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Study of the prevalence of bovine babesiosis with Babesia bovis and Babesia bigemina isolated in the livestock of Bamako and its peri-urban area between 2018 and 2023

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Abstract
Bovine babesiosis is one of the most widespread tick-borne diseases in Mali. The present study was carried out on cattle farms in the peri-urban area of Bamako District. Its aim was to determine the prevalence of Babesia bovis and Babesia bigemina bovine babesiosis during the period from January 2018 to November 2023. 555 blood samples were taken from the jugular vein of cattle from 33 localities spread over five (05) axes. These were the Bamako Siby axis, the Bamako- Kati axis, the Bamako- Koulikoro axis, the Bamako- Ségou axis and the Bamako- Sikasso axis. The samples collected were analyzed in the laboratory using a diagnostic technique based on the examination of GIEMSA-stained blood smears. This technique yielded a prevalence rate of 15.86% for bovine babesiosis. Statistical analysis revealed a significant difference between the five axes ($p<0.05$) for Babesia bigemina, while there was no significant difference between the axes ($p>0.05$) for the prevalence rate of Babesia bovis. With regard to the gender-related risk factor, there was no significant difference between male and female cattle in the prevalence rates of Babesia bovis and Babesia bigemina. The prevalence rates of Babesia bovis and Babesia bigemina were significantly higher in older cattle than in younger cattle ($p<0; 05$). The present study revealed that bovine babesiosis is on the increase in the District of Bamako.

Keywords: Tick, bovine babesiosis, prevalence, Mali

1. Introduction
Mali is one of the major cattle-producing countries in West Africa [4]. Peri-urban livestock farming plays a key role in meeting the needs of urban populations for meat, dairy and poultry products [15]. Bovine babesiosis is considered to be the most important parasite of livestock, due to the losses it causes to cattle [12].
Serological studies carried out in the District of Bamako and in the Sikasso region have revealed that cattle breeding is facing serious problems due to ticks and tick-borne diseases [14, 15]. This led us to carry out the present study to improve knowledge of the situation of Babesia bovis and Babesia bigemina babesiosis in the District of Bamako and in the peri-urban area.

2. Materials and Methods
The study took place in the District of Bamako and in the District's peri-urban zone.
The research team travelled at the request of the breeders at the sites visited. For this reason, there was no specific choice of sites during the present study. In addition, the survey sites were spread over five axes: the Bamako Siby axis, the Bamako- Kati axis, the Bamako- Koulikoro axis, the Bamako- Ségou axis and the Bamako- Sikasso axis. 555 blood samples were taken from cattle from 33 localities. Blood was drawn from the jugular vein of cattle using a sampling needle in vacuum Vaccutainer tubes containing EDTA anticoagulant. Each tube was marked with the site code, animal identification number and date of collection. The tubes containing the collected blood were placed in a rack and then stored in a cooler containing ice chips, which was sent to the Laboratoire Central Vétérinaire in Bamako.

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In Au, smears were prepared on glass slides with a margin at one end. Smears were fixed in methanol solution for 5 minutes, then stained in Rapid GIEMSA solution for 5 minutes. They were then washed under a stream of tap water and dried. After receiving 2 or 3 drops of immersion oil, the dried smears were placed under the X100 objective of a binocular electric microscope to test for Babesia bovis and Babesia bigemina in red blood cells. Raw data were entered using Microsoft EXCEL 2010 and analyzed using Stata version 12.1. The "chi 2" test was used to compare the different variables.

3. Results
3.1. Prevalence of Babesia bovis by Axis
88 cattle were positive for Babesia bovis out of a total of 555 examined on all the Axes, giving an overall prevalence rate of 15.86%. In descending order of prevalence, the Axis rates were as follows: 20.69% for the Bamako-Sikasso axis, 18.82% for the Bamako-Kati axis, 15.87% for the Bamako-Siby axis, 15.33 for the Bamako-Koulikoro axis and 12.50 for the Bamako-Segou axis (Table 1). Statistical analysis revealed no significant difference between axes (P> 0.05).

Table 1: B. bovis prevalence rates by Axis

Axis	Negative	Positive	Total	prevalence (%)
Kati	69	16	85	18,82
Koulikoro	127	23	150	15,33
Ségou	119	17	136	12,50
Siby	106	20	126	15,87
Sikasso	46	12	58	20,69
Total	467	88	555	15,86

Pearson chi2(4) = 2.7556 Pr = 0.600

3.2. Prevalence of Babesia bigemina by Axis
269 cattle were positive for Babesia bigemina out of 555 examined on the five axes of the study, giving an overall prevalence rate of 48.47%. The prevalence rates recorded on the axes were, in descending order, 60.34% for the Bamako-Sikasso axis, 58.73% for the Bamako-Siby axis, 56.62% for the Bamako-Ségou axis, 37.65% for the Bamako-Kati axis and 34.00% for the Bamako-Koulikoro axis (Table 2). Statistical analysis revealed a significant difference between the five axes, ($P < 0.05$).

Table 2: B. bigemina prevalence rates by Axis

Axis	Negative	Positive	Total	prevalence (%)
Kati	53	32	85	37,65
Koulikoro	99	51	150	34,00
Ségou	59	77	136	56,62
Siby	52	74	126	58,73
Sikasso	23	35	58	60,34
Total	286	269	555	48,47

Pearson chi2(4) = 28.7609 Pr = 0.000

Table 3: B.bovis prevalence rates by sex

B. bovis / Sex	Ngative	Positive	Total	prevalence (%)
Female	369	63	432	14,58
Male	98	25	123	20,33
Total	467	88	555	15,86

Pearson chi2(1) = 2.3659 Pr = 0.124

3.3. Babesia bovis prevalence rates by sex
The survey revealed the presence of 432 females versus 123 males out of a total of 555 cattle examined. The B. bovis prevalence rate was 20.33% in male cattle, versus 14.58% in females. There was no significant difference between male and female cattle by the chi2 test, ($p > 0.05$) (Table 3).

3.4. Prevalence rate of Babesia bigemina by sex
The prevalence rate of B. bigemina was 50.46% in female cattle, compared with 41.46% in males. There was no significant difference between male and female cattle by chi2 test, ($p > 0.05$) (Table 4).

Table 4: B. bigemina prevalence rates by sex

B. bigemina/Sex	Negative	Positive	Total	Prevalence (%)
Femelle	214	218	432	50,46
Mâle	72	51	123	41,46
Total	286	269	555	48,47

Pearson chi2(1) = 3.1046 Pr = 0.078

3.5. Babesia bovis prevalence rates by age group
The prevalences of B. bovis observed in the different age groups are, in decreasing order of: 29.58% in Bullocks, 19.00% in Heifers, 14.74% in Cows, 8.00% in Bulls and Steers, 7.41% in Calves and 6.03% in Calves. Analysis of these results showed a significant difference between the different age groups (p<0.05), (Table 5).

Table 5: B. bovis prevalence rates by age category

Category	Negative	Positive	Total	prevalence (%)
Genisse	179	42	221	19,00
Taureau_Boeuf	23	2	25	8,00
Taurillon	50	21	71	29,58
Vache	81	14	95	14,74
Veau	25	2	27	7,41
Velle	109	7	116	6,03
Total	467	88	555	15,86

Pearson chi2(5) = 22.7386 Pr = 0.000

3.6. Babesia bigemina prevalence rates by age category
Statistical analysis of B. bigemina prevalence in different age groups revealed a significant difference between these groups, ($p < 0.05$). The prevalence rates observed in these age categories are, in descending order, as follows: 68.00% in Bulls and Steers, 65.26% in Cows, 49.32% in Heifers, 42.25% in Bullocks, 40.52% in Calves and 14.81% in Calves. Analysis of these results showed a significant difference between the different age groups (p<0.05), (Table 6).

Table 6: B.bigemina positivity rates by age category

Category	Negative	Positive	Total	prevalence (%)
Genisse	112	109	221	49,32
Taureau_Boeuf	8	17	25	68,00
Taurillon	41	30	71	42,25
Vache	33	62	95	65,26
Veau	23	4	27	14,81
Velle	69	47	116	40,52
Total	286	269	555	48,47

Pearson chi2(5) = 30.8886 Pr = 0.000

4. Discussion
The overall prevalence rate of Babesia bovis on all Axes was 15.86%. Our results are similar to those obtained by Farougou et al [5]; Djakaridia et al [3]. These authors obtained Babesia bovis prevalence rates ranging from 13.16% to 59.33% in West Africa. Our results are lower than those of YEO et al [19], who recorded a Babesia bovis prevalence of 45.83%. Different results were obtained by Wodajnew et al [18], Haben et al [8] and Namomsa et al [16] with Babesia bovis prevalence

rates of 1.24%, 6.17% and 3.91% respectively.

The overall prevalence rate of Babesia bigemina over the five axes of the study was 48.47%. This result is comparable to that obtained by Farougou *et al*., 2007, who recorded a 57% prevalence of Babesia bigemina. Our results are superior to those of YEO *et al.* [18]. who recorded a prevalence of 13.61% for Babesia bigemina. Wodajnew *et al.* [18], also obtained lower prevalence rates of 0.248%, 15.53% and 1.30% respectively.

The prevalence of Babesia bovis was significantly higher in male cattle (20.33%) than in females (14.58%), while the prevalence of Babesia bigemina was higher in female cattle (50.46%) than in males (41.46%). In both cases, analysis of the results revealed no statistically significant difference between male and female cattle using the chi2 test, (p>0.05). Similar results were found by Wodajnew *et al.* [18], Fethu *et al* [6], Hika *et al.* [10] and Namomsa *et al.* [16]. Our results differ from those of Hamsho *et al*. [9], who found a significant difference (p<0; 05) between male and female cattle.

The prevalence rates of Babesia bovis and Babesia bigemina were significantly higher in older than in younger cattle (p<0; 05). This could be explained by the fact that animals have an acquired immunity that protects them against certain parasitic aggressions until adulthood. In both cases, statistical analysis of the results showed a significant difference (p<0; 05) between cattle of different age categories. Our results are similar to those found by Bipin *et al.* [1], Wodajnew *et al.* [18], and differ from those obtained by Namomsa *et al* [16], Fethu *et al.* [6], Tembue *et al.* [17] and Hika *et al.* [10].

5. Conclusion
The present study has revealed that bovine babesiosis is on the increase in the Bamako District. It is therefore necessary to strengthen the diagnostic capacities of veterinary laboratories and to consolidate the intervention capabilities of the country's veterinary services.

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