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INCIDENCE OF CLINICAL AND SUB-CLINICAL *THEILERIA PARVA* INFECTION IN SANGA CATTLE KEPT UNDER DIFFERENT ECF CONTROL STRATEGIES IN ZAMBIA

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En Juin 1992, une épizootie de fièvre de la côte orientale fut déclarée dans la Province Centrale de la Zambie. Cette situation incita à l'étude de différentes méthodes de contrôle de la maladie applicables au secteur traditionnel. Cinq stratégies différentes, impliquant la lutte contre les tiques et/ou la vaccination par la méthode "d'infection et de traitement", furent évaluées sur des troupeaux de bovins Sanga pendant une période de deux ans et demi.

Deux troupeaux étaient protégés contre les tiques par des douches hebdomadaires d'acaricide et deux autres troupeaux n' étaient pas protégés contre le vecteur. Dans les deux cas, un seul des deux troupeaux était également vacciné. Un cinquième troupeau était vacciné et protégé contre les tiques par 18 douches d'acaricide par an. Seuls les animaux séro-négatifs aux anticorps de shizontes de Theileria parva (test IFAT) furent inclus dans l'étude. Les animaux furent testés sérologiquement chaque année en avril et en octobre (après les saisons d'adultes et de nymphes de Rhipicephalus appendiculatus). A la fin de l'expérimentation, en août 1995, tous les animaux furent artificiellement infectés pour déterminer leur état immunologique.

Toutes les méthodes de contrôle ont significativement réduit l'incidence des cas cliniques. Aucune différence de protection n'a pu être observée entre les animaux vaccinés et ceux protégés hebdomadairement contre les tiques. Les résultats de l'infection artificielle montrent qu' une proportion importante des animaux du groupe de contrôle (60%) mais aussi du troupeau protégé contre les tiques hebdomadairement (50%) ont été en contact avec le parasite sans développer de signes cliniques. Tous les animaux vaccinés ont résisté à l'inoculation d'une dose léthale de Theileria parva .

INTRODUCTION

Tick-borne diseases such as anaplasmosis, babesiosis and cowdriosis are known to be endemically stable in the Lutale area, while East Coast fever (ECF) had not been recorded in the area before 1992. Therefore, ECF represented an enormous danger for the cattle population and high losses could be expected once the disease was introduced. In June 1992, an outbreak of ECF occurred in an experimental herd used for the evaluation of different tick control strategies. This situation prompted the investigation into different ECF and tick control strategies applicable in the traditional sector.

MATERIALS & METHODS

The field trial was carried out in a typical woodland savannah area, at Lutale, 175 km west of Lusaka in Central Province of Zambia. Important tick species present are *Amblyomma variegatum*, *Boophilus decoloratus* and *Rhipicephalus appendiculatus*.

The trial was designed to compare the incidence of ECF and ECF-specific mortality under different combinations of tick control and ECF immunisation. The following 5 experimental herds were formed from the cattle used by de Castro *et al.*,(*in press*) in a previous tick control trial:

- ECF immunisation and no tick control (UI).
- ECF immunisation and seasonal treatment against ticks (SI).
- ECF immunisation and weekly treatment against ticks (TI).
- No ECF immunisation and weekly treatment against ticks (TNI).
- No ECF immunisation and no tick control (UNI).

Each group was composed of 41 animals and their offspring. Only animals which were serologically negative for *T. parva* schizont antibody in the IFA test were eligible for inclusion in the study. The trial was designed to cover the 1992-93, 1993-94 and 1994-95 tick seasons.

All cattle from groups UI, SI and TI were immunised against ECF by the infection and treatment method described by Radley *et al.* (1981). The vaccine used was the trivalent stabilate produced by the FAO "East Coast fever vaccine production and quality control" project in Lilongwe, Malawi. This stabilate, known as "Muguga cocktail", contains three different *T. parva* strains: *Muguga*, *Kiambu 5* and *Serengeti* transformed.

The initial immunisation was carried out on the 28/10/92. All calves born after 28/10/92 and belonging to an immunised group were vaccinated in November each year (starting in November 1993). The minimum age of calves at immunisation was 4 weeks.

In the intensively sprayed herds (TNI and TI), all animals were sprayed weekly with a pyrethroid acaricide cypermethrin (Barricade®, Shell Zambia Ltd.) throughout the trial. Animals in the strategic group (SI) were sprayed, with the same acaricide, every four weeks in the dry season, from April to September, and every two

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weeks in the wet season, from October to March. On the day of spraying, animals from the unsprayed groups (UI and UNI) were kept in their respective kraals during acaricide application and were thereafter grazed separately.
The experimental animals were kept under a traditional rangeland grazing system simulating as closely as possible the typical production system of the area.
The animals in all herds received chemoprophylaxis against trypanosomiasis with Samorin® (2%) at intervals of approximately six months. All animals also received an annual treatment with Berenil® to remove any Samorin®-resistant trypanosome strains (No breakthroughs of trypanosomiasis were observed). Specific treatments were administered to individual animals on diagnosis of disease problems other than ECF. Diagnosed cases of ECF were left untreated.
Blood samples were collected from all animals twice a year at the end of October (end of the nymphal challenge) and at the end of April (end of the adult challenge). Sera were tested for antibodies to *T. parva* with the IFA test using schizont antigen at the laboratory of the Belgian Animal Development Co-operation project in Chipata. Titres ≥ 1:40 were considered positive.
At the end of the field trial, an artificial challenge experiment to determine the immunological status of each group was undertaken. Twenty adult cattle, present at the beginning of the experiment (on the 28/10/92), were randomly selected from each of the trial groups. A control group of 10 pure breed Friesian steers (150 to 250 kg) was purchased from an ECF-free dairy farm in Southern Province and transported to Lutale. These animals were negative for *T. parva* antibodies (IFA tested) and *Theileria* piroplasms could not be detected in their blood.All animals were dewormed one week before the experimental challenge and kept under intensive tick control by a weekly pour-on application of Renegade®. On 9 July 1995, all animals were inoculated subcutaneously in the mid-neck with 1 ml of undiluted stabilate (Batch 10).
Rectal temperatures of each animal were recorded every day for 35 days. Blood smears and lymphnode biopsies were prepared from each animal with a body temperature exceeding 39.5°C but no treatment was carried out. East Coast fever reactions in cattle were classified as described by Norval *et al.*, 1992. Serum for schizont antibody IFA testing were collected from all animals on days 0, 21, 28 and 35 and frozen at -20°C
Animals with non apparent or mild reactions were considered to be protected from ECF whereas moderate and severe reactions were interpreted as indicative for naive animals.

RESULTS

After the initial outbreak, the incidence of ECF was not constant and only the first tick season after immunisation was of importance. The highest incidence of clinical ECF, 0.0035 cases/day, occurred in the first adult *R. appendiculatus* season in 1993, just after immunisation. The incidence of ECF declined markedly over the next two seasons of adult activity, and no new cases were seen during the last observed season of adult *R. appendiculatus* challenge between January and March 1995. During the two periods of nymphal challenge the incidence of clinical ECF was 0.0003 cases/day.
In April 1993, after the first adult tick challenge, eight animals (25.8%) of the survivors of the control group were seropositive. Since thirteen animals died of ECF during this period, 43% of the initial UNI group became infected with *Theileria parva* and 38% of those went through sub-clinical theileriosis. Only 34% of the total number of immunised animals were found positive five months after immunisation. Despite the fact that the UI group were under no tick control and that a relatively high incidence of the disease occured during this period, only 50% of the animals were found sero-positive. The overall sero-prevalence was even lower in the next surveys, following the general pattern of the disease incidence. Only 10% of the control group and 12% of the immunised animals were seropositive in September 1994.
The overall mortality rate was higher in calves than in adults while the ECF-specific mortality was higher in adults than in calves. Very high mortality was observed in the UNI group with adult annualised mortality of 26% and calf annualised mortality of 90%. All adult mortality in this group was due to ECF while only 10% of the calf mortality was from confirmed infection with *T. parva*. No significant difference in mortality could be observed between animals protected from ECF by immunisation or by tick control. No clinical cases of ECF were seen in calves from any of the treated groups and no case of ECF occurred in the TI group.
The results of the artificial challenge showed that 12 (60%) and 10 (50%) animals from the UNI and TNI group respectively did not react to the inoculation of a lethal dose of *Theileria parva.* In the control group, naive Friesians, all animals were severely affected while only one immunised animal showed a moderate reaction and then recovered.

DISCUSSION

Cattle immunised by infection and treatment are solidly immune to challenge with the homologous parasite for at least three years (Burridge *et al.*, 1973). Indeed, all immunised animals resisted reinfection with a homologous challenge of *Theileria parva.* Although there was no significant difference between immunised groups, four mild reactions and one moderate were recorded in group TI (but all animals recovered) while in groups SI and UI not a single animal showed a reaction to the artificial challenge. This emphasises the importance of maintaining a low tick infestation on immunised animals to boost their immunity. This could have been more marked if the experimental period had been longer.
About half of the non-immunised animals were not susceptible to ECF three years after the introduction of *T. parva* into the area. The Sanga cattle breed is known to be very resistant to a number of diseases but, although this innate resistance might partly extend to ECF, it is believed that most of the resistance displayed in the challenge experiment is due to immunity acquired silently during the three-year field trial. The low ECF incidence in the second part of the experiment resulting in a low number of infected ticks and/or ticks with a low number of infected acini may have played a role in those mild infections. The relatively low tick burden and the proportion of

animals with a low piroplasm parasitaemia level in the grazing area may have contributed to this natural immunisation.

The comparison between the non-immunised groups under different acaricidal treatments suggests that, despite the intensive dipping programme, animals were still exposed to the disease as reported before by Morzaria *et al.* (1988). This may be due to inefficient control of ticks or to very rapid transmission of *T.parva* sporozoites soon after tick attachment (Young, 1981). The high ambient temperature observed in the area contributed to this ability for quick transmission of the disease (Young and Leitch, 1981) once ticks attached to their host.

The high proportion of protected animals in the area led to a reduction of the incidence of the disease over time. The persistence of East Coast fever challenge is demonstrated by the growing seroprevalence in non-immunised cattle. The large immunisation campaign seems to have accelerated the progress to an endemic status in the area. The degree of stability of this endemicity relies mainly on the equilibrium between tick burden and proportion of naive animals in the area. Any dramatic change in either of these components could jeopardise the maintenance of endemic stability.

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REFERENCES

Burridge, M.J. and Kimber, C.D. (1973). Duration of serological response to the indirect fluorescent antibody test of cattle recovered from *Theileria parva* infection. *Research in Veterinary Science* **14**: 270-271.

de Castro, J.J., James, A.D., Minjauw, B., Di Giulio, G.U., Permin, A., Pegram, R.G., Chizyuka, H.G.B. and Sinyangwe, P. (In Press). Long-term studies on the economic impact of ticks in Zambia. *Experimental and Applied Acarology*

Minjauw, B. (1996). Epidemiology and control of East Coast fever in traditionally managed Sanga cattle. PhD Thesis, University of Reading. 197 pp.

Morzaria, S.P., Irvin, A.D., Wathanga, J., D' Souza, D., Katende, J., Young, A.S., Scott, J. and Gettinby, G. (1988). The effect of East Coast fever immunisation and different acaricidal treatments on the productivity of beef cattle. *Veterinary Record* **123**: 313-320

Radley, D.E. (1981). Infection and treatment method of immunization against theileriosis. In: *Advances in the Control of Theileriosis: Proceedings of an International Conference Held at ILRAD, Nairobi, 9-13th February, 1981.* Editors, A.D. Irvin, M.P. Cunningham and A.S. Young, Martinus Nijhoff Publishers, The Hague, pp. 227-237.

Young, A.S. (1981). The epidemiology of theileriosis in East Africa. In: *Advances in the Control of Theileriosis: Proceedings of an International Conference Held at ILRAD, Nairobi, 9-13th February, 1981.* Editors, A.D. Irvin, M.P. Cunningham and A.S. Young, Martinus Nijhoff Publishers, The Hague, pp. 38-55.

Young, A.S. and Leitch, B.L. (1981). Epidemiology of East Coast fever: some effects of temperature on the development of Theileria parva in the tick vector, Rhipicephalus appendiculatus. *Parasitology* **83**: 199-211.