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Survival period of field isolates of *Trypanosoma vivax* in refrigerated blood

Short communication

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The simplest and most reliable parasitological technique for the diagnosis of *T. vivax* and *T. congolense* infections in domestic animals is to centrifuge unclotted blood in capillary tubes and look for the parasites packed between the buffy coat and the red cell layer (Murray et al., 1977). The content of this section of each capillary tube is expressed onto a slide and examined using phase contrast or darkground microscopy. The level of parasitaemia can be determined by a scoring system (Paris et al., 1982). However, in field situations, there can be considerable time lag between blood collection and examination unless a portable generator is available to provide electricity. The usual practice, therefore, is for blood samples to be cooled with ice and transported to the laboratory for examination. Paris and others working with cattle recommended that samples should be examined within 12 h if the parasites are to survive in reasonable numbers (Paris et al., 1982; Murray et al., 1983). Our studies indicate that the time could be much longer without any appreciable change in the trypanosome score, at least when examining small ruminants.

During routine screening of animals at Fasola in southwest Nigeria for trypanosomiasis 9 sheep and 9 goats naturally infected with *T. vivax* were identified. 5 ml of blood was obtained from each animal using EDTA vacutainer tubes. Samples were immediately cooled in ice and transported over 70 km to the laboratory in Ibadan where they were refrigerated at 4° C. Two, 16 and 24 h after collection, trypanosome scores were determined as described by Paris et al. (1982) and shown on Table 1. Parasites were found to be viable beyond 24 h although there was some reduction in the trypanosome scores (Table 2). Another set of infected blood samples from 3 sheep and 4 goats was then examined as before but at 6 hourly intervals until the 68th hour after collection.

The results are shown in Table 3. It was found that the mean score remained constant for 20 h in sheep and 26 h in goats infected with *T. vivax* Even at the 50th hour the mean score was still above 50% of the initial value with both species of animals. The results thus indicate that, with small ruminants infected with *T. vivax*, reliable trypanosome scores can be obtained up to 24 h after sample collection so long as the samples are properly refrigerated. Qualitatively, infection could be detected up to the third day after collection; trypanosomes survived better in goat blood than sheep blood. Similar studies with other species of trypanosomes and livestock would be worthwhile.

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Table 1. Darkground/phase contrast buffy coat parasitaemia scoring system (Paris et al., 1982)

Score	Trypanosomes/field*	Estimated parasitaemia trypanosomes/ml
6+	Swarming >100	$>5\times10^6$
5+	>10	$>5\times10^5$
4+	1–10	10^4–5×10^5
3+	1 per 2 fields to 1 per 10 fields	5×10^3–5×10^4
2+	1–10 per preparation	10^3–10^4
1+	1 per preparation	10^2–10^3

* magnification = 250

Table 2. Mean trypanosome scores with time in refrigerated blood samples from sheep and goats naturally infected with *T. vivax* (Experiment 1)

	Hours after blood collection		
	2	16	24
Sheep (n = 9)	3.8 ± 1.2	3.1 ± 1.2	2.2 ± 1.1
Goat (n = 9)	4.7 ± 1.1	3.7 ± 1.4	2.7 ± 1.3

Table 3. Mean trypanosome scores with time in refrigerated blood samples from sheep and goats naturally infected with *T. vivax* (Experiment 2)

Hours after blood collection	Sheep (mean ± SD) (n = 3)	Goat (mean ± SD) (n = 4)
2	4.3 ± 1.9	4.5 ± 1.1
8	4.3 ± 1.9	4.5 ± 1.1
14	4.3 ± 1.3	4.5 ± 1.1
20	4.3 ± 1.3	4.5 ± 1.1
26	3.6 ± 1.3	4.5 ± 1.1
32	3.6 ± 1.0	3.5 ± 1.1
38	3.3 ± 1.0	3.5 ± 1.1
44	3.3 ± 1.0	3.5 ± 1.1
50	2.3 ± 1.0	3.3 ± 1.5
56	2.9 ± 0.8	3.0 ± 1.2
62	1.3 ± 0.4	2.5 ± 1.1
68	1.0 ± 0.4	2.5 ± 1.1

SD = Standard deviation