World Rabbit Science

World Rabbit Sci. 2017, 25: 407-414
doi:10.4995/wrs.2017.4107

WILD RABBIT RESTOCKING: SUITABLE ACCLIMATION CONDITIONS FOSTER ADAPTIVE BEHAVIOUR AND IMPROVE SURVIVAL OF CAPTIVE REARED RABBITS

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Abstract: Wild rabbit is a very important species in the Mediterranean region. Its relevance is both ecological, being a keystone species —prey for several predators— and economic, as it is the most important and the most managed small game species among mammals in the Iberian Peninsula. Although both researchers and game managers claim restocking operations have moderate to low success, it is still one of the management measures often used by conservationists and hunters to recover or boost wild rabbit populations. The high mortality usually verified in the first days after release is the main limiting factor in rabbit restocking. Based on practical research, protocols have improved, pointing out good practices and suggestions to increase success. Adopting soft instead of hard-release protocols has been proven to increase short-term survival after liberation. Aware that purchased rabbits seldom display a quick adaptation to the field, we performed two restocking experiments with different conditions (acclimation period and park size) in order to compare the outcomes in terms of survival rate. The rabbit batch that experienced a longer acclimation period in a larger park showed higher survival rates, as well as more frequent and much larger spatial movements. These results suggest that proper conditions during acclimation may contribute to the success of wild rabbit restocking operations.

Key Words: rabbit, *Oryctolagus cuniculus*, space use, game management, wildlife conservation.

INTRODUCTION

Wild rabbit is a key species in the Mediterranean region, as it is an important prey for more than 30 predators (Delibes-Mateos *et al.,* 2008a). Among them are the Iberian lynx (*Lynx pardinus*), currently classified as "Endangered", and the imperial eagle (*Aquila adalberti*), categorised with "Vulnerable" status (IUCN, 2017), whose survival depends on high-density populations of wild rabbits (Delibes and Hiraldo, 1981). Besides its relevant role in the Mediterranean food webs, the wild rabbit is also regarded as an ecosystem engineer due to the beneficial consequences of its natural presence and activity. The wild rabbit modifies vegetation through grazing, acts as a disperser for seeds of many plant species, provides refuge for other animals that use its warrens (Gálvez-Bravo *et al.,* 2009), and improves soil fertility (Willot *et al.,* 2000). In addition, in the Iberian Peninsula the wild rabbit is the most important sedentary small game in numbers of specimens hunted annually (Garrido, 2012; ANF, 2012). Game activity includes several sources of revenue, such as hunting licences, hunting-day or season fees, hunting equipment, and management measures like restocking or habitat improvement. The wild rabbit alone generates around 900 000 000 € of income every year in Spain (Villafuerte *et al.,* 1998) and makes a considerable contribution to the 365 000 000 € generated annually by hunting activities in Portugal (Paixão *et al.,* 2009). Nevertheless, rabbit populations have declined significantly in

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https://doi.org/10.4995/wrs.2017.4107

the Iberian Peninsula over the last 50 yr, and general information on the relationship between the density of rabbits and their ecological roles in ecosystems is still lacking (Ferreira, 2012). Two diseases, myxomatosis and rabbit haemorrhagic disease, acting together with human-induced habitat changes (mainly intensification of agriculture, habitat loss and fragmentation) have accelerated the decline and even caused local extinction of rabbits in many regions (Moreno and Villafuerte, 1995; Moreno *et al.,* 2007). After a few years of apparent population stabilisation, in 2013 an unusually high mortality was recorded in the Iberian Peninsula, which was attributed to a new variant of rabbit haemorrhagic disease virus (Abrantes *et al.,* 2013). This new sort of virus, against which rabbits seem to have little or no natural defences, has been concerning both hunters and conservationists.

Wild rabbit is the main prey for Iberian lynx, and its scarcity, together with habitat loss or degradation and human-caused mortality, constitute the main reasons for lynx decline (Delibes *et al.,* 2000; Ferrer and Negro, 2004; Rodríguez and Delibes, 2004). During the second half of the past century, the Iberian lynx distribution area declined by 98% (Rodríguez and Delibes, 1992; Guzmán *et al.,* 2004). The species used to be found in most parts of the Iberian Peninsula (Delibes, 1979), but is now absent in most of Spain (Guzmán *et al.,* 2002). Recently restricted to two separate regions of southwest Spain, the species' geographical range is now being increased via reintroduction efforts in new locations (Rodriguez and Calzada, 2015). Likewise, in Portugal, releases began in late 2014 and have been taking place since then (Life+IBERLINCE, 2014).

Being a major threatened species, the lynx became heavily studied. Ecological aspects and fundamental requirements for Iberian lynx to prosper are well known (Delibes, 2002), and the maintenance of viable wild rabbit populations is one of them. In order to recover rabbit populations, a variety of management measures is frequently implemented by both conservationists and hunters. One often used is restocking (Calvete *et al.,* 1997). According to Delibes-Mateos *et al.* (2008b), from 1993 to 2002 thousands of rabbits were restocked in almost half of the hunting estates in central Spain. Between 1993 and 1995, in Doñana National Park only, 4300 rabbits were released for conservation goals (Villafuerte *et al.,* 2001). There are successful examples even in areas where the species was not present before, although the habitat was suitable (see Guil *et al.,* 2014). Yet this is not the usual outcome, especially if the number of rabbits to restock is low. In fact, researchers and game managers share the opinion that restocking operations usually have a low success rate (Calvete *et al.,* 1997; Letty *et al.,* 2002).

The major problem associated with restocking is the low survival rate in the first few days after implementation. According to Calvete *et al.* (1997), high rabbit mortality during the 10 d immediately following release appears to be the main limiting factor in rabbit restocking. This drawback reduces the breeding stock, risking the viability of the population (Letty *et al.,* 2008). Stress induced by capture, handling, captivity, transportation, and release is one of the factors responsible for the mortality frequently verified in restocking actions (Teixeira *et al.,* 2007; Waas *et al.,* 1997). To minimise this effect, a soft-release protocol is often followed, in which the animals have the opportunity to adapt to the new environment before being effectively free. This principle is opposed to that of hard release, in which there is no acclimation period.

Despite the large amount of information on game management in general and restocking procedures in particular, plenty of hunting estates, especially those without a game manager, still implement unplanned and improvised restocking operations. Often, small groups of wild rabbits are confined in small enclosures, put inside existing burrows or warrens, or just released in the chosen place (Calvete *et al.,* 1997). The decline in the wild populations led to the development of captive rearing of wild rabbits on game farms. The rabbits used for restocking are often acquired from a producer, as it is much easier than capturing wild rabbits in an area where they are abundant. Producers can breed and keep wild rabbits in a variety of conditions, ranging from a virtually absolute wild regime (rabbits living in colonies in enclosures) to very artificial ones (rabbits raised in cages) (González-Redondo & Sánchez-Martínez, 2014).

For this reason, it is important to consider the degree of wild behaviour the rabbits display, as it is likely to influence their adaptability to the new environment. Unplanned or barely prepared restocking operations are far from ideal, but as they are often implemented, we aimed at gaining useful knowledge from experiments with similar characteristics. Our goal was to test if different restocking protocols, concerning acclimation conditions, could produce different results in restocking operations using small groups of captive reared wild rabbits.

MATERIALS AND METHODS

Study area

Field work was conducted in a 1224 ha hunting estate in the Caldeirão mountain range in southern Portugal (37°13'N, 7°58'W) (Figure 1). The area is characterised by a Mediterranean mesothermic temperate climate with hot and dry summers and mild winters and belongs to the Mediterranean Ibero-Atlantic province (Rivas-Martínez and Loidi, 1999). The entire hunting area lies within the Natura 2000 Network, with 986 ha belonging to the Caldeirão Nature 2000 site and 238 ha to the Barrocal Nature 2000 site. The altitude ranges from 160 to 430 m and shallow leptosols are dominant, covering about 78% of the area. Cork oak (*Quercus suber*) forest is the major cover, occupying about 41.6% of the area; bushes (35.3%), agricultural fields (orchards and crops; 20.6%), and *Pinus* sp. (2.3%) take up the remaining area. Previous surveys showed wild rabbits are present in the area in scattered and low density populations.

Study sites, materials, and construction scheme

We used 2 different sites (restocking parks A and B) to perform the restocking operations. Both places were located within large patches of scrubland dominated by *Cistus ladanifer* with some pasturelands nearby. To accommodate the rabbits, we built artificial warrens and sowed mixed pastures of graminae and fabaceae in the surrounding areas. The artificial warrens were built using wood pallets and vegetal remains from the surroundings and then covered by soil.

Site A consisted of three individual small warrens fenced together, forming an enclosure of 0.025 ha. In site B, we built a single but larger warren. After being fenced, it formed an enclosed area of 0.004 ha. Both enclosures were secured against terrestrial carnivores by electrical fences and against aerial predators by protective nets placed on the top. We placed four doors in site A and two doors in site B.

We used a third enclosure (acclimation park) for rabbit acclimation before transfer to the restocking park in the first experiment. The enclosure area was about 0.4 ha, with 3 internal divisions that allowed rabbit confinement and capture. We also provided artificial warrens, food, and water and installed an electrical fence for protection against terrestrial predators.

Restocking procedures

We conducted 2 restocking operations using rabbits brought from a certified breeder, who kept the animals in a semi-natural regime, i.e. living outside in enclosures but with food supplementation and used to human presence, 440 km away from the study area. Before travelling, the rabbits were sexed, weighed, vaccinated, ear-tagged and fed. The

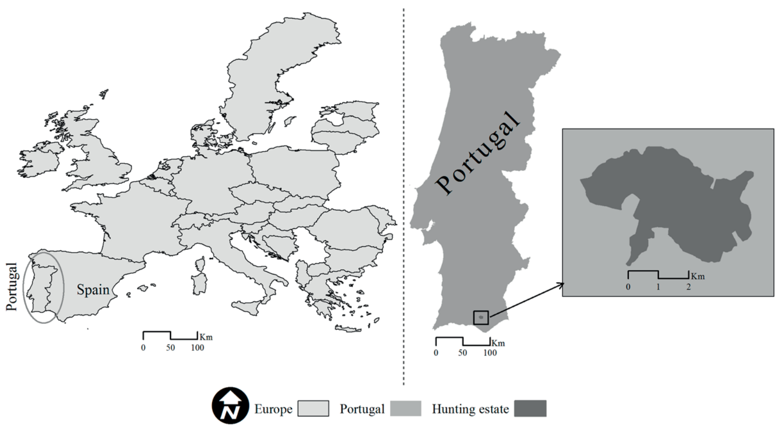

Figure 1: Study area.

trips were made during the night and took approximately 5 h. The animals travelled in small groups inside 2 cages. The first restocking took place in December 2010 and was characterised by a long confinement period, first in the acclimation park and then in the restocking park. In the second restocking, which occurred in November 2011, the rabbits were directly put inside the restocking park, skipping the acclimation park, and the confinement period was much shorter.

First restocking

A total of 50 rabbits (12 males and 38 females) arrived before morning and were placed inside the acclimation park warrens. After 27 d, 9 rabbits were captured (1 male and 8 females), equipped with radio-tracking collars and released in restocking site A, during the early hours of the morning. We provided water and food during the captivity period, which lasted 65 d before we opened the doors. This captivity period represented an extended acclimation stage in the place where the actual release would occur (Figure 2).

Second restocking

A total of 9 rabbits (2 males and 7 females) arrived before sunrise, were fitted with radio collars and placed directly in restocking site B. After 17 d of confinement, the passages were opened to allow the batch to move freely (Figure 2).

Rabbit survey by radio tracking

The rabbits were equipped with radio collars weighing about 30 g and containing an activity sensor (BIOTRACK, Wareham, UK). There was no predetermined tracking period, as the intention was to monitor the rabbits as long as possible. The locations were obtained mainly at night, and the operator always approached the survey area on foot, in order to reduce disturbance.

Data analysis

Concerning survival analysis, unusual locations or lasting lack of activity were interpreted as potential deaths, and whenever that occurred the technician searched for the animals. Causes of death were classified as *predation* or *other causes*. *Predation* was assigned to terrestrial carnivores (when incisor marks on collars, scat, rabbit caecum, and buried or half-buried corpses were found) or to raptors (evidence of feathers, characteristic tufts of torn-out hair, and remains of long bones) (Calvete and Estrada, 2004). *Other causes* included individuals dead due to disease,

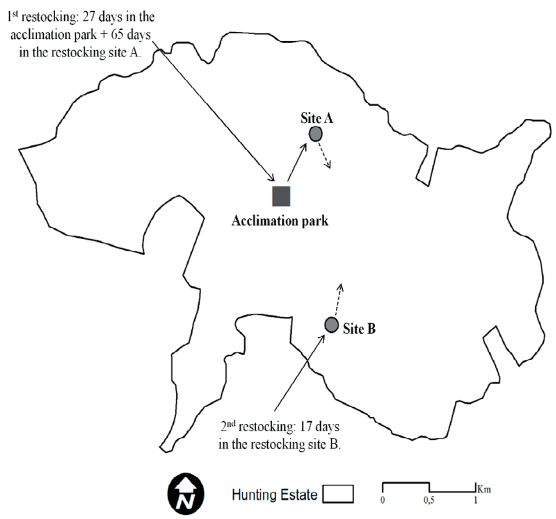

Figure 2: Restocking spatial representation.

aggression associated with social interactions, non-recovered carcasses inside warrens or other unknown motives. Following Rouco *et al.* (2010), we also considered animals found dead on the nth day after release had survived n–1 d.

Survival at a given moment was assessed by calculating the ratio between the number of living rabbits and the total number of rabbits released: Sr=lr/tr; Sr, survival ratio; lr, living rabbits; tr, total rabbits.

Additionally, by calculating the survival ratio daily, the global survival rate of the batch was obtained.

As to the spatial behaviour, in order to analyse the dispersal, 3 indicators were calculated –maximum distance travelled (MDT), mean dispersal distance from the release point (DRP) and home range total area (HR)– for each individual and batch. MDT is the distance from the release point to the furthest recorded point, and DRP is the average distance of all locations of each rabbit from the release point (Moreno *et al.,* 2004). Home range total area sizes were estimated by the minimum convex polygon method (MCP; Mohr, 1947), using all locations of each individual (100% MCP). These variables were calculated using the outputs from the radio-tracking monitoring, the software ArcGis 9.3.1. (ESRI 2009) and Hawth's Analysis Tools (Beyer, 2004).

RESULTS

Restocking procedures

Rabbit survey by radio tracking

During the confinement period, 4 deaths occurred, 2 in each experiment, diminishing the number of animals effectively tracked to 14 (7 in each experiment). The survey period of the first restocking lasted 65 d after release, until the radio collar batteries went out, in which 28 monitoring sessions were conducted and 168 locations registered. The survey of the second restocking lasted 54 d, in which 162 locations were registered during 46 monitoring sessions.

Data analysis

In the first experiment, 2 rabbits died during the confinement period. There was no mortality in the first days after release. The first death occurred 28 d after the park was opened. After that, 3 more deaths were recorded. A total of 6 deaths were verified, 1 due to *predation* by terrestrial carnivores and 5 to *other causes*. At the end of the survey there were 3 living rabbits, which represented a survival rate of 0.33 (Figure 2).

In the second experiment, 2 rabbits died during the confinement period. There was no high mortality immediately after the doors were open as the first death only occurred 7 d afterwards. However, after 53 d all the rabbits were dead, 4 due to *predation* by terrestrial carnivores and 3 to *other causes* (Figure 3).

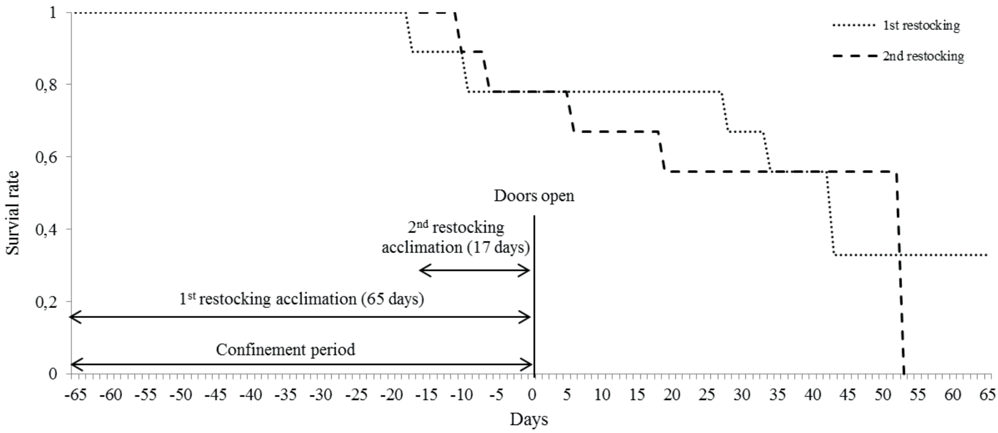

Figure 3: Daily survival rate of the rabbits in both restockings.

Table 1: Spatial movement results of the rabbits in both restockings.

1st Restocking				2nd Restocking			
Individual	HR (m^2)	MDT (m)	DRP (m)	Individual	HR (m^2)	MDT (m)	DRP (m)
A	3848	64	50	H	156	18	16
B	5167	180	112	I	117	16	13
C	2595	59	46	J	156	34	5
D	11337	151	97	K	0	13	13
E	2683	67	40	L	0	7	7
F	8702	106	67	M	0	0	0
G	1995	139	110	N	0	0	0
Batch				Batch			
Min	1995	59	46	Min	0	0	0
Max	11337	180	112	Max	156	34	16
Average	5190	109	75	Average	72	15	9

HR: home range; MDT: maximum distance travelled; DRP: mean dispersal distance from the release point.

All the dispersion-related measurements –HR, MDT, and DRP– were much higher in the first restocking, whether the analysis was based on each individual or at the batch level (Table 1).

Concerning the second restocking, HR measuring for individuals K, L, M, and N was not viable because there were not enough locations outside the park. In fact, individuals M and N both died inside the restocking park and were never found outside the park during the monitoring sessions. As for rabbits K and L, they were located outside the park only once before being found dead by predation.

DISCUSSION

Aware that longer acclimation periods favour successful restocking operations (Rouco *et al.,* 2010), we tested restocking viability using captive reared wild rabbits by exposing them to longer acclimation periods than usual. The results suggest that a suitable acclimation period allows wild rabbits to endure after a restocking operation.

The first batch of rabbits (27 d of acclimation followed by translocation and 65 d of confinement before actual release) achieved better survival rates and explored the surroundings much more than the second batch (17 d of confinement before release). The first batch had a survival rate of 0.3 after 65 d of monitoring, whereas the second batch were all dead after 53 d of radio tracking.

According to the literature, a peak of rabbit mortality usually occurs in the days immediately following release, which is considered the main limiting factor in rabbit restocking (Calvete *et al.,* 1997). In both experiments, we applied longer confinement periods than found in similar studies (Calvete *et al.,* 1997; Letty *et al.,* 2002; Calvete and Estrada, 2004; Rouco *et al.,* 2010). We presume that the extra time rabbits had to adapt and get to know the environment, even though the parks were still closed, is the main reason why higher rabbit mortality during the first few days after confinement did not occur.

The individuals of the first batch seemed to have acquired greater ability to explore the environment than the second batch individuals. In the first experiment, all rabbits were located several times outside the park, while in the second experiment 2 rabbits never left the park, another 2 were located outside the park once, and only 3 rabbits were frequently found in the park surroundings. As stated before, the exploration of the surroundings was also very different in the two experiments. The spatial movements observed are characterised by the more frequent and larger movements of the first batch against the fewer and shorter movements of the second batch.

To our knowledge, there are no previous studies presenting wild rabbit home range estimations of low-density populations in areas like the Caldeirão mountain range. We used non-local rabbits for restocking purposes rather than local rabbits to track their movements. That is probably why the HR values we recorded are notably inferior to those found in other studies. For instance, Devillard *et al.* (2008), working with wild rabbit low-density populations in

France, reported HR of 20492 m^2 (local rabbits; 95% MCP). In our case, also working with low-density populations, yet tracking non-local rabbits, the higher average HR was 5190 m^2, obtained during the first experiment. Even compared with those reported for higher density populations by Devillard *et al.* (2008) (7333 m^2 and 6878 m^2), our results still show smaller HR.

Concerning both survival rates and spatial behaviour, besides the differences in confinement period in the restocking parks, we suspect that the time spent in the acclimation park was responsible for the different outcomes. Before being released into the restocking park, the rabbits used in the first experiment spent 27 d in the acclimation park, an enclosure of 0.4 ha, where they could move freely. This period spent in a wide and partially protected space could have enhanced the rabbits' ability to explore the environment.

It is important to stress the 2 experiments took place in near but different sites, which makes them hard to compare to the full extent. However, the area where the second batch was released was better suited for wild rabbit because it had more pastures and fewer predators (parallel field work results). The only advantage the first batch had compared to the second was a better and longer acclimation, which supports our conclusions.

CONCLUSIONS

This study provides helpful information regarding wild rabbit reinforcements or re-introductions and turns out to be even more relevant because it took place in the Caldeirão mountain range, once a territory of the Iberian lynx. Adding to the lynx recovery efforts now being made, including captive breeding programmes and lynx releasing, our results can contribute to increasing the density of its main prey, the wild rabbit. This information can also be applied to other important areas for Iberian lynx recovery, such as the Western Sierra Morena-Guadiana (southwest Spain and southeast Portugal) and Algarve-Odemira region (southwest Portugal; see Sarmento *et al.,* 2009). This information is also valuable for hunters intending to recover or boost wild rabbit populations in their hunting estates via restocking.

We do not encourage unplanned and improvised population management actions, and we clearly prefer translocations from near areas and the use of adapted rabbits over supplementations using foreign and unadapted rabbits. The results achieved in the experiments, and the consequent conclusion extracted, should not be interpreted as support or incentive to use unadapted rabbits instead of adapted ones.

In summary: i) the results corroborate the already known fact that longer acclimation periods are more beneficial than shorter ones when performing a wild rabbit restocking (Rouco *et al.,* 2010), and ii) testing with unadapted rabbits and extending considerably the confinement periods, we arrived at the conclusion that restocking can also be viable using such animals.

Acknowledgements: We would like to thank the Associação de Caçadores de Querença for their valuable support with the fieldwork.

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