EAZWV
Heidelberg 2002 Meeting

ZOO HEIDELBERG

Conference Report for the fifth meeting of the European Wildlife Disease Association

EWDA EAZWV EAZA

General

Following an invitation from Dr. P. Dollinger, Secretary of the European Association of Zoo and Wildlife Veterinarians (EAZWV), the 5th EWDA meeting was held from 8 to 12 May 2002 together with the EAZWV at the German Cancer Research Center DKFZ in Heidelberg (Germany), with "Diseases in European Wildlife" as one of the subjects.

The business meeting of the European Section of the Wildlife Disease Association (EWDA) was called to order by the Secretary, Christian Gortázar at about 2 p.m. on Friday, 10 May 2002 at the DKFZ facilities. In attendance were the Board-members and the delegates that attended the 5th EWDA meeting at Heidelberg. C. Gortázar informed the members about the health of the Chairman, Torsten Mörner, and wished him a speedy recovery. He also explained the new position of Dr Mörner, as Vice-President of the Wildlife Disease Association. Finally, C. Gortázar gave a general explanation of the special nature of EWDA membership (without fees or other formal documents) and the relationship of the EWDA and the WDA.

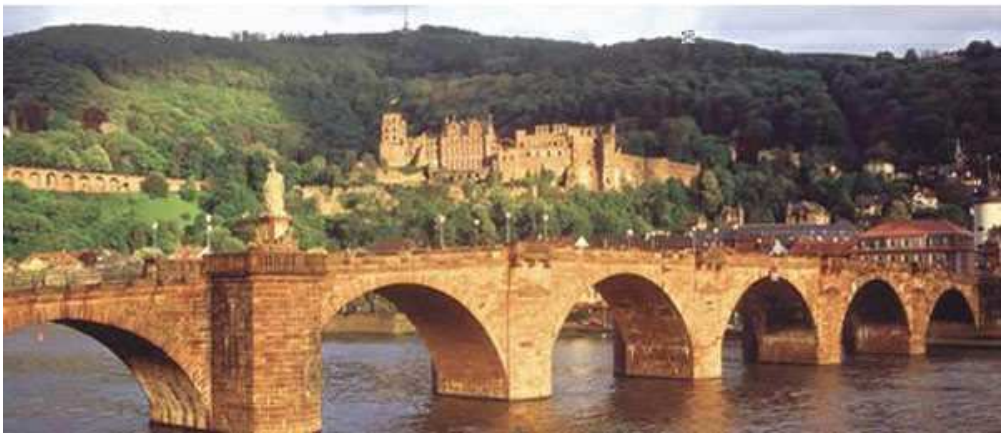
Presentations

The papers and posters presented by EWDA members at Heidelberg are listed in the table below. Abstracts of these presentations (in pdf format) can be viewed or downloaded by clicking on the appropriate title.

LEAD AUTHOR	TITLE
Alessandra Piccirillo (S)	A re-emerging disease of hares (*Lepus europaeus*):hare fibromatosis.
Alonso Aguirre	Fibro-sarcoma in a green turtle.
Paolo Tizzani	Presence of infectious agents and parasites in wild populations of Cottontails (*Sylvilagus floridanus*).
L. Belloy	Molecular epidemiology of *Mycoplasma conjunctivae* in Caprinae: transmission across species in natural outbreaks.
A. Caron (S)	Relation between bovine TB prevalence and herd-level indicators in Buffalo in the Kruger National Park.
Christian Gortázar	Rabbit viral diseases: recombinant vaccines or natural recovery?
Dolores Gavier-Widen	*Mycobacterium tuberculosis* infection in Asian elephant.
Elvira Schettler	Severe feather loss and abnormalities ('pinching off') in a juvenile white–tailed eagle from Northern Germany.
Emanuelle Fromont	Spatio-temporal variations in seroprevalence of Chlamydiosis and Q fever in mountain ungulate populations: retrospective detection
Geraldine Boseret (S)	Septicaemic infection in a harbour porpoise.
Ignasi Marco	Noninvasive monitoring of cortisol metabolites in captive and free-ranging roe deer *(Capreolus capreolus*).
Ignasi Marco	Haematologic and serum biochemistry values of the Southern Chamois (*Rupicapra pyrenaica pyrenaica*) captured by drive-net.
Ignasi Marco	Two cases of Pineal Meningioma in free-ranging Southern Chamois (*Rupicapra pyrenaica*).
Javier Millan (S)	Marked differences in the nematode fauna between wild and farmed red-legged partridges.
Javier Millan	First record of *Haemoproteus* sp. parasitising red-legged partridge (*Alectoris rufa*).
Jean Hars	The annual serological survey in wild boar populations in France 2000-2001 Report.
Joaquín Vicente (S)	TB pathology in the European wild boar.
Joseph S. Masangkay	Tumours of the carapace of the Philippine Green Sea Turtle (*Chelonia mydas*).
Kai Frölich	First report of European Brown Hare Syndrome (EBHS) in brown hares from Switzerland and Argentina.
Marc Artois	Puumala (Hantavirus) epizootiology in the Ardennes region (France).
Marc Artois	Surveillance and control of wildlife diseases in Europe.
Marino García	Postmortem findings in wild bustards (*Otis tarda*) from Spain: a clinical approach.
Martin Janovsky	*Mycoplasma conjunctivae* is maintained in domestic sheep but not in Alpine Chamois in the Swiss alps.
Nicole Latz (S)	Canine parvovirus infection in free-ranging carnivores from Germany.
Oliver Krone	The liver fluke *Metorchis bilis* - A new threat for the White-tailed Sea Eagle in middle Europe?
Paul Duff	'Wildlife disease in the UK - a short history.
Richard Delahay	Bovine TB in British wild mammals.
Richard Delahay	Applications of GIS in the Research and Management of Wildlife diseases.
Richard Delahay	The spatio-temporal distribution of *Mycobacterium bovis* in a badger population.
Richard Delahay	Use of farm buildings by badgers and associated risks of bovine TB transmission to cattle.
S. Rehbein	Endoparasites of Red Deer *(Cervus elaphus L.)* of Northrhine-Westfalia (Germany).
Sophie Rossi (S)	Survey of brucellosis involving Brucella suis biotype 2 in wild boars.
Marie-Eve Terrier	Abnormal mortality in roe deer.
Thijs Kuiken	Pathology of common eiders from the Dutch Wadden Sea in winter 1999-2000.
Torsten Mörner	Diseases and mortality in free ranging lynx, brown bear, wolf and wolverine in Sweden.
Ursula Höfle	Mycoplasma and Avian Polyoma virus infection in captive Spanish Imperial Eagles.
Vic Simpson	Dental lesions and bite wounds in Eurasian Otters (*Lutra lutra*).
Vittorio Guberti	Pseudorabies in wild boar.
Vittorio Guberti	Wolf mortality and demography in Italy.
Walter Glawischnig	*Mycobacterium avium subsp. avium* infection in red deer (*Cervus elaphus hippelaphus*) from Austria.

Student Awards

As decided in Zaragoza 2000, three awards were given by the Treasurer, Marc Artois, to the best student presentations at Heidelberg 2002. All papers entered for the award are denoted in the table above by the letter 'S', including the three prize winning entries. The first prize was given to Alex Caron, and two second awards were shared by Sophie Rossi and Nicole Latz. The high quality of all EWDA student presentations was emphasised.



EWDA Board

A proposal for a new EWDA Board and the establishment of an EWDA Advisory Board was discussed and accepted with general agreement. The following members were elected - M. Artois, Chairman; K. Frölich, Vice-Chair; U. Höfle, Treasurer; C. Gortazar, Secretary; P. Duff, Newsletter Editor; R. Delahay, Web-site responsibilities. The members of the EWDA Advisory Board are G. Boseret, D. Gavier-Widen, M. Giacometti, T. Kuiken, T. Mörner and R. Orusa.

Website

Richard Delahay covered progress in setting up the EWDA website. This now has several items with more about to be introduced in the near future, and Richard will be helped by Marco Giacometti. **Could all EWDA members who would like their details on the web-site please send Richard or Marc their name, institute and research interests; their conference abstract and any other item for the website.**

EWDA Working Groups

Marc Artois suggested three particular areas where the EWDA might concentrate efforts in the future. (1) to improve wildlife disease surveillance in Europe in order to get a more formal way of reporting and surveying wildlife disease. (2) to develop European wildlife disease research, including possible topics such as tuberculosis, hares, wild boar, foxes, or migration and diseases. (3) to review the current teaching in wildlife diseases in European countries, in order to produce wildlife disease teaching support. M. Artois, K. Frölich and C. Gortázar, respectively, accepted positions to take the leads in these three fields.

Chairman's Address

Dear colleagues and EWDA fellows,

It is for me a great honor to become chairman of the WDA European Section board. I thank you all for your support. It is nevertheless a hard task to succeed Torsten MORNER who has created this Section and led it for so many years with enthusiasm. It has been Torsten's honor to obtain recognition of our group both at the European level and overseas. When this story started in the early 90's, the few of us who were already involved in wildlife disease studies in Europe were persuaded to join the crew with enthusiasm. Since then many new people have been attracted to the organization and enrolled. They are now its rich resource and are prepared for the new challenges ahead.

In the name of all of us, I would thank Torsten for his dedication. I wish him further success in his new duties. We trust that Torsten will remain a supporter and a pre-eminent active member of our Section, mainly acting for the enforcement of our relationship with the main-section of the WDA in North America.

At the same time, the European Union is now increasing its partnership to new country members and one can expect that exciting new trails will be explored by our European Section. But, since on the one hand we are not yet formally organized and on the other, not so numerous, we have to be modest and remain realistic - although to be lucid does not impede creativity and innovation! It is my guess that the citizens of this continent, biologists and wildlife managers as well as our veterinary officers, would expect us, wildlife disease specialists, to organize ourselves in three main domains: - improving wildlife disease monitoring, developing sound science research projects and implementing specific education programs. During the coming years as EWDA chairman, I would like to spend my energy in approaching these three goals, with your help and with contributions from the new board members. I trust that our imagination and involvement shall soon allow our organization to find the necessary official support to achieve these goals. I am confident that our next meeting (possibly in Scandinavia) will give us an opportunity to measure our progress.

Thank you all again for coming to Heidelberg. Thanks to the organizers of this meeting, namely the chairman of our scientific committee, Christian GORTAZAR, to have offered us this nice opportunity to meet each other and share some interests with our zoo colleagues. Congratulations to all the students, who entered the student award, for their excellent presentations. I wish you all a safe trip back home and look forward to meeting you again soon.

Prof. Marc ARTOIS

Our next meeting?

It was suggested that the next EWDA meeting could be in a Scandinavian country, perhaps in Uppsala, Sweden. After the Heidelberg meeting, which was fully organised by the EAZWV without any EWDA input, the members underlined their wish for the next meeting to be organised by the EWDA and to focus on wildlife disease rather than on problems of animals in captivity.

European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting,
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May 8-12, 2002, Heidelberg, Germany.

LOW GRADE FIBROSARCOMAS IN GREEN TURTLES; IS FIBROPAPILLOMATOSIS GOING AMUCK?

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Abstract

The green turtle (*Chelonia mydas*) is protected under the U.S. Endangered Species Act and the Wildlife Laws of the State of Hawaii. Fibropapillomatosis (FP) is a disease of marine turtles characterized by multiple cutaneous masses ranging from 0.1 to more than 30 cm in diameter that has primarily affected green turtles. The disease has a worldwide distribution and has been observed in all major oceans and all species of marine turtles that are considered endangered of extinction. Where present, prevalence of the disease varies among locations, ranging from as low as 1% to as high as 90%. Although several viruses have been identified associated with the tumors, including herpesviruses, a retrovirus and a papilloma-like virus, the primary etiological agent remains to be isolated and identified. Concurrent infections of FP and cardiovascular trematodiasis have been recognized as the most important mortality factors of Hawaiian green turtles considerably reducing the survival of the species. The neoplastic processes previously observed by our previous studies and more recently during gross and histopathologic examination of 14 turtles collected in the Hawaiian Islands with FP suggested a synergistic effect of cardiovascular trematodes and the primary agent of FP. Tumors in the internal organs of some turtles were characteristic of fibropapillomas, fibromas, myxomas, and low-grade fibrosarcomas. This study suggested that when occurring together, spirorchidiasis and GTFP represent a debilitating and fatal syndrome of Hawaiian green turtles.

We describe the histopathology of cutaneous and internal spindle cell tumors found in green turtles from the Hawaiian Islands, and present histopathological and molecular evidence of the presence of low-grade fibrosarcomas. Histologically, some tumors of the nasopharynx, mouth and temporomandibular tissues appear to have an aggressive, invasive behavior. These masses are well demarcated from adjacent tissues but demonstrate infiltration of surrounding stroma and bone lysis. Although there is no evidence of vascular invasion or high mitotic activity, these tumors have been classified as low grade fibrosarcomas. The precancer to cancer sequence in the progression of benign to malignant tumors has been documented in other species with similar tumors, e.g. papillomas transforming into squamous cell carcinomas.

Although there is no evidence of vascular invasion or high mitotic activity, further research is necessary to demonstrate whether the visceral lesions are the result of metastasized external papillomas, or indeed are multiple independent processes. In other species, the precancer to cancer sequence in the progression of canine oral papilloma to carcinoma has been documented. We are trying to establish the biological behavior and molecular characterization of these tumors. Current retrospective and prospective studies are in progress to determine the implications of these novel findings.

Zusammemnfassung (??)

Résumé (??)

Key words: green turtle, fibropapilloma, fibrosarcoma, Hawaiian Islands

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European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting, joint with the annual meeting of the European Wildlife Disease Association (EWDA) May 8-12, 2002, Heidelberg, Germany.

PUUMALA (HANTAVIRUS) EPIZOOTIOLOGY IN THE ARDENNES REGION (FRANCE)

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Abstract

Human cases of the so called " haemorrhagic fever with renal syndrome" (HFRS) were routinely recorded at the CHR (regional hospital) of Charleville, (Ardennes) France. This hospital has registered the maximum number of human disease cases in this country. Epidemic outbreaks of acute infection have been recorded every third year, on a regular basis since the year 1990.

During the three years preceding the last peak of the disease (1999), Bank vole (*Clethrionomys glareolus*) the rodent reservoir, was monitored by trapping and antibody screening. Voles were box-trapped on standard trap-lines in a randomised selection of wooded habitats near the city of Charleville. This surveillance was aimed to test the hypothesis that the population level increase in the reservoir will amplify the virus prevalence in voles, and the infection hazard in exposed human beings afterward.

If the sampled rodents are representative of the whole population of reservoir to which humans are exposed, the data are in accordance with a synchrony of the infection rate of both species during the three years epidemiological cycle. But the assumption that the vole demographic population peak precedes the epidemic outbreak in humans is not supported by the data.

During the whole study, the demography of the monitored vole population can be described by a global trend of increase (x by four) with seasonal fluctuations (top in September and bottom in spring). But the prevalence rate reached a maximum 8/10 % level already during the autumn of 1998 (year 2) and remained at this level up to the population peak in September 1999. It can be speculated that the amount of available virus in voles was at its highest level in between September 1998 and 1999.

In accordance with previous records, the greatest number of HFRS was registered during this period at Charleville CHR. Provided that the data of this preliminary study are accurate, the temporal correlation between the infection rates in the human victim and the reservoir host strongly suggest a common source of infection. We hypothesise that mechanism of virus excretion is related to the social structure of the reservoir. In combination with an indirect transmission route it can explain the observed infection figure in voles. These hypotheses are to be challenged during ongoing epidemiological studies.

Zusammenfassung(??)

Résumé (??)

Key words:

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SURVEILLANCE AND CONTROL OF WILDLIFE DISEASES IN EUROPE

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Key words : Infectious diseases – Control – Europe – Wildlife

Abstract

During the last thirty years new epidemiological patterns have emerged as free-ranging wildlife have become progressively more involved in the epidemiology of both common and emerging infectious diseases of humans and domestic animals. This has been seen in rabies, bovine tuberculosis and more recently in wild-boar classical swine fever. Emerging diseases are of interest for veterinarians as well as public health officials. Attempts to control these diseases have not always been successful as in wildlife populations control of either host or pathogen can present particular problems. Lessons should be learnt from previous experiences to help in the management of new emerging diseases in the future.

Zusammenfassung (??)

Résumé (??)

ERYSIPELOTHRIX RHUSIOPATHIAE INFECTION IN STRANDED HARBOUR PORPOISE (*Phocoena phocoena)* AND HARBOUR SEAL *(Phoca vitulina).*

G.BOSERET *, T.JUNIAUX# , J.MAINIL *

Abstract :

An adult female harbour porpoise (*Phocoena phocoena)* and a juvenile male harbour seal have been found dead on a Belgian beach in autumn 2001. The two bodies were in good condition. Pure and abundant growth of a small rod-shaped, Gram-labile bacterium was obtained aerobically and anaerobically on Columbia blood-agar from the heart blood, the mouth, the pharynx, the lungs, the intestine and the anus of the porpoise, and from the intestine, the pharynx, the mouth, the nose and the anus of the seal. The colonies were surrounded by a narrow zone of alpha-hemolysis. The catalase- and peroxydase-tests gave negative results.
Rapid ID 32 Strepto (Biomérieux, France) sugar tests applied on porpoise's heart blood, lungs and intestine, and on seal's intestine and pharynx identified this isolate to *Erysipelothrix rhusiopathiae*.
E.rhusiopathiae is not reported as a common cause of infection and death in wild cetaceans and wild pinnipeds in opposite to respectively captive dolphins and sea lions.
Nevertheless, *E.rhusiopathiae* can be considered as the cause of death of the stranded harbour porpoise as it was present in heart blood and internal organs, and the seal was carrying the bacterium with lesions of enteritis which coulg be associated with *E.rhusiopathiae.*

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BOVINE TB IN BRITISH WILD MAMMALS.

R. J. DELAHAY

Bovine tuberculosis caused by *Mycobacterium bovis* is a zoonotic infection with a wide range of mammalian hosts. In parts of the UK *M. bovis* infection in cattle is a persistent problem. The European badger (*Meles meles*) is implicated in the transmission of *M. bovis* to cattle, and is widely believed to constitute the most important reservoir of infection in British wildlife. TB is endemic in the British badger population. Infected individuals may excrete bacilli in sputum, faeces, urine and/or pus from wounds and burst abscesses. However, infected badgers can survive for relatively long periods and breed successfully, and the impact of the disease on population size and structure appears to be minimal. The foraging habits of badgers may bring them into close contact with pasture and farm buildings, providing opportunities for disease transmission to cattle. However, few studies have been carried out on the status of *M. bovis* infection in other wild mammals in the UK. Infection has been identified in several species of deer, foxes, moles, rats, mink, a feral ferret, farm cats and a field vole. Although the evidence from these studies does not support the existence of a significant self-maintaining reservoir of infection in any wild mammal other than the badger, there is a clear lack of sufficient data to rule out the involvement of other species. This paper describes the methodology and preliminary findings from a contemporary survey for *M. bovis* infection in wild mammals in south-west England, and discusses its limitations and the interpretation of results.

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USE OF FARM BUILDINGS BY BADGERS (*MELES MELES*) AND ASSOCIATED RISKS OF BOVINE TB TRANSMISSION TO CATTLE.

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Key words: (??)

The extent of farm visits by badgers (*Meles meles*) and the consequent opportunities for transmission of *Mycobacterium bovis* (bovine TB) to cattle were investigated on dairy and beef farms at Woodchester Park, Gloucestershire, England. Home range use of infected and uninfected badgers was also compared. Two infected badgers were radio-tracked within social group territories containing cattle farms. The core activity areas of both badgers included farm buildings. One infected badger had two such core areas representing 17% of her active fixes. A third infected badger that could not be collared was also observed in farm buildings. Feed sheds, cattle sheds, slurry pits, cattle troughs, barns, haystacks and farmyards were used by at least 26 badgers for a range of food items. Visits were most frequent on dry nights, when farms provided alternative food sources to earthworms deep in the ground. Foraging at cattle feed accounted for 56% of visits. Within buildings badgers came into close proximity of sick, injured or pregnant cattle, and calves housed overnight. Badger faeces were found in cattle feed on troughs and on the ground, and both farms suffered herd breakdowns in the same year. Cattle trough climbing experiments revealed that badgers are capable of climbing up to 115cms, well beyond the current safety guideline of 80cms. These visits to farm buildings and facilities may constitute a serious risk of *M. bovis* transmission to cattle and other badgers. Where such visits occur, improvements to farm security and husbandry could be effective means of controlling badger to cattle transmission of bovine TB.

Zusammenfassung(??)

Résumé (??)

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TITLE WILDLIFE DISEASE IN THE UK: A SHORT HISTORY

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Key words: (??)

Abstract

This paper first discusses the history of wildlife disease in the UK, why the subject should be examined, sources of information and the possible significance of wildlife disease in the UK down the centuries.

The history of wildlife disease is examined in three periods – pre 1800, 1800-1900 and 1900 to the present day. The diseases considered are then discussed in three general categories, zoonotic disease, diseases of importance to domesticated stock and thirdly, diseases of importance to wildlife, social structures and the environment.

A further aspect that will be considered are the institutes, organisations and scientists that have been involved in wildlife disease investigations in more recent years. The development of investigation techniques is followed together with discussion on the current status of wildlife investigation in the United Kingdom.

ZusammenfassungJ (??)

Résumé (??)

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FIRST REPORT OF EUROPEAN BROWN HARE SYNDROME VIRUS (EBHSV) IN FREE-RANGING EUROPEAN BROWN HARES (*LEPUS EUROPAEUS*) FROM SWITZERLAND AND ARGENTINA

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Key words (??)

Extended Abstract

In 1989, Lavazza and Vecchi (1989) found viral particles in European brown hares (*Lepus europaeus*) which had died from European brown hare syndrome (EBHS) by negative staining immune electron microscopy of the liver. The causative agent of EBHS is a small (30 to 35 nm) icosahedral, non-enveloped and hemagglutinating virus (Gavier-Widen and Mörner, 1991) and is classified as a calicivirus (Ohlinger and Thiel, 1991). European brown hare syndrome has been demonstrated in European brown hares and in mountain hares (*Lepus timidus*), and has been reported in many European countries (Morisse, 1988; Eskens and Volmer, 1989; Marcato et al., 1989; Henriksen et al., 1989; Okerman et al., 1989; Chasey and Duff, 1990; Sostaric et al., 1991; Gavier-Widen and Mörner, 1993; Salmela et al., 1993; Steineck and Nowotny, 1993; Nauwynck et al., 1993; Gortazar and de Luco 1995; Frölich et al., 1996; Slamecka et al., 1997). However, until now neither EBHSV-antigen nor corresponding pathology has been found in lagomorphs in Switzerland. Only one report exists for antibodies against caliciviruses in European brown hares in this country (Büttner, 1996). Our objective was to determine, whether free-ranging European brown hares and mountain hares were naturally infected by EBHSV in Switzerland. From 1997 to 2000, complete necropsy and histopathological investigations were performed on 157 free-ranging European brown hares found dead throughout Switzerland. Organ samples of all these individuals (157 livers and 107 spleens available) were tested for European brown hare syndrome virus (EBHSV)-antigen by enzyme-linked immunosorbent assay (ELISA) test kit (Frölich et al., 1996). Furthermore, 60 blood samples available were tested for antibodies against EBHSV by ELISA. In addition, liver samples of 87 free-ranging mountain hares hunted in 1996 were tested for EBHSV-antigen. In two European brown hares from southern Switzerland lesions suggestive of changes induced by EBHSV were present, and high titers of EBHSV-antigen were detected in both liver and spleen samples of the same animals. The two liver samples were positive up to a dilution of 1: 1,000 and the two spleen samples were positive when diluted 1: 10,000 and 1:1,000, respectively. Major histological lesions were restricted to the liver parenchyma and characterized by midzonal- and perilobular, large extensive to coalescing areas of coagulative and lytic necrosis and partial replacement of the parenchyma by erythrocytes. Additional lesions were characterized by multiple subendocardial hemorrhages and tubular nephrosis. Based on negative staining electron microscopy investigations of liver and spleen homogenates, we observed calicivirus in one antigen-positive hare. These viral particles morphologically resemble those observed by Poli et al. (1991) in spleen and liver of hare with acute hepatic necrosis. Low EBHSV-antigen titers were found in three additional European brown hares from central and western Switzerland, but EBHS-

lesions were absent. Antibodies against EBHSV were not detected in any of the sera of European brown hares, and EBHSV-antigen was not found in the samples of mountain hares. The two individuals showing EBHS originated from a region south of the Alps. This local subpopulation unit is part of the European brown hare metapopulation of northern Italy, where the disease is known to be widespread (Scicluna et al., 1994).

EBHS has been reported in many different European countries (Frölich et al., 2001) but until now it was not known outside Europe (Lenghaus et al., 2001). In 1888, imported European brown hares were released for hunting in the province of Santa Fé in Argentina. Genetic studies on European brown hares from Argentina might indicate their European origin (Faber, pers. comm.). However, detailed genetic investigations about the origin of European brown hares from Argentina do not exist until now. Due to the presence of optimal habitat the hare populations increased rapidly and in 1907 the European brown hare was officially declared a pest. As a consequence, commercial hunting started in the early 1930's (Kujawski, 1998). Our objective was to determine whether European brown hares in Argentina were naturally infected with EBHSV. From 1998 to 2000, serum samples of 80 shot European brown hares from Argentina were examined for antibodies against European brown hare syndrome virus (EBHSV) and 80 spleen samples were investigated for EBHSV-antigen by enzyme linked immunosorbent assay (ELISA). Nine hares were positive for EBHSV-antigen and antibodies against EBHSV were detected in only one individual. Based on negative staining electron microscopy investigations of spleen homogenates, we observed calicivirus in one of five EBHSV-antigen-positive hares. However, EBHS has not been reported to have caused any mortality. Consequently, a less pathogenic variant of EBHSV may be present in the population which does not cause clinical symptoms suggestive of EBHS.

In conclusion, this is the first report of EBHS in Switzerland and of specific antibodies to EBHSV, EBHSV-antigen and electron microscopy findings in free-ranging European brown hares from South America.

Zusammenfassung(??)

Résumé(??)

References

1. Büttner S. (1996): Gesundheitszustand der Feldhasen (*Lepus europaeus*) in der Schweiz. Thesis, Veterinary Medical University of Berne, Berne, Switzerland, 99 pp.
2. Chasey. D and P. Duff (1990): European brown hare syndrome and associated virus particles in the UK. The Veterinary Record 126, 623-624.
3. ESKENS, U., and K. VOLMER. 1989. Untersuchungen zur Ätiologie der Leberdystrophie des Feldhasen (*Lepus europaeus*, Pallas). Deutsche Tierärztliche Wochenschrift 96: 464-466.
4. FRÖLICH, K., H. H. D. MEYER, Z. PIELOWSKI, L. RONSHOLT, S. v. SECK-LANZENDORF, and M. STOLTE (1996): European brown hare syndrome in free-ranging hares in Poland. Journal of Wildlife Diseases 32, 280-285.
5. FRÖLICH, K., G. HAERER, L. BACCIARINI, M. JANOVSKY, M. RUDOLPH, and M. GIACOMETTI. 2001. European brown hare syndrome in free-ranging European brown and Mountain hares from Switzerland. Journal of Wildlife Diseases. 37, 803-807.
6. GAVIER-WIDEN, D., and T. MÖRNER (1991): Epidemiology and diagnosis of the European brown hare syndrome in Scandinavian countries: Revue Scientifique et technique. Office International des Epizooties 10, 453-458.
7. GAVIER-WIDEN, D., and T. MÖRNER (1993): Descriptive epizootiological study of European brown hare syndrome in Sweden. Journal of Wildlife Diseases 29, 15-20.
8. GORTAZAR, C., and D. F. de LUCO (1995): La enfermedad hemorrgica de la liebre. Trofeo 295, 30-34.
9. HENRIKSEN, P., D. GAVIER, and F. ELLING (1989): Acute necrotising hepatitis in Danish farmed hares. The Veterinary Record 125, 486-487.
10. KUJAWSKI, O. GRAF. 1998. Die neue Wild Küche. 2. Aufl., Leopold Stocker Verlag, Graz, Germany, 72 pp.
11. LAVAZZA, A., and G. VECCHI (1989): Osservazioni su alcuni episodi di mortalità nelle lepri. Evidenziazione al microscopio elettronico di una particella virale. Nota preliminare. Selezione Veterinaria 30, 461-467.
12. LENGHAUS, C., M. J. STUDDERT, AND D. GAVIER-WIDEN. 2001. Calicivirus infections. In Infectious diseases of wild mammals, 3nd Edition, E. S. Williams, and I. K. Barker (eds.). Iowa State University Press, Ames, Iowa, pp. 280-291.
13. MARCATO, P. S., C. BENAZZI, M. GALEOTTI, and L. SALDA DELLA (1989): Infective necrotic hepatitis of leporids. Rivista di Coniglicultura 26, 41-50.
14. MORISSE, J.-P. (1988): Hemorrhagic septicemia syndrome in rabbits: First observations in France. Le Point Vétérinaire 20, 79-83.

15. NAUWYNCK, H., P. CALLEBAUT, J. PEETERS, R. DUCATELLE, and E. UYTTEBROEK (1993): Susceptibility of hares and rabbits to Belgian isolate of European brown hare syndrome virus. Journal of Wildlife Diseases 29, 203-208.
16. OHLINGER, V. F., and H. J. THIEL (1991): Identification of the viral hemorrhagic disease virus of rabbits as a calicivirus. Revue Scientifique et technique. Office International des Epizooties 10, 311-323.
17. OKERMAN, L., P. van de KERCKHOVE, S. OSAER, L. DEVRIESE, and E. UYTTEBROEK (1989): European brown hare syndrome in captive hares (Lepus capensis) in Belgium. Vlaams Diergeneeskundig Tijdschrift 58, 44-46.
18. POLI, A., M. NIGRO, D. GALLAZZI, G. SIRONI, A. LAVAZZA, and D. GELMETTI. 1991. Acute hepatosis in the European brown hare (*Lepus europaeus*) in Italy. Journal of Wildlife Diseases 27, 621-629.
19. SALMELA, P., K. BELAK, and D. GAVIER-WIDEN (1993): The occurrence of European brown hare syndrome in Finland. Acta Veterinaria Scandinavia 34, 215-217.
20. SCICLUNA, M. T., A. LAVAZZA, and L. CAPUCCI. 1994. European brown hare syndrome in northern Italy: results of a virological and serological survey. Revue Scientifique et Technique, Office International des Epizooties 13, 893-904.
21. SLAMECKA, J., P. HELL, and J. RATISLAV (1997): Brown hare in the Westslovak Iowland. Acta Scientarium Naturalium Academiae Scientarum Bohemicae Brno 31, 1-115.
22. SOSTARIC, B., Z. LIPEJ, and R. FUCHS (1991): The disappearance of free living hares in Croatia: 1. European brown hare syndrome. Veterinarski Arhiv 61, 133-150.
23. STEINECK, TH., and N. NOWOTNY (1993): European Brown Hare Syndrome (EBHS) in Östereich: Epizootiologische Untersuchungen. Tierärztliche Umschau 48, 225-229.

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POSTMORTEM FINDINGS IN WILD GREAT BUSTARDS (*Otis tarda*) FROM SPAIN:A CLINICAL APROACH.

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Abstract

Causes of death and morbidity are reported for 13 free-living great bustards (Otis tarda) from Spain. The main mortality cause for the adult birds and two juveniles was collision with power lines or fences. One wild adult bustard died of Aspergillus fumigatus generalised infection. Other causes of death for juveniles were predation, septicemia, parasitic obstruction of small intestine by cestodes, ventriculus impaction, and trauma with agricultural vehicles. Causes of morbidity were skin injuries, fractures, soft tissue and liver trauma, pneumonia of different etiology, ectoparasites and hemoparasites.

Zusammenfassung(??)

Résumé(??)

Key words: Great bustard, *Otis tarda*, mortality, morbidity, postmortem.

Introduction

There are 22 species of bustards (family Otididae), some of them are endangered species. The great bustard (*Otis tarda*) is a large, highly sexually dimorphic, globally endangered bird. Their numbers have declined considerably during the present century and current populations inhabit cereal steppes of Europe and Asia (2). This decline has been attributed to habitat changes caused by human population growth, farming practices, changes in agricultural practices and hunting pressure (1).

The Iberian population is the largest, being the size of the Spanish population at around 17.000-19.000 birds. Most of the Spanish great bustard nuclei seem to be stable perhaps with a very slight tendency to increase in some particularly well conserved areas (1).
Other bustard species (*Chlamydotis undulata, Ardeotis kori, Eupodotis ruficrista…*) have attracted much attention from the veterinary profession, especially in the Middle East, where a good amount of information about management and clinical aspects of bustards exists (3-7,9,12,17).
Most of the references regarding great bustards are based on ecological studies, and there are some published papers on captive great bustard diseases(13-15) but the authors could only found three references on veterinary aspects (parasites) of free-living great bustards (10,11,16), none of them being published in an international journal.
Clinical management of great bustards is not an easy task due to the strength, weight, present injuries and stressful nature of these birds. The knowledge of diseases affecting free-living great bustards could help in the daily treatment and follow up of clinical cases. The aim of this retrospective study is to provide an overview of morbidity and mortality causes in the wild Spanish great bustard population.

Material and method

We included for this retrospective study 13 postmorten cases of free-living great bustards (6 adults and 7 juveniles) that were necropsied in the period 1998-2001. Five of the 6 adults and 3 of the 7 juveniles were males. In one juvenile we could not determinate the sex. Included in this paper are only free-living birds that were found dead or died within 24 hours in the rehabilitation centre.
Standard avian postmorten examination techniques were used(8), including two X-ray projections, samples for histopathology, parasitology, and microbiology when appropriate and biometry. The condition of each bird was recorded as emaciated, poor, fair, good or obese, based on the degree of pectoral muscle wasting. Birds with emaciated and poor pectoral muscle scores were included in the pectoral muscle wasting group (4). For histopathology special stains were used when *Chlamydophila* spp. or tuberculosis were part of the differential diagnosis.
Cause of death was determined from consideration of the clinical history, clinical observations, laboratory findings, and significant postmortem findings.
Endoparasites were washed in distilled water and preserved in 70% ethanol until processed for identification. When possible a blood smear was prepared to search for hematozoa and faeces were screening by flotation method with zinc sulphate. Arthropods collected for identification were fixed and stored in 70% ethanol.

Results

Causes of death in wild great bustards are summarised in table 1.

Table 1. Causes of death in 7 juvenile and 6 adult free-living great bustards.

Cause of death	Adult	Juvenile	Total
Trauma-electric transmission power lines, fences	5	2	7
Trauma-vehicle	0	1	1
Aspergillosis	1	0	1
Predation	0	1	1
Parasitic obstruction of small intestine by cestodes	0	1	1
Septicemia	0	1	1
Gizzard impaction	0	1	1
Total	6	7	13

Trauma trough collisions with electric power lines or fences was responsible for 83.3% of adult and 42.9% of juvenile deaths in wild great bustards. Aspergillosis accounted for 16.7% of adult mortality. Other mortality causes of juvenile birds were predation (14.3%), septicemia (14.3%), gizzard impaction (14.3%) and obstruction of small intestine with cestodes (14.3%).
Other postmortem findings in the present study are summarised in table 2.

Table 2. Postmortem findings in 7 juvenile and 6 adult free-living great bustards.

Postmortem finding		Adult	Juvenile	Total
Intestinal parasitism-cestodes		6	4	10
Trauma-skin		5	4	9
Fractures		4	3	7
	humerus	1	1	2
	radius	1	0	1
	ischium	1	0	1
	femur	0	1	1
	tibiotarsus	0	1	1
	pubic bone	0	1	1
	scleral ring	1	0	1
	skull	1	0	1
Pneumonia		4	3	7
	bacteria	0	2	2
	fungus	1	0	1
	aspiration	3	1	4
Pectoral muscle wasting		2	4	6
Hemoparasites		2	2	4
Ectoparasites		2	1	3
Trauma-liver		2	1	3
Intestinal parasitism-nematodes		1	1	2
Sternal bone deformity		1	0	1

All the adult bustards (100%) and 57.1% of juveniles presented intestinal parasitism by cestodes. These cestodes were identified as *Otiditaenia conoides* and *Idiogenes otidis.* The nematodes present in the caecum of one adult and one juvenile bird were identified as *Heterakis isolonche*.
Also in the faeces of that juvenile *Capillaria* spp. and *Trichostrongylus* spp. eggs were detected, but no adults were recovered.
Skin injuries were a morbidity cause in 83.3% of adult and 57.1% of juvenile great bustards.
Bone fractures occurred in 66.7% of adult birds and 42.9% of juveniles, being open fractures of the humerus the most common fractures.
Pneumonia was observed in 66.7% of adult and 42.9% of juvenile birds. Foreign body inhalation pneumonia was recorded for 50% of adult bustards and 14.3% of juvenile birds. Pneumonia of bacterial origin (*Pasteurella* spp.) was found in 28.6% of juvenile bustards, and *Aspergillus fumigatus* pneumonia and air sacculitis was responsible for the death of one adult (16.7%) great bustard.
Hemoparasites of the specie *Haemoproteus telfordi* and *Haemoproteus tendeiroi* were seen in blood films from 2 adult and 2 juvenile great bustards. However it was not possible to make a blood smear from every bird.
Ectoparasites were found in 33.3% of adults and 14.3% of juveniles. *Qtilipeurus turmalis* (Mallophaga, Insecta) and a tick from the genus *Hyalomma* were identified.

Discussion and conclusions

Mortality causes of free-living juvenile great bustards from Spain has been cited previously (2), although it was not the objective of the study. Predation was the main mortality cause in juveniles and collision with power lines was found in two occasions. In this study only one young was found to be predated by a raptor, while other was hit by an agricultural vehicle. The later cause has been reported as common in bustards chicks as they look after cover in crop fields (18).
In a previous work in Madrid province (18) the authors found that collision with power lines was responsible of the death of 30 wild great bustards in the period 1999-2000. Trauma trough collisions with electric power lines or fences accounted for 63.1% mortality in wild great bustards in the present study. Bone fractures and dislocations, skin and soft tissue injuries, liver rupture, aspiration pneumonia and lung haemorrhage were the most frequent causes of morbidity in such cases. When such cases are admitted to rehabilitation a reasonable valuation and decisions must be done before treatment is accomplished.
A frequent postmortem finding was a high parasitic burden caused by cestodes (*Otiditaenia conoides* and *Idiogenes otidis*). These species of cestodes have been previously reported in great bustards (wild and captive) in Spain (10,16). They also report the death of one bustard following obstruction of the small intestine by cestodes. This condition has been also described in captive houbara bustards in UAE (4,12). In our study the prevalence of cestode infections in bustards was higher than the previously reported by Reina et al. (11.4%) in Spain and Jones et al. (25.6%) in the UAE. This could be due to many different causes as our small sample size and the different origin of birds. Great bustards admitted to rehabilitation centres are usually weakened, dehydrated, traumatised, and in poor body condition (authors, unpubl. data), conditions that may increase the susceptibility to the pathologic effects of cestode infections (12). The clinician must consider these observations when dealing with this specie. Nematodes were seen in low numbers in the caecum of 2 bustards.
Gizzard impactions and foreign-body obstruction have been reported as an important cause of death of captive juvenile rufous-crested bustards and houbara bustard chicks (4,5). One juvenile bird of our study died after a gizzard impaction of unknown aetiology.

Gram-negative bacterial diseases were the most important cause of death over the first 180 days of captive bustard chicks in UAE (5). In our study one week old chick died of septicemia where *E. coli* was culture from different organs. The isolation of *E. coli* from bustards at necropsy was found on many occasions in one study involving captive houbara, rufous-crested and kori bustards (9).

After collision with fences or power lines most of the birds could live for sometime and also walk away for many metres before die (pers. obs.). Aspiration pneumonia was found in 50% of the adult birds as result of aspiration of food (normally seeds) from the ventriculus after the collision.

Aspergillosis has been described as a common cause of euthanasia and postmortem finding in captive and imported adult houbara bustards and also caused mortality in juvenile kori and houbara bustards (4). One adult female died four hours after presentation prostrated and with obvious signs of dyspnea. At necropsy fungal granulomas were seen in trachea, syringe, lungs, pericardium, air sacs, kidney and pelvic nerve roots. A cream-coloured ovoid plaque (7 cm minimum diameter, 11 cm maximum diameter) was recovered surrounding the abdominal viscera. A pure culture of *Aspergillus fumigatus* was obtained from the granulomas. These bird also presented signs of external parasitism, but only a tick from the genus *Hyalomma* was recovered. A high cestode burden was another incidental finding in this bird. To the authors knowledge this is the first report of aspergillosis in a wild adult great bustard.

Klebsiella spp. pneumonia and pneumonia of unknown etiology have described in captive bustards fron the UAE (4). Bacterial pneumonia has been also a morbidity cause (28.6%) in free-living juvenile great bustards. In one occasion *Pasteurella* spp. was cultivated from the lungs of a juvenile bird. Other young great bustard had a focal bacterial pneumonia and liver lipidosis based on histopathology. Unfortunately no microbiology results are available for this case. Some authors have stated that aggressive care of bustards during the first 30 days after hatching is clearly important (5).

Hemoparasites from the genus *Haemoproteus* were detected in 4 of the bustards, but it was not possible to obtain a blood film for each bird. Studies of wild great bustards hemoparasites species and prevalence are been carried out by the authors since 1998 in the Spanish population and will be reported in the future.

Management of wild great bustards clinical cases posses a challenge to the veterinarian. The results presented in this paper could help understanding the morbidity causes when attending such patients.

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References

1. Alonso JC and Alonso JA. The Great Bustard in Spain: Present Status, Recent Trends and Evaluation of Earlier Censuses. Biological Conservation 1996; 77: 79-86.
2. Alonso JC, Martín E, Alonso JA and Morales MB. Proximate and ultimate causes of natal dispersal in the great bustard *Otis tarda*. Behavioral Ecology 1997; 9; 3: 243-252.
3. Bailey TA, Naldo J, Samour JH, and Howlett JC. Bustard Therapeutics. ERWDA External Report No5. 1997.
4. Bailey TA, Naldo J, Samour JH, and Howlett JC. Post-mortem Findings in Bustards in the United Emirates. Avian Diseases 1996; 40: 296-305.
5. Bailey TA, Naldo J, Samour JH, Sleigh IM, and Howlett JC. Bustard pediatric diseases: a review of clinical and pathologic findings. Journal of Avian Medicine and Surgery 1997; 11(3): 166-174.
6. Bailey TA, Nicholls PK, Wernery U, Samour JH, Cooper JE, Path C, and O´Leary MT. Avian paramixovirus type 1 infection in Houbara bustards (*Chlamydotis undulata macqueenii*): clinical and pathological findings. Journal of Zoo and Wildlife Medicine 1997; 28 (3): 325-330.
7. Bailey TA, Silvanose CD, Naldo J, and Howlett JH. Pseudomonas aeroginosa infections in Kori bustards (Ardeotis kori). Avian Pathology 2000; 29: 41-44.

8. Cooper J.E. 1989. Disease and Threatened Birds. ICBP Thecnical Publications.
9. Dáloia MA, Bailey TA, Samour JH, Naldo J, and Howlett JC. Bacterial flora of captive houbara (*Chlamydotis undulata*), Kori (*Ardeotis kori*) and rufous-crested (*Eupodotis ruficrista*) bustards. Avian Pathology 1996; 25: 459-468.
10. Illescas-Gómez MP, and Gómez MP. First citation in Spain of Schistometra conoides (Bloch 1782) Skrjabin 1914, parasitizing *Otis tarda* L. Proceedings of the third European Multicolloquium of Parasitology, 1980, Cambridge, U.K.
11. Illescas-Gómez MP, and Gomez MP. Idiogenes otidis, intestinal parasite of Otis tarda. Revista Ibérica de Parasitología 1981; 41 (4): 475-484.
12. Jones A, Bailey TA, Nicholls PK, Samour JH, and Naldo J. Cestode and acanthocephalan infections in captive bustards: New host and location records, with data on pathology, control, and preventive medicine. Journal of Zoo and Wildlife Medicine 1996; 27 (2): 201-208.
13. Körmendy B, Illes J, Glavits R, and Sztojkov V. An outbreak of *Yersinia pseudotuberculosis* infection in a bustard (*Otis tarda*) flock. Acta Veterinaria Hungarica 1988; 36 (3-4): 173-176.
14. Körmendy P, Sztojkov V, Ivanics E, Illes J, and Csatori G. Pseudotuberculosis in a Great bustard population and comparison with the changes after an experimental tuberculosis infection. Verh. ber. Erkr. Zootiere 1982; 24: 299-302.
15. Kozakiewicz, B. Infection with endoparasites of bustards (*Otis tarda*) under breeding conditions. Wiadomosci-Parazytologiczne 1984; 30 (4): 489-492.
16. Reina D, Habela M, Serrano F, Nieto CG, Brena M, Pérez E, Navarrete I, Hernandez-Rodriguez S. Contribución al conocimiento de la parasitofauna de los animales silvestres y de vida libre en la provincia de Cáceres (España). In Memoriam al Prof. Dr. Francisco de Paula Martínez Gómez 1992. Universidad de Córdoba, 407-428.
17. Sleigh I and Samour JH. Bird care manual: Management techniques for a collection of bustards. NARC, 1996.
18. Traverso JM. Agricultura y tendidos afectan a las avutardas del este de Madrid. Quercus 2001; diciembre: 62-63.

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MYCOBACTERIUM TUBERCULOSIS INFECTION IN ASIAN ELEPHANTS (ELEPHAS MAXIMUS) IN SWEDEN

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Extended abstract

Key words: tuberculosis, mycobacteria, M. tuberculosis, elephant, zoonosis,

Tuberculosis caused by *Mycobacterium tuberculosis* is an important zoonotic disease of captive Asian elephants. Elephant tuberculosis has acquired renewed attention following the outbreaks in USA in the past years. It is estimated that 3% of the captive elephant population in North America is infected (1). Although tuberculosis was recognised in captive elephants in Europe as early as in the 18th century, only limited information, mostly isolated reports, is available about the tuberculous status in captive elephants in Europe.

Recently two infected Asian elephants were detected in a Swedish zoo during a routine control based on mycobacterial culture of trunk lavage. The elephants were a 30-years-old female born in Burma, and a 26-years-old female. Both had arrived to the zoo in the late 1980-s, after having spent 12 and 10 years respectively at European circuses. The two elephants had shown occasional weak bovine reaction to the comparative tuberculin skin test, the 30-years old in 1993 and the 26-years-old in 1997. Thereafter their skin test was negative or inconclusive four times and twice respectively. Both animals seroconverted between 1995 and 1997 and developed a weak positive antibody titre at the ELISA (ELIB and MB70- antigen, at ID-Lelystad, The Netherlands). Culture of trunk lavage has been made in all the elephants of the zoo since 1998.

In late summer 2001 the 30-years-old female became tired, developed polydipsia but retained normal appetite and body weight. A significant increase in beta- and gamma globulins, and a drop in albumin/globulin-quota was noted, starting in early 2001. The trunk lavage cultured positive for *M. tuberculosis* and she was euthanised. The 26-year-old elephant showed positive culture of all three trunk washes in December 2001, while the cultures from lavage obtained in October and November had been negative. This animal is to the date (end of January 2002) in excellent physical condition but will be euthanised in February 2002.

The post mortem examination of the 30-years-old elephant revealed lesions consistent with pulmonary tuberculosis. Solid granulomatous pneumonia involved approximately one fourth of the left lung. Two large (20 x10 x10 cm diamater) granulomas were present in the right lung. The rest of both lungs' parenchyma had widespread numerous small foci. Lesions were also found in bronchomediastinal lymph nodes and in an axillary lymph node. Multiple granulomas of up to 10 mm, were observed along the mucosa of the trachea and the epiglottis.

Histopathology showed diffuse granulomatous consolidation, formed by whirls of epithelioid cells surrounded by macrophages, eosinophils and lymphocytes. There were multiple small foci of central caseous necrosis and several microabscesses within the consolidation. Large organised epithelioid cell granulomas were present in the lungs, lymph nodes and spleen. These lesions had irregular encapsulation, cellular outer layers and large necrotic centres. Calcification was mild. Some of the lesions were extending into the lumen of the smaller airways in the lungs. Large necrotic granulomas with abundant acid fast bacilli were present in the mucosa of the trachea, opening through the epithelium into the lumen. A few acid fast bacilli were found in the lung lesions. Multinucleated giant cells were not observed. *Mycobacterium tuberculosis* was isolated from the lesions on Löwenstein-Jensen medium.

Tuberculosis in elephants is most frequently of pulmonary forms (2). The lesions observed in this case were indicative of shedding via aerosol and respiratory secretions. It is generally recognised that the diagnosis of pre-clinical tuberculosis in elephants is difficult. The present diagnostic tests can detect exposure to mycobacteria and immunological response, but do not provide information on the progression of the infection. The definitive diagnosis can be made by culture of clinical samples first when shedding is in place, which appears to occur at later stages, when well established and productive lesions have developed in natural passages, such as respiratory tract. Tuberculosis acquired by humans from elephants is considered to result most often from close and repeated contact, such as that with elephant handlers. There is also a potential risk for the transmission of *M. tuberculosis* from elephants to other exotic species. It is important that monitoring and control programs for the detection of tuberculous elephants in captivity is implemented in order to prevent infection in humans and animals.

References

1. Mikota SK., Larsen RS and Montali R. (2000). Tuberculosis in elephants in North America. Zoobiology 19 (5), 393-403.

2. Montali R.., Mikota SK and Cheng LI. (2001) *Mycobacterium tuberculosis* in zoo and wildlife species. Rev. sci. tech. Off. Int. Epiz, 20 (1), 291-303

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MYCOBACTERIUM AVIUM SUBSP. AVIUM INFECTION IN RED DEER (CERVUS ELAPHUS HIPPELAPHUS) FROM AUSTRIA

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Key words: (??)

Since January ten cases of *Mycobacterium avium subsp. avium* infections in free living red deer originating from different geographical regions in Austria were observed. The affected animals, nine adults (4 hinds, 5 stags) and one female fawn, showed diarrhoea and severe weight loss. They were shot by local hunters and the carcasses were submitted for post mortem to veterinary laboratories.
At necropsy, lymphadenitis associated with enlarged intestinal lymph nodes was present in all animals. While in some individuals lesions were restricted only to intestinal lymph nodes, in others lymph nodes and tissues of different organs such as those from the respiratory tract were affected. In Ziehl-Neelsen stained smears acid-fast bacilli were demonstrated in large numbers. One male animal showed singular tubercle (3-4 mm in diameter) localised in the subcutaneous muscle tissue.
Strains were identified as *M. avium subsp. avium* by cultural characteristics such as mycobactin dependency, growth on egg medium, or growth stimulation by pyruvate and pH 5.5. Isolates were additionally characterized by variety of PCR-based molecular techniques. Typing of strains was performed by PCR detection of insertion sequence IS901 and the virulence-associated *mig* (macrophage-induced gene), inverted repeat (IR) typing (IS1245/IS1311) and random amplified polymorphic DNA (RAPD) analysis.
Reasons for increasing numbers of *M. avium* infections in red deer and possible predisposing conditions are not known, so far. In mammals *M. avium* is generally considered to be a opportunistic pathogen and infection is mostly seen in immunocompromised individuals. The epidemiology of *M. avium* infections is poorly understood but infections are mostly regarded as environmentally acquired. A possible contamination of feeding places by droppings of birds has to be considered.

Zusammenfassung (??)

Résumé (??)

European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting,
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RABBIT VIRAL DISEASES: RECOMBINANT VACCINES OR NATURAL RECOVERY?

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Abstract

The additive effect of myxomatosis and rabbit haemorrhagic disease (RHD) reduced European wild rabbit (*Oryctolagus cuniculus*) numbers in most of its historical range, specially in the ecologically less-favourable habitats. This had adverse effects both on rural economy and on conservation throughout Spain, where the rabbit is important for the survival of a high number of endangered carnivores and birds of prey, but also a relevant game species. Thus, the decline in rabbit numbers had direct consequences on the reproductive effort of predators, and may even have caused an increase of illegal predator persecution.

In an effort to improve rabbit populations, several management tools have been applied in the last decade. These include the recent development of a recombinant Myxoma-virus (MV) that expresses the RHDV major capsid protein VP60. Concern about the field use of such horizontal transmisible recombinant virus for wild rabbit management, sometimes with opposing goals, has already been expressed (Angulo 2001). Nevertheless, little is known on the current status of Spanish wild rabbit populations. Here we use a large-scale monitoring scheme to state that rabbit abundances are already increasing, and to discuss wether tools such as recombinant vaccines are really needed.

In order to monitorize the abundance of game species, the Government of Aragon (northeastern Spain) set up in 1992 a large-scale survey based on spotlight counts at 17 sites. Rabbit relative abundances increased from 20 rabbits per 100 km seen in 1992, to 66 in 2001. If we group those localities with an average of 20 or more rabbits detected per 100 km in the first three years of monitoring ("rabbit-rich" localities), and compare them with the remaining ones ("rabbit poor"), it comes out that interannual differences are important, that the differences in abundance between "rich" and "poor" sites are also significant, and that an interaction between both factors exists.

These data suggest that wild rabbit populations are recovering naturally, and that their recovery is more evident in sites with higher densities. Most probably, the "rabbit-rich" areas are those where the lagomorph is most important as a hunting species and as a prey. Now, if the rabbit is recovering by itself in those "good" areas, some costy and risky management actions, such as the use of transgenic vaccines, may not be justified. Moreover, the success of a horizontal transmisible recombinant MV depends on a certain

host density. If the new vaccine is not effective in low-density rabbit populations it will be of little use, and the risks may outweigh its eventual benefits to economy and conservation.

Zusammenfassung

Myxomatose und Haemorrhagische Kaninchenkrankheit (Rabbit haemorrhagic disease RHD) haben die Europäischen Wildkaninchenpopulationen (*Oryctolagus cuniculus*) reduziert. Dies hat, besonders in Spanien, starke sozioökonomische und ökologische Folgen.

Eine der Massnamen, die der Kaninchenkrise entgegenkommen soll, ist ein rekombinanter Impfstoff. Es handelt sich um ein Myxoma-virus (MV), welches das RHDV-capsid Protein VP60 ausspricht?#expresar. Obwohl zur Zeit weit über die benutzung rekombinierter Viren diskutiert wird, ist zudem noch wenig über die Entwicklung der Kaninchenbestände nach der RHD bekannt. Wir zeigen, das die Kaninchennummern sich in Spanien zum Teil bereits natürlich erholen, und fragen ob rekombinante Impstoffe wirklich notwendig sind.

Nächtliche Scheinwerferzählungen wurden an 17 Orten der Region Aragón (Nordostspanien) seit 1992 durchgeführt. Die relative Häufigkeit stieg von 1992 bis 2001 von 20 bis 100 Kaninchen pro 100 Km. Orte an denen man zwischen 1992 und 1994 bereits über 20 Kaninchen per 100 Km zählte, zeigen eine starke Erholung der Kaninchenbestände, während Orte an denen weniger als 20 Kaninchen pro 100 Km angetroffen werden konnten, keinerlei Bestandserholung zeigen.

Wenn aber die Wirksamkeit des neuen rekombinanten Impstoffs von der Kaninchendichte abhängt, dann nützt er nur an den Orten wo bereits eine natürliche Erholung der Wildkaninchen beobachtet wird. Orte dagegen, in denen die Kaninchen sehr niedrige Bestände haben, können von der neuen Vaccine wenig erwarten. Eventuell könnten dann die Risikos des neuen Impstoffs seine Vorteile auswiegen.

Résumé (??)

Key words: European wild rabbit, Monitoring, Rabbit haemorrhagic disease, Recombinant vaccine

References

Angulo, E. (2001). When DNA research menaces diversity. Nature 410:739.

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PSEUDORABIES IN WILD BOAR

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Abstract

During the period 1997-1999 three Italian wild boar populations have been investigated in order to detect antibodies against Aujeszky Disease using an ELISA test. The number of examined samples (N), their seroprevalencies (P) and standard errors (SE) were:
1) Maremma, Tuscany (Grosseto) (N=265; P= 47% SE=3)
2) Presidential Estate of Castel Porziano (Rome) (N=136; P=0%)
3) Northern Apennines (Bologna) (N=89; P=6%; SE=2.4)
In the study area 2) according to sampling intensity and population number (about 800 animals in an enclosed area of 50 sqkm) the maximum undetected prevalence might be 2%, an unrealistic prevalence for maintaining the infection in a such high-density populations.
No differences were observed according to gender whereas prevalence significantly increases with age showing (for the maremma population only) and endemic infection. In this population, using age stratified seroprevalence, the force of infection results in 0,00153/wild boar/day. The importance of latency in maintaining the infection in the wild is discussed. The infection in the wild boar appears endemic where population are large and long lasting and when simpatry with free roaming domestic pigs have been reported in past.

Zusammenfassung (??)

Résumé (??)

Key words: hare, poxvirus, hare fibromatosis, re-emerging disease

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MORTALITY, AGE STRUCTURE AND FERTILITY OF A NON RANDOM SAMPLE OF 206 DEAD WOLVES RETRIEVED IN ITALY DURING 1986-2001

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During the period 1986-2001, 206 wolf carcasses retrieved in Italy have been examined.
For each dead wolf data and locality of retrieval were available. Causes of mortality were assessed by standard necropsies and specific examinations. Age was obtained by tooth eruption, cementum-annuli count and cranial sutures. Female fertility was assessed by pregnancy or uterine scares count. Yearly and monthly retrieval trends were calculated. Age structure of the sample has been defined, along with the number of reproductive females and their mean number of uterine scares.
No apparent trend have been observed by years, whereas the autumn/winter months are more likely in having dead wolves. Male-female sex ratio was 1.04; when age was clumped in three main classes, 38.6% of the animals were younger than one year, 18.4% ranged from one to two years and 43% were older than three years. Five of the females older than three years were pregnant (N=1) or showed uterine scares (N=4), representing 5.1 % of the whole females collected in the sample. The mean number of uterine scares or foetus was 4.4 (SD 1.67). The main mortality causes was poaching (illegal shooting and poisoning). The data are discussed and criticised.

Key words: Wolf, Mortality, Age structure, Fertility

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THE ANNUAL SEROLOGICAL SURVEY IN WILD BOAR POPULATIONS IN FRANCE 2000-2001 REPORT

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Abstract (? ?)

Zusammenfassung(? ?)

Resumé (? ?)

Key words : (? ?)

The French Ministry of Agriculture (MAP) and the National Hunting and Wildlife Agency (ONCFS) have been concerned increasingly with the risk that wildlife could constitute to the health of reared animals and man in France.

For three reasons, the wild boar has become a subject of special attention:the population figures have been highly increasing for the past 25 years, particularly for the last 10 years (x 350 %).

Wild boars are or could be a reservoir of several infectious or parasitic diseases, which are very important from an economic or public-health point of view because they are transmissible to the pig (which belongs to the same *Sus scrofa* species), to other domestic species and man.

The development of open-air pig breeding increases the chances of contact with wild boars, and consequently the risks of transmission of pathogenic agents.

A national programme for the serological surveillance of the wild boar population was launched in 1991 by the MAP. It was based on the blood sampling of hunter-killed boars, supervised in each French *département* (sub-regional division, 95 of them in France) by the official Veterinary Services in collaboration with the local hunters associations. The analyses were carried out by the respective national reference laboratories in the French Agency for Food Safety (AFSSA) and the National Veterinary School in Alfort. .

Until 1999, sera were only analysed for Classical Swine Fever and Aujeszky Disease antibody detection. An average of 1000 sera per year have been collected in 60 *départements* (i.e. 10 to 20 analysed sera per *département*). Since the 2000-2001 hunting season, the programme's protocol has been revised by the ONCFS and the MAP so that it be better adapted to the epidemiological situation, but also to increase the reliability and precision of the results. On one hand, the surveillance part was extended to include two other diseases: brucellosis and trichinellosis. On the other hand, the sampling procedure was thoroughly changed (objective: 100 sera per *département* in the 22 *départements* selected according to epidemiological criteria related to the 4 diseases, as

observed in previous surveys in wild or domestic swine populations). In 2000-2001, the objective had exceeded our expectations since 2,548 sera were collected.

Results

For classical swine fever, no spread of the virus has been revealed among the wild boar population in France, except for the Northern Vosges former outbreak (in the Moselle and Bas-Rhin *départements*) which, in 2001, is in the process of natural extinction;
For Aujeszky's disease, the virus is carried by wild boars in several *départements,* with variable infection rates;
For brucellosis, the *Brucella suis* 2 infection appears extremely widespread, with very high prevalence rates (mean seroprevalence rate = 29 %);
for trichinellosis, several seroprevalences which may amount to 16% have been detected in certain *départements*. This, undoubtedly, reveals an understimation of the presence of *Trichinella*, which, according to the official trichinoscopic verifications, are only exceptionally shown in wild boar
In order to follow the evolution of the incidence and the geographical distribution of the four diseases in France, it is planned to repeat the sero-surveys in the course of next hunting seasons.

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MYCOPLASMA AND AVIAN POLYOMA VIRUS INFECTION IN CAPTIVE SPANISH IMPERIAL EAGLES (*AQUILA ADALBERTI*)

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Summary
Both avian polyomavvirus (APV) and *mycoplasma spp*. can cause respiratory disease in birds. In birds of prey the detection of either of the two pathogens in relation to disease has been scarce until now. A Spanish Imperial Eagle (*Aquila adalberti*) that died despite treatment from severe anemia was found positive for *mycoplasma spp*. in all organs. Although cultures were negativ for cytopathogenic viruses, avian polyomavirus could be demonstrated by PCR (Polymerase Chain Reaction) in spleen and kidney. *Mycoplasma spp*. of yet unidentified species was cultured from 12 (67%) out of 18 captive Spanish Imperial Eagles examined subsequently, and APV could be isolated from two (11%) of the 18 healthy eagles. Mycoplasma or APV or both may have played a role in respiratory diseases such as Aspergillosis observed frequently in captive and free-living Spanish Imperial eagles in which analysis for these pathogens had not been performed. There is a need to better understand the role of these pathogens in captive and free living populations of eagles, especially the Spanish Imperial eagle.

Zusammenfassung
Aviäre Mykoplasmen und aviäre Polyomaviren (APV) können schwere, häufig mit respiratorischen Symptomen verbundene Erkrankungen bei verschiedenen Vögeln hervorrufen. Bei Greifvögeln werden derartige Infektionen bisher nur selten diagnostiziert. Ein in Gefangenschaft gehaltener Spanischer Kaiseradler (*Aquila adalberti*) starb trotz Behandlung an einer schweren Anämie. Postmortal konnte aus allen Organen des Adlers hochgradig *Mycoplasma spp*. isoliert werden. Obwohl in der Zellkultur keine zytopathogenen Viren isoliert werden konnten, wurde mittels PCR (Polymerase-Ketten-Reaktion) in der Niere und der Milz des Adlers aviäres Polyomavirus nachgewiesen. Von 18 weiteren, in menschlicher Obhut befindlichen, klinisch gesunden Kaiseradlern wurden Rachen-, Tracheal- und Bindehautsacktupfer untersucht. Dabei konnten bei 12 (67%) der Adler Mykoplasmen und bei zwei Adlern (11%) APV nachgewiesen werden. Die Spezies der isolierten Mykoplasmen konnte bisher nicht bestimmt werden. Sowohl Mykoplasmen als auch APV könnten eine Rolle bei den, bei freilebenden und in Gefangenschaft befindlichen Kaiseradlern häufig auftretenden Atemwegserkrankungen, meist Aspergillosen, spielen, bei denen bisher keine Untersuchung auf diese Erreger durchgeführt wurde. Weitere Untersuchungen zur Bedeutung von Mykoplasmen und APV für frei lebende und in menschlicher Obhut befindliche Kaiseradler wären von Interesse.

Résumé
La mycoplasmose et l'infection avec le polyomavirus aviaire sont des maladies graves souvent respiratoires dans les oiseaux. Au contraire ce ne sont pas de diagnoses fréquents chez les rapaces. Un aigle imperial espagnol (*Aquila adalberti*) qui présentait une anémie grave s'était mort malgré le traitement d'émergence. Les cultures de mycoplasmes étaient positives des toutes les organes de l'aigle. Utilisant des fibroblastes des embrions de poule il n'était pas possible d'isoler des virus cytopatogènes, mais avec le PCR (Polymease chain reaction) nous avons trouvé du APV dans le rein y la rate de l'aigle. Des echantillons de 18 aigles imperiaux ont éte examinés en culture par mycoplasmes et dans des cultures des fibroblastes des embrions de poule. Dans 12 (67%) des oiseaux nous avons isolé du mycoplasme des espèces sans identifier, et dans les cultures du pharynx de deux aigles (11%) il y' avait APV. Dans les aigles imperiaux sauvages et on captiveté il y a regulièrement des maladies respiratoires, fréquentement des aspergilloses. Les deux patogènes isolés dans cette étude pouvraient avoir un rôle dans ces episodes, mais jusqu' á maintenant il n'y avait pas d'analise de mycoplasme ou de APV. Il faut des études supplémentaires pour mieux entendre l'importance de ces deux patogènes par l'aigle imperial sauvage et on captivité.

Keywords: Birds of prey, Spanish Imperial Eagle, Avian Polyomavirus, Mycoplasma spp.

Extended Abstract

Avian polyomavirus virus (APV) infections recently have been demonstrated to affect birds of prey (2), such as the common buzzard (*Buteo buteo*) and the common kestrel (*Falco tinnunculus*). The virus was described originally as budgerigar fledgling disease virus and is known to cause severe fatal disease in psittacine and non psittacine birds worldwide (3, 6).
Mycoplasmosis is an important disease in poultry that is also known in birds of prey (1). Recent investigations have led to the detection of a number of species-speficic new Mycoplasma species from birds of prey with respiratory disease (5). Demonstration of Mycoplasma infection on cytology or by other direct methods is difficult, as well as the isolation and identification of *Mycoplasma spp*. (4). As a result mycoplasmosis seems to be an underdiagnosed disease especially in free-living birds of prey admitted to rehabilitation centres (5).
This extended abstract describes a case of mycoplasmosis and APV infection in a captive Spanish Imperial eagle and summarizes briefly the results on the prevalence of Mycoplasma and APV in 18 captive Spanish imperial eagles.

An apparently healthy adult female Imperial eagle was found to be severely anaemic during a routine health survey. The bird was in very good body condition and showed very moderate respiratory distress. Despite the application of emergency treatment including homologous blood transfusions the eagle died within 24 hours. Macroscopic lesions observed during necropsy included the presence of numerous small (0.5 to 1 cm) granulomatous lesions in the fascia adjacent to the left shoulder joint, as well as in all air sacs (including the clavicular air sacs) and a very pale and atrophic spleen. Microscopically severe haemosiderosis was observed in the liver, in combination with a moderate diffuse infiltrate of mono- and polymorph-nuclear cells. In the lung few large pale eosinophilic intranuclear inclusions could be observed in the present macrophages

and some epithelial cells. Samples were processed for microbiology, isolation and PCR of *Mycoplasma spp*. as well as for virus isolation and PCR to detect avian polyomavirus DNA.

Mycoplasma spp. was cultured in large amounts from all organs, as well as from swabs from the mentioned shoulder joint. The isolate could not yet be identified, but using antisera against *M. synoviae* and *M. gallisepticum* it proved to be neither. Inoculated cultures of chicken embryo fibroblasts (CEF) were negative for cytopathogenic viruses. Using PCR, APV could be detected in the spleen and kidney of the bird. No pathogenic bacteria or fungi were found in the microbiological cultures.

Subsequently, tracheal, choanal and conjunctival swabs of 18 captive Spanish Imperial eagles were examined for the presence of *Mycoplasma spp*. by culture and PCR. In 12 (67%) of the analysed, clinically healthy birds *Mycoplasma spp*. were detected. In five birds *Mycoplasma spp*. could be grown from choanal as well as from tracheal swabs, while in five eagles only the choanal and in one bird only the tracheal swabs were positive. Only on one occasion *Mycoplasma spp*. could be cultured from a conjunctival swab. Pharyngeal and cloacal swabs of the same 18 eagles were examined for the presence of cytopathogenic viruses in CEF cultures. APV was isolated from pharyngeal swabs of two eagles (11%), while no cytopathogenic virus was found in the other samples. The two APV positive eagles had both presented respiratory symptoms upon admission at the rehabilitation and captive breeding centre a few month ago, but were clinically healthy at the time of sampling.

Both, *Mycoplasma spp*. and APV are known to cause respiratory disease in their hosts (1, 2, 3, 4). However, *Mycoplasma spp*. have also been isolated from wild injured or debilitated raptors that did not show signs of respiratory disease or affection of joints (4). A common kestrel from which APV could be isolated showed anaemia, ascites and fatty degeneration of the liver (2). In previous years, cases of Aspergillosis with severe pulmonary fibrosis have been observed in young, free-living Imperial eagles found debilitated or injured, in which viral cultures were negative but no PCR for APV or cultures for *Mycoplasma spp*. had been performed. Either one or both of the afore mentioned pathogens may have played a role in these cases. Additional analysis are under way in order to better understand the importance, pathogenesis and epidemiology of APV and *Mycoplasma spp*. infections in captive and free-living populations of the Spanish Imperial eagle.

1 Heidenreich M. Greifvögel: Krankheiten, Haltung, Zucht. Blackwell Wissenschafts-Verlag, Berlin 1996; 123.

2 Johne R and Mueller H. Avian Polyomavirus in wild birds: genome analysis of isolates from Falconiformes and Psittaciformes. Arch Virol 1998 143: 1501-1512.

3 Khan MS, Johne R, Beck I, Pawlita M, Kaleta EF and Muller H. Development of a blocking enzyme-linked immunosorbent assay for the detection of avian polyomavirus-specific antibodies. J Virol Methods 2000; 89: 39-48.

4 Lierz M, Schmidt R, Goebel T, Ehrlein J and Runge M. Detection of Mycoplasma spp. in raptorial birds in Germany In: Lumeij, J.T., Remple, J.D.

Redig, P.T., Lierz, M., Cooper, J.E. Raptor Biomedicine III. Zoological Education Network, Inc. Lake Worth (FL) 2000; 25-33.

5 Poveda JB, Giebel J, Flossdorf J, Meier J and Kirchhoff H. Mycoplasma buteonis sp. nov., Mycoplasma falconis sp. nov. and Mycoplasma gypis sp. nov., three species from birds of prey. Intern J Sys Bacteriol 1994; 44: 94-98.

6 Sandmeier P, Gerlach H, Johne R and Muller H. Polyomavirus infections in exotic birds in Switzerland. Schweiz Arch Tierheilkd 1999; 141: 223-229.

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MYCOPLASMA CONJUNCTIVAE IS MAINTAINED IN DOMESTIC SHEEP BUT NOT IN ALPINE CHAMOIS IN THE SWISS ALPS

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Key words: Alpine chamois, sheep, ELISA, PCR, culture, epidemiology, infectious keratoconjunctivitis, *Mycoplasma conjunctivae*, *Rupicapra rupicapra rupicapra*, serodiagnosis, Switzerland.

Extended abstract

Infectious keratoconjunctivitis (IKC) caused by *Mycoplasma conjunctivae* is a highly contagious ocular infection which is common in domestic sheep and goats. In the European Alps, IKC is often observed in Alpine chamois *Rupicapra r. rupicapra*) and in Alpine ibex (*Capra i. ibex*), but the disease has also been described in other wild Caprinae in the Pyrenees and in New Zealand. The infection is characterised by inflammation of the conjunctiva and cornea, and in the most advanced stages, the cornea is opaque or even perforated. In IKC outbreaks in chamois and ibex, spontaneous recovery is the most prevalent outcome of the disease. However, mortality can occasionally reach 30% (2). On alpine meadows in Switzerland, IKC occurs at the same time in the same regions in several host species. To assess which host species maintains the *M. conjunctivae* infection in Switzerland, we performed bacteriological and serological investigations in both, domestic sheep and Alpine chamois.

Among a sample of 69 sheep showing clinical signs of IKC in 3 Swiss cantons, *M. conjunctivae* was identified 53 times (76.8%). An indirect ELISA based on a membrane fraction of M. conjunctivae, prepared by industrial standards (1) was used to detect M. conjunctivae antibodies in 674 sera of adult sheep. We analysed a stratified random sample of 123 sheep herds from 25 out of the 26 Swiss cantons. At least one positive animal was detected in 89.4% of the herds. In positive herds (n3D 110), 57.1% of the individual animals tested positive. To assess the importance of sheep's age in the spread of M. conjunctivae, 209 sera of adult sheep and 93 lamb sera among 8 sheep herds were analysed using the indirect ELISA. Seroprevalence in 2-6 month old lambs was 50.5%, indicating that the IKC agent is spread in sheep flocks during raising. Lambs experimentally infected with *M. conjunctivae* carried the agent for 8 and 23 weeks, respectively, depending on the strain used for challenge. We concluded that *M. conjunctivae* is widespread in domestic sheep in Switzerland. In this country, mycoplasmal IKC was found to be endemic and self-maintained in the domestic sheep population (4). In alpine chamois, the occurrence of IKC was assessed in 1950-1999in Grisons, a canton in eastern Switzerland. First IKC outbreaks were reported in the decade 1950-1959. Since then, the number of affected subpopulations constantly increased

and, by the year 1999, IKC outbreaks were reported in 39 out of 51 (77%) chamois subpopulations. In 1992-1999, a total of 243 chamois which died of the consequences of IKC were recorded. The number of cases differed between years, and a distinct seasonal trend was observed. IKC was more common during summer and autumn, with 48% of the cases recorded in August-October. Juveniles (< 4 years of age) were mostly represented. To verify the presence of *Mycoplasma conjunctivae* in chamois we have analysed conjunctival swabs taken from animals affected with IKC. Among a sample of 28 affected chamois, *M. conjunctivae* was identified 14 times (50%). An indirect ELISA was developed to detect specific *M. conjunctivae* antibodies in the sera of alpine chamois with infectious qeratoconjunctivitis using serospecific antigens of *M. conjunctivae*. In subpopulations with ongoing IKC outbreaks, seroprevalence was low (8%). Seroprevalence was even lower in subpopulations with recent IKC outbreaks (3%). We concluded that the *M. conjunctivae* infection is not maintained in the chamois population of the eastern Swiss Alps (3), and transmission of the agent from sheep living in proximity during summer may be the source of epidemics in chamois.

Prevention of IKC in wild Caprinae should focus on preventing the spill-over of *M. conjunctivae* from livestock. However, studies are required to evaluate the distribution of *M. conjunctivae* infection in domestic sheep in several countries, and molecular markers should be developed to trace spill-over of *M. conjunctivae* from domestic animals to wildlife populations. In addition, immunological studies should be performed to develop tools which could lead to the control of *M. conjunctivae* infection in domestic sheep.

Zusammenfassung(??)

Résumé (??)

References

1. Belloy L, Giacometti M, Abdo El-M, Nicolet J, Krawinkler M, Janovsky
2. M and Frey J. Detection of specific Mycoplasma conjunctivae antibodies in
3. sera of sheep with infectious keratoconjunctivitis. Vet Res 2001; 32: 155-164.
4. Giacometti M, Janovsky M, Belloy L and Frey J. Infectious keratoconjunctivitis of ibex, chamois and other Caprinae. Rev sci tech Off
5. int Epiz, in press.
6. Giacometti M, Janovsky M, Jenny H, Nicolet J, Belloy L, Goldschmidt-Clermont E and Frey J. Mycoplasma conjunctivae infection is not maintained in alpine chamois in eastern Switzerland. J Wildl Dis, in press.
7. Janovsky M, Frey J, Nicolet J, Belloy L, Goldschmidt-Clermont E and
8. Giacometti M. Mycoplasma conjunctivae infection is self-maintained in the
9. Swiss domestic sheep population. Vet Microbiol 2001; 83: 11-22.

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THE LIVER FLUKE *METORCHIS* BILIS – A NEW THREAT FOR THE WHITE-TAILED SEA EAGLE (*HALIAEETUS ALBICILLA*) IN MIDDLE EUROPE?

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Extendet abstract:

The white-tailed sea eagle (*Haliaeetus albicilla*) an endangered species in Germany suffers a lot of threats in the highly civilised landscape. Amongst the main causes of death such as traumata due to interference with human structures, i.e. collisions with trains, wire, electrocution, and poisoning, parasites do also play an important role in the health status of these birds (1). Liver flukes belonging to the genus *Metorchis*, diagnosed at a prevalence of 51% (n=112) in the eagles, were thought to be responsible for frequently detected alterations of the liver bile duct system.
To identify flukes at species level and to understand the biology of this trematode the life cycle was established in the laboratory. As known from the genus *Metorchis* (2) the life cycle includes two intermediate hosts: a water snail as first and a cyprinid fish as second intermediate host. Parallel histological and toxicological examinations were performed to reveal the liver alterations and the cause of death in the eagles.
During routine post mortem examination of a white-tailed sea eagle which recently died, adult specimen of *Metorchis* sp. were collected from the gall bladder and the bile ducts. Following cleaning the still living trematodes were put into tap water and stored in a refrigerator (5-7°C) for two days where the parasites excreted fully developed eggs. These eggs were fed to *Bithynia tentaculata* (Prosobranchia) in which the miracidum hatches to undergo an asexual multiplication. Within 5 weeks p.i. rediae were found in the hepato-pankereas and two weeks later cercaria started to leaf the snail in search for the next intermediate host. The excretion of cercariae could be enhanced by exposing the water basin with the snails under a lamp. The pleurolopho cercariae with a membranous tail reacted positively photo- and chemotactic which helped them to find their next host the fish. Ids (*Leuciscus idus*) were used as second intermediate host. After putting snails into fish basins, cercariae penetrated the skin of the fish to become encysted metacercariae in the fins, muscle or even in the cornea. A reduced visual faculty may result in a higher possibility to fell victim of the definitive host, where the parasite maturates. The shape and measurements of the metacercaria revealed the species *Metorchis bilis*. Regardless the high prevalence and intensities (up to 908) only two sea eagles died due to infections with this liver fluke. Histological examinations showed hypertropia of the epithelium cells of the bile duct walls, partial or total obstruction of the bile ducts resulting in thickening of the bile and liver swelling. Very similar organ alterations consisting of stasis of the bile in the gall bladder and the bile ducts and liver swelling could be attributed to high lead levels in the liver (3), which was also often the cause of death (17%, n=112) in these birds. Other eagles which died due to traumata, e.g. territorial fight (n=10) showed liver alterations in four cases due to an infection with *Metorchis bilis* which make them more likely to be the loser.

Zusammenfassung (??)

Résumé (??)

Key words: white-tailed sea eagle, *Haliaeetus albicilla,* causes of death, trematode*,* liver fluke, *Metorchis bilis,* life cycle

References

1 Krone O, Langgemach TP, Sömmer P and Kenntner N. Causes of mortality in white-tailed sea eagles from Germany. Proceedings of the Conference Sea eagle 2000 (in press), Björko, Sweden.
2 Raschmaschkin DA. On the species position of metacercariae of the genus *Metorchis* (Trematoda, Opistorchidae) from fishes of west Siberia (in russian). Parasitologija 1978; 12: 68-78.
3 Kenntner N, Tataruch F and Krone O. Heavy metals in soft tissue of white-tailed eagles found dead or moribund in Germany and Austria from 1993 –2000. Environmental Toxicology and Chemistry 2001; 20: 1831-1837.

European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting,
joint with the annual meeting of the European Wildlife Disease Association (EWDA)
May 8-12, 2002, Heidelberg, Germany.

PATHOLOGY OF COMMON EIDERS FROM THE DUTCH WADDEN SEA IN WINTER 1999-2000

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In December 1999, there was unexplained high mortality of common eiders (*Somateria mollissima*) in the Dutch Wadden Sea. Gross necropsies, supplemented by histologic, virologic, bacteriologic, parasitologic, and toxicologic analyses, were performed on 13 eiders. All birds were severely emaciated and had a multifocal enteritis caused by infection with the acanthocephalan parasite *Profilicollis botulus*. The abundance of this parasite varied from 20 to 1833 (average: 865). The primary cause of death was considered to be starvation or *P. botulus* infection, although the latter was considered less likely. There was no evidence of other diseases, including those caused by viral or bacterial infections, or intoxication as the primary cause of mortality in these eiders. Based on analysis of weather and food resources, we hypothesize that the eider mortality was caused by a combination of factors, including overharvesting by the shellfish industry, a series of mild winters, and decreased shellfish quality. This pathological investigation was hampered by lack of historic data of body weights and *P. botulus* burdens of eiders in this part of their range. To rectify this, we recommend the establishment of a national wildlife health surveillance system. The activities of such a system could include (1) detection of disease or disease-causing agents in wildlife, including eiders; (2) diagnosis, or precise identification, of the diseases; (3) collection and analysis of the information gained from detection and diagnosis; and (4) use of surveillance information in making decisions and policies.

Zusammenfassung(??)

Résumé (??)

Key words: (??)

European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting,
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CANINE PARVOVIRUS INFECTION IN FREE-RANGING CARNIVORES FROM GERMANY

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Abstract

Binn et al. (1970) first described a so-called minute virus of canids (MVC, CPV-1). This so far unknown virus was isolated from apparently healthy dogs. Years later Eugster and Nairn (1977) described a new virus that was pathogenic for dogs, causing haemorrhagic enteritis and myocarditis. In the last twenty years this original canine parvovirus (CPV-2) was detected in many countries in domestic and free-ranging carnivore species and underwent several mutations. Presently, it occurs in two new types (CPV-2a, CPV-2b).
To assess whether wild carnivores play a potential role in the epidemiology of CPV in domestic dogs in Germany, the seroprevalence against CPV in free-ranging carnivores (n=1.669) was determined. Our objective was to compare the prevalence of antibodies against CPV among free-ranging carnivores in selected urban (Berlin ∅ 3.818 man/km², cities in Nordrhein Westfalen 1.242-2.376 man/km²) and selected silvatic areas (Mecklenburg-Vorpommern 41-49 man/km², Brandenburg ∅ 51 man/km²). We assume that human population density is positively correlated with the density of domestic dogs (Frölich et al. 2000). Moreover, dogs were suspected to contaminate the habitat of free-ranging carnivores, and therefore, dog density was assumed to influence the seroprevalence of CPV antibodies in wild carnivores.
In sera from red foxes (*Vulpes vulpes*) (106 of 1593 [6.7%]), raccoon dogs (*Nyctereutes procyonides)* (2 of 37 [5.5%]) and stone martens (*Martens foina*) (4 of 13 [31%]) antibodies were detected by using haemagglutination inhibiting test. Antibodies against CPV were found in all four areas. Preliminary results indicate that the seroprevalence in foxes was not significantly higher in urban compared to silvatic areas suggesting an independent infection process among free-ranging foxes.
Furthermore, we tested 494 tissue samples (small intestine, spleen, mesenterial lnn.) for the presence of CPV nucleic acid by using polymerase chain reaction (PCR). However, until now no positive sample could be detected.

Zusammenfassung (??)

Résumé (??)

Key words: (??)

References

1. Carmicheal, lE. Et al. (1980): hemagglutination by canine parvovirus: serologic studies and diagnostic application. Am j vet res 41, 784-90
2. Binn, lN. et al. (1970): recovery and characterization of a minute virus of canines. Infect. Immun. 1, 503-8
3. Eugster, AK.and Nairn, C. (1977): Diarrhea in puppies: parvovirus-like particles demonstrated in their faeces. Swest vet. 30, 50-60
4. Steinel, A et al. (2001): Parvovirus infection in wild carnivores j wildl. Dis. 37(3), 594-607
5. Steinel A., Munson, L, Van Vuuren M and Truyen U. (2000): Genetic characterization of felineparvovirus sequences from various carnivores j general virology 81, 345-50
6. Truyen U. (1996): Die Evolution des caninen Parvovirus: der Verlust und Rückgewinn des wirtes Katze. Tierärztl. Prax. 24, 316-18
7. Truyen U et al. (1998): Survey on viral pathogens in wild red fox (*vulpes vulpes*) in germany with emphasis on parvoviruses and analysis of a dna sequence from a red fox parvovirus. Epidemiol. Infect. 121, 433-40

8. Truyen U, Platzer G, Parrish CR, Hänichen T, Hermanns W and Kaaden OR. (1994): Detection of canine parvovirus dna in paraffin- embedded tissues by polymerase chain reaction.
9. J. Vet. Med. B. 41, 148-52

European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting, joint with the annual meeting of the European Wildlife Disease Association (EWDA) May 8-12, 2002, Heidelberg, Germany.

NONINVASIVE MONITORING OF CORTISOL METABOLITES IN CAPTIVE AND FREE-RANGING ROE DEER (*CAPREOLUS CAPREOLUS*).

J. MONTANÉ, J. LÓPEZ-OLVERA, D. PERPIÑÁN, I. MARCO, X. MANTECA, R. PALME and S. LAVÍN.

Abstract

Fecal 11,17-dioxoandrostanes (11,17-DOA) were measured in 13 free-ranging and 14 captive roe deer (*Capreolus capreolus*) in order to evaluate the effect of captivity on this parameter and its usefulness as an indicator of chronic stress in this species. Furthermore, fecal 11,17-DOA were analyzed before and after a nine hours transport in 20 roe deer to evaluate the transport stress response and the effect of acepromazine on it. The 11,17-DOA enzyme immunoassay described by Palme and Möstl (1997) was used for the analyses.

Zysammenfassung (?)

Résumé (?)

Key words

Cortisol, fecal, monitoring, roe deer, *Capreolus capreolus*.

European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting, joint with the annual meeting of the European Wildlife Disease Association (EWDA) May 8-12, 2002, Heidelberg, Germany.

HAEMATOLOGIC AND SERUM BIOCHEMISTRY VALUES OF THE SOUTHERN CHAMOIS (*RUPICAPRA PYRENAICA PYRENAICA*) CAPTURED BY DRIVE-NET.

J. LÓPEZ-OLVERA, J. MONTANÉ, D. PERPIÑÁN, S. LAVÍN AND I. MARCO.

Abstract

Haematological and serum biochemical values were determined for a total of 47 southern chamois (*Rupicapra pyrenaica pyrenaica*) in the Cadí-Moixeró Natural Park (Catalonia, North-East Spain) in 2000 and 2001. The chamois (32 adult males, 8 yearling males and 7 adult females) were captured by drive-net and physically restrained to obtain blood samples.

The haematological parameters studied were: red blood cell count (RBC), packed cell volume (PCV), haemoglobin concentration, mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), white blood cell count (WBC), differential leukocyte count and platelet count.

The biochemical parameters studied included cortisol, glucose, lactate, total bilirubin, urea, creatinine, cholesterol, triglycerides, aspartate aminotransferase, alanine aminotransferase, creatine phosphokinase, lactic dehydrogenase, alkaline phosphatase, chloride, sodium and potassium.

To our knowledge, these are the first haematological and biochemical values presented for this species.

Zysammenfassung (?)

Résumé (?)

Key words

Chamois, *Rupicapra pyrenaica*, haematology, serum biochemistry, reference values.

**European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting,
joint with the annual meeting of the European Wildlife Disease Association (EWDA)
May 8-12, 2002, Heidelberg, Germany.**

TWO CASES OF PINEAL MENINGIOMA IN FREE-RANGING SOUTHERN CHAMOIS (*RUPICAPRA PYRENAICA*).

E. VIDAL, I. MARCO, S. LAVÍN AND M. PUMAROLA.

Abstract

Meningiomas are neoplastic proliferations of the cells forming the meninges and only a few cases have been recorded in human affecting the pineal gland. This paper records two cases of pineal meningiomas affecting two free-ranging Southern Chamois and provides a comprehensive histological and immunohistochemical characterisation that leads us to classify them as Meningothelial Meningiomas of the Pineal Gland.

Zysammenfassung (?)

Résumé (?)

Key words

Meningioma, pineal gland, chamois, *Rupicapra pyrenaica*.

European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting,
joint with the annual meeting of the European Wildlife Disease Association (EWDA)
May 8-12, 2002, Heidelberg, Germany.

TUMORS OF THE CARAPACE OF PHILIPPINEGREEN SEA TURTLE (*Chelonia mydas*)[1]

J. S. MASANGKAY[2] and C. NALO-OCHONA[3]

Abstract

Ten tissue samples of fibropapilloma on the carapace of ten individual turtles were subjected to tissue processing and stained with Hematoxylin and Eosin, Masson's Trichrome, and Alcian blue. Based on the tissue sections examined, all samples showed structures resembling the mammalian skin.

In the epidermis, there was moderate to severe hyperplasia comprising of about 10-25 layers of epidermal cells. Orthokeratotic hyperkeratosis with hypertrophy of epithelial cells was present. There were multifocal areas of vacuolation in the stratum granulosum down to stratum basale. There were focal areas of necrosis in the stratum basale.

The epidermis rests on a stroma resembling the dermis, which contains small blood vessels and well-differentiated fibroblasts haphazardly arranged in interlacing bundles of collagen. In the papillary layer of the dermis, increase in downward growth of epidermal hyperplasia with regular to irregular pattern of dermal papillae were observed forming the rete ridges. The reticular layer of the dermis contains more abundant and thicker well-differentiated fibroblasts than the papillary layer. Few mitotic figures and no signs of malignancy or anaplastic changes were observed.

Zusammenfassung (??)

Résumé (??)

Key words: (??)

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European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting,
combined with the annual meeting of the European Wildlife Disease Association (EWDA)
May 8-12, 2002, Heidelberg, Germany.

MARKED DIFFERENCES IN THE NEMATODE FAUNA BETWEEN WILD AND ARMED RED-LEGGED PARTRIDGES (*Alectoris rufa*)

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Abstract

The introduction of new diseases and parasites has been claimed as one of the main problems associated with the introduction of exotic species (1). It has been proposed that an autochthonous pheasant-species can be displaced by an introduced one through parasite-mediated competition (3). However, there is a lack of research regarding the hazard of introducing autochthonous farm-reared individuals without sanitary guarantees. More than 4 million farm-reared juvenile red-legged partridges (*Alectoris rufa* L.) are released yearly in Spain to supplement stocks for shooting (2). Most of them are released without sanitary control. In this study, we compared the nematode fauna of 135 farmed partridges (belonging to ten facilities placed in distant parts of the country), and 159 wild partridges from seven hunting areas where releases of farm-reared birds have not taken place recently. Nine different nematode species were found, but both groups shared not one. In breeding facilities we found *Eucoloeus contortus* (5 farms), *Ascaridia* sp. (5 farms), *Aonchoteca caudinflata* (2 farms), *Heterakis gallinarum* (4 farms), and *Heterakis tenuicauda* (1 farm). In hunting areas we found *Cheilospirura gruweli* (4 areas), *Seurocyrnea seurati* (1 area), and *Trichostrongylus tenuis* (2 area). Implications of the findings for the conservation and management of the red-legged partridge and its habitats are discussed.

Zusammenfassung (????optional?????)

Über vier Millionen junge gezüchtete Rothühner (*Alectoris rufa* L.) werden jährlich auf Spanischen Jagdrevieren ausgelassen, davon die Meisten ohne einer Kontrolle bezüglich ihres Gesundheitszustandes. In dieser Studie werden die Nematoden von 135 Zuchtrothühnern (aus neun Zuchtanstalten aus verschiedenen Regionen), mit denen von 159 wilden Rothühnern aus sieben Jagdrevieren verglichen. In keinem der Untersuchungsreviere wurden in den letzten Jahren Zuchthühner ausgelassen. In den Zuchtvögeln fanden wir *Eucoloeus contortus* (5 Zuchten), *Ascaridia* sp. (5 Zuchten), *Aonchoteca caudinflata* (2 Zuchten), *Heterakis gallinarum* (4 Zuchten), und *Heterakis tenuicauda* (1 Zucht). In Jagdrevieren fandern wir dagegen *Cheilospirura gruweli* (4 Reviere), *Seurocyrnea seurati* (1 Revier), und *Trichostrongylus tenuis* (2 Reviere). Die Implikationen für die Erhaltung und Hege des Rothuhns in dessen natürlichen Habitaten werden diskutiert.

Résumé (??)

Key words: (??)

References

1. Gebhart H. Ecological and economic consequences of introductions of exotic wildlife (birds and mammals) in Germany. Wildl Biol 1996; 2: 205-11.
2. MAPA. Hunting statistics. Annual report. Ministerio de Agricultura, Pesca y Alimentación, 1993.

3. Tompkins DM, Dickson G and Hudson PJ. Parasite-mediated competition between pheasant and grey partridge: a preliminary investigation. Oecologia 1999; 119: 378-82.

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European Association of Zoo- and Wildlife Veterinarians (EAZWV) 4th scientific meeting,
joint with the annual meeting of the European Wildlife Disease Association (EWDA)
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FIRST RECORD OF *HAEMOPROTEUS* SP. PARASITING RED-LEGGED PARTRIDGES (*ALECTORIS RUFA*)

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Abstract
During a health status survey in a Spanish red-legged partridge (*Alectoris rufa*) breeding facility, blood smears were obtained from 40 juvenile birds (26 males and 14 females). In four of the males (15.4 %) typical elongate and sausage-shaped, partial or totally circling host cell nucleus gametocytes were observed in red blood cells. It was identified as an *Haemoproteus* sp. gametocyte. The intensity of infection in males was 5.0±24.0 gametocytes/1000 erythrocytes. No females were infected. To the best of our knowledge, this is the first description of an *Haemoproteus* sp. infection in this species. Morphometric characteristics did not allow us to identify the parasite species, since very few is known on this topic in European phasianids. Parasited partridges showed a marked splenomegaly. Infected erythrocytes presented smaller nuclei, with a nuclear displacement ratio of 0.80. The only heavily parasited individual (122.8 gametocytes/1000 erythrocytes) showed in addition lower serum urea and erythrocyte and leukocyte counts, and a lighter liver.

Zusammenfassung

Während einer Untersuchung über den Gesundheitszustand einer spanischen Rothuhn (*Alectoris rufa*) Zucht, wurden von 40 jungtieren (26 männlich und 14 weiblich) Blutaufstriche gewonnen. In vier der männlichen Vögel (15.4 %) wurden innerhalb der roten Blutzellen typische längliche, wurstförmige Gametozyten beobachtet, die zum Teil oder ganz den Kern der Wirtszelle umringen. Diese Blutparasiten wurden als *Haemoproteus* sp. Gametozyten identifiziert. Die Infektionsintensität war 5.0±24.0 Gametozyten/1000 Erythrozyten. Kein weibliches Rothuhn war infiziert. Nach unseren Daten, handelt es sich um den ersten Bericht über einen *Haemoproteus* sp. Blutparasiten in dieser Vogelart. Die morphometrischen Eigenheiten erlaubten die Spezies-identifizierung nicht, da noch wenig über Blutparasiten der Europäischen Phasianiden bekannt ist. Die infizierten Erythrozyten hatten kleinere Kerne, mit einem Kernverschiebungsverhälnis von 0.80. Das einzige stark parasitierte Rothuhn (122.8 Gametozyten/1000 Erythrozyten) hatte einen niedrigen Harnstoffwert und geringere Erythrozyten und Leukozyten, sowie eine leichtere Leber.

Résumé

Key words: *Alectoris rufa*, blood parasite, *Haemoproteus*, splenomegaly.red-legged partridge, Spain,

Introduction
Haemoproteus sp. is an obligate heteroxenous blood protozoan parasite of birds which uses blood-sucking dipterans of the families Hippoboscidae and Ceratopogonidae for its

transmission. The infections are characterised by schyzogony (merogony) in visceral endothelial cells, gametocyte development in circulating erythrocytes, and presence of pigment in granules in infected erythrocytes (12). Haemoproteids are not commonly associated with disease, and occur widely in avian populations, including Phasianidae (2). In the Iberian peninsula, 8 *Haemoproteus* species have been reported in 18 avian hosts (4).
The red-legged partridge (*Alectoris rufa*) is a medium-sized Phasianidae, subfamily Phasianinae, native from the Iberian peninsula. There are about 1,7-3,6 millions of reproductive pairs (8). In addition, more than 4 million farm-reared juveniles are released yearly to supplement stocks for shooting (9). Despite the ecological and economical importance of this species, few workers have studied the prevalence and intensity of haemoparasites in these birds, only *Plasmodium relictum* having previously been reported (4).

Material and Methods

In November, 1999, 40 apparently healthy young farm-reared red-legged partridges (26 males and 14 females) were surveyed in order to study the sanitary status of a breeding facility (Lugarnuevo, Jaén, southern Spain). Blood samples for the preparation of blood smears were obtained from the brachial vein. The air-dried blood smears were subsequently fixed in absolute methanol and stained with Giemsa's solution (10). The slides were examined microscopically at a magnification of 1000x under immersion oil by the same person (JM). 10,000 red blood cells (RBC) were counted from 5 distant fields of the smear. *Haemoproteus* linear dimensions were obtained as described in (1). Measurements are expressed as means followed by standard deviations. The number of specimens measured is indicated by *n* and the nuclear displacement ratio by NDR.
Packed cell volume (PCV), was measured in heparinized capillary tubes. The RBC count was carried out in a Neubauer chamber, and the number of white blood cells (WBC) was estimated counting the number of leukocytes per 10,000 erythrocytes in the blood smear. Serum proteins (SP) were measured with a refractometer, and serum urea (SU) by spectrophotometry. Spleen weight (SW) and liver weight (LW) were recorded to the nearest milligram.
A Mann-Whitney U test was used to compare erythrocyte morphometric characteristics and haematological and physiological values depending on the presence or absence of the parasite.

Results

During RBC counting, an deep blue elongate and sausage-shaped body was observed into the cytoplasm of erythrocytes. It had golden brown pigment granules and circled partial or totally the host cell nucleus, which was displaced from the central position in most of the cases (NDR=0.80±0.18, table 1). Despite the RBC dimensions were not altered, the nuclei from infected erythrocytes were smaller than those from uninfected ones (U test, $z=5.08$, $p<0.001$ for nucleus length, and $z=4.93$, $p<0.001$ for nucleus width, table 1). Some of the gametocytes were immature, usually in lateral position and with an ameboid outline.
These bodies were identified as a *Haemoproteus* sp. gametocytes (see picture 1). Their morphometric characteristics are reflected in table 1. The absence of measures of one of the samples is due to the fact that this partridge only hosts immature gametocytes. However, the measurements did not allow us to identify the species (see below). To the best of our knowledge, this is the first description of an *Haemoproteus* species parasiting red-legged partridges.
The prevalence of infection was 0.1 (I.C. 95%= 0.028-0.237) in the whole sample, and 0.154 (I.C. 95%=0.027-0.410) in the case of males. No females were infected. Intensity of the parasitaemia was 5.0±24.0 infected cells per 1000 RBC (range: 0-122.8).
Splenomegaly was the most common finding among parasited partridges. In fact, those parasited showed a heavier spleen than those non-parasited (U test, $z=-2.41$, $p<0.05$, table 2). Only one of the affected partridges suffered from a markedly heavily parsitaemia (122.8 gametocytes per 1000 erythrocytes). This individual showed not only a marked splenomegaly,

but RBC and WBC counts, SU and LW were markedly lower than in uninfected males (see table 2).

Discussion

Despite the importance of red-legged partridge as ecological and economic resource, *Haemoproteus* sp. had not been previously described. This could be de to i)few research on this topic, ii)absence of sanitary control of partridge breeding facilities, iii)low prevalence of this parasite in partridges or iv)absence of clinical signs associated to its presence which may keep the parasite undiagnosed. Another possibility for the absence of previous records could be that gametocytes of most species of *Haemoproteus* disappear from the blood in autumn (13), when red-legged partridges are released and/or hunted, and therefore when most of the researches are carried out.

Many authors agree that this parasite is not commonly associated with clinical signs in its hosts (3). However, despite the parasited birds were not apparently sick, we found splenomegaly, which is one of the commonly cited lesions in haemoproteosis (5).

The current classification of *Haemoproteus* spp. is based primarily on host taxonomy since gametocytes have few distinctive features. The philosophy of familiar specificity among the haemoproteids is nowadays adopted (2). There are currently six haemoproteids accepted for the Phasianidae, two of them inhabiting Phasianinae from North America and Asia (2). However, there is a lack of research on this topic in Paleartic Galliformes (8). The only *Haemoproteus* species accepted in Europe is *H. mansoni*, parasite of the red grouse (*Lagopus lagopus scoticus*) in Scotland (U.K.) (14), but Fallis and Bennett (7) demonstrated that it could not be transmitted to members of other subfamilies of Phasianidae. Therefore, the haemoproteid we describe here could be a new species.

To Bennett et al. (3) species of *Haemoproteus* are apparently relatively benign. However, there are some reports of major mortality among flocks of galliform and anseriform birds due to this species (e.g. 11). In fact, we did find some clinical changes associated with the parasite. Thus, we believe more research should be carried out to study the prevalence and effect of this parasite in red-legged partridge populations. The identification of the *Haemoproteus* species with molecular techniques would also help in this topic.

References

1. Bennett GF and Campbell AG. Avian Haemoproteidae. I. Description of *Haemoproteus fallisi* n. sp. and a review of the haemoproteids of the family Turdidae. Can J Zool 1972; 50: 1269-1275.
2. Bennett GF and Pierce MA The haemoproteids of the avian family Phasianide. Can J Zool 1989; 67: 1557-1565.
3. Bennett GF, Peirce MA and Ashford RW. Avian Haematozoa: mortality and pathogenicity. J Nat Hist 1993; 27: 993-1001.
4. Cordero del Campillo M, Castañón L and Reguera A. Índice-catálogo de zooparásitos ibéricos, 2ª ed. Secretariado de Publicaciones, Universidad de León, León, Spain 1994; 584-585.
5. Cordero del Campillo M and Rojo FA. Parasitología veterinaria. McGraw-Hill-Interamericana, Madrid, Spain. 1999; 815-816.
6. Fallis, AM and Bennett GF. Description of Haemoproteus canachites n. sp. (Sporozoa: Haemoproteidae) and sporogony in Culicoides (Diptera: Ceratopogonidae). Can J Zool 1960; 38: 455-464.
7. Kučera J. Blood parasites of birds in central Europe. 3. *Plasmodium* and *Haemoproteus*. Folia Parasit 1981; 25: 303-312.
8. Lucio A. Perdiz roja (*Alectoris rufa*). In: Atlas de la aves de España (1975-1995), SEO-Birdlife, Lynx Edicions, Spain 1997.
9. MAPA. Estadísticas de caza. Annual report. Ministerio de Agricultura, Pesca y Alimentación, 1993.
10. Melhorn H, Düwell D and Raether W. Atlas de Parasitología Veterinaria. GRASS ediciones, Spain 1992; 308.
11. Morii T. A review of *Leucocytozoon caulleryi* infection in chicks. Journal of protozoological research 1992; 2: 128-133.
12. Springer WT. Other blood and tissue protozoa. In: Diseases of poultry, Calnek, B. W. (Ed.), 10th edition. Iowa State University Press, Ames, Iowa, USA, 1997; 167-178.
13. Valkiunas G. Ecological implications of hematozoa in birds. Bull Scand Soc Parasitol 1996; 6: 103-113.
14. White, EM and Bennett GF. Avian Haemoproteidae. 12. The haemoproteids of the grouse family Tetraonidae. Can J Zool 1979; 57: 1465-1472.

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DISEASES AND MORTALITY IN FREE-RANGING LYNX (*Lynx lynx*), BROWN BEAR (*Ursus arctos*), WOLF *(Canis lupus*) AND WOLVERINE (*Gulo gulo*) IN SWEDEN

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Abstract

One hundred and forty-six lynx, 98 brown bears, 20 wolves and 27 wolverines submitted to the National Veterinary Institute, Uppsala, Sweden in the years 1987 to 2001 were investigated for diseases and causes of mortality. The most common cause of death in lynx was sarcoptic mange, most likely acquired from infected red foxes that had been killed by the lynx. The second most common cause of death was traumatic injuries; animals killed in road accidents or by trains, with 34 cases (23%). Lynx moves over large areas and frequently follow roads or railway tracks, especially in winter when the snow cover is deep. Sarcoptic mange was also common in wolves. The most common cause of death in wolves was traumatic injuries; road accidents with cars or lorries, with seven animals (35%) which died of this reason. The most frequent cause of natural death in brown bears was intraspecies fights (infanticide): with 16 (16%) bears killed by other bears. Territory defending male bears are believed to be the animals causing this mortality. Infectious diseases were not frequently observed in any species and only a few cases were observed. Traumatic injuries, originating from road or railway accidents, were also a common cause of death in wolves and in brown bears. Infanticide, i.e. intraspecies killing, was also observed as a cause of mortality in young wolverines. Malformation of the spinal cord was observed in one lynx and one wolf. It is not known if these malformation are hereditary or not and if they are of importance in the future management of these small and endangered populations. Parasites were only found in a few animals and in all cases the parasitic burden was low. A large number of lynx, brown bears, wolves and wolverines were examined as forensic cases since poaching was suspected. The illegal hunting can be a problem for the future management of these large predator populations, since the illegal killing can be a threat to the future status of the population.

Zusammenfassung (??)

R;esum;e (??)

Key words:

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A RE-EMERGING DISEASE OF HARES (*Lepus europaeus*): HARE FIBROMATOSIS

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Key words: hare, poxvirus, hare fibromatosis, re-emerging disease

Abstract

Since 1961 (Leinati *et al*.) fibromatosis in hares has not been reported in the Northern part of Italy. After 40 years, in the same geographic area an outbreak of fibromatosis in game hares occurred. Two hundred and fifty (25%) hares showed typical skin lesions, with no difference in prevalence related to the age. Skin tumours of 1 to 3 cm in diameter were present on legs and ears. After 4-6 weeks skin lesions disappeared and the animals spontaneously recovered. Hystologically many large cells with large nuclei and numerous periodic acid-Schiff inclusion bodies in their abundant cytoplasm were observed. E.M. confirmed the presence of poxviruses. Cross-infection trials and serological examinations on challenged rabbits, hares and cottontails are in progress to confirm origin and nature of the virus.

Zusammenfassung (??)

Résumé (??)

Key words: hare, poxvirus, hare fibromatosis, re-emerging disease

Introduction

Fibromatosis is a disease, different from mixomatosis, typical of hares. It was for the first time described in Italy (4) and France (3), even if at that time there had already been several reports of its occurrence, some of them long time before mixomatosis had been introduced in Europe (6, 7).
It is caused by a leporipoxvirus antigenically related more closely to the agent of rabbit (Shope) fibroma than to myxoma virus (1, 5).
The clinical disease is characterised by single or multiple protruding solid tumours, occurring mostly on ears and legs. The virus is probably mechanically transmitted by arthropods, but animals could also become infected by direct contact. The presence of skin microlesions or trauma is considered a predisposing factor. Morbidity is usually high but mortality is low and most of adult hares spontaneously recover within 1-3 months. Since 1961 in Northern Italy and 1964 in the rest of Europe fibromatosis in hares has not being reported anymore (2).

Clinical cases

At the end of January 2001, in the same geographic area where the disease was first detected in 1959 (Pavia province, Lombardia region; lat. 44,916, long. 9,0146), we observed an outbreak of fibromatosis in a farm of game hares. Two hundred and fifty (25% of the effective) hares showed typical skin lesions, with no difference in prevalence related to the age. Skin tumours of 1 to 3 cm in diameter were present on ears and legs (**Figures 1-2**). After 4-6 weeks skin lesions changed: tumours reduced in size and sometime spontaneously detached. Bleeding scars or dry crusts were then observed and at last most of the animals spontaneously recovered (**Figure 3**). Four hares died for concurrent infections (pasteurellosis and staphylococcosis) and then submitted to necropsy and laboratory examinations. More recently two affected hares showing typical lesions were hospitalised in the animal facilities of IZSLER, kept in isolated rooms and checked daily. At present they are being used for serological controls and experimental trials in order to reproduce the disease.
At the end of 2001 similar lesions were detected in a single hare (**Figure 4**) imported from Hungary and released in an area of Novara province, close to Pavia province, for restocking of hunting areas.

Material and methods

Samples of typical tumours taken during necropsy were submitted to routine examinations i.e. bacteriological, histological (EE staining and Alcian PAS) and virological (identification by electron microscopy using the negative staining drop method and isolation on chicken embryo and RK13 cell culture).
An attempt of reproducing experimentally the disease is still in course. Rabbits, hares and cottontails (*Sylvilagus floridanus)* have been infected using a 1:10 (v/v) PBS homogenate of a nodule (1.5 cm in diameter). The animals of each species were infected through eye inoculation, intradermal injection or direct contact with affected hares. Blood samples were taken from all animals before the challenge and at 1 and 2 months post infection. Two other time-point bleeding are scheduled at 3 and 5 months p.i.; the whole set of sera will be then examined using serological methods (competition ELISA) set up to detect anti-rabbit mixomatosis antibodies.

Results

Bacteriological examination resulted negative on solid nodules, whilst from ulcerated ones *Staphylococcus* sp. and other environmental bacteria were isolated.
Histologically we observed areas of consistent proliferation of new connective tissue in which large spindle or star-shaped fibroblasts with large nuclei and numerous periodic acid-Schiff (PAS) inclusion bodies in their abundant cytoplasm were evident. The vascular hyperplasia, characterised by new blood vessels showing dilated walls and containing large amounts of erythrocytes, was evidence of a recent fibroblastic proliferation. A particular amount of large PAS-positive fibroblasts was sometime visible around the wall of the wider new blood vessels.
Electron microscopic examination was systematically carried out on the homogenates of the nodular tissues taken from all the infected hares showing typical lesions. It permitted to observe several viral particles morphologically resembling poxvirus (**Figure 5**). Similar particles, but in a lower amount, were found when we examined by EM crust material scraped from the scar of an infected hare 4 weeks after the housing in the isolation room.
The attempts of *in vitro* isolation were positive with regard to embryo egg inoculation, i.e. typical pocks were observed on chorion-allantoic membrane (CAM) after three passages and poxvirus particles were observed examining CAM extracts. On the contrary we did not observe any cytopathic effect (CPE) on RK13 cell culture nor positivity for poxvirus by EM, even after 7 passages.
The results of serological investigations will be available at the end of the experimental trial.

Discussion

This outbreak of hare fibromatosis occurred in a “close” farm of game hares, with a good hygienic standard. The morbidity in the farm was quite high and this could be due to passive transfer of virions through equipment and cages, and favoured by microlesions on the skin of captive hares.
It is difficult to determine the origin of the disease because no new animals had been recently introduced, neither the disease had been diagnosed in free living hares of the same area. Nevertheless the detection of similar lesions in a hare imported from abroad for restocking of hunting areas suggests the hypothesis of a re-appearance of the virus in Italy by this way.
Pavia province was among those at the beginning considered as endemic for hare fibromatosis by Leinati et al. (1959). Thus, the possibility that other species could act as reservoirs cannot be excluded. It has to be noted that in this province as well as in other neighbouring province of Piedmont region the population of *Sylvilagus floridanus* is sharply increasing and it is well known that cottontails are naturally resistant to leporipoxviruses such as myxomatosis virus.
The re-appearance of hare fibromatosis after 40 years should draw our attention to the risk that it could become widespread on the territory, causing serious problems and taking an epidemiological relevance. In fact, even if it usually causes low mortality rates, the hares become weaker and could become affected by secondary infections more easily. In addition, they remain carrier for a long time (>1 month) and so could spread large amount of viral particles, increasing the rate of diffusion of the disease in the field.
In conclusion, specific prophylaxis and control measures must be implemented for all the three categories of hares involved in hunting management and mainly used for restocking hunting areas. These hares, i.e. captive hares from close farms, imported hares and free-living hares from the so called “restocking areas” are normally moved to areas even far away, and thus a strict control of their health status and a permanent monitoring of the “receiving areas” are needed to avoid the dissemination of fibromatosis as well as any other infectious disease.

References

1. Fenner F. Viruses of the myxoma-fibroma subgroup of the poxviruses. II. Comparison of soluble antigens by gel diffusion tests, and a general discussion of the subgroup. Aust J Exp Biol Med Sci 1965; 43: 143-56.
2. Fenner F. Hare Fibroma Virus - Chapter 11 in: Osterhaus ADME (Ed.) Virus Infections of Rodents and Lagomorphs. Elsevier Science B.V.. Amsterdam 1994; 77-9.
3. Lafenêtre H, Cortez A, Rioux JA, Pages A, Vollhardt Y and Quatrefages H. Enzootie de tumeurs cutanées chez le lièvre. Bull Acad Vet France 1960; 33: 379-89.
4. Leinati L, Mandelli G and Carrara O. Lesioni cutanee nodulari nelle lepri della pianura padana. Atti Soc Ital Sci Vet 1959; 13: 429-36.
5. Leinati L, Mandelli G, Carrara O, Cilli V, Castrucci G and Scatozza E. Ricerche anatomo-istopatologiche e virologiche sulla malattia cutanea nodulare delle lepri padane. Boll. Inst. Sieroter. Milan 1961; 40: 295-328.
6. Mello U. Di una affezione neoplasica ad andamento epizootico nelle lepri. Ann Staz. sper per la lotta mal inf bestiame Piemonte-Liguria 1929; 2: 47-59.
7. von Dungern E and Coca AF. Über Hasensarkome, die in Kaninchen wachsen und über des Wesen der Geschwulstimmunität. Z. Immunforsch. Exp Ther 1909; 2: 391-98.

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ENDOPARASITES OF RED DEER (*CERVUS ELAPHUS* L.) OF NORTHRHINE-WESTFALIA (GERMANY)

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Abstract

The endoparasite fauna of 85 red deer (37 calves <1 year, 18 ~1 year, 30 >1 year) - 76 complete gastrointestinal tracts, 77 livers, 83 lungs - which were shot during the hunting seasons 1998/99, 1999/2000 and 2000/01 was examined. The red deer originated from four areas from the south of Northrhine-Westfalia. Two of the areas are located close to the German-Belgian border, the other two areas are south of Cologne.
In all, 2 cestode species (*Moniezia benedeni* and *M. Expansa*) and 24 species of nematodes (21 species of gastrointestinal nematodes, two lungworm species and *Setaria cervi* in the abdominal cavity) were recorded. All deer harboured gastrointestinal nematodes, 80.7% *Dictyocaulus eckerti* (1-162 lungworms) and 12% *Varestrongylus sagittatus*. *Elaphostrongylus* larvae were found in 47% of the faecal samples of the deer.
The prevalence of nematodes in the abomasum, small intestine and large intestine was 100%, 65.8% or 97.4%, respectively. The most prevalent species were *Spiculopteragia böhmi* (100%), *Oesophagostomum venulosum* (86.8%), *Ostertagia leptospicularis* (83.5%), *Oesophagostomum sikae* (81.6%), *Skrjabinagia kolchida* (72.2%), *Cooperia pectinata* and *Capillaria bovis* (43.4% each), *Nematodirus roscidus* (35.5%) and *Rinadia mathevossiani* (31.6%). *Apteragia quadrispiculata*, *Hyostrongylus rubidus* and *Nematodirus battus* were recorded for the first time as parasites of German red deer. The abomasum (adults + mucosal fourth-stage larvae) haboured the highest worm burden, followed by small intestine and large intestine. The geometric mean gastrointestinal nematode counts of these sections were 863, 14 or 13 nematodes, respectively. Calves harboured 735, approximately one year old animals 2041 and the animals older than 1 year 1032 gastrointestinal nematodes (geometric means). Worm counts of abomasum and large intestine were highest in the appr. One year old animals, small intestinal worm counts and *Dictyocaulus* lungworm counts decreased with higher age. Neither liver flukes nor rumen flukes were seen.

Zusammenfassung

Die Endoparasitenfauna von 85 Stücken Rotwild (37 Kälber <1 Jahr, 18 ~1 Jahr, 30 >1 Jahr) - 76 vollständige Verdauungskanäle, 77 Lebern, 83 Lungen - aus den Jagdjahren 1998/99, 1999/2000 und 2000/01 aus 4 Herkunftsgebieten im südlichen Nordrhein-Westfalen wurde untersucht. Zwei der Herkunftsgebiete sind an der deutsch-belgischen Grenze gelegen, die anderen beiden südlich von Köln.
Dabei wurden 2 Zestodenarten (*Moniezia benedeni* und *M. Expansa*) und 24 Nematodenarten nachgewiesen: 21 Arten im Magen-Darm-Kanal, 2 in der Lunge und eine Spezies in der Bauchhöhle (*Setaria cervi*). Alle Tiere waren mit Magen-Darm-Nematoden befallen, 80,7% mit 1 bis 168 großen Lungenwürmern (*Dictyocaulus eckerti*) und 12% mit kleinen Lungenwürmern, *Varestrongylus sagittatus*. In 47% der Enddarmkotproben wurden *Elaphostrongylus*-Larven festgestellt.
Alle Rothirsche waren mit Labmagennematoden infiziert, 65,8% mit im Dünndarm und 97,4% mit im Dickdarm parasitierenden Nematoden. Die am häufigsten gefundenen Nematodenarten im Verdauungskanal waren *Spiculopteragia böhmi* (100%), *Oesophagostomum venulosum* (86,8%), *Ostertagia leptospicularis* (83,5%), *Oesophagostomum sikae* (81,6%), *Skrjabinagia kolchida* (72,2%), *Cooperia pectinata* und *Capillaria bovis* (jeweils 43,4%), *Nematodirus roscidus* (35,5%) und *Rinadia mathevossiani* (31,6%). Erstmals als Parasiten des einheimischen Rotwildes wurden nachgewiesen *Apteragia quadrispiculata*, *Hyostrongylus rubidus* und *Nematodirus battus*. Der Labmagen (adulte Nematoden + histotrope Larven 4) war der am stärksten parasitierte Abschnitt des Verdauungskanals, gefolgt von Dünn- und Dickdarm, die nahezu gleichstarke Wurmbürden aufwiesen (geometrisches Mittel der Befallsintensität: 863, 14 bzw. 13 Nematoden). Kälber beherbergten durchschnittlich (geometrisches Mittel) 735, die etwa einjährigen Tiere 2041 und die >1 Jahr alten Tiere 1032 Magen-Darm-Nematoden. Die Parasitierung von Labmagen und Dickdarm war am stärksten bei den etwa

einjährigen Stücken; der Wurmbefall des Dünndarms sowie der mit großen Lungenwürmern ging mit zunehmendem Alter zurück. Leberegel und Pansenegel waren nicht nachweisbar.

Zusammenfassung (??)

Résumé (??

Key words: Red deer, Germany, Northrhine-Westfalia, endoparasites, helminths

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SURVEY OF BRUCELLOSIS INVOLVING BRUCELLA SUIS BIOTYPE 2 IN WILD BOARS, ARC-EN-BAROIS FOREST, FRANCE

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In France, since 90's free ranging wild boars had been identified as a reservoir of *B. suis* 2 and a threat for outdoors piggeries. A national survey has been implemented in order to study the extend of the disease in the French wild boars populations. Parallel to this national survey we have studied more particularly transmission mechanism of porcine brucellosis in wild boars inhabiting Arc-en-Barois forest. There, on a 11 000 ha forest, 500 to 1000 wild boars are shot every year. On 3000 ha capture recapture are performed by the National Game and Wildlife Office for biological studies, that allowed a dynamic serologic survey of marked individuals.
Serological and bactériological status of shot wild boars (N~295) was studied in relation to age, gender, reproduction success and corpus lutea observations. Overall seroprevalence is around 20% while bacterioprevalence does not raise 3%. Very few lesions and clinical signs were recorded. Youngs are significantly less seropositive than older animals ($p<0.05$), male are more seropositive than female when subadults or adults, but this former tendency is not significant ($p>0.05$). We also notice that teats are seldom developped in seropositive subadult female (12 to 24 months old), what reveal that such individuals seldom reproduce. Nevertheless seropositive animals are recorded in the whole area, and we cannot see any difference in seroprevalence before and after mate. In capture animals antibodies can disappear between 4 and 8 months old, and around farrowing period in adult females. No clinical signs were observed on these animals except some orchitis in males. Impact on reproduction success is still unclear.
Our conclusions are that, contrary to chamois brucellosis, porcine brucellosis is widely spread over this wild boar population. This situation could not be maintained by domestic pigs that does not range on this area. Moreover hare populations are very low and could not explain the extend of brucellosis in wild boar. Thus, contrary to chamois, we have to admit intra-wild boars transmissions. In this enzootic situation sexually mature animals seem more susceptible to infection, even we cannot exclude a transcient disappearing of antibodies until sexual maturity in young animals infected as piglets. Paradoxically mate does not seem to play a determinant role in transmission. Regarding brucellosis impact on population dynamics, we cannot see any impact of infection on individual survival. And even if subadults reproduction success seems lessened by brucellosis, we cannot detect a true impact of the disease on wild boar dynamics.

Zusammenfassung(??)

Résumé

Key words:

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SEVERE FEATHER LOSS AND ABNORMALITIES („PINCHING OFF“) IN A JUVENILE FREE-LIVING WHITE-TAILED EAGLE (*HALIAEETUS ALBICILLA*) FROM NORTHERN GERMANY

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Extended abstract

One of 28 free-living white-tailed eagles (*Haliaeetus albicilla*) that hatched in the federal state Schleswig-Holstein (northern Germany) in 2001 suddenly showed a severe symmetrical loss of flight feathers on both wings at the age of 10 weeks. However, loss of body contour feathers was not observed. No obvious preceded incidence could have explained these clinical signs which are described as „pinching off“ (1). The „Pinching off“ syndrome has been observed in several raptors (2), among them two free-living young white-tailed eagles from Schleswig-Holstein (3). However, a scientific explanation for this phenomenon is still missing. Trauma, pathological agents, hormonal disorders or genetic depression are discussed to be involved in the aetiology. In this study we focussed on pathological agents such as parasites and viruses. In the following the clinical signs are documented and preliminary results of several investigations are presented.

Being unable to fly, the white-tailed eagle described in this study was captured at the age of approximately 3 months and brought to the raptor rehabilitation centre at the Wildlife Park Eekholt. A thorough clinical examination including x-ray was performed. Except for the feather loss and abnormalities in both wing- and tail feathers no clinical signs could be diagnosed. However, numerous biting lice of the genus *Colpocephalum* were found especially on the wings. The bird was successfully treated with 0,01% Heptenofos (Ragadan□) solution. A possible correlation between the biting lice and the feather abnormalities was excluded because the clinical signs were still observed after this treatment.

Nearly every second primary feather was missing on both wings. The remaining primaries as well as the secondaries and also the tail feathers showed significant changes in their structure. In most of these feathers not only the calamus and rachis was deformed but also the typical pennaceous character of the vane was missing. Moreover, the feathers were easily bendable and the rachis often showed longitudinal splitting. Lost primaries were permanently replaced by new abnormal feathers, which are also moulted after a while. The abnormal secondaries have not yet been moulted.

Biopsy feather material and blood samples were taken for parasitological and virological investigations.

A small number of quill mites were detected by histological examination of the pulp. Consequently, the bird was treated with 0.2 mg/kg Ivermectin 5 times in two-week intervals, and after a 6 weeks brake, three times with 0.4 mg/kg Ivermectin in weekly intervals. However, this treatment did not

improve the feathering conditions. Therefore, the mites are presumably not the causal agents of the „pinching off“ syndrome.
Blood chemistry showed no abnormalities except for a rise of lactate dehydrogenase (LDH). However, an endoscopic investigation did not give evidence for any pathological process.
Furthermore, no evidence for the presence of viruses like avian polyomavirus (APV) or circovirus was found by preliminary electronmicroscopical examinations. Neither could APV be detected in feather biopsy material or in blood by polymerase chain reaction (PCR). Blood and feather material is currently tested for circovirus using PCR and sequence analysis.
In conclusion, clinical signs of severe feather loss and abnormalities („pinching off“) of yet unknown aetiology in a free-living white-tailed eagle are presented and preliminary results will be discussed.

Zusammenfassung

Ein freilebender Seeadler (*Haliaeetus albicilla)* aus Schleswig-Holstein zeigte im Alter von 10 Wochen plötzlich einen Verlust mehrerer Handschwingen an beiden Flügeln. Dieses klinische Bild wurde in der Vergangenheit mehrfach bei Greifvögeln (2), u. a. bei zwei Seeadlern aus Schleswig-Holstein (3) beobachtet und ist als “Pinching off” Syndrom beschrieben worden (1).
Die klinische Untersuchung des flugunfähigen Seeadlers ergab, abgesehen von den Federverlusten an den Schwingen und am Stoß, keine auffälligen Befunde. Ein starker Befall mit Mallophagen (Gattung *Colpocephalum)* konnte erfolgreich behandelt werden. Eine Korrelation zwischen den Mallophagen und den Federabnormalitäten wird ausgeschlossen.
Die klinische Untersuchung des Federkleides ergab, dass fast jede zweite Handschwinge fehlte, und die noch vorhandenen Schwungfedern, sowie die Stoßfedern auffällig in ihrer Struktur verändert waren.
Histologisch wurde eine geringe Anzahl von Federspulmilben in einem Blutkiel gefunden. Nach einer Therapie mit Ivermectin konnte jedoch keine Verbesserung des Gefiederzustandes festgestellt werden. Ein ursächlicher Zusammenhang zwischen den Federspulmilben und dem klinischen Erscheinungsbild des Seeadlers wird daher nicht angenommen.
Weder das große Blutbild noch eine endoskopische Untersuchung ergaben Hinweise auf ein pathologisches Geschehen. Darüber hinaus konnten elektronenmikroskopisch keine Anzeichen für ein Vorhandensein von Viren, wie aviären Polyomaviren (APV) oder Circoviren, gefunden werden. Auch eine Untersuchung auf APV mittels PCR verlief negativ. Derzeit wird Biopsiematerial und Blut auf Circoviren (PCR) untersucht.
Zusammenfassend werden die klinische Symptome einer ausgeprägten Gefiederstörung von bisher ungeklärter Ätiologie (“Pinching off” Syndrom) bei einem freilebenden Seeadler präsentiert und vorläufige Untersuchungsergebnisse vorgestellt.

Résumé (??)

Key words: *avian polyomavirus, biting lice, circovirus, endoscopy, electronmicroscopy, feather abnormalities, feather loss, Haliaeetus albicilla, PCR, pinching off syndrome, quill mites, white-tailed eagle*

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Literature:

1 Cooper JE. Veterinary aspects of birds of prey. The Standfast Press, Gloucestershire 1985; 158.
2 Grünhagen H. Federanomalien bei Greifvögeln. In: Deutscher Falknerorden (Ed) Greifvögel und Falknerei. Neumann Neudamm 1989; 73-76.
3 Robitzky U, Artenhilfsprogramm für den Seeadler *Haliaeetus albicilla* in Schleswig-Holstein. In: Meyburg BU and RD Chancellor (Eds.) Eagle studies. World Working Group on Birds of Prey, Berlin, London & Paris 1996; 73-95.

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DENTAL LESIONS AND BITE WOUNDS IN EURASIAN OTTERS (*Lutra lutra*)

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Abstract (??)

Zusammenfassung (??)

Résumé (??)

Key words:

As part of a long-term study to monitor the health status of otters in south west England post mortem examinations were carried out on 242 submitted between December 1988 and January 2002. The great majority (80%) had died in road traffic accidents but bite wounds were the second most common cause of death (1, 2). This report describes observations on bite wounds and dental lesions.

Bite wounds were commonly present, with 23 out of 102 females (22.5 %) and 35 out of 140 males (25 %) showing lesions. Bites to the head usually involved the cheek and/or lip. In some cases the wounds showed evidence of infection, with extensive cellulitis and necrosis involving the subcutaneous tissues and muscles. Bites to the feet were mostly confined to the central pad and/or the digits. In some cases the digits were amputated. Most bites in the perineal area were around the anus but often also involved the vulva or scrotum.

The consistent pattern of the bite wounds suggests stylised fighting and it is believed that the great majority were due to fighting with other otters (1, 2). In a few cases the size and spacing of the wounds were smaller and could have been inflicted by another species, possibly American mink, *Mustela vison*. Two cubs were killed by dogs and in these cases the lesions were distinct, with large puncture wounds, extensive bruising and fractured ribs.

All ages were affected, but it was noticeable that although two female cubs had died as a result of bite wounds, no male cubs of less than 4 kg were bitten. There was evidence of a higher incidence of wounds in otters in particular weight ranges. This was particularly noticeable in males between 5 and 5.9 kg where 60 % were affected. The incidence then dropped off progressively and was lowest in the heaviest otters. A similar, though less striking, pattern was seen in females with bites present in around 29% of animals in the 4 to 6 kg. range. As with males, lesions then became less frequent in the heavier animals.

The degree of wear on the teeth was recorded and any lesions noted. The general health of teeth was good, with very little calculus and no cases of caries. However, fractures and loss of teeth were common, with 46 animals (19%) affected.

Fractures or loss of incisors were recorded in 15 males and 7 females (9% overall). It might be expected that, as the teeth cannot be replaced, the incidence of lesions would increase with increasing age. However, this was not the case and lesions were fairly evenly distributed through the weight range of both sexes. Lesions were twice as common on the left side of the mouth as on the right.

Lesions affecting canine teeth were more common and fractures or loss were recorded in 11.6% of otters (19 males and 9 females). Frequently there was either a slab fracture or the tooth was

sheared off near the base, leaving the pulp cavity exposed. In most cases only one canine was broken but in several otters two or more were fractured and in some cases an entire tooth had been lost. Young animals were not affected and no lesions were seen in females of less than 4 kg or in males of less than 5 kg. As was observed with bite wounds, there was then a peak in the incidence of lesions as the body weights increased, followed by a decline in incidence in heavier animals. In both sexes the highest incidence of canine damage was in the weight range 6 to 6.9 g where around 28% of males and 21% of females were affected.

In some otters the canines on one side of the mouth were more worn than on the other, suggesting that they may be 'handed'. It was also observed that attrition of the enamel on the anterior/lateral aspect near the base of the lower canines was very common. It is assumed that this is caused by some aspect of feeding behaviour.

Lesions involving the cheek teeth were uncommon but often important. Fractures involving the fourth upper premolars, or carnassials, were seen in six otters, five males and one female. As with domestic dogs, this is a significant lesion and in two cases it resulted in a root abscess and osteomyelitis. The infection tracked through the bone dorso-laterally and led to severe cellulitis and necrosis of facial tissues, septicaemia and death. The largest otter examined, a male weighing 11.3 kg., also appeared to have died as a result of a tooth abscess. Unfortunately it was too autolysed for detailed examination. It had fractures to several lower incisors and premolars and a fracture involving the root of the lower right first molar. The latter had become infected, resulting in osteomyelitis and thickening of the ramus. This was the only lesion seen in a molar tooth.

It seems likely that that fractures to carnassial teeth are due to otters biting on hard material but fractures and loss of incisors and canines may often be due to fighting. In several cases fresh bite wounds were seen in otters that also had fresh tooth damage, particularly to canines. Ten out of the 28 otters with canine lesions (36%) had bite wounds, mostly to the head, feet and perineum, and 62% of otters with incisor damage had bite wounds. The fact that canine damage was less common in heavier, and therefore generally older, male otters is perhaps an indication that animals which have suffered serious damage as young adults are less likely to survive to full maturity.

The prevalence of bite wounds appeared to be fairly constant at around 15 - 17 % up until 1999 but in the year 2000 it was 32 %. This was large due to a marked increase in bitten females, four of which died as a result of their wounds. Mortality from bite wounds in 2000 was 12.5% whereas in earlier years it has been in the range 4 - 6.5 %. This figure is almost certainly an under estimate of the true mortality caused by fighting, as otters dying in road accidents are much more likely to be seen and submitted for examination than otters dying in the countryside. Mortality in otters from intraspecific aggression could be as important as road accidents.

References

1. Simpson V R. Health status of otters (*Lutra lutra*) in south-west England based on postmortem findings. Veterinary Record 1997; 141: 191-197

2. Simpson V R and Coxon K E. Intraspecific aggression, cannibalism and suspected infanticide in otters. British Wildlife 2000; 11: 423-426

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PRESENCE OF INFECTIOUS AGENTS AND PARASITES IN WILD POPULATION OF COTTONTAIL (*SYLVILAGUS FLORIDANUS*) AND CONSIDERATION ON ITS ROLE IN THE DIFFUSION OF PATHOGENS INFECTING HARES

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Abstract
In three study areas located in Alessandria province (Piedmont, Italy), 271 *Sylvilagus floridanus* were captured for parasitological and serological survey. The parasitological research has shown the presence of ecto (ticks, fleas) and endoparasites (nematodes, protozoa), and in particular the infestation with exotic species (*Euhoplopsyllus glacialis*, *Obeliscoides cuniculi*, *Trichostrongylus calcaratus*, *Passalurus nonannulatus*). Sera were tested to detect antibodies against the following diseases: EHBS and RHD, myxomatosis, leptospirosis, brucellosis, tularemia, Lyme borreliosis, toxoplasmosis and encephalitozoonosis. Experimental infection of 6 cottontails and 6 domestic rabbits (all sero-negatives for RDHV and EBHSV) with virulent RDHV strain, induced mortality only in domestic rabbits. Experimental infection of 4 cottontails and 2 hares (all sero-negatives for RDHV and EBHSV) with virulent EBHSV strain, induced mortality in a cottontail. The results and their sanitary aspects are discussed.

Zusammenfassung (??)

Résumé (??)

Key words: cottontail rabbit, *Sylvilagus floridanus*, infectious diseases, parasitological diseases.

Introduction

The eastern cottontail (*Sylvilagus floridanus*), a native American lagomorph, was introduced in different European countries: France (1953), Italy (1966), Spain (1980), Switzerland (1982). The largest "Italian" population lives in Piedmont, North Italy, where the eastern cottontail is felt as a sort of pest for agriculture and a biological competitor of the brown hare (*Lepus europaeus*)(17). Since few data are available on the diffusion of infectious agents and parasitic agents of this species in our country and in Europe we carried out a seroepidemiological survey and parasitological analysis with the aim to determine their role as hosts, vehicles or reservoirs of pathogens common to hares.

Area of study
We considered three different areas in the province of Alessandria (lat. 44,916; long. 8,6148), Italy, with the following criteria: i) typical habitat of all territories occupied by *S. floridanus*; ii) high density of *S. floridanus* (>100 animals/Km^2); iii) contemporary presence of brown hares; iv) no hunting allowed.

Materials and methods

Fifteen individuals were shot in each study area at bimonthly intervals, between July 1999 and August 2000. A total number of 271 animals (131 males, 140 females) were collected. Each animal was submitted to both serological and parasitological analyses.

Parasitological analyses:

Detection and identification of: ectoparasites in animals' fur, protozoa in faeces, helminths in the digestive tract, nematodes in the respiratory system and trematodes in the liver.

In each area, for each species of nematodes we determined the prevalence (P), the relative abundance (RA) and the abundance (A).

Serological examination:

The serum was analysed to detect antibodies to the following infections using the reported tests and the relative "cut-off" indicated in brackets:

- Rabbit Haemorrhagic Disease (RHD): Competitive ELISA (≥1/10)
- European Brown Hare Syndrome (EBHS): Competitive ELISA (≥1/10)
- Mixomatosis: Competitive ELISA (≥1/10)
- Leptospirosis (8 serovars of *Leptospira interrogans*): MicroAgglutination Test-MAT (≥1/100)
- Brucellosis (*B.abortus-melitensis*): Complement Fixation Test-CFT (≥20 U.I.)
- Tularemia: MicroAgglutination Test-MAT (≥1/40)
- Lyme Borrelliosis: Indirect Immuno Fluorescence Test-IFAT (≥1/80)
- Toxoplasmosis: Latex Agglutination Test-LAT (≥1/32)
- Encefalitozoonosis (*Encephalitozoon cuniculi*): Carbon Immuno Assay-CIA (≥1/40).

Experimental trials

We checked the reproducibility of RHD and EBHS in seronegative cottontails under experimental conditions, by inoculation of RHDV and EBHSV virulent strains. In the first trial 2 cottontails and 3 rabbits were infected with the RHDV reference strain BS89 by oro-nasal route, 2 cottontails and 2 rabbits were administered by the same route and inoculum but inactivated, the remaining 2 cottontails and 2 rabbits were maintained as non infected controls. The animals were kept in three separated rooms, checked daily for two weeks and serologically controlled, for RHDV IgM and IgG, after 14 and 28 days post infection (p.i.). In the second experiment 4 seronegative cottontails, and 2 seronegative hares were infected by oro-nasal route with a virulent suspension of EBHSV. The animals were kept in separated cages in the same room, checked daily for two weeks and serologically controlled for EBHSV IgM and IgG after 11 and 32 days p.i..

Results and discussion

Parasites

Three nematodes have been collected from the digestive tract: *Obeliscoides cuniculi*, *Trichostrongylus calcaratus* and *Passalurus nonannulatus.* Their prevalence and abundance are shown in Table 1. Moreover we have found two different genera of cestodes whose identification is in progress.

Seven different *Eimeria* were found: *E. honessi, E. paulistana, E. environ, E. magna, E. leporis, E. andrewsi* (15).

As regards ectoparasites, we have found ticks (*Ixodes ricinus*) and three different fleas: *Ctenocephalides canis*, *Euhoplopsyllus glacialis* and *Ctenophtalmus agyrtes.* Their prevalence is reported in Table 2. No lung or liver parasite was found.

Tab. 1 e 2

The nematodes found are first reports in Italy. Rabbits (*Oryctolagus cuniculus*) infected by *O. cuniculi* have been reported in India and China, while in North America the parasite has been found in *S. floridanus*, *Lepus americanus* and *Lepus capensis* (9; 7; 1; 10; 6; 11; 12; 13). *T. calcaratus* has been reported in North America in *S. floridanus*, *L. americanus*, *Sciurus niger* and *Odocoileus virginianus* (16; 2; 9; 4; 3). *P. nonannulatus* has been reported in North America in *S. floridanus* (2).

The flea *E. glacialis* is another first report in Italy. It has been previously reported in North America in *Sylvilagus audubonii, Lepus californicus, L. americanus* and *Urocyon cinereoargenteus* (8; 14; 5).

Our results point out that the importation of an allochthonous species implied the introduction of new parasites in Italy. This might have unpredictable consequences on the sympatric populations of autochthonous lagomorphs. Recently, we have collected *O. cuniculi* and *E. glacialis* in hares come from two of our three study areas (in prep.).

Serology

The positive results are summarised in the table 3. No antibodies were detected against *Brucella* spp. and *E. cuniculi*. Indeed, the results obtained induce to exclude a relevant role of *Sylvilagus* in the dissemination and transmission to hare and other mammals, including humans, of infectious agents such as *F. tularensis, T. gondii, Brucella spp., L. interrogans, E. cuniculi, B.burgdorferi*

Tab. 3 e 4

More interesting are the results concerning the three main viral diseases of lagomorphs (Table 4). The high prevalence for mixomatosis in the absence of known clinical cases (some sera had titres as high as 1/1280) confirms that cottontail that *Sylvilagus* could be infected by this virus. It probably acts as wild reservoir and could play an active role in the epidemiology of mixomatosis of this viral infection in the wild.

Since RHD and EBHS are both considered host-specific diseases, it is more difficult to explain the serological results detected i.e. an overall seroprevalence of 17.8% for EBSV and 33.3% for RHDV antibodies. Indeed, a relevant number of sera had high titres for EBHSV (up to 1/1280), whilst the highest RHDV titre in a single serum was 1/80. These results, compared to those detected in the sera of RHD recovering-convalescent rabbits and EBHS-convalescent hares, indicate that some of the cottontails could have been naturally infected by the EBHSV, developing a strong immunity. Conversely, the titres for RHDV antibodies were always too low to be considered specific or directly induced by one of the two known calicivirus of lagomorphs.

Experimental trials

The inoculation of RHDV caused an acute form of RHD only in the three seronegative rabbits, which all died within 72-hrs p.i. Showing typical signs and lesions. The RHDV positivity was confirmed by sandwich ELISA test. All the remaining animals remained healthy during the 14 days of observation and in their sera, collected at 14 and 28 days p.i., no antibodies (IgM and IgG) were detected using the RHDV ELISAs. These data confirm that cottontails are not susceptible to experimental infection with RHDV, as previously postulated, but also that the inactivated RHDV do not stimulate the immune system of rabbits when administered by oro-nasal as well as it did when injected intramuscularly. During the second trial one cottontail died at 4 days p.i. and at necropsy it had typical gross lesions referable to EBHS. The diagnosis was confirmed by virological ELISA, and by the relevant seroconversion (IgM up to 1/40000 and IgG up to 1/1280) detected in the sera of the hare and the three cottontails that survived. These results demonstrate transmissibility of EBHSV to *S. floridanus* and support the conclusions of the sero-epidemiological survey. In particular, the cottontail could play a role in the spreading of EBHS.

Further epidemiological studies will be pointed to contemporarily detect the incidence of EBHS in sympatric and even syntopic populations of hares and *Sylvilagus* as well as to determine the characteristics (morbility, lethality, lesions etc) of such disease in *Sylvilagus,* since it could be less severe or totally silent.

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References

1. Bloomer-SEM; Willebrand-T; Keith-IM; Keith-LB. Impact of helminth parasitism on a snowshoe hare population in central Wisconsin: a field experiment. Canadian-Journal-of-Zoology 1995; 73 (10): 1891-98.
2. Boggs-JF; McMurry-ST; Leslie-DM Jr.; Engle-DM; Lochmiller-RL. Influence of habitat modification on the intestinal helminth community ecology of cottontail rabbit populations. Journal-of-Wildlife-Diseases 1990; 26 (2): 157-69.
3. Clemons-C; Rickard-LG; Keirans-JE; Botzler-RG. Evaluation of host preferences by helminths and ectoparasites among black-tailed jackrabbits in Northern California. Journal-of-Wildlife-Diseases 2000, 36 (3): 555-8.
4. Coyner-DF; Wooding-JB; Forrester-DJ. A comparison of parasitic helminths and arthropods from two subspecies of fox squirrels (Sciurus niger) in Florida. Journal-of-Wildlife-Diseases 1996; 32 (3): 492-97.
5. Durden-LA, Kollars-TM Jr. The fleas (Siphonaptera) of Tennessee. Journal-of-Vector-Ecology 1997; 22 (1): 13-22.
6. Ives-AR; Murray-DL. Can sublethal parasitism destabilize predator-prey population dynamics? A model of snowshoe hares, predators and parasites. Journal-of-Animal-Ecology 1997; 66 (2): 265-78.

7. Jin-ZhaoQing; Lu-JunJie; Tang-LiQuan; Yang-Min. Investigations of parasites of domestic rabbits used for experiments and their control with drugs in Lanzhou region. Chinese-Journal-of-Veterinary-Science-and-Technology 1994; 24 (7): 13-4.
8. Kucera-JR. Additional records of fleas (Siphonaptera) from Utah. Great-Basin-Naturalist 1995; 55 (1): 92-4.
9. Lepitzki-DAW; Woolf-A; Bunn-BM. Parasites of cottontail rabbits of southern Illinois. Journal-of-Parasitology 1992; 78 (6): 1080-83
10. Malakondaiah-P; Hafeez-M. Prevalence of helminthic infections in certain laboratory animals in and around Tirupati. Cheiron 1996; 25 (1-2): 40-2.
11. Murray-DL; Keith-LB; Cary-JR. The efficacy of anthelmintic treatment on the parasite abundance of free-ranging snowshoe hares. Canadian-Journal-of-Zoology 1996; 74 (9): 1604-11.
12. Murray-DL; Cary-JR; Keith-LB. Interactive effects of sublethal nematodes and nutritional status on snowshoe hare vulnerability to predation. Journal-of-Animal-Ecology 1997; 66 (2): 250-64
13. Murray-DL; Keith-LB; Cary-JR. Do parasitism and nutritional status interact to affect production in snowshoe hares? Ecology 1998; 79 (4): 1209-22.
14. Patrick-MJ, Harrison-RL. Fleas on gray foxes in New Mexico.Journal-of-Medical-Entomology 1995; 32 (2): 201-4.
15. Poglayen G., Meneguz P.G., Tizzani P., Gaglio G., Ferlazzo M.. Coccidia (Eimeridae) of Sylvilagus floridanus in Italy: presence and space-temporal distribution. Sixth National Conference of Parasitology, Sofia, 5-7 october 2001.
16. Pursglove SR, Prestwood AK, Nettles VF, Hayes FA. Intestinal nematodes of white-tailed deer in southeastern United States. J Am Vet Med Assoc 1976;169(9):896-900
17. Silvano F., Acquarone C. e Cucco M.. Distribution of the eastern cottontail *Sylvilagus floridanus* in the province of Alessandria. Hystrix 2000; 11 (2): 75-8.
18. Spagnesi M., Toso S. Iconografia dei mammiferi d'Italia. Istituto Nazionale per la Fauna Selvatica "Alessandro Ghigi" 1999, 200 pp.

Area of study	**"Roleto"**					**"Tollara"**					**"Sezzadio"**				
	P	RA	A	min	max	P	RA	A	min	max	P	RA	A	min	max
O. cuniculi	93	18,0	77,5	1	430	84	16,3	52,7	1	770	5	0,7	23,4	1	90
T. calcaratus	90	52,4	234,2	3	1778	88	83,7	287	3	2550	89	99,3	194	1	1560
P. nonannulatus	62	29,7	191,9	1	1920	0					0				

Table 1 – Gastrointestinal nematodes in *S.floridanus*: prevalence (P), relative abundance (RA) and abundance (A) in the three study area.

Area of study	**"Roleto"**	**"Tollara"**	**"Sezzadio"**
I. ricinus	8,8	6,6	5,5
Euhoplopsyllus glacialis	73,3	8,8	96,7
Ctenocephalides canis	1,1	1,1	1,1
Chtenophtalmus agyrtes	0	2,2	1,1

Table 2 – Prevalence of ectoparasites in the three study areas

Area of study	***F.tularensis***	***B.burgdorferi***	***T.gondiii***	***L. interrogans***
Roleto	0/83	3/83^	2/81°	0/83
Tollara	0/87	2/87^	2/84°	0/87
Sezzadio	1/83*	1/83^	1/82°	1/83"
Total	*1/253*	*6/253*	*5/247*	*1/253*
Prevalence	*0,4%*	*2,4%*	*2,0%*	*0,4%*

Table 3 - Results of the seroepidemiological investigation (bacterial diseases). Legenda: *1:40, ^1:80, °1=1:64, 2=1:128, 2=1:256 "1:6400 serovar Australis Bratislava

Area of study	**Mixomatosis**	**EBHS**	**RHDV**
Roleto	41/82	8/82	27/82
Bormida	28/83	16/83	25/83
Tollara	44/88	21/87	33/87
Totale	*113/253*	*45/252*	*85/252*
Prevalence	*44,6*	*17,8*	*33,7*

Table 4. Results of the seroepidemiological investigation (viral diseases)

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PATHOLOGY OF BOVINE TUBERCULOSIS IN THE EUROPEAN WILD BOAR (*SUS SCROFA*)

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Bovine tuberculosis, infection by *Mycobacterium bovis,* affects a wide range of wildlife, and it is one of the most important re-emerging zoonotic diseases in Europe. Bovine TB has recently been described in the European wild boar, *Sus scrofa* (Mignone et al, 1991). Transmission of infection could occur amongst cattle, deer and wild boar (Aranaz et al, 1996). Humans may contract infection by handling or consuming tuberculous wild boar. Characterising the pathology of tuberculosis in the wild boar is necessary to establish the diagnosis. Moreover, the distribution of the tuberculous lesions can provide indication of the mode of shedding and the route of infection, which are important from the point of view of the epidemiology (de Lisle et al, 2001). This communication describes the gross and histopathological features of bovine TB in European wild boars shot during hunting.

All the animals included in this study presented gross tuberculous lesions, and infection by *Mycobacterium bovis* was confirmed by culture in Lowestein Jensen medium with addition of piruvate and identification by PCR. Most of the tuberculous wild boars had no clinical signs and were in normal nutritional state. Caseocalcareous tubercles from one milimeter to five centimeter in diameter were the most frequently found lesions. They were mainly located in lymph nodes, and only a few (7%) individuals showed visceral lesions. Lesions were consistently found in mandibular lymph nodes (100 % of animals with gross lesions). Thoracic lymph nodes showed lesions in 56% of the cases and abdominal lymph nodes in 49%. One third of the cases (34 %) presented macroscopical lesions exclusively located in mandibular lymph nodes. Caseous lesions were predominant, while calcification was less abundant (see table 1).

Tuberculous lesions were confirmed histopathologically in mandibular, bronchial, mediastinal, and mesenteric lymph nodes, as well as in liver, spleen, lungs and mandibular salivary gland. The tuberculous changes consisted of were well organized granulomas with abundant central necrosis and calcification, encased in a thick fibrotic capsule. A cellular layer composed of macrophages, lymphocytes, epithelioid cells and scarce giant cells was present subjacent to the capsule and surrounding the necrotic centres. The giant cells were of irregular shape and size, giant cells of Langhans type were rarely observed. The smaller granulomas were more cellular while the larger lesions were predominantely necrocalcific.

The large size and chronicity of the lesions in the head, such as in the mandibular lymph nodes, suggest these sites as early involvement, while the small granulomas in various organs, such as liver, spleen and lungs, suggest a secondary haematogenous origin. The sites of primary involvement give indications of route of

infection, which in the wild boar of this study appears to be alimentary. Shedding of mycobacteria is likely to occur in saliva in the wild boar, as gross lesions were often observed in the mandibular salivary gland, and were histologically confirmed as growing and breaking into the lumen of excretory ducts. Since little is known about the epidemiology of bovine tuberculosis in the European wild boar, more studies are needed to elucidate the role of this species in the spread of *M. bovis*.

Table 1. Components of the gross lesion in different locations of affected lymph nodes in European wild boars.

References

1. Aranaz, A., Liebana E., Mateos A., Dominguez L., Vidal D., Domingo Rodriguez-Ferri E. F., Bunschoten A. E., Van Embden J.D. and Cousins D. (1996). Spacer oligonucleotide typing of *Mycobacterim bovis* strains..and other animals: a tool for studying epidemiology of tuberculosis. J Clin Microbiol ., 34 (11): 2734-40.
2. Mignone, W., Ercolini, C., Fisichella, S. y Dondo, A. (1991). Osservazioni preliminari su alcuni episodi di tuberculosi nel cinghiale (*Sus scrofa*). Estratto da Selezzione Veterinaria 32(5):843-849.
3. De Lisle, G. W., Mackintosh, C. G. and Bengis, R. G. (2001). *Mycobacterium bovis* in free-living captive wildlife, inlcluding farmed deer. Rev. Sci. Tech. Off. Int. Epiz. 20 (1): 86-111.

Lesion type	Head lymph nodes	Thoracic lymph nodes	Abdominal lymph nodes
Caseous necrosis	83 %	65%	50%
Calcification	27 %	17%	25%
Miliary foci	27%	35%	40%