DOI: 10.1515/ap-2015-0049
© W. Stefański Institute of Parasitology, PAS
Acta Parasitologica, 2015, 60(2), 350–355; ISSN 1230-2821

DE GRUYTER

RESEARCH NOTE

Coccidiosis in European rabbit (*Oryctolagus cuniculus algirus*) populations in the Iberian Peninsula

Sofia Marques Silva[1,2,*], Catarina Ferreira[2,3,4], Joana Paupério[2], Rodolfo Miguel Silva[5], Paulo Célio Alves[2,6,7] and Armando Lemos[5]

[1]Departamento de Zoologia, Museu Paraense Emílio Goeldi/Universidade Federal do Pará, Belém, PA, Brazil; [2]Centro de Investigação em Biodiversidade e Recursos Genéticos, Universidade do Porto, InBIO Laboratório Associado, Vairão, Portugal; [3]Department of Biology, Trent University, Peterborough, Ontario, Canada; [4]Instituto de Investigación en Recursos Cinegéticos, Universidad Castilla La Mancha (IREC; CSIC-UCLM-JCCM), Ciudad Real, Spain; [5]Instituto de Ciências Biomédicas Abel Salazar, Universidade do Porto, Vairão, Portugal; [6]Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto, Portugal; [7]Wildlife Biology Program, University of Montana, Missoula, USA

Abstract

The European rabbit *Oryctolagus cuniculus* is a keystone species from the Iberian Peninsula where viral diseases have played a prominent role in regulating their populations. Coccidiosis, a parasitic disease caused primarily by *Eimeria* spp., is also thought to have important negative effects. However, few studies have investigated the impact of coccidia on wild European rabbit populations on the Iberian Peninsula. Here we estimate coccidian prevalence in rabbit faecal samples collected along transects established in two ecological regions. Six *Eimeria* species, with different pathogenicity, were identified (*E. coecicola*, *E. perforans*, *E media*, *E. magna*, *E. irresidua* and *E. flavescens*). Species diversity varied significantly between regions although mean oocyst excretion levels were generally low in both areas (57.61 s.d.±78.07 and 17.03 s.d.±27.72, oocyst per gram of rabbit faeces). This study is the first to describe the composition of the *Eimeria* spp. assemblage for wild rabbit populations on the Iberian Peninsula and provides fundamental information for future studies on the potential interaction of viral and parasitic diseases.

Keywords

Eimeria spp., Lagomorpha, Portugal, faecal pellets, protozoans, host-parasite interactions

The European rabbit *Oryctolagus cuniculus* is a keystone species native to the Iberian Peninsula (Ferrand 2008). The density of this species has suffered a steep decline over the last decades being current levels less than 10% of early 20th century estimates (Delibes-Mateos *et al.* 2009). Myxoma (MV) and rabbit haemorrhagic disease viruses (RHDV) have been identified as the main factors with several studies investigating the impact of viral infections in rabbit populations (e.g. Villafuerte *et al.* 1995; Delibes-Mateos *et al.* 2009; Abrantes *et al.* 2013). However, parasitic diseases, such as coccidiosis, can also impact rabbit populations (e.g. Bertó-Moran *et al.* 2013). Lello *et al*. (2005) reported that coccidiosis can cause significant weight losses and reduce the overall physical condition of wild rabbits, in some instances even more than myxomatosis. Notably, in France, 20% mortality due to coccidiosis has been recorded, sometimes exceeding the percentage of deaths due to RHDV (Marchandeau *et al.* 1999).

Coccidiosis is most frequently caused by *Eimeria* species and eleven species have been described to parasitize rabbits (Hobbs and Twigg 1998; Pakandl 2009). In Iberia, coccidiosis has been recorded in both wild and domestic rabbit populations (Maldonado *et al.* 2004; RIPAC 2004; Rosell *et al.* 2009; Bertó-Moran *et al.* 2013), however no studies described

*Corresponding author: sofiamarques1@gmail.com

Eimeria beyond genus. Identification of coccidia at the species level is relevant due to the distinct pathogenicity of each species, which can determine its potential to interact with other host diseases (Lello *et al.* 2005; Pakandl 2009; Bertó-Moran *et al.* 2013; Boag *et al.* 2013).

Data on the prevalence of coccidiosis in Iberian wild rabbit populations are scarce. In Spain, *Eimeria* spp. oocyst excretion has been quantified in rabbit restocking enclosures in the wild (Bertó-Moran *et al.* 2013), and the only study available for Portugal states that in a southern wild population about 13% of rabbits found dead presented coccidiosis (RIPAC 2004).

The present study is the first to describe the composition of the *Eimeria* spp. assemblage in wild rabbit populations from its native range, the Iberian Peninsula. We present new data on the prevalence of *Eimeria* species in two rabbit populations in Portugal under different environmental conditions, and compare the monthly variation of oocyst excretion within and between populations. We hope this study may contribute, in the future, for studies on the potential interaction between viral and parasitic diseases, a topic that has gained a new breath over the last years (Bertó-Moran *et al.* 2013; Boag *et al.* 2013).

The two rabbit populations selected are located in two natural parks: Serra da Estrela Natural Park (PNSE) in north-east Portugal, and Sudoeste Alentejano and Costa Vicentina Natural Park (PNSACV) in south-western Portugal (Fig. 1). PNSE is a mountain region, with Meso and Supramediterranean climate, marked by cold wet winters, and frequent occurrence of snow. PNSACV climate is Mediterranean, strongly influenced by the proximity of the sea, and regularly affected by summer droughts.

From December 2004 to May 2005, we collected rabbit faecal samples along pellet count transects in the two study areas (for more details on the pellet count method see Ferreira and Alves 2009). The assessment of coccidia typically relies on the analysis of fresh faecal samples, and this is usually ensured by the collection of faeces from animals within enclosures, after direct observation of defecation, or directly from dead animals (e.g. Stodart 1968; Hobbs and Twigg 1998). Because our sampling design was targeted primarily to monitor rabbit abundance, the rabbit faeces collected for this study may have been exposed to weather conditions for a certain period of time. For that reason, only those pellets appearing fresh were collected which hampered data collection in April and May 2005 in PNSE, when the climate was exceptionally dry and all pellets found were visually desiccated. In the field, the rabbit faecal pellets collected were wrapped in humid paper, stored in plastic bags and transported to the laboratory in a cooler. Samples were kept frozen until analysis in the ICBAS-UP/ Campus of Vairão laboratory.

Approximately 1.0 g of faeces was macerated and added to a sugar solution with a density of about 1.2 g/cm^3 (Jarvinen 1999). After admixing, the solution was filtered with fine mesh gauze and left standing for a few minutes. As oocysts were

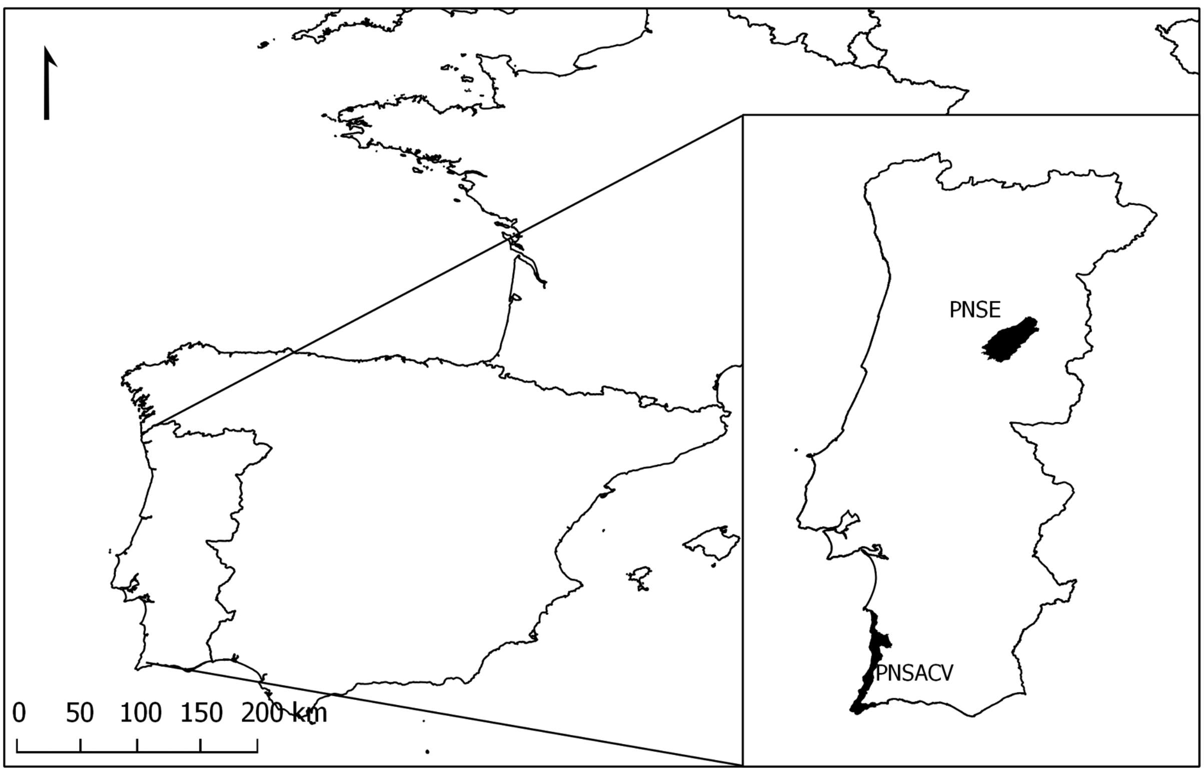

Fig. 1. Location of the two study areas in the Iberian Peninsula. PNSE – Serra da Estrela Natural Park and PNSACV – Sudoeste Alentejano and Costa Vicentina Natural Park

separated from faecal matter by flotation, the solution was collected on the surface and transferred to a slide for species identification and to a McMaster slide for oocyst counts (Gordon and Whitlock 1939). *Eimeria* species identification was performed using an optic microscope at 400x magnification, and considering oocyst morphology (length, width, colour and shape), according to the descriptions of Pellerdy (1974) and Hobbs and Twigg (1998). This procedure was performed for at least three replicates per study area per month. Diversity of *Eimeria* species in each study area was estimated using Shannon's diversity index.

Oocyst excretion was measured as the number of oocysts per gram (OPG) of rabbit faeces: OPG = (volume of sugar solution × number of oocysts)/(amount of faeces/slide volume). The relationship between oocyst excretion and rabbit abundance in each study area (expressed as pellets/m^2; Ferreira and Alves 2009) was assessed by Spearman correlation tests. Differences in the oocyst excretion between areas and months were assessed by Mann-Whitney-Wilcoxon tests. Analyses were performed using basic and vegan R packages (R Development Core Team 2012; Oksanen *et al.* 2013 and references therein).

Six *Eimeria* species were observed (Fig. 2): *E. coecicola* which is considered non-pathogenic; *E. perforans* slightly pathogenic; *E media*, *E. magna* and E. *irresidua* pathogenic; and *E. flavescens* is highly pathogenic (reviewed by Pakandl

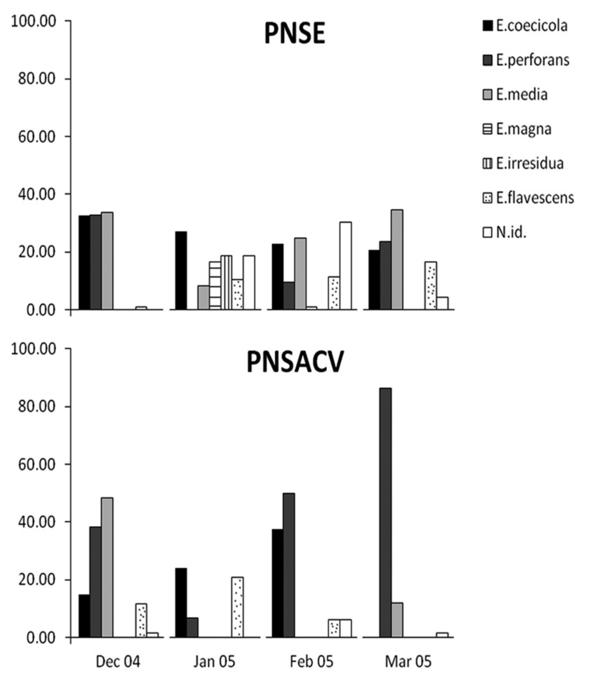

Fig. 2. Percentage of *Eimeria* species identified within PNSE – Serra da Estrela Natural Park and PNSACV– Sudoeste Alentejano and Costa Vicentina Natural Park. N. id. – Species not identified. Note that species are represented according to their level of pathogenicity. Results are only presented from December 2004 to March 2005 since in April and May 2005 no samples could be collected in PNSE, and no oocysts were observed in PNSACV samples

2009). Of these, *E. magna* and *E. irresidua* were only detected in PNSE. In addition to these confirmed species, three other species were suspected to occur, *E. piriformis* and *E. intestinalis*, found only in PNSACV; and *E. stiedae* found only in PNSE on March 2005. However, due to the undefined observed shape of the oocysts it was not possible to reach an exact species identification, and so these and other oocysts that were too degraded to be confidently assigned to any species, were classified as "not identified" (Fig. 2), and were excluded from calculations of the species diversity indexes (Fig. 3). Seasonal and regional variation in the diversity of coccidian were observed (Fig. 2), with higher species diversity recorded in the winter and in PNSE (Fig. 3). This result is in accordance with previous findings from regions with long and dry summers, such as PNSACV, where only *Eimeria* species with a shorter sporulation time may be able to complete their life cycles (Stodart 1968; Hobbs and Twigg 1998; Bertolino *et al.* 2003). Accordingly, *E. perforans*, the species with the shortest sporulation time (Hobbs and Twigg 1998), was the most frequent species in PNSACV (Fig. 2).

Mean oocyst excretion ± s.d. estimated for PNSE was 57.61±78.07 OPG, and for PNSACV 17.03±27.72 OPG (Fig. 4). No statistically significant differences were found between months within each study area (PNSE: $p = 0.624$; PNSACV: $p = 0.121$; Fig. 4). Considerably higher oocyst excretion estimates were reported in rabbits in restocking enclosures in southern Spain (over 500 OPG, Bertó-Moran *et al.* 2013), than those reported herein (lower than 170 OPG; Fig. 4). According to Hobbs *et al.* (1999), oocyst counts are density dependent, and so the discrepancies found between our results and Bertó-Moran *et al.* (2013) may be attributed to the "high rabbit population density" in the restocking enclosures, which contrasts with the low rabbit abundances found in our study areas. Mean rabbit abundance ± s.d. in PNSE between December 2004 and May 2005 was 4.06±3.02 pellets/m^2 and 1.51±1.14 pellets/m^2 in PNSACV. Furthermore, oocyst counts of *Eimeria* species in rabbit wild populations can vary considerably across months and years (Stodart 1968; Hobbs *et al.* 1999; Foronda *et al.* 2005). Thus, oocyst excretions in our study areas might also

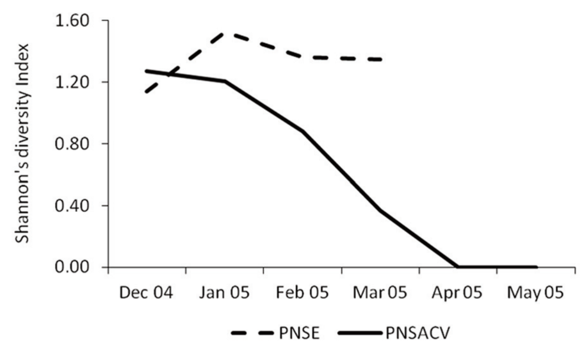

Fig. 3. Monthly variation of Shannon's diversity index of *Eimeria* spp. found in rabbit faecal pellets in PNSE – Serra da Estrela Natural Park and PNSACV– Sudoeste Alentejano and Costa Vicentina Natural Park

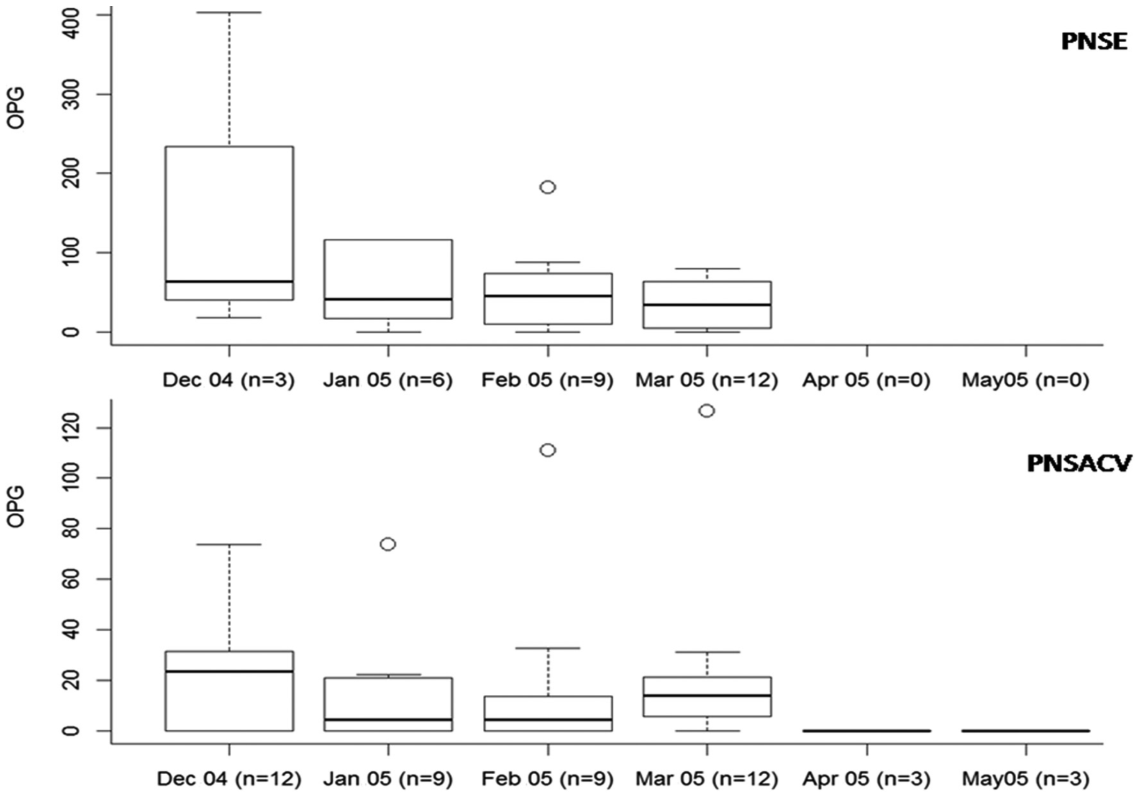

Fig. 4. Monthly variation of oocyst excretion estimates, expressed as the number of *Eimeria* oocysts per gram (OPG), found in rabbit populations in PNSE– Serra da Estrela Natural Park and PNSACV– Sudoeste Alentejano and Costa Vicentina Natural Park. Number of replicates of 1.00g of fresh rabbit pellets analysed in brackets. In April and May 2005 no samples could be collected in PNSE, and no oocysts were observed in PNSACV samples

be distinct from the ones reported for Spain (Bertó-Moran *et al.* 2013) just due to natural seasonality. Finally, we cannot exclude the possibility that the oocyst excretion values reported in our study were affected by our sampling strategy, namely the use of pellet count method to collect rabbit faecal samples. As mentioned above, despite the effort to collect fresh pellets, the pellets collected may have been exposed for some time to weather conditions, leading to desiccation and therefore to an underestimation of oocyst excretion.

Irrespective of the potential sampling bias, PNSE clearly showed higher oocyst excretion values comparing to PNSACV (W = 377.5, $p = 0.0002$). Although we found no positive correlation between rabbit abundance and oocyst excretion within PNSE (S = 137.918, $p = 0.325$), and just a slight correlation in PNSACV (S = 360.912, $p = 0.0334$), rabbit abundance was significantly higher in PNSE than in PNSACV (W = 34, $p = 0.0082$). Hobbs *et al.* (1999) concluded that it is difficult to find evidences for the relation between oocyst counts and host density in rabbit populations in the wild, particularly in short-term studies. So we cannot rule out the possibility that the higher oocyst excretion described for PNSE could be due to the higher host abundance in the study area. Alternatively, the differences reported on oocyst excretion estimates of both study areas may be justified by their distinct climate. Our results agree with those presented by Stodart (1968), who described higher oocyst counts in colder and wetter climates (similar to the climate described for PNSE) than in Mediterranean climatic conditions (as those described for PNSACV).

To our knowledge, this study is the first to describe the composition of the *Eimeria* spp. assemblage in wild rabbit natural populations from the Iberian Peninsula, a region where this species is native and plays a keystone role (Ferrand 2008). In addition, our work highlights the importance of gathering information on the impact of coccidia in European rabbit populations, especially in a moment when understanding the potential interaction with viral diseases is particularly critical. In 2012, a new variant of the RHDV was found in several wild rabbit populations distributed across Spain and Portugal (Abrantes *et al.* 2013). Unlike the classical form of RHDV, this variant kills rabbits as young as 11 days old, and affects individuals vaccinated against classic RHDV (Abrantes *et al.* 2013; Dalton *et al.* 2014). The effects of this new variant have been devastating, with rabbit populations in the wild potentially reaching a new historical low in 2013 in Iberia (Delibes-Mateos *et al*. in press). The potential for an interaction between viral and parasitic diseases is particularly important at present, when rabbit populations may be further at risk raising concern for their conservation in this region. Moreover, correlation between seasonal dynamics of myxomatosis outbreaks and coccidiosis has been recently hypothesized (Bertó-Moran *et al*. 2013; Boag *et al*. 2013). Lastly, new molecular techniques to identify *Eimeria* species are now available (Oliveira *et al.* 2011) which will facilitate the investigation of these topics. Finally, we strongly recommend that parasite load estimations should be taken into consideration in rabbit monitoring programs.

Acknowledgments. Authors wish to thank F. Matos (PNSE) and J. Nunes (PNSACV), N. Ferreira, J. Vicente (IREC-UCLM, Spain) for support in different aspects of this work, and C. Gortázar Schmidt and an anonymous reviewer for providing suggestions to previous versions of this manuscript. We also thank D. Jenkins for reviewing the English. This work was partially funded by the Instituto da Conservação da Natureza e das Florestas (ICNF, Portugal). S.M. Silva currently holds a Programa Nacional de Pós Doutorado/Coordenação de Aperfeiçoamento de Pessoal de Nível Superior fellowship at the Department of Zoology in the Museu Paraense Emílio Goeldi/Universidade Federal do Pará. C. Ferreira currently holds a post-doctoral fellowship (Ref. SFRH/BPD/88643/2012) funded by the Fundação para a Ciência e a Tecnologia of the Ministério da Educação e Ciência, Portuguese government. J. Paupério is currently supported by Project "Genomics and Evolutionary Biology" co-financed by North Portugal Regional Operational Programme 2007/2013 (ON.2 – O Novo Norte), under the National Strategic Reference Framework (NSRF), through the European Regional Development Fund (ERDF).

References

Abrantes J., Lopes A.M., Dalton K.P., Melo P., Correia J.J., Ramada M., Alves P.C., Parra F., Esteves P.J. 2013. New variant of rabbit hemorrhagic disease virus, Portugal, 2012-2013. *Emerging Infectious Diseases*, 19, 1900–1902. DOI: 10.3201/eid1911.130908

Bertolino S., Wauters L.A., De Bruyn L., Canestri-Trotti G. 2003. Prevalence of coccidia parasites (Protozoa) in red squirrels (*Sciurus vulgaris*): effects of host phenotype and environmental factors. *Oecologia*, 137, 286–295. DOI: 10.1007/s00442-003-1345-x

Bertó-Moran A., Pacios I., Serrano E., Moreno S., Rouco C. 2013. Coccidian and nematode infections influence prevalence of antibody to myxoma and rabbit hemorrhagic disease viruses in European rabbits. *Journal of Wildlife Diseases*, 49, 10–17. DOI: 10.7589/2011-12-343

Boag B., Hernandez A.D., Cattadori, I.M. 2013. Observations on the epidemiology and interactions between myxomatosis, coccidiosis and helminth parasites in a wild rabbit population in Scotland. *European Journal of Wildlife Research*, 59, 557–562. DOI: 10.1007/s10344-013-0704-0

Dalton K.P., Nicieza I., Abrantes J., Esteves P.J., Parra, F. 2014. Spread of new variant RHDV in domestic rabbits on the Iberian Peninsula. *Veterinary Microbiology*, 69, 67–73. DOI: 10.1016/j.vetmic.2013.12.015

Delibes-Mateos M., Ferreras P., Villafuerte, R. 2009. European rabbit population trends and associated factors: a review of the situation in the Iberian Peninsula. *Mammal Review*, 39, 124–140. DOI: 10.1111/j.1365-2907.2009.00140.x

Delibes-Mateos M., Ferreira C., Carro F., Escudero M.A., Gortázar C. In press. Ecosystem effects of variant rabbit hemorrhagic disease virus, Iberian Peninsula. *Emerging Infectious Diseases*. DOI: 10.3201/eid2012.140517

Ferrand N. 2008. Inferring the evolutionary history of the European rabbit (*Oryctolagus cuniculus*) from molecular markers. In: (Eds. P.C. Alves, N. Ferrand and K. Hackländer) Lagomorph biology: Evolution, ecology and conservation. Springer, Berlin, Heidelberg, 47–63

Ferreira C., Alves P.C. 2009. Influence of habitat management on the abundance and diet of wild rabbit (*Oryctolagus cuniculus algirus*) populations in Mediterranean ecosystems. *European Journal of Wildlife Research*, 55, 487–496. DOI: 10.1007/s10344-009-0257-4

Foronda P., Figueruelo E., Ortego A., Abreu N., Casanova, J. 2005. Parasites (viruses, coccidia and helminths) of the wild rabbit (*Oryctolagus cuniculus*) introduced to Canary Islands from Iberian Peninsula. *Acta Parasitologica*, 50, 80–84

Gordon, H.M., Whitlock, H.V. 1939. A new technique for counting nematode eggs in sheep faeces*. Journal of the Council for Scientific and Industrial Research*, 12, 50–52

Hobbs R.P., Twigg L.E. 1998. Coccidia (*Eimeria* spp) of wild rabbits in southwestern Australia. *Australian Veterinary Journal*, 76, 209–210

Hobbs R.P., Twigg L.E., Elliot A.D., Wheeler A.G. 1999. Factors influencing the fecal egg and oocyst counts of parasites of wild European rabbits *Oryctolagus cuniculus* (L.) in Southern Western Australia. *Journal of Parasitology*, 85, 796–802

Jarvinen J.A. 1999. Prevalence of *Eimeria macusaniensis* (Apicomplexa: Eimeriidae) in midwestern *Lama* spp. *Journal of Parasitology*, 85, 373–376

Lello J., Boag B., Hudson, P.J. 2005. The effect of single and concomitant pathogen infections on condition and fecundity of the wild rabbit (*Oryctolagus cuniculus*). *International Journal of Parasitology*, 35, 1509–1515. DOI: 10.1016/j.ijpara.2005.06.002

Maldonado J., Pagés A., Alfonso M., Martínez E., Artigas C. 2004. Técnicas de aislamiento de *Clostridium spiroforme* a partir de casos de enteritis en conejos. In: *XXIX Symposium de Cunicultura de ASESCU*, 31 Mach – 1 April, 2004, Lugo, Spain, 85–87

Marchandeau S., Aubineau J., Baudron P., Blanchet L., Chauvet C., Legros E., Merlet C., Suret, H. 1999. La pathologie de Lapin du garenne dans l'Ouest de la France. *Bulletin Mensuel de l'Office National de la Chasse*, 245, 10–16

Oksanen J., Blanchet F.G., Kindt R., Legendre P., Minchin P.R., O'Hara R.B., Simpson G.L., Solymos P., Stevens M.H., Wagner, H. 2013. Vegan: Community Ecology Package. R package version 2.0–10. World wide web electronic publication. Available from: http://CRAN.R-project.org/package = vegan

Oliveira U.C., Fraga J.S., Licois D., Pakandl M., Gruber, A. 2011. Development of molecular assays for the identification of the 11 *Eimeria* species of the domestic rabbit (*Oryctolagus cuniculus*). *Veterinary Parasitology*, 176, 275–280. DOI: 10.1016/j.vetpar.2010.10.054

Pakandl M. 2009. Coccidia of rabbit: a review. *Folia Parasitologica*, 56, 153–166

Pellerdy LP. (Ed.) 1974. Coccidia and Coccidiosis. Verlag Paul Parey, Berlim, Germany, 657 pp.

R Development Core Team 2012. R: A language and environment for statistical computing. R Foundation for Statistical Computing. Vienna, Austria

RIPAC. 2004. *Rede de Recolha e Identificação de Patologias em Animais Cinegéticos –Relatório Final* 2004. Federação de Caçadores do Algarve e Direcção Regional de Agricultura do Algarve. World wide web electronic publication. Available from: http://www.fcalgarve.pt/rede.htm

Rosell J.M., de la Fuente L.F., Badiola J.I., Fernández de Luco D., Casal J., Saco M. 2009. Study of urgent visits to commercial rabbit farms in Spain and Portugal during 1997-2007. *World Rabbit Science*, 17, 127–136. DOI: 10.4995/wrs.2009.652

Stodart E. 1968. Coccidiosis in wild rabbits, *Oryctolagus cuniculus* (L.), at four sites in different climatic regions in eastern Australia I. Relationship with the age of the rabbit. *Australian Journal of Zoology*, 16, 69–85

Villafuerte R., Calvete C., Blanco J.C., Lucientes J. 1995. Incidence of viral hemorrhagic disease in wild rabbit populations in Spain. *Mammalia*, 59, 651–660. DOI: 10.1515/mamm.1995.59.4.651

Received: May 20, 2014
Revised: October 24, 2014
Accepted for publication: December 1, 2014

ERRATUM

Erratum to:

DOI: 10.1515/ap-2015-0049

Acta Parasitologica, 2015, 60(2), 350–355; ISSN 1230-2821

RESEARCH NOTE

Coccidiosis in European rabbit (*Oryctolagus cuniculus algirus*) populations in the Iberian Peninsula

Sofia Marques Silva[1,2]*, Catarina Ferreira[2,3,4], Joana Paupério[2], Rodolfo Miguel Silva[5], Paulo Célio Alves[2,6,7] and Armando Lemos[5]

[1]Departamento de Zoologia, Museu Paraense Emílio Goeldi/Universidade Federal do Pará, Belém, PA, Brazil; [2]Centro de Investigação em Biodiversidade e Recursos Genéticos, Universidade do Porto, InBIO Laboratório Associado, Vairão, Portugal; [3]Department of Biology, Trent University, Peterborough, Ontario, Canada; [4]Instituto de Investigación en Recursos Cinegéticos, Universidad Castilla La Mancha (IREC; CSIC-UCLM-JCCM), Ciudad Real, Spain; [5]Instituto de Ciençias Biomédicas Abel Salazar, Universidade do Porto, Vairão, Portugal; [6]Departamento de Biologia, Faculdade de Ciençias, Universidade do Porto, Porto, Portugal; [7]Wildlife Biology Program, University of Montana, Missoula, USA

The original version of the article unfortunately contained a mistake. The published sentence:

"**Acknowledgments.** Authors wish to thank F. Matos (PNSE) and J. Nunes (PNSACV), N. Ferreira, J. Vicente (IREC-UCLM, Spain) for support in different aspects of this work, and C. Gortázar Schmidt and an anonymous reviewer for providing suggestions to previous versions of this manuscript. We also thank D. Jenkins for reviewing the English. This work was partially funded by the Instituto da Conservação da Natureza e das Florestas (ICNF, Portugal). S.M. Silva currently holds a Programa Nacional de Pós Doutorado/Coordenação de Aperfeiçoamento de Pessoal de Nível Superior fellowship at the Department of Zoology in the Museu Paraense Emílio Goeldi/Universidade Federal do Pará. C. Ferreira currently holds a post-doctoral fellowship (Ref. SFRH/BPD/88643/2012) funded by the Fundação para a Ciência e a Tecnologia of the Ministério da Educação e Ciência, Portuguese government. J. Paupério is currently supported by Project "Genomics and Evolutionary Biology" cofinanced by North Portugal Regional Operational Programme 2007/2013 (ON.2 – O Novo Norte), under the National Strategic Reference Framework (NSRF), through the European Regional Development Fund (ERDF)."

should be corrected as follows:

Acknowledgments. Authors wish to thank F. Matos (PNSE) and J. Nunes (PNSACV), N. Ferreira, J. Vicente (IREC-UCLM, Spain) for support in different aspects of this work, and C. Gortázar Schmidt and an anonymous reviewer for providing suggestions to previous versions of this manuscript. We also thank D. Jenkins for reviewing the English. This work was partially funded by the Instituto da Conservação da Natureza e das Florestas (ICNF, Portugal). S.M. Silva currently holds a Programa Nacional de Pós Doutorado/Coordenação de Aperfeiçoamento de Pessoal de Nível Superior fellowship at the Department of Zoology in the Museu Paraense Emílio Goeldi/Universidade Federal do Pará. C.F. was funded by a Post-Doctoral grant (Ref. SFRH/BPD/88643/2012) from the Fundação para a Ciência e Tecnologia of the Ministério da Ciência, Tecnologia e Ensino Superior, Portuguese government, and by a Marie Curie Outgoing International Fellowship for Career Development (PIOF-GA-2013-621571) within the 7th Framework Programme of the European Union.