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Research article

Prevalence and genetic diversity of rodent-associated *Bartonella* in Hulunbuir border regions, China

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ABSTRACT

Bartonella spp. are globally distributed gram-negative facultative intracellular bacteria that infect a wide range of hosts. Rodents are natural reservoirs of many *Bartonella* species, some of which are also pathogenic to humans. The rapid development of transportation and tourism has highlighted the risk of *Bartonella* transmission to humans. Thus, it is essential to maintain surveillance of *Bartonella* spp. infections in rodents. In China, *Bartonella* spp. infections have been monitored in various areas; however, these have not included the Hulunbuir border regions. In the present study, we monitored the prevalence and genetics of rodent-associated *Bartonella* spp. in the Hulunbuir border regions. Eleven rodent species were captured at five ports. Eight species were confirmed as *Bartonella*-positive using quantitative PCR assay, with an overall positivity rate of 20.05 %. *Lasiopodomys brandtii* was the predominant rodent species captured for *Bartonella* detection. Sequencing and phylogenetic analysis (using the maximum likelihood method) revealed the presence of three *Bartonella* species in these rodents, including two pathogenic to humans, namely, *Bartonella alsatica* and *Bartonella grahamii*. *B. grahamii* was the predominant *Bartonella* species identified in the rodents. Taken together, these results highlight the prevalence and genetic diversity of *Bartonella* spp. in rodents in the Hulunbuir border regions, indicating the need for risk assessment of human spillover.

1. Introduction

Bartonella spp. are gram-negative facultative intracellular bacteria, first identified in patients in Peru in 1909 and classified as rickettsia before 1993 [1]. *Bartonella* spp. belong to the group Alphaproteobacteria of the family *Bartonellaceae.* The host animals of *Bartonella* spp., including *Rodentia, Chiroptera, Artiodactyla, Lagomorpha, Carnivora,* and *Insectivora* are diverse [2]. *Bartonella* spp. mainly parasitize red blood cells or vascular endothelial cells and are transmitted by blood-sucking arthropods, including sandflies,

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fleas, lice, mites, and ticks. Humans can get infected with *Bartonella* through bites by blood-sucking arthropods, or through traumatic exposure to contaminants [3]. To date, at least 50 recognized and 10 candidate *Bartonella* species have been identified, of which more than 18 have been identified as zoonotic species [4–6].

Rodents belong to the order Rodentia and are considered natural reservoirs of *Bartonella* [5,7]. With their vast diversity and robust reproductive capabilities, rodents are ubiquitous and remarkably adaptable to a wide range of natural environments. As carriers of numerous pathogenic microorganisms, and with their close interactions with humans and other animals, rodents have become a focal point in the study and prevention of various environmental diseases [8,9]. In 1940, *Bartonella vinsonii* subsp. *arupensis* was first detected in rodents in the United States [10]. Approximately half a century later, *Bartonella* infection was detected in rodents in the UK, followed by Portugal [7], Canada [11], Japan [12], Thailand [13,14], China [15], Australia [16], Germany [17], Malaysia [18], India [19], and Switzerland [20]. *Bartonella*-positive rates in Portugal, Thailand, China, Malaysia, India, and other regions were >50 % [7, 13–15,18,19]. Currently, the members within genus *Bartonella* have been isolated from more than 98 rodent species in seven families,

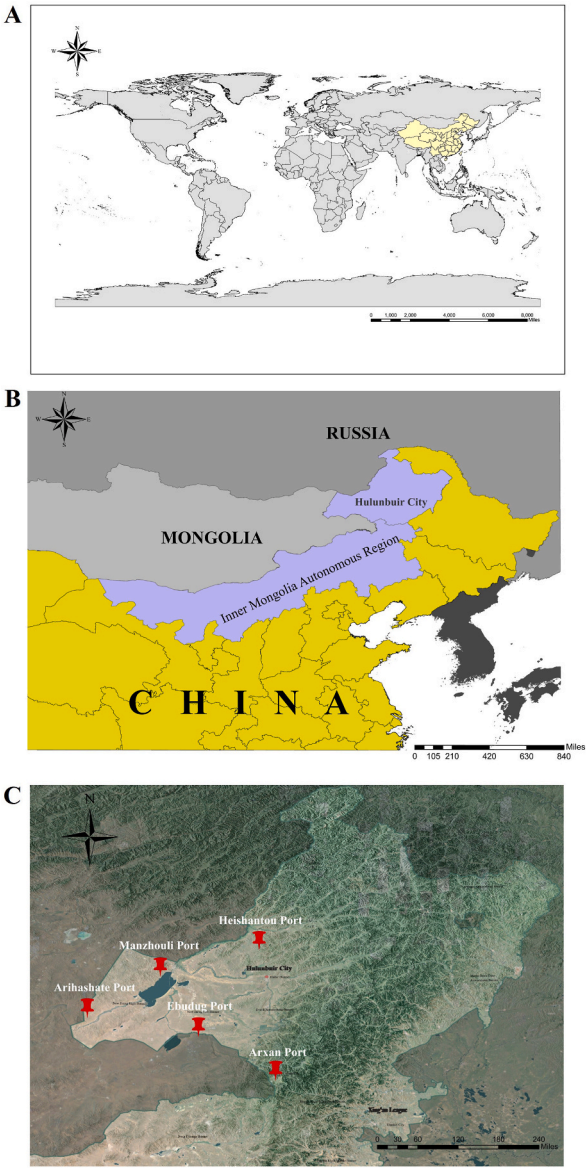

Fig. 1. Geographic maps of the rodent sampling sites in the Hulunbuir border regions. These maps were plotted using ArcGIS 10.2 (Environmental Systems Research Institute Inc., CA, USA). (A) World map showing the location of China. (B) Map showing the locations of Inner Mongolia Autonomous Region and Hulunbuir City. (C) Enlarged map of Hulunbuir City; the red symbols indicate the sampling regions. Arihashate Port: latitude 48.4439°N, longitude 115.8815°E; Ebudug Port: latitude 50.2424°N, longitude 120.1909°E; Arxan Port: latitude 47.1771°N, longitude 119.9431°E; Heishantou Port: latitude 40.2340°N, longitude 122.0775°E; Manzhouli Port: latitude 49.5800°N, longitude 117.4500°E. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

and approximately 22 rodent-related *Bartonella* and their subspecies have been identified [21]. Eight species or subspecies of rodent-related *Bartonella*, namely *Bartonella alsatica, Bartonella doshiae, Bartonella elizabethae, Bartonella grahamii, Bartonella rattimassiliensis, Bartonella tribocorun, Bartonella vinsoni* subsp.*vinsonii*, and *B. vinsoni* subsp.*arupensis*, have been reported to be pathogenic to humans and can cause myocarditis, endocarditis, lymphadenitis, hepatitis, and other diseases [5]. Therefore, investigating the prevalence of rodent-related *Bartonella* infection is of great significance for controlling *Bartonella* infection in humans.

China is located in East Asia along the west coast of the Pacific Ocean, with a vast territory and complex and diverse terrain. Rodents are widely distributed throughout China. Investigations into rodent infection with *Bartonella* in China began in 1999 when Ying et al. isolated *B. elizabathae* from rodent blood [15]. Subsequently, studies were carried out in 15 provinces, including Yunnan [22], Heilongjiang [23], Taiwan [24], Zhejiang [25], Fujian [26], Hebei [27], Shanxi [28,29], Guangdong [30], Jiangxi [31], Guizhou [32], and Sichuan [33]. Hulunbuir is situated to the south of Russia and the east of Mongolia. Arihashate Port, Ebudug Port, Arxan Port, Heishantou Port, and Manzhouli Port are important border crossings in this region. Rodents have a strong tendency to move along the border and can freely migrate across it, which poses a significant risk for the introduction and spread of diseases transmitted by rodents [34]. Hulunbuir is one of the most beautiful grasslands in China. With 80,000 square kilometers of natural grassland, it is one of the four major grasslands worldwide and an important part of the eastern Mongolian grassland, creating favorable conditions for the growth and reproduction of rodents [35]. In recent years, with the continuous development of tourism and animal husbandry industries in Hulunbuir, human activities have gradually expanded, giving rise to greater opportunities for direct or indirect contact with rodents, increasing the risk of transmission of bartonellosis [36]. Therefore, in the present study, we explored the prevalence and genetics of rodent-associated *Bartonella* species in the Hulunbuir border regions to provide basic data for understanding the risk of zoonotic spillover and control of *Bartonella* infection in humans.

2. Materials and methods

2.1. Ethics statement

The protocols were approved by the Institutional Ethics Committee of Shenyang Agricultural University (No. 2021040701). All animal experiments were conducted in strict accordance with the animal husbandry guidelines of the Shenyang Agricultural University (Liaoning, China).

2.2. Rodent sample collection

The rodents were captured from the Hulunbuir border regions between June 2021 and November 2022. Five sites were selected for rodent trapping; their geographical distributions are shown in Fig. 1. The rat traps with peanuts dipped in sesame oil as bait were arranged every 5 m according to the movement trajectory or feces of the rodents. The traps were placed at 6:30 p.m. and retracted at 6:00 a.m. the next morning. The cages where the rodents were captured were brought to the laboratory and placed in a bucket of water, to which excess ether was subsequently added, and the bucket was then sealed. Each captured rodent was then transferred to a white cloth bag and transported to the Manzhouli International Travel Health Care Center for breed identification. Rodent species were identified and recorded in accordance with the Chinese Manual of Important Medical Animal Identification. The liver, spleen, lung, and kidney tissues of the rodents were collected aseptically and stored at −80 °C for further pathogenic detection.

2.3. Quantitative PCR-based Bartonella screening

Approximately 200 mg of mixed liver, spleen, lung, and kidney tissues from rodents were placed in a 2 mL RNase-free tube. To this, 5 zirconium beads and 500 μL of PBS were added for grinding. After grinding, the mixture was centrifuged at 6000 rpm for 10 min and 200 μL of the supernatant was used for nucleic acid extraction. A DNA extraction kit (TransGen Biotech Co., LTD, Beijing, China) was used to extract nucleic acids, following the manufacturer's instructions. *Bartonella* was detected using genus-specific quantitative PCR (qPCR) by amplifying a fragment of 301 bp within the transfer mRNA (*ssrA*) gene [37]. AceQ Universal U + Probe Master Mix V2 (Nanjing Vazyme Biotech Co., Ltd., Nanjing, China) was used for *Bartonella*-specific qPCR according to the manufacturer's instructions.

2.4. Molecular characterization of Bartonella spp.

In positive samples detected by qPCR, the citrate synthase (*gltA*), RNA polymerase subunit (*rpoB*), and *ssrA* gene fragments of *Bartonella* were further amplified by PCR and sequenced using Sanger's method [37–39]. PCR was performed using 2 × Taq Plus Master Mix II (Nanjing Vazyme Biotech Co., Ltd.). The amplification products of *gltA*, *rpoB*, and *ssrA* were 379, 866, and 301 bp in length, respectively, and were identified using 1.2 % agarose gel electrophoresis. The PCR products were purified and transported to Sangon Biotech Co., Ltd. (Shanghai, China) for Sanger sequencing.

2.5. Phylogenetic analysis

The sequences obtained were assembled using DNASTAR Lasergene v7.1 and then deposited in GenBank (Accession Number: OR739188-OR739220). The genotypes of the sequences were initially identified using the Basic Local Alignment Search Tool (BLAST) program of the National Center for Biotechnology (NCBI). For the phylogenetic analysis, reference sequences of different *Bartonella*

genotypes were downloaded from the GenBank database. A phylogenetic tree was constructed using the maximum likelihood (ML) method in MEGA X (https://www.megasoftware.net). A bootstrap test with 1000 bootstrap replicates was conducted to estimate the confidence.

2.6. Statistical analysis

The composition of rodent species in the Hulunbuir border regions and the detection rate of *Bartonella* infection in different collection areas and species were evaluated using the chi-square test or Fisher's exact test with SPSS 23.0 (SPSS, Inc., Chicago, IL, USA). Differences with a value of $p < 0.05$ were considered statistically significant.

3. Results

3.1. Rodent species captured in Hulunbuir border regions

A total of 878 rodents were collected from five collection areas in the Hulunbuir border regions between June 2021 and November 2022 (Table 1). Of these, 410 rodents were collected at the Arihashate Port, 162 from the Ebudug Port, 159 from the Arxan Port, 74 from the Heishantou Port, and 73 from the Manzhouli Port (Table 1). The captured rodents belonged to four families, nine genera, and eleven species (Table 1). *Lasiopodomys brandtii* showed the highest abundance, followed by *Apodemus agrarius* and *Microtus gregalis,* accounting for 61.5 % (540/878), 14.69 % (129/878), and 12.19 % (107/878) of the total rodents, respectively (Table 1 and Fig. 2A). Rodent species compositions varied significantly among the five ports ($P < 0.001$, $\chi^2 = 637.9$). The distribution of various rodent species across the five ports is presented in Table 1. The proportions of *Lasiopodomys brandtii* at Arihashate Port and Ebudug Port were significantly higher than those at the other three ports, at 88.53 % (363/410) and 90.12 % (146/162), respectively. Arxan Port was mainly inhabited by *Apodemus agrarius*, accounting for 81.13 % (129/159). Heishantou Port was mainly inhabited by *Microtus gregalis*, accounting for 63.51 % (47/74). *Meriones unguiculatus* was predominant at the Manzhouli Port, accounting for 60.27 % (44/73). *Apodemus agrarius* and *Apodemus peninsulae* captured at Arxan Port were not present at the other ports, whereas *Rattus norvegicus* was only collected from Heishantou Port, and *Cricetulus barabensis* was only found at Manzhouli Port (Table 1), suggesting that the compositions of rodent species in these three areas was more diverse than those in the other two ports.

3.2. Bartonella prevalence in captured rodents

qPCR was performed to detect *Bartonella* infection in tissue samples from the captured rodents. We found that 176 of the 878 samples were positive, with a positivity rate of 20.05 %. *Bartonella* was detected at all five ports, indicating that its wide distribution in the Hulunbuir border regions. The *Bartonella* infection rate was the highest in rodents at Ebudug Port, with 39.51 % (64/162), followed by Arihashate Port and Arxan Port, with positivity rates of 19.02 % (78/410) and 15.72 % (25/159), respectively. *Bartonella* infection rates at Heishantou Port and Manzhouli Port were lower, with 6.76 % (5/74) and 5.48 % (4/73), respectively (Table 1 and Fig. 2C). The difference in positivity rates among the different collection areas was significant ($P < 0.001$, $\chi^2 = 165.28$).

Eight of the 11 captured rodent species were infected with *Bartonella* (Table 1 and Fig. 2B). The positivity rate of *Bartonella* infection was the highest in *Allactaga sibirica*, with a positivity rate of 40 % (6/15), followed by 25.74 % (139/540) in *Lasiopodomys brandtii*,

Table 1
Positivity rates of *Bartonella* infection in rodents captured in Hulunbuir border regions in this study.

Family	Genus	Species	Arihashate Port	Ebudug Port	Arxan Port	Heishantou Port	Manzhouli Port	Total
Muridae	*Apodemus*	*Apodemus agrarius*	0	0	14/129 (10.85 %)	0	0	14/129 (10.85 %)
		Apodemus peninsulae	0	0	1/7 (14.29 %)	0	0	1/7 (14.29 %)
	Rattus	*Rattus norvegicus*	0	0	0	1/14 (7.14 %)	0	1/14 (7.14 %)
	Mus	*Mus musculus*	0	0	0	0/1	0/2	0/3
Cricetifae	*Cricetus*	*Cricetulus barabensis*	0	0	0	0	0/1	0/1
	Microtus	*Lasiopodomys brandtii*	69/363 (19 %)	59/146 (40.41 %)	9/18 (50 %)	0/3	2/10 (20 %)	139/540 (25.74 %)
		Microtus gregalis	7/38 (18.42 %)	3/8 (37.5 %)	1/3 (33.33 %)	0/47	0/11	11/107 (10.28 %)
	Phodopus	*Phodopus sungorus*	0	0	0/2	0	0/1	0/3
	Meriones	*Meriones unguiculatus*	1/4 (25 %)	0	0	0	2/44 (4.55 %)	3/48 (6.25 %)
Dipodidae	*Allactaga*	*Allactaga sibirica*	1/5 (20 %)	1/1 (100 %)	0	4/9 (44.44 %)	0	6/15 (40 %)
Sciuridae	*Citellus*	*Spermophilus dauricus*	0	1/7 (14.29 %)	0	0	4	1/11 (9.09 %)
Total			78/410 (19.02 %)	64/162 (39.51 %)	25/159 (15.72 %)	5/74 (6.76 %)	4/73 (5.48 %)	176/878 (20.05 %)

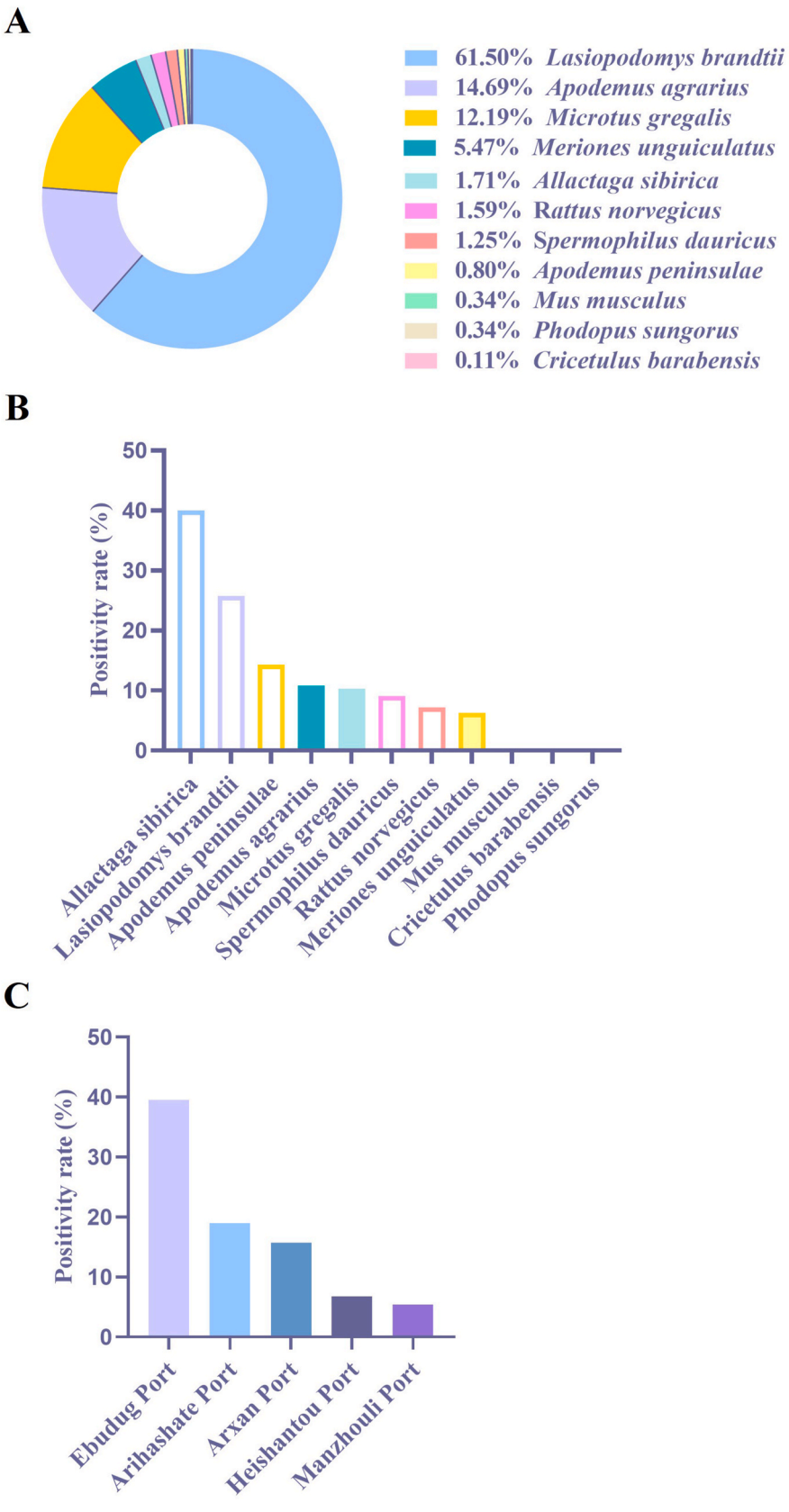

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Fig. 2. Composition of small rodent species and positivity rates of *Bartonella* infection in rodents captured in the Hulunbuir border regions. The graphs were generated using the GraphPad Prism version 8.0 (Graph Pad Software Inc., CA, USA). (A) Composition of small rodent species captured in the Hulunbuir border regions. (B) Positivity rates of *Bartonella* infection among different rodent species. (C) Positivity rates of *Bartonella* infection in rodents at different collection sites.

14.29 % (1/7) in *Apodemus peninsulae*, 10.85 % (14/129) in *Apodemus agrarius*, and 10.28 % (11/107) in *Microtus gregalis. Spermophilus dauricus*, *Rattus norvegicus*, and *Meriones unguiculatus* had positivity rates of <10 %. The positivity rates among the different rodent species were significant ($P < 0.001$, $\chi^2 = 33.2$).

3.3. Molecular characterization of Bartonella species in captured rodents

The qPCR-positive samples were further amplified and sequenced using the *gltA*, *rpoB*, and *ssrA* gene fragments of *Bartonella*. Phylogenetic trees were created based on sequences obtained by the ML method using MEGA X software. The analysis showed that three *Bartonella* species were detected at five sites in the Hulunbuir border regions. The *gltA*, *rpoB*, and *ssrA* gene fragments from the three ports (Arihashate Port, Heishantou Port, and Manzhouli Port) clustered with *B. grahamii* (Figs. 3–5), with 96–99 % identity to *gltA*, 98–100 % to *rpoB*, and 96–98 % to *ssrA*. The samples from Arxan Port grouped with *B. Japonica* (Figs. 3–5), with 96.8 % identity with *gltA*, 99.8 % with *rpoB*, and 98 % with *ssrA*. The *gltA* gene fragments of samples from Ebudug Port grouped with *B. alsatica* and *B. heixiaziensis* (Fig. 3), sharing 99.39 % identity with *B. alsatica* and 98.64 % identity with *B. heixiaziensis* at the nucleotide level. The *ssrA* and *rpoB* sequences also shared a high genetic similarity (98.6 % and 99.51 %) with those of *B. alsatica* (Figs. 4 and 5).

4. Discussion

Bartonella is a zoonotic pathogen with rodents as its natural host [5]. With the development of transportation, tourism, and the construction of some key projects, there has been an increase in opportunities for human contact with wildlife, particularly disease-carrying vectors, which may lead to the transmission of bartonellosis to humans. Therefore, it is necessary to investigate the prevalence of rodent-associated *Bartonella* spp.

In this study, 878 rodents were collected from five ports in the Hulunbuir border areas and tested for *Bartonella* infection. The overall infection rate of *Bartonella* at the five ports was 20.05 %, which was lower than that in most areas in China. The *Bartonella*-positivity rate in rodents at the Ebudug Port was the highest at 39.51 %, which is comparable to that found in the Qaidam Basin (38.6 %, 39/101 [40]), Shangdang Basin (37.4 %, [55/147] [29]), Qinghai-Tibetan Plateau (30.1 %, [31/103] [41]), and Zhejiang province (31.4 % [134/427] [25]), of China. The positivity rates at Arihashate Port and Arxan Port were 19.02 % and 15.72 %, respectively, similar to those found in rodents from Hebei (17 %, [38/223] [27]), Guizhou (16.05 % [13/81] [32]), and southeastern China (14.86 %, [169/1137] [42]). The positivity rates of rodent-associated *Bartonella* at Heishantou Port and Manzhouli Port were low, at 6.76 % and 5.56 %, respectively, which are similar to that in Guangzhou (6.4 % [30]) and eastern China (8.4 % [43]).

A total of 11 rodent species were captured from the Hulunbuir border areas. No *Bartonella* infection was detected in three rodent species: *Mus musculus*, *Phodopus sungorus*, and *Cricetulus barabensis*. Since only three individuals of each of these three rodent species were collected, the absence of *Bartonella* infection may not accurately reflect the true infection rate in these species. Among rodents positive for *Bartonella* infection, *Allactaga sibirica* had the highest positivity rate (40 %). *Bartonella* infections in *Allactaga sibirica* have been reported in the Qaidam Basin (100 %, [3/3] [40]), and Shaanxi Province (20 %, [1/5] [44]), of China. The other seven *Bartonella*-positive rodent species were *Lasiopodomys brandtii* (25.74 %), *Apodemus peninsulae* (14.29 %), *Apodemus agrarius* (10.85 %), *Microtus gregalis* (10.28 %), *Spermophilus dauricus* (9.09 %), *Rattus norvegicus* (7.14 %), and *Meriones unguiculatus* (6.25 %). *Apodemus peninsulae* captured from the Shaanxi Province (17.65 %, [3/17] [44]), Zhejiang Province (29 %, [2/7] [25]), the Qinghai-Tibetan Plateau (37.93 % [22/58] [41]), and Zhongtiao Mountain (100 % [1/1] [28]), were *Bartonella*-positive. *Apodemus agrarius* and *Rattus norvegicus*, which are widely distributed in China, have also been found to be infected with *Bartonella* in many areas of the country. *Bartonella* was also detected in *Microtus gregalis* collected from the Qinghai-Tibetan Plateau [41] and *Meriones unguiculatus* collected from Inner Mongolia [45]. *Lasiopodomys brandtii* has not previously been reported to harbor *Bartonella.*

Three species of *Bartonella* were identified in rodents from the Hulunbuir border areas. The samples from Arihashate Port, Heishantou Port and Manzhouli Port harbored *B. grahamii*, those from the Arxan Port harbored *B. Japonica*, and those from the Ebudug Port harbored *B. alsatica*. Therefore, the distribution of *Bartonella* was likely influenced by areas. Among the three *Bartonella* species, *B. alsatica* and *B. grahamii* have been reported to cause lymphadenopathy, endocarditis and neuroretinitis in humans [46–51]. To date, there have been four reported cases of human infections by *B. alsatica* [48–51]. The first and third patients were rabbit breeders and both presented with endocarditis post-infection [48,51]. The second case involved a rabbit butcher who accidently scratched her finger during work, leading to lymphadenopathy [50]. The fourth patient with a *B. alsatica* infection had a history of regularly hunting wild rabbits, and his clinical manifestation was acute renal failure due to a membranoproliferative glomerulonephritis [49]. To date, only two cases of *B. grahamii* infection in humans have been reported, one case of lymphadenitis caused by a cat scratch and one case of neuroretinitis in a dog owner [46,47]. Given that two species of human pathogenic *Bartonella* have been detected in the Hulunbuir border areas, it is crucial to continue monitoring *Bartonella* infection in this region. It is also essential to monitor antibody levels in the human population to prevent the transmission of *Bartonella* to humans.

In conclusion, we investigated the prevalence and genetic diversity of rodent-associated *Bartonella* in the Hulunbuir border regions in China. Eight of the 11 rodent species captured were *Bartonella*-positive, with an overall positivity rate of 20.05 %. Three *Bartonella*

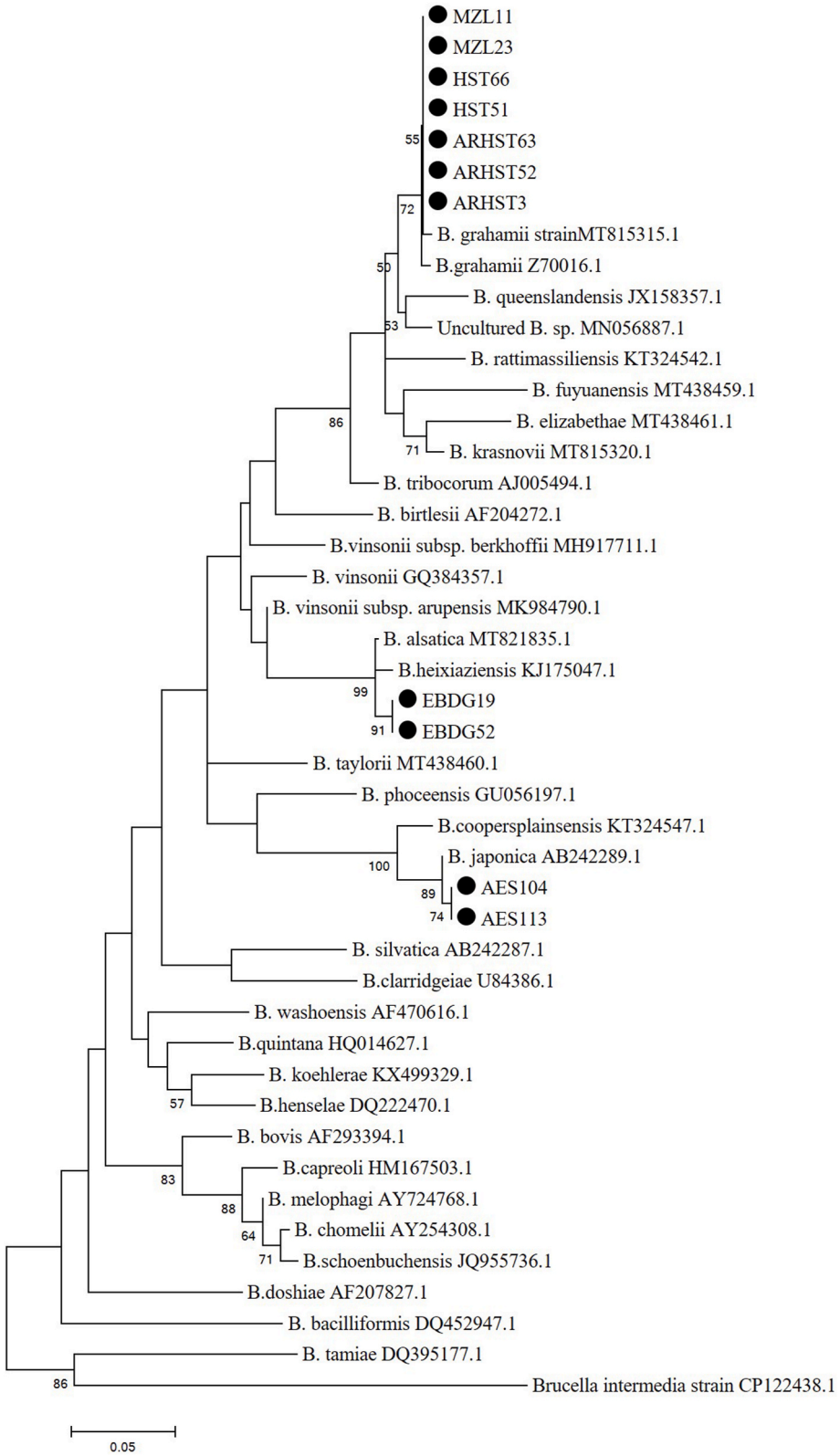

Fig. 3. Phylogenetic analysis of *gltA* gene fragments of *Bartonella*. Sequences of *Bartonella* determined in this study are marked with dark dots. EBDG, AES, HST, ARHST, and MZL correspond to the sampling locations Ebudug Port, Arxan Port, Heishantou Port, Arihashate Port and Manzhouli Port. The scale bar indicates the number of nucleotide substitutions per site.

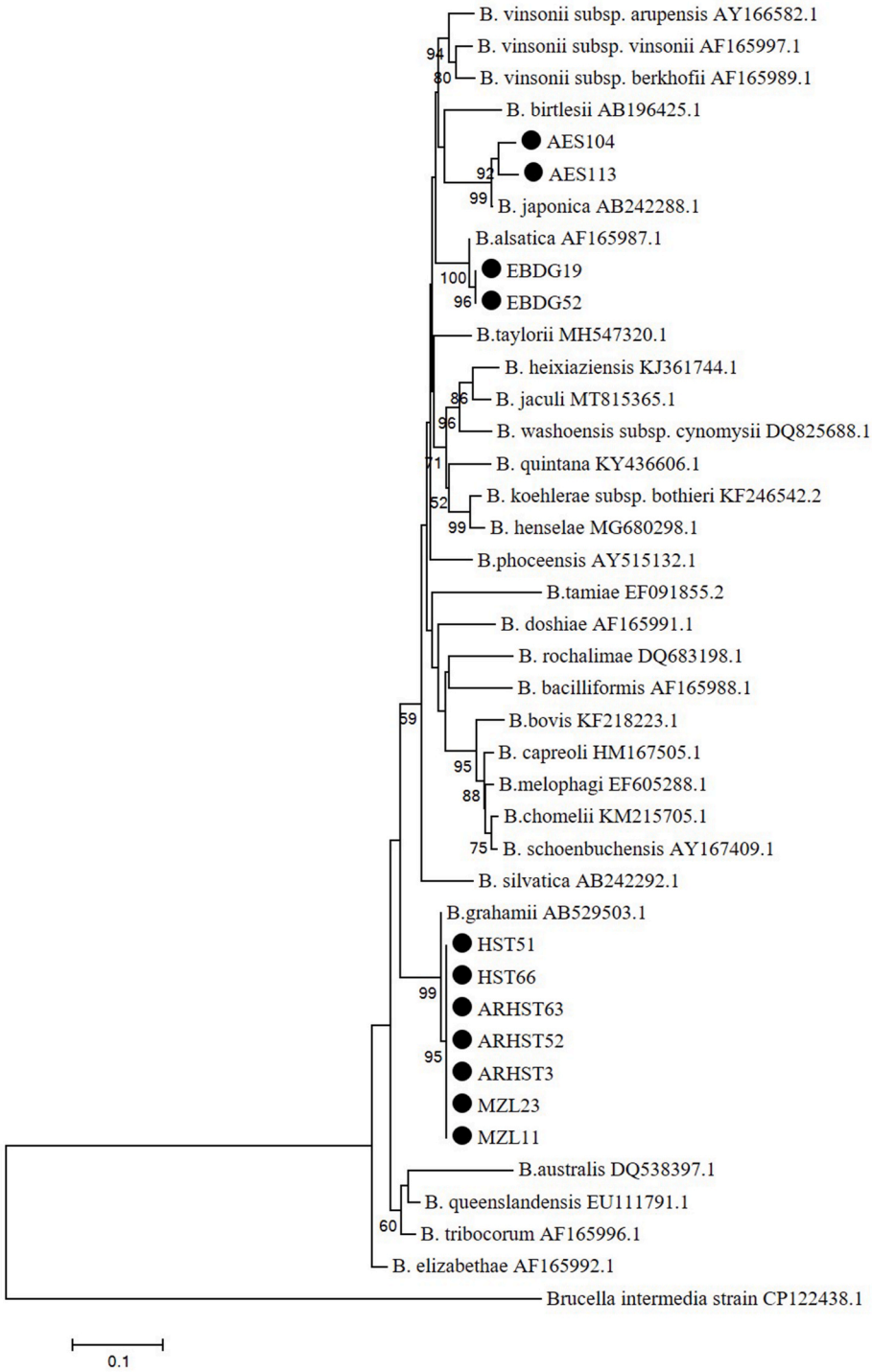

Fig. 4. Phylogenetic analysis of *ropB* gene fragments of *Bartonella*. Sequences of *Bartonella* determined in this study are marked with dark dots. EBDG, AES, HST, ARHST, and MZL correspond to the sampling locations Ebudug Port, Arxan Port, Heishantou Port, Arihashate Port and Manzhouli Port. The scale bar indicates the number of nucleotide substitutions per site.

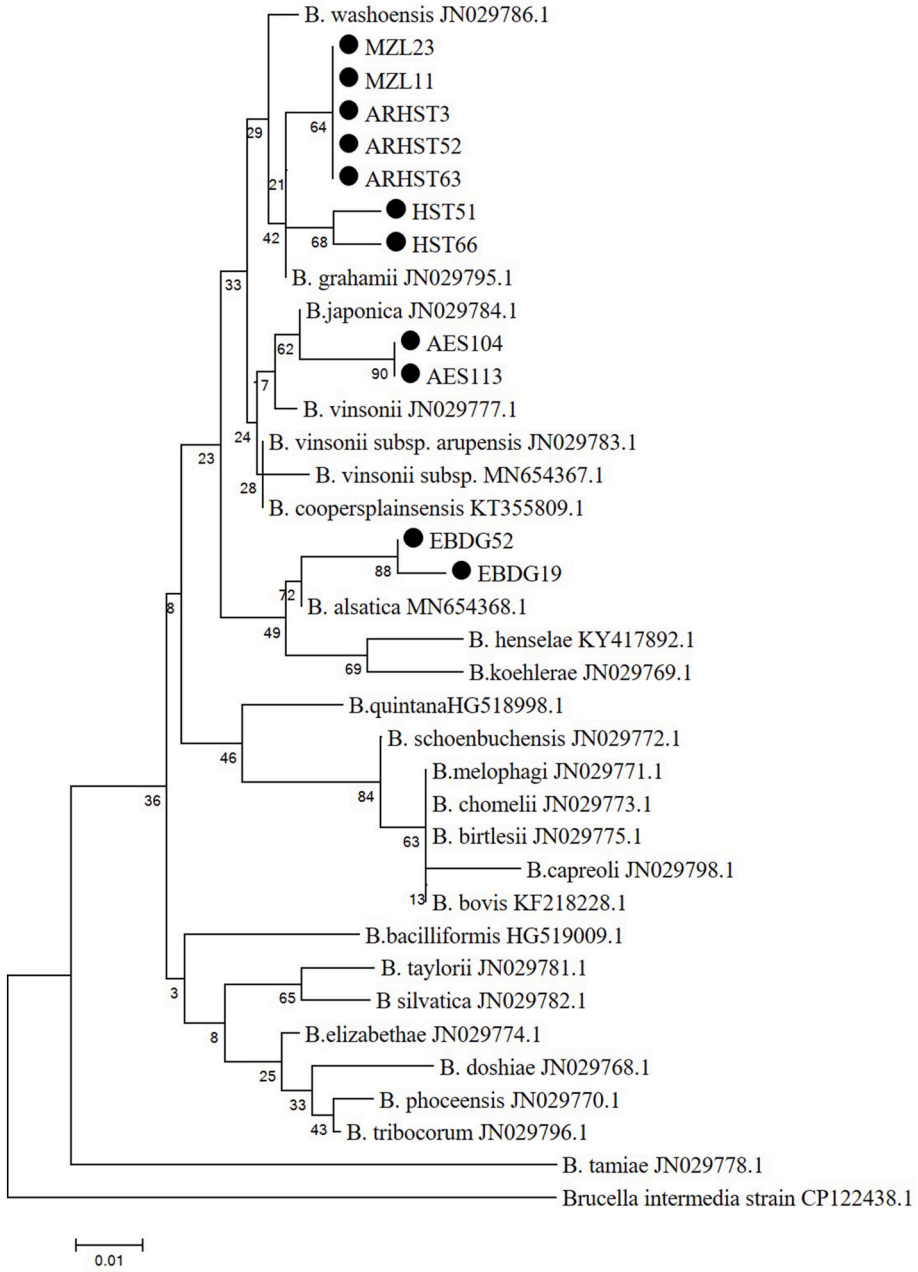

Fig. 5. Phylogenetic analysis of *ssr*A gene fragments of *Bartonella*. Sequences of *Bartonella* determined in this study are marked with dark dots. The identification EBDG, AES, HST, ARHST, and MZL correspond to the sampling locations Ebudug Port, Arxan Port, Heishantou Port, Arihashate Port and Manzhouli Port. The scale bar indicates the number of nucleotide substitutions per site.

species, including two pathogenic to humans, *B. alsatica* and *B. grahamii*, were detected in the captured rodents. *Lasiopodomys brandtii* was the dominant rodent species captured for the detection of *Bartonella,* whereas *B. grahamii* was the predominant *Bartonella* species in the captured rodents. Our results highlight the prevalence and genetic diversity of *Bartonella* species in rodents in the Hulunbuir border regions, indicating the need for risk assessment of human spillover.

Data availability statement

The original contributions presented in the study are included in the article, further inquiries can be directed to the corresponding authors.

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CRediT authorship contribution statement

Xuexia Wen: Writing – original draft, Investigation. **Yaoqi Fang:** Investigation. **Feng Jiang:** Investigation. **Yixin Wang:** Data curation. **Qijun Chen:** Writing – review & editing, Funding acquisition. **Zeliang Chen:** Writing – review & editing. **Yuhan Wu:** Data curation. **Qing Xin:** Data curation. **Xiaohu Han:** Writing – original draft, Conceptualization. **Hua Deng:** Investigation.

Declaration of competing interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

References

[1] E.B. Breitschwerdt, R.G. Maggi, B.B. Chomel, Bartonellosis: an emerging infectious disease of zoonotic importance to animals and human beings, J. Vet. Emerg. Crit. Care 20 (2010) 8–30, https://doi.org/10.1111/j.1476-4431.2009.00496.x.
[2] T.S. Il'ina, A.I. Irkhin, [Bacteria of Bartonella genus: characteristics, pathogenicity factors and methods of molecular-genetic investigation], Mol Gen Mikrobiol Virusol (2004) 3–10.
[3] B. Goodman, P. Whitley-Williams, Bartonella, Pediatr. Rev. 41 (2020) 434–436, https://doi.org/10.1542/pir.2019-0198.
[4] U. Okaro, A. Addisu, B. Casanas, Bartonella species, an emerging cause of blood-culture-negative endocarditis, Clin. Microbiol. Rev. 30 (2017) 709–746, https://doi.org/10.1128/CMR.00013-17.
[5] M. Krugel, N. Krol, V.A.J. Kempf, Emerging rodent-associated Bartonella: a threat for human health? Parasit Vectors 15 (2022) 113, https://doi.org/10.1186/s13071-022-05162-5.
[6] K.E. Mullins, J. Hang, R.J. Clifford, Whole-genome analysis of bartonella ancashensis, a novel pathogen causing verruga peruana, rural ancash region, Peru, Emerg. Infect. Dis. 23 (2017) 430–438, https://doi.org/10.3201/eid2303.161476.
[7] B.A. Ellis, R.L. Regnery, L. Beati, Rats of the genus Rattus are reservoir hosts for pathogenic Bartonella species: an Old World origin for a New World disease? J. Infect. Dis. 180 (1999) 220–224, https://doi.org/10.1086/314824.
[8] E. Hardgrove, D.M. Zimmerman, M.E. von Fricken, A scoping review of rodent-borne pathogen presence, exposure, and transmission at zoological institutions, Prev. Vet. Med. 193 (2021) 105345, https://doi.org/10.1016/j.prevetmed.2021.105345.
[9] B.G. Meerburg, G.R. Singleton, A. Kijlstra, Rodent-borne diseases and their risks for public health, Crit. Rev. Microbiol. 35 (2009) 221–270, https://doi.org/10.1080/10408410902989837.
[10] P. Houpikian, P.E. Fournier, D. Raoult, Phylogenetic position of Bartonella vinsonii subsp. arupensis based on 16S rDNA and gltA gene sequences, Int. J. Syst. Evol. Microbiol. 51 (2001) 179–182, https://doi.org/10.1099/00207713-51-1-179.
[11] C. Jardine, G. Appleyard, M.Y. Kosoy, Rodent-associated Bartonella in Saskatchewan, Canada, Vector Borne Zoonotic Dis. 5 (2005) 402–409, https://doi.org/10.1089/vbz.2005.5.402.
[12] H. Kabeya, K. Inoue, Y. Izumi, Bartonella species in wild rodents and fleas from them in Japan, J. Vet. Med. Sci. 73 (2011) 1561–1567, https://doi.org/10.1292/jvms.11-0134.
[13] K. Klangthong, S. Promsthaporn, S. Leepitakrat, The distribution and diversity of bartonella species in rodents and their ectoparasites across Thailand, PLoS One 10 (2015) e0140856, https://doi.org/10.1371/journal.pone.0140856.
[14] D. Pangjai, S. Maruyama, S. Boonmar, Prevalence of zoonotic Bartonella species among rodents and shrews in Thailand, Comp. Immunol. Microbiol. Infect. Dis. 37 (2014) 109–114, https://doi.org/10.1016/j.cimid.2013.12.001.
[15] Y. Bai, M. Kosoy, A. Martin, Characterization of Bartonella strains isolated from black-tailed prairie dogs (Cynomys ludovicianus), Vector Borne Zoonotic Dis. 8 (2008) 1–5, https://doi.org/10.1089/vbz.2007.0136.
[16] N.A. Dybing, C. Jacobson, P. Irwin, Bartonella species identified in rodent and feline hosts from island and mainland western Australia, Vector Borne Zoonotic Dis. 16 (2016) 238–244, https://doi.org/10.1089/vbz.2015.1902.
[17] C. Silaghi, M. Pfeffer, D. Kiefer, Rodents Bartonella, Fleas and ticks: a molecular field study on host-vector-pathogen associations in saxony, eastern Germany, Microb. Ecol. 72 (2016) 965–974, https://doi.org/10.1007/s00248-016-0787-8.
[18] K.R. Blasdell, D. Perera, C. Firth, High prevalence of rodent-borne bartonella spp. in urbanizing environments in sarawak, Malaysian borneo, Am. J. Trop. Med. Hyg. 100 (2019) 506–509, https://doi.org/10.4269/ajtmh.18-0616.
[19] B.R. Ansil, I.H. Mendenhall, U. Ramakrishnan, High prevalence and diversity of Bartonella in small mammals from the biodiverse Western Ghats, PLoS Negl Trop Dis 15 (2021) e0009178, https://doi.org/10.1371/journal.pntd.0009178.
[20] S. Divari, M. Danelli, P. Pregel, Biomolecular investigation of bartonella spp. in wild rodents of two Swiss regions, Pathogens 10 (2021) 1331, https://doi.org/10.3390/pathogens10101331.
[21] J.P. Buffet, M. Kosoy, M. Vayssier-Taussat, Natural history of Bartonella-infecting rodents in light of new knowledge on genomics, diversity and evolution, Future Microbiol. 8 (2013) 1117–1128, https://doi.org/10.2217/fmb.13.77.
[22] D.M. Li, Q.Y. Liu, D.Z. Yu, Phylogenetic analysis of Bartonella detected in rodent fleas in Yunnan, China, J. Wildl. Dis. 43 (2007) 609–617, https://doi.org/10.7589/0090-3558-43.4.609.
[23] D.M. Li, Y. Hou, X.P. Song, High prevalence and genetic heterogeneity of rodent-borne Bartonella species on Heixiazi Island, China, Appl. Environ. Microbiol. 81 (2015) 7981–7992, https://doi.org/10.1128/AEM.02041-15.
[24] J.W. Hsieh, K.C. Tung, W.C. Chen, Epidemiology of Bartonella infection in rodents and shrews in Taiwan, Zoonoses Public Health 57 (2010) 439–446, https://doi.org/10.1111/j.1863-2378.2009.01234.x.
[25] Q. Liu, J. Sun, L. Lu, Detection of bartonella species in small mammals from Zhejiang Province, China, J. Wildl. Dis. 46 (2010) 179–185, https://doi.org/10.7589/0090-3558-46.1.179.
[26] X. Ye, G.W. Li, M.L. Yao, [Study on the prevalence and genotypes of Bartonella species in rodent hosts from Fujian coastal regions], Zhonghua Liuxingbingxue Zazhi 30 (2009) 989–992.
[27] R. Jian, Q. Ren, J. Xue, Genetic diversity of Bartonella infection in residential and field rodents in Hebei, China, Front. Microbiol. 13 (2022) 1039665, https://doi.org/10.3389/fmicb.2022.1039665.
[28] J. Yu, X.Y. Zhang, Y.X. Chen, Molecular detection and genetic characterization of small rodents associated Bartonella species in Zhongtiao Mountain, China, PLoS One 17 (2022) e0264591, https://doi.org/10.1371/journal.pone.0264591.

[29] J. Yu, B. Xie, G.Y. Bi, Prevalence and diversity of small rodent-associated Bartonella species in Shangdang Basin, China, PLoS Negl Trop Dis 16 (2022) e0010446, https://doi.org/10.1371/journal.pntd.0010446.
[30] X.Y. Yao, H. Liu, J. Sun, Epidemiology and genetic diversity of bartonella in rodents in urban areas of Guangzhou, southern China, Front. Microbiol. 13 (2022) 942587, https://doi.org/10.3389/fmicb.2022.942587.
[31] H. Hu, Z. Liu, R. Fu, Detection and phylogenetic analysis of tick-borne bacterial and protozoan pathogens in a forest province of eastern China, Acta Trop. 235 (2022) 106634, https://doi.org/10.1016/j.actatropica.2022.106634.
[32] M. Lu, G. Tang, Z. Ren, Coxiella Ehrlichia, Bartonella infections in rodents from Guizhou province, southwest China, Ticks Tick Borne Dis 13 (2022) 101974, https://doi.org/10.1016/j.ttbdis.2022.101974.
[33] L. Hao, D. Yuan, L. Guo, Molecular detection of Bartonella in ixodid ticks collected from yaks and plateau pikas (Ochotona curzoniae) in Shiqu County, China, BMC Vet. Res. 16 (2020) 235, https://doi.org/10.1186/s12917-020-02452-x.
[34] D. Simons, L.A. Attfield, K.E. Jones, Rodent trapping studies as an overlooked information source for understanding endemic and novel zoonotic spillover, PLoS Negl Trop Dis 17 (2023) e0010772, https://doi.org/10.1371/journal.pntd.0010772.
[35] G.E. Garcia-Pena, A.V. Rubio, H. Mendoza, Land-use change and rodent-borne diseases: hazards on the shared socioeconomic pathways, Philos. Trans. R. Soc. Lond. B Biol. Sci. 376 (2021) 20200362, https://doi.org/10.1098/rstb.2020.0362.
[36] M.M. Islam, E. Farag, A. Mahmoudi, Rodent-related zoonotic pathogens at the human-animal-environment interface in Qatar: a systematic review and Meta-analysis, Int. J. Environ. Res. Public Health 18 (2021) 5928, https://doi.org/10.3390/ijerph18115928.
[37] M.H. Diaz, Y. Bai, L. Malania, Development of a novel genus-specific real-time PCR assay for detection and differentiation of Bartonella species and genotypes, J. Clin. Microbiol. 50 (2012) 1645–1649, https://doi.org/10.1128/JCM.06621-11.
[38] A.F. Norman, R. Regnery, P. Jameson, Differentiation of Bartonella-like isolates at the species level by PCR-restriction fragment length polymorphism in the citrate synthase gene, J. Clin. Microbiol. 33 (1995) 1797–1803, https://doi.org/10.1128/jcm.33.7.1797-1803.1995.
[39] P. Renesto, J. Gouvernet, M. Drancourt, Use of rpoB gene analysis for detection and identification of Bartonella species, J. Clin. Microbiol. 39 (2001) 430–437, https://doi.org/10.1128/JCM.39.2.430-437.2001.
[40] H. Rao, S. Li, L. Lu, Genetic diversity of Bartonella species in small mammals in the Qaidam Basin, western China, Sci. Rep. 11 (2021) 1735, https://doi.org/10.1038/s41598-021-81508-w.
[41] J. Yu, Q. Li, L. Lu, Detection and genetic diversity of Bartonella species in small mammals from the central region of the Qinghai-Tibetan Plateau, China, Sci. Rep. 12 (2022) 6996, https://doi.org/10.1038/s41598-022-11419-x.
[42] H. Liu, T. Han, W. Liu, Epidemiological characteristics and genetic diversity of Bartonella species in rodents from southeastern China, Zoonoses Public Health 69 (2022) 224–234, https://doi.org/10.1111/zph.12912.
[43] X.R. Qin, J.W. Liu, H. Yu, Bartonella species detected in rodents from eastern China, Vector Borne Zoonotic Dis. 19 (2019) 810–814, https://doi.org/10.1089/vbz.2018.2410.
[44] C.H. An, B.B. Chen, W. Lyu, Bartonella species investigated among rodents from Shaanxi province of China, Biomed. Environ. Sci. 33 (2020) 201–205, https://doi.org/10.3967/bes2020.028.
[45] J. Li, C. Zhang, M. Lu, The diverse genetic genotypes of Bartonella species circulating in rodents from Inner Mongolia, Northern China, PLoS Negl Trop Dis 17 (2023) e0011462, https://doi.org/10.1371/journal.pntd.0011462.
[46] F.T. Kerkhoff, A.M. Bergmans, A. van Der Zee, Demonstration of Bartonella grahamii DNA in ocular fluids of a patient with neuroretinitis, J. Clin. Microbiol. 37 (1999) 4034–4038, https://doi.org/10.1128/JCM.37.12.4034-4038.1999.
[47] J. Oksi, S. Rantala, S. Kilpinen, Cat scratch disease caused by Bartonella grahamii in an immunocompromised patient, J. Clin. Microbiol. 51 (2013) 2781–2784, https://doi.org/10.1128/JCM.00910-13.
[48] D. Raoult, F. Roblot, J.M. Rolain, First isolation of Bartonella alsatica from a valve of a patient with endocarditis, J. Clin. Microbiol. 44 (2006) 278–279, https://doi.org/10.1128/JCM.44.1.278-279.2006.
[49] M. Puges, A. Menard, X. Berard, An unexpected case of Bartonella alsatica prosthetic vascular graft infection, Infect. Drug Resist. 12 (2019) 2453–2456, https://doi.org/10.2147/IDR.S206805.
[50] E. Angelakis, H. Lepidi, A. Canel, Human case of Bartonella alsatica lymphadenitis, Emerg. Infect. Dis. 14 (2008) 1951–1953, https://doi.org/10.3201/eid1412.080757.
[51] D. Jeanclaude, P. Godmer, D. Leveiller, Bartonella alsatica endocarditis in a French patient in close contact with rabbits, Clin. Microbiol. Infect. 15 (Suppl 2) (2009) 110–111, https://doi.org/10.1111/j.1469-0691.2008.02187.x.