

THE EXPERIMENTAL TRANSMISSION OF *THEILERIA OVIS* BY *RHIPICEPHALUS EVERTSI MIMETICUS* AND *R. BURSA*

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ABSTRACT

NEITZ, W. O. The experimental transmission of *Theileria ovis* by *Rhipicephalus evertsi mimeticus* and *R. bursa*. *Onderstepoort J. vet. Res.* 39 (2) 83-86 (1972).

The successful transmission of *Theileria ovis* Rodhain, 1916 to splenectomized sheep by *Rhipicephalus evertsi mimeticus* Dönitz and *R. bursa* Canestrini & Fanzago is recorded. The protozoon is acquired by the immature stages of these two ticks and transmitted by the ensuing adults. The course of the disease is accompanied by pyrexia and a transitory appearance of schizonts in films prepared from swollen lymph nodes while endoglobular parasites are demonstrable during the reaction and for long periods after recovery.

INTRODUCTION

Benign ovine and caprine theileriosis, which is caused by *Theileria ovis* Rodhain, 1916 (syn. *Theileria recondita* Lestoquard, 1929) is widely distributed in Africa, Asia and Europe. Countries in which the disease has been established are recorded by Neitz (1957) and in the FAO-WHO-OIE Animal Health Yearbook (1970). Investigations on the biological transmission are limited to those conducted in the Russian Federated Socialist Republic and South Africa (Table 1).

The object of this paper is to record that *Rhipicephalus bursa* from the northern hemisphere is capable of transmitting the South African *T. ovis* strain and that *R. evertsi mimeticus* from South West Africa can serve as a vector of this protozoon.

MATERIALS AND METHODS

1. Origin of the ticks

Larvae, the progeny of two *R. bursa* females, were supplied by Dr. C. R. Lambert of the Ciba-Geigy Research Laboratory, Domaine de la Barge, Switzerland.

Two engorged *R. e. mimeticus* females, taken off sheep, were obtained from Dr. J. D. Bezuidenhout, Veterinary Investigation Centre, Windhoek, South West Africa.

2. Tick-breeding

The procedures for rearing ixodid ticks, described by Neitz, Boughton & Walters (1971) were employed. Both tick species were maintained in the acaridarium at 25 to 26°C and 85 to 90% RH. They were kept in glass tubes plugged with cotton wool and examined daily until metamorphosis had taken place. Tick-breeding observations were recorded in a register.

3. Tick hosts

Larvae and nymphae of both two-host tick species were fed on the ears of splenectomized sheep harbouring a microscopic *T. ovis* infection as shown in Table 2. The ensuing adults were fed on the left ear of three fully susceptible splenectomized sheep recorded in Table 3. Ear-bags, secured around the base of the ears with Unna's paste, were used to retain the ticks on the hosts. From the third day after infestation the ear-bags

TABLE 1 Biological transmission of *T. ovis*

Vector	Country	No. of hosts	L	N	I	Authors
<i>Rhipicephalus bursa</i> Canestrini & Fanzago	Transcaucasia (USSR) + South Africa	2	X—X—)	X—X—)	X—X—)	Rastégaieff, 1934* Neitz, 1972 (present studies)
<i>Rhipicephalus evertsi evertsi</i> Neum.	South Africa	2	X—X—)	X—X—)	X—X—)	Jansen & Neitz, 1956
<i>Rhipicephalus evertsi mimeticus</i> Dönitz	† South Africa	2	X—X—)	X—X—)	X—X—)	Neitz, 1972 (present studies)
<i>Dermacentor marginatus</i> Olenov	Transcaucasia	3		X—X—)	X—X—)	Rastégaieff, 1940 (Cited by Bitukov, 1953)
<i>Dermacentor silvarum</i> Sulzer	Transcaucasia	3		X—X—)	X—X—)	Rastégaieff, 1937
<i>Haemaphysalis sulcata</i> Canestrini & Fanzago	Kazakhstan (USSR) . .	3		X—X—)	X—X—)	Bitukov, 1953
<i>Ornithodoros laborensis</i> Neum.	Transcaucasia	Many		X—X—)	X—X—)	Rastégaieff, 1935, 1936
	Kazakhstan		?)—?)	?)—?)	?)—?)	Bitukov, 1953

Legend: X = Stage in which infection is acquired
) = Stage in which infection is transmitted
 L = Larva; N = Nympha; I = Imago
 * = See remarks in text
 + = Investigations with ticks from Giba-Geigy, Switzerland
 † = Investigations with ticks from South West Africa

TABLE 2 Attempts to infect rhipicephalid ticks with *T. ovis*

No. of splenectomized <i>T. ovis</i> carrier	Infested with immature stages of	Batch No.	Fed for days	No. of engorged nymphae collected	Premoulting period in days	Prefeeding period in days	Ensuing adults fed on sheep No. (Table 3)
S 15535 . . .	<i>R. bursa</i>	3317 Ba	15-20	59	19	123	2476
S 15535 . . .	<i>R. (e.) mimeticus</i>	3349 Aa	13-28	119	39	29	2191
S 2476 . . .	<i>R. (e.) mimeticus</i>	3349 Ca	14-24	30	38	29	2212

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TABLE 3 Biological transmission of *T. ovis* by *R. bursa* and *R. e. mimeticus*

Sheep			Infestation with adult ticks			Clinical signs and microscopical examination of the left parotid lymph node (L1) and blood films											
No. Sex Age	History	Batch No.	History	Fed for days	No. collect.	IP in days	Days after infest.	Temp. °C	L1, degree of enlargement	Koch bodies	<i>T. ovis</i> e.-forms	Remarks					
2476 ♂ 204 days	Splenuctomized; fully susceptible	3317 Ba1	<i>R. bursa</i> LL and NN fed on <i>T. ovis</i> carrier (Table 2)	7-9	7 ♀♀ 9 ♂♂	13	12	39.4	Mild anaemia - anisocytosis, basophilia and polychromasia seen on Day 11 after first appearance of <i>T. ovis</i> and persisted until Day 34; PCV of 30% on Day 1 decreased to 21% on Day 20 and rose to 29% on Day 64				
							13	40.6	+	0	0	0					
							14	40.2	+	0	0	0					
							15	40.4	+	+	+	+					
							16	39.6	+	+	+	+					
							17	38.6	+	+	+	+					
							18	38.6	+	+	+	+					
19	38.9	+	+	+	+												
20	39.0	0	0	0	0	+	+	+	+	+	+						
2191 ♀ 415 days	Splenuctomized; fully susceptible	3349 Aa1	<i>R. e. mimeticus</i> LL and NN fed on <i>T. ovis</i> carrier (Table 2)	7-9	11 ♀♀ 20 ♂♂	13	8	38.5	Mild anaemia - anisocytosis, basophilia and polychromasia seen on Day 11 after first appearance of <i>T. ovis</i> and persisted until Day 26; PCV of 30% on Day 1 decreased to 24% on Day 19 and rose to 29% on Day 33				
							9	38.8	
							10	39.6	+	+	+	+	+	+	+	+	+
							11	38.9	+	+	+	+	+	+	+	+	+
							12	39.5	+	+	+	+	+	+	+	+	+
							13	40.8	+	+	+	+	+	+	+	+	+
							14	40.8	+	+	+	+	+	+	+	+	+
							15	40.6	+	+	+	+	+	+	+	+	+
							16	39.2	+	+	+	+	+	+	+	+	+
							17	38.4	+	+	+	+	+	+	+	+	+
2212 ♀ 404 days	Splenuctomized; fully susceptible	3349 Ca1	<i>R. e. mimeticus</i> LL and NN fed on <i>T. ovis</i> carrier (Table 2)	8-9	5 ♀♀ 16 ♂♂	14	8	38.5	0	.	.	.	Mild anaemia - anisocytosis, basophilia and polychromasia on Day 11 after first appearance of <i>T. ovis</i> and persisted until Day 26; PCV of 30% on Day 1 decreased to 18% on Day 20 and rose to 26% on Day 37				
							9	39.2	+	
							10	39.0	+	+	+	+	+	+	+	+	+
							11	38.9	+	+	+	+	+	+	+	+	+
							12	38.4	+	+	+	+	+	+	+	+	+
							13	39.4	+	+	+	+	+	+	+	+	+
							14	40.4	+	+	+	+	+	+	+	+	+
							15	41.0	+	+	+	+	+	+	+	+	+
							16	40.0	+	+	+	+	+	+	+	+	+
							17	39.0	+	+	+	+	+	+	+	+	+
18	38.4	0	0	0	0	+	+	+	+	+							
19	39.2	0	0	0	0	+	+	+	+	+							
20	38.2	0	0	0	0	+	+	+	+	+							

Legend: LL = Larvae; NN = Nymphae
 IP = Incubation period
 e.-forms = Endoglobular forms
 + = Moderate enlargement
 ++ = Fairly marked enlargement
 +++ = Marked enlargement
 0 = Absent
 . = Not examined
 PCV = Packed cell volume

were opened daily for the collection of detached ticks. After detachment of the females the males were removed and the left lymph nodes were then palpated daily to determine the degree of enlargement. Smears were prepared from the swollen nodes during the febrile reaction as shown in Table 3. Blood smears were prepared daily during and after the reaction period. The films were stained with Giemsa stain and examined for the presence of Koch bodies and endoglobular parasites.

RESULTS

Table 2 gives the duration of the feeding periods of the immature stages, the number of nymphae that fed to repletion on the *T. ovis* carriers and the duration of the nymphal premoult and imaginal prefeeding periods.

The feeding of the ensuing adults of both species on three fully susceptible splenectomized sheep and their response to the infestation are recorded in Table 3. All the sheep developed a febrile reaction which commenced 13 to 15 days after tick attachment and persisted for 4 to 5 days. The parotid lymph nodes showed a distinct swelling for periods varying from 7 to 11 days. Smears prepared from them showed an increased lymphocytic and histiocytic activity and very few Koch bodies on Days 15 to 17 after the *R. bursa* infestation of Sheep 2476 and on Day 15 after the *R. e. mimeticus* attachment on Sheep 2191 and 2212. The endoglobular *T. ovis* parasites were seen 3 days after the appearance of schizonts in the lymph node films. The infection of the erythrocytes was never more than 1.50%. A variable degree of anaemia developed in all the sheep and persisted for several weeks as recorded in Table 3.

DISCUSSION AND CONCLUSIONS

Although larvae of both species readily attached on the ears of the three *T. ovis* carriers, the nymphal harvests were poor in two of them. The reason for this is attributed to the inclination of larvae to attach in large numbers at localized areas at the base and other sites of the inner surface of the ear. As feeding progresses a variable degree of suppuration develops at the attachment sites. Many of the engorged larvae and others in the process of moulting are then smothered by the exudate from the cutaneous lesions.

The transmission of the South African *T. ovis* strain by adults of both species was successful. *R. e. mimeticus* as a vector is established for the first time. Theiler (1962) records this tick species from South West Africa, Botswana, Zaïre and Angola. Her records also show that the proven vector of *T. ovis*, *R. e. evertsi*, is present in the first three named countries but not in Angola. *T. ovis* occurs in Angola (De Mello & Carbal, 1923) and Zaïre (Rodhain, 1916) but up to the present it has not been described from sheep in the potential enzootic regions of South West Africa and Botswana.

The results of the current investigations not only show that *R. bursa* is a vector of *T. ovis* but they also cast doubt on the claim of Rastégaieff (1934) that her biological transmission by this tick species was successful. She infested the ears of an entire sheep with 105 females and 76 males and 3 days later endoglobular *T. ovis* parasites appeared in smears prepared from the peripheral blood circulation. In contrast Jansen &

Neitz (1956) and the current investigations have established that the prepatent period of the endoglobular *T. ovis* parasites following the infestation with three *Rhipicephalus* spp. varied from 17 to 20 days. Consideration of these divergent results suggests that the entire sheep, employed by Rastégaieff (1934), harboured a latent *T. ovis* infection and that the infestation by an extremely large number of *R. bursa* adults resulted in a relapse.

SUMMARY

1. The known vectors of *T. ovis* and the countries, where the transmission by ticks has been conducted, are presented in tabular form.
2. Observations on the current transmission experiments are listed in two tables.
3. *T. ovis* is acquired by the larvae and nymphae of *R. e. mimeticus* and *R. bursa* and transmitted by the ensuing adults.
4. The reaction in all the sheep was accompanied by an increased lymphocyte and histiocyte production in the parotid lymph nodes, a fever and a relatively mild anaemia.
5. Schizonts appeared in smears prepared from lymph nodes and endoglobular forms in blood smears.

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