Trypanosoma brucei and mixed *T. brucei/T. congolense* infections in cattle: their development and transmission

Peter Van den Bossche, Reginald De Deken, J. Brandt, D. Geysen, D. Berkvens, N. Speybroeck and Stanny Geerts

Institute of Tropical Medicine, Veterinary Department, Nationalestraat 155, 2000 Antwerp, Belgium

Introduction

Recent studies in Uganda have shown the important role cattle can play in the transmission and dynamics of *Trypanosoma brucei rhodesiense* human sleeping sickness (HSS) (Fèvre *et al.*, 2001). The role of cattle as reservoirs of human pathogenic trypanosomes may also be of considerable importance in many areas of southern and eastern Africa where, due to pressure for land, people and their livestock encroach into tsetse-infested, protected game areas that are known foci of HSS. Such a situation occurs, for example, in Malawi where people and cattle have settled along the edge of the Kasungu National Park and the Nkhotakota Game Reserve both well-known foci of human trypanosomiasis (Van den Bossche *et al.*, 2000).

Because of the potential implication of cattle in the epidemiology of HSS, it is important to determine the course of *T. brucei* infections in cattle and their transmissibility at the various stages of infection. Furthermore, since cattle are often subject to challenge with other trypanosome species such as *T. congolense* it is of particular importance to evaluate the effect of such an additional infection on the development and transmission of *T. brucei*. To clarify some of those important questions some cattle were infected experimentally and the development and transmission of *T. brucei* were studied. For security reasons use was made of *T. b. brucei*.

Material and methods

The experiment was conducted at the experimental stable of the Veterinary Department of the Institute of Tropical Medicine. Ten head of cattle (Belgian breed) were infected with *T. b. brucei* EATRO 1125, a strain isolated from a bushbuck in Uganda (Van Meirvenne *et al.*, 1975), through the bites of infected tsetse (*Glossina morsitans morsitans*). About two months after the *T. b. brucei* infection, eight of the ten animals (and one challenge control animal) were challenged with tsetse infected with *T. congolense* TRT 15 (savannah-type), a strain isolated from cattle in eastern Zambia. The development of the single and mixed infection was monitored using parasitological (buffy coat) and molecular tools (Geysen *et al.*, 2003). Concurrently, the packed cell volume (PCV) was measured and the body temperature recorded.

Once weekly, teneral tsetse (*G. m. morsitans*) were fed on the flank of the infected animals. These flies were bred at the tsetse rearing unit of the Institute of Tropical Medicine and are known to have a high vectorial capacity (Van den Abbeele, 2001). Engorged flies were retained and their infection status was determined 30 days later using the method described by Lloyd and Johnson (1924). To be able to identify mixed infections in tsetse, the mouthparts of all flies with trypanosomal infections and infections in the salivary glands were subjected to molecular analysis (Geysen *et al.*, 2003).

Results

About five days after the challenge by the tsetse infected with *T. b. brucei*, all experimental animals developed a parasitaemia which peaked about 10 days post infection. From day 55 onwards, it became difficult to detect parasites with parasitological diagnostic tools but animals remained positive on PCR.

Concurrent with the appearance of *T. b. brucei* in the jugular blood, animals developed an acute fever (Figure 1). From day 20 onwards, temperature was again

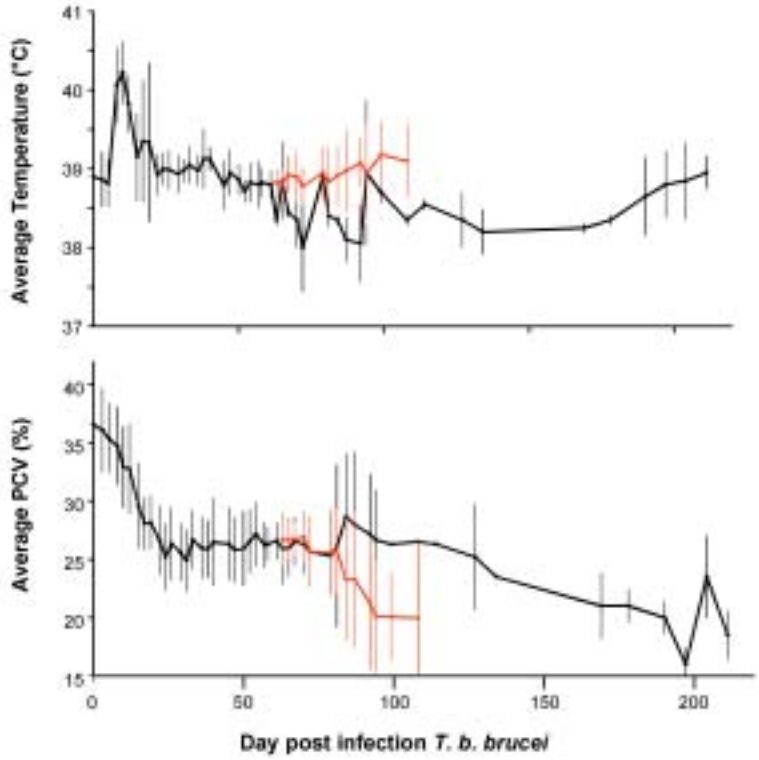

Figure 1. *Average temperature and average PCV of animals infected with a* T.b.brucei *(in black) and a mixed* T.b.brucei/ T.congolense *infection (in red)*

within the normal range and remained normal throughout the chronic phase of the *T. b. brucei* infection. The PCV declined steeply from an average of 36% to an average of 26% during the first 20 days of infection (Figure 1). It remained at this lower value in the *T. b. brucei* control animals.

not differ significantly throughout the observation period. For a period of about 8 months, tsetse continued to pick up trypanosome infections from the *T. b. brucei* control animals. During most of that time no trypanosomes were detected by parasitological means but the animals remained intermittently positive on PCR.

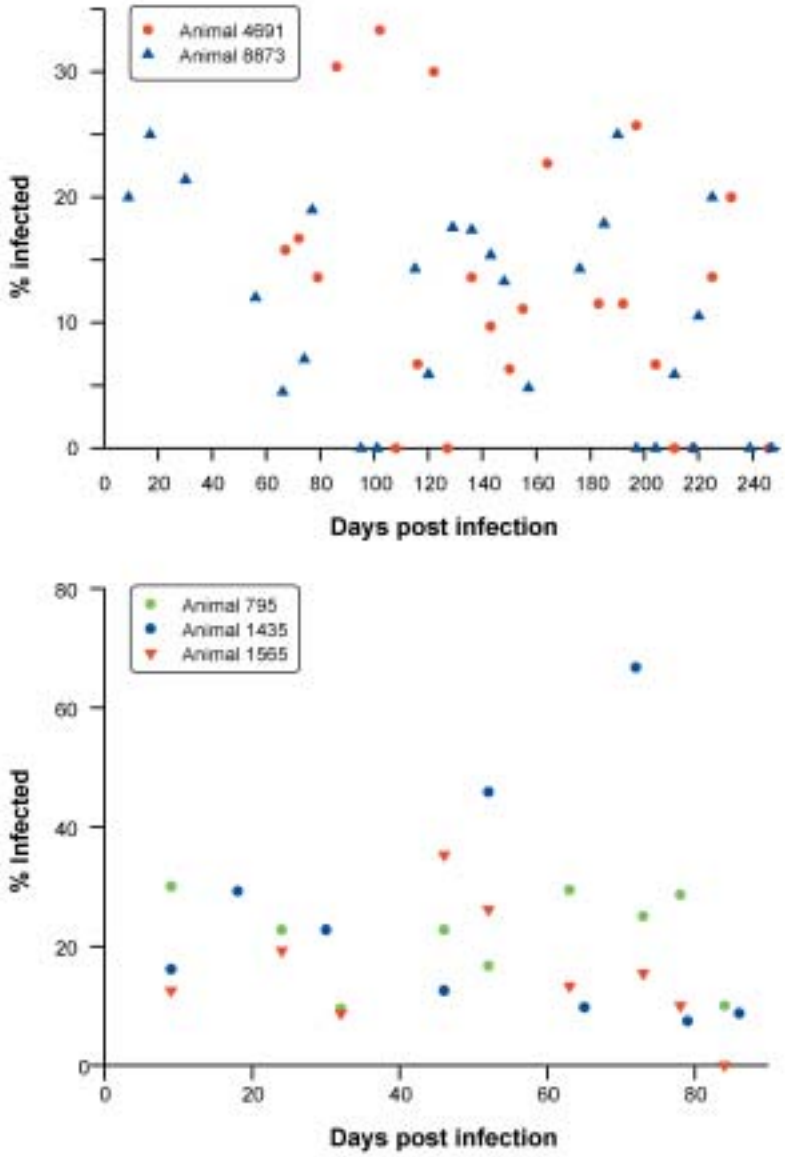

Figure 2. *Proportion of* G. m. morsitans *developing a metacyclic infection after being infected on animal with a single* T. b. brucei *(animals 4691 and 8873) or mixed* T. b. brucei/T. congolense *infection (animals 795, 1435 and 1565)*

After challenge with *T. congolense*, it took about 19 days before the parasites could be detected in the blood. The *T. congolense* infection caused an increase in body temperature and another sharp decline in the PCV (Figure 1). All animals were treated with diminazene aceturate (Berenil®, Hoechst) at 7.0 mg/kg body weight when the PCV reached a value below 19%.

The average proportion of tsetse that developed a metacyclic infection in the salivary glands was 18.8% (Figure 2). This proportion was not affected by the animal on which the flies were infected and did

Challenge with *T. congolense* and the subsequent development of a mixed *T. b. brucei/T. congolense* infection did not affect the *T. b. brucei* transmission rate. About 28% of the tsetse with a metacyclic infection were infected with a mixed *T. b. brucei/T. congolense* infection. The high proportion of mixed *T. b. brucei/T. congolense* infections was explained best by a model implying that if a fly is refractory to *T. congolense*, it is also refractory to *T. b. brucei* and vice versa.

Conclusions

According to our observations, the course of a *T. b. brucei* infection in cattle can be divided into two distinct phases. During the first or acute phase, which lasts for about 2 to 3 weeks, animals develop fever and the PCV declines steeply. In the subsequent or chronic phase of infection, which can last for more than a year, body temperature remains within the normal range and the PCV declines slowly. Parasitaemia is usually low and, from day 50 post-infection onwards, parasites become difficult to detect using the available parasitological diagnostic tools. Hence conventional diagnostic tools will most likely detect *T. b. brucei* infections in cattle in the early phase of infection. During the chronic phase of infection molecular tools are required. Despite low parasitaemias, cattle with such a chronic infection are important reservoirs and continue to infect tsetse. Infection with another trypanosome species such as *T. congolense* has little effect on the development and transmission of such a chronic *T. b. brucei* infection. Moreover, our results indicate that tsetse are effective vectors of mixed *T. b. brucei/T. congolense* infections. Ultimately, however, the steep decline in PCV and bad condition due to *T. congolense* may result in death of the infected animal.

References

Fèvre,E.M. et al. (2001). *Lancet* **358,** 625-628.
Geysen,D. et al. (2003). *Veterinary Parasitology* **110,** 171-180.
Lloyd,L.L. & Johnson,W.B. 1924. *Bulletin of Entomological Research* **14,** 225-227.
Van den Abbeele, J. 2001. PhD Thesis, Antwerp University, 121pp..
Van den Bossche, P. et al. (2000). *Veterinary Parasitology* **88,** 163-176.
Van Meirvenne, N. et al. (1975). *Annales de la Société Belge de Médicine Tropicale* **55,** 1-23.