SEROLOGICAL STUDIES WITH PESTE DES PETITS RUMINANTS AND RINDERPEST VIRUSES IN NIGERIA

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SUMMARY

One hundred and ninety-five goat and 67 sheep sera collected from various parts of southern Nigeria were screened for neutralising antibodies to both the peste des petits ruminants (PPR) and rinderpest viruses. Neutralising antibodies against both viruses were found in the sheep and goat sera examined. Parallel titration of samples which neutralised both viruses indicated a primary infection with the PPR virus (PPRV). However, some samples which failed to neutralise PPRV neutralised the rinderpest virus (RV) indicating RV activity in sheep and goats in Nigeria. These findings are discussed in relation to the diagnosis of PPRV infection and the recent reappearance of bovine rinderpest in Nigeria.

INTRODUCTION

Although natural rinderpest in sheep and goats is rare, there are reports of rinderpest in sheep grazed with infected cattle in Nigeria (Johnson, 1958) as well as in sheep, goats and cattle in the Sudan (El Hag Ali, 1973). The isolation of peste des petits ruminants very close to and shortly after the isolates described by El Hag Ali (1973) in Sudan has led to the speculation that the outbreak he described may have been PPRV infection (Taylor, pers. comm.). In India outbreaks involving only sheep and goats but later spreading to cattle and buffaloes have been reported (Narayanaswamy and Ramani, 1973). Zwart and Macadam (1967a, b) showed that cattle to sheep and goat transmission occurs and that sheep and goats can transmit the infection to cattle but only over a short period of time. Although Zwart and Rowe (1966) found that 18.8 and 15.2% of the sheep and goat sera they examined in parts of Nigeria had rinderpest neutralising antibodies, the result was seen as indicative of primary PPRV activity although these workers observed that the number of positive samples increased after contact with rinderpest infected cattle. The serological survey by Taylor (1979a) showed that rinderpest did not appear to exist in small ruminants in Nigeria. In view of the recent reappearance of bovine rinderpest in Nigeria it was decided to reassess the situation.

MATERIALS AND METHODS

Sera from 195 goats and 67 sheep were collected from Ihialla/Okija in Anambra State, Badeku, Abulle/Idiata, Abadina and the National Cereals Research Institute, Moor Plantation in Oyo State and Ondo in Ondo State and stored at -20°C until examined.

The PPRV used was the Vom isolate, NIG.75/1 (Taylor and Abegunde, 1979) which had undergone 40 passages in Vero cells. The rinderpest virus (RV) was the Kabete “O” strain (RBOK) (Plowright and Ferris, 1962) which had received 95 passages in bovine kidney (BK) cells (RBOK/BK 95).

PPR neutralisation tests were performed in roller tube monolayers of Vero cells. The growth medium for these cells consisted of Glasgow modified Eagle's
medium containing 10% ox serum and tryptose phosphate broth (Eagles/TPB\textsubscript{10}/
OS\textsubscript{20}) while the maintenance medium was medium 199 in Earle's balanced salt
solution with 5% ox serum. RV neutralisation tests were carried out in microtitre
plates using secondary BK cells grown in Eagle's medium containing 20% ox serum
(Eagles/OS\textsubscript{20}).

PPRV antibody screening tests were done according to the method of Taylor
(1979a) with slight modifications. Forty goat and 10 sheep sera with neutralising
antibody titres to RV (see below) and which completely neutralised PPRV in
screening tests were titrated against PPRV. The sera were inactivated at 56°C for
30 min and then diluted in 10-fold steps with medium 199 in Hanks balanced salt
solution to cover a range from 10\textsuperscript{-1} to 10\textsuperscript{-5}. The serum–virus mixture was held at
37°C for 2 h after which four tissue culture tubes were each inoculated with 0.2 ml
of the serum–virus mixture followed immediately by the addition of 1.0 ml of
growth medium containing 10\textsuperscript{8} freshly suspended cells as described by Taylor
(1979a). Tubes were sloped at 37°C for four days after which the medium was
changed to maintenance and the tubes were rolled. Further medium changes were
done every four days and the tubes were examined for cytopathic effect (CPE) on
days 5, 8, 11 and finally on day 13 when the test was terminated. Complete
protection of even one out of the four tubes per sample was interpreted as an
evidence of neutralising antibodies. Serum neutralisation titres were estimated
after Karber.

All 195 goat and sheep sera were screened and titrated against the RV
(RBOK/BK 95) simultaneously using a microneutralisation test. Fifty millilitres of
Eagle's medium was put into each of the 96 wells of the microtitre plates (Sterilin,
Middlesex, UK) and an equal volume of inactivated test serum added to the first row
using two wells per sample. Using a multi-channel Finnpipette doubling dilutions of
the sera were made to cover a range from 10\textsuperscript{-0.3} to 10\textsuperscript{-2.6}. An equal volume of RV
diluted in TPB to contain an estimated 10\textsuperscript{3.3} TC\textsubscript{50/ml} was then added to each of the
wells and the mixtures held at 37°C for 2 h. Thereafter 50 \mu l of freshly suspended BK
cells in Eagle's medium containing about 10\textsuperscript{5.6} cells/ml was added to each well, the
plates were sealed and incubated at 37°C. Microscopic readings for CPE were done
between days 4 and 6 and the final reading was done after the plates were fixed in
10% formalin in PBS. Serum neutralisation titres were estimated after Karber.

RESULTS

Out of the 195 goat sera which were screened for antibodies against both PPRV
and RV, 41 (21%) neutralised only PPRV, 77 (39.5%) neutralised both viruses
while 38 (19.5%) neutralised only RV. Of the 38 samples which neutralised only
RV 33 had titres of 10\textsuperscript{1.1} while three had titres of 10\textsuperscript{1.4} and two others had titres of
10\textsuperscript{2.1}. Forty of the goat sera which neutralised both viruses when titrated against
both had higher titres against PPRV than RV. While the PPRV antibodies ranged
between 10\textsuperscript{1.3} and 10\textsuperscript{3.8} RV antibody titres ranged between 10\textsuperscript{0.3} and 10\textsuperscript{2.1}.

Out of 67 sheep sera screened for antibodies to both viruses 10 (14.9%)
neutralised only PPRV, 29 (43.3%) neutralised both viruses while four (6%)
normalised only rinderpest virus. All four samples that neutralised RV only had
titres of 10\textsuperscript{1.1}. Titration of 10 of the sheep sera that neutralised both viruses
revealed that the PPRV antibody titres were also consistently higher than those to
RV. While the PPRV antibody titres ranged between 10\textsuperscript{2.1} and 10\textsuperscript{4.1} those to RV
were between 10\textsuperscript{0.8} and 10\textsuperscript{2.1}.