INTRODUCTION

In South Africa, the distribution of tsetse has undergone considerable changes over the last century (Phelps & Lovermore 2004). The rinderpest epidemic of 1896 and intensive aerial spraying operations between 1946 and 1952 have resulted in the eradication of two savannah species, *Glossina morsitans morsitans* and *Glossina pallidipes*. However, two other tsetse species, *Glossina austeni* and *Glossina brevipalpis*, persisted in KwaZulu-Natal Province (Kappmeier, Nevill & Bagnall 1998). They are confined mainly to shaded areas along rivers as well as in forests or thickets. In 1990, a widespread trypanosomosis outbreak in cattle occurred in areas surrounding the then Hluhluwe Game Reserve (now incorporated in the Hluhluwe-iMfolozi Park) (Kappmeier, Nevill & Bagnall 1998). The outbreak was controlled by introducing pyrethroid dipping of cattle and the treatment of sick animals with diminazene aceturate or homidium bromide (Kappmeier, Nevill & Bagnall 1998). After a period of 2 years the outbreak was brought under control. Between 1992 and 1993 a trial to control *G. brevipalpis* was conducted in a 55 km² area of the Hluhluwe-iMfolozi Park. The trial only had limited success and prompted further research in improved control methods for this tsetse species. Since the nagana outbreak of the 1990s, little information is available on the prevalence of the disease in cattle. To obtain updated data on and assess the contribution of trypanosomosis to the disease burden of cattle kept at the edge of the Hluhluwe-iMfolozi Park, a survey was conducted at Mvutshini Dip. Use was made of a purposeful sampling strategy by restricting sampling to animals that the livestock owner considered to be in poor condition. Of a total of 76 blood samples collected, 26 were parasitologically positive and 46 were positive on PCR/RFLP. Almost all infections were due to *Trypanosoma congolense* savannah subgroup. A total of 63 animals had a PCV < 24% and were considered to be anaemic. Results from the survey show that trypanosome infections contribute significantly to the overall burden of disease in the area. Further research is required to develop appropriate control methods.

Keywords: Bovine trypanosomiasis, KwaZulu-Natal, *Trypanosoma congolense*
assess the contribution of nagana to the disease burden of cattle kept at the edge of the Hluhluwe-iMfolozi Park, this survey was conducted.

MATERIAL AND METHODS

Sampling site and sample selection

About 300 adult (> 12 months of age) communal cattle (Angoni breed), herded adjacent to the northeastern edge of the tsetse-infested (*G. brevipalpis*) Hluhluwe-iMfolozi Park, were presented at Mvutshini Dip situated at 32°09’ E and 28°08’ S. A total of 76 adult animals belonging to 32 herds were sampled. Since the aim of the survey was to establish the contribution of trypanosomosis to the disease burden in communal cattle herded at the edge of the Park, preference was given to a purposeful sampling strategy by restricting sampling to animals that the livestock owner considered to be in poor condition.

Diagnostic methods

From each animal, a sample of jugular blood was collected in vacutainer tubes coated with EDTA as anticoagulant. In the laboratory blood from each sample was decanted into plain microhaematocrit centrifuge capillary tubes that were sealed with “Cristaseal” (Hawksley) and centrifuged in a microhaematocrit centrifuge for 5 min at 9 000 rpm. After centrifugation, the packed cell volume (PCV) was determined. As in other countries of southern Africa with cattle of the same breed, animals with a PCV < 24 % were considered to be anaemic (Van den Bossche, Shumba & Makhamba 2000). The buffy coat and the uppermost layer of red blood cells of each specimen were extruded onto a microscope slide and examined for the presence of motile trypanosomes. Samples were examined with a phase-contrast microscope with a 40x objective lens.

For each blood sample, a second buffy coat was extruded onto a filter paper (Whatman no. 3, Whatman®). Filter papers were stored in sealed plastic bags containing silica gel at –18 ºC. The samples were further analysed using the PCR-RFLP described by Geysen, Delespaux & Geerts (2003).

All statistical analyses were performed using the statistical package SPSS (SPSS Inc. 2004).

RESULTS

A total of 26 trypanosomal infections (34.2 %) were detected using the buffy coat method. With the ex-
ception of one infection (mixed *Trypanosoma congolense* and *Trypanosoma vivax*), they were all *T. congolense*. Using the PCR-RFLP, a total of 46 (60.5%), infections (all *T. congolense* Savannah subgroup) were detected. All the parasitologically positive animals were positive on PCR-RFLP.

Of the 76 animals sampled, 63 animals were anaemic. Thirty-nine (84.8%) of the 46 PCR-RFLP positive animals were anaemic. The average PCV of all animals sampled was 19.8 ± 4.2%. The average PCV of parasitologically positive animals (18.6 ± 3.8%) differed little (P > 0.05) from the PCV of the parasitological negative animals (20.5 ± 4.4%). Similarly, the average PCV of animals negative on PCR-RFLP (20.2 ± 4.3%) differed little (P > 0.05) from the PCV of animals positive on PCR-RFLP (19.6 ± 4.3%). The distribution of the PCV-values of all animals sampled and of the PCR-RFLP positive and negative animals are presented in Fig. 1.

**DISCUSSION**

Results of this survey show that bovine trypanosomosis is prevalent in the vicinity of the Hluhluwe-iMfolozi Park. A high proportion of the cattle that the owner considered to be in poor condition were infected with trypanosomes. The proportion of animals infected with trypanosomes almost doubled when the samples were analysed using molecular tools. This is not surprising considering the high specificity but low sensitivity of the buffy coat method (Paris, Murray & McOdimba 1982). Although no exact estimates of the prevalence can be made, this finding suggests that trypanosomosis contributes significantly to the overall burden of disease in cattle in the area. One of the most typical signs of bovine trypanosomosis is the development of anaemia which is best measured by determining the PCV (Murray & Dexter 1988). It is thus not surprising that a large proportion of the infected animals were anaemic. However, an equally large proportion of the PCR-RFLP negative animals also had low PCV values. The reason for this is not clear but it suggests that other factors (such as certain tick-borne diseases) causing anaemia are also present in the area, which may aggravate the outcome of trypanosome infections. Further research is required to determine the causes of this anaemia.

Effective control of the trypanosomosis situation requires a good understanding of the epidemiological circumstances in the areas surrounding the Hluhluwe-iMfolozi Park. Firstly, an extensive survey should be conducted to determine the distribution of nagana in the areas surrounding the Park. The outcome of the survey may clarify the origin of tsetse challenge in the area of interest. The effectiveness of various control options relies on the distribution of the tsetse challenge. Indeed, pyrethroid-treatments of cattle to control tsetse will only be effective when cattle are mainly challenged by tsetse from populations established outside the Park which are largely dependent on cattle for their survival. If, on the other hand, cattle are challenged mainly by tsetse that are highly dependent on the game animals other tsetse control options need to be considered. In the meantime, strategic use of trypanocidal drugs may alleviate the disease situation in the cattle in the region.

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**REFERENCES**


