# ORIGINAL PAPER

# Sanitary risks of red-legged partridge releases: introduction of parasites

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Abstract We studied the helminth community and body condition of 99 hunter-harvested red-legged partridges (Alectoris rufa) from Ciudad Real (Central Spain). Fortysix were sampled in two game estates where an important number of farm-reared red-legged partridges are released yearly. The remaining 53 were obtained from natural wild populations adjacent to one of the estates with releases. Four nematode species (Heterakis gallinarum, Aonchoteca caudinflata, Eucoleus contortus and Cheilospirura gruveli) and two cestode species (Raillietina (R.) tetragona and Skryabinia bolivari) were identified. The managed areas showed higher parasite diversity, with higher prevalences and intensities for all helminths found. Three of these species were typical of farm-bred partridges and two of these, A. caundinflata and S. bolivari, were found parasitizing adult partridges. This suggests introduction of these helminths into the breeding population of managed states. The birds sampled in the nonmanaged estates showed a better body condition, but no relation with parasite infection was found. Our results suggest that the release of farmreared red-legged partridges, a strategy that is becoming a common practice in Spanish hunting areas, poses risk to

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Gestión Ambiental Viveros y Repoblaciones de Navarra, C/Padre Adoain, 219 bajo, E- 31015 Pamplona, Spain wild populations because of introducing parasites. However, these results also suggest that simply stopping releases may be a good way to remove locally those parasites from populations, as the establishment of the introduced parasites seems limited.

**Keywords** *Alectoris rufa* · Condition · Helminths · Restocking · Spain

# Introduction

The red-legged partridge (*Alectoris rufa*) is the most abundant game bird in the Iberian Peninsula, and its hunting is one of the most important economical and social activities in central Spain (Bernabeu 2000). During the last decades, natural populations of this game bird have declined in most of its distribution range (Aebischer and Potts 1994). In Spain, only declines have been documented (Gortázar et al. 2002, and references therein). The strategy of many hunting estate managers to overcome this decline has been releasing farm-reared birds.

Previous studies evidenced differences between the parasites found in farmed and those found in wild redlegged partridges (e.g., Millán et al. 2004a). Regarding helminths, monoxenous species were more abundant in farms and heteroxenous parasites in natural populations (Millán et al. 2004b, c). These differences can negatively affect autochthonous populations in the managed areas, because of the probably limited previous contact with these pathogens. In fact, potential disease transmission from released animals to wild populations is one of the most important points to be considered in restocking programs (Viggers et al. 1993). As an example, the annual release of farmed ring-necked pheasants (*Phasianus colchicus*) in the

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UK is believed to maintain or even increase *Heterakis* gallinarum burdens in the wild pheasant population (Draycott and Sage 2005), which in turn could be one of the factors involved in the gray partridge *Perdix perdix* decline (Tompkins et al. 2001). However, the survival of released red-legged partridges is low (see Birkan 1977; Capelo and Castro Pereira 1996; Gortázar et al. 2000; Duarte and Vargas 2004), and this could be limiting pathogen transmission between farmed and wild birds.

Thus, the aim of the present study was to confirm if parasites could be effectively introduced into the field by farm-reared red-legged partridge releases.

### Materials and methods

## Study area

The study was performed in four game estates, ranging from 1,009 to 3,145 ha, located in Ciudad Real (Central Spain, 30T, X419903/Y4313925). Habitat is characterized by undulated farmland of wheat and barley crops with olive trees and vineyard patches. Most cereals are cultured by a traditional 2-year rotation system. No site differences where evident between the study estates.

The red-legged partridge is the most important game species in these hunting areas, although other game species like the Iberian hare (*Lepus granatensis*) or the feral pigeon (*Columba livia*) are present. The red fox (*Vulpes vulpes*) is the most abundant predator, but feral cats (*Felis catus*) and dogs (*Canis familiaris*), montagu's harrier (*Circus pygargus*), marsh harrier (*Circus aeruginosus*), buzzards (*Buteo buteo*), and golden eagle (*Aquila chrysaetos*) can prey on red-legged partridges. Moreover, the study area holds important populations of several species of steppe birds, such as great bustard (*Otis tarda*), little bustard (*Tetrax tetrax*), pin-tailed sandgrouse (*Pterocles orientalis*), or stone curlew (*Burhinus oedicnemus*).

Two of the sampled game estates followed an intensive management model with more than 2,000 farm-reared partridges released yearly. The other two consisted of wild populations where no restocking with captive-bred game birds was performed in the last 10 years and that were adjacent to one of the managed states.

## Study animals

A total of 99 hunter-harvested red-legged partridges were sampled, 46 from the managed areas and 53 from the wild populations. All birds were weighed using a 1,000-g Pesola<sup>®</sup> precision balance ( $\pm 5$  g), and left tarsus length was measured using a caliper ( $\pm 0.01$  mm). All measures were taken by the same person (Pérez-Rodríguez). Sex and

age were determined in the field according to the morphological description of Sáenz de Buruaga et al. (2001) but were confirmed by examination of the gonads and the bursa of Fabricius. We tried to obtain a sex and age ratio next to 50% in our sample.

Partridge body condition was estimated using two different measurements: the pectoral muscle thickness (PMT), measured using a portable ultrasound meter (Krautkramer USM 22 device; Pérez-Rodríguez et al. 2006), and the residuals of the regression of the body mass on the cube of tarsus length (RBMTL; Andersson 1992).

Laboratory methods

The digestive tract was opened longitudinally, and the content was collected for parasite isolation. The digestive tract was soaked overnight in water to allow the liberation of any parasite that could be attached to the mucosa. The liver was cut into 3-mm slices and put into water to allow the visual inspection for trematodes. The samples obtained were examined by means of a stereomicroscope and the detected worms were counted and stored in 70% ethanol before identification. The identification of the parasites was done according to López-Neyra (1947); Skryabin (1991), and Melhorn et al. (1992).

#### Statistical analysis

We used Mann–Whitney's U test to evaluate the influence of management (natural vs restocking), sex and age on parasite diversity, parasite burdens, and body condition and  $\chi^2$  tests to evaluate the influence of these factors on parasite prevalence. The possible relationship between the parasite burdens and body condition was tested with Spearman's correlation test.

#### Results

Four nematode species (*H. gallinarum*, *Aonchoteca caudinflata*, *Eucoleus contortus*, and *Cheilospirura gruveli*) and two cestode species (*Raillietina (R.) tetragona* and *Skryabinia bolivari*) were identified. No trematodes were detected.

*H. gallinarum, C. gruveli*, and *R. (R.) tetragona* were found in both different management strategies. *A. caudin-flata, E. contortus*, and *S. bolivari* were only found in the managed areas. Helminth diversity was significantly higher in the managed areas  $(0.38\pm0.58$  species per host) than in the natural ones  $(0.06\pm0.23; Z=-3.53; p<0.001)$ .

In the case of parasites found in both management models, their prevalence was higher in the managed areas, but these differences reached the significance level only in

Table 1 Helminth prevalences found in red-legged partridges from two different management models and significance level of the differences ( $\chi^2$  test)

	Wild ( <i>n</i> =53, %)	Managed ( <i>n</i> =45, %)	Mean	p value
Heterakis gallinarum	3.77	17.39	5.01	< 0.05
Aonchoteca caudinflata	0	6.52	3.56	ns
Eucoleus contortus	0	2.17	1.16	ns
Cheilospirura gruveli	1.89	10.87	3.49	ns
Raillietina tetragona	3.77	6.66	0.42	ns
Skryabinia bolivari	0	4.44	2.4	ns

the case of *H. gallinarum* (Table 1). The intensities of these parasites followed the same trend, but in this case, the significance level was reached by *H. gallinarum* and *C. gruveli* (Table 2).

No relationships were found between parasite burdens and the two body condition indexes PMT and RBMTL (Spearman correlation tests,  $r_s$ =-0.31 and -0.28, p>0.05). PMT was higher in the wild populations (26.5±2.4 mm) than in the managed areas (24.3±3.1 mm; Fig. 1), but this difference was only significant in the case of juvenile males (Table 3). No differences were found in the RBMTL.

# Discussion

All parasite species found in our study have been previously described parasitizing the red-legged partridge (López-Neyra 1947; Carvalho-Varela and Ferradeira 1997; Cordero del Campillo and Rojo 1999; Calvete et al. 2003; Millán et al. 2004a, b).

The species richness detected in this study (six species in the managed area and only three in the natural one) is lower than those reported in two previous large-scale studies (13 different species by Calvete et al. (2003) or 14 different species by Millán et al. (2004a) but similar to the richness reported by Millán et al. (2004c) in a small study area in southern Spain (four species). However, prevalences and abundances are markedly lower than those reported in these studies.

Millán et al. (2004a, b) found that the parasite community in wild populations of red-legged partridges was different from that of farm-bred birds. They suggested that new parasites could be introduced into the field with the release of farm-reared birds. Three species identified in our study, *H. gallinarum*, *C. gruveli*, and *R. (R.) tetragona*, are usually found in natural populations (López-Neyra 1947; Tarazona et al. 1979; Millán et al. 2004b, c). However, the other three ones (*E. contortus*, *A. caudinflata*, and *S. bolivari*), are more usually found in farm-reared birds (see Millán et al. 2004a). These were found in our study parasitizing birds sampled in the managed areas only.

Furthermore, two of these species were found in adult birds in our study. Partridges released for hunting are usually juveniles. Thus, the presence of these parasites in adult birds suggests (1) that there had been a transmission from farmed to wild birds, (2) that we found birds released in the previous year that had kept the infection for a whole year, or (3) that some adult birds were released. All of these possibilities would be consistent with the introduction of these parasites into the field through releases of their farmbred hosts. Although the two last options cannot demonstrate themselves in the transmission of these parasites between farm-reared and wild birds, the period of time spent by the farm-released partridges in the field, at least 2 months, could imply a high risk of transmission because of an increased parasite excretion after releasing, as experimentally shown in the case of the pheasant (Villanúa et al. 2006a). This in turn could be due to the absence of antiparasite treatment and the stress immunodepression that follows releasing into an unknown habitat.

The introduction of new pathogens into a wild population can be an important conservation problem because natural populations could have lower resistance to a pathogen to which they have had no previous contact (Newton 1998). In our study area, some endangered steppe birds share the habitat with the red-legged partridge.

Table 2Helminth burdens found in red-legged partridges from the two different management models and significance level of their differencesby the Mann–Whiney U test

	Wild ( <i>n</i> =53)			Managed (n=45)			F value	p value
	Mean	Min–max	SD	Mean	Min-max	SD		
Heterakis gallinarum	0.04	0-1	0.19	0.36	0–5	0.93	-1.22	< 0.05
Aonchoteca caudinflata	0.00	0	0.00	0.18	0–3	0.68	-1.89	ns
Eucoleus contortus	0.00	0	0.00	0.04	0–2	0.30	-1.08	ns
Cheilospirura gruveli	0.13	0-7	0.96	1.89	0-30	6.82	-2.17	< 0.05
Raillietina tetragona	0.13	0–4	0.68	0.40	0-8	1.61	-0.67	ns
Skryabinia bolivari	0.00	0	0.00	0.91	0–30	4.72	-1.54	ns



Fig. 1 Body condition in the two different management models measured with the regression residuals of the body mass on the cube of tarsus length (*RBMTL*) and the pectoral muscle thickness (*PMT*)

Therefore, the introduction of new nonspecific parasites supposes an additional problem for their conservation (Villanúa et al. 2007a).

The detection of higher parasite burdens in the managed areas in this study could be an indicator of the low effectiveness of the preventive measures to reduce parasites in the released animals. Current preventive measures applied before the release of farm-reared game birds (if they exist) are based on (1) a coprological analysis performed in feces collected in the aviary soil and (2) antiparasite treatment following the same protocol that is used in the case of poultry (Villanúa et al. 2007b). The first point of this protocol has the problem that there are a lot of factors such as host reproductive status (Ruíz de Ybañez et al. 2004), weather (Vicente et al. 2005), season (Kumba et al. 2003), random day-to-day variations (Giver et al. 2000), phase of the parasitic infection (Giver et al. 2000), or hour of sampling (Villanúa et al. 2006b) that can modify propagule excretion and are not always considered. In addition, the effectiveness of antiparasite protocols in game species is insufficient to prevent introducing parasites into the field as recently shown in the case of Albendazole treatment against A. caudinflata and H. gallinarum in redlegged partridges (Villanúa et al. 2007b).

If the preventive protocols are not enough to eliminate the parasites from the released birds, the other option taken is antiparasite treatments administered in the field. Previous studies evaluated the effect of administering anthelminthic drugs in the field together with the supplementary food (Woodburn et al. 2002; Draycott and Sage 2005). These authors found that the birds given anthelmintic treatment and supplementary food had significantly lower worm burdens and increased their productivity. Nevertheless, some authors think that treatment of free-living birds is not practical (Cole 1999) and the pros and cons of the administration of drugs to wild birds have to be considered carefully, taking ethical and public health concerns into account (Höfle et al. 2004).

On the other hand, we must remark that the two sampled game states with wild partridges were adjacent to one of the managed states. Thus, our results indicate that wild partridge populations have lower parasite diversity, preva-

 Table 3
 Mann-Whiney U test results for the body condition indexes (pectoral muscle thickness, PMT, and regression residuals of the body mass on the cube of tarsus length, RBMTL) found in the different age and sex groups in two different management models

	RBMTL				PMT				
	Males		Females		Males		Females		
	z value	p value	z value	p value	z value	p value	z value	p value	
Adult Juvenile	-0.71 0.10	ns ns	-1.84 2.01	ns ns	1.60 2.40	ns <0.05	0.52 1.65	ns <0.1	

lence, and burdens than adjacent populations where thousands of farm-bred partridges are released yearly, that is, that the transmission of parasites from farmed birds to wild birds is relatively limited to the hunting lands where releases are performed but may not affect so much in neighbor populations. This may be due to natural factors (poor dispersal ability of partridges in general, or of farmed birds in particular) or to management actions (maintenance of released partridges within the boundaries of game states by pursuing them or intense hunting removing most released partridges before they may disperse). In any case, this result suggests that when releases are interrupted in a given hunting land, the problem of parasite transmission may be largely avoided. In fact, this may be the main reason explaining why when sampling wild and released populations in Spain we still find differences in parasite communities, albeit millions of partridges have been released during the last decades all over Spain.

As we have shown, lower body condition found in juvenile partridges from the managed areas was not apparently related with parasites. It can be explained by two different causes: (1) a deficient muscle development caused by the farm-rearing (in case a significant part of the juvenile birds sampled in these areas were farm-reared partridges) or (2) the effect of other pathogens such as bacteria or viruses that may equally be introduced along with released birds. However, more specific studies are needed to test this hypothesis.

In conclusion, while the present study does not refute the hypothesis that hunting estates with partridge releases are introducing helminth parasites into the ecosystem, neither does it support it. The observation that wild populations adjacent to managed populations do not carry the parasite species found only in managed populations strongly suggests that such transfer to the breeding population on the managed estate is limited. Anyway, improved controls are desirable to prevent associated risks for sustainable game management and conservation.

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