

1 MYAGMARSUREN ET AL – SERO–EPIDEMIOLOGY OF EQUINE
2 PIROPLASMOSIS
3 **A SEROEPIDEMIOLOGICAL SURVEY OF *THEILERIA EQUI* AND *BABESIA***
4 ***CABALLI* IN HORSES IN MONGOLIA**

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21 **ABSTRACT**

22 Equine piroplasmosis caused by *Theileria equi* and *Babesia caballi* is an economically
23 important disease with a worldwide distribution. The objective of the present study was
24 to investigate the seroepidemiology of *T. equi* and *B. caballi* in horses reared in various
25 Mongolian provinces. Serum samples prepared from blood collected from horses in 19
26 Mongolian provinces were screened for antibodies specific to *T. equi* and *B. caballi*
27 using enzyme-linked immunosorbent assays based on recombinant forms of *T. equi*
28 merozoite antigen-2 and the *B. caballi* 48-kDa merozoite rhoptry protein, respectively.
29 Of 1,282 horses analyzed, 423 (33%) and 182 (14.2%) were sero-positive for *T. equi*
30 and *B. caballi*, respectively. Additionally, 518 (40.4%) were positive for at least 1
31 parasite species, of which 87 (16.8%) were co-infected with both parasites. Both *T. equi*
32 and *B. caballi* were detected in all surveyed provinces, and on a per province basis the
33 positive rates ranged from 19.0%–74.2% and 4.5%–39.8%, respectively. *Theileria equi*-
34 and *B. caballi*-positive rates were comparable between male (31.9% and 14.1%,
35 respectively) and female horses (34.5% and 14.3%, respectively). However, the positive
36 rates were higher in the >3-yr-old age group (37.7% and 15.6%, respectively) compared

37 with the 1–3-yr-old age group (19.4% and 10.0%, respectively). These findings
38 confirmed that *T. equi* and *B. caballi* infections are widespread among horses all over
39 Mongolia, and that horse age is a risk factor for infection in this country. Our results
40 will be useful for designing appropriate control measures to minimize *T. equi* and *B.*
41 *caballi* infections among Mongolian horses.

42

43 **KEY WORDS**

44 *Babesia caballi*, ELISA, Epidemiological Mapping, Equine Piroplasmosis, Horses,
45 Mongolia, *Theileria equi*

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47 Equine piroplasmosis is an acute, subacute, or chronic tick-borne disease of the
48 Equidae (horses, donkeys, mules, and zebras) caused by hemoprotozoan parasites
49 *Theileria equi* and *Babesia caballi* (Bruning, 1996). It is generally characterized by
50 fever, anemia, jaundice, edema, and, in some cases, death (Schein, 1988; de Waal,
51 1992; Bruning, 1996). Although both *T. equi* and *B. caballi* cause clinical disease, the
52 former is associated with more severe disease (Camacho et al., 2005).

53 Equine piroplasmosis is common in tropical and subtropical regions, including
54 parts of Africa, the Middle East, Asia, Central and South America, the Caribbean, and

55 Europe, with 27 countries reporting disease incidence according to the World
56 Organisation for Animal Health (OIE, 2017). Once infected with *T. equi* and *B. caballi*,
57 horses become persistent carriers for a long period (Knowles et al., 1997). Detection of
58 these carrier animals is vital because they can be a source of infection for transmission
59 to uninfected horses. Therefore, epidemiological surveys have been conducted in
60 several endemic countries to identify such chronically infected carrier animals (Butler et
61 al., 2012; Oduori et al., 2015; Sumbria et al., 2016; Guven et al., 2017; Díaz-Sánchez et
62 al., 2018).

63 Mongolia is an agricultural country in which the livestock sector is key to its
64 economic growth. The horse population in Mongolia was estimated to be about 3.9
65 million head in 2017 (National Statistics Office of Mongolia, 2017). However, the
66 productivity of Mongolian horses has been severely undermined for various reasons,
67 including the presence of infectious diseases (Odontsetseg et al., 2005; Pagamjav et al.,
68 2011; Sukanuma et al., 2017). This also limits the export market for Mongolian horses
69 and their products (World Bank., 2009).

70 The causative agents of equine piroplasmiasis, *T. equi* and *B. caballi*, have been
71 reported in Mongolia using microscopy (Rüegg et al., 2007), the indirect fluorescence
72 antibody test (IFAT) (Avarzed et al., 1997, Rüegg et al., 2007), enzyme-linked

73 immunosorbent assays (ELISAs) (Ikadai et al., 2000; Boldbaatar et al., 2005;
74 Munkhjargal et al., 2013), and PCR assays (Rüegg et al., 2007; Sloboda et al., 2011;
75 Munkhjargal et al., 2013). However, these studies were limited to only a few provinces
76 and no countrywide survey has been conducted, despite the fact that such investigations
77 will equip the veterinary authorities in Mongolia with the means of designing and
78 implementing a risk-based control strategy. In the present study, therefore, we prepared
79 serum samples from blood collected from horses in 19 of 21 Mongolian provinces, and
80 screened for *T. equi*- and *B. caballi*-specific antibodies using parasite-specific ELISAs.

81 **MATERIALS AND METHODS**

82 **Serum samples**

83 Blood samples were collected from a total of 1,282 horses in 19 of 21
84 Mongolian provinces in 2013–2017 (Table I). Sampling was not carried out in Orkhon
85 and Darkhan-Uul provinces, as the livestock farming is uncommon in these 2 urban
86 areas. Approximately 2 ml of blood was collected from each animal into a vacutainer
87 tube, without anticoagulant (Zhejiang Gongdong Medical Technology Co. Ltd., Taizhou,
88 Zhejiang, China). All sampled animals were apparently healthy during sampling. Serum
89 samples were prepared from blood and stored at –20 C until use. All animal procedures
90 were approved by the Animal Care and Use Committee of Obihiro University of

91 Agriculture and Veterinary Medicine, Japan (approval number: 29-1).

92 **ELISAs**

93 All serum samples were screened for *T. equi*- and *B. caballi*-specific antibodies
94 using previously described ELISAs (Huang et al., 2003; Ikadai et al., 1999). Briefly, the
95 truncated form of *T. equi* merozoite antigen-2 (EMA-2t) and the 48-kDa merozoite
96 rhoptry protein (BC48) of *B. caballi* were expressed as GST-fusion proteins
97 (rGST-EMA-2t and rGST-BC48, respectively) in *Escherichia coli*, then purified as
98 described by Huang et al. (2003) and Ikadai et al. (1999), respectively. Each well in
99 96-well ELISA microplates was coated with 5 µg/ml of rGST-EMA-2t, rGST-BC48, or
100 rGST antigen diluted in a carbonate–bicarbonate buffer, then incubated at 4 C overnight.
101 After discarding unabsorbed antigens, each well was blocked with 100 µl of 3%
102 skimmed milk in phosphate-buffered saline (PBS) with 0.05% Tween 20 (blocking
103 solution) at room temperature for 1 hr. After washing once with 200 µl of PBS
104 containing 0.05% Tween 20 (wash buffer), 100 µl of horse serum sample diluted 1:200
105 in blocking solution was added to each well. The ELISA plates were incubated at 37 C
106 for 1 hr, then each well was washed 6 times with wash buffer. Next, 100 µl of
107 horseradish peroxidase-conjugated goat anti-horse immunoglobulin G (Sigma-Aldrich,
108 St. Louis, Missouri) diluted 1:5,000 in blocking solution was added to each well, and

109 the plates were incubated at 37 C for 1 hr. After washing 6 times with wash buffer, 50
110 μ l of tetramethylbenzidine (TMB) substrate solution (Sigma-Aldrich) was added to each
111 well, then incubated at 37 C for 30 min. After adding 50 μ l of TMB stop solution
112 (Sigma-Aldrich), the optical density (OD) value was measured at 450 nm.

113 For each serum sample, the net OD value was calculated by subtracting the OD
114 value of the rGST-coated well from that of rGST-EMA2t- or rGST-BC48-coated wells.
115 A sample was considered to be positive for the *T. equi* or *B. caballi* antibody if the net
116 OD value was higher than the mean OD value + 3 standard deviations of 10 negative
117 serum samples.

118 **Statistical analyses**

119 The 95% confidence intervals for positive rates were calculated using an
120 OpenEpi software program (<http://www.openepi.com/Proportion/Proportion.htm>) based
121 on the Wilson score interval (Wilson, 1927). *P* values to compare positive rates between
122 female and male horses, as well as between 1–3-yr-old and >3-yr-old age groups, were
123 calculated using an “N-1” chi-squared test
124 (https://www.medcalc.org/calc/comparison_of_proportions.php) (Campbell, 2007;
125 Richardson, 2011). A *P* value < 0.05 was considered to indicate a significant difference
126 between the positive rates.

127 **RESULTS**

128 We screened a total of 1,282 horses in 19 Mongolian provinces for the presence
129 of antibodies to *T. equi* and *B. caballi* using ELISAs (Table I). Among them, 423
130 (33.0%) and 182 (14.2%) were positive for *T. equi* and *B. caballi*, respectively. In
131 addition, 518 (40.4%) horses were positive for at least 1 parasite species, and 87
132 (16.8%) among them had co-infection with *T. equi* and *B. caballi*. The overall positive
133 rate of *T. equi* was significantly higher than that of *B. caballi* ($P < 0.0001$). On a per
134 province basis, the positive rates of *T. equi* and *B. caballi* ranged from 19.0%–74.2%
135 and 4.5%–39.8%, respectively. In each province, except for Dundgovi, the positive rate
136 of *T. equi* was higher than that of *B. caballi* (Table I). Although the reason for the
137 higher rate of *B. caballi* positivity compared with that of *T. equi* in Dundgovi is not very
138 clear, the finding may not be conclusive as the sample size was relatively small.

139 Geographically, high rates of *T. equi* positivity (>25%) were observed in all
140 provinces in central (Arkhangai, Tov, and Ovorkhangai) and Govi (Bayankhongor,
141 Dundgovi, Govisumber, Omnogovi, and Dornogovi) regions, as well as in Bayan-Ulgii
142 in the western region, Bulgan and Selenge in the northern region, and Dornod and
143 Sukhbaatar in the eastern region (Fig. 1). The geographical variation of *B.*
144 *caballi*-positive rates was comparable with that of *T. equi* (Fig. 1). All provinces with

145 high rates of *T. equi* positivity (>25%) also had relatively high rates of *B. caballi*
146 positivity (>10%). However, *B. caballi*-positive rate in Govisumber was <10%,
147 compared with >25% *T. equi* positivity, while in Khovsgol, *B. caballi*-positive rate was
148 >10%, compared with <25% *T. equi* positivity (Fig. 1).

149 We next compared the positive rates between male and female horses, and
150 between 1–3-yr-old and >3-yr-old age groups. Between males and females, the overall
151 positive rates of both *T. equi* (31.9% and 34.5%) and *B. caballi* (14.1% and 14.3%,
152 respectively) were comparable (Table II). In addition, positive rates of these parasite
153 species in each surveyed province were also comparable between males and females.
154 On the other hand, overall positive rates of *T. equi* and *B. caballi* were significantly
155 higher ($P < 0.0001$ and 0.012, respectively) in the >3-yr-old age group (37.7% and
156 15.6%, respectively) compared with those in the 1–3-yr-old age group (19.4% and
157 10.0%, respectively) (Table III).

158 **DISCUSSION**

159 Seroepidemiological surveys of *T. equi* and *B. caballi* are very important to
160 estimate the risk of infections in horses in endemic countries. The objective of the
161 present study was to investigate the seroepidemiology of *T. equi* and *B. caballi*
162 infections in horses reared throughout Mongolia. ELISA based on BC48 is widely used

163 for sero-diagnosis of *B. caballi*, while EMA-1 and EMA-2 are the 2 most commonly
164 used antigens in *T. equi*-specific ELISA (Salim et al., 2008; Munkhjargal et al., 2013;
165 Rosales et al., 2013). However, a previous study found that EMA-2-based ELISA could
166 detect *T. equi*-specific antibodies in infected horses 6-12 days earlier compared with
167 EMA-1 ELISA (Huang et al., 2003). In the present study, therefore, BC48- and
168 EMA-2-based ELISAs were employed for the sero-survey of *B. caballi* and *T. equi*,
169 respectively. Our findings demonstrated that horses in all of the surveyed provinces had
170 been exposed to both *T. equi* and *B. caballi*. The overall positive rate of *T. equi*
171 infection was significantly higher than that of *B. caballi*. This observation is an
172 agreement with the findings from previous studies conducted in Mongolia (Boldbaatar
173 et al., 2005; Munkhjargal et al., 2013).

174 The virulence of *T. equi* is known to be higher than that of *B. caballi* (Camacho
175 et al., 2005). Therefore, the high rate of *T. equi* positivity suggests that horse
176 populations throughout Mongolia are at risk of clinical piroplasmiasis. Indeed, a recent
177 study found evidence to suggest *T. equi* is the causative agent of severe equine
178 piroplasmiasis among Mongolian wild horses (Przewalski's horse) (Tarav et al., 2017).
179 Possible reasons for observed differential positive rates of *T. equi* and *B. caballi* include
180 differences in the density of infected tick vectors and waning of immunity following

181 parasite clearance. A previous study identified *Dermacentor nuttalli*, the most abundant
182 tick species in Mongolia, as a vector of both *T. equi* and *B. caballi* in Mongolia
183 (Battsetseg et al., 2001). Notably, however, the rate of *T. equi*-infected *D. nuttalli* was
184 higher than that of *B. caballi*-infected ticks (Battsetseg et al., 2001). Compared with *B.*
185 *caballi*, the *T. equi* infection usually persists in the host for longer, probably throughout
186 its life, acting as a source of infection for ticks vectors (Zweygarth et al., 1996;
187 Mehlhorn and Schein, 1998). Moreover, complete elimination of *T. equi* from such
188 chronically infected horses is extremely difficult (Friedhoff and Soule, 1996). These
189 observations could explain why the *T. equi*-positive rate was higher than the *B.*
190 *caballi*-positive rate in the present study.

191 We also observed differences in the positive rates of *T. equi* and *B. caballi*
192 infections among Mongolian provinces. Differences in the density of *D. nuttalli* in
193 different Mongolian regions might explain these geographical variations; however, the
194 geographical distribution of *D. nuttalli* in Mongolia is not completely understood.
195 Therefore, future studies should focus on analyzing the relative abundance of *D. nuttalli*
196 in various provinces of this country. As well as *D. nuttalli*, several other species of ticks
197 are known to exist in Mongolia (Tuvshintulga et al., 2015; Boldbaatar et al., 2017).
198 Thus, the investigation of *T. equi* and *B. caballi* infections in other tick species could

199 help understand the epidemiology of these parasite species in Mongolian horses.

200 Our findings also showed that the positive rates of both *T. equi* and *B. caballi*
201 infections were comparable between male and female horses. In Mongolia, both horse
202 sexes are reared together under the same management system, which may explain the
203 comparable positive rates of infection (Munkhjargal et al., 2013). In contrast, *T. equi*-
204 and *B. caballi*-positive rates of infection were higher in older horses compared with
205 younger animals. This is likely to reflect the greater chance of being exposed to infected
206 ticks with increasing horse age (Rüegg et al., 2007).

207 A limitation of the present study is relatively small sample size, which was not
208 defined statistically, compared with the horse population in each province. Previous
209 studies found potential strain variations among *T. equi* and *B. caballi* isolates (Bhoora,
210 et al., 2009; Munkhjargal et al., 2013). The genetic variations were also observed
211 among gene sequences encoding EMAs in *T. equi* and BC48 in *B. caballi* (Bhoora, et al.,
212 2010a, 2010b). However, impact of such genetic variations on our ELISA results was
213 not considered, and this is also a limitation of the present study.

214 In summary, the present study found that horses bred throughout Mongolia
215 were exposed to both *T. equi* and *B. caballi* infections. We also found that the positive
216 rates of both *T. equi* and *B. caballi* varied among the surveyed provinces. The present

217 findings must be useful for designing a risk-based control strategy with the objective of
218 minimizing *T. equi* and *B. caballi* infections in horses in Mongolia.

219 **ACKNOWLEDGMENTS**

220 The authors assert all applicable international, national, and/or institutional
221 guidelines for the care and use of animals were followed. We thank the owners and staff
222 of the horse farms involved in the present study. We also thank the local veterinarians
223 for their kind support in sampling. This study was supported by a grant from the Japan
224 Agency for Medical Research and Development in collaboration with Japan
225 International Collaboration Agency (AMED/JICA) Science and Technology Research
226 Partnership for Sustainable Development (SATREPS) project (grant number
227 17jm0110006h0005). All authors declare no conflicts of interest associated with the
228 present study.

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357 **FIGURE 1.** Epidemiological mapping of *Theileria equi* and *Babesia caballi*.
358 Epidemiological maps were prepared to illustrate geographical variations of the
359 seropositive rates of (A) *T. equi* and (B) *B. caballi* infections among Mongolian horses,
360 using ArcGIS v10.1 software program (Environmental Systems Research Institute,

361 Redlands, California). The differential prevalence rates are indicated by different colors.

362

Table I. Positive rates of *Theileria equi* and *Babesia caballi* infections in horses in 19 Mongolian provinces.

Province	No. samples	<i>T. equi</i>		<i>B. caballi</i>		Co-infection [†]	
		No. positive	% (CI*)	No. positive	% (CI)	No. positive	% (CI)
Arkhangai	54	20	37.0 (25.4-50.4)	7	13.0 (6.4-24.4)	5	22.7 (10.1-43.4)
Bayankhongor	57	18	33.3 (21.0-44.5)	7	12.3 (6.1-23.3)	4	19.0 (7.7-40.0)
Bayan-Ulgii	105	45	43.0 (33.8-52.4)	17	16.2 (10.4-24.4)	10	19.2 (10.8-31.9)
Bulgan	20	6	30.0 (14.5-51.9)	2	10.0 (2.8-30.1)	0	0.0 (0.0-32.4)
Dornod	93	69	74.2 (64.5-82.0)	37	39.8 (30.4-49.9)	30	39.5 (29.3-50.7)
Dornogovi	57	16	28.1 (18.1-40.8)	9	16.0 (8.5-27.4)	4	19.0 (7.7-40.0)
Dundgovi	33	10	30.3 (17.4-47.3)	12	36.3 (22.2-53.4)	4	22.2 (9.0-45.2)
Govi-Altai	29	17	59.0 (40.7- 74.5)	7	24.1 (12.2-42.1)	4	20.0 (8.1-41.6)
Govisumber	25	7	28.0 (14.3-47.6)	2	8.0 (2.2- 24.9)	2	28.6 (8.2-64.1)
Khentii	98	24	24.5 (17.1-33.9)	7	7.1 (3.5-14.02)	1	3.3 (0.6-16.7)
Khovd	110	21	19.0 (12.8-27.4)	5	4.5 (1.9-10.2)	3	13.0 (4.5-32.1)
Khovsgol	62	15	24.1 (15.2-36.1)	9	14.5 (7.8-25.3)	3	14.3 (5.0-34.6)
Omnogovi	57	20	35.0 (24.0-48.1)	12	21.0 (12.5-33.3)	6	23.1 (11.0-42.1)
Ovorkhangai	26	11	42.3 (25.6-61.1)	3	11.5 (4.0-28.9)	0	0.0 (0.0-21.5)
Selenge	64	13	26.5 (12.3-31.7)	9	14.0 (7.6-24.6)	1	4.8 (0.8-22.7)
Sukhbaatar	136	42	31.0 (23.8-39.1)	15	11.0 (6.8-17.4)	4	7.5 (3.0-17.9)
Tov	48	23	48.0 (34.5-61.7)	6	12.5 (5.8-24.7)	3	11.5 (4.0-29.0)
Uvs	75	17	23.0 (14.7-33.3)	5	6.6 (2.9-14.7)	2	10.0 (2.8-30.1)
Zavkhan	133	29	22.0 (15.6- 29.6)	11	8.2 (4.7-14.2)	1	2.6 (0.5-13.2)
Total	1,282	423	33.0 (30.5-35.6)	182	14.2 (12.4-16.2)	87	16.8 (13.8-20.3)

* 95% confidence interval

[†] Expressed as a percentage of the number of animals infected with at least one parasite species (No. *T. equi*-positive + No. *B. caballi*-positive – No. co-infected).

Table II. Positive rates of *Theileria equi* and *Babesia caballi* infection in male and female horses in 19 Mongolian provinces.

Province	No. Samples		<i>T. equi</i>					<i>B. caballi</i>				
	Male	Female	Male		Female		<i>P</i> value	Male		Female		<i>P</i> value
			No. positive	% (CI*)	No. positive	% (CI)		No. positive	% (CI)	No. positive	% (CI)	
Arkhangai	30	24	10	33.3 (19.2-51.2)	10	41.7 (24.5-61.2)	0.5292	4	13.3 (5.3-29.7)	3	12.5 (4.3- 31.0)	0.9313
Bayankhongor	28	29	10	36.0 (20.7-54.2)	8	27.6 (14.7-45.7)	0.4996	5	17.8 (7.9-35.6)	2	6.9 (1.9- 21.9)	0.2137
Bayan- Ulgii	40	65	13	32.5 (20.1-47.9)	32	49.2 (37.5-61.1)	0.0947	7	17.5 (8.7-31.9)	10	15.4 (8.6- 26.0)	0.7777
Bulgan	12	8	5	42.0 (19.3-68.0)	1	12.5 (2.2-47.1)	0.1700	1	8.3 (1.5-35.4)	1	12.5 (2.2-47.1)	0.7648
Dornod	65	28	49	75.4 (63.7-84.2)	20	71.4 (52.9-84.7)	0.6875	24	36.9 (26.2-49.1)	13	46.4 (29.5-64.2)	0.3931
Dornogovi	32	25	4	12.5 (4.9-28.1)	12	48.0 (30.0-66.5)	0.0033	5	15.6 (6.9-31.7)	4	16.0 (6.4-34.6)	0.9675
Dundgovi	4	29	2	50.0 (15.0-85.0)	8	27.6 (14.7-45.7)	0.3682	2	50.0 (15.0-85.0)	10	34.5 (19.9-52.6)	0.5520
Govi- Altai	22	7	14	63.6 (42.9-80.3)	3	42.8 (15.8-74.9)	0.3390	5	22.7 (10.1-43.4)	2	28.6 (8.2- 64.1)	0.7548
Govisumber	17	8	3	17.6 (6.2-41.0)	4	50.0 (21.5-78.5)	0.0990	1	5.9 (1.0-26.9)	1	12.5 (2.2-47.1)	0.5785
Khentii	55	43	12	21.8 (12.9-34.4)	12	27.9 (16.7-42.7)	0.4881	5	9.1 (3.9-19.6)	2	4.6 (1.3-15.4)	0.3926
Khovd	79	31	13	16.4 (9.9-26.1)	8	25.8 (13.7-43.2)	0.2609	5	6.3 (2.7-13.9)	0	0.0 (0-11.0)	0.1545
Khovsgol	35	27	7	20.0 (10.0-35.9)	8	29.6 (15.8-48.5)	0.3853	5	14.3 (6.3-29.4)	4	14.8 (5.9-32.5)	0.9562
Omnogovi	47	10	17	36.2 (12.1-64.6)	3	30.0 (10.8-60.3)	0.7116	8	17.0 (8.9-30.1)	4	40.0 (16.8-68.7)	0.1086
Ovorkhangai	9	17	3	33.3 (23.9-50.5)	8	47.0 (26.2-69.0)	0.5094	1	11.1 (1.9-43.5)	2	11.8 (3.3-34.3)	0.9685
Selenge	40	24	8	20.0 (10.5-34.8)	5	20.8 (9.2-40.5)	0.9370	5	12.5 (5.4- 26.1)	4	16.7 (6.7-35.9)	0.6426
Sukhbaatar	78	58	20	25.6 (17.3-36.3)	22	37.9 (26.6-50.8)	0.1259	10	12.8 (7.1-22.0)	5	8.6 (3.7-18.6)	0.4407
Tov	9	39	7	77.8 (45.3-93.7)	16	41.0 (27.1-56.6)	0.0487	1	11.1 (1.9-43.5)	5	12.8 (5.6-26.7)	0.8857
Uvs	46	29	12	26.1 (15.6-40.3)	5	17.2 (7.6-34.5)	0.3731	2	4.3 (1.2-14.5)	3	10.3 (3.6-26.4)	0.3120
Zavkhan	66	67	18	27.3 (18.0-39.0)	11	16.4 (9.4-27.1)	0.1295	5	7.6 (3.3-16.5)	6	8.9 (4.2-18.2)	0.7861
Total	714	568	228	31.9 (28.6-35.4)	196	34.5 (30.7-38.5)	0.3258	101	14.1 (11.8-16.9)	81	14.3 (11.6- 17.4)	0.9188

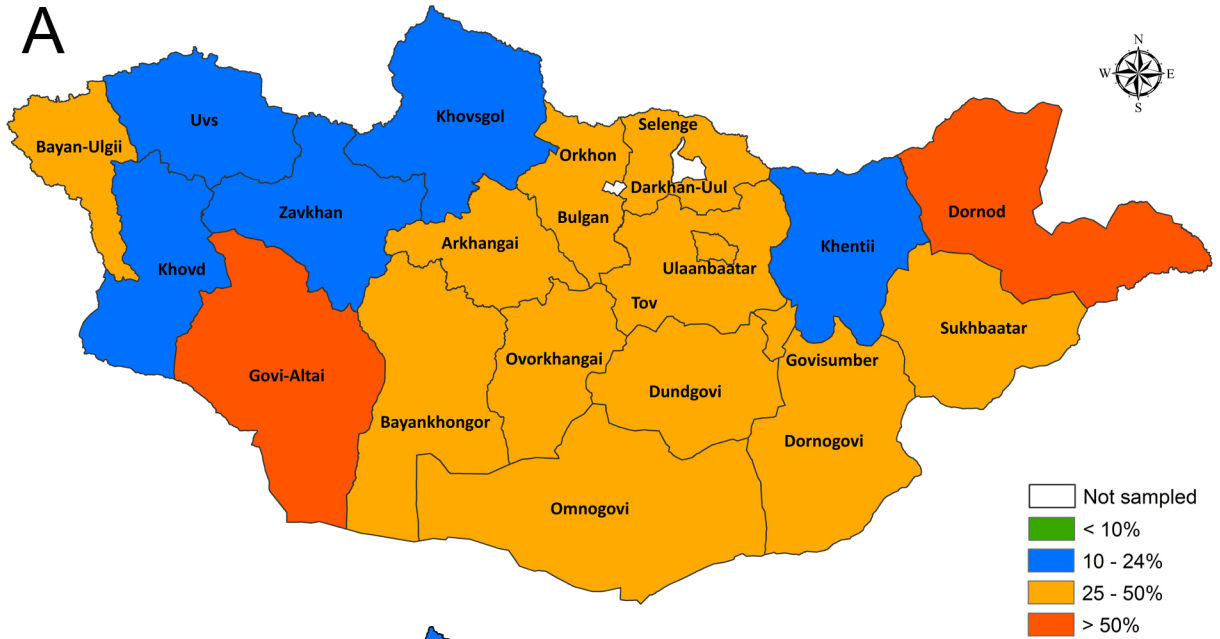
* 95% confidence interval

Table III. Positive rates of *Theileria equi* and *Babesia caballi* infection in 1–3-year-old and >3-year-old horse age groups in 19 Mongolian provinces.

Province	No. Samples		<i>T. equi</i>				<i>B. caballi</i>					
	1-3 years	>3 years	1-3 years		>3 years		<i>P</i> value	1-3 years		>3 years		<i>P</i> value
			No. positive	% (CI*)	No. positive	% (CI)		No. positive	% (CI)	No. positive	% (CI)	
Arkhangai	15	39	10	66.7 (41.7-84.8)	10	25.6 (14.6-41.1)	0.0055	4	26.7 (10.9-51.9)	3	7.7 (2.6-20.3)	0.0652
Bayankhongor	11	46	0	0.0 (0-25.9)	18	39.1 (26.4-53.5)	0.0130	0	0.0 (0-25.9)	7	15.2 (7.6-28.2)	0.1712
Bayan- Ulgii	17	88	2	11.8 (3.3-34.3)	43	48.9 (38.7-59.1)	0.0049	1	5.9 (1.0-26.9)	16	18.2 (11.5-27.5)	0.2099
Bulgan	11	9	4	36.4 (15.2-64.6)	2	22.2 (6.3-54.7)	0.5017	1	9.1 (1.6-37.7)	1	11.1 (1.9-43.5)	0.8851
Dornod	24	69	18	75.0 (55.1-88.0)	51	73.9 (62.5-82.8)	0.9160	10	41.7 (24.5-59.3)	27	39.1 (28.5-50.9)	0.8236
Dornogovi	18	39	3	16.7 (5.8-39.2)	13	33.3 (20.6-49.0)	0.1987	0	0.0 (0-17.6)	9	23.1 (12.6-38.3)	0.0276
Dundgovi	6	27	2	33.3 (9.7-70.0)	8	29.6 (15.8-48.5)	0.8605	1	16.7 (3.0-56.3)	11	40.7 (24.5-59.3)	0.2763
Govi- Altai	10	19	3	30.0 (10.8-60.3)	14	73.7 (51.2-88.2)	0.0256	0	0.0 (0-27.8)	7	36.8 (19.1-58.9)	0.0305
Govisumber	10	15	0	0.0 (0-27.8)	7	46.7 (24.8-69.9)	0.0126	0	0.0 (0-27.8)	2	13.3 (3.7-37.9)	0.2388
Khentii	11	87	2	18.2 (5.1-47.7)	22	25.3 (17.3-35.3)	0.6078	0	0.0 (0-25.9)	7	8.0 (3.9-15.7)	0.2388
Khovd	23	87	1	4.3 (0.8-20.9)	20	22.9 (15.4-32.9)	0.0442	0	0.0 (0-14.3)	5	5.7 (2.5-12.8)	0.2435
Khovsgol	13	49	2	15.4 (4.3-42.2)	13	26.5 (16.2-40.3)	0.5593	5	38.5 (17.7-64.5)	4	8.2 (4.4-21.8)	0.0063
Omnogovi	22	35	3	13.6 (4.7-33.3)	17	48.6 (33.0-64.4)	0.0075	3	13.6 (4.7-33.3)	9	25.7 (13.4-40.1)	0.2794
Ovorkhangai	7	19	1	14.3 (2.6-51.3)	10	52.6 (31.7-72.7)	0.0855	0	0.0 (0-35.4)	3	15.8 (5.5- 37.6)	0.2729
Selenge	20	44	2	10.0 (2.8-30.1)	11	25.0 (14.6-39.4)	0.1702	3	15.0 (5.2-36.0)	6	13.6 (6.4-26.7)	0.8821
Sukhbaatar	42	94	5	11.9 (5.2-25.0)	37	39.4 (30.1-49.5)	0.0014	1	2.4 (0.4-12.3)	14	14.9 (9.1-23.5)	0.0323
Tov	11	37	2	18.2 (5.1-47.7)	21	56.7 (40.9-71.3)	0.0264	1	9.1 (1.6-37.7)	5	13.5 (5.9-27.9)	0.7014
Uvs	26	49	2	7.7 (2.1-24.1)	15	30.6 (19.5-44.5)	0.0251	2	7.7 (2.1-24.1)	3	6.1 (2.1-16.5)	0.7927
Zavkhan	32	101	2	6.2 (1.7-20.1)	27	26.7 (19.1-36.1)	0.0147	1	3.1 (0.5-15.7)	10	9.9 (5.5-17.3)	0.2252
Total	329	953	64	19.4 (15.5-24.0)	359	37.7 (34.6-40.8)	<0.0001	33	10.0 (7.2-13.7)	149	15.6 (13.5-18.1)	0.0120

* 95% confidence interval

A



B

