J. Hyg., Camb. (1985), **95**, 191–196
Printed in Great Britain

A study of the host range and distribution of antibody to Akabane virus (genus bunyavirus, family *Bunyaviridae*) in Kenya

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(*Received* 22 *October* 1984; *accepted* 2 *February* 1985)

SUMMARY

Serum neutralizing antibody to Akabane virus (genus bunyavirus, family *Bunyaviridae*) was found in a high proportion (50–95 %) of cattle sampled in Kenya, while sheep and goats had fewer positive (13–33 %). Camel and horse sera also contained antibody to the virus (70 % and 50 % respectively). The antibody was found in animals from the high altitude temperate type of grasslands, drier bushed and wooded grasslands and the semi-desert. No arthrogryposis nor hydranencephaly has been encountered in Kenya which might be related to this widespread virus infection.

A wide range of Kenyan wild ruminants had antibody to Akabane virus in their sera, as also did zebra.

INTRODUCTION

A strain of Akabane virus (genus bunyavirus, family *Bunyaviridae*) was isolated from *Anopheles funestus* mosquitoes trapped in a coastal forest belt in Kenya (Metselaar & Robin, 1976). Another virus strain related to Akabane was isolated from culicoides midges in South Africa (Theodoridis *et al.* 1979). Akabane virus has been shown to be the cause of abortions and a congenital arthrogryposis hydranencephaly syndrome in cattle and sheep in Japan, Australia and Israel (Kurogi *et al.* 1975; Markusfeld & Mayer, 1971; Hartley *et al.* 1975). This syndrome has not been described in Africa other than as a consequence of the vaccination of sheep with an attenuated Rift Valley fever vaccine (F. G. Davies, unpublished data; Coetzer & Barnard, 1977). A close surveillance for such congenital malformations has been maintained in Kenyan cattle for 15 years but none has been encountered (F. G. Davies, unpublished observations).

Bluetongue (genus orbivirus, family *Reoviridae*) and ephemeral fever (family *Rhabdoviridae*) viruses are thought to be transmitted by *Culicoides* spp. in Kenya (Davies & Walker, 1974*a*; Davies & Walker 1974*b*; Davies, Shaw & Ochieng, 1975). They show a wide ecological distribution and a high prevalence of antibody in the domestic and wild ruminant populations. The work described in this paper was carried out to examine the possibility that Akabane is another common and widespread African virus which involves the natural fauna in its maintenance cycle.

MATERIALS AND METHODS

Virus strain

The virus strain used was the MP 496 strain isolated by Metselaar & Robin (1976). This is serologically closely related to, if not identical with Akabane virus. No pathogenicity studies have been carried out with the strain in early pregnant sheep and cattle. The strain was adapted to Vero cells by three passages at limiting dilutions. A stock virus was prepared and stored at −70 °C.

Serum neutralization

The serum neutralization tests were carried out in a microtitre system. Sera were inactivated at 56 °C for 30 min and diluted 1 in 5 with serum free Eagle's minimum essential medium (MEM), 0·025 ml aliquots were transferred to the test plate and mixed with a further 0·025 ml of MEM. A second dilution was made so that each test used wells, containing a 1 in 10 and 1 in 20 dilution of the original serum. Tests were carried out in triplicate in early experiments and later in duplicate. To these, approximately 100 $TCID_{50}$ of the virus was added in 0·025 ml volumes and the plates held at 37 °C for 1 h and then overnight at 4 °C. Vero (or PS cells, a continuous porcine kidney line, David-West & Porterfield, 1974 in early experiments) cells were added in 0·1 ml volumes per well and the plates sealed and held at 37 °C. The plates were examined after 3 days for evidence of cytopathic effects and the results read at 5 days either microscopically or by staining the cultures with crystal violet. Sera were considered to be positive when 80% or more of the cytopathic effects of the challenge virus were suppressed.

Sera

Sera were collected from cattle, sheep, goats and other animals over a period of several years and stored at −20 °C. The wild animal sera were largely from the collections made by the Wildlife Section at the Veterinary Research Laboratories, Kabete. The origins of the the sera were related to the ecological zones of Kenya described by Pratt, Greenway & Gwynne (1966). They are based upon the climatic and vegetation characteristics, which are relevant to vector distribution and agricultural potential. Positive control sera were prepared in adult mice by repeated inoculations with 10% suspensions of MP 496 infected mouse brain.

RESULTS

The results of neutralization tests in sera from domestic animals are shown in Table 1. Serum neutralizing antibody to MP 496 was found throughout all the diverse ecological zones of Kenya from coastal or high altitude forest to the dry *Commiphora* bushed scrublands of the semi-desert. Cattle appeared to be strongly preferred feeding hosts for the vector of the virus in Kenya, with 90% of many populations of adult animals containing antibody to the virus.

A group of sentinel cattle were first bled at 2–6 months of age, when most contained antibody to Akabane virus in their sera. This was presumably colostral in origin. Some showed seroconversions each year, until at 3–4 years of age, most had antibody to the virus (Table 2). This was the situation with most cattle populations where older animal groups were sampled whenever possible.

Table 1. *Antibody to Akabane virus in domestic animals in Kenya*

	Neutralizing antibody [number positive/number tested (%)] in animals from ecological zone*					Totals
	II	III	IV	V	VI	
Cattle	177/231 (76)	241/280 (86)	160/193 (83)	28/40 (70)	—	606/744 (81)
Sheep	12/47 (26)	35/133 (26)	17/186 (9)	—	—	64/366 (17)
Goats	—	6/46 (13)	31/92 (33)	—	—	37/138 (26)
Camels	—	—	9/12 (75)	49/56 (87)	19/42 (45)	77/110 (70)
Horses	—	—	12/24 (50)	—	—	12/24 (50)
Totals	189/278 (68)	282/459 (61)	229/507 (45)	77/96 (80)	19/42 (45)	796/1382 (58)

* II Moisture index, −10 or greater (forest and derived grasslands & bushlands).
III Moisture index, −10 to −30 (*Combretum* savanna and moist bushed grasslands).
IV Moisture index, −30 to −42 (semi-arid *Acacia* savanna, dry rangelands).
V Moisture index, −42 to −50 (arid *Commiphora* dry thornbush and scrub).
VI Moisture index, −51 to −57 (very arid dwarf shrub grasslands).

https://doi.org/10.1017/S0022172400062422 Published online by Cambridge University Press

Table 2. *Antibody to Akabane virus in a group of sentinel cattle, first bled at 2–6 months*

Age	Number tested	Number (%) with neutralizing antibody
under 5 months	30	25 (83)
1 year	30	1 (3)
2 years	29	6 (20)
3 years	21	18 (85)
4 years	23	22 (95)

Table 3. *Antibody to Akabane virus in wild game animals in Kenya*

	Neutralizing antibody [number positive/number tested (%)] in animals from ecological zone*			
	II	III	IV	Totals
Wildebeest	—	4/5	26/81	30/86 (34)
Kongoni	—	—	37/57	37/57 (60)
Waterbuck	—	—	7/40	7/40 (17)
Buffalo	37/48	8/10	17/56	62/114 (54)
Topi	—	10/10	—	10/10 (100)
Eland	—	—	10/13	10/13 (77)
Bushbuck	1/4	—	—	1/4 (25)
Impala	—	7/12	7/22	14/34 (41)
Grant's Gazelle	—	—	4/11	4/11 (36)
Thomson's Gazelle	—	—	12/51	12/51 (24)
Zebra	—	—	25/108	25/108 (23)
Totals	38/52 (73)	29/37 (78)	145/439 (33)	212/528 (40)

* See Table 1 for definition of zones. No wild animals from zones v and VI were tested.

Sheep and goats have a much lower antibody prevalence than cattle. Camels which are long lived animals had a higher proportion with antibody to the virus. Antibody was also found in sera from horses, zebra and a whole range of wild ruminant species (Table 3). A number of positive sera from each species group of wild ruminants were titrated to determine their antibody levels to the virus. The titres were in the range of 20 to 320, with most below 80. Positive cattle, sheep and goat sera gave similar titres.

DISCUSSION

The results obtained appear to support the hypothesis that Akabane virus is endemic in those parts of sub-Saharan Africa which share ecological characteristics with parts of Kenya. The wild game animals in the region have evidence of contact with the virus, for many contain serum neutralizing antibody. They may have played a part in the maintenance and amplification of the virus in those areas where domestic stock have only relatively recently been introduced. Antibody was also found in camel, horse and zebra sera. It is suggested that Akabane is, like bluetongue and ephemeral fever, an indigenous African virus, and not an introduction to the region.

Cattle are a preferred feeding host for *Culicoides* spp. in Kenya (Davies & Walker, 1974*a*; Walker & Boreham, 1976). They would also appear to be favoured by the vector of Akabane virus in Kenya. A high antibody prevalence to Akabane virus was found in cattle populations of zones II–V, which covers a range of habitat from moist grasslands to semi-desert. Camel sera from zone V showed 87% with antibody, and 45% from zone IV which is virtually desert. A similar wide distribution of antibody was found with bluetongue and ephemeral fever in Kenya (Davies & Walker, 1974*a*; Davies, Shaw & Ochieng, 1975). Both these diseases are thought to be transmitted by *Culicoides* spp. The latter are considered to be the vectors of Akabane virus in Australia. The distribution of *Anopheles funestus* from which MP 496 was originally isolated, does not correlate with that of the antibody. It is not considered to be a candidate vector of any great importance.

Cattle do not breed in the drier rangelands of Kenya until they are at least three years of age. Consequently, only a few are likely to remain susceptible to the virus, when they are in early pregnancy. Many sheep and goats are however susceptible to the virus during early pregnancy, for the proportions of their populations which are immune to Akabane virus is much lower than with cattle. The explanation as to why they are not affected, must await the results of pathogenicity studies with MP 496 in the indigenous strains of sheep and goats which are found in Kenya. It is possible that, as with bluetongue and Rift Valley fever viruses, they are relatively insusceptible.

This paper is published by kind permission of the Director of Veterinary Services, Kenya. The authors wish to acknowledge the support of Research Grant R 3792, from the Ministry of Overseas Development, London.

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