PREVALENCE OF ANTIBODIES TO PARAINFLUENZA-3 VIRUS IN VARIOUS WILDLIFE SPECIES AND INDIGENOUS CATTLE SHARING THE SAME HABITATS IN KENYA

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Abstract: Sera from various wild ruminants co-existing in the same habitats with cattle had haemagglutination-inhibition (HI) antibodies as high as those in the cattle. Sera from wildebeest *(Connochaetes taurinus),* kongoni *(Alcelaphus cokei),* Thomson's gazelle *(Gazella thomsonii),* eland *(Taurotragus oryx)* and the African buffalo *(Syncerus caffer)* had titers ranging from 64 to 128. Zebra *(Equus burchelli),* bushbuck *(Tragelaphus scriptus),* warthog *(Phacochoerus aethiopicus),* and oryx *(Oryx* spp.) sera were negative to the HI test. It was suggested that some of the wild ruminants with high titers could be a possible reservoir of parainfluenza-3 virus.

INTRODUCTION

Bovine parainfluenza-3 (PI-3) virus affects the respiratory tract of cattle and together with *Pasteurella* has been associated with the shipping fever complex.[6] In East Africa, PI-3 virus or a related agent was first isolated from lymph nodes of cattle.[11] A serological survey has shown that haemagglutination-inhibition antibodies to PI-3 virus are widespread in cattle in East Africa,[7] as well as wildlife species in other parts of Africa.[2] Kaminjolo *et al.*[9] confirmed the results of Plowright[11] by isolating PI-3 virus from an acute respiratory disease of cattle in Kenya. In an extensive survey of wildlife sera from several African countries, Hamblin and Hedger[4] demonstrated neutralising antibodies to PI-3 virus in 20 of the 44 species examined and suggested the African buffalo *(Syncerus caffer)* might be a particularly important reservoir of infection in the wild. Natural infection of the African buffalo was later confirmed by isolations of PI-3 virus from 3 of 51 clinically normal buffalo in Zimbabwe,[5] whereas Thorsen *et al.*[12] isolated the virus from one fallow and one mule deer and four of the free-ranging pronghorns in Canada. In both Kenya and Tanzania, Masai cattle graze together with numerous species of wild game, some of which are resident while others are on a seasonal migratory route. Furthermore, on many ranches within East Africa, most of the cattle co-exist with many species of wildlife in the same habitat. In view of this a survey for antibodies to PI-3 in wildlife species co-habitating the same areas with cattle was deemed necessary.

MATERIALS AND METHODS

Serum samples. Bovine sera were collected from herds located along migratory routes of wildlife and in game reserves where cattle and various wildlife species intermingled. The wildlife sera were collected in the same geographical areas from immobilized game animals or during game cropping programmes from ranches in the same areas. All wildlife were immobilized with a combination of xylazine, etorphine hydrochloride and acepromazine maleate.[3] Serum samples were stored at -20 C or -70 C until tested.

Virus antigen. A strain of PI-3 virus isolated from Kenya was used.[9] The virus was propagated in fetal bovine kidney

cell cultures grown in Eagle's minimum essential medium (MEM)[1] containing 5% bovine fetal calf serum, 100 I.U. penicillin and 25 mg mycostatin per ml. The maintenance medium contained the same ingredient with 2% horse serum replacing the fetal calf serum. The virus was harvested after the full cytopathic effects had been observed and stored at -70 C until used. The virus was used at 4 haemagglutinating units (HU) per ml.

Erythrocytes. Heparinised blood was obtained from bovine and guinea pig donors and centrifuged at 400 × g for 10 min. The erythrocytes (RBCs) were washed three times in phosphate buffered saline (PBS) pH 7.2 and bovine RBCs resuspended to a concentration of 10% for serum adsorption. Guinea pig RBCs were resuspended to 0.5% and 1.0% concentration for the haemagglutination-inhibition test after Kaminjolo and Paulsen.[8]

Treatment of sera. Both bovine and wildlife sera were adsorbed: 0.2 ml of serum were added to 0.6 ml of PBS pH 7.2 and 0.8 ml of 10% bovine RBCs. The mixture was incubated at 37 C for 15 min and then centrifuged to recover the serum which was inactivated in a waterbath at 56 C for 30 min.

Hemagglutination-inhibition test. World Health Organization (WHO) hemagglutination plates were used for HI test using the method of Rentscher *et al.*[2] modified by Kaminjolo and Paulsen[8] in which the inactivated sera were diluted in two-fold series. The HI titer was taken as the reciprocal of the highest serum dilution which completely inhibited 4 HU.

RESULTS

Antibody titers of up to 128 were detected in the sera from wildebeest *(Connochaetes taurinus)*, kongoni *(Alcelaphus cokei)*, Thomson's gazelle *(Gazella thomsonii)* and the African buffalo *(Syncerus caffer)*, whereas the sera of eland *(Taurotragus oryx)* had a titer of 512 (Table 1). Zebra *(Equus burchelli)*, bushbuck *(Tragelaphus scriptus)*, warthog *(Phacochoerus aethiopicus)* and oryx *(Oryx* spp.) sera were negative to the HI test. Most bovine sera obtained from two areas where wildlife lived or migrated through had high HI titers

Table 1. Prevalence of haemagglutination-inhibition (HI) titers to parainfluenza-3 (PI_3) virus in various wildlife species.

	Reciprocal serum antibody titers								
Animal species	<16	16	32	64	128	256	512	1024	Total
Wildebeest	10	2	5	2	3	—	—	1	23
Kongoni	9	3	2	3	1	—	—	—	18
Thomson's gazelle	7	1	3	2	2	—	—	—	15
Buffalo	7	1	—	6	3	—	1	—	18
Impala	15	—	2	1	—	—	—	—	18
Eland	9	1	3	—	—	—	1	—	14
Topi	1	2	—	—	—	—	—	—	3
Zebra	5	—	—	—	—	—	—	—	5
Bushbuck	3	—	—	—	—	1	—	—	4
Warthog	2	—	—	—	—	—	—	—	2
Oryx	3	—	—	—	—	—	—	—	2
Totals	69	10	15	14	9	1	2	1	121

[1] Wellcome (Kenya) Ltd.

[2] Dr. Rentscher *et al.*, Bakteriolongisches Institut, 6958, Laupheim, West Germany.

corresponding to those observed for the wildlife species (Table 2). One wildebeest *(Connochaetes taurinus)* serum and one bovine serum (both obtained from the Masai Mara region which is on the yearly migratory route of the wildebeests) had a titer of 1024. On examining all bovine and wildlife serum samples it was apparent that HI antibody titers were of the same order for both types of animals.

DISCUSSION

Cottral[1] believes that in bovine HI titers of 10 and 40 show evidence of previous infection by PI-3 virus, whereas bovine with clinical disease would show titers of 320 or higher. Kaminjolo *et al.*[9] reported that cattle which were seronegative to PI-3 on the HI test or had low titers (e.g., 4) later showed titers of 512 and 1024, respectively, after an interval of 5 weeks following the outbreak of disease in the herds. Thus it would appear that the titers reported herein indicate previous exposure to PI-3 virus for both the bovine and wildlife. The presence of HI antibodies to PI-3 virus in both the domestic and wild species indicates that the virus is ubiquitous throughout the geographic areas shared by these ruminants.

Table 2. Prevalence of haemagglutination-inhibition titers to parainfluenza-3 (PI_3) virus in bovine sera from wildlife habitats.

	Reciprocal serum antibody titers								
	<16	16	32	64	128	256	512	1024	Totals
Kilifi	10	6	14	13	2	—	3	—	48
Maasai Mara	25	5	8	9	7	1	16	1	72
Totals	35	11	22	22	9	1	19	1	120

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