Prevalence and incidence of bovine trypanosomosis on the Adamaoua plateau in Cameroon ten years after the tsetse eradication campaign

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Abstract

Between March 2004 and February 2005 the monthly incidence of trypanosome infections was measured in cattle from nine sentinel herds in the Adamaoua province of Cameroon. Three herds of 20 cattle each were kept on the plateau which has been cleared from tsetse flies about ten years ago, three other herds were grazing in the tsetse infested valley whereas the last three were herded in the buffer zone. The cross-sectional study showed that the initial trypanosomosis prevalence was 1.8, 5.2 and 2.0 % on the plateau, in the buffer zone and the valley, respectively. During the longitudinal study the trypanosomosis incidence was high in the valley (3.7 to 20%) and the buffer zone (1.8 to 13.4%), whereas it was significantly lower (0 to 2.1%) on the plateau. Tsetse flies, mainly *Glossina morsitans submorsitans* and a few *G. tachinoides*, were caught in the valley and the buffer zone, but none on the plateau. The data indicate a low trypansomosis risk on the plateau. Further entomological studies, however, are required to clarify the origin of the trypanosome infections on the plateau.

Key words: trypanosomosis, Cameroon, incidence, sentinel herds, *Glossina* spp., risk
Introduction

Following the appearance of tsetse flies (*Glossina morsitans* submorsitans, *G. fuscipes* fuscipes and *G. tachinoides*) on the Adamaoua plateau in 1950 (Banser, 1979; Hurault, 1993; Martin, 2002), the government organized large-scale trypanocidal treatment campaigns in the affected regions of the Adamaoua, North and East Cameroon from 1960 to 1975. These campaigns provided preventive treatment at the start of the transhumance and curative treatment upon return or in the case of illness (Hamadama, 2001). An average number of 300,000 doses of diminazene aceturate (Berenil®, Hoechst) and 600,000 doses of Isometamidium chloride (Trypamidium®, Merial) were administered annually (Banser, 1979). In 1976, with the objective of controlling the vectors of trypanosomosis, a tsetse control unit was created in the far north of the country. This unit launched a ground-spraying campaign with DDT (Dichlorodiphenyl triclorethane) (Banser, 1979). Under the auspices of the lake Tchad Basin commission the operational capacities of the unit were intensified through a regional project between Cameroon and Nigeria. The two projects merged in 1979 to form a special tsetse eradication unit (Mission Spéciale d’Eradication des Glossines des Sciences, MSEG) with two field stations (one in the far north and the other in Adamaoua). The first field station continued to ground spray with DDT while the second resorted to aerial spraying with helicopters. However, despite the control efforts another invasion of tsetse occurred in 1990 spreading over the Adamaoua region from Tignere to Galim and in Mayo Banyo (Boutrais & Cuisance, 1995; Hurault, 1993).

Two aerial spraying campaigns on the Adamaoua plateau in 1991-1992 and 1994 resulted in effective control of tsetse flies from the re-invaded zones (Cuisance and Boutrais, 1995). To prevent reinvasion of tsetse flies from the valley a barrier consisting of targets and traps was put in place after aerial spraying. However, bush fires destroyed most of the targets and traps soon after deployment in 1994. Thereafter the barrier was replaced by a program of insecticide-treatments of cattle. At the end of 1994, a preliminary evaluation of the tsetse control activities in Adamaoua showed that the eradication campaign on the plateau had not been 100% effective and that some pockets of *G. m. submorsitans* and *G. f. fuscipes* had survived (Cuisance & Boutrais, 1995). Since then, no further entomological and parasitological information is available.
The present study aims to assess the situation of bovine trypanosomosis on the Adamaoua plateau in Cameroon ten years after the end of the tsetse control interventions.

**Material and methods**

**Study area**

The Adamaoua plateau is situated in the northern part of Cameroon and covers more than 72,000 km$^2$. Suchel (1972) describes two types of climate in the region: one covering the northern part of the Adamaoua plateau, the other covering the meridional reserve. The Faro & Deo division, our study area, is located in the northern part and has a Soudano-Sahelian climate. This area lies at an altitude of 1000-1100m above sea level with an average rainfall of 1800 mm. The rainy season lasts from March/April to October and most of the rainfall occurs between June and September. The Adamaoua plateau is covered with savannah consisting for more than 90% of *Daniellia olivert* and *Lophira lanceolata*. (Letouzey, 1969). The environment is very suitable for intensive cattle rearing. The zebu cattle (white and red Fulani and Goudali or Peuhl of the Adamaoua) are kept under traditional extensive husbandry systems with communal herding. In the dry season many cattle herds transhumate from the plateau to the tsetse infested-valley.

The Faro & Deo division, which covers 11,000 Km$^2$, counts 74,559 head of cattle (PACE, unpubl. report, 2005) and has 67,413 inhabitants (PNVRA, unpubl. report, 2001). At the end of the tsetse eradication campaigns in 1994 the territory was divided in the following three zones situated from south to north (Fig. 1):

1. The plateau: most of tsetse flies (*G. m. submorsitans*), with the exception of some small pockets, were cleared from this zone in 1994. Large ranches with big cattle herds are present on the plateau. There is only limited transhumance to the tsetse-infested valley in the dry season often involving only part of the herd. Cattle are regularly dipped during the rainy season.

2. Buffer zone: The MSEG has identified this zone to act as a barrier to tsetse invasion from the valley to the Adamaoua plateau (Boutrais & Cuisance, 1995). Therefore it was advised that all stationary herds should be treated regularly with
insecticides (pyrethroids). According to the MSEG and our own observations insecticides are used quite frequently, although the treatment frequency varies according to the breeder's financial resources. Some treat all the year round and others only during the dry or the rainy season. The most commonly used products are pyrethroids (Butox®, Cypermil®, Didétéki®, Ectopor® and Eradick®).

3. The valley: The plain of Koutine is an agricultural zone where the cattle keepers of the plateau and some herds from neighboring Nigeria pass the dry season (transhumance). The biggest tsetse threat is originating from the forest area where the flies have never been controlled and which is bordering the tsetse-infested game reserves of Faro (Boutrais & Cuisance, 1995).

In 1994 *Glossina morsitans submorsitans*, *G. fuscipes fuscipes* and *G. tachinoides* were present in the valley. *Glossina morsitans submorsitans* was present in the buffer zone and on the plateau (Cuisance and Boutrais, 1995).

**Trypanosomosis surveillance**

To assess the trypanosomosis risk nine sentinel herds, three in each zone of interest, were established and monitored between March 2004 and February 2005. Each sentinel herd consisted of 20, randomly selected and ear-tagged, adult Zebu cattle, kept under traditional village management (Fig 1). Sentinel cattle did not leave their respective zones during the observation period.

At the start of the surveillance period, all the sentinel animals received a curative, intramuscular treatment with diminazene aceturate (Berenil®, Hoechst) at a dose of 7mg/kg body weight. Blood was collected monthly from an ear vein into heparinized microhaematocrit centrifuge capillary tubes and onto glass slides, as thick and thin blood smears. The capillary tubes were sealed with “Cristaseal” (Hawksley, Lancing, UK) and centrifuged immediately in a microhaematocrit centrifuge for 5 min, at 7 500 g. After centrifugation, the PCV was determined. The buffy coat and the uppermost layer of red blood cells of each specimen were extruded onto a microscope slide and examined with a phase-contrast microscope with a x 40 objective lens for the presence of motile trypanosomes (Murray et al., 1977). Giemsa-stained thick and thin blood smears were examined under x 100 oil immersion objective lens for the presence of trypanosomes.
Animals, which were infected with trypanosomes, received a curative treatment with diminazene aceturate, as indicated above. The monthly trypanosomosis incidence was calculated as the number of new positive cases expressed as a percentage of the total number sampled, without taking into account the prophylactic effect of the drug.

**Tsetse monitoring**

Between April 2004 and March 2005 a total of 15 geo-referenced biconical traps (Challier et al., 1977) were used to monitor the tsetse population in the study area. Four traps were deployed on the plateau; 7 in the buffer zone and 4 in the valley (Figure 1). From April 2005 until September 2005 the number of traps was increased to 7 on the plateau, 10 in the buffer zone and 5 in the valley. Traps were checked twice a month. Records were kept of the number, species and sex of the tsetse captured in each trap. A monthly mean index of abundance (IA) of tsetse was calculated as the mean number of flies (males and females) captured per trap per day.

**Sero-conversion during transhumance**

To determine the level of challenge of cattle during transhumance in the valley, the sero-conversion of calves originating from the plateau was monitored after a first transhumance to the valley. For this purpose, hundred seronegative calves (age: 5 to 11 months old) belonging to 6 different herds were ear-tagged and went on transhumance between November 2004 and March 2005. Their blood was checked for the presence of anti-trypanosomal antibodies after transhumance. Anti-trypanosomal antibodies were determined on blood samples collected on filter paper (Whatman no. 4). Eluted blood spots were screened for the presence of trypanosomal antibodies using an indirect ELISA with *Trypanosoma congolense* as antigen (Rebeski et al., 2000). Negative control samples were obtained from non-infected cattle in the tsetse-free zone of Kousseri in Cameroon. Positive control sera were obtained from experimentally infected cattle with *T. congolense*. Samples giving a percentage positivity (PP) which exceeded the mean (PP) of negative samples plus three standard deviations were considered positive (Desquesnes et al., 2001).
**Statistical analysis**

Logistic regression analysis was carried out in order to detect any significant differences between the average trypanosomosis incidence of the sentinel herds in the 3 zones of the study area. Analysis of variance was used to compare the PCV of the cattle on the plateau, in the valley and in the buffer zone.

**Results**

**Trypanosomosis surveillance**

A total of 170 blood samples from the sentinel cattle was examined in March 2004. Trypanosomes were detected in 5 animals. Four infections were due to *T. congolense*, one to *T. brucei*. The prevalence on the plateau, in the buffer zone and in the valley was 1.8, 5.2 and 2.0%, respectively. During the eleven months surveillance period, a total of 60 trypanosomal infections were detected in the sentinel cattle: 3 on the plateau, 25 in the buffer zone and 32 in the valley. In all zones *T. congolense* (n=52; 86.7%) was the most common. *Trypanosoma vivax* was detected in 10% (n=6) of the cases and *T. brucei* in 1.7% (n=1). The remainder were mixed infections of *T. congolense* and *T. vivax* (n=1; 1.7%). The monthly incidence for each of the three zones is shown in Fig 2. It varied from 3.7 to 20.0 and from 1.8 to 13.4% in the valley and buffer zones, respectively. On the plateau the incidence remained very low (0-2.1%).

The average trypanosomosis incidence in the 3 sentinel herds of the plateau during the whole observation period was significantly lower than in the valley and the buffer zone (p<0.001). Furthermore, there was also a significant difference between the average incidence of the sentinel herds in the valley and in the buffer zone (p<0.05). In both zones the maximum incidence was recorded in September 2004 (rainy season). The analysis of variance of the PCVs of the cattle in the three zones (Fig 3) showed no significant differences (p = 0.304). Nevertheless the cattle on the plateau had a slightly higher mean PCV of 32.47. In the three zones, there was an increasing tendency of mean PCVs between June and July. Afterwards the means slightly decreased and levelled out.

**Tsetse monitoring**
A total of 744 *G. m. submorsitans* (263 females and 481 males) and 5 *G. tachinoides* (2 females and 3 males) were captured in the valley between April 2004 and September 2005. Only 6 *G. morsitans submorsitans* (1 female and 5 males) were captured in the buffer zone and none on the plateau. The monthly mean index of abundance is shown in Fig. 4.

**Sero-conversion during transhumance**

From the 100 sero-negative calves only 78 could be sampled again upon return from transhumance. In twelve (15%) animals anti-trypanosomal antibodies were detected. The proportion of animals that sero-converted varied according to the herd and the area where the herd had stayed during transhumance.

**Discussion**

Results from the trypanosomosis monitoring indicate that 10 years after the tsetse control operations in the Adamaoua region the incidence of trypanosomosis on the plateau is still relatively low (0-2.1%). This finding is in accordance with the observation that no tsetse flies were detected on the plateau. The mean PCV of the animals on the plateau would be expected to be higher than in the buffer zone and the valley, but this was not the case. The lower PCV on the plateau suggests the concomitant occurrence of other anaemia causing factors, presumably tick infestation, helminthosis, haemoparasitosis (other than trypanosomosis) and nutritional deficiencies. Frequent treatments of cattle with insecticide in the buffer zone and the valley might contribute of lower densities of haematophagous insects, notably *Stomoxys* spp., than on the plateau and might also have an impact on the PCV values. (B. Bauer pers communication)

Given the relatively high incidence of trypanosomosis both in the valley and in the buffer zone, it is surprising that only very few tsetse (n=6) were captured in the latter zone contrary to the valley. This might be explained inter alia by the fact that cattle are the most likely host of tsetse in the buffer zone (few game animals) and thus receive higher challenge compared to cattle kept in the valley where game animals are present. With the exception of a few *G. tachinoides*, the tsetse survey showed almost exclusively *G.m. submorsitans*. This proves that two of the 3 tsetse species, which were present in the Adamaoua region in 1950 (Banser, 1979; Hurault, 1993; Martin,
2002), are still there. Whether or not *G. f. fuscipes* has survived, needs to be further explored. However, we have to take into account an underestimation of the abundance of tsetse flies due to the use of biconical traps, which are not optimal to catch *G. m. morsitans* and *G. f. fuscipes*. Due to the small number of sentinel herds and traps which were used in this study these results have to be interpreted with caution and larger surveys need to be carried out in order to confirm these results.

The overall level of sero-conversion (15%) during transhumance, which was observed in this study, is similar to the peak trypanosomosis incidence rate (12-20%) of the sentinel herds in the valley. Given the important differences in sero-coversion from one herd to the other, however, the challenge in the tsetse infested valley is probably very different from one place to the other. This might be linked to the presence of areas with higher tsetse densities near to game reserves.

**Conclusion**

The combined use of entomological, parasitological and serological methods to assess the trypanosomosis risk in the 3 study zones has clearly shown that there is a significantly lower risk of infection on the plateau than in the buffer zone and the valley. This indicates that the results of the tsetse eradication campaign which took place in the early 1990 are still relevant. The regular insecticide treatment of the cattle herds in the buffer zone is probably contributing to prevent reinvasion of the plateau. Further entomological studies are required, however, to clarify the origin of the trypanosomosis challenge on the plateau.

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