Onderstepoort J. vet. Res., 54, 211–213 (1989)

RESEARCH COMMUNICATION

COMPARATIVE INFECTION RATES OF *THEILERIA PARVA LAWRENCEI* IN SALIVARY GLANDS OF *RHIPICEPHALUS APPENDICULATUS* AND *RHIPICEPHALUS ZAMBEZIENSIS*

E. F. BLOUIN and W. H. STOLTSZ, Veterinary Research Institute, Onderstepoort 0110

ABSTRACT

BLOUIN, E. F. & STOLTSZ, W. H., 1989. Comparative infection rates of *Theileria parva lawrencei* in salivary glands of *Rhipicephalus appendiculatus* and *Rhipicephalus zambeziensis*. *Onderstepoort Journal of Veterinary Research*, 56, 211–213 (1989).

Three cattle, which had been experimentally infected with *Theileria parva lawrencei* and maintained as carriers of the infection, were each infested simultaneously with clean nymphal *Rhipicephalus appendiculatus* and *Rhipicephalus zambeziensis* in ear bags on separate ears. After moulting, the ensuing adult ticks were fed on rabbits for 4 days and their salivary glands were examined for infective stages of the parasite. Microscopic examination revealed significantly higher infection rates in the salivary glands of *R. zambeziensis* than in *R. appendiculatus* which may indicate an increased vector efficiency of *R. zambeziensis* for *T. p. lawrencei*.

INTRODUCTION

Corridor disease, caused by *Theileria parva lawrencei,* is transmitted from buffalo *(Syncerus caffer)* to cattle by *Rhipicephalus appendiculatus* (Neitz, 1955). In the Republic of South Africa (RSA) it is a controlled disease and is endemic to the Kruger National Park and some of the Zululand game reserves (Potgieter, Stoltsz, Blouin & Roos, 1988). Sporadic outbreaks occur on farms bordering on, or in close proximity to, these endemic areas and have thus far always been associated with the presence of buffalo (Bigalke, De Vos & Barrowman, 1976).

De Vos (1981) indicated that *R. appendiculatus* was still considered to be the only known tick species capable of transmitting Corridor disease. *Rhipicephalus zambeziensis* was described by Walker, Norval & Corwin (1981) as a new tick species from eastern and southern Africa, including the RSA, which closely resembled *R. appendiculatus*. *R. zambeziensis* is generally found in hotter, drier areas than *R. appendiculatus* and the 2 species have often been confused in the past (Norval, Walker & Colborne, 1982). In the RSA, the distribution of *R. zambeziensis* has not been extensively surveyed but it has reportedly occurred on game species in the Kruger National Park (Horak, Potgieter, Walker, De Vos & Boomker, 1983). It is likely that this species is more widely distributed than present data indicate.

Lawrence, Norval & Uilenberg (1983) demonstrated that *R. zambeziensis* was an experimental vector of *T. p. lawrencei* and that infection rates in salivary glands were high. They also suggested that in certain parts of Zimbabwe *R. zambeziensis* may be the only vector of *T. p. lawrencei*. In similar trials, a South African strain of *R. zambeziensis* has been successfully used in the transmission of *T. p. lawrencei* between cattle (Potgieter *et al.*, 1988) and infection rates in ticks were also found to be high. Since infection rates in *R. appendiculatus* fed on cattle are generally low (Young & Purnell, 1973), a comparison of the infection rates in the 2 tick species was conducted, using 3 *T. p. lawrencei* carrier cattle. This report presents the results of those trials.

MATERIALS AND METHODS

Three cattle *(Bos taurus* and *Bos indicus* crosses*)*, reared under tick-free conditions at the Veterinary Research Institute, were infected with the Hluhluwe 3 isolate (De Vos, 1982) of *T. p. lawrencei*. Details of the infection of individual animals are given in Table 1.

Ticks were reared and fed according to the methods of Neitz, Boughton & Walters, 1971. Each of the animals was infested with 300 nymphal *R. appendiculatus* (Rietvlei cross) (De Vos & Roos, 1981) on 1 ear and 300 *R. zambeziensis* (Killkenny) (Potgieter *et al.*, 1988) on the other ear. Engorged nymphae were collected and allowed to moult in an acaridarium at 25 °C and 85 % relative humidity.

At approximately 6 weeks post-moulting, 150 of the ensuing adults of each species from each feeding were fed on rabbits. At 4 days post-infestation the ticks were removed. Each tick was placed into a drop of RPMI 1640 medium[1] and its dorsal exoskeleton was cut away. The salivary glands of individual ticks were dissected out and teased onto glass slides. The glands were fixed in Carnoy's fixative, stained with methyl green/pyronin (Irvin, Boarer, Dobbelaere, Mahan, Masake & Ocama, 1981) and examined with a light microscope for infective stages of *T. p. lawrencei*.

RESULTS

Despite the relatively small numbers of ticks recovered and examined (refer to Table 2), it was clear that significantly higher infection rates were obtained with *R. zambeziensis* ticks which fed as nymphae on Animals 9512–9 and 9418–5 (Table 2). The mean number of infected acini per infected tick was also higher in *R. zambeziensis* than in *R. appendiculatus*. No infective stages could be found in any of the ticks fed as nymphae on Animal 9666–0. This result was not surprising, since piroplasms could not be demonstrated microscopically in blood smears of the animal examined during this period. Subinoculation of blood from Animal 9666–0 to a splenectomized ox did, however, result in a patent parasitaemia in the latter animal, thus confirming the carrier status.

DISCUSSION

Chemotherapeutic treatment of cattle involved in Corridor disease outbreaks is not recommended, as this may result in the creation of carrier animals

Received 6 March 1989—Editor

[1] Highveld Biological (Pty) Ltd, P.O. Box 488, Kelvin, 2054, RSA

TABLE 1 History of the infection of experimental cattle with *Theileria parva lawrencei* prior to tick feeding

Animal No.	Method of infection	Chemotherapy	Outcome
9512–9 (S)	Transmission with adult *R. appendiculatus*	Parvaquone[1] 10 mg/kg IM on days 11, 13, 26, 28 and 30 p.i. Buparvaquone[2] 5 mg/kg IM at 24 months p.i.* (5 treatments at days 0, 2, 4, 7 and 10)	Patent piroplasm parasitaemia 130 days p.t. 1/1000 rbc** Temporary disappearance of patent parasitaemia. Piroplasms reappeared 150 days p.t. < 1/10000 rbc**
9660–0	Transmission with adult *R. appendiculatus*	Parvaquone[1] 10 mg/kg IM on days 11 and 13 p.i.	Schizonts intermittently observed up to 124 days p.i. No patent piroplasm parasitaemia***
9418–5 (S)	Subinoculation of blood from a carrier ox†	No treatment required	Piroplasm parasitaemia 1/3000 rbc**

p.i. = post-infection
S = splenectomized
rbc = red blood cells
p.t. = post-treatment
IM = intramuscularly

* Attempt to sterilize piroplasm infection
** Parasitaemia during trial
*** Carrier status confirmed after subinoculation of blood to a splenectomized ox
† Details of infection given in Potgieter *et al.*, 1988

[1] Clexon, Coopers Animal Health Ltd, England
[2] BW 720C, Coopers Animal Health Ltd, England

TABLE 2 Comparative infection rates of *Theileria parva lawrencei* in the salivary glands of *Rhipicephalus appendiculatus* and *Rhipicephalus zambeziensis* fed on carrier cattle

Tick species	Animal No.	Adult ticks fed on rabbits		Mean No. of infected acini per infected tick
		No. infected/ No. examined	% infection	
R. appendiculatus	9512–9	0/35	–	–
	9512–9*	0/25	–	–
	9418–5	4/72	5	4
	9666–0	0/60	–	–
R. zambeziensis	9512–9	31/62	50	18,7
	9512–9*	2/28	7	3
	9418–5	40/43	93	28
	9666–0	0/57	–	–

* Second tick feeding 4 months after treatment with buparvaquone

(Potgieter, Roos & De Vos, 1985). When such methods are used to establish experimental *T. p. lawrencei* infections in cattle, piroplasm parasitaemias are usually very low. Infection rates in *R. appendiculatus* fed on these carriers are also low. The initial results presented here suggest that *R. zambeziensis* may be more efficient in picking up low level infections of at least certain isolates of *T. p. lawrencei* than *R. appendiculatus*. Higher yields of tick-derived parasite material for serological and immunological studies have thus been obtained by feeding *R. zambeziensis* on experimental carrier animals, but only when using animals with microscopically detectable parasitaemias.

The role that *R. zambeziensis* plays in the epidemiology of Corridor disease in the RSA can only be evaluated after further information on its distribution and abundance is obtained. It has been noted in Zimbabwe that, in the absence of tick control, *R. zambeziensis* appears to maintain lower levels of abundance than *R. appendiculatus* (Norval *et al.*, 1982). The primary concern is that a more efficient vector might enhance the transmission of *T. p. lawrencei* between cattle and increase the possibility of the parasite changing its behaviour to that of *T. p. parva* as reported by Barnett & Brocklesby (1966) and Young & Purnell (1973). This phenomenon has thus far not been observed in the RSA in field or laboratory observations despite the fact that *R. zambeziensis* has probably existed in endemic areas for many years.

ACKNOWLEDGEMENTS

The authors wish to thank Dr F. T. Potgieter for his valuable advice and criticism during the preparation of the manuscript and Mr B. R. Boersema for his assistance in the tick feedings.

REFERENCES

BARNETT, S. F. & BROCKLESBY, D. W., 1966. The passage of *Theileria lawrencei* (Kenya) through cattle. *British Veterinary Journal*, 122, 396–409.

BIGALKE, R. D., DE VOS, A. J. & BARROWMAN, B. R., 1976. The control of some tick-borne diseases in South Africa. *Bulletin Office International des Épizooties*, 86, 89–100.

DE VOS, A. J., 1981. *Rhipicephalus appendiculatus:* Cause and vector of diseases in Africa. *Journal of the South African Veterinary Association*, 52, 315–322.

DE VOS, A. J., 1982. The identity of bovine *Theileria* spp. in South Africa. M. Med. Vet. thesis, University of Pretoria, Pretoria, South Africa.

DE VOS, A. J. & ROOS, J. A., 1981. Observations on the transmission of *Theileria mutans* in South Africa. *Onderstepoort Journal of Veterinary Research*, 48, 1–6.

HORAK, I. G., POTGIETER, F. T., WALKER, J. B., DE VOS, V. & BOOMKER, J., 1983. Ixodid tick burdens of various large ruminant species in South African nature reserves. *Onderstepoort Journal of Veterinary Research*, 50, 221–228.

IRVIN, A. D., BOARER, C. D., DOBBELAERE, D. A. E., MAHAN, S. M., MASAKE, R. & OCAMA, J. G. R., 1981. Monitoring *Theileria parva* infection in adult *Rhipicephalus appendiculatus* ticks. *Parasitology*, 82, 137–147.

LAWRENCE, J. A., NORVAL, R. A. I. & UILENBERG, G., 1983. *Rhipicephalus zambeziensis* as a vector of bovine theileriae. *Tropical Animal Health and Production*, 15, 39–42.

NEITZ, W. O., 1955. Corridor disease: A fatal form of bovine theileriosis encountered in Zululand. *Bulletin of Epizootic Diseases of Africa,* 3, 121–123.

NEITZ, W. O., BOUGHTON, F. & WALTERS, H. S., 1971. Laboratory investigations on the life cycle of the Karoo paralysis tick *(Ixodes rubicundus* Neumann, 1904*). Onderstepoort Journal of Veterinary Research,* 38, 215–224.

NORVAL, R. A. I., WALKER, J. B. & COLBORNE, J., 1982. The ecology of *Rhipicephalus zambeziensis* and *Rhipicephalus appendiculatus* (Acarina, Ixodidae) with particular reference to Zimbabwe. *Onderstepoort Journal of Veterinary Research,* 49, 181–190.

POTGIETER, F. T., ROOS, J. A. & DE VOS, A. J., 1985. Implications of chemotherapy of *Theileria lawrencei* infections (Corridor disease) in cattle (Abstract). *South African Journal of Science,* 81, 44.

POTGIETER, F. T., STOLTSZ, W. H., BLOUIN, E. F. & ROOS, J. A., 1988. Corridor disease in South Africa: a review of the current status. *Journal of the South African Veterinary Association,* 59, 155–160.

WALKER, J. B., NORVAL, R. A. I. & CORWIN, M. D., 1981. *Rhipicephalus zambeziensis* sp. nov., a new tick from eastern and southern Africa, together with a redescription of *Rhipicephalus appendiculatus* Neumann, 1901 (Acarina, Ixodidae). *Onderstepoort Journal of Veterinary Research,* 48, 87–104.

YOUNG, A. S. & PURNELL, R. E., 1973. Transmission of *Theileria lawrencei* (Serengeti) by the ixodid tick, *Rhipicephalus appendiculatus. Tropical Animal Health and Production,* 5, 146–152.