PAPERS AND ORIGINALS

Tanapox: A New Disease Caused by a Pox Virus

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Summary

Two epidemics of a new virus disease, tanapox, occurred in 1957 and 1962 among the Wapakomo tribe along the Tana River in Kenya. Several hundred people were affected by a short febrile illness with headache and prostration and the disease was characterized by a single pock-like lesion on the upper part of the body. A pox virus, unrelated to the vaccinia-variola group, has been incriminated as the causative agent. The virus has a limited host range and has been grown only in human and monkey tissue cultures, and so far the only animals that have proved susceptible in the laboratory have been monkeys. The characteristic lesions have been reproduced in a human volunteer. Histopathological and electron microscopic studies indicate that the virus belongs to the pox group, but serological tests show that it differs from other animal pox viruses, including yabapox virus of monkeys. A similar if not identical pox virus has caused epidemics in primate colonies in the U.S.A. It is suggested that tanapox is a zoonosis and that the disease is transmitted from monkeys to man in Kenya.

Introduction

Two epidemics of an acute febrile illness associated with pox-like skin lesions occurred in 1957 and 1962 among the people living along the Tana River in Kenya. Biopsy specimens of the skin lesions were sent to Liverpool in 1962 and a pox virus was isolated. A preliminary report on the disease and the virus was given at a meeting of the Pathological Society of Great Britain and Ireland in 1963 and a summary of the findings was recorded by Allison (1965). The virus has now been shown to differ in many ways from other pox viruses and has been named tanapox. In this paper a more detailed account of the disease and the causative virus is presented.

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Epidemiology

The disease was first seen in September 1957 in the village of Ngau, which is situated on a small hill in the swamp forest flood plain of the lower reaches of the Tana River in Kenya. A small epidemic occurred among the children attending the local mission school. In 1962 a more widespread epidemic occurred along the middle reaches of the Tana River between Garissa and Garsen. About 50 cases were seen and all age groups and both sexes were affected. There were probably several hundred cases and the disease was apparently confined to the riverine Wapakomo tribe, who were living in scattered family groups along the river. In both years there were extensive floods and the people with their domestic animals and many wild animals were isolated on islands in the middle of extensive flood waters. In 1957 Ngau became an island in a swamp that was an ideal breeding site for *Mansonia* mosquitoes, and at the time of the epidemic the evening biting rate of *Mansonia uniformis* and *M. africanus* on man exceeded more than 700 per hour. Though the Wapakomo tribe were in contact with other nomadic tribes living in the surrounding semi-desert area there was no evidence of transmission among these people. Also there was no transmission to other people when infected patients were transferred to the Provincial Hospital in Mombasa for further investigations.

The absence of previous or subsequent reports does not mean that the disease is necessarily a new entity or that it has died out. This is a very isolated area where a non-fatal illness of short duration is unlikely to attract the attention of the very few medically qualified people visiting the area. The disease was discovered fortuitously when people with the characteristic lesions were seen in the course of surveys for filariasis and schistosomiasis. The people, however, had no name for the disease and they indicated that it was something new. The restricted distribution and occurrence of the epi-

demics at a time of extensive and prolonged flooding suggested that this was a zoonosis resulting from the major ecological disturbance occurring at the time. This hypothesis gains some support from subsequent studies of the properties of the tanapox virus and closely related if not identical viruses isolated in outbreaks of disease in primate colonies in the U.S.A. (Casey *et al.*, 1967; Hall and McNulty, 1967; Espana, 1969).

Clinical Features of the Disease in Man

The clinical features of the disease have received very little attention. The incubation period is unknown. In most cases the infection started with a short febrile illness lasting three to four days, sometimes with severe headache and backache and pronounced prostration. In every case pock-like lesions appeared on the skin during the course of the febrile illness. The pock lesions were at first similar to those of smallpox starting as a papule and developing into a raised perfectly circular vesicle which then became umbilicated (Fig. 1), but there was no pustulation and most characteristically there was usually only a single lesion and never more than two. The lesions occurred mainly on the upper arm, face, neck, and trunk; they were not seen on the hands, legs, or feet. In an individual patient when first seen it might be difficult to distinguish this condition from modified smallpox in a vaccinated individual who develops only one or two lesions; but the larger size, the firm solid nature of the lesions, their slower evolution, and absence of pustulation were distinctive features.

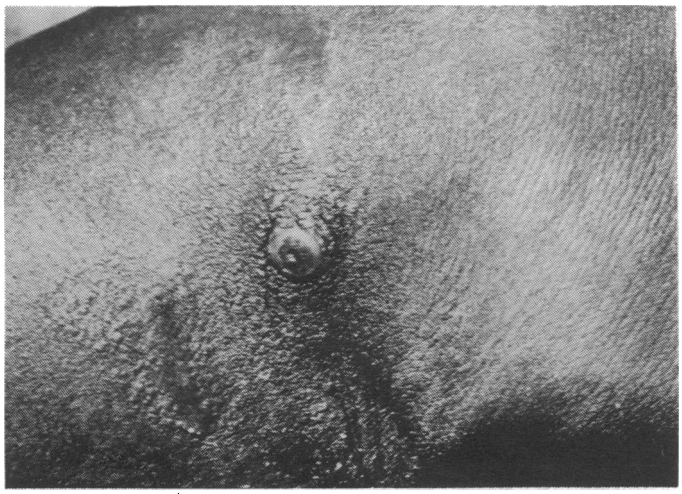

FIG. 1—Raised firm lesion showing umbilication.

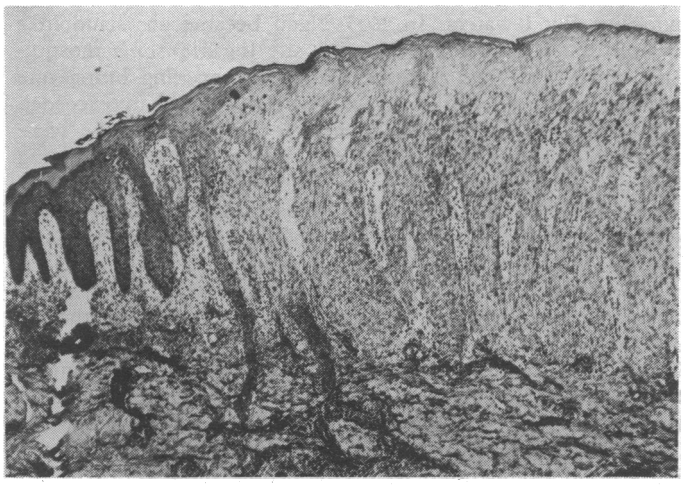

FIG. 2—Section of biopsied lesion from African patient. (Haematoxylin and eosin. × 34.)

Laboratory Investigations

Examination of stained sections of a biopsied skin lesion suggested that a pox virus might be the causal agent (Figs. 2 and 3). There was very pronounced hyperplasia of the skin

FIG. 3—High-power view of Fig. 2 showing cytoplasmic inclusions (arrows). (Haematoxylin and eosin. × 322.)

epithelium with comparatively little involvement in the underlying dermis. The epithelium showed little of the destructive changes seen in the skin lesions caused by vaccinia and variola viruses, and infiltration of the epithelium by inflammatory cells was slight. In the cytoplasm of the hypertrophied epithelial cells faintly acidophilic inclusion bodies often rather irregular in outline were present, and many cell nuclei appeared to be vacuolated. The histological appearances were more reminiscent of swinepox than of smallpox.

Two biopsied skin lesions—7 days and 10 days old—were received on 27 April 1962. Smears made from one of the lesions stained by Gutstein's alkaline methylviolet technique showed numerous elementary bodies such as those from vaccinia or variola lesions. A little of an extract, to which formalin to make 2% had been added, was sent to Dr. M. Epstein at the Middlesex Hospital. In spite of imperfect fixation a shadowed preparation showed in electronmicrographs appearances similar to those of vaccinia virus. Concentrated extracts from the biopsy specimens, however, failed to fix complement or to precipitate in agar gel when tested with a high-titre rabbit antivaccinial serum. It appeared therefore that this pox virus, though morphologically similar, was serologically unrelated to the vaccinia-variola group.

Cultivation of the Virus

Material from the biopsy specimens was inoculated into fertile eggs, into primary tissue cultures of human thyroid, and intra-

dermally on the foot pad of a guinea-pig and intracerebrally into three 1-day-old mice. No lesions were produced on the chorioallantois of chick embryos after several passages, nor were any detectable pathological effects caused by inoculation of material into the yolk sacs of chick embryos. No signs of inflammation appeared in the guinea-pig foot pad and the baby mice showed no ill effects and continued to thrive.

In monolayer cultures of human thyroid definite cytopathic changes were noted about 10 days after inoculation of an extract of the biopsied specimen. There appeared focal changes shown by increased granularity in the cytoplasm of the cells. These small foci slowly enlarged over succeeding days and the cells in the centres tended to become rounded off and detached from the glass, so that in the cell sheet small holes appeared surrounded by flattened cells with intensely granular cytoplasm. Inoculation of the culture fluid into fresh tissue-culture tubes at first failed to produce infection, but subculture to fresh tubes was successful if intact cells or cell suspensions treated by freezing and thawing or disintegrated by sonic vibration were used as inoculum. Subculture from the thyroid-tissue-culture tubes to primary cultures of human amnion produced similar cytopathic effects and with repeated subculture focal lesions could be first seen five to six days after inoculation.

Fig. 4 shows the appearance of an early focus in cultures of human amnion cells (unstained) and Fig. 5 the appearances at a higher magnification of cells in a more advanced lesion stained by haematoxylin and eosin. Cell nuclei tended to become vacuolated in older lesions, and inclusion bodies staining with eosin were to be seen in the cytoplasm. From culture tubes in which cytopathic effects had spread to involve the greater part of the cell layer, cell-free culture fluids had low titres of virus rarely exceeding 10^3 focus-forming units (f.f.u.) per ml, while sonicated cell suspensions had infective titres up to 10^5 or 10^6 per ml. For the preparation of high-titre virus suspensions, cells were collected into 10 ml of medium from 6 to 10 12-oz (340-g) tissue-culture bottles showing widespread involvement of the cell sheets after 10 to 14 days' incubation; these cell suspensions were sonicated, the cell debris thrown down by low speed centrifugation, and the supernatant fluid used as virus stocks. These were kept frozen at −70°C.

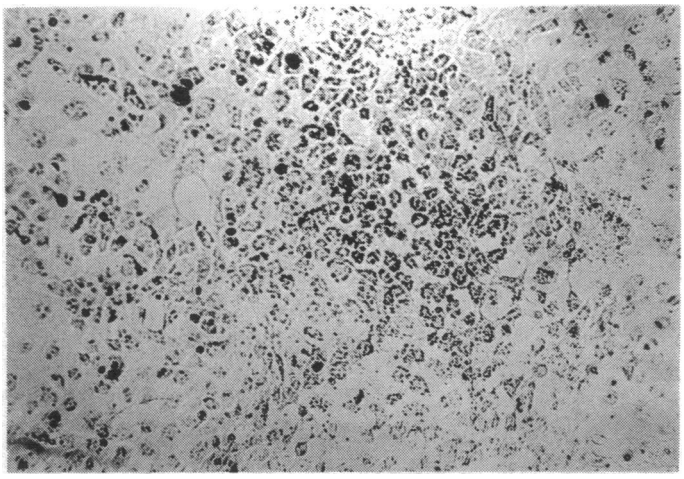

FIG. 4—Tissue culture of human amnion. Early focus of intensely granular cells some of which are beginning to round up. (Unstained. × 140.)

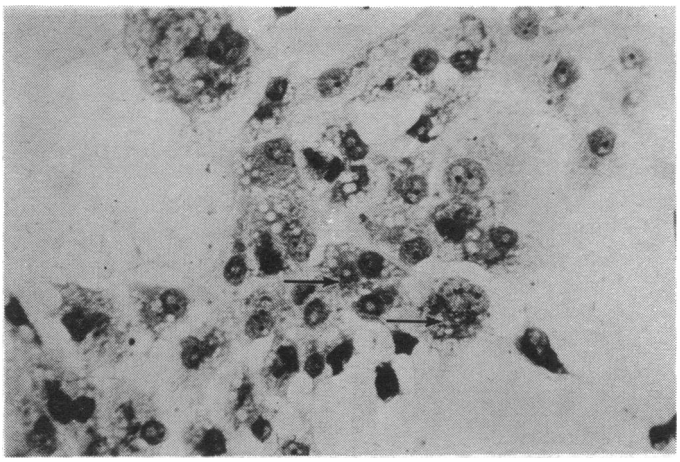

FIG. 5—Tissue culture of human amnion. Infected cells with vacuolated cytoplasm and inclusion material (arrows). (Stained haematoxylin and eosin. × 350).

In addition to primary tissue cultures of human thyroid and amnion, the human cell line, HEp2, and cell strain WI-38 were also found suitable for cultivation of the virus. Similarly typical cytopathic changes occurred in primary cultures of vervet monkey kidney and patas monkey kidney and in the continuous monkey cell lines GLV3A, GMK-AH-1, and Vero. For much of the later work with the virus, particularly testing of sera for neutralizing antibody, cultures in Vero cells were used. In this cell line focal lesions usually appeared on the fifth or sixth day after inoculation and were usually counted from the 8th to the 11th day, when most primary foci were well developed but secondary lesions had not appeared. Foci could be accurately counted in unstained cell sheets in Leighton tubes at a magnification of 10 times with a low-power dissecting microscope. No cytopathic effects were observed in tissue cultures of chick embryo fibroblasts, nor could virus be recovered from the chick fibroblasts after 10 days' incubation by subculture to tissue cultures of human cell lines. Similarly no specific changes were observed in cultures of primary rabbit kidney, a continuous line of rabbit kidney cells (GLRK13), or in primary cultures of bovine embryo kidney.

Attempts to Reproduce the Disease

After the virus had been passed three times in tissue cultures of human thyroid and four times in human amnion a pool was prepared for animal inoculation. The medium—199+2% calf serum—was removed from the virus-infected cell sheets after 12 days' incubation at 35°C. The cells were washed with Dulbecco's phosphate-buffered saline scraped off the glass tubes and suspended in fresh phosphate-buffered saline. The suspension was then sonicated by a Mullard drill for 30 to 40 seconds, centrifuged at 2,000 r.p.m. to remove cell debris, and the supernatant used as virus suspension. To this suspension sterile glycerol was added to make 40% and the mixture kept at −20°C. Over a period of six months this suspension had a titre of 5×10^5 f.f.u. per ml in tissue cultures of human amnion.

This suspension was used to inoculate a calf, a lamb, two young pigs, and a goat. Each animal was injected intradermally with 0·2 ml at two skin sites and on an adjacent scarified area of skin. The animals were observed over a period of one month but no lesion developed at any of the inoculation sites. The animals had been bled for serum before inoculation and subsequent specimens of blood were collected four to five weeks later. None of the sera showed any neutralizing antibody to the virus when tested in tissue cultures. It has been noted above that a virus suspension from the original biopsy specimen had been without effect in the skin of a guinea-pig and intracerebrally in baby mice. Tissue-culture virus was also without effect when injected intradermally or intramuscularly in rabbits.

MONKEYS

Two rhesus and two vervet monkeys were inoculated with virus on different occasions as shown below.

Rhesus 1 was inoculated intradermally at two sites and on an adjacent scarified skin area with the virus suspension used for inoculation of the calf, sheep, goat, and pigs mentioned above. Three small papules were seen at inoculation sites on the third day but these did not increase greatly in size though induration was still present on the eighth day and slight scabbing was present at the site of scarification.

Rhesus 2 was inoculated intradermally with 0·1 ml at each of 14 points. The virus suspension used was prepared in tissue cultures of Vero cells grown in 199 medium containing 2% vervet serum. The virus had previously been passed three times in tissue cultures of human thyroid, four times in human amnion, nine times in WI-38 cells (a cell strain derived from human embryo lung), and three times in Vero cells. The suspension had an f.f.u. titre in Vero cells of about 4×10^6 per ml. Papules appeared at all inoculation sites on the fifth day and increased in size until the ninth day, when the largest was raised, firm, and measured just over 1 cm in diameter. There was no vesiculation. Thereafter the lesions regressed and were barely detectable by the 14th day.

Vervet 1 received three different virus suspensions at the same time each being injected once intradermally and inoculated on to scarified skin. The first suspension was that used for rhesus 1; the second suspension was prepared from cultures in human amnion that had been inoculated with material from the human experimental infection recorded below; and the third suspension had been prepared from extracts of the skin biopsy material received from Africa. The first and second virus preparations produced small papular lesions by the third day after inoculation but these were regressing by the eighth day. The third suspension produced rather larger lesions, which had reached a diameter of 6-7 mm by the eighth day with slight surrounding redness of the skin. These lesions were regressing by the 12th day.

Vervet 2 was inoculated intradermally at 12 sites with the same virus suspension that had been used for rhesus 2. On the fourth day small papules 5 mm in diameter were present but these did not increase in size and seemed to be regressing by the seventh day. The fact that these lesions were less pronounced than those in rhesus 2 may have been related to the presence of low-titre antibody in the serum of this vervet monkey at the time of inoculation (see below).

All four monkeys were bled for serum before inoculation and again three to six weeks later. Neutralizing antibody was not detected in the sera of rhesus 1, but appeared in the sera of rhesus 2 and vervet 1 after inoculation, and increased in titre in the sera of vervet 2 as mentioned later. After they had been bled rhesus 2 and vervet 2 each received three intravenous injections at six-day intervals of high-titre virus grown in Vero cells in a medium containing 2% vervet serum and partially purified by repeated high-speed centrifugation. The animals were bled six days after the last intravenous injection and the immune sera used for later serological studies.

INOCULATION OF A HUMAN VOLUNTEER

The volunteer was an adult male who had been repeatedly vaccinated against smallpox. The virus used had been passed three times in human thyroid and four times in human amnion tissue cultures. The suspension used as inoculum was prepared from sonicated amnion cells in the same manner as that used for the domestic animals mentioned above, except that the virus was deposited from the stock suspension by centrifugation at 10,000 r.p.m. for 30 minutes and was then resuspended in phosphate-buffered saline. The titre of the suspension in human amnion tissue cultures was 2×10^5 f.f.u. per ml. A control suspension was prepared in exactly the same manner from uninfected amnion cell cultures.

All inoculations were made on the skin of the flexor aspect of the forearm. Two intradermal injections of 0·1 ml of the virus suspension and two of the control suspension were made; in addition, the virus suspension was inoculated into the scarified skin at two adjacent sites. No reaction appeared at the sites of injection of the control material. The intradermal injections of virus suspension produced two areas of erythema with slight central thickening by the fourth day. The appearance had not changed on the fifth day, but by the seventh day the lesions were fading, leaving a readily palpable underlying nodule. No obvious change was apparent at the scarification sites on the fourth day, but by the seventh day each showed several tiny papules with a surrounding zone of erythema about 1 cm in diameter. From one of the papules scraped gently with a sterile needle smears were made and stained by Gutstein's method. Numerous virus particles were present in the smear. On the eighth day, just before excision of the lesion, there was slight axillary adenitis but no fever or general upset.

The tissue removed under local anaesthesia on the eighth day was divided; one piece was fixed in Zenker and used to prepare paraffin sections and the other was fixed in osmic acid for sectioning and examination by electron microscopy. From a tiny piece of the excised lesion virus was recovered by culture in human amnion and thyroid cells. After the excision the edges of the sutured wound became thickened and slightly inflamed but this subsided after eight to nine more days. The histological appearance of the excised lesion under low power is shown in Fig. 6. There is considerable epithelial thickening though less than that shown in the section of the naturally occurring lesion in an African child (Fig. 2). Under high power (Fig. 7) the lower layers of the

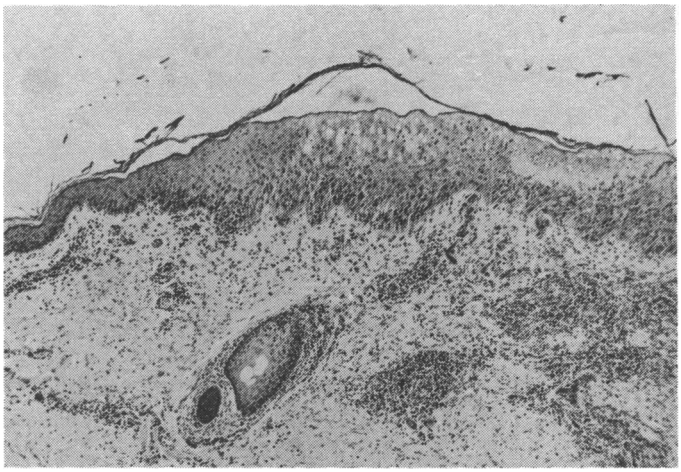

FIG. 6—Eighth day after virus inoculation. Section through lesion. (Stained haematoxylin and eosin. × 45.)

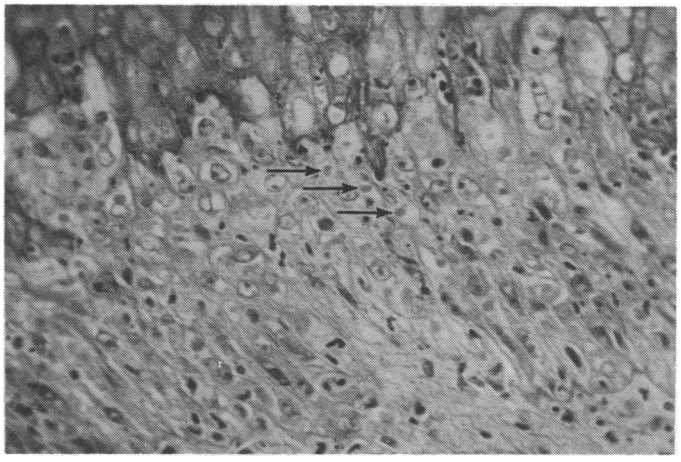

FIG. 7—Eighth day after virus inoculation. Section of lesion showing cytoplasmic inclusions (arrows). (Stained Giemsa. × 344.)

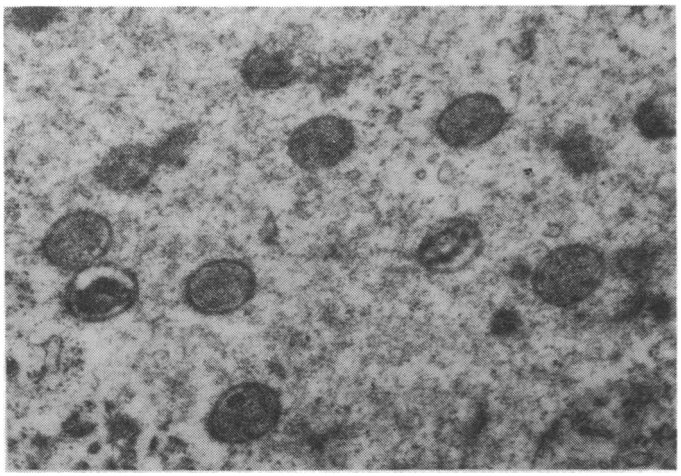

FIG. 8—Eighth day after virus inoculation. Section of tissue examined by electron microscope to show virus particles in cytoplasm of infected cell. (× 37,333.)

epithelium show cytoplasmic inclusions similar to those in Fig. 3. The appearance of the virus particles in thin sections of the lesion examined by the electron microscope are shown in Fig. 8. They resemble in size and appearance vaccinia virus particles in tissue sections prepared in the same way. Blood samples were taken from the volunteer just before inoculation of virus and again 14 and 23 days later. The results of examination of these sera are given below.

Serology

Tests for antibody in sera have been made by neutralization, complement fixation, and precipitation in agar-gel techniques. Neutralization tests were made in Vero cell monolayers in Leighton tubes. Virus suspension diluted to give 50-100 f.f.u. in 0·1 ml was mixed with equal volumes of serum dilutions in rubber-stoppered tubes. These were then placed in a water-bath at 37°C for two hours; of the mixtures 0·2-ml volumes were added to the monolayer culture tubes from which medium had been removed; 1·2 ml of medium was added to each tube and the tubes were then incubated at 35°C. Three or four tubes were used for each mixture and a normal serum devoid of antibody and dilutions of an immune monkey serum were included in each test. When the control tubes inoculated with normal serum-virus mixture showed well-marked focal lesions—usually from the 8th to the 11th day after inoculation—lesions were counted in each tube. Fifty per cent. reduction in mean focus count as compared with the controls was taken as the end point of neutralization.

In complement fixation tests the antigen used to test for antibody in sera was prepared from sonicated Vero cells from monolayer cultures showing numerous virus lesions. Mixtures of serum dilutions, antigen, and complement (two and a half full haemolytic doses) were kept at 4°C overnight before adding sensitized cells and incubating the tubes at 37°C for 30 minutes. An extract from sonicated non-infected Vero cell cultures was used as a negative control "antigen."

It has been noted above that the sera of calf, lamb, goat, and pigs which had been inoculated with tissue-culture virus intradermally and on the scarified skin failed to neutralize the virus in a dilution of 1/10, the lowest serum dilution tested. This was not surprising as there was no clinical evidence to suggest that the inoculated virus multiplied in these animals. The results of tests on human and monkey sera are shown in the Table. In the first two sera from African patients no antibody was demonstrable with the techniques used. The third patient's serum showed a low titre of complement-fixing antibody, which had increased in the later sample, but neither showed neutralizing antibody. The results of tests on the serum of the experimentally infected individual suggest that serum antibody may develop slowly in response to infection; it may be that the sera were collected too early from the African patients but the day of illness on which they were collected is uncertain.

No antibody was detected in the serum of the first rhesus monkey 41 days after intradermal injection, but the virus inoculum in this animal was considerably smaller than that given to subsequent animals and the lesions which were produced were rather insignificant. The vervet monkeys showed much smaller lesions after intradermal infection than did rhesus 2. This may have been a consequence of low susceptibility in this species and in vervet 2 to the presence of neutralizing antibody in the serum before infection. The antibody titres in the sera of rhesus 2 and vervet 2 were considerably increased by subsequent intravenous injections of virus.

That the virus isolated in culture was responsible for the lesions in the African children was shown by serological tests made with the high-titre immune serum from vervet monkey 2. This serum was used to test saline extracts of the African biopsy lesions for viral antigen by complement fixation and precipitation in agar gel tests. The central and superficial parts of the lesions were excised; the excised tissue was cut finely with scissors and ground with glass powder in 0·4 ml of saline. The watery extract obtained after low-speed centrifugation was tested for antigen. Crusts from a case of herpes zoster were treated in the same way to serve as control. With the monkey immune serum used at a constant dilution of 1/20 the extract of the tanapox lesion fixed complement up to a dilution of 1/64 but gave no fixation with the preimmunization serum from the same monkey. The zoster crust extract gave negative results in dilutions from 1/4 upwards with both monkey sera. In a precipitation in agar gel test with the undiluted extracts and undiluted monkey sera the tanapox extract gave a line of precipitate with the immune monkey serum but none with the preimmunization serum, while the zoster crust extract failed to precipitate with either serum.

Relation of Tanapox Virus to Other Poxviruses

Tanapox virus, like many pox viruses, is resistant to 40% glycerol in the cold and in phosphate-buffered saline retains its titre for months at −20° or −70°C. Virus suspension with an infective titre of 10^7 f.f.u. per ml and concentrated extracts of virus-infected tissue cultures failed to agglutinate red cells from a vervet monkey, a baboon, a fowl, baby chicks, a rabbit, a guinea-pig, and man (group O). That this virus belongs to the pox group is evident from the clinical

Serological Response of Humans and Monkeys to Infection with Tanapox Virus

Subject	Before Infection		Days after Infection	C.F. Titre	Neut. Titre
	C.F. Titre	Neut. Titre			
A.B.			N.K.	<1/5	<1/10
C.D.			N.K.	A.C.	<1/10
E.F. "Acute"			N.K.	1/10	<1/10
E.F. Convalescent			N.K.	1/20	<1/10
Volunteer	<1/5	<1/10	14	1/10	1/10
			23	1/20	1/50
Rhesus 1	<1/5	<1/10	41	<1/5	<1/10
Rhesus 2	<1/5	<1/10	19	1/80	1/500
			3 i.v. injections of virus		
			38	1/160	1/5000
Vervet 1	<1/5	<1/10	44	1/20	1/50
			22	1/80	1/800
Vervet 2	<1/5	1/40	3 i.v. injections of virus		
			40	1/320	1/6000

C.F. = Complement fixation. Neut. = Neutralization. A.C. = Anticomplementary serum. N.K. = Not known.

and histological appearance of the lesions and its size and structure as shown by the electron microscope; in addition, the virus can readily be shown to reactivate heat-inactivated vaccinia virus in tissue cultures of human or monkey cells (to be published).

The virus is serologically not related to the vaccinia-variola group of pox viruses to which monkey pox belongs. A high-titre antivaccinial serum prepared in the rabbit failed to precipitate or fix complement with tanapox antigen and failed to neutralize the virus in tissue culture. The human volunteer had good neutralizing antibody against vaccinia virus before infection but none against tanapox. Comparative studies with other animal pox viruses have not been made, but antisera to many of them have been tested for neutralizing antibody against tanapox in tissue culture. The following sera failed to neutralize tanapox virus when tested in a dilution of 1/10: human sera obtained after infection with molluscum contagiosum and with orf; serum from a sheep after infection with contagious pustular dermatitis; and immune sera prepared in the rabbit with tissue-culture grown viruses of goat pox, Rumanian sheep pox, lumpy skin disease of cattle, contagious pustular dermatitis, and fowl pox.

Discussion

The occurrence of the outbreak of mild pox disease in a remote area of Kenya, without record of a similar occurrence elsewhere, and the history of a similar outbreak several years before, suggested that the source of the infection might have been some species of animal. The fact that the disease occurred under conditions of flood, when mosquitoes were particularly active, and that the lesions occurred on exposed parts of the skin suggested that the infection might be transmitted by mosquitoes by direct mechanical transfer from animal to man in a manner similar to the transmission of myxomatosis among Australian rabbits. In the laboratory the virus could be grown only in tissue cultures of human or monkey cells, and transmission of infection by inoculation of virus succeeded only in man and monkeys. These observations suggest that the reservoir of infection in the Tana River valley is probably some species of monkey. At least five species of primates are present in the locality where the outbreak occurred (*Papio doguera*, *Cercopithecus mitis*, *Cercopithecus aethiops*, *Colubus abyssinicus*, and *Cercocebus galeritus*) and of these one at least (*C. aethiops*) has proved to be susceptible to experimental infection. As yet, however, no attempt has been made to transmit infection by mosquitoes in the laboratory, nor have specimens of blood from monkeys in the Tana River valley been examined for antibody, which might provide evidence of the occurrence of the virus infection in these animals.

In 1966 there were outbreaks of mild pox infection in primate colonies in three different parts of the United States of America—Oregon, California, and Texas. The infection was apparently derived from monkeys held by one importer, and macacus monkeys were mainly affected (Schmidt, 1970). The lesions in the monkeys were similar in appearance and in their histology to the lesions of tanapox in African children. They were hypertrophic in character, slow in development, with little evidence of vesiculation, and in these respects were quite different from the lesions of monkey pox. In at least two of the American centres infection was contracted by those working with the monkeys. In these contact human infections the lesions both clinically and histologically bore a definite similarity to the lesions of tanapox in Kenya (McNulty *et al.*, 1968). From the monkey and human infections in these American primate centres pox viruses were isolated which appeared to have growth characteristics like those of tanapox virus. By the co-operation of Dr. McNulty, of Portland, Oregon, and of Dr. Espana, in Davis, California, strains of their viruses and of tanapox virus have been exchanged. While the comparative study of the viruses has not been completed it seems that the American strains are very similar if not identical with tanapox virus—a finding which strengthens the case for the monkey as the reservoir of infection for the disease in Africans in the Tana River area. Further studies, however, are necessary to provide conclusive evidence for this suggestion.

The virus isolated from the tanapox cases, like those from the outbreaks in monkey colonies in the U.S.A., appears to be different from other pox viruses so far described. Its host range, seemingly limited to man and monkeys, the slow development and evolution of hypertrophic lesions involving mainly epidermis, the restricted range of cells permitting growth in tissue culture, and the nature of the cytopathic changes observed are features that serve to distinguish these pox viruses from most of the others. The failure of a wide range of antisera to other pox viruses to inhibit growth of tanapox virus also serves to set this virus apart. Tanapox virus has some resemblances to yabapox virus. Both produce infection in monkeys and man, but the lesions caused by yabapox involve the production of slowly developing tumour-like masses of mesodermal tissue. Preliminary serological studies have been made with sera from monkeys convalescent from yabapox infection and an immune antiyabapox serum prepared in the rabbit and kindly supplied by Dr. Yohn, of Cleveland, Ohio. In complement fixation tests there is some cross-reaction between tanapox and yaba viruses but the antiyaba sera have so far failed to neutralize tanapox virus in tissue culture. Further work on the relationship of yabapox and tanapox is, however, desirable.

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