quite similar. However, hunters showed a higher difficulty than the general public in identifying the benefits that the Iberian lynx reintroduction could entail. In hunters' opinion, several aspects of the Iberian lynx reintroduction project would contribute to solve important problems

Reasons in favour	HORNACHUELOS (N = 343)	GUADALMELLATO (N = 368)	GUARRIZAS (N = 365)
It is an autochthonous animal	15.70%	19.90%	18.70%
To preserve the species	15.80%	11%	13.90%
General pro-environment values	12.50%	12.30%	9.20%
It is a beautiful animal	10.60%	8.60%	9.50%
It would improve the situation of the country and the landscape	8.70%	9.40%	7.60%
It holds the ecosystem's natural balance	6.20%	10.20%	7.90%
To watch lynxes in my region	7.90%	6.50%	6.20%
It would increase the tourist interest of the area	6%	4.10%	7.90%
The habitat in my region is adequate	7.30%	4.80%	5.10%
It is not a damaging animal	3.10%	3.50%	4.60%
The lynx is a legacy for future generations	2%	2.10%	1.30%
Others	2.30%	3.50%	5.70%
No response	2%	4%	2.40%

Table 1. Percentage of agreement among the general public with several statements in favour of the reintroduction of the Iberian lynx in the three surveyed areas.

of the hunting sector. Thus, rabbit restocking would alleviate the scarcity of shooting species and an increase of the surveillance would contribute to control poachers.

Among the troubles that some polled hunters related to the reintroduction project, those which contradict some benefits identified by other hunters are remarkable (mainly decrease in the number of rabbits and increase in surveillance). There is a group of hunters that relate the Iberian lynx reintroduction to hunting or shooting restrictions in different levels. Regarding the perception of the factors that threaten the Iberian lynx, hunters of all three areas agree in pointing rabbit scarcity out as the main threat, followed by roads and, in lesser magnitude, by illegal hunting methods. Hunters consider that expansion of urbanization and abandonment of the countryside are greater threats to the lynx than spare time activities and expansion of olive tree plantations. Younger hunters are in general more sensible to the threats the

Iberian lynx can suffer due to habitat changes or illegal hunting methods (see table 2).

Hunters show the same optimistic attitude to the possibility of avoiding the Iberian lynx extinction as that showed by the general public of those areas. Three out of every four polled hunters consider both preserving the Mediterranean scrubland and restocking rabbits as "very important". The third measure chosen by its efficacy level is the prohibition of leg holds and snares. Nevertheless, the support level expressed by hunters to every measure focused on protecting the Iberian lynx is remarkably lower than that expressed by the general public.

Check these web pages for further information:

www.iesa.csic.es

www.juntadeandalucia.es/medioambiente/LIFE_lince

www.lifelince.org

Benefits derived from the Iberian lynx reintroduction	HORNACHUELOS	GUADALMELLATO	GUARRIZAS
	(N = 112)	(N = 111)	(N = 105)
Rabbit restocking	18.7%	18%	16.2%
Recovery of the species in the area	10.3%	3.9%	7.8%
Control of other predators	4.9%	6.8%	5.9%
Natural ecosystem's balance	5.9%	5.9%	2.5%
Increase surveillance, control of poachers	4.9%	5.4%	4.9%
Improvement of the shooting prey quality	4.9%	3.4%	5.9%
Care and conservation of the habitat	1.5%	4.4%	3.4%
Help the shooting, in general	2.5%	2.4%	2.9%
Increase the tourist attraction	1%	2%	1.5%
Others	0.5%	2%	0.5%
Troubles derived from the Iberian lynx reintroduction	(N = 77)	(N = 87)	(N = 97)
Decrease in number of rabbits	15.8%	18%	18.1%
Increase in surveillance	3.4%	6.8%	3.9%
Shooting prohibition	2.00%	3.9%	7.4%
Shooting period or surface restrictions	3.4%	4.4%	5.4%
Hunting restrictions, in general	3.9%	1%	2.9%
Increase in poaching	2%	2%	2%
Attacking livestock risk	2%	2.4%	1.5%
It demands an increase in the caution	1%	1.5%	1%
Road kills and accident risk	1.5%	0%	1.5%
Others	3%	2.4%	3.9%

Table 2. Percentage of agreement among hunters with different potential benefits and troubles derived from the Iberian lynx reintroduction in each one of the three surveyed areas.

Communication program in the Iberian lynx reintroduction areas in Andalusia: Communicating, our eternal problem

Silvia Saldaña¹ and Miguel A. simón²

1. Iberian lynx LIFE conservation project. EGMASA.
c/ Pepe Espaliú, 2. 14004. Córdoba.

2. Iberian lynx LIFE conservation project. Consejería de Medio Ambiente, Junta de Andalucía.
c/ Fuente del Serbo, 3. 23071. Jaén.

Communication

An overview of the draft copy of the Communication program in the Iberian lynx reintroduction areas in Andalusia is presented here.

Reintroduction and recovery programs of threatened species are unlikely to succeed if they do not consider and actively incorporate values, attitude, behaviour and wishes of the local community (Reading & Kellert, 1993). Communication with the people involved in conservation projects is therefore essential, especially that with local actors living in the territory where these projects are going to be carried out.

In cases like this Life Nature project (Conservation and reintroduction Iberian lynx in Andalusia), there is a great variety of actors: land owners, hunters, various administrations, general public, international public, mass media, tourists, NGOs. This fact added to the challenge that reintroduction itself entails, shows how complex the relationships among all implied people can be.

So that, making a sociologic study in three potential areas (*Hornachuelos* and *Guadalmellato* in *Córdoba* and *Guarrizas* in *Jaén*) arose in order to select together with habitat data, the best area for reintroduction. The Institute of Social Advanced Studies (IESA) was entrusted with this work, which allowed knowing about the attitude of the local population towards the Iberian lynx reintroduction by means of telephonic surveys.

Once the future areas for reintroduction were selected (*Guadalmellato* in *Córdoba* and *Guarrizas* in *Jaén*), the results of this study came

also in useful for preparing the draft copy of the "Program for Environmental Communication in the Reintroduction Areas".



Figure 1. Front cover of the Program for Environmental Communication in the Reintroduction Areas

This program is part of a general "Environmental Communication Plan", and it is structured into two great blocks: (1) Spreading inside the reintroduction area, that is to say in the localities with municipal district inside the

reintroduction area, and (2) Spreading outside the reintroduction areas, that is both in the areas with current lynx occurrence and in other national and international areas. Both blocks themselves are organized into three sections according to the sort of action: (1) Communication actuations, (2) Awareness actuations and (3) Participation actuations.

Likewise, the general objectives of the block 1 are highlighted: Spreading inside the reintroduction area,

- **To facilitate communication pathways** between the groups directly implied in the reintroduction project and the headship of the project.
- **To attend** proposals, claims and other suggestions of the local population.
- To pass on the importance of reintroduction as a **conservation tool**.
- To promote the **participation** of the local population and *vice versa*.

And those of the block 2: Spreading outside the reintroduction areas,

- **To inform** the population about the reasons of the reintroduction of the Iberian lynx.
- To announce the **selection process** of the reintroduction area.
- To transmit the needing of **conservation of our environment**,

the Mediterranean scrubland, using the lynx as a standard bearer.

- To open **collaboration lines and systems**.
- To achieve a **good international projection**.

The following issues are some examples of the corresponding actions:

- Travelling exposition "Our Great Cat", together with complementary elements such as scale models, camera traps, documentation, etc.
- Specific awareness and information conferences focused on different groups: Teachers of Doñana and Sierra Morena, administration, land owners.
- Layout and updating of information points called "*InfoLince*" in the areas current lynx occurrence and reintroduction areas.
- Celebration of special events in collaboration with populations (i.e. World's Environment Day June 5, 2008 in *La Carolina*).
- Annual volunteering in Sierra Morena and Doñana.

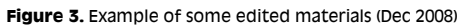
Moreover, general and specific materials about the lynx and the reintroduction have been or are close to be edited (Fig. 2).

MATERIALS	PUBLIC
Re-edition of the teaching poster "The Iberian lynx's world"	General public
Iberian lynx stick	General public
Re-edition of the leaflet "More lynx, more hunting"	Hunters
Good practices leaflets for visitors of lynx areas	Tourists, visitors
Leaflet about the Iberian lynx reintroduction	General public
Leaflet "FAQ" about the Iberian lynx reintroduction	General public
Leaflets and panels about the relationship between the farming sector and the conservation of the Iberian lynx.	Agricultural and stockbreeding sectors
PowerPoint presentations adapted to the different publics	Schoolchildren, farmers, administration
Teaching booklets	Schoolchildren

Figure 2. General and specific materials about the Iberian lynx and its reintroduction edited by the Iberian lynx LIFE conservation project

torralba, G. López, M.P. Delgado, R. García, I. Tenorio and M. Vara) are making all this work possible.

Reading, RP. And Kellert, SR. 1993. Attitudes Toward a Proposed Reintroduction of Black-Footed Ferrets (*Mustela nigripes*). Conservation Biology 7: 569-580



Block E: “Working groups: Conclusions”



Group 1: Strategy and mid- and long-term objectives

GOAL

"Advice objectives based on the Spanish National Strategy and define aims for the reintroduction"

VISION

All necessary works for the first reintroduction, as well as the selection of the places for the following reintroductions through the Iberian Peninsula must be performed within the framework of the Spanish National Strategy.

SUMMARY RECOMMENDATIONS:

Planning

1. It is necessary and also urgent to approve regional recovery plans in those communities which lacks them, and also in Portugal. These plans must have concrete goals and an agenda.
2. An Iberian plan based on the Spanish National Strategy must be approved to impulse and coordinate all reintroduction initiatives. Besides the selected sites in Andalusia, the identification and preparation of other potential areas must go on and be accelerated.

Coordination

3. Iberian coordination must be reinforced by means of the existing working group, through the designation of an external advisor who must speed up the most urgent works.

In situ conservation and reintroduction

4. All studies and actions that are necessary to preserve and expand current breeding nuclei must be continued and reinforced with priority.
5. The reintroduction action in Guadalmellato is considered as the reinforcement of the Sierra Morena meta-population, and is able to be started in the breeding season 2009/10.
6. Before beginning this activity in Guadalmellato, it is necessary to rely on a better definition of some aspects of the reintroduction plan. Also, it is necessary both to rely on some information elements (PHVA, habitat and population viability analysis) and to reinforce in situ preparation policies (Agreements with owners, information and public awareness).

7. Although wild individuals have shown better results in previous carnivore reintroductions, it is necessary to combine the release of animals coming from both populations. Regarding the individuals extracted from the field, it is necessary to carefully evaluate the effect of the extractions over the donor population. Animals bred in captivity will be released up to 2010, once they receive a proper training.
8. The reintroduction in Guarrizas or other sites must not begin until the goal of reaching 6 breeding females, and after a careful evaluation of the results.
9. The instruments for general public information must be reinforced, mainly concerning to the foreseeable death of some released individuals. The information must be especially fluid among the different actors of the project, including the neighbouring Communities. To achieve this, effective information and communication mechanisms must be established.

Ex-situ conservation

10. The *ex-situ* program must be completed as soon as possible by being provided with means and techniques necessary to the adaptation of captive bred individuals to a release in the wild.

Group 2: Rehabilitation and release protocols*

GOAL

Define short-term goals for Iberian lynx reintroduction and delineate pre-release training protocols.

(During the plenary session it was decided that this Group would also address short-term reintroduction goals. This change largely influenced the group's discussion)*

VISION

The first Iberian lynx releases must be oriented towards learning and shall be carried out with extreme care in order to ensure success and to help reinforce social support for future reintroduction efforts.

SUMMARY

This Group recommends that the first Iberian lynx releases should be carefully designed to ensure an initial success that also allows learning for future reintroduction efforts. The first reintroduced lynxes should be wild-caught animals allowed to acclimate in on-site preconditioning pens. Further releases could use captive stock and also test different release methods. Reintroduction details at the individual level (eg., pre-release preparation), population level (eg., age and sex structure, etc.), reintroduction site and other levels need to be discussed in a specific workshop that will require thorough planning to maximize expert contributions. This workshop should happen in the very near future, before reintroduction efforts begin. Provided that agreements and protocols were in place, reintroduction could begin throughout 2009 to ensure that lynxes would be acclimated and settled at the release site for the 2010 reproductive season.

SUMMARY RECOMMENDATIONS:

1. To maximize success in the first reintroduction it is recommended to use wild-born Iberian lynxes acclimated to the release site in preconditioning pens. Careful monitoring and control of all reintroduced lynxes is a crucial part of the learning process.
2. Mid- and long-term, once there are lynxes settled in an area, exhaustive control can be relaxed and there should be possibilities of using simpler protocols, including hard releases.
3. The reintroduction process must follow an adaptive management approach in all regards, including the design of goals, the development of viability studies, as well as adaptations in the design and implementation of protocols. First releases should be considered experimental, designed to learn and refine techniques for future reintroduction efforts. Therefore, viability studies for the extraction of individuals from the donor population are not as critical in this initial experimental release as it will be in future releases.

4. To evaluate the specific details of the document "Plan for the Reintroduction of the Iberian Lynx in Andalusia" is a task that cannot be tackled in the short time dedicated for discussion in this Working Group. The Group recommendation is to organize a meeting in the near future to discuss specific details (at the individual, population, site and other levels, respectively).
5. This workshop should be planned in advance and participants should be carefully selected to ensure that each person can contribute important and complementary aspects to the discussion. Specific questions regarding planning for reintroduction should be circulated in advance among participants in order to give each person time to think about the issues and prepare information to discuss during the meeting. The scheme could include –but not be limited to- questions regarding (1) Individual adaptation (hunting skills, avoidance of humans, etc.); (2) Spatial and social population structure (number, age, sex ratio, releases in time and space); (3) Release site characteristics (habitat quality, prey quality, anthropogenic threats), among other questions. Aspects related to viability studies (both in the donor population as well as at the reintroduction site) will be important issues to discuss and define during this workshop.
6. Independently of specific planning details, this Group recommends that the first experimental releases aim at learning how to settle the lynxes at the reintroduction site, as well as how to achieve reproduction as soon as the reintroduced animals are mature enough to breed.
7. It is considered premature to begin reintroductions in February 2009; yet, it is recommended to commence later, throughout 2009 –once all important planning details are in place-- so the reintroduced lynxes could be settled at the release site for the 2010 reproductive season. For this purpose, the Group recommends to start the construction of release enclosures as soon as possible so they can be ready to hold lynxes within the near future.
8. It is important to use the experiences in Sierra Morena as an example of what can happen at the reintroduction site. It is recommended to take into account dispersing individuals as possible reintroduction candidates. Therefore, it is important to radio-tag as many potentially dispersing juveniles from the Sierra Morena population and to conduct a thorough monitoring of these individuals.

Group 3: Post-release monitoring

GOAL

"To define both systems and monitoring periods of the released lynxes, as well as the control mechanisms for all the possible mortality causes."

VISION

Lynx releases must be controlled in order to obtain the maximum information, which will be applicable to next reintroductions.

SUMMARY RECOMMENDATIONS:

Coordination

1. It is considered necessary that Andalusia foster its relationships with Castilla-La Mancha in order to increase the implication of the second one on the Iberian lynx conservation. The multilateral commission must be promoted to achieve that, so we demand the Environment Ministry to lead the process.

Reintroduction monitoring:

2. The post-release monitoring objective is to allow a proper monitoring to the released individuals. It is considered as necessary to intensively monitor (10-15 locations per day) individuals, especially at night, until their hunting range is established. Once it happens, one location per day will be enough, although monitoring will be more intensive during breeding season. An management adapted to the circumstances is recommended.
3. The most sophisticated available transmitters should be used (re-programmable GPS locators which send information via GSM). Computer means to store a great information volume is needed.
4. Once home ranges are established, the same monitoring protocols as the ones used in the current populations will be applied.
5. The F1 generation will be also radio-tagged, and the rest of generations until achieve the marked objectives.

Post-release

6. A protocol to decrease mortality causes will be designed. The participation of the authority agents is essential, mainly in the border of the reintroduction areas. Especial attention will be paid to the known mortality causes.

7. It is necessary and right to adopt measures that include compensations to the lost caused by attacks to hens and livestock.
8. This process is considered as a chance to improve capture methods, so that new systems must be explored.
9. Results will be evaluated at least annually.

Financing

10. It is advised to manage and to properly plan the post-LIFE. To achieve this, it is necessary to explore new Financing sources.

Group 4: Communication plan and public awareness

GOAL

"To define systems of fluid communication among project partners, implied people, mass media, and, especially, with the people inhabiting reintroduction areas".

VISION

An obvious communication problem among implied actors is shown: generalized mistrust, lack of fluidity, transparency and credibility, a global image of the lynx is not projected (although it does of the Andalusian lynx), a clear leader to whom speak to is lacking as well as a lack of communication protocol for crisis situations and the value of communication is not appreciated.

SUMMARY RECOMMENDATIONS

To try to solve some of the problems lighted before, the next recommendations are proposed:

SMART⁽¹⁾ Objectives

Sub-group 1

1. To organize a workshop during the first trimester of 2009, with the following objectives:
 - a. To admit that communication among communities and countries is important
 - b. To mark communication protocols and to agree on the creation of a single communication department.
2. Appointment of a communication team on national level.
3. To make a global communication plan for the strategy of every country.

Sub-group 2

1. To organize by January, 2009 an intern workshop to clearly define the role and the task of every program participant.
2. A material package for media must exist by January 2009, relying on homogeneous and updated data.

3. By January 2009, an internal communication protocol must be defined, agreed on and set off.
4. To organize a seminar for the training of journalists and other groups linked to the first Iberian lynx reintroduction.

Sub-group 3

1. To organize a workshop by mid January 2009 for understanding and facing key issues in communication.
2. By the end of January, 2009, to create a position of information and communication coordinator, with experience in communication and conflict resolution, in order to facilitate communication and create transparency and trust.
3. Team building organized by an external company during a weekend by the end of February 2009, in order to improve trust, communication and relationships among project participants.

⁽¹⁾ **S.M.A.R.T.:** Specifics **M**easurable **A**ttainable **R**ealistic **T**imed

Block F: “Iberian lynx reintroduction plan in Andalusia”



Iberian lynx reintroduction plan in Andalusia

Miguel A. simón¹, Urs Breitenmoser², Astrid Vargas³, Chistine Breitenmoser², José M. Gil-Sánchez⁴, Manuela Von Arx², Fridolin Zimmermann², Andreas Ryser², Marie-Pierre Ryser-Degiorgis⁵, Gema Ruiz⁶, Guillermo López⁷ and José García⁴.

1. Iberian lynx LIFE conservation project. Consejería de Medio Ambiente, Junta de Andalucía. c/ Fuente del Serbo, 3. 23071. Jaén. miguelangel.simon@juntadeandalucia.es
2. KORA. Thunstrasse 31. CH-3074. Muri
3. Programa de Conservación *Ex-situ* del lince ibérico. Centro de cría "El Acebuche". Parque Nacional de Doñana. 21760. Matalascañas, Huelva.
4. Iberian lynx LIFE conservation project. EGMASA. Avd. Andalucía, 104 Esc 3, 1º. 23006. Jaén.
5. Centre for Fish and Wildlife Health. Vetsuisse Faculty. University of Berne Box 8466, CH-3001 Berne.
6. Iberian lynx LIFE conservation project. EGMASA. c/ Johan Gutenberg, 1. 41092. Sevilla.
7. Iberian lynx LIFE conservation project. EGMASA. c/ Pepe Espaliú, 2. 14004. Córdoba.

Abstract

One of the principal goals of the Iberian lynx LIFE conservation project is to initiate the first reintroduction of the species in recently extinct nuclei. The previous work to select the best area for the reintroduction of the Iberian lynx in Andalusia began in 2005. The two selected areas to perform the reintroduction into are :Guadalmellato and Guarrizas valleys, both areas showing optimal habitat structure and resources abundance. A brief abstract of the previous works and a planning of the future steps in the Iberian lynx reintroduction are presented in this communication.

Introduction

The Iberian Lynx (*Lynx pardinus*) is the species of the genus *Lynx* which historically, has manifested a more limited distribution, as it is endemic to the Iberian Peninsula (Ferrer & Negro 2004). Although it was found in practically all parts of the peninsula up until the 19th century, during the 20th century numbers diminished alarmingly to the point where in the 1980s there remained a series of isolated nuclei in the mountain ranges in the south east of the peninsula (Rodríguez & Delibes, 1992; Fig. 1). During the following decade there was to be a continued decline which would continue up until 2001 with the identification of two populated nuclei for the species: Doñana and oriental Sierra Morena, with between them no more than 150 exemplars (Guzmán et al. 2004; Fig. 1). The causes that have led the species to this scenario, and which in turn have led the IUCN to catalogue the species as "critically endangered", has been the decline in the rabbit population (its basic prey) due to viral diseases, destruction of habitat (the lynx needs Mediterranean scrub) and mortality of an antropic nature (road casualties, poaching, etc.) (Rodríguez & Delibes, 1992; Guzmán et al. 2004).

The Iberian lynx is a territorial feline whose diet is composed of about 95% wild rabbit (*Oryctolagus cuniculus*) (Gil-Sánchez et al. 2006) and whose optimum habitat is a 55% cover of Mediterranean scrubland (Palomares et al. 2000). The size of the territory of the Iberian lynx is variable depending on the quality of the habitat and the abundance of resources, oscillating between 300 hectares in optimum conditions and 12.000 Ha in sub-optimal conditions. The fact that there are only two nuclei of breeding populations means that the species is extremely vulnerable. One necessary step to reduce the risk of extinction is the creation of new populations, either by natural settlement or via reintroduction.

Spanish National Strategy for the conservation of the Iberian lynx: Point 7.5 in the National Strategy refers to the increase in the number of the lynx population, and the objective set out is to achieve the presence of at least a viable breeding population in every autonomous region included in the Strategy document. This is four-stage plan. First is the "localisation and selection of the areas" which

must be done, according to UICN criteria, within the area that the species occupied in the 1980s about which there is very precise information. The second phase is the "adaptation of the reintroduction areas", which consists of a series of measures to ensure that the chosen location is fit for purpose and viable in the long term. The third stage included in the strategy is the "selection and preparation of the lynx", whereby studies are carried out to determine the origin, genetic aspects and the most suitable social structure for the founding of new breeding populations. Ideally, specimens for the programme should be taken from the wild, as far as this is possible in the donor populations. The final phase of the strategy is the execution of the reintroduction projects, setting as an objective for 2012 the inclusion of all the autonomous regions mentioned in the National Strategy document, with each region having in place a reintroduction programme with political backing and adequate evaluation at each stage of execution.

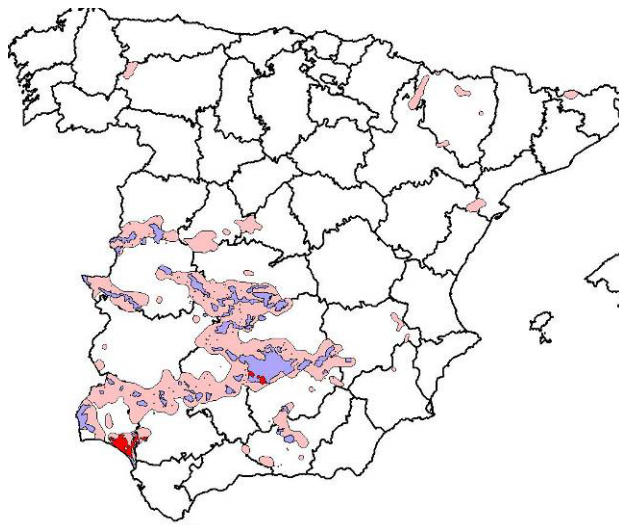


Figure 1. Distribution of the Iberian lynx in Spain in 1960 (pink), in 1990 (blue) and present day (red)., according to Rodríguez & Delibes (1992) and Guzmán et al. (2004)

LIFE Project: "Conservation and Reintroduction of the Iberian lynx in Andalusia" (LIFENAT 06/E/209): Within the group of actions for conservation of the species included in this project is the section referring to the creation of a new breeding nucleus through reintroduction. The execution of the objective is designed based on the National Strategy document and follows directives of the UICN in reintroduction. The first stage designated in

the strategy, referring to localisation and selection of areas for reintroduction, has been based on the work "Análisis de los hábitat adecuados para el linco ibérico en Andalucía", (Analysis of adequate habitats for the Iberian lynx in Andalusia) developed within the framework of the Project LIFE "Recuperación de las poblaciones de Lince Ibérico (*Lynx pardinus*) en Andalucía" (Recuperation of the Iberian lynx populations (*Lynx pardinus*) in Andalusia which was carried out between 2002 y 2006 (LIFENAT 02/E/8609). In this work three areas in Sierra Morena were selected by a multi-criteria analysis as being most suitable to host an Iberian lynx reintroduction project. Habitat, availability of resources, the level of protection in the studied zones and the historic distribution of the species in the 1990s, were all taken into account. The resulting selection of zones was from the southern sector of the Sierra de Hornachuelos, and the environs of the rivers Guadalquivir and Guadalmellato and Guarrizas.

Identification and selection of areas

Identification of the areas of reintroduction: As already mentioned, the task of the identification of the Iberian lynx reintroduction areas in the autonomous region of Andalusia was carried out within the framework of the LIFENAT 02/E/8609 project and thereby the three aforementioned areas were identified. Nevertheless, and although this document will serve as a base for the reintroduction to be carried out during the duration of the current LIFE project, during its first year a similar task was carried out (based on a multi-criterion analysis) for the identification in the Iberian peninsular as a whole, as it was already agreed with our partner the Junta de Extremadura.

Selection of the final area for reintroduction in Andalusia: Once the optimal areas for the reintroduction of the Iberian lynx in the autonomous region of Andalusia were identified, , we proceeded with carrying out a detailed study of multiple field variables, within the LIFENAT 06/E/209 project. This study had the aim of realising a definitive selection of the best area, so several variables were taken into account such as microhabitat suitability, abundance of trophic resources, current threats to the species, connection with current lynx populations and finally the social attitude of the local population before the reintroduction of the lynx. It was determined that although there were three suitable zones

the ideal zone to carry out the reintroduction of the Iberian lynx within the framework of the up and running LIFE project is the zone surrounding the river Guadalquivir, in the province of Cordoba, as it presents a better structure of micro-habitat than the zone around the Guarrizas, which remained in second place. Hornachuelos took last place due to its smaller capacity and currently fewer possibilities for metapopulation integration. Hence, the planned reintroduction in the LIFE Project will take place in Guadalquivir, even though, as stipulated in the Project, adaptation actions will begin in parallel in Guarrizas with a view to prepare for following reintroductions which will remain dependent on the preliminary results obtained in Guadalquivir.

Adaptation of reintroduction areas

Improving the habitat: The areas Guadalquivir and Guarrizas, selected for the reintroduction, present an optimal habitat structure for the presence of the Iberian lynx without having to perform any improvements, this having been

the most consistent criteria when choosing these areas. However, within the LIFE NAT 06/E/209 project an improvement plan has been developed with the aim to increase the density of rabbits in concrete zones in each area which presents less abundance, and thus maximising capacity for the future. With this in mind execution has begun of the "Improvement plan for the habitat in the areas of reintroduction of the Iberian lynx in Andalusia" in which a series of actions such as spot clearings, the implementation of sowing and improving grazing, or the reduction of cattle pressure are included. This improvement plan also includes the area of Guarrizas as in the present project preparation of this area is included. The abundance of rabbits and then habitat structure in the zone of Guadalquivir has such positive values that failure to realise this improvement would not hamper the reintroduction of the lynx in the zone, but once completed it is hoped that capacity will have increased over and above current values.

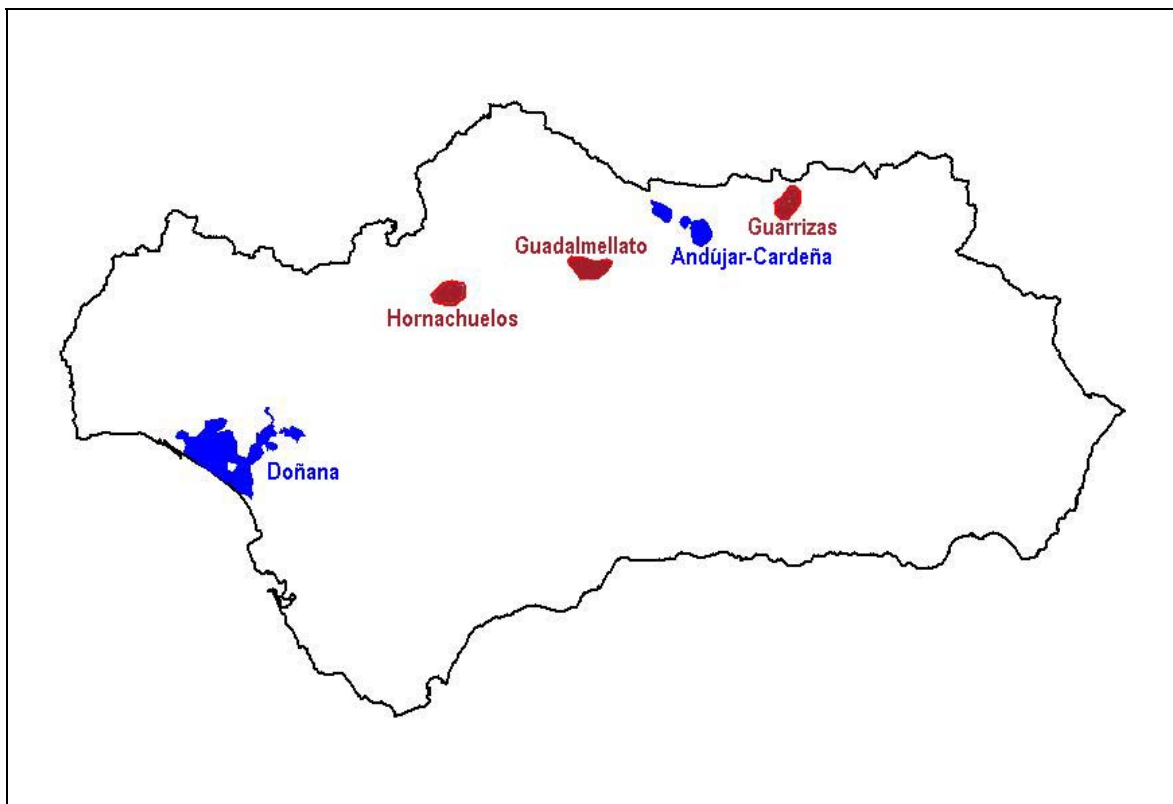


Figure 2. Schematic representation of the three areas pre-selected for the Iberian lynx reintroduction in Andalusia (in red). Current distribution range is represented in blue.

Increase the resources abundance: The areas de Guadalmellato and Guarrizas manifest adequate levels of rabbit population for the presence and reproduction of the Iberian lynx, these being higher in some zones even than in the zones presently inhabited by the species. Even so, all the actions included above in C.1 are focused on increasing the rabbit population. In the improvement plan, moreover, other actions to be executed are included such as the renting out of the rights to small game hunting in some zones, with the aim of increasing even more the abundance of resources in the zone.

Selection and preparation of specimens

Origin of the specimens: The use of wild specimens in the reintroduction of carnivores has shown to give better results than the use of individuals born in captivity (Jule et al. 2008). Thus, depending on the possibility of extraction of the species from its two subpopulations, it would be convenient to transfer wild animals as a first option, especially in the first releases (as once the first animals are settled, conspecific attraction is expected to be an important factor in the settling in of posterior releases, as has happened with other lynx species). In successive years the releases would include animals from the breeding programme in captivity, once sufficient adaptation experience of those released and taking into account genetic aspects, with the aim of creating a founding nucleus with the greatest diversity possible. Extractions of individuals from a wild population, especially from one of a critically endangered species, have intrinsic risks which should be carefully evaluated in order not to jeopardize the actual population. As the basis for evaluating these risks on the Iberian lynx populations, an evaluation of the effect of the extraction of specimens from the wild Iberian lynx populations for potential reintroductions was performed by 2001-2002, which results showed that the risk of extracting up to four cubs (two of each subpopulation) per year could be assumed reasonably (Palomares et al. 2002). This study has been followed for six years in the extractions of individuals made for the captive breeding program. Between 2002 and 2008, 36 Iberian lynxes have been extracted from the field (4 juveniles per year plus 12 individuals extracted for other reasons), and both subpopulations seem not to have suffered any negative effect because of that. Given that

2008 was the last year of extractions for the captive breeding program, these extractions can be continued for reintroductions 2009 on. That study, however, lacked data of Sierra Morena subpopulation, and conservative Doñana data were used for both subpopulations. Given that a good knowledge of the status of Sierra Morena subpopulation has been reached nowadays, a new PVA analysis is to be performed to re-evaluate the effect of the extractions. Current demography data on the Sierra Morena Iberian lynx population are much positive than expected in 2001 (and the population has been growing since that), so this study will probably much increase that value of four individuals per year. The population in Sierra Morena is apparently a suitable donor source of specimens for reintroduction. Firstly, the effect of the extractions (almost 6 individuals per year) seems not to have had any incidence on the Sierra Morena population increasing rate. Moreover, accumulated data during the last five years suggests that the population is regulated by the carrying capacity (estimated by the inter-annual availability of rabbit) and manifests indices of saturation (regulation density-dependent on reproduction, frequent aggressions, sedimentation of youngsters in suboptimal periphery areas). The population seems to have reached the carrying capacity by 2008. This situation is a consequence of the high survival levels of territorial specimens, thus, the reproductive success observed originates inter-annually an apparently surplus fraction, with the whole of the metapopulation area acting as a source area. Added to this, is a sanitary risk which comes with having the best population of Iberian lynx confined in a small space and with a high density, as the effects of an infectious epidemic could be devastating. Under this scenario, extraction of lynx individuals from Sierra Morena is going to be continued as in previous years, but the destiny of the animals will be the reintroduction. On the other hand, Doñana subpopulation has remained stable during the last decade, as well as it has suffered a dramatic decrease inside the National Park, so this population has been currently discarded as a donor until the results of this new study come out. When the new PVA is performed, a re-evaluation of the potentiality of the donor subpopulations will be carried out: the possibility of increasing the number of extractions from Sierra Morena and

of beginning the extractions from Doñana will be considered. The use of captive born individuals will permit at all times the re-adaptation of the genetic plan of the population attending to any losses that are produced.

Preparation of the specimens: Specimens from natural environments will be released via a system of soft releases, after realising protocolary quarantine. Specimens from the breeding programme in captivity should receive preparation in semi-free conditions and will only be released once their ability to survive in the natural environment is demonstrated (predatory abilities, resource search). Lynx born in captivity that are to be selected for reintroduction programmes can be identified according to genetic criteria – even before their birth and, from the first moment, they will receive a type of handling which includes minimum interaction with humans and fosters natural conditions. The age of transfer will depend on different factors, although it is recommended that the younger the better as they will have better abilities to adapt to the new environment and developed immune defences to face the dangers in the natural environment. Before transferring to the pre-release cages, lynx born in captivity will undergo a sanitary examination to ensure their good health. The time these specimens are in the pre-release cages will depend on various factors including the age of the animal (never before it is able to hunt and defend itself) and its behaviour. The infrastructures for the pre-release of animals born in captivity will be the same as those transported from their natural environment. In the medium to long term (2010 onwards) it is expected that the breeding programme will provide some 20-40 lynx per year for reintroduction programmes. The planning of the release method should include an increase in the number of fenced areas, and a decrease in their size. A large part of the design of future releases will depend on the lessons learned from the first ones. Depending on this information a comparative study could also be planned on the differences between hard release and soft release for individuals from the wild.

Execution of the reintroduction

For the elaboration of the release methodology, meetings were held with the

members of the LIFE team, with KORA, Iberian Lynx *Ex-situ* Breeding program.

Infrastructures for release: Each release unit will consist of three installations of soft release of between 4 and 6 Ha forming a 3km- sided triangle (half the average distance between Iberian lynx territories in Sierra Morena). Each installation will consist of a minimum of 40% dense scrub and at least 20% grassland. (The installations will have a minimum of 3 structures which can be used as a shelter (tree stumps, large shrubs, rocky shelters or artificial shelters), likewise two supplementary feeding stations and a water point. The installations should provide an abundance of rabbits. Because of its abundance of rabbits and good habitat structure, the first release unit will be in the estates of El Cotillo and Nava Llanas, according to the plan and whose precise location will be determined in the field.

Release procedure: A soft release method will be carried out. The annual release units will release three pairs (male and female) simultaneously per year, until the re-evaluation of the new PVA is performed. To do it, three enclosures for acclimatisation (1 pair per enclosure) will be used. Two of the pairs will be sexually mature specimens (extracted from the field when they are still not territorial) and the third will not. According to the selection study, in the framework of the current Life Project (2006/2011) releases will begin in the area of Guadalmellato and will continue with Guarrizas area as soon as possible. The intention is to set up one release unit in 2009, with the precise analysis of the results of this first release the successive releases will be planned. Two of the installations will house the reproductive pairs of Iberian lynx which will have to be introduced at least in December (one month in advance of mate season), although preferably it will be done during the autumn. The third installation will house 2 youngsters (regarding the description the two release scenarios the more conservative option was chosen, that of two juveniles of the opposite sex), which will be introduced at any moment of the year and will be liberated after two months, sufficient time, a priori, to adapt to the release site and recognition of the presence of adult lynx in enclosures. The installations for breeding will house the pair for two weeks before the birth, in which moment the male will be removed to the exterior to prevent its presence in a small

area at the moment of birth and will enable the female to move freely. Thus it is hoped that the female will serve as an element of fixation for the male and will prevent negative interaction with the cubs. The installation will be opened five weeks after the birth, sufficient time for the cubs to grow enough to avoid a critical period of fighting.

Objetives:

a) Short term: (1) 1st year, begin liberation in 2009 in Guadalmellato, (2) 2nd year, achieve stabilisation of released lynx, and (3) 3rd -4th years: begin releases in Guarrizas.

b) Medium term: (1) Achieve 15 established territorial females, (2) reach maximum genetic variability, and (3) begin releases in Hornachuelos.

c) Long term: (1) Achieve carrying capacity for 30 territorial females in Guadalmellato and Guarrizas, and (2) 15 established territorial females in Hornachuelos.

Previous information on the demographic evolution of the species: **The case of the River Yeguas nucleus.** The River Yeguas Valley nucleus of population has experienced a recuperation process between 2003- 2008, which could serve as an example to apply in areas of reintroduction for the design of a potential scenario. This nucleus has been continuously monitored by means of photo-trapping since 2003. The location of the cameras, linked to the rabbit sources (supplementary feeding stations, rabbit release enclosures, areas with high rabbit densities, etc), has allowed monitoring almost the 100% of the individuals living there along the years. All individuals in the nucleus are thus individually known, and familiar relationships were well known (maternal relationships were unambiguously known, whereas paternal were supposed according to male-female contact during mating season). This nucleus, with an average survival of territorial females of 79.9% (based in recaptures between years as an inter-annual mean value of survival rate), has increased in four years as described in the figure below. The increase came about exclusively due to the local production of cubs and was associated with an increment in the carrying capacity (probably thanks to the recuperation programme), since rabbit

population increased x3 during that period. Moreover, an important supplementary feeding program is being carried out since 2003. The dynamics of the recuperation can fit a linear or a quadratic function (see figure 3).

Application of the available information to the design of two possible scenarios

Objective: To liberate Iberian lynxes until achieving the settlement of 15 territorial females in Guadalmellato and Guarrizas, equivalent to half the carrying capacity. Once 15 territorial females occur, detain releases unless from a genetic point of view it is

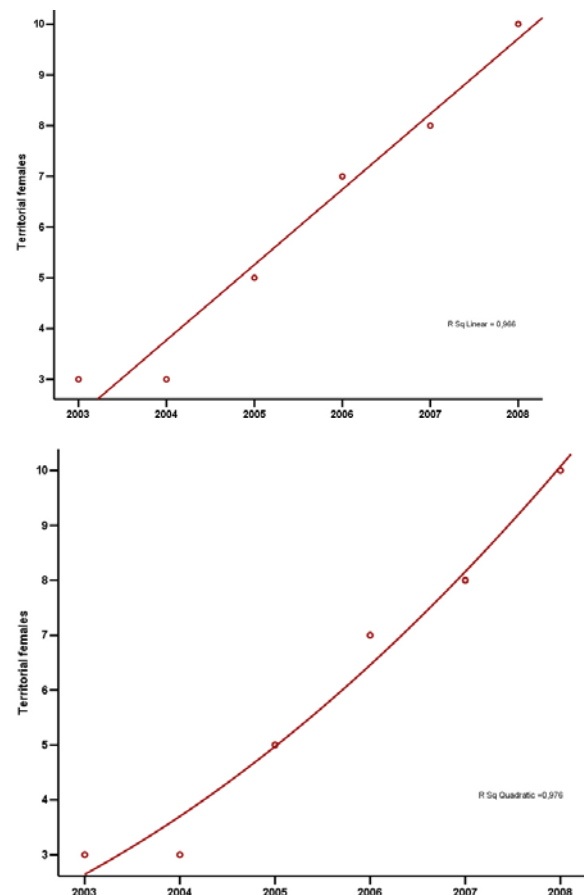


Figure 3. Linear and quadratic adjustment of the increasing trend in territorial female Iberian lynxes living in the River Yeguas Valley population nucleus

necessary to add new individuals. The justification of this objective is that it is estimated that a sufficient number of territorial females to enable natural growth and to reach capacity limit (without taking into account genetic management, which will

depend on the results of the genetic monitoring of the reintroduced population).

Scenarios of acclimatisation of specimens

- Scenario A: Survival rate and settlement of founding specimens similar to that obtained in the River Yeguas area: 79.9%.

- Scenario B: Survival rate and settlement in founding specimens 50%.

In both scenarios it is considered that the acquisition of 6 territorial females can suppose a starting point for the beginning of a population nucleus. It is considered that 6 is a prudent number since it is double the number of territorial females which began the Yeguas nucleus.

Release plan

Scenario A: Liberation of three pairs per year in the first two years, one pair per year during the following two years (these last two to compensate for fortuitous losses) and the necessary specimens to complete the genetic management. The objective of 6 founding territorial females will be reached in 2 years as settlement is expected from the first year of liberation. Also, according to the exponential growth model observed in Yeguas, once 6 territorial females have been acquired three

more years will be needed to achieve the objective of 15 territorial females.

Scenario B: Liberation of three pairs per year during the first four years, one pair per year during the following four years (these last four are to compensate for fortuitous losses) and the necessary specimens to complement the genetic management).

The objective of 6 territorial females will be achieved in 4 years (survival rate and settlement of 50%). Also, once the 6 territorial females are acquired, according to the exponential growth model observed in Yeguas, 3 more years will be needed to reach the objective of 15 territorial females. In any case, the evolution of the liberated specimens in the first two years will define the type of real scenario and subsequent provision of releases.

Spatio-temporal planning

Short to medium term: see table 1.

Long term: On the basis of the success obtained in the two selected areas in the present programme, between years 5 and 9 programmes of reintroduction could be initiated in other suitable areas in Andalusia; in particular in the southern sector of Sierra de

YEAR	GUADALMELLATO		GUARRIZAS	
	Num. Pairs		Num Pairs	
	Scenario A	Scenario B	Scenario A	Scenario B
1	3	3	-	-
2	3	3	3	3
3	1	3	3	3
4	1	3	1	3
5	Genetic management	1	1	1
6	Genetic management	1	Genetic management	1
7	Genetic management	1	Genetic management	1
8	Genetic management	1	Genetic management	1
9	Genetic management	Genetic management	Genetic management	Genetic management
...	Genetic management	Genetic management	Genetic management	Genetic management

Table 1. Short to medium goals of the Iberian lynx reintroduction program.

Hornachuelos and in the rest of the areas with a potentially apt habitat, according to the model realised with the LIFENAT 02/E/8609 project, but were rejected due to the fact that rabbit density was inferior to the selected areas, social surveys and improvement programmes would be needed until the

necessary requisites were achieved to be included as an area apt for releases.

Monitoring

Sanitary monitoring: Sanitary monitoring will be carried out both on the reintroduced lynx population and on the associated fauna which represents a sanitary risk for the lynx. The

sanitary monitoring consists of a programme of pre-release surveillance, a continuous programme of observation of risk agents and a follow up of post release of the liberated animals.

Lynx population monitoring: The monitoring of the lynx population will be carried out according to the habitual protocol of work on the LIFE project. The methods used will be photo-trap, radio tracking and tracking by indirect means. All liberated specimens will be radio tagged with GPS emitters, and during at least one week after release in the natural environment, will be subjected to 24 hour intense surveillance.

Rabbit population monitoring: The monitoring of the wild rabbit population will be carried out according to the habitual protocol set out in the LIFE project.

Public awareness

In order to carry out the correct dissemination of the reintroduction strategy of the Iberian lynx, on the one hand a sociological study was carried out by the Instituto de Estudios Sociales Avanzados (IESA-CSIC) for the selection of the best areas of reintroduction and on the other a specific communication programme for the reintroduction zones. The sociological study made highlighted the positive acceptance of the local stakeholders with regard to the Iberian lynx reintroduction programme, likewise it gave an insight into some of the weak points which could be considered among some more sensitive sectors as in the case of the hunting sector. This will help for later work on sensitisation which will be carried out. On the other hand we have a specific programme of communication which forms part of a larger plan which spans the whole communications strategy for the reintroduction and

conservation project of the Iberian lynx in Andalusia. Organised in two large blocks: (1) Communication of the reintroduction process to areas outside the reintroduction zones and (2) communication of the reintroduction process inside the reintroduction zone, a necessary tool which will help to communicate this process in an ordered and more efficient way.

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