

Communications

Continuing prevalence of African horse sickness in Nigeria

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ADEYEFA (C.A.O.), HAMBLIN (C.) Permanence de la prévalence de la peste équine africaine au Nigeria. *Revue Elev. Méd. vét. Pays trop.*, 1995, 48 (1) : 31-33

Les anticorps contre la peste équine africaine ont été recherchés au moyen du test ELISA dans les sérums d'équins provenant de dix régions du Nigeria nettement séparées les unes des autres. Les animaux testés comprenaient des chevaux importés ou exotiques, des chevaux de race indigène ou croisés localement et des ânes africains. Un pourcentage élevé des sérums (79,8 p. 100) était positif, ce qui confirme la permanence de la prévalence des anticorps contre la peste équine africaine chez les équins nigériens.

Mots clés: Âne - Cheval - Virus peste équine africaine - Epidémiologie - Anticorps - Test ELISA - Nigeria.

Introduction

African horse sickness (AHS) is an infectious but non-contagious arthropod borne virus disease of equidae caused by an Orbivirus belonging to the family Reoviridae. *Culicoides imicola*, the only proven field vector of African horse sickness virus (AHSV) is abundant in Nigeria (1, 5). Although all 9 serotypes of AHSV have been identified in the sub-saharal regions of Africa and are considered to be enzootic in these regions (9), only AHSV serotype 9 has been confirmed in Nigeria.

Early reports of AHS in the country (7, 8) were diagnosed almost entirely on clinical observations. In later years, however, clinical diagnoses were supplemented by small-scale serological surveys, confirming AHSV infection in the horse population (3, 4).

The first recorded isolation of AHSV in Nigeria was made from a horse that died in Zaria in 1970 (11). This isolate was subsequently identified as AHSV serotype 9. Kemp (10) later demonstrated some neutralizing activity against this virus isolate in sera from 138 of 144 horses (95.8 %) and 14 of 14 donkeys (100 %) collected from animals in the western part of the country. In 1975, Best *et al.* reported an outbreak of disease which had occurred in Kano (2) the previous year. During this epizootic approximately 20 imported horses died. Isolated viruses were again confirmed as AHSV serotype 9. Later, Nawathe *et al.* (12) demonstrated precipitating antibodies against AHSV in sera of 86 of 99 (86.7 %) local horses and 32 of 47 (68.1 %) imported horses. These authors suggested that

since only the imported horses were likely to have been vaccinated the high prevalence of antibodies in sera from local animals was probably due to continual exposure to AHSVs. More recently, Oladosu *et al.* (13) reported an outbreak of AHS involving two horse stables in Lagos. AHSV was isolated from three blood samples taken from sick and in-contact horses. These authors also demonstrated complement fixing antibodies against AHSV in 35 of 39 (89.7 %) of the horse sera collected from the infected stables.

This article reports the presence of antibodies against AHSV in 254 horse and 58 donkey sera collected between January 1991 and September 1993 from ten widely separated regions throughout the country (table I).

Materials and methods

Sera were examined by competition ELISA using methods similar to those described by Hamblin *et al.* (6). Briefly, 50 µl/well of concentrated, cell extracted AHSV antigen, diluted in carbonate/bicarbonate buffer (pH 9.6), was passively adsorbed overnight at 4°C onto the solid phase of U-well ELISA plates (Dynatech).

Fifty microlitre amounts of each test serum, diluted 1 in 5 in phosphate buffered saline (PBS) containing 5 % Marvel® milk powder and 1 % adult bovine sera (blocking buffer), were added to duplicate wells of a 96-well carrier, transfer plate (40 sera per plate). Fifty microlitres of the optimal dilution of guinea-pig anti-AHS infectious sub-viral particles, prepared in blocking buffer, were then added to each well.

The ELISA plates were washed five times and dried by inversion onto absorbent paper. Serum mixtures were transferred onto the washed ELISA plates and incubated at 37°C for 1 h on an orbital shaker. The ELISA plates were washed as before. Bound guinea-pig antibodies were measured following the addition of 50 µl/well of rabbit anti-guinea-pig immunoglobulins conjugated to horse radish peroxidase enzyme. Plates were again incubated at 37°C for 1 h on an orbital shaker. After washing, 50 µl/well of orthophenylene diamine/H₂O₂ were added. Colour development was stopped after 10 min by the addition of 50 µl/well of 1M H₂SO₄.

Absorbance values were determined at 492 nm. Controls on each plate included duplicate wells containing one strong positive, two weak positive and one negative horse sera. Test sera were recorded positive where the absorbance values in the duplicate test wells were less than 50 % of the mean absorbance value recorded in 8 virus control wells containing guinea-pig antisera in the absence of competing test sera.

Results

Table I shows the regions sampled, the origin of the species tested, the range of ages and, where known, the vaccination history of the animals. Overall, there was a high

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Reçu le 22.7.1994, accepté le 23.5.1995.

TABLEAU 1
Detection of AHSV antibodies by ELISA in equine sera from Nigeria

Region	Origin	Species	Age (yr) range	Number +/Total	% Positive	Vaccination history
Sokoto	Imported	Horse	9	1/1	100.0	Unknown
Sokoto	Local*	Horse	2-13	22/28	78.6	None
Sokoto	Local	Donkey	3-6	34/50	68.0	None
Maiduguri	Imported	Horse	6-10	6/9	66.7	Unknown
Maiduguri	Local	Horse	4-13	31/32	96.9	None
Kano	Imported	Horse	4-11	5/5	100.0	Unknown
Kano	Imported	Horse	8	2/2	100.0	None
Kano	Local	Horse	8-12	3/3	100.0	None
Kaduna	Imported	Horse	7-10	6/6	100.0	Vaccinated
Kaduna	Local	Horse	4-10	6/7	85.7	None
Jos	Imported	Horse	7-12	3/3	100.0	Unknown
Jos	Local	Horse	4-13	14/15	93.3	None
Ilorin	Local	Horse	2-11	3/8	37.5	None
Ilorin	Local	Donkey	3-6	0/2	0.0	None
Ibadan	Imported	Horse	5-15	7/10	70.0	Unknown
Ibadan	Local	Horse	4-16	26/31	83.9	None
Ibadan	Local	Horse	6 Months	1/2	50.0	None
Ibadan	Local	Donkey	5-7	1/6	16.7	None
Nsukka	Local	Horse	2-11	9/13	69.2	None
Ikeja	Local	Horse	4-13	27/35	77.1	None
Lagos	Imported	Horse	8-11	12/12	100.0	Vaccinated
Lagos	Imported	Horse	7-11	16/17	94.1	Probably
Lagos	Imported	Horse	5-8	1/2	50.0	Unknown
Lagos	Local	Horse	8	2/2	100.0	Vaccinated
Lagos	Local	Horse	4-8	3/3	100.0	Probably
Lagos	Local	Horse	4-13	8/8	100.0	Unknown
Total				249/312	79.8	

* Local = indigenous and locally cross-bred animals.

prevalence of antibodies in equine sera collected from all regions except Ilorin. One hundred and fifty-five (82.9 %) of the 187 sera collected from indigenous and local, cross-bred horses (*i.e.* local in table I) aged 6 months to 13 years were recorded positive by ELISA. A similar high percentage of the sera from the imported horses 88.1 % (59/67) aged between 4 and 15 years, was also recorded positive. Unfortunately, the vaccination history for these imported horses was not always available. As a consequence, only 18 of the 67 (26.8 %) sera tested were from horses known to have been vaccinated, while a further 17 (25.4 %) were from animals which were probably vaccinated. Only two (3.0 %) of the imported horses were known not to have been vaccinated. Both were eight years of age and had been imported from North Africa, although the actual country and the dates of importation were not known. In addition, it was not known whether these two animals had ever exhibited clinical signs of AHS or were survivors of a larger group, some of which might have died of AHS without being reported. No information was available for the remaining 30 (44.8 %) imported horses. There was no significant difference between the prevalence of AHSV antibodies detected in local and imported horses ($\chi^2 = 0.9945$, $p > 0.3187$). Positive levels of antibodies by ELISA were detected in 35/58 (60.3 %) of the sera collected from donkeys.

Discussion

Since there is no routine, annual vaccination programme in Nigeria, the indigenous and local cross-bred horses are rarely, if ever, vaccinated but despite this they appear to have a high innate resistance to infection with AHSVs (2). In contrast, horses imported into Nigeria, particularly from AHS free areas, are susceptible to infection and are therefore usually vaccinated before importation. However, once established in the country they are seldom re-vaccinated.

The high prevalence of antibodies recorded in sera from horses and donkeys of all ages from widely separated areas and in the absence of vaccination suggests that antibody levels are being boosted and maintained, probably by continual exposure to AHSVs. Such a high prevalence would also explain the relatively few confirmed outbreaks of disease recorded in recent years.

Conclusion

The data presented here supports earlier work and confirms the continued prevalence of AHSV antibodies in horses and donkeys throughout Nigeria. Further, all non-traumatic, sudden deaths in equines as well as suspected

cases of AHS should be investigated fully to determine which, if any, of the other eight AHSV serotypes are circulating in Nigeria.

Acknowledgements

The authors are grateful to Dr P.P.C. Mertens who kindly supplied the purified infectious sub-viral particles of AHSV serotype 9 used for the production of guinea-pig antiserum.

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ADEYEFA (C.A.O.), HAMBLIN (C.) Continuing prevalence of African horse sickness in Nigeria. *Revue Elev. Méd. vét. Pays trop.*, 1995, 48 (1) : 31-33

Equine sera collected from 10 widely separated regions throughout Nigeria were tested for antibodies against African horse sickness viruses (AHSV) using a competitive enzyme-linked immunosorbent assay (ELISA). The animals sampled included imported, exotic horses, indigenous and locally cross-bred (local) horses and African donkeys. A high percentage of the sera (79.8 %) were positive, confirming the continued prevalence of AHSV antibodies in Nigerian horses and donkeys.

Key words: Ass - Horse - African horse sickness virus - Epidemiology - Antibody - ELISA - Nigeria.

Effect of three different routes of administration on the immunogenicity of infectious bursal disease vaccine

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KEMBI (F.A.), DELANO (O.O.), OYEKUNLE (M.A.). Effet de trois voies différentes d'administration sur le pouvoir immunisant des vaccins contre la bursite infectieuse. *Revue Elev. Méd. vét. Pays trop.*, 1995, 48 (1) : 33-35

Les auteurs ont comparé la réponse immunitaire de trois groupes de 10 poussins vaccinés à l'âge de 2 semaines contre la bursite infectieuse (maladie de Gumboro) par voie orale, intramusculaire et oculaire, à l'aide d'un vaccin préparé par le NVRI (Vom, Nigeria). Tous les poulets sont restés séronégatifs 3 semaines après la primovaccination. Cependant, des anticorps précipitants étaient présents sur ces volailles après un rappel à l'âge de 6 semaines. Chez les poulets vaccinés par voie oculaire, cette séroconversion a été observée sur 70 p. 100 des sujets à l'âge de 7 semaines ; ce taux s'est élevé à 80 p. 100 dans les 2 semaines suivantes puis abaissé à 55,6 p. 100 à la 10^e semaine. Dans les groupes vaccinés par voies orale et intramusculaire, ces taux étaient respectivement de 30 et 33,3 p. 100 à la 7^e semaine mais ils s'élevaient pour les deux groupes jusqu'à 87,5 p. 100 à la 10^e semaine. Si l'on considère le facteur âge dans la sensibilité des poulets à la bursite infectieuse, la voie oculaire semble être la plus efficace.

Mots clés : Poussin - Volaille - Maladie de Gumboro - Vaccin - Nigeria.

Introduction

Infectious bursal disease (IBD) was first reported in birds about 3-7 weeks old in Nigeria (5). Since then the disease has been a major threat to the Nigerian poultry industry.

If chicks can be protected against the disease, especially in the first 4 weeks of life, the economic returns will be satisfactory. At present the majority of vaccines used in Nigeria are being produced by the National Veterinary Research Institute (NVRI), Vom, Nigeria. Although the NVRI recommended that the vaccine could be administered through the oral, ocular and intramuscular routes, owners and veterinary staff often use the oral one. But despite vaccination of flocks, persistent field outbreaks have occurred (1). This is of great economic importance because of the mortality and morbidity it causes. Various factors such as poor vaccine storage, transportation and interference of maternal antibodies have been alleged (4).

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Reçu le 15.11.1994, accepté le 28.3.1995.