3.2.8 Rabbit restocking operations

Rabbit haemorrhagic disease (RHD) caused severe mortalities that reduced wild rabbit numbers in most of its historical range in Spain and Portugal, specially in the ecologically less-favourable habitats. Mostly due to this disease, in Serra da Malcata rabbit density is extremely low and is a limiting factor to the success of a reintroduction plan for the Iberian lynx, Estimated density is approximately 2 rabbits per hectare and the species is absent from a significant percentage of the territory

In an attempt to solve the generalized rabbit regression problem, restocking operations are being commonly applied since the last ten years. These operations are made, not only for economic proposes (e.g. game management), but also with conservation objectives (Arthur 1989, Mauvy *et al.* 1991, Alves *et al.* 1998, Letty *et al.* 1998). In most of the above refereed studies, a significant portion of the animals suffered mortality, which was mainly concentrated during the first days of the operations. Therefore, to continued applying this sort of technique it was necessary to review restocking protocols, in order to increase success and understand ecological factors associated with low success.

According to several authors (Mauvy *et al.* 1991; Letty *et al.* 1998) is necessary to take the following factors into account:

- The importance of social behaviour and structure on the introduced population;
- Introduction area characteristics (feeding supply, weather conditions, shelter, etc.);
- Density and specific composition of the predator community;
- Dispersion patterns on the introduced populations.

It is also important to note that the patterns of rabbit distribution and abundance on the Nature Reserve (low density and isolated populations), influence the strategic option for using this technique for restoring the population levels.

Experimental protocol

From 1999 to 2000, seven restocking operations, in five geographic areas, were conducted based on the following protocol:

From 1999 to 2000, seven restocking operations were carried out in five geographic areas according to the following protocol:

1.Capture site selection:

- Respect for the subspecies of the native rabbit (*Oryctolagus cuniculus algirus*);
- Presence of a high density of rabbits that could permit high capture rates;
- Low incidence of viral pathologies.

2.Capture techniques:

- In areas with abundant warrens, rabbits were captured using ferrets (*Mustela furo*);
- In areas without warrens, rabbits were captured with the use of nets.

3.Quarantine

- Captured animals, identified with tattoos, were installed in individual boxes, weighed, and sex and age was determined;
- Rabbits were vaccinated against myxomatosis and VHD using the vaccines described in 3.2.7;
- Animals were fed with rabbit commercial food and natural vegetation;
- Quarantines lasted a minimum of 9 days;

- Mortality causes were evaluated in the UTAD Veterinarian School, using anatomopathological and histopathological tests.

4.Release procedures

- Rabbits were introduced in 120 m^2 fenced areas, with artificial warrens and a suitable natural food supply;
- Fenced areas were maintained during a minimum period of 15 days and the areas were protected against predators by using *Tomahawk*® box-traps (Tomahawk Live Trap Co, USA);
- Ten to 35 rabbits were introduced in the fenced area with a sex-ratio in favour of females.

5.Monitorisation

- Two operations were monitored by marking approximately 20% of the individuals with radio-tags;
- Five operations were monitored using a method of pellet counting as described bellow.

For the last two restocking operations, and following the recommendations of the Iberian lynx Work Group, the protocols were changed in the following aspects:

1- Increase rabbit numbers per operation to about 100;
2- Increase fenced areas to approximately 5000 m^2;
3- Base monitoring techniques on a combination of pellet mapping and camera trapping.

In Tables 4 and 5 the data of the nine restocking operations is described. After the first two actions we decided to stop using radio-tracking as the monitoring method. Although it is a more informative method, since it precisely determines the causes of mortality and survival rates, it is a highly costly method in terms of human effort and, at the same time, can produce negative effects on released animals such as dilacerations on skin and muscles (Cypher 1997, Swenson *et al.* 1999, Pérez *et al.* 2001). Therefore, we developed a monitoring method based on pellet mapping and counting, in order to reduce the effects of radio-tags.

Restocking operations

In Tables 4 and 5 the data of the nine restocking operations is described. After the first two actions we decided to stop using radio-tracking as the monitoring method. Although it is a more informative technique, since it allows to determine, precisely, the mortality causes and survival rates, is a highly costly method in terms of human effort and, at the same time, can produce negative effects on release animals such as dilacerations on skin and muscles (Cypher 1997, Swenson *et al.* 1999, Pérez *et al.* 2001). Therefore, we developed a monitoring method based on pellet mapping and counting, in order to reduce the effects of radio-tags.

Table 4 – Sites, dates and monitorization methods of the nine restocking operations conducted in Serra da Malcata during the Life project.

.

Reference	Site	Date	Monitoring method
QM0699	Quinta do Major	June 1999	Radio-tracking
QM0899	Quinta do Major	August 1999	Radio-tracking
QM0999	Quinta do Major	September 1999	Radio-tracking
CO0100	Concelhos	January 2000	Radio-tracking
VE0600	Ventosa	June2000	Pellet counting
RC0600	Ribeiro da Casinha	June 2000	Pellet counting
PO1100	Poio	Novembre 2000	Pellet counting
QU0703	Quartos	July 2003	Pellet counting + camera trapping
GI0703	Gingeiras	July 2003	Pellet counting + camera trapping

For the pellet counting method, we used a grid composed of 1 m^2 sampling units (*n*=25), placed in the neighbourhood of the fenced release area (Figure 6), so as to total a sampling area of 375 m^2. The grid characteristics and sampling methodology were based on the description methods by Forus & Humphrey (1997). Density was calculated using the following formula:

$$\delta = ([10\,000\ m^2/ha] * \mu\ ) / (\rho * T * A)$$, where:

δ = rabbit density/ha,
μ = medium number of pellets/sampling unit,
ρ = defecation rate (nº of produced pellets/24 h),
T= time interval between samples (days),
A= sampling unit area (m^2).

Using the described formula and considering that we sampled a total area of 375 m^2, we determined the absolute rabbit density on the analysed area, in five sampling occasions during a period of 4 months.

To study rabbit dispersion, we mapped the latrines in a radius of 500 m from the release point, using a GPS navigator (Garmin ® GPS 12; Garmin ® GPS 12 xl; Garmin ® GPS etrex vista; Garmin ® GPS etrex summit; Garmin ® GPS etrex venture). This procedure was applied after the 4 months period of pellet counting and repeated in time intervals of 8 to 12 months.

Data was recorded on a GPS in UTM coordinates and downloaded to a PC using the programme Arcview ® 3. To study spatial distribution we used the 95% Adaptative Kernel method (Worton 1989).

In the restocking operations monitored by radio-tracking, animals were fitted with 75 g Biotrack® radio-collars (Biotrack International, RU) with mortality sensors. Every radio-tagged rabbit was located from the ground once a day. When a mortality sign was received, the animal was located and stored for future analyses of mortality causes. Rabbits were monitored during a 2-month period since the release procedures.

Figure 5 – Geographic localization of the nine restocking operations conducted during the project. (10x10 km UTM grid)

Table 5 – Descriptive data of the nine restocking operations conducted during the project.

Reference	Rabbits origin	Released	Tagged (%)	Juveniles (%)	*Sex ratio*
QM0699	Breeding centre	10	6 (60)	0 (0)	0.67 (4 M;6 F)
QM0899	Sintra	22	8 (37)	5 (23)	0.83 (10 M;12 F)
QM0999	Sintra	21	8 (36)	4 (19)	1.62 (13 M;8 F)
CO0100	Mafra	13	5 (38)	0 (0)	0.62 (5 M;8 F)
VE0600	Infantado	16	-	4 (24)	1.14 (8 M;7 F)
RC0600	Infantado	15	-	3 (20)	1.29 (8 M;7 F)
PO1100	Infantado	34	-	6 (17)	1.13 (18 M; 16 F)
QU0703	Portel	96	-	56 (58)	0.44 (42 M;54 F)
GI0703	Portel	103	-	49 (48)	0.57(59 M;44 F)
Total		**330**	**-**	**12 (39)8**	**0.51 (167 M;163 F)**

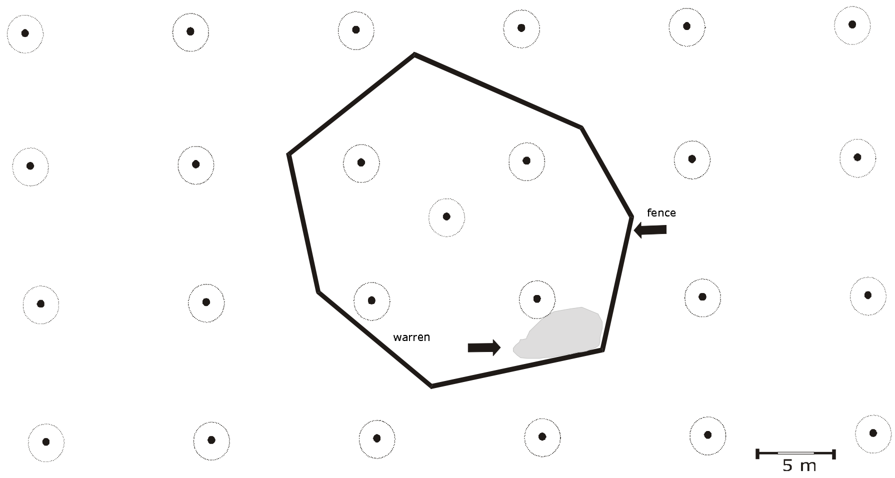

Figure 6 –Release structure and sampling units grid (*n*=25) used to monitored the VE0600, RC0600 and PO1100 restocking operations.

Results and Discussion

Restocking operations monitorized with radio-tracking

Medium survival rate in the restocking operations monitorized with radio-tracking was estimated in 60.5 %. Highest success of 100% survival was obtained for the operation QM0899,were all the animals remained in the release structure during a significant period (Table 6). Habitat suitability of the release area, which is a critical factor for the viability of any translocation operation, was probably the major factor for the high success. Low altitude and high levels food resources and shelter could had a direct influence on the survival rate.

The operation CO0100, had the lowest success of all, with 0% of survival. The null success of this restocking, was probably influenced by release area high altitude (1050 m) and by severe climatic conditions (Table 6). Mortality causes were related with physiological stress and low physical condition.

In terms of temporal evolution of survival rate, is possible to verify in the QM0699, QM0999 and CO0100 operations, the direct influence of the defensive effect of the fence (Figure 7), which, by restricting dispersion constitutes a fundamental tool for defending rabbits against predators during the sensitive period of adaptation to new environment.

Table 6 – Medium dispersion and survival rates for the rabbits released in radio-tracking monitored restocking actions a) fenced area period ; b) post-fenced area period.

Reference	Survival a)	Survival b)	Medium dispersion (m)
QM0699	5 (83 %)	4 (67 %)	50 (0-350 m)
QM0899	8 (100 %)	8 (100 %)	0
QM0999	8 (100 %)	6 (75 %)	0
CO0100	0 (0 %)	0 (0 %)	0

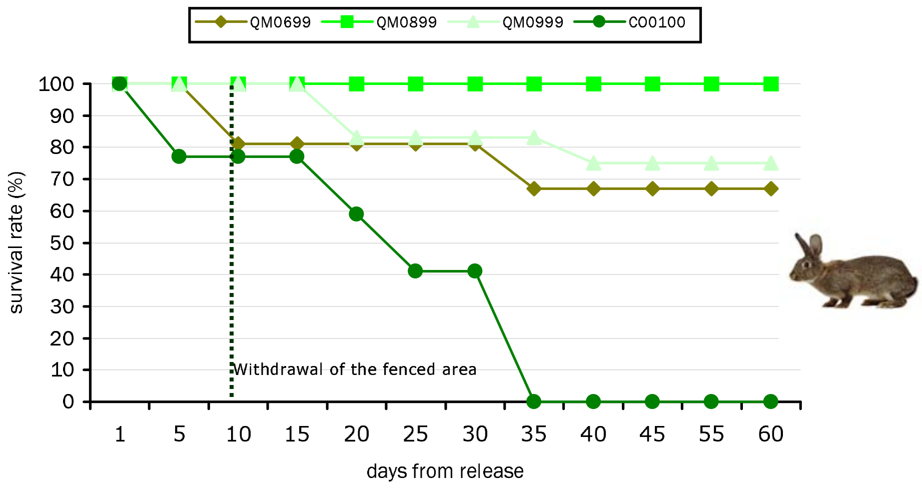

Figure 7 – Temporal evolution of rabbits survival rate on the four restocking operations, conducted during the project, which were monitored by radio-tracking.

Restocking operations monitored by the pellet counting (2000-2002)

RC0600

In restocking operation RCO600 we verified a significant decrease in rabbit density on the release site after the introduction date. Before the introduction, rabbit density was estimated at 0 animals/hectare and a total of 15 animals were released in the fenced area (Figure 8). After the fence was withdrawn density decreased gradually. This phenomenon could be originated by two factors: mortality or dispersion. We detected two cases of mortality originated by undetermined causes.

The dispersion rate of this restocking operation was very high over the following years. Presently, this area has a suitable rabbit density, estimated at 5.80 individuals per hectare, in an area of approximately 10.10 hectares (Adaptative Kernel 95%), inside the sampling circle (corresponding to 12%) (Figure 9; Table 7).

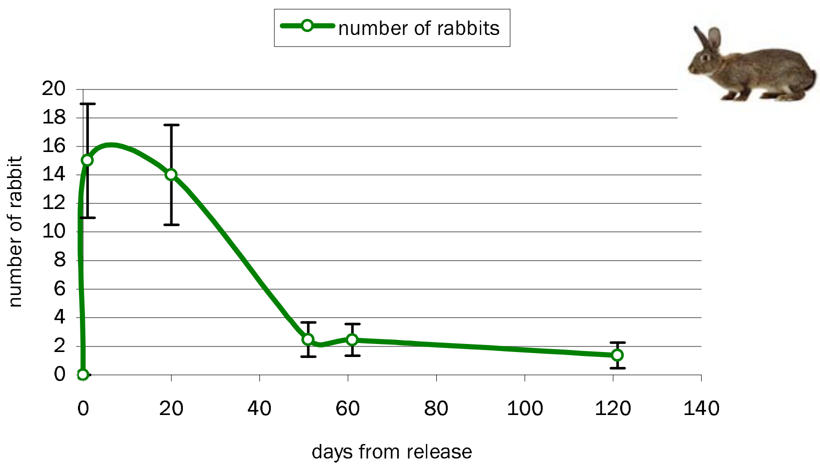

Figure 8 –Number of rabbits in the fenced area and proximities, estimated by pellet counting, during the restocking operation RQ0600.

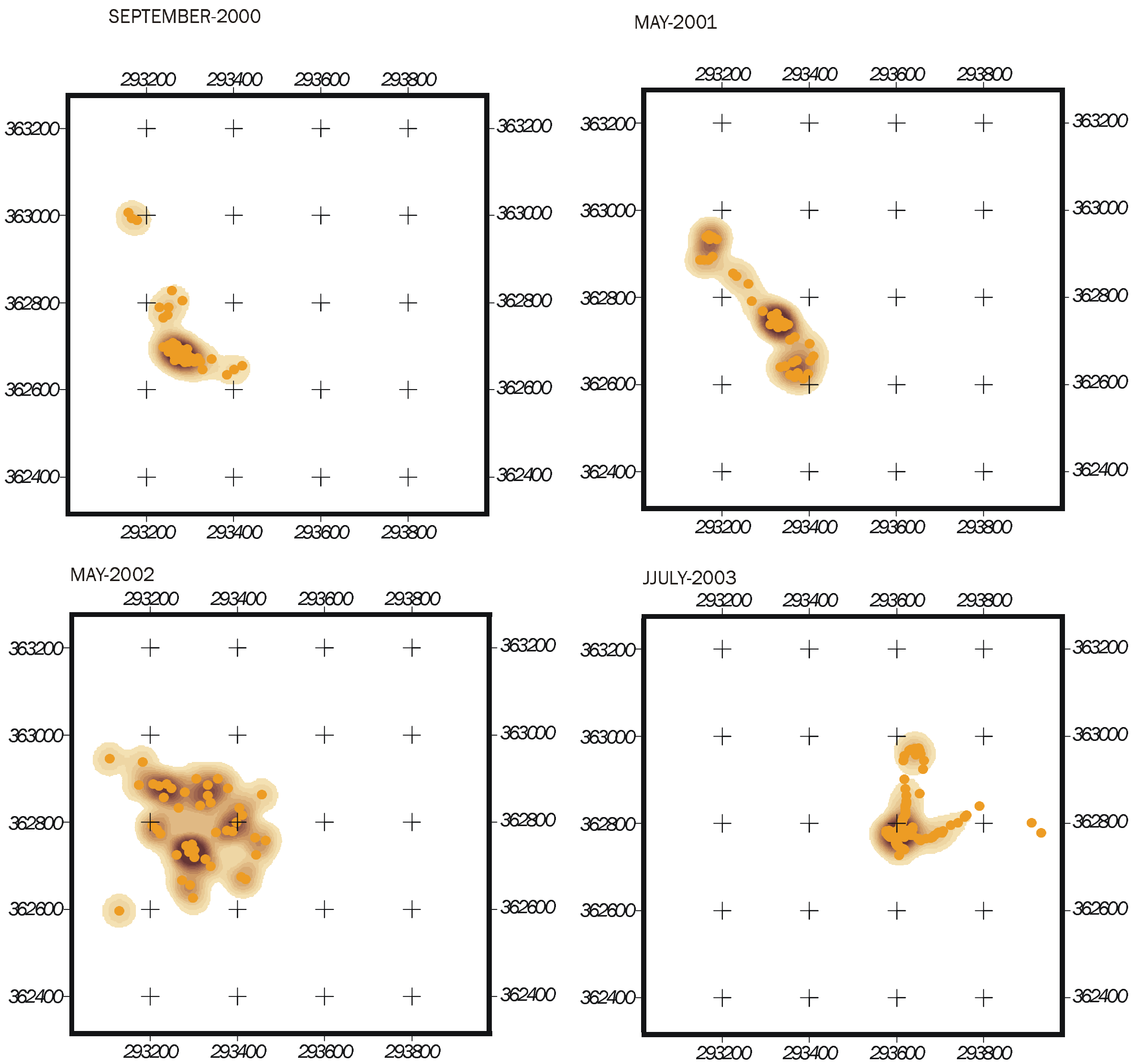

Figure 9 – Dispersion analysis of the introduced rabbit obtained for the operation RC0600. a) September 2000; b) May de 2001; c) May 2002, d) May 2003.

VE0600

During the operation VE0600, we also verify, during the pellet counting monitoring period, a gradual decrease on the rabbit density. Starting from an estimated null density, we introduced 16 animals in the fenced area, decreasing this density, after a 2 months period to a density of 5.8 rabbits on the sampled area (Figure 10). During the restriction period we observed the mortality of three individuals due to the effects of physiological stress.

Till May 2002, rabbits recolonized uniformly the release area and estimated density reached a satisfactory level of 4.11 individuals/hectare in total area of 6.29 hectares (Adaptative Kernel 95%) (Figure 11), inside the sampling circle (8%). In last monitorization, conducting in the summer of 2003, we verify that rabbits were completely absent from the area. This drastic decrease on population levels was probably caused by a VHD outbreak. Although we have no proofs than can support this explanation, we can speculate that only an epizootic phenomenon could cause the disappearance of rabbits.

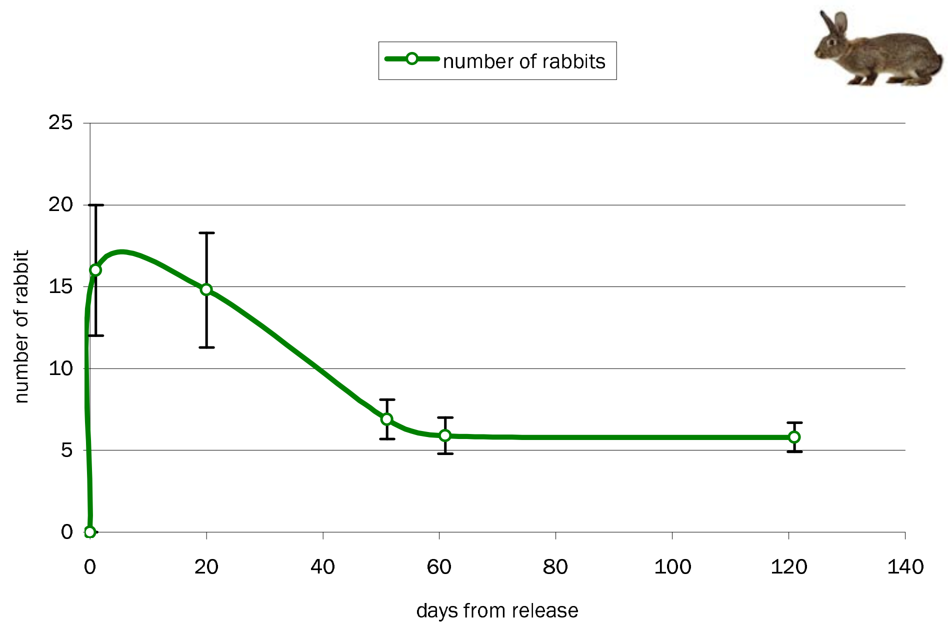

Figure 10 –Number of rabbits in the fenced area and proximities, estimated by pellet counting, during the restocking operation VE0600.

PO0100
PO0602

Due to climatic conditions it was not possible to monitor the first stage of the PO1100 restocking operation. During sequential GPS latrine mapping surveys, we analysed the dispersion and density rates of the introduced populations. Since November 2000 until now, the rates of rabbit re-colonisation of this area were quite high. In 2003, we estimated an occupied area of x km^2 (Adaptative Kernel 95%) (Figure 12) within the monitored 500 m radius area measured from the release fence. Although, we cannot assume that rabbit occupation of this area is only due to the restocking operations, it seems clear that this technique helps the re-colonisation phenomenon since rabbits were absent from that area and neighbouring populations were more than 3 km apart.

New protocol restocking operations (2003)

QUO703

The QU0703 restocking operation was conducting in an area of high density of predators. In previous surveys we capture and/or photograph wildcats, foxes (*Vulpes vulpes*), martens (*Martes foina*), genets (*Genetta genetta*) and mongooses (*Herpestes ichneumon*). In order to avoid high mortality rates due to predation, we prolong the adaptation period and so when this report was writing the area remains close and accordingly we don't have data on dispersion and post-release survival. During the adaptation period, we detected mortality due to avian predation (n=1), small carnivore predation (n=8) and physiological stress (n=3).

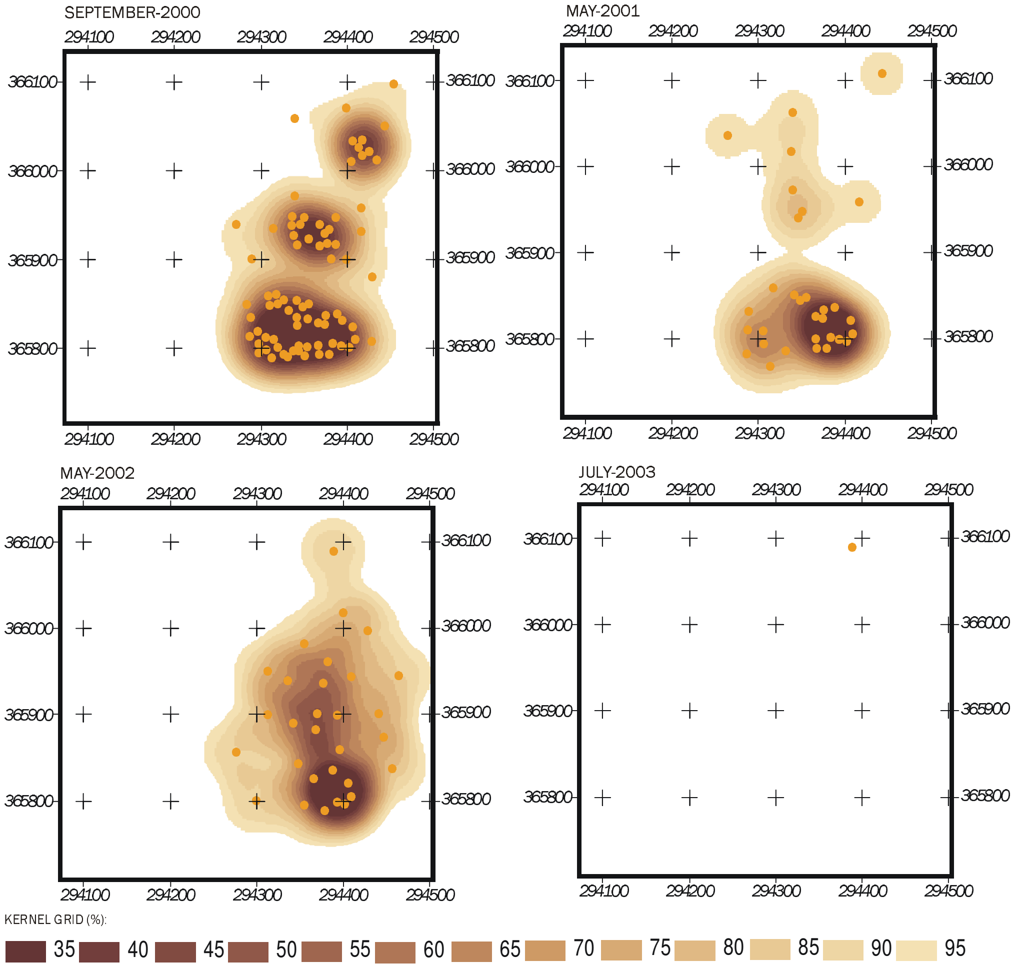

Figure 11 – Dispersion analysis of the introduced rabbit obtained for the operation VE0600.

GI0703

GI0703 operation was conducted in a high altitude plateau (approximately 1000 m), in area with low mammalian carnivore density. Vegetation is typical of Malcata high altitude areas, being dominated by extend scrubland of *Erica* spp.. Previously to release, the area was intensively managed for increasing rabbit density, by opening pastures and installing artificial warrens. Although we identified rabbit populations on the proximity, during years the area was not naturally recolonized and so, we opted to apply the restocking technique as a mean of increasing rabbit occupation and population levels. Restriction period lasted 17 days and, in the end of this period, we opened entrances on the fence, in order to promote dispersion. Carnivore presence and rabbit survival were analysed using camera-trapping. Besides the presence of booted eagles (*Hieretus pennatus*) that caused mortality in, at least, two rabbits, we also detected a marten and a resident juvenile fox, that remained in the area during the entire monitorization period. Detected mortality was related with avian predation (*n*=2), fox predation (*n*=2), physiological stress (*n*=8) and unknown causes (*n*=2) In August 2003, we analysed dispersion and a colonization area of 1,84 hectares was observed. Although preliminary results seem to be satisfactory, only future monitoring actions can clarified the potential positive result of this operation in terms of rabbit recovery.

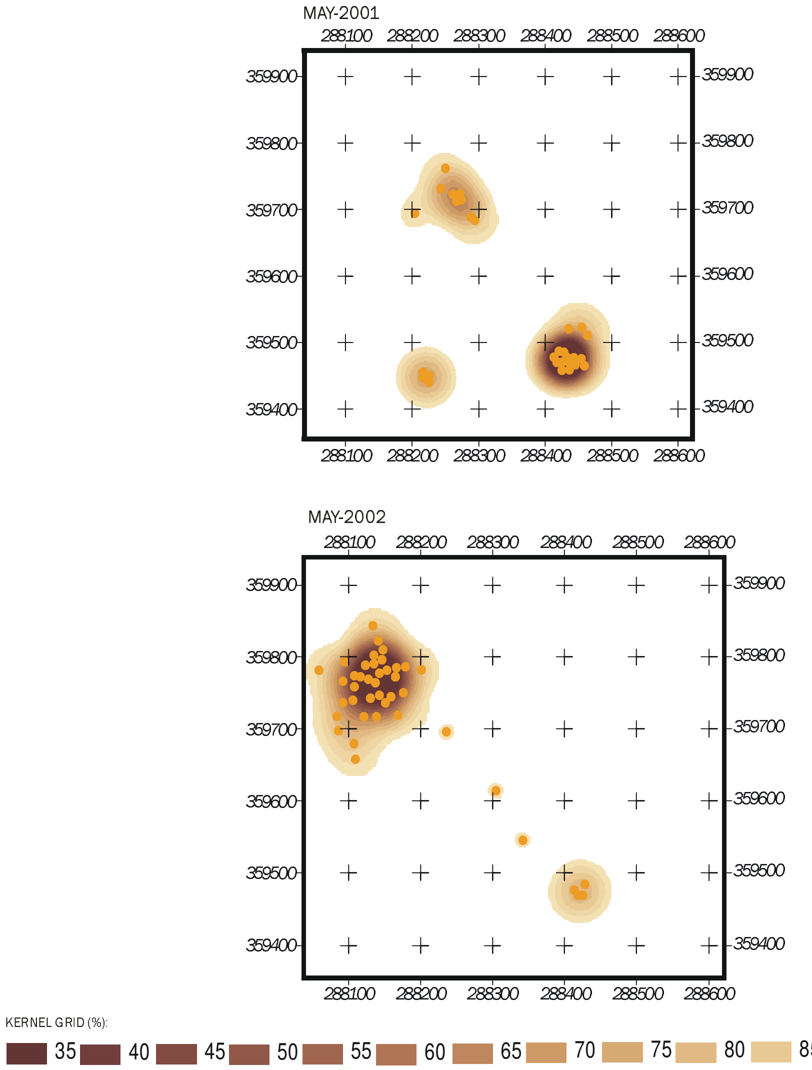

Figure 12 – Dispersion analysis of the introduced rabbit obtained for the operation PO0100 and PO0602.

▲ Rabbit release during the QU0703 operation photographed on a camera-trap

▲ Juvenile red fox on predatory activity inside the fenced area of restocking GI0703.

Mortality causes – global analysis

Rabbit mortality causes were influence by several factors, such as physiological stress, low fitness levels and, mainly, by the presence of predators (Figure 14). From a total sample of 45 deaths, we observed 17 cases of predation related mortality, caused mostly by mammalian carnivores.

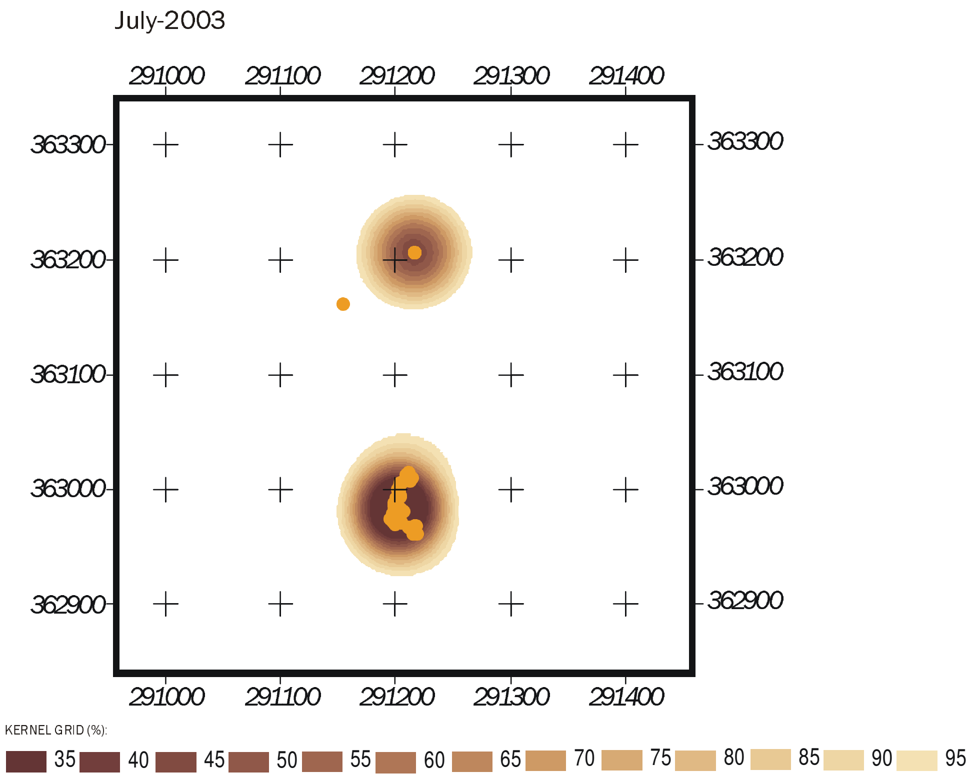

Figure 13 – Dispersion analysis of the introduced rabbits, obtained for the operation GI0703

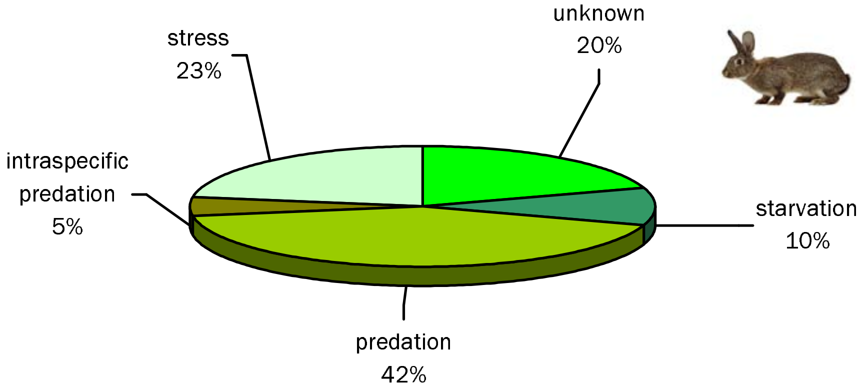

Figure 14 – Proportion of mortality causes observed during the nine restocking operation conducted during the project.

Restocking contribution for increasing rabbit density

Square 6-5 case tudy

On square 6-5, where QM0699, QM0899 and QM0999 restocking operations were carried out, it was possible to observe the effects of this management technique upon rabbit density (Figure 15). Considering that this area was submitted to habitat restoration in 1997, and since then the only management action carried out was rabbit restocking, it is possible to conclude that rabbit recovery resulted from the restocking operations.

Initially, we had an estimated rabbit density of 1.30 individuals per hectare and by the end of the project the density was 12.33 rabbits/hectare. This fact emphasises the importance of continuing restocking operations in several parts of Serra da Malcata, and the crucial part this management tool plays in rabbit recovery.

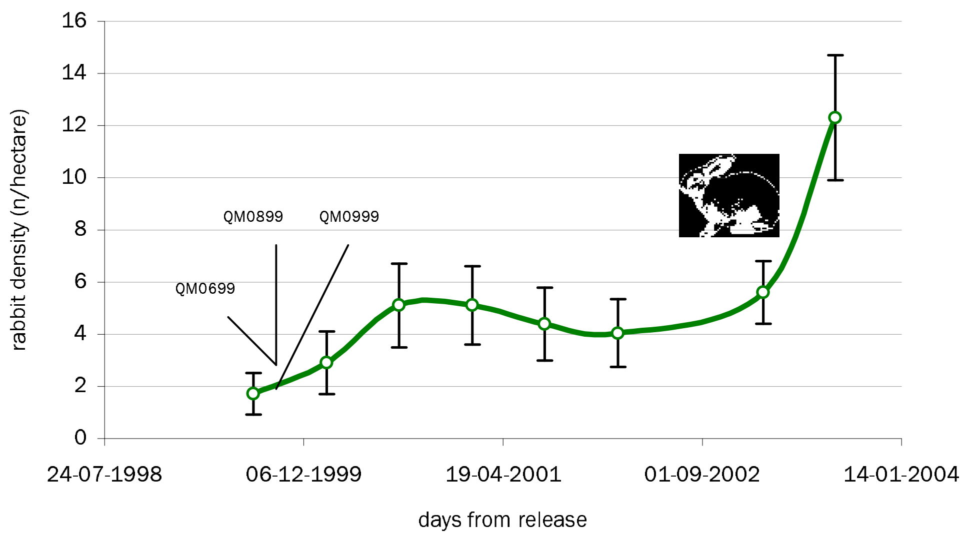

Figure 15 – Influence of the restocking operations upon the rabbit density on square 5-6 (see chapter 3.4.2 for details on the density evaluation method).

In translocation operations it is difficult to distinguish the more important success factors, since a high degree of interdependence can be observed. However, factors such as habitat quality of release areas, particularly at the food supply and shelter levels, can be crucial for achieving success. Rabbit fitness levels and seasonality are also important factors that cannot be neglected.

Using the conclusions of this project and the recommendations of the Lynx Work Group the following conclusions should be pointed out:

1. It is fundamental to restrict dispersion and eliminate predation during the acclimatisation period;
2. Operations should not be undertaken in extreme seasons (mid-summer and mid-winter);
3. Release areas should have enough food and shelter available (artificial when necessary);
4. Release numbers should be high (approximately 100 rabbits) and fenced release areas should be big enough to support them (approximately 1 hectare) during the first and most critical period.

3.2.9 Analysis of the incidence of rabbit epizootic diseases

Since 1999, it was possible to collect blood and tissue samples from 23 rabbits. These samples were submitted to ELISA tests and immunoassays analyses, with the purpose of detecting the VHD calicivirus and anti-bodies (analysis conducted in ICETA – Vairão). As a result, it was possible to detect the VHD virus in three individuals exhibiting VHD symptoms. Due to the low samples obtained, these results are not conclusive regarding the effect of this pathology on the population dynamics of Malcata rabbit populations.

▲Booted eagle preying upon a rabbit on the QU0600 restocking area.

3.3 Public awareness and dissemination of results

3.3.1 Educational actions

During the project several education actions were conducted focused on different target classes as described on Table 7

Table 7 – Descriptive data on the educational actions conducted during the project.

Action	Dates	Total hours	Target public	
Living environmental. From conception to action	11 to 13 of October 2000	15	High school teachers	Dissemination of the objectives, actions and project results. Awareness campaign on the conservation of the Iberian lynx
Environmental education	7 to 11 of June 2001	35	High school teachers	Dissemination of the objectives, actions and project results. Awareness campaign on the conservation of the Iberian lynx
Green week	29 of April to 3 of May 2002	6	All types of public	Divulgation of the Natura 2000 network. Explicit references on the project importance for the management of the Malcata area
Environmental education and conservation actions	11 to 15 of November 2002	35	ICN nature guards	Importance of environmental education for the conservation of natural resources. Dissemination of the objectives, actions and project results
Green week	28 of April to 2 of May 2993	6	All types of public	Divulgation of the Natura 2000 network. Explicit references on the project importance for the management of the Malcata area

3.3.2 Information bulletin

Initially there it was predicted the publication of 12 numbers of the information bulletin 'HabitatMalcata". However, since we are unable to have access a significant percentage of EU funding, due to bureaucratic problems, it was only possible to publish six numbers, with the information described on Table 8.

Table 7 – Descriptive data on the educational actions conducted during the project.

number	Date	Printing numbers	Contents
1	May 2000	500	Project summary. Actions and objectives
2	September 2000	1000	Conservation of Mediterranean woodland
3	December 2000	1000	Rabbit restocking
4	March 2001	1000	Serra da Malcata mammalian carnivores
5	May 2001	1000	Environmental education
6	September 2001	1000	Illegal hunting and habitat recovery

3.3.3 Exchange of information with Spanish teams

Since the project is develop in the Portuguese-Spanish border and Serra da Malcata contacts with two Spanish autonomic regions (Castilla-León and Extremadura) is quite important to maintain a permanent contact with Spanish teams involved in lynx conservation. So, during the project several meetings and field trips were conducted in order to increase the informational exchange (Table 9).

Table 8 – Descriptive data on the educational actions conducted during the project.

Data	Place	Context
9/02/2000	Soto del Real (Madrid/Spain)	First meeting of the Spanish Iberian Lynx Work Group. Actions and objectives of the project were presented
17/02/2000	Salamanca (Spain)	Seminar on the conservation of the Iberian lynx in Castilla-León
17/10/2001	Penamacor (Portugal)	Work meeting for exchanging information on Iberian lynx status and rabbit recovery. Meeting attended by Castilla-León and Extremadura lynx teams and by the Spanish Lynx Conservation Coordinator
12/12/2001	Sierra da Gata (Spain)	Field trip to Sierra da Gata to observe rabbit management actions conducted by Castilla-León and Extremadura lynx teams
14/05/2002	Madrid (Spain)	Meeting of the Spanish Iberian Lynx Work Group
12/06/2003	Acebuche (Doñana NP/Spain)	Meeting of the Spanish Iberian Lynx Work Group
23/09/2003	Penamacor (Portugal)	Work meeting for exchanging information on Iberian lynx status and rabbit recovery. Meeting attended by Castilla-León and Extremadura lynx teams.

3.3.4 Other actions

The project was also divulgated in the following congresses and seminars:

- Seminar "Reproduction and integrated science". London Zoological Society. London. November 2000. Presentation of the poster "Recovery of the habitat and preys of the Iberian lynx. EU Life project";

- Seminar "Resources management in border natural areas". Faculty of Sciences of the Lisbon University. Lisbon. March 2001. Oral presentation entitled "Recovery of habitats and preys for the conservation of the Iberian lynx in Serra da Malcata";

- II International Symposium on Wildlife". University of Trás-os-Montes e Alto-Douro. Vila Real. April 2001. Presentation of the poster "Recovery of the habitat and preys of the Iberian lynx. EU Life project";

- Agrarian School of Castelo Branco. Castelo Branco. June 2002. Presentation of the Conference "Iberian lynx: The most endangered cat species in the world".;

- International Seminar on the Iberian lynx. IUCN/CSPG;WWF, MIMAM. Andújar. Octubre 2002. Oral presentations: Status survey of Iberian lynx in Portugal". "Proposal of Action Plan for the Conservation of Iberian lynx in Portugal".

- XXVI th Congress of the International Union of Game Biologists. IUGB. Braga. September 2003. Oral presentation entitled "The conservation of the Iberian lynx in Portugal. Dilemmas and possible solutions".

The project was also disseminated in several articles of national and international press and in two TV documentaries.

3.4 Overall project operation and Monitoring

3.4.1 Study of ecological parameters of the Iberian lynx and the Carnivore community

3.4.1.1 Abundance and spatial distribution

Detecting changes in abundance on endangered species is a key issue for there conservation and for a basis of decision making in wildlife management. Because of their relatively low population sizes and elusive nature, carnivores are notoriously difficult to survey. Fortunately, sophisticated non-invasive methodologies for furthering our knowledge of carnivores are increasingly available. The cost-effective development of biochemical techniques such as DNA analyses permits accurate identification of an individual animal from remains such as scats and shed hairs. Camera-trap photography, track plates, scent stations, hair snagging, and genetic analyses can now be used in conjunction with traditional techniques -- such as snow tracking, trapping, and radio-telemetry -- to monitor carnivores. Because no one survey method is suitable for all species, we are using a suite of non-invasive techniques, found to be effective in Iberian lynx (Guzmán *et al.*, 2002) and other feline species . One important issue when sampling such a rare species as the Iberian lynx is using a method with high reliability that could detect with substantial precision presence and declines in population size. So, we decided not using sighting reports as proofs of lynx existence since this method trends to give a fictional scenario of lynx numbers (Guzmán *et al.*, 2002). Personal interviews and questionnaires most times include misidentification of lynx, considerable errors on the sighting date and place of observation.

For conducting this survey we choose a combination of methods with high reliability and that could constitute a suitable data base for future lynx conservation. These methods were:

1- Sign searching;
2- Camera trapping;
3- Box trapping.

We also made a description of observational data in order to maximize the information, although this data was not considered as reliable proofs of lynx presence.

Goals

1. To map the Iberian lynx presence and other mammalian carnivore species and explain this spatial distribution according to ecological parameters;
2. To define each carnivore population status;
3. Define target areas for the implementation of the management actions.

Sighting records

Some interviews were carried out to local people to ensure that all kind of information was included in the presence evaluation of the Iberian lynx. Evaluation criteria were applied to these sightings in order to define their degree of authenticity, and the sightings were rated in four classes (Vandel, 1997):

1. **Confirmed sighting**– it was made by the authors or by a co-worker or even by a third part if there is a conclusive proof with it (photo, footprint, scat or skin);
2. **Probable sighting** – it was made by a third part and there are no faults in his testimony;
3. **Doubtful sighting** – it was made by a third part and there are some faults in his testimony although they are not sufficient to reject the sighting;

4. **Rejected Sighting** – the testimony has many faults such as the description of some morphological aspects or behavioural different from those described for the Iberian lynx.

Snow tracking

On the 14^{th} 15^{th} and 16^{th} January, 2000, a snow tracking survey was carried out to detect any sign of the Iberian lynx and of the other carnivores. There was only enough snow in the northern areas and at the higher altitude. Becker *et al.* (1998) biometric methodology, was used to avoid double counting.

Sign searching

The sign searching methodology was conducted using a geographic framework of 2 x 2 km UTM grid squares, defined upon the study area.. The field work consisted of a search for signs of lynx and mammalian carnivores presence and was undertaken from 1999 to 2003, with two annual samplings (February and August of each year).

Focus zones

In order to augment potential results and to obtain a more coarse-grain survey we defined focus zones inside the UTM squares. Focus zones are areas of particular interest where intensive effort was conducted; they are irregular in shape and they present natural characteristics suitable for lynx, namely:

1- Presence of Mediterranean scrubland patches with areas superior to 100 hectares;
2- Potential ecological corridors between suitable patches;
3- Medium or high rabbit density

Collected data

In focus zones searching efforts were conducted on roads, trails and rocky areas since the lynx uses mostly these areas for territorial marking. Mudded areas and snow covered territories were also searched for potential tracks.

When a potential sign was detected its UTM geographic coordinates were recorded using a GPS navigator and deposition location was characterized by recording the following data, in a 20 m radium circle: 1) percentage of rocky cover; 2) percentage of grass cover; 3) percentage of shrubs cover; 4) percentage of trees cover; 5) trees species. Percentage class were defined using the following scale:

0 (0%); 1 (1-10%); 2 (11-20%); 3 (21-40%); 4 (41-60%); 5 (61-80%); 6 (81-100%)

Lynx potential scats were collected using a protocol suitable for DNA analyses, in order to determinate their specific taxonomic origin (Pires & Fernandes, 2001; Palomares *et al.*, 2000). Recent advances in DNA techniques have opened the door for more accurate assessment of lynx distribution and so in the current project we gave emphasis to this methodology in our survey. Carnivore scats were identify by shape, colour and odour by experiences field workers.

Camera-trapping

The use of camera trapping as the advantage of causing low perturbation effect in detecting cryptical animals with inconspicuous habits (Wilson, 2001;). Currently, photographic trapping is successfully used in Spain in the Iberian lynx national census and other ecological studies (Guzman, 2001; Guzman, 2002), and is gaining popularity as a suitable method for this species detection. Recent results, obtain in Spain, attribute to this technique a substantial importance on determine lynx abundance and presence, since animals can be identified by natural features (e.g., pelage characteristics) which is an evidence that camera-trapping survey methodology is reliable for the purpose at hand.

Photographic devices

Two distinct camera devices were used: pressure plate triggering device and sensor heat activated CamTracker® In the first camera system the triggering device is as pressurised plate (dimensions of 30 × 30 cm) which is connected to a simple 35 mm automatic camera (equipped with fixed great angular objective and automatic flash), through an electric wire buried in the ground. The plate is protected by a hermetic plastic bag to avoid damages to the system due to humidity. The circuit inside is opened. When an animal steps on the plate, closes the circuit and makes the camera shoot a photograph. The set device is placed inside a wooden box opened at the front to avoid direct impact by the rain and sun. The camera is set at about 15 cm height (average) above the ground and distanced 2 to 4 meters of the pressure plate. This system is equivalent to the one described by York (2001) and used in Spain by Guzman (2001; 2002).

The second photographic system is totally automatic and the photograph shots are activated by differences of intensity of heat. The sensor memorizes the temperature in the detection area and when a heat source makes the temperature vary the camera is activated and a photograph is taken. The sensor is associated to a 35 mm camera with similar characteristics to the one described above The unit is compact and is protected by a water-resistant container. The sensor was programmed to detect 24 hours/day and once activated a photograph is shot every 20 seconds until the temperature returns to normal. The attractant was distanced 2 to 4 meters from the camera.

Attractants

The attractant scent station consisted in a wooden stake with a piece of cork-tree bark attached at 40-50 cm above the ground (Guzman, 2002). The lure was sprayed on the bark.

The lure used in the scent-stations, associated to the camera-traps, was Iberian lynx urine which, according to Guzmán *et al.* (2002) is considered to be the most effective attractant for the species A scent lure was preferred instead of food-bait, in order to avoid the deterioration of the station and the attraction of non-target species (which could eat or destroy the bait-station). Another disadvantage of the use of food-bait is the possible contamination of the target species with pathogens due to bait degradation.

The Iberian-lynx urine was kindly offered by the Doñana National Park lynx team and was collected from captive specimens held in El Acebuche reproduction centre.

Box-trapping

Several methods of trapping have been applied for carnivore species such as padded foot-hold traps and snares and box-traps. Foot-hold traps have been used to capture Iberian lynx by a various number of authors (Castro, 1992; Palomares, 2001), however, because of its possibility of inducing serious injuries on the captured species (Phillips, 1996 and confirmed by the experience gained by several years of captures) its use has been forbidden for capturing wild-living animals. Therefore, although box-trapping is not an injury free method it is the safest and commonly used for capturing these kinds of felids (Palomares, 2001).

The cage-traps used were wire-cages with two guillotine doors activated by a pedal. The dimensions of the traps were approximately 180 × 80 × 80 cm. A live pigeon was used as bait and was placed at the centre of the cage-trap inside a protection box. Tomahawk® live-traps 1,14 m × 0.38 m × 0.51 m were also used with a similar bait. Traps were disguised with natural vegetation such as *Cistus ladanifer* (Maddock, 1993) and the box floor was covered with dirt and herbaceous species in order to replicate a natural ground cover. Lynx urine was also used as scent lure to increase the capture probability.

As Iberian-lynxes tend to use trails to move through their territory, the box-traps were placed in paths and trail intersections.

Traps were visited everyday at dawn to avoid the exposure of a captured animal to high temperatures and to prevent an excessive stressful situation that could result in death by capture myopathy.

Trapping efforts were only made when it was considered that there was a strong possibility of Iberian-lynx presence at that location. This possibility was evaluated by other prospecting methods such as potential excrement and foot-print collection in favourable habitat. Every trapping effort was of at least 20 capture days / trap.

Hair snares

We used the methods described by McDaniel *et al*. (2000), for estimating the limit ranges of the Canadian lynx (*Lynx canadensis*) geographic distribution. Hair snares transects had a total length of 2 km. Each station consisted of a pie pan dangling from fishing line as an attractor, a pad baited with FAS and catnip oil, and a Velcrolike patch nailed to a nearby tree to snare the hair of any rubbing lynx. Any cat hairs collected was submitted to a morphological analysis.

Results

Using the methods describe above, it was possible to determined, with a significant amount of precision, the abundance and spatial distribution patterns of mammalian carnivore species of Serra da Malcata (Table 7).

Red fox

Red fox is most abundant carnivore in Malcata, being detected in the totality of the sample squares (*n*=58) (Figure 16). During the field work, we photograph a minimum of 23 animals for a total sampling area of 60.97 km^2 (trapping polygon plus buffer area), which allows us to calculate a minimum population of 63 animals (assuming one fox per 2.65 km^2). If we assume that we were able to photograph 75% of the resident foxes, maximum density could be estimated in population a population of 95 individuals (0.56 foxes per km^2). Assuming these number, fox medium density could be estimated at 0.47 foxes/km^2 .