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Seroprevalence of Borna disease virus antibodies in alpacas and llamas in Austria

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Summary

Borna disease (BD) is a fatal neurological disease that frequently affects horses and sheep. The bicolored white-toothed shrew (*Crocidura leucodon*) has been confirmed as the natural virus reservoir. We analysed the seroprevalence of anti-Borna disease virus (BoDV) antibodies in the Austrian New World Camelid population. We took blood samples from 445 animals in 86 herds from all Austrian federal states and tested the serum samples for antibodies by an indirect immunofluorescence assay.

We tested 445 samples (266 from alpacas and 179 from llamas). For seven serum samples (3 from alpacas and 4 from llamas), no results could be obtained. We found antibodies in 52 samples (11.9 %), with llamas showing a seroprevalence of 12.6 % (22/175) and alpacas of 11.4 % (30/263). Breaking the results down by federal state, Vienna had a higher positive test rate than Salzburg (p=0.0086) and Upper Austria (p=0.0332). Burgenland (p=0.0284), Lower Austria (p=0.0152), Styria (p=0.0229) and Carinthia (p=0.0483) had a significantly higher seroprevalence than Salzburg.

Zusammenfassung

Untersuchungen zur Seroprävalenz von Antikörpern gegen das Virus der Bornaschen Krankheit bei Alpakas und Lamas in Österreich

Einleitung

Die Bornasche Krankheit (Borna disease, BD) ist eine akut bis subakut, selten chronisch verlaufende Meningoenzephalomyelitis, die außer bei Equiden und Schafen, gelegentlich auch bei anderen Spezies auftreten kann. Der Erreger, übertragen über die Feldspitzmaus (*Crocidura leucodon*), ist ein einzelsträngiges, behülltes RNA-Virus mit negativer Polarität und wird in der Ordnung *Mononegavirales* der Familie *Bornaviridae* zugeordnet.

Das Ziel der Untersuchung war, die Seroprävalenz von Antikörpern gegen das Virus der Bornaschen Krankheit (Borna disease virus, BoDV) an einer größeren Stichprobe von Serumproben von Neuweltkameliden in Österreich zu ermitteln.

Material und Methode

Es wurden 445 Blutproben von Tieren aus 86 Herden in Österreich genommen. Die Serumproben von 266 Alpakas und 179 Lamas wurden mittels eines indirekten Immunfluoreszenztests auf Antikörper gegen das Virus der Bornaschen Krankheit (Borna disease virus; BoDV) untersucht.

Ergebnisse

Anti-BoDV Antikörper konnten bei 11,9 % (52/438) der Tiere nachgewiesen werden. Sieben Proben (3 von Alpakas und 4 von Lamas) waren wegen unspezifischer Reaktionen im Zellkern nicht auswertbar. Die Titer lagen zwischen 1:10 und 1:640. Alpakas hatten eine Seroprävalenz von 11,4 % (30/263) und Lamas von 12,6 % (22/175). Positive Tiere kamen in Wien (5/21) häufiger vor als in Salzburg (0/38) (p=0,0086) und Oberösterreich (3/66) (p=0,0032). Im Burgenland (4/24; p=0,0284), der Steiermark (15/100; p=0,0229), Niederösterreich (10/64; p=0,0152) und Kärnten (8/61; p=0,0483) waren die Nachweishäufigkeiten signifikant höher als in Salzburg. Antikörper wurden auch bei Tieren in Tirol (3/48) und Vorarlberg (1/17) nachgewiesen.

Um nähere Angaben zu den Tieren mit Antikörpern gegen das Virus der Bornaschen Krankheit zu

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(Only statistically significant differences displayed.)

The study confirmed that alpacas and llamas are susceptible to BoDV infection. BoDV antibodies were found in 11.9 % of clinically healthy animals in all Austrian federal states, with a higher rate in the eastern provinces. There is a good agreement between the high rates of seroprevalence and the increased occurrence of the bicolored white-toothed shrew in eastern Austria.

Abbreviations: BoDV = Borna disease virus; FITC = Fluorescein isothiocyanate; IHC = Immunohistochemistry; LARA = Lama- und Alpakaregister Austria; MDCK = Madin-Darby canine kidney cells; NWC = New World camelids

erhalten, wurden 33 Fragebögen an die Halter der Herden verschickt und 27 wurden ausgefüllt zurückgesendet.

Bei 85 % der Halter (23/27) war die Bornasche Krankheit unbekannt. Ein Drittel gab an, Symptome, die bei der Bornaschen Krankheit auftreten können, wie Lahmheit, Gleichgewichtsstörungen, Apathie oder Gewichtsverlust, bemerkt zu haben. Knapp ein Viertel verzeichnete auch plötzliche Todesfälle, die mit neurologischen Störungen einhergingen, jedoch wurde in keinem Krankheitsfall die Bornasche Krankheit nachgewiesen. Außerdem konnten keine Unterschiede zwischen Herden, in denen anti-BoDV Antikörper und in denen keine nachgewiesen wurden, festgestellt werden. Acht von 27 Betrieben (31 %) berichteten über ein erhöhtes Schadnageraufkommen in der letzten Zeit. Jedoch nur knapp über die Hälfte der Befragten führte eine Bekämpfung durch.

Diskussion

Die Studie bestätigt, dass Alpakas und Lamas in Österreich Kontakt mit dem BoDV hatten. Bei 11,9 % der klinisch unauffälligen Tiere konnten Antikörper gegen BoDV nachgewiesen werden. Die höhere Seroprävalenzrate in den östlichen Bundesländern lässt sich mit einer stärkeren Verbreitung der Feldspitzmaus (*Crocidura leucodon*) in diesen Regionen erklären.

Introduction

Borna disease (BD) is a fatal neurological disease in a variety of mammals such as horses, sheep, goats, cats, dogs, cattle, donkeys and mules (Rott & Becht 1995; Richt & Rott 2001; Herrsche 2007). The name originated from an epidemic among cavalry horses near the town of Borna in the state of Saxony (Germany) in 1894–1896. The aetiological agent, Borna disease virus (BoDV), is a single-stranded non-segmented negative-sensed and enveloped RNA virus classified within the order *Mononegavirales*, family *Bornaviridae*, species *Mammalian 1 Orthobornavirus*. It has two genotypes (BoDV-1, BoDV-2) (Amarasinghe et al. 2019), with BoDV-1 the most prevalent bornavirus in mammals.

BD is not caused by the virus itself but by a T-cell-mediated immunopathological process in the central nervous system (Stitz et al. 2002). The zoonotic potential of the virus was confirmed in human BD cases, with fatal encephalitis induced by BoDV-1 (Korn et al. 2018; Schlottau et al. 2018). The persistently BoDV-1 infected bicolored white-toothed shrew (*Crocidura leucodon*, *C. leucodon*) is the natural reservoir host (Hilbe et al. 2006; Bourg et al. 2013; Dürrwald et al. 2014).

Clinical BD is geographically restricted to Germany, Switzerland, Liechtenstein and Austria. The first case of BD in Austria was diagnosed in a dog in 1993 in Vorarlberg (Weissenböck et al. 1998). Cases of acute BD in horses have been reported in Vorarlberg in 1993 and 1997 and in Styria in 1998 (Suchy et al. 2000). The BoDV isolate of the horse in Styria was given the name BoDV-2 because the nucleotide sequence differed more than 15 % from that of the reference strains (Nowotny et al. 2000). In 2015 and 2016, four horses with BD were documented in Upper Austria and BoDV-1 was found in two species of shrew (Weissenböck et al. 2017). The seroprevalence study lists only BoDV without specifying the genotype, because both genotypes, BoDV-1 and BoDV-2, are documented in Austria; to date, BoDV-2 has only been recorded in Austria.

BoDV-1 mainly infects horses and sheep, very rarely other animal species such as rabbits, cattle, goats, dogs and zoo animals such as New World Camelids (NWC) (Richt & Rott 2001). In 1993, two cases of BD in alpacas in a zoological garden were described histologically (Altmann et al. 1976), with the results subsequently confirmed by immunohistochemistry in one case (Schüppel et al. 1994). BD has also been found in NWC in Germany and Switzerland (Herrsche 2007; Jacobsen et al. 2010; Schulze et al. 2020; Malbon et al. 2022).

There is no epidemiological information on BoDV infections in NWC in Austria, although the number of domesticated NWC (alpacas and llamas) in Austria has increased rapidly over the past years (Bauerstatter et al. 2018). As registration of NWC in Austria is not obligatory, the exact number of animals is not known. The voluntary llama and alpaca register (LARA) in Austria estimates about 9,000 animals, of which approximately 40 % are llamas (Scheiber, personal communication).

We have examined serum samples from NWC in Austria for antibodies against BoDV and determined the seroprevalence. We compare the results with the geographic prevalence of *C. leucodon* (Spitzenberger 2001), the natural reservoir of BoDV. By means of questionnaires, we solicited information on age, gender, breed and herd size of seropositive NWC compared to seronegative animals and on the knowledge of Borna disease among NWC breeders and owners.

Materials and Methods

Study population and blood samples

Blood samples were taken for a 2015 study of the prevalence of infectious diseases, collecting data on management and health measures in Austrian NWC herds after approval from the ethics committee of the Vetmeduni Vienna and the national Austrian authority in accordance with §26 of the Animal Welfare Act (68.205/0171-II/3b/2013).

At that time, the NWC population in Austria was estimated to be about 4,000–6,000 animals (Bauerstatter et al. 2018). We visited a total of 86 herds in all Austrian federal states, collecting information on herd structure, feeding and management and taking blood samples. The herds were divided into four group sizes (<5 animals, 5–10 animals, 11–20 animals, >20 animals). The numbers of herds and animals were adjusted according to the estimated numbers of animals in the federal states, the species (alpaca and llama), the age and the gender to make the study sample as representative as possible. We aimed to include approximately 10 % of the New World camelids. The selection of the herds and animals and sample preparation was described in more detail by Stanitznig et al. (2016). Samples were stored at -80 °C until analysis.

Questionnaire

Additionally, we sent a questionnaire to the owners in 2020–2021 with questions on their knowledge of Borna disease, rodent infestation, pest control and previous examinations to diagnose BD. The owners were also asked whether they had observed symptoms such as lameness, changes in behaviour, reduced feed intake, apathy, loss of weight and libido without obvious reason, neurological signs such as imbalance, or (sudden) death in their herds. We also requested the owners to provide information on animal contact and movements, such as breeding of the mares in other herds, visiting shows and exhibitions, and hosting guest animals. The questionnaire gives retrospective information on the herd management practices of previous years, which may have influenced the herd conditions. However, we acknowledge that there was a time difference between sampling and the questionnaire.

Serology

We tested the sera for antibodies against BoDV at the Institute of Virology at the Justus-Liebig-University Giessen using an indirect immunofluorescence assay (Narayan et al. 1983). Serum samples were incubated on slides with acetone-fixed MDCK cells (Madin-Darby canine kidney) persistently infected with a BoDV isolate from an alpaca. After incubation for 40 minutes, cells were exposed for 40 minutes to fluorescein isothiocyanate (FITC)-conjugated goat anti-alpaca IgG antibodies (Dianova, Germany). Negative and positive sera were used as controls in each test, along with uninfected MDCK cells. Sera were diluted fourfold from 1:10 to 1:2560. Sera with anti-BoDV antibodies caused a brilliant granular fluorescence in the nucleus. The diagnostic procedure (IIFT) and the laboratory (VIRO VET DIAGNOSTIK GIESSEN) have been accredited by DAkkS since 2015.

Statistical analysis

Data and serological results and responses to the questionnaire were documented in spreadsheets using Microsoft Excel 2021 (Version 16.50). We used the same program for descriptive statistics and to prepare figures and calculate frequency distributions. We used the Fisher exact test to compare the results for age, gender, breed and species. Differences with $p<0.05$ were considered statistically significant.

Results

Serology

We had 445 serum samples of NWC of Austria available to determine anti-BoDV antibodies. We could evaluate 438 of them with an indirect immunofluorescence assay: 263 samples from alpacas and 175 from llamas (Tab. 4). Seven samples could not be evaluated due to non-specific reactions in the nucleus. The titres ranged between 1:10 and 1:640 (Tab. 1). Nearly 12 % of the serum samples (52/438) showed anti-BoDV antibodies. Antibodies could be determined in 30 alpacas and 22 llamas; the proportions did not differ substantially (llamas: 22/175, 12.6 %; alpacas: 30/263, 11.4 %).

Alpaca blood samples (which could be evaluated) were generally taken from females (158/263, 60 %), while the llama blood samples more often came

Tab. 1: Distribution of anti-BoDV antibody titres in alpacas and llamas / Verteilung der anti-BoDV Antikörpertiter in Alpakas und Lamas

Titer/Titer	Number of positive animals / Anzahl der positiv getesteten Tiere	Percentage of all animals tested (n=52) / Prozentualer Anteil bezogen auf alle getesteten Tiere (n=52)
1:10	8	15.4 %
1:40	33	63.5 %
1:160	8	15.4 %
1:640	3	5.7 %
Total/Gesamt	52	100.0 %

from males (102/175, 58.3 %). These proportions represent the gender frequencies in alpaca and llama herds. Twenty-two of 158 female alpacas (14.0 %) and 7/104 male alpacas (6.7%) had anti-BoDV antibodies, while 11/72 female llamas (15.3 %) and 11/102 male llamas (10.8 %) were seropositive. Anti-BoDV antibodies were found in 15.5 % (18/116) of the Classic Llamas, in 11.7 % (30/257) of the Huacaya alpacas, in 7 % (4/58) of the Woolly Llamas but in none of the Suri Alpacas (n=5). Seroprevalence was higher in the five- to nine-year-old group (25/157, 15.9 %) (p=0.0337) than in the group of animals younger than five years (18/231, 7.8%); 6/44 (13.6 %) of the older animals (≥10 years) were seropositive. The age distribution did not allow further analyses.

Data for herd sizes indicated that 15.6 % of the animals from larger herds (more than 20 animals) were seropositive. 13.5 % of the animals kept in small herds (less than 5 animals) had anti-BoDV antibodies, while 8.0 % llamas and alpacas from herds with 5–9 animals and 8.5 % of groups with 10–20 animals were seropositive (Tab. 2). The differences between seroprevalence according to the herd size were not statistically significant.

Figures 1 and 2 show the numbers of positive and negative animals in the federal states, as well as the geographic distribution of the herds and their sizes. About half of the herds in Styria (8/15 herds), Burgenland (3/5 herds), Vienna (4/6 herds), Lower Austria (8/16 herds) and Carinthia (6/12 herds) had seropositive animals, while less than half of the herds in Upper Austria (4/11 herds), Tyrol (3/9 herds), Vorarlberg (1/5 herds) and Salzburg (0/6 herds) had seropositive animals. In total, we identified 37/86 herds (44 %) animals with anti-BoDV antibodies. Vienna had a seroprevalence of 24% (5/21), Burgenland 17% (4/24), Styria 15 % (15/100), Lower Austria 16 % (10/64) and Carinthia 13 % (8/61). Upper Austria (3/66), Tyrol (3/48) and Vorarlberg (1/17) had a seroprevalence between 5 % and 6 %, while no seropositive animal could be detected in Salzburg (0/38). Statistically significant differences between the seroprevalences were found for some federal states. Vienna had a higher seroprev-

Tab. 2: Animals with anti-BoDV antibodies broken down by herd size / Tiere mit anti-BoDV Antikörpern bezogen auf die Größe der Herde

Herd size (animals) / Herdengröße (Tiere)	Number of animals with antibodies / Anzahl der Tiere mit Antikörpern	Percentage of animals in their age group with antibodies / Anteil der Tiere mit Antikörpern bezogen auf Altersgruppe
<5	10	13.5 % (10/74)
5–9	9	8.0 % (9/113)
10–20	10	8.5 % (10/118)
>20	20	15.6 % (20/128)
No information / keine Angaben	3	60.0 % (3/5)
Total / Gesamt	52	11.9 % (52/438)

Tab. 3: Questionnaire: Answers to selected questions from herds with and without animals with anti-BoDV antibodies / Fragebogen: Antworten auf ausgewählte Fragen zu Herden mit und ohne anti-BoDV Antikörper

	Herds with BoDV antibodies / Herden mit BoDV Antikörpern (n=5)		Herds without BoDV antibodies / Herden ohne BoDV Antikörper (n=22)		
Number of animals / Tieranzahl	23		106		
Age group <5 years / Altersgruppe <5 Jahre	11		60		
Age group 5–9 years / Altersgruppe 5–9 Jahre	7		30		
Age group ≥10 years / Altersgruppe ≥10 Jahre	3		9		
No information on age / Ohne Angabe des Alters	2		7		
	yes/ja	no/nein	yes/ja	no/nein	No answer/ keine Antwort
Attending shows or exhibitions / Teilnahme an Shows und Ausstellungen	1	4	3	19	0
Keeping guest animals / Unterbringung von Gasttieren	0	5	1	21	0
Mating mares in other herds / Stuten in anderen Herden decken lassen	2	3	1	21	0
Symptoms possibly associated with BoDV-1 infections / Symptome, die bei BoDV-1 Infektionen auftreten können	1	4	8	14	0
Sudden deaths / Auftreten plötzlicher Todesfälle	2	3	5	17	0
Increase in number of rodents / Zunahme an Schadnagern	3	2	5	16	1

alence than Salzburg (p=0.0086) and Upper Austria (p=0.0032). Burgenland (p=0.0284), Lower Austria (p=0.0152), Styria (p=0.0229) and Carinthia (p=0.0483) had a higher seroprevalence than Salzburg (Fig. 1 and 2). Other comparisons yielded p-values in the range of 0.1175–0.7662.

Responses to the questionnaire

The questionnaire was completed by 27 farm owners of 86 farms (31 %), with the remaining owners not responding or having given up keeping animals. Five of the owners had seropositive animals in their herd, while 22 responses came from seronegative herds. The small number of seropositive herds did not allow a statistical evaluation (Tab. 3). Animal contact with other herds was infrequent and restricted to during shows and exhibitions, while hosting guest animals or while mating mares in other herds (Tab. 3). Tab. 3 provides further details.

Discussion

The sero-epidemiological study showed an anti-BoDV seroprevalence of 11.9 % in healthy alpacas and llamas in Austria at the time the blood samples were taken. We found no significant differences in seroprevalence between alpacas and llamas, between breeds of alpacas, between age groups or between herd structures. There is a good agreement with sero-epizootiological studies in Germany, which found a seroprevalence of 11.5 % in healthy horses, although the figure increased to 22.5 % in endemic areas and up to 50 % in stables with acute BD cases (Richt et al. 2000).

We did find a significant difference in the seroprevalence of animals in the eastern and southern part of Austria compared to the western and northern part. Regions with a higher percentage of seropositive alpacas and llamas (Vienna 24 %; Burgenland 16 %; Styria 15 %; Lower Austria 14%; Carinthia 13%) had more *C. leucodon* (Spitzenberger 2001), while we

Tab. 4: Overview: Numbers of positive, negative and unspecified (n.sp.) animals, broken down by species, breed and gender (f = female, m = male) / Übersicht: Daten aller positiven, negativen und Tiere ohne Angabe (o.A.), unterteilt nach Tierart, Rasse und Geschlecht (w = weiblich, m = männlich)

Species / Tierart		Total/ Gesamt	Positive	Negative	N.sp./ o.A.	Positive plus negative animals / Positive und negative Tiere
Alpaca						
Total / Gesamt		266	30	233	3	263
Breed / Rasse	Huacaya	261	30	228	3	258
	Suri	5	0	5	0	5
Gender / Geschlecht	f/w	160	22	136	2	158
	m	105	7	97	1	104
	n.sp./o.A.	1	1	0	0	1
Llama						
Total / Gesamt		179	22	153	4	175
Breed / Rasse	Classic Llama	119	18	98	3	116
	Woolly Llama	59	4	54	1	58
	Lama n.sp./o.A.	1	0	1	0	1
Gender / Geschlecht	Llama f/w	75	11	61	3	72
	Llama m	103	11	91	1	102
	Llama n. sp./o.A.	1	0	1	0	1
All animals / Alle Tiere		445	52	386	7	438

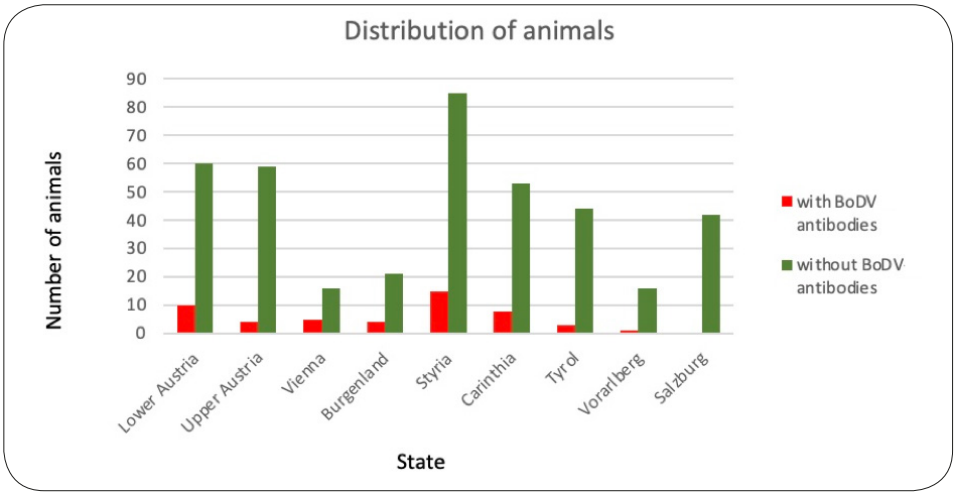

Fig. 1: Number of animals with or without anti-BoDV antibodies in the Austrian federal states / Anzahl der Tiere mit und ohne anti-BoDV Antikörper(n) in den österreichischen Bundesländern

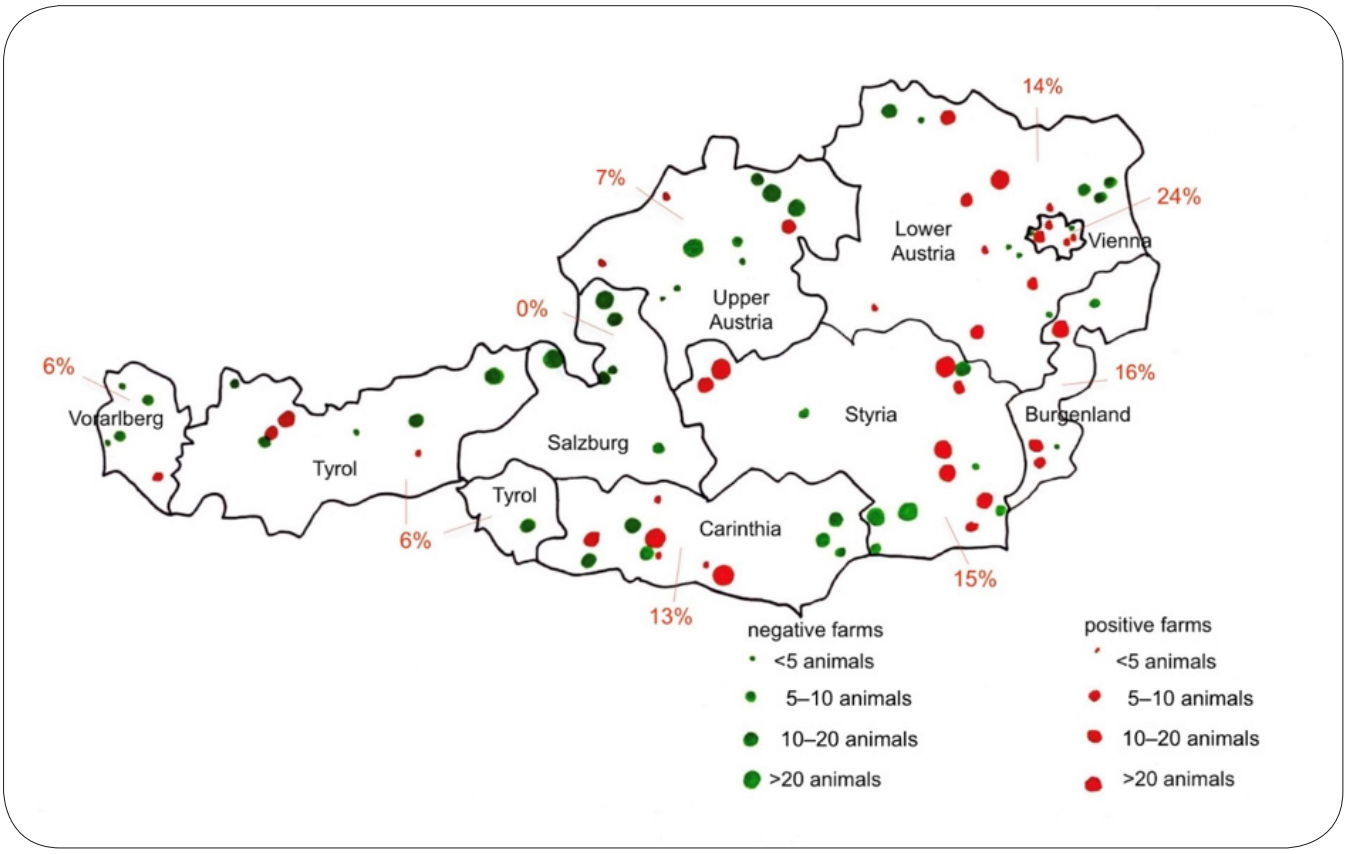

Fig. 2: Location of herds with (red dots) and without (green dots) anti-BoDV antibodies. The diameter of the dot represents the herd size. For each federal state, the percentage of antibody-positive animals in the studied population is given. Please note that the figure does not display the precise geographical locations of the herds. / Lage der Herden mit (rote Kreisflächen) und ohne (grüne Kreisflächen) anti-BoDV Antikörper(n). Der Durchmesser der Kreisfläche gibt die Herdengröße an. Für jedes Bundesland ist der Prozentsatz der Tiere mit anti-BoDV Antikörpern bezogen auf die untersuchten Tiere angegeben. Die Darstellung erlaubt keine Rückschlüsse auf die exakte geographische Lokalisation der Herden.

found a low percentage of seropositive animals (Upper Austria 7%; Vorarlberg 6%; Tyrol 6 %; Salzburg 0 %) in regions with a low occurrence of *C. leucodon* (Fig. 2). The southern and eastern parts of Austria provide optimal habitat for *C. leucodon*: warm and dry areas, not higher than 680 m above sea level and mainly urban and arable land (Kraft 2008). For practical reasons, we reported the seroprevalence in alpacas and llamas according to federal state; further epidemiological studies might explicitly consider for each farm, if the farm environment is a favourable habitat for *C. leucodon*. The first and only time that BoDV-1 in *C. leucodon* was found in Austria was in 2015. The infected animals were collected in a region with maximal distance of about 17 km to four horses with BD in Upper Austria (Weissenböck et al. 2017).

As the blood samples were taken in 2015, we sent questionnaires in 2021 to the farm owners to learn more about the seropositive alpacas and llamas. Although the information was retrospective, there might have been changes in herd management between sampling and completion of questionnaire that might be not reported. This general limitation of retrospective analysis is likely to be the case in the present study. Unfortunately, the small number of questionnaires returned did not allow a significant statistical analysis. One focus of the questionnaire was on BD: the knowledge of the owners about BD, observations of animals with symptoms that could occur with BD or the proof of BD. About 15 % of the breeders had knowledge of BD and half of them had seen clinical signs known for animals with BD.

The incubation period for BD after natural infection in horses ranges from two to six months. Typical clinical signs are simultaneous or consecutive changes in psyche, sensorium, sensibility and mobility. The most common clinical signs are depression with apathy, somnolence and stupor. The neurological signs are variable and complex (Richt et al. 2000). To complicate the picture further, BoDV-1 infection can lead to different forms of BD. Peracute BD leads to death 1–3 days after the onset of initial signs and acute BD 1–6 weeks after the onset of clinical signs (Richt et al. 2000). Because the clinically suspicious animals were not examined further, either serologically *intra vitam* or with virological methods *post mortem*, we could not draw any conclusions about the clinically noticeable cases.

The incidence of BD in endemic areas in Germany ranges from 0.02 % to 0.04 %. There is no explanation for the discrepancy between the high seroprevalence in horses and the low incidence of BD. It is assumed that

various factors cause BD after infection with BoDV-1: age, immune status, genetic factors of the animal and genetic characteristics of the virus strain. Many cases of BoDV-1 infection are clinically inapparent (Richt et al. 2000) and it is not known whether the presence of serum antibodies in clinically healthy horses indicates a former BoDV-1 infection with viral clearance (Herden et al. 1999). As far as we are aware, these issues are also unknown in New World camelids.

About 30 % of the owners of the farms observed an increase in the number of rodents during previous years. *C. leucodon* belongs to the order *Eulipotyphla*, family *Soricidae* and we cannot make any definitive statement on the occurrence of *C. leucodon* for the farms on which rodents were observed.

In conclusion, our study confirms the presence of BoDV in Austria and shows for the first time that alpacas and llamas contain antibodies to BoDV, with a seroprevalence of 11.9 %. It remains of interest to prove cases of BD in alpacas and llamas and to find BoDV-infected shrews in regions other than Upper Austria. It also seems advisable to continue to monitor the seroprevalence in other countries and to investigate animals that have been euthanized or have died with neurological symptoms. Hygienic precaution should be considered when handling potentially contaminated material to protect animals and humans. In addition to the clinical investigation and diagnostic procedures in animals and humans, a surveillance of the animal reservoir in the environment is necessary for risk assessment and disease prevention.

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Fazit für die Praxis:
Bei Vorstellung von Neuweltkamelen mit Krankheitssymptomen wie Apathie, Gleichgewichtsstörungen, Lahmheit, Gewichtsverlust ohne ersichtlichen Grund, verminderter Futteraufnahme, Verhaltensstörungen oder Libidoverlust bei Hengsten sollte eine Infektion mit dem BoDV als Differentialdiagnose in Betracht gezogen werden.

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