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Incidence of bluetongue virus precipitating antibodies in sera of some domestic animals in the Sudan

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SUMMARY

To determine the presence and prevalence of bluetongue (BT) infection in a variety of domestic animal species in different geographical regions of the Sudan, a serological study using the agar gel precipitation technique was initiated. A total of 2142 serum samples were examined. Of the numbers tested approximately 28% of sheep, 11·2% of goats, 8% of cattle and 4·9% of camels were positive for group-specific antibodies to BT virus antigen, indicating previous exposure to BT infection. None of the samples tested from horses or donkeys were positive. The findings suggest that the disease is widely distributed in most parts of the Sudan where possible insect vectors prevail and may be endemic in sheep in Juba District, Equatoria Province, Southern Region. Goats appeared to have some degree of resistance to infection compared with sheep, and there seemed to be no significant differences in positive rates between farm and free-range cattle.

It is concluded that BT infection may cause clinical disease in sheep, while it is probably subclinical or inapparent in goats, cattle and camels of the Sudan.

INTRODUCTION

Bluetongue (BT) is an infectious arthropod-borne viral disease principally affecting ruminants, to which sheep in particular are most susceptible. It is transmitted by mosquitoes and biting gnats or midges, Culicoides species (Neitz, 1948; Price & Hardy, 1954; Foster, Jones & McCrory, 1963; Luedke, Jones & Jochim, 1967; Lee, Causey & Moore, 1974; Luedke, Walton & Jones, 1976; Luedke, Jones & Walton, 1977).

Serologically BT virus is represented by at least 16 distinct types (Andrews & Pereira, 1972). Since Klontz, Svehag & Gorham (1962) first described the precipitation of BT antigen by specific antibodies, the gel precipitation test has been employed in numerous serological surveys to determine the extent of BT virus exposure (Trainer & Jochim, 1969; Metcalf & Jochim, 1970; Soliman, Hafez & Ozawa, 1972; Hafez & Ozawa, 1973; Afshar & Kayvanfar, 1974; Moore & Kemp, 1974; Taylor & McCausland, 1976).

Although the disease has long been recognized and is widespread in Africa (Howell, 1963) its existence in the Sudan has not been well reported and the various

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aspects of its epizootiology not studied. The first reference to a disease in sheep suspected to be BT was made in 1953 (Anon, 1953), when an outbreak occurred at the Gezira research farm in Wad Medani and the 'infective material' sent to Onderstepoort Research Laboratory, South Africa, was shown to contain a BT virus. The antigenic virus type, however, was not specified, but a vaccine introduced from South Africa was claimed to be effective against the local strain (Anon, 1957). Subsequently Pillai (1961) reported on suspected cases of BT in a flock of sheep at the Khartoum University farm. Attempts at virus isolation were unsuccessful but a presumptive BT diagnosis was established based on the clinical symptoms and gross lesions as well as the general picture of the disease. Ever since the disease diagnosed on these grounds has been reported sporadically from different parts of the country. On several occasions virus isolation was attempted but has not so far been achieved.

The present serological study was undertaken to assess the status and significance of BT infection in some domestic animal species of the Sudan as part of a series of investigations aimed at determining the role of the common viral pathogens in Sudanese livestock.

MATERIALS AND METHODS

Bluetongue virus antigen

Tissue culture precipitating antigen prepared from type 10 BT8 strain as described by Jochim & Chow (1969) together with reference antiserum were provided by Dr M. M. Jochim, Animal Disease Research Laboratory, United States Department of Agriculture, Denver, Colorado. These were stored at 4 °C until used for serum testing.

Serum samples

All sera were collected from apparently healthy animals in various regions of the Sudan. The majority of ovine, caprine and some of the bovine sera were obtained from free-range animals during 1975 in connexion with the animal census and disease survey programme. Additional bovine sera representing farm animals were obtained in 1976 and 1977 from Kuku cooperative dairy, Khartoum North, and Nisheshiba breeding centre in Wad Medani, respectively. Camel sera came from slaughtered animals at Tambool market abattoir, Gezira Province, during 1972. Equine sera were derived from several areas including Khartoum, Gedaref and Obeid. No attempt was made to record the age, sex or breed of the donor animals. All sera were stored at −20 °C before testing.

Serum testing

Individual sera were examined for precipitating antibodies to BT virus antigen using the agar gel precipitation test. This was carried out by a modification of the technique described by Ouchterlony (1949). The gel diffusion medium consisted of 1% (w/v) purified agar (Difco) in distilled water. Warm agar was poured into

Table 1. *Results of testing sheep sera from various parts of the Sudan for precipitating antibodies to BT virus antigen*

Serum origin	No. sera	No. positive	Positive %
Rufaa	429	162	37·8
Omdurman	198	33	16·7
Zalingi	134	7	5·2
Obeid	19	4	21·0
Dongola	18	0	0
Atbara	32	7	21·9
Juba	13	8	61·5
El Nuhud	19	10	52·6
Malakal	25	12	48·0
Shendi	23	6	26·1
El Dein	20	4	20·0
Bara	20	7	35·0
Soba Research Laboratory	30	16	53·3
Total	980	276	28·2

plates* 100 × 15 mm at 20 ml volumes per plate. After the agar had solidified four sets of wells were punched in each plate using a template locally made to the desired pattern. The wells, which were approximately 6 mm in diameter, were arranged in a hexagonal pattern around a central well of a similar diameter with the centre of a middle well 1 cm from the centre of each outside well. Individual wells were sealed with 0·03 ml volumes of molten agar and the plates kept at 4 °C until required for serum testing.

Bluetongue virus antigen was pipetted in the central wells and undiluted test serum in the peripheral wells at 0·03 ml. In each plate undiluted reference antiserum and normal antigen were included as controls. The plates were held at between 22 and 27 °C in a humid air-tight container for 48–72 h before they were examined for precipitin lines under reflected light using a magnifying glass.

RESULTS

The results of testing sheep sera from various parts of the Sudan are shown in Table 1. Of 980 samples examined 276 (28%) were positive. The positive rates ranged from 5·2% in Zalingi, Southern Darfur Province in the west, to 61·5% in Juba District, Southern Region. None of the samples from Dongola, Northern Province, was positive.

The incidence of BT virus precipitating antibodies in the sera of goats, cattle, camels, horses and donkeys is shown in Table 2. The highest positive rates among these species were detected in goats followed by cattle and camels. All equine sera were negative throughout. Of 98 serum samples from goats 11·2% were positive with the highest incidence (23·5%) in goats from El Dein area, Southern Darfur Province, and lowest (8·3%) in those from Malakal District in the south. All of

* Schott Mainz, Jener Glas, West Germany.

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Table 2. *Results of testing caprine, bovine, camel and equine sera for precipitating antibodies to BT virus antigen*

Species	Origin	No. sera	No. positive	Positive %
Goats	Juba	18	4	22·2
	El Dein	17	4	23·5
	Nyala	16	2	12·5
	Malakal	12	1	8·3
	Soba Research Laboratory	35	0	0
	Total	98	11	11·2
Cattle	Kuku dairy	344	15	4·4
	Nisheshiba breeding centre	278	43	15·5
	Juba	102	3	2·9
	Gedaref	66	5	7·6
	Malakal	84	4	4·8
	Total	874	70	8·0
Camels	Tambool	102	5	4·9
Horses	Various	53	0	0
Donkeys	Various	35	0	0
	Totals	1162	86	7·4

Table 3. *Overall incidence of precipitating antibodies to BT virus antigen in sera of different animal species*

Species	No. sera	No. positive	Positive %
Sheep	980	276	28·2
Goats	98	11	11·2
Cattle	874	70	8·0
Camels	102	5	4·9
Horses	53	0	0
Donkeys	35	0	0
Total	2142	362	16·9

the 35 sera from goats at Soba Research Laboratory, Khartoum, were negative. Of 874 bovine serum samples tested 70 (8%) were positive; the highest positive rate (15·5%) being detected in Nisheshiba breeding centre, Gezira Province, and lowest (2·9%) in free-range animals from Juba District. Of the 102 camel sera five (4·9%) were positive. Table 3 shows the total incidence of BT virus precipitating antibodies in the different animal species surveyed.

DISCUSSION

The agar gel precipitation test has been shown to be advantageous in detecting BT virus group-specific antibodies, which commonly persist for a long time after infection (Bowne, 1971). The test as described in this paper was found to be reliable, practicable and more rapid and simpler to conduct than the one described by Jochim & Chow (1969) and later improved by Jochim (1976), while at the same time being almost as economical.

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Since the object of this study was mainly to screen the various animal species qualitatively for BT virus antibodies and obtain an estimate of the prevalence and distribution of infection, no attempt was made to monitor antibody titres for seroconversions in any one herd or flock. Neither was the possible effect on the results of such factors as the time of sampling, age or breed of sampled animals considered. Also as BT is predominantly a disease of ruminants of which sheep are generally considered most susceptible (Howell, 1963), particular emphasis in this survey has been made on ovine serum samples.

Considering that no vaccination against BT is practised in the Sudan, the results of this serological survey would clearly indicate that BT infection occurs in all domesticated ruminants of the Sudan as evidenced by the demonstration of specific antibodies in the sera of sheep, goats, cattle and camels, but not horses or donkeys. The absence of such antibodies in the equine species may lend support to the disease being mainly of ruminants. However, with the detection of BT antibody in pigs (Afshar & Kayvanfar, 1974) this point yet remains to be unequivocally confirmed.

The results also indicate that sheep are more exposed to BT infection than goats, cattle or camels, and that the infection might well be enzootic in sheep in Juba District, Equatoria Province, where a positive rate of 61·5% was recorded in this species. However, this finding is not surprising in view of the heavy annual rainfall and abundance of mosquitoes and other biting insects in this region, which may serve as virus vectors. Further BT infection generally appears to be widely distributed in most parts of the Sudan where such possible insect vectors prevail. Hence the absence of infection in Dongola and the very low incidence in Zalingi District may be explained by the relatively cold weather and scarcity of rains in the former, which is in the desert zone, and the altitude of the latter which lies very close to the Marra mountain. As such the conditions in both areas are unfavourable for insect population to multiply.

Goats appear to have some degree of resistance to BT infection, when compared with sheep. Thus whereas the positive rate in sheep from Juba District was 61·5% it was 22·2% in goats from the same area. Similarly the positive rate was 53·3% in sheep and 0% in goats kept under the same conditions at Soba Veterinary Research Laboratory. By virtue of their lower antibody contents, cattle and camels appear to be even more resistant to BT infection than goats. Apart from the relatively high incidence of BT antibody recorded in cattle from Nisheshiba breeding centre no significant differences seemed to exist between farm and free-range cattle with regard to previous exposure to BT infection. The demonstration of BT virus antibodies in goats, cattle and camels, taken with the complete lack of reports of clinical disease in these species indicates that the infection in them is subclinical or inapparent.

Concluding from the results of this survey BT infection is generally widespread in the Sudan and occurs in all domestic ruminants, with sheep being most commonly exposed to infection. As has been recently reported in other countries (Hafez & Ozawa, 1973; Afshar & Kayvanfar, 1974; Taylor & McCausland, 1976) ruminant species are also involved in the maintenance of BT virus in the Sudan.

While BT infection may be manifested clinically in sheep, it is probably subclinical or inapparent in goats, cattle and camels. Further studies including intensified research on the virus types involved and the role of insect vectors and wild ruminants in the epizootiology of the disease will be required if eventually control measures are to be considered.

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