



Some epidemiological and economic aspects of a bluetongue-like disease in cattle in South Africa—1995/96 and 1997

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ABSTRACT

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In December 1995 to March 1996 and the early summer of 1997 South Africa experienced above average rainfall which favoured the occurrence of *Culicoides* transmitted diseases. During this period several outbreaks of an uncommon disease of cattle occurred over a large part of the country. The clinical signs were similar to those of infection with the viruses of bluetongue (BT) and epizootic haemorrhagic disease of deer (EHD). Virus isolation from cattle and *Culicoides* yielded both viruses. Dual infections occurred on several farms. Typing of BT isolates yielded types 2, 3, 6 and 8. On at least two farms more than one BT virus serotype was involved. On one farm only EHD virus could be isolated from cattle and *Culicoides*. Serological tests confirmed that on this farm the disease was caused by EHD. In 1932/33, when a similar disease was reported conditions were vastly different. Rainfall figures show that the 1932/33 season was exceptionally dry. Techniques available at that time could not identify EHD and the cause was reported to be BT. The occurrence of BT in a dry season and over a much wider area than the distribution in South Africa of *Culicoides imicola*, the only proven vector for BT, is a clear indication that other species less dependent on high rainfall are involved. The present isolation of BT virus from three of five pools of parous *C. bolitinos* is evidence that this species, which breeds in cattle dung, may be an additional vector for BT.

Keywords: Bluetongue, bluetongue-like disease, *Culicoides*, epizootic haemorrhagic disease of deer, South Africa

INTRODUCTION

In December 1995 to March 1996 and in the early summer of 1997 an uncommon disease of cattle occurred in South Africa. The clinical signs (Gerdes, Nesser, Barnard & Larsen 1996) were similar to those of infection with the viruses of bluetongue (BT) (Metcalf & Luedke 1979) and epizootic haemorrhagic disease of deer (EHD) (Metcalf, Luedke & Jochim 1991).

Bluetongue, a disease of domestic and wild ruminants, is seen mostly in sheep. Clinical disease is rarely seen in cattle which act as reservoirs and amplifiers of the virus (Metcalf & Luedke 1979). Distribution of the disease is dependent on the presence of suitable species of *Culicoides* in large enough numbers and is therefore most prevalent during the summer months, particularly in wet seasons (Du Toit 1944).

In 1959/60 during a bluetongue-like epizootic among cattle in Japan a virus was isolated and named Ibaraki virus (Inaba 1975). In subsequent studies this virus was shown to be identical to epizootic haemorrhagic disease of deer (EHD2) virus (Campbell, Barber & Jochim 1978; Campbell & St George 1986). Although the Ibaraki strain of EHD was only retrospectively confirmed to be an EHD virus, the Japanese epizootic in 1949–1951 (Omori 1970) is now recognized as being the first description of EHD infection

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in cattle. The disease, which primarily affects deer, was first described in 1955 in New Jersey in the United States of America (Shope, MacNamara & Mangold 1955, 1960). It has been isolated on a number of occasions from the blood of cattle convalescing from disease and the serological evidence of EHD infection in the absence of BT resulted in a presumptive diagnosis of an EHD epizootic (Metcalf *et al.* 1991). Despite the extensive epizootic in Japan (Inaba 1975) and serological evidence of a worldwide occurrence of the virus (Gumm, Taylor, Roach, Alexander, Greiner & Gibbs 1984; Campbell & St. George 1986) experimental infection of cattle with EHD virus has usually failed to produce clinical disease (Uren 1986).

In South Africa, both viruses have been isolated on occasion from single cases of cattle suffering from a BT-like disease as well as from clinically normal cattle (B.J.H. Barnard, unpublished data 1984) and from *Culicoides* (Nevill, Erasmus & Venter 1991).

This investigation reports on aspects of an epizootic in South Africa in 1995/96 and 1997 and compares some epidemiological features with those of a similar outbreak in 1932/33 (Bekker, De Kock & Quinlan 1934).

MATERIAL AND METHODS

The outbreak

In the summer of 1995/96 outbreaks were recorded over a large part of the country (Fig. 1). Cattle of all ages and all breeds on affected farms were involved. In February 1996, outbreaks on three farms in the Delareyville District in the North West Province were investigated and in January 1997, a localized outbreak in Gauteng was studied in a herd of 60 calves on the farm Donkerhoek in the Bronkhorstspuit District. Twenty-four randomly selected 5 to 8-month-old calves were examined on this farm on two occasions, 28 d apart. Additional information and samples were obtained from veterinarians. Information was also obtained by means of a questionnaire sent to farmers.

Samples

Seventy-two blood and/or organ samples were received for routine virus isolation. The history and clinical signs reported by senders suggested that 13 of the samples were collected from cattle suffering from other conditions. Although isolation of BT or EHD viruses from these 13 samples was regarded as unlikely, they were processed in exactly the same way.

Virus isolation and identification

Standard laboratory procedures were used to process the samples after which preparations were in-

jected intravenously into embryonating chicken eggs, intracerebrally into infant mice and inoculated on to CER cell monolayers. When deemed necessary this was followed by further passages in eggs, mice or on CER cell cultures. Virus isolation from *Culicoides* was done on VERO cell monolayers. Isolated viruses were identified by complement fixation (CF) and BT virus isolates were typed by plaque reduction.

Serology

Presence of group specific antibodies against BT and EHD viruses in cattle shortly after the onset of the disease was tested for by ELISA and CF, respectively. Results for BT virus were compared with results of routine samples of cattle from the same area tested at irregular times after the occurrence of the outbreak.

Milk production

Milk production records of one herd in Delareyville were obtained from the owner, and a milk-processing firm provided records for different regions. On request of the firm, the figures of the volume of milk handled are not shown.

Culicoides

During the outbreaks *Culicoides* were caught in light traps during three nights on the Delareyville farms and during two nights at Donkerhoek. In addition, collections were made on Kaalplaas, a farm adjoining the Onderstepoort Veterinary Institute (OVI) where no cases of the disease had been observed. The catches were sorted into species and divided into pools of parous females for virus isolation.

Rainfall

Rainfall figures for the periods before and during the outbreaks of four widely separated localities in the affected area were obtained from the South African Weather Bureau.

Information on the 1932/33 outbreak

Data for the 1932/33 outbreak, with the exception of climatic data, are those published previously (Bekker *et al.* 1934)

RESULTS

Affected area

In early summer (November 1995) the first cases of an isolated outbreak in the northern part of the country were reported (Fig. 1). In December and January the disease erupted on numerous farms over an extensive area. The incidence increased sharply,

reached a peak in February/March and declined towards the end of April. In the summer of 1997 a few isolated outbreaks occurred in the same area. The vast majority of cases occurred in the central part of the country in an area of approximately 32000 km². The 1932/33 outbreak occurred in the same general area (Fig. 1).

Morbidity

The morbidity based on clinical signs observed by farmers, varied from less than 1% to 10%.

The clinical signs which have been described previously (Gerdes, Neser, Barnard & Larsen, 1996) are

compatible with those of BT in sheep. According to the owner of the calf herd six of the 60 calves showed signs of the disease. However, on closer examination of 24 randomly selected calves it became clear that the signs may be inconspicuous and could only be seen after careful inspection. Lacrimation occurred in a number of them. Rectal temperatures ranged from 39,5–41,0°C, and approximately 10% were lame. Inspection of the feet of lame calves and also many of those that appeared normal revealed a coronitis; the area of skin immediately above the dew claw was red, swollen and hot to the touch. A few 1–2 mm focal haemorrhages could be seen in unpigmented coronary bands of both Jersey and Holstein calves. Ulcers were present on the muzzle,



FIG. 1 Areas in which outbreaks of a BT/EHD-like disease occurred in South Africa in 1932/33 and in 1995/96 and 1997

lips and/or gums of almost all calves. The ulcers ranged in size from being just visible to 10 mm in diameter. In two calves confluent ulcers covered the entire upper gum. Necrosis was seen in the unpigmented skin of a single calf. When the calves were examined 28 d later, some of the lesions were still visible.

In addition to lesions, a decrease in milk yield was a common feature in all affected dairy herds. The drop in milk production in a herd from which both BT and EHD viruses were isolated is shown in Fig. 2. Milk production dropped 42%, from 560 kg to 323 kg within days after the appearance of the first cases. One month later the production was still well below the level of production before the onset of the disease. Records from a milk-processing firm showed similar decreases in areas from which cases had been reported. The fall in affected areas was markedly more than in areas from which no clinical cases were reported.

Viruses isolated from cattle

Twenty-one (36%) of 59 samples from cattle judged to be infected yielded virus while no virus was isolated from the 13 samples originating from cattle manifesting clinical signs suggestive of other conditions (Table 1). Bluetongue virus types 2 and 8 were isolated from four samples each and single isolations were made of BT3 and BT6. EHD was isolated from 9 samples. Two BT virus isolates and the EHD virus isolates were not typed. In two instances both BT and EHD viruses were isolated from cattle on the same farm and both were recovered from *Culicoides*. In one instance on a farm in the Delareyville District, BT2 and EHD viruses were isolated from cattle as well as from midges. The different viruses were found in widely separated places (Fig. 1).

Viruses isolated from *Culicoides*

Eleven *Culicoides* spp. were collected (Table 2). Affected farms yielded two BT virus isolates, one from *C. imicola* and one from *C. bolitinos*, while EHD virus was isolated from *C. nevillei* and *C. cornutus* collected on two different farms. Both BT and EHD viruses were isolated from midges in the Delareyville District. Only EHD virus could be isolated from Donkerhoek. *C. imicola* and *C. bolitinos* from Kaalplaas, where no cases of the disease had been detected, yielded 12 and two isolates of BT virus, respectively.

Serology

Serological tests (Table 3) on serum samples, collected from affected cattle at the onset of the disease, showed that at the start of the outbreak 22% and 92% were negative for BT and EHD, respectively. After the outbreak 126 (84%) of 149 routinely tested serum samples originating from the affected area tested positive for BT.

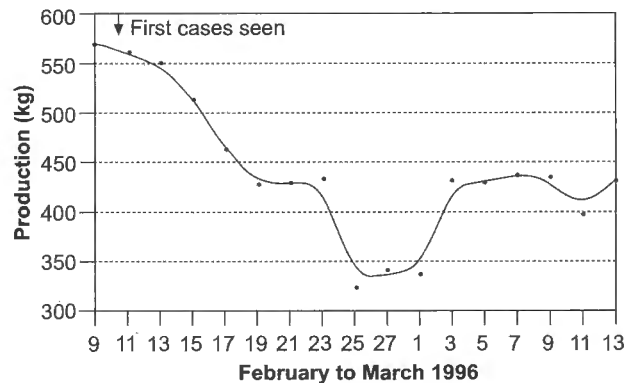


FIG. 2 Milk production in a herd in the Delareyville District from which BT and EHD viruses were isolated

TABLE 1 Viruses isolated from cattle with and without signs of BT/EHD and from *Culicoides* collected during the outbreak—South Africa, 1995/96 and 1997

Origin of sample	Virus isolated from cattle with/without signs of disease				Virus isolated from <i>Culicoides</i>	
	With			Without	BT	EHD
	BT	EHD	Any			
	Serotype	Number	Number	Number	Number	Number
Delareyville	2	2	2	0	2	2
	8	1	—	—	—	—
Donkerhoek	0	0	1	0	0	1
Kaalplaas	—	—	0	0	14	0
Various	2	2	—	—	—	—
	3	1	—	—	—	—
	6	1	—	—	—	—
	8	3	—	—	—	—
	NT ^a	2	6	—	—	—
Positive/tested	—	12/59	9/59	0/13	—	—

^a Serotype not determined

TABLE 2 *Culicoides* collected and viruses isolated from parous females caught during an outbreak of a BT/EHD-like disease in cattle in South Africa—1995/96 and 1997

<i>Culicoides</i> species	Virus isolated from <i>Culicoides</i> collected on farms with and without affected cattle					
	With signs of BT/EHD-like disease				Without signs	
	Donkerhoek		Delareyville		Kaalplaas	
	Virus isolated	Number of midges and pools	Virus isolated	Number of midges and pools	Virus isolated	Number of midges and pools
<i>imicola</i>	0	141 (2)	BT x1	200 (10)	BT x12	940 (47)
<i>bolitinos</i>	0	73 (2)	BT x1	20 (1)	BT x2	40 (2)
<i>cornutus</i>	—	— ^a	EHD x1	100 (5)	—	—
<i>nevilli</i>	EHD x1	27 (1)	—	—	—	—
<i>magnus</i>	0	260 (2)	—	—	—	—
<i>zuluensis</i>	0	22 (1)	—	—	0	28 (2)
<i>pycnostictus</i>	—	—	0	20 (1)	—	—
<i>nivosus</i>	—	—	0	140 (7)	—	—
<i>milnei</i>	0	11 (1)	—	—	—	—
<i>leucostictus</i>	—	—	—	—	0	—
sp. # 107	—	—	—	—	0	42 (2)

^a No specimens of relevant sp. collected

TABLE 3 Presence of group specific antibodies against the viruses of BT and EHD in cattle—South Africa 1996/97

Number of cattle tested	Group specific antibodies in cattle against		
	BT (ELISA)		EHD (CF)
	At onset of outbreak	After the outbreak	At onset of outbreak
Tested	53	149	53
Negative	12	3	49
Suspicious	4	12	0
Positive	37	126	4
% negative	22	2	92

Twelve of 24 calves bled on Donkerhoek were negative for BT antibodies when they were first tested (Table 4). Twenty-eight days later the antibody titres of six positive calves had decreased, six suspicious reactions turned negative and the twelve negative calves were still negative. Over the same period antibody titres against EHD virus of the five positive calves had increased and 18 of 19 calves which tested negative on day 0 seroconverted to positive.

Rainfall

The rainfall (Fig. 3) in 1995/96 in four widely separated localities within the affected area was well above the average and markedly higher than recorded in the preceding two seasons. In contrast, the rainfall in 1932/33 and the preceding two years was considerably lower than the average.

DISCUSSION

Diagnosis

The disease features of BT and EHD are similar in their clinical manifestations, pathology, seasonality, known vector (Couvillion, Nettles, Davidson Pearson & Gustafson 1981), low morbidity and occasional epizootics (Metcalf, Lomme & Beal 1980; Metcalf Luedke & Jochim 1991). Furthermore, dual infections have been reported in white-tailed deer (Prestwood, Kistner, Kellog & Hayes 1974; Thomas, Willis, & Ruckerbauer 1974) and cattle (Foster, Metcalf, Barber, Jones & Luedke 1980). As both viruses can be isolated from clinically normal cattle it is important to combine the virological and serological results to establish an aetiological diagnosis of BT/EHD-like disease in cattle.

On the farm Donkerhoek EHD virus was isolated from acutely ill calves as well as from *Culicoides*. The antibody levels of the calves against EHD increased over the test period and negative calves became positive. The opposite was true for BT. It could not be isolated from affected cattle or *Culicoides*. Antibodies against BT virus in the calves declined, positive calves became negative and negative calves remained negative. These results clearly indicate that on this farm the disease was caused by EHD virus.

No specific diagnosis could be made on other farms, as suitable serum samples were not available. However, the high rate of virus isolation from cattle with typical signs compared to no isolation from cattle with signs suggestive of some other infection indicates that the isolations were not coincidental. Furthermore, the

TABLE 4 Serological change in 24 5 to 8-month-old calves with signs of a BT/EHD-like disease on the farm Donkerhoek in South Africa 1997

Test result	Number with antibodies against			
	BT		EHD	
	Day 0 (%)	Day 28 (%)	Day 0 (%)	Day 28 (%)
Positive/suspicious	12 (50)	6 (25)	5 (21)	23 (96)
Negative	12 (50)	18 (75)	19 (79)	1 (4)

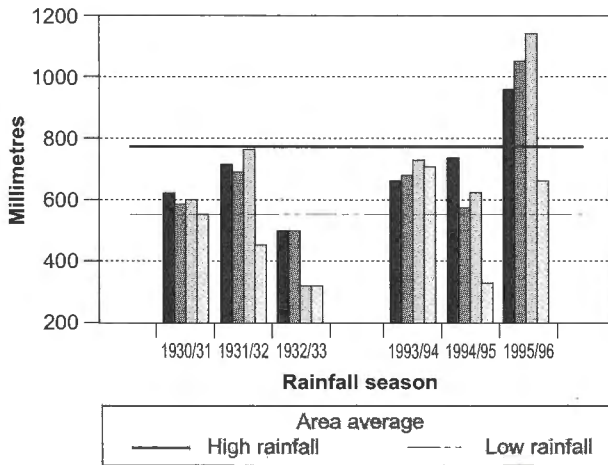


FIG. 3 Rainfall at four widely separated localities during two outbreaks of a BT/EHD-like disease in South Africa: 1932/33 and 1995/96

increase in the number of cattle with antibodies against both viruses from the early stages of the outbreak to the end of the season, together with the relatively high isolation rates for BT (20%) and EHD (15%) viruses strongly indicates that one or both viruses were involved. The simultaneous involvement of both viruses is further implied by the isolation of BT and EHD viruses from cattle in the same herd and from *Culicoides* collected on the same farm.

Outbreaks of the disease in cattle in 1932/33 occurred sporadically while in sheep it was present even where it was previously almost unknown. On some farms the disease in cattle and BT in sheep occurred simultaneously (Bekker *et al.* 1934); this outbreak was the first in which an aetiological agent was demonstrated (Bekker *et al.* 1934). The diagnosis was based on the development of BT in sheep inoculated with blood collected from affected cattle, as present-day diagnostic techniques were not available. It is thus likely that EHD was involved in those cases where BT virus could not be demonstrated.

Epidemiological aspects

Climatic conditions in 1995/96 were, according to present beliefs, most favourable for BT to occur. Al-

most the entire country experienced above average rainfall resulting in lush pastures and the development of swampy areas which had been dry for previous years. Record numbers of *Culicoides*, mostly *C. imicola*, a proven vector for BT virus (Du Toit 1944), were caught in light traps in widely separated localities (R. Meiswinkel, OVI, unpublished results 1996). Both viruses were isolated from this species.

Climatic conditions experienced before and during the 1932/33 outbreak were vastly different from those currently believed to favour outbreaks of *Culicoides* transmitted viral diseases. Low rainfall records proof that the 1932/33 season, as well as the previous two seasons, was exceptionally dry. However, according to Bekker *et al.* (1934) the telluric conditions were probably favourable for the occurrence of the outbreak.

The occurrence of BT over a much wider area than the distribution in South Africa of *C. imicola*, the only proven vector for BT, is a clear indication that other species, less dependent on high rainfall are involved. The present isolation of BT virus from three of five pools of parous *C. bolitinos* is evidence that this species, which breeds in cattle dung, may be a vector for BT virus. Furthermore, the isolation of EHD from parous *C. nevillei* and *C. cornutus* suggests that they too may also be potential vectors for BT and indicates that more research is needed to define vector capacity and to understand which climatic conditions give rise to large numbers of these species.

In the present outbreak BT virus was isolated from 12 and EHD virus from nine cattle from widely separated localities while *Culicoides* collected on affected farms yielded two isolations each of both viruses. *Culicoides* collected on Kaalplaas, where no cases of the disease were seen, yielded BT only. The reason why no disease was seen on Kaalplaas is not clear.

Economic consideration

The importance of the disease, based on the low morbidity rate (< 1–10%) reported by farmers, is largely underestimated. The reason for such low morbidity rate reports can be ascribed to the inexperience of

farmers in observing slight clinical signs and inconspicuous lesions. On the farm Donkerhoek careful inspection showed that virtually all calves were affected. A high morbidity in an affected dairy herd was implied by a drop of 42% in milk production in a dairy herd within days of the first appearance of clinical cases. The drop in milk production was a general phenomenon revealed by the decrease in the volume of milk handled by a milk-processing firm. The diminished production was most marked in areas where the disease was reported. These findings support previous observations that sometimes a loss in milk yield is the first sign of malaise in a dairy herd (Bekker *et al.* 1934), and can lead to considerable financial loss. The detrimental effect of infection on beef production is unknown but cannot be ignored in a disease which can occur over an extensive part of a country and which can affect a high percentage of cattle.

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