STUDIES ON SCHISTOSOMIASIS. 6. A FIELD OUTBREAK OF BILHARZIA IN CATTLE

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ABSTRACT

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An outbreak of bilharzia (Schistosoma mattheei) infestation involving about 100 oxen on the farm Otthilie near Tolwe in northern Transvaal is described. Infestation of the cattle appears to have occurred per os from a single drinking trough.

The cattle showed clinical signs and pathological lesions of both the reinfestation and the acute

syndromes. The worm burdens were the highest recorded hitherto in naturally infested cattle. Severe macroscopic clay pipe stem periportal fibrosis, granulomata of the ureters and severe grey pigmentation of the lungs, are described for the first time in naturally infested cattle. There was more marked liver pigmentation than had been encountered previously in this host. A striking feature in every case was conspicuous congestion and enlargement of the ileo-cecal valve, which was dark red in colour.

Treatment of affected animals with injectable trichlorphon controlled the outbreak without causing mass embolism and liver infarcts. At least 11 treatments at a dosage level of 10-12 mg/kg

Faecal examinations for eggs or miracidia cannot be used to assess the worm burden or the clinical state of the animal. Moreover, it is suggested that this disease may be confused with other conditions and this probably accounts for the rarity of reports of outbreaks in ruminants.

INTRODUCTION

Although infestations with Schistosoma mattheei occur in up to 70 to 90% of the cattle in the Transvaal Lowveld (Pitchford, 1958; 1959) and 69,15% of those examined in Rhodesia (Condy, 1960), there are few reports of outbreaks of clinical bilharzia in ruminants, particularly cattle, in southern Africa. Reinecke (1970) described an outbreak in sheep and cattle in Zululand in 1964 and stated that this was the first report of large scale deaths of cattle from this parasite in southern Africa.

In Kenya Dinnik & Dinnik (1965) recovered "many specimens of S. leiperi and S. bovis" from a 3-month-old calf but they did not state whether this animal had died from bilharzia. They also mentioned heavy losses on the farm concerned, but did not specify the species of animal affected. Non-fatal cases of bilharzia in cattle in southern Africa have been described by Veglia & Le Roux (1929), Strydom (1963), McCully & Kruger (unpublished report, 1965), Hurter & Potgieter (1967) and Lawrence & Schwartz (1969).

This paper describes a severe outbreak of bilharzia which occurred in September, 1971 in cattle on the farm Otthilie No. 283 near Tolwe in the northern Transvaal. In addition three lightly infested cattle originating from Amoskuil near Ellisras in northern Transvaal were used for comparing results of treatment and diagnosis with the severely infested cattle from Otthilie. These light infestations of S. mattheei were also acquired in the field.

HISTORY OF THE OUTBREAK

Otthilie

In April or May 1971 the farmer noticed that about 100 head of cattle were losing condition. He found numerous unidentified fresh water snails in the drinking trough of the camp and suspected fascioliasis. The drinking trough was treated with CuSO4 and all the cattle were dosed with hexachlorophene* and moved to another camp.

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Subsequently the 30 oxen in best condition were dosed once per os with trichlorphon† and placed in a feedlot. The remaining cattle were not dosed with trichlorphon nor did they receive supplementary fodder until August, when the natural grazing was in a very poor state. The fodder this group received consisted of poor quality hay.

At the beginning of September 1971, after 10 or 12 oxen had died, the owner treated the remainder with thiabendazole**. When the deaths continued, however, veterinary help was sought and one of us (P. J. G.) performed an autopsy examination on an ox. On finding numerous schistosomes in the liver of this animal, he suspected bilharzia to be the cause of death and informed the other authors.

On the first visit to the farm by the other authors (14th September 1971), an animal that was in extremis was killed and examined post mortem. Seven oxen that were showing clinical signs were transferred to the Veterinary Research Institute, Onderstepoort, for further studies.

Towards the end of September good rains fell on the farm and grazing improved markedly. On 21st October 1971 treatment of the remaining oxen was commenced as described below.

Amoskuil

The oxen examined were part of about 30 transferred to Brits, Transvaal, where they became heavily infested with nematodes. Routine faecal examinations revealed both schistosome and nematode eggs and 4 of the animals were acquired for trials.

CLINICAL SIGNS

Otthilie

The condition of the 30 animals dosed once with trichlorphon before being placed in the feedlot did not improve. Some of them had moderately sunken eyes and their faeces were poorly formed.

The majority of the other cattle were very emaciated and showed severely sunken eyes, diarrhoea, lustreless haircoat, weakness and ataxia (Fig. 1, 2). These signs agree with those described by McCully & Kruger

* Bis 356, Cooper & Nephews † Dylox, Bayer Agrochem ** Bovizole, Merck, Sharpe & Dohme

(1969) in an experimentally infested ox (Van Wyk & Bartsch, 1971; Hussein, 1971, 1973). Some of them, however, exhibited clinical signs similar to those of cattle which were challenged after an immunizing infestation (Van Wyk & Bartsch, 1971): staring, anxious expressions, hypermetria when running and disorientation, while one animal (C3) which escaped at Onderstepoort attacked people outside their house about 1 km away.



FIG. 1 Ox C2, in extremis but still alive and showing marked emaciation



FIG. 2 Ox C2 with the eyes so markedly sunken that the eyelids did not close properly, causing conjunctivitis

Blood cell counts* in some of the oxen revealed varying degrees of anaemia (Table 1).

TABLE 1 Blood cell counts on 16.9.71

Ox	RCC(1)	WCC(2)	
Control Group:			
C1	4 549	11 865	
C3	5 629	5 380	
C4	3 506	6 258	
Treated Group:(3)			
T1	4 732	5 213	
T2	3 779	6 053	
T3	3 610	4 860	

¹⁾ Red cell count in thousands per mm3

Amoskuil

The calves were emaciated, dehydrated and weak, their eyes were sunken and they suffered from diarrhoea, but the clinical signs were ascribable mostly to the heavy nematode infestation.

Pathological Findings

Otthilie

While 1 ox (C2) (Fig. 1, Fig. 2) killed in extremis on the farm and 4 of the 7 cattle moved to Onderstepoort (C1 and C3-C5) served as untreated controls, the 3 others (T1-T3) as well as those remaining on the farm were treated with trichlorphon injectable** (see Treatment below). After treatment T1-T3 and 2 further cattle on the farm (T4 and T5) were killed for worm recovery.

Prior to being killed by stunning (captive bolt or 0,22 calibre rifle) and exsanguination, 150 000 i.u. heparin† was injected intravenously into each animal. The worms were recovered by perfusion by a modification of the method described by McCully & Kruger (1969). The animals were subsequently necropsied and tissue sections prepared by standard histopathological techniques.

Pentobarbital sodium†† was added to the perfusion fluid to aid in the recovery of the worms (Foster, Cheetham & Mesmer, 1968). Subsequent to perfusion more barbiturate was added to the suspension and the worms were killed by heating to 60° C in a waterbath with intermittent vigorous shaking to relax the worms and release females from the gynaecophoric

Pathological findings common to the control and treated groups

With the exception of one animal, C2, (Fig. 1 and 2) which was in extremis prior to necropsy, the untreated animals were variably emaciated but ambulatory. At necropsy the treated animals were in noticeably better condition than the untreated controls.

Marked dehydration and atrophy of skeletal muscles characterized their overall appearance. Mild hydropericardium occurred in all the control animals. The pathological findings in Animal C2 were complicated by traumatic reticulitis and a liver abscess $(10 \times 3 \text{ cm diameter}).$

Granulomas were observed macroscopically throughout the mucosa and serosa of the gastrointestinal tract from the abomasum to the anus. They varied in size from foci 1-2 mm in diameter to areas 15-20 cm long, were grey-white in colour, about 5 mm high and associated with petechiae and ecchymoses. Many perivascular haemorrhages and circumscribed nodules 1-2 cm in diameter were found at the intestinal-mesenteric attachment in assocation with thrombosed mesenteric veins.

Granulomatous lesions of the rectum generally took on a linear appearance; they were up to 10 cm long, 1 cm in width and became polypoid towards the anus. In one case (C5) these polypoid growths were approximately 2 cm long and 5 mm in diameter. The rectal granulomas were associated with petechial and ecchymotic haemorrhages.

White cell count per mm

⁽³⁾ Cell counts before commencement of treatment

^{*} Coulter Electronics Model B Coulter Counter

^{**} Dylox Injectable, Bayer Agrochem

[†] Heparin, 10 000 i.u./ml, Medical and Hospital Supplies †† Sagatal, Maybaker

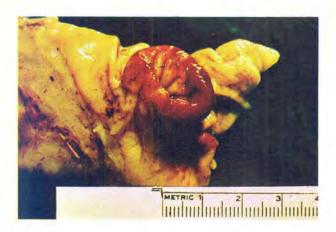
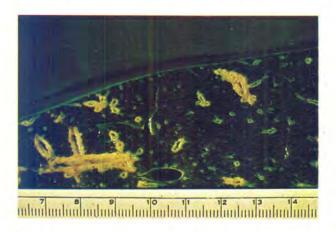




FIG. 3 Ileo-cecal valve demonstrating mild enlargement and haemorrhage

FIG. 4 Another severely enlarged, granulomatous and haemorrhagic ileo-cecal valve. This structure was about 4 cm in diameter and very obvious in protrusion into the proximal colon



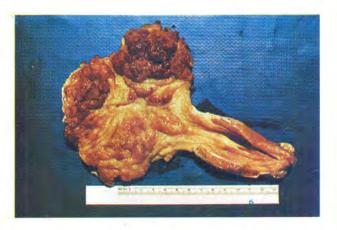


FIG. 5 Transverse section of liver with severe periportal fibrosis especially involving the medium and large-sized portal tracts. There is also obvious grey pigmentation of the parenchyma

FIG. 6 Severely affected urinary bladder. The large areas of polypoid granulomatous patches have become villous and papillomatous in areas

All 10 cases showed conspicuous enlargement and haemorrhage of the *ileo-cecal valve*; it was dark red in colour, up to 4 cm thick and protruded as much as 3 cm into the cecum (Fig. 3 and 4).

All animals revealed some degree of hepatic pathology. The most noticeable lesion was a grey, sometimes almost black discolouration (Fig. 5). White granulomas 1–2 mm in diameter were scattered throughout the liver parenchyma of 2 cases (T3 and C5). Prominent macroscopic periportal fibrosis occurred in 5 of the 10 animals (Controls N1, N3 and N4 and treated T3 and T5). This lesion (Fig. 5) took the form of white fibrous thickening of the large, the medium-sized and to a lesser degree the smaller portal vessels; both macro- and microscopically these resembled Symmers' clay pipe stem fibrosis of human bilharziasis (Symmers, 1904).

There was also a grey discolouration of the *lungs*, which, although very prominent, was moderate compared to the dark hepatic pigmentation.

Granulomas 3 or 4 cm in diameter and about 5 mm high were found in the *gall bladders* of 6 animals (C1, C2, C3, C5, T2 and T5), They were greyish-white in colour and were not associated with haemorrhages.

Macroscopic lesions were seen in the urinary tract of 7 animals (C1, C2, C3, C4, C5, T2 and T3). In the urinary bladders, greyish-white raised granulomata were present either as scattered, discrete foci, 1-2 cm in diameter, or as nearly confluent lesions, involving up to about 80% of the bladder mucosa (Fig. 6). These bladder granulomas were always associated with petechiae and ecchymoses. Except in C4, the ureters of these animals contained granulomatous patches, which were usually scattered throughout the length of the ureter and often involved its entire circumference. The granulomas were also greyishwhite, slightly raised and associated with similar haemorrhages. Small linear granulomas extending several centimetres were also seen in the ureters of these animals. Dilatation of the ureter in areas containing granular patches was common but hydronephrosis was not observed.

Pathological findings peculiar to treated cattle

In the treated animals there was a reduction in the intensity of the granulomatous lesions as well as in the pigmentation of the liver and lungs. The distribution of the granuloma, however, resembled that of the untreated cases.

Thrombi containing partially resorbed parasites were present in the larger portal vessels, but there were no foci of hepatic infarction.

In 3 of the 5 treated animals minute black foci gave the hepatic parenchyma a speckled appearance. Microscopic examinations showed these foci to be large accumulations of haematin.

Comment

A detailed description of the microscopic pathology will be published elsewhere.

Van Wyk & Bartsch (1971) reported *macroscopic* Symmers' fibrosis in experimentally infested cattle but it has not been recorded previously in naturally infested animals other than man. The clinical significance of this periportal fibrosis was not investigated, but it probably contributed to the overall debility of the animals.

The very prominent enlargement and reddening of the ileo-cecal valve (Fig. 3 and 4) found in all of these animals, may serve as a good indicator of the presence of bilharziasis in cattle. While it is uncertain as yet whether this lesion is pathognomonic for bovine bilharzia, it has also been found in all experimentally infested oxen harbouring more than about 4000 schistosomes (Van Wyk & Bartsch, unpublished data). Although this is the first record of the lesion in cattle infested with S. mattheei, it was already noted in 1886 or 1887 in cattle infested with other schistosomes of uncertain identity in India (Bomford, 1886; 1887, cited by Montgomery, 1906). Montgomery (1906) comments as follows: "Bomford found the large intestine to be congested and oedematous notably in the region of the ileo-cecal valve, in fact to such an extent that this structure had the appearance of a dark, cushion-like ring.

The incidence of lesions in the urinary tract was considerably higher in this outbreak than in those described previously (Pitchford, 1958; Hussein, 1968; McCully & Kruger, 1969). In Rhodesia, Condy (1960) found 69,15% of 2 509 cattle infested with bilharzia and states that the main lesions were in the urinary bladder. Unfortunately the incidence of these lesions is not given, but as eggs were demonstrated in only 3% of 1 000 bladders, it was apparently low. This is the first report of bilharzial granulomata in the ureters of cattle. These lesions of the urinary their constant association and haemorrhages suggest that haematuria is probably a common clinical sign of bovine bilharziasis. Wery (1950) reported chronic haematuria in 30 cattle of which 15 were proved to be infested with S. bovis and McCully & Kruger (1969) found mild haematuria in a few cattle infested with S. mattheei.

DIAGNOSIS

Method

Before Oxen C3, C4 and CL8 were killed, miracidial hatching tests (Kruger & Heitmann, 1967) and faecal egg counts (Lawrence, 1970) were carried out.

An indirect fluorescent antibody test for *S. mattheei* infestation (Du Plessis & Van Wyk, 1972) was conducted on the sera of the untreated cattle C1, C3 and C4.

Results

The results of the faecal examinations and the titres obtained on serological examination are presented in Tables 2 and 3.

Ox C3, which had 24 732 female schistosomes in the mesentery, had an egg count of 35 eggs per gram (e.p.g.) of faeces while that of C4, with 23 818 female schistosomes, was 77 e.p.g. (Table 2). Ox CL8, with only 1 172 females in the mesentery (Table 3), passed 300 e.p.g. of faeces.

The antibody titre for these animals, determined by the indirect immunofluorescent method, varied from 1/810-1/7 290.

Comment

Ox CL8 was in extremis from heavy nematode infestation and suspected babesiosis when the faecal sample was taken and probably passed only a small amount of faeces. This may account in part for the relatively high egg count compared to those of the more heavily infested animals. In another investigation, however, cattle harbouring 4 000–6 000 female schistosomes in the mesentery after a single experimental infestation passed more than 1 000 e.p.g. of unformed faeces at the peak of egg production (Van Wyk, unpublished data). In cattle, therefore, there is apparently no correlation between the schistosome burden and the faecal worm egg count.

The total number of eggs voided per day would probably be a more reliable indicator of the shistosome burden but this is difficult to determine in cattle under field conditions, especially when diarrhoea is present (Lawrence, 1973b).

These findings were to be expected from the results of other workers. Lawrence (1973a, Fig. 1) showed that in cattle infested with a single dose of cercariae the schistosome egg production dropped rapidly after the peak concentration of eggs in the faeces had been reached. Twenty five weeks after infestation the total number of schistosomes recovered from these cattle had dropped by only about 30-35% (in comparison with maximum worm development percentage at ±7 weeks), whereas the reduction in e.p.g. count (compared to peak production) was more than 75%. At 40 weeks 50% of the original worm burden remained but the egg concentration in the faeces was only 5% of the peak concentration. Apparently the decrease in worm burden was not due to a disproportionate decrease in female parasites because the author concluded that the lower egg concentration "must be largely caused by a reduction in egg production by the female schistosome". Reinfestation of some of these cattle did not result in "any increase in egg count" in 3 out of 4 animals examined despite the fact that the percentage of worms that developed following the reinfestation was comparable to that of the primary infestation. Moreover, Hussein, Saeed & Nelson (1970) reported that the schistosomes of a challenge dose in cattle previously immunized with heterologous schistosome cercariae produced markedly fewer eggs per female worm than those of a primary infestation.

DIFFERENTIAL DIAGNOSIS

Various diseases of cattle may be confused with the bilharzia syndrome described by Bomford (1886; 1887, cited by Montgomery, 1906), McCully & Kruger (1969), and Hussein (1971; 1973). Bomford killed 2 oxen suffering from bilharzia because he suspected rinderpest. Reinecke (1970) stressed the importance of differentiating bilharzia from nagana, while other conditions such as severe coccidiosis, arsenical poisoning and Johne's disease also have clinical signs similar to those found in bilharzia.

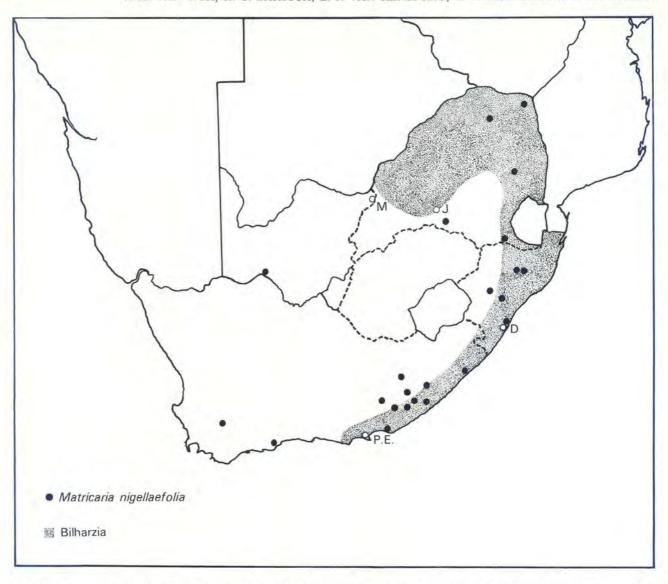


FIG. 7 The distribution of bilharzia* (from Van Eeden & Combrinck, 1966) and Matricaria nigellaefolia (reprinted with the permission of the Director, Botanical Research Institute, Pretoria) in the Republic of South Africa. Key:
P.E. — Port Elizabeth, D — Durban, J — Johannesburg, M — Mafeking

* Distribution of Bulinus (Ph.) spp. snails

The reinfestation syndrome of bovine bilharzia (Van Wyk & Bartsch, 1971) may be confused with poisoning caused by the plant *Matricaria nigellae-folia* (Staggers weed or, in Afrikaans, "Stootsiektebossie"; Steyn, 1949), the geographical distribution of which closely resembles that of bilharzia in South Africa (Fig. 7).

The following clinical signs occur in both these conditions: severe ataxia and weakness with knuckling over the fetlocks; aimless wandering around; "staggers" and a tendency to lean against solid objects, e.g. fences; marked emaciation; gnashing of the teeth; salivation and constipation.

In the bilharzia reinfestation syndrome various other signs which differentiate it from *Matricaria* poisoning, are also seen, such as sunken eyes; a staring, anxious expression, alternating with a sleepy appearance without constriction of the pupils; marked intermittent straining and hypermetria. Moreover, the bilharzia case shows less tendency to lean against solid objects.

TREATMENT

Otthilie. As described above, Oxen C1-C5 from Otthilie served as untreated controls while T1-T3 were treated in the laboratory. Treatment consisted of a 50% trichlorphon solution administered intramuscularly (i.m.) at 3-4 day intervals as follows:

Ox T1: Three injections of 4 ml (8 mg/kg); 2 of 6 ml (12 mg/kg); 2 of 8 ml (16 mg/kg) and 1 of 10 ml (20 mg/kg).

Ox T2: Eight injections of 8 ml, i.e. 16 mg/kg. Ox T3: Eleven injections of 4 ml, i.e. 8 mg/kg.

Ox T1 was killed in extremis 4 days after the last treatment while T2 and T3 were killed 23 and 14 weeks respectively after treatment.

The farmer treated the rest of the herd with 6 ml trichlorphon per injection (10-15 mg/kg) on 8 occasions at intervals of 4-6 days. On completion of this treatment, 2 animals (T4 and T5) of the small number in poor condition were killed for worm recovery.

TABLE 2 Otthilie: Treatment and diagnosis in cattle heavily infested with S. mattheei

Ox lotal in	Females	Total		Worm distribution			Treatment (trichlor-	**	t	
	mesentery	Females	Males	Mes.	Liver	Lungs	phon mg/kg)	Titre	E.P.G.	
Control Group:				12-1	%	0/2	0/0			
C1	64 434†	22 347	27 542	36 892	80,3	2,6	17,2		1/7290	İ
C2	41 654*	14 622	19 256	22 398	74,3	2,7	23,0	-	† †	-
C3	56 833†	24 732	26 873	29 960	93,4	0,8	5,8	-	1/2430	35 77
C4	54 603†	23 818	24 698	29 905	96.4	2,8	0,8	_	1/810	77
C3	71 083*	28 696	31 795	39 288	90,0	2,6	7,4	_	_	-
Mean	57 721	22 843	_			-	_		_	-
reated (Lab.):										
T1	15 896*	513	9 007	6 889	6,3	90,8	3,0	8-20 (×8)		1
T2	24 751*	10 633	12 822	11 929	88,0	3,4	8,5	16 (×8)		-
T3	1 517	685	840	677	89,4	0,7	9,9	7 (×11)	_	_
Mean	14 055	3 944	-	-	61,2	31,7	7,1	_	1	
Treated (Farm):		76.57				132	1 1	100000		
T4	1 815*	17	528	1 287	2,8	97,2	0,0	10-15	-	_
		1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1				1 3 4 5 1	2.0	(×8)		
T5	46 342†	8 972	21 530	24 812	42,1	52,2	5,7	10-15	-	_
20-0		70.00	100000	100	100	0.0	10.6	(×8)		15
Mean	24 079	4 495	-	_	22,4	74,7	2,9		-	

Total counts

Estimated from counts of 12 aliquots From Du Plessis & Van Wyk (1972) Eggs per gram of faeces

†† Not tested

Total counts were done on the worms recovered from 6 head of cattle (C2, C5, T1, T2, T3, and T4), while the worm burdens of the other 4 were estimated by counting the schistosomes in aliquots (Appendix Table 1).

Amoskuil. One of 4 oxen from Amoskuil was given 11 treatments of trichlorphon at 11,5 mg/kg i.m. at intervals of 3 or 4 days while the other 3 served as untreated controls.

Results

Otthile. The maximum number of S. mattheei recovered from any animal was 71 083 (C5, Table 2). In the control group the mean number of schistosomes was 57 721, of which 22 843 were females in the mesentery. In the cattle treated at the Institute there was a mean of 14 055, of which 3 954 were females in the mesenteric veins. The 2 animals (T4 and T5) examined after treatment on the farm had a total of 46 342 and 1 815 schistosomes respectively.

The treatment of T1 was designed to test the maximum dose the animal would tolerate in its weakened state. The final dose (10 ml) was higher than the maximum recommended by the manufacturer for a single treatment of cattle and caused an adverse reaction. Muscle fasciculations, sialosis and inco-ordination were soon noticed and later decubitus developed. Atropine therapy failed to relieve the condition and the animal was killed when in extremis 3 days after the final dose of trichlorphon.

Ox T2 received the maximum dose (8 ml) per injection recommended by the manufacturer for a single treatment. The first 4 treatments had a marked effect on the animal: diarrhoea became projectile and anorexia, inco-ordination and ataxia developed. Its mass (Fig. 8) fell during treatment but rose sharply thereafter.

Ox T3 was kept under observation for 9 weeks before treatment was commenced. During this period its mass remained constant but it decreased when treatment started, then increased again while therapy was still in progress. After treatment its increase in mass closely parallelled that of T2 (Fig. 8).

No further mortalities occurred on the farm after treatment had commenced. The farm was not revisited subsequent to a second visit immediately after treatment had terminated but apparently the herd improved so much that all but a few animals were sold after 4 months. The few that did not improve after the initial treatment were treated again and were apparently in good condition when they were slaughtered at a later date.

Amoskuil. The mean number of schistosomes from the 3 controls was 2 660, 1 234 of which were mature females in the mesentery (Table 3). The treated ox harboured 1 055 worms, of which 323 were mature female from the mesentery; the percentage reduction was therefore 60,4% (total worms) or 69,4% (females in the mesentery).

TABLE 3 Worm burdens of control and treated cattle from Amoskuil

Ox	Total worms*	Females in mesen- tery	Males in mesen- tery	Total females	Total males
Control:	100		2.32	C.ET	
CL 6	3 457	1 578	1 533	1 729	1 728
CL 7	2 145	952	966	1 147	998
CL 8	2 3 7 9	1 172	1 183	1 195	1 184
Control mean	2 660	1 234	1 227	1 357	1 303
Treated TL 6	1 055	323	287	739	316

^{*} S. mattheei

Comment

Numerous drugs have been shown to be very effective against bilharzia in ruminants. Hurter & Potgieter (1967) and Reinecke (personal communication, 1973) showed that niridazole* was very effective in sheep at a dosage rate of 100 mg/kg for 3 days. Hycanthone** was found to be effective in

^{*} Ambilhar, Ciba ** Etrenol, Winthrop

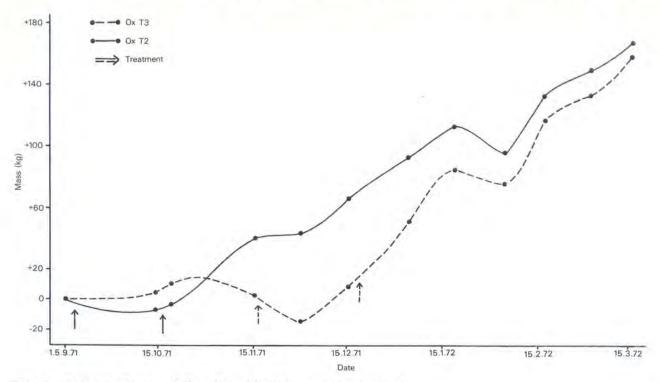


FIG. 8 Changes in the mass of Oxen T2 and T3 before and after treatment

sheep (Lawrence & McKenzie, 1970) and Stibophen*** in cattle (Lawrence & Schwartz, 1969). McCully & Kruger (1969) treated cattle and sheep with tartar emetic.

With the possible exception of stibophen, these drugs are all highly effective against schistosomes, which are killed *en masse*, and the resulting emboli cause portal occlusion and focal hepatic infarction (McCully & Kruger, 1969). The magnitude of this process is better understood if one considers the size of the worms acting as emboli and the large numbers of parasites present in the mesenteric venous system. Reinecke (1970) stated: "The indiscriminate treatment of infested animals may produce more serious consequences than the disease itself".

Reinecke (unpublished report, 1964) expressed the opinion that animals heavily infested with bilharzia can be treated successfully only if the worms are killed over an extended period, thus enabling the animal to cope with the gradual accumulation of dead worms. Trichlorphon was therefore tested to gauge its effect in this respect.

Medda (1969) and Medda, Reinhardt & Muscas (1969) obtained good results in sheep and goats with 4 or more oral treatments alternately of 100 and 120 mg/kg trichlorphon given 3 or 4 days apart. Dinnik (1967) reported good results in 5 head of cattle infested with S. bovis, treated with trichlorphon at a dosage rate of 50 or 75 mg/kg per os for 4 to 6 treatments. One animal that was slaughtered had only 9 parasites, but there were apparently no untreated controls. This work was repeated by Lawrence & Schwartz (1969) with "catastrophic" results as 2 of 5 cattle died after the second treatment. The disparity in results was possibly due to differences in absorption of the orally administered drug as these authors did not starve the cattle or withdraw concentrates.

The parenteral preparation of trichlorphon used in the present investigation has the advantage that absorption is not dependent on the diet of the animal or physiological conditions of the alimentary tract. The maximum tolerated blood concentration of drug is therefore more easily and safely obtained.

The animals used in these trials were apparently more heavily infested than cattle treated by other workers. Lawrence & Schwartz (1969) counted more than 1 000 parasites in an untreated control, but used an *in situ* counting technique which may not have accounted for all the worms actually present. Dinnik (1967), on the other hand, did not have untreated control cattle.

This trial shows that the drug has an antischistosomal effect. The effect in Ox T3 which received 11 treatments at a low dosage rate, was more favourable than in Oxen T1 and T2, which received 8 treatments at higher dosage levels. This response could be due to individual variation but demonstrates that the relatively low dose can be effective.

The numbers of worms recovered from Oxen T4 and T5, that were treated on the farm, differ considerably. Unfortunately the treatment could not be supervised, with obvious implications. The fact that the other animals recovered within 4 months suggests, however, that the treatment was effective on a herd basis and that most animals were probably treated consistently.

EPIZOOTIOLOGY

Otthilie

A river, the Mogalakwena, which is known to be infested with bilharzia (Hanford, personal communication, 1972) passes through the farm on which the outbreak occurred. Although the river is fenced and the oxen did not have access to it during the year preceding the outbreak, they were supplied with water pumped from under the sand in the river bed.

^{***} Fantorin, Glaxo-Allenbury's

Method and locus of infestation

There is only one drinking trough in the camp and, according to the farmer, there are no pans which

contain water for any length of time.

At the time of the investigation on the farm, a few live *Bulinus (Physopsis)* spp. snails, the intermediate host of *S. mattheei*, were found in the drinking trough (Fig. 9). The farmer had, however, treated the trough shortly before with CuSO₄. There were many shells of these snails in the trough and the reservoir that supplied it contained extremely large numbers of the snails.



FIG. 9 The drinking trough where the cattle apparently became infested

It would appear, therefore, that the infestation had its origin in the drinking trough. Since this trough is relatively deep and not one into which cattle would climb (Fig. 9), the infestation probably occurred per os. Fairley & Jasudasan (1929), however, only recovered 290 schistosomes from a goat that had been dosed with 98 000 cercariae of Schistosoma spindale intraruminally 86 days previously. The cercarial suspension, 80 to 120 ml in volume, was dosed in 4 daily portions with a stomach tube. On the other hand, massive infestations developed when the suspension was dropped slowly onto the tongue and buccal mucous membranes of 2 goats, which died before the worms became patent. These observations were confirmed by Reinecke & Kruger (personal communication, 1973), who recovered only 4 S. matheei from a sheep infested intra-ruminally with 15 000 cercariae, while sheep infested percutaneously became heavily infested.

If these findings hold true under field conditions, most of the cercariae must have penetrated the skin or mucous membranes (e.g. lips and chin) before reaching the rumen. In this case the fact that more than 71 000 worms developed in a single animal indicates an extremely high concentration of cercariae

in the trough.

Another possibility which requires investigation is the effect of dilution of the ruminal contents of cattle or sheep on intraruminal infestation. From the work of Reinecke & Kruger (personal communication, 1973) and Fairley & Jasudasan (1929), who infested sheep and goats intraruminally with small volumes of cercarial suspension and found that only very few cercariae (0,03 and 0,3% respectively) developed, it appears that the ruminal fluid adversely affects the cercariae. Under prevailing extensive farming conditions in the northern Transvaal cattle usually drink

only once a day, when each consumes a large volume of water, which may dilute the ruminal contents to such an extent that the penetration of cercariae is facilitated*.

A massive build-up of cercariae is theoretically possible under the prevailing conditions on the farm. The water level in the drinking trough is regulated with a ball valve and the snails in it are not therefore subjected to floods such as occur in streams. Under these conditions the snail population would increase rapidly in summer until the maximum number that could be sustained by the available food and water supply in the trough is reached.

If it is changed frequently 51 water is sufficient to maintain more than 100 snails (Van Wyk, unpublished data). Since the capacity of the trough is approximately 1 000 1 and 100 oxen drink more than 4 000 1 water a day, the water in the trough would be replaced 4 times a day. In addition the growth of algae provides an adequate food supply. Theoretically, therefore, this trough could support at least 20 000 snails at any one time and, if one lightly infested animal should defaecate in it, several thousand snails could become infested.

Under outdoor conditions in summer a single snail may "shed" 1000 S. mattheei cercariae daily between 06h00 and 08h00 (Pitchford, Meyling, Meyling & Du Toit, 1969). If there are 4000 infested snails in the trough, about 4 million cercariae would be available, concentrated under the surface of the water each morning to infest the animals that drink there.

In the other camps on Otthilie no snails were found in the drinking troughs, which are supplied with water from boreholes.

Since the development of the snail intermediate host, as well as that of the miracidium to the cercaria, is retarded in autumn and winter (Pitchford & Visser, 1965), a build-up of cercariae is possible during the warm summer months only (Pitchford, 1963). When cattle are infested experimentally with a lethal single infestation of cercariae, they show no clinical signs before Day 45 after infestation, i.e. 10–14 days before fatalities may occur (Van Wyk, unpublished data). Furthermore, when such an infestation is preceded by a small immunizing infestation, clinical signs do not occur before 100 days (Van Wyk, unpublished data). These oxen were losing condition in April or May, therefore they were probably infested not later than in February or March, 1971.

Nevertheless, a few cattle showed both clinical signs and *post mortem* lesions of the multiple infestation syndrome (Van Wyk & Bartsch, 1971), suggesting that infestation took place even earlier, i.e. in about December 1970. Some of the oxen, infested with as many as 71 083 schistosomes, therefore survived at least 6 months, and probably up to 9 months, on very poor grazing.

^{*} This possibility has subsequently been examined in sheep (Van Wyk, unpublished data) and 43,5 per cent cercariae developed in a sheep receiving a large volume of water to dilute the ruminal contents before intraruminal infestation compared to 13,6 per cent in similar infestation without dilution and 72,2 per cent in percutaneous infestation.

CONTROL

Strydom (1963) outlined the control of schistosomiasis in farm animals. On Otthilie cattle do not have access to the river and the control of bilharzia is therefore neither complicated nor expensive. Snails or their egg packets will rarely be pumped into the reservoir and drinking trough because the water is pumped from under the sand of the river bed. Hence infrequent, but regular, treatment of the reservoir and drinking trough with molluscicides should prevent accumulation of the snails and thus another outbreak of bilharzia.

DISCUSSION AND CONCLUSIONS

More schistosomes were recovered from cattle in this outbreak than have been reported previously. This may account for the fact that some pathological changes, e.g. *macroscopic* clay pipe stem periportal fibrosis and granulomas of the ureters, are described for the first time in cattle.

The clinical signs and pathological changes, including the macroscopic Symmers' fibrosis, associated with reinfestation of immune cattle was unknown until this syndrome was produced experimentally (Van Wyk & Bartsch, 1971). At the time it appeared improbable that cattle would be exposed to infestation of the magnitude required to produce the syndrome under field conditions, but this outbreak confirmed that large numbers of parasites are required to produce it and, moreover, showed that it can develop in the field.

Such a concentration of cercariae is probably rare but it is possible that similar cases were missed in the past for various reasons. Firstly, the syndrome resembles other conditions which produce staggers (e.g. Matricaria plant poisoning). Secondly, post mortem diagnosis is complicated by a high incidence of bilharzia in animals in enzootic areas, difficulties associated with estimation of the schistosome burden ante- or post mortem, entrapment of worms in blood clots and the similarity between post mortem pseudomelanosis and the grey pigmentation associated with bilharziasis.

The absence of mass embolism and large scale hepatic infarction after treatment of these heavily infested cattle indicates that trichlorphon had the desired effect, in that worms died slowly and thus enabled the body to become adapted to a steady accumulation of dead parasites. Despite the poor results obtained in one animal treated on the farm, the drug appears to have been effective in controlling the outbreak and greatly reduced the numbers of worms. At least 11 parenteral treatments of trichlorphon should be given to cattle at a dosage of 10 to 12 mg/kg treatment i.m. at 3–5 day intervals. Such prolonged treatment is a distinct disadvantage but the drug is very cheap and with the correct control measures it should not be necessary to give more than one series of treatments on any one farm.

This investigation shows that there is at present no method to determine the numbers of bilharzia worms ante mortem in cattle. It is well known that there is no correlation between schistosome numbers and the titre determined by the immunological tests (Sadun, 1967; Du Plessis & Van Wyk, 1972; Smithers, 1973) and in this outbreak faecal examinations for eggs and miracidia proved equally ineffective. Post mortem worm recovery is accurate, but time consuming and therefore of little value in extensive surveys. Moreover, this type of survey is done at

abattoirs, where animals in good condition are slaughtered, and the heavily infested animal is thus not encountered. Tests for worm metabolic products in the serum or excreta of the host (Supronova, Soprunov & Lur'e, 1973) may offer a solution.

The prevailing conditions on the farm suggest that in cattle *per os* infestation is possible and can result in extremely heavy worm burdens.

In this outbreak on Otthilie the bilharzia was uncomplicated by other disease conditions. As with other internal parasitic infestations, it is probable that the most frequent and important effect of the schistosome parasite at subclinical to subacute levels of infestation is a temporarily decreased growth rate and lowered resistance to other diseases. In a bilharzia enzootic area, therefore, it is important that schistosome should be specifically sought as a possible complicating factor in outbreaks of other diseases.

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APPENDIX

A method for estimating numbers of adult schistosomes. The schistosomes are placed in a 21 Erlenmeyer flask with indented sides (Fig. 10) and the suspension made up to 1 500 ml volume.

The flask is swirled vigorously and, while the contents are still well agitated, an aliquot of 15 to 100 ml is decanted into a 100 ml measuring cylinder, fitted with a funnel to prevent spillage. This process is repeated until about 20 to 30% of the suspension has been sampled. Thereafter the volume of each aliquot is recorded and the schistosomes in each counted.

This method was tested on 2 suspensions containing known numbers of adult *S. matheei* schistosomes (Appendix Table 1, 2). In Suspension 1 (Appendix Table 1) the total number of schistosomes was placed in the flask and 10 aliquots decanted. The volume decanted was 30% of the total. In Suspension 2 the schistosomes were too numerous for 1 flask and were therefore divided into 2 portions, each of which was sampled separately until 20% of it had been examined.

APPENDIX TABLE 1 Suspension 1

		Estimates			
	Actual total	Aliquot 30% of total	Aliquot 20% of total	Aliquot 15% of total	
Males	15 511 15 041	+7,69* +3,78*	+10,44 +6,99	+8,77 +6,22	
Total	30 552	+5,81*	+8,77	+7,54	

^{*} Percentage difference between actual and estimated total



FIG. 10 A 21 flask with indented sides for mixing the worm

APPENDIX TABLE 2 Suspension 2

	Actual total	Estimate (20% aliquot)		
		Number of worms	Percentage difference	
Males	34 431 28 311	36 295 30 157	+5,41 +6,13	
Total	62 742	66 452	+5,59	

Results (Appendix, Tables 1 en 2)

In both cases the estimated total was higher than the true count for both male and female worms. In Suspension 1 2 out of the 10 estimates (male worms) and 3 out of 10 (females) were lower than the true count, while the rest were higher. The totals estimated (30% total aliquots) were 7,69% and 3,78% too high respectively for the males and females.

In Suspension 2 these estimates (20% aliquot) were respectively 5,41% and 6,13% too high.

Comment

In the past few workers recovered so many schistosomes from animals that total counts were impractical. Lawrence (1973a) infested calves with 5 000 to 45 000 cercariae and recovered up to 60% after their development to adults, but he does not mention the method he used to count them.

The schistotomes are relatively large and dense and hence difficult to suspend for taking aliquots. In order to improve turbulence the sides of the Erlenmeyer flask used for mixing were indented but it did not overcome this problem. Nevertheless, the errors obtained were relatively small and the method is of value especially in field outbreaks where worm burdens usually vary considerably.

More work is clearly indicated; possibly a suspending fluid with a S.G. higher than water will improve distribution of the worms during mixing and hence estimates by the aliquot method.

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