

A four-year survey of African swine fever in Malawi

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SUMMARY

A serological survey of free-ranging domestic pigs in the Central and Southern Regions of Malawi, together with laboratory data on confirmed cases of African swine fever (ASF) and data from interviews with pig owners, undertaken over a four-year period from 1981-4, has enabled the ASF enzootic area of Malawi to be identified. The area covers much of the western part of the Central Region and includes Mchinji district and parts of Kasungu, Ntchisi, Dowa and Lilongwe districts. Mortality is substantially less than 100% in outbreaks within the enzootic area but approaches 100% in outbreaks outside this area, as shown by both the serological investigation and the interview data.

INTRODUCTION

In most reports from Africa, African swine fever (ASF) is described as a peracute disease with mortality approaching 100% (Montgomery, 1921, reviewed by DeTray, 1963; Hess, 1971). The published reports of ASF in Malawi describe devastating outbreaks, particularly in the Southern Region, including one in Nsanje and Chikwawa districts starting in 1959 in which more than 12000 domestic pigs were lost (Matson, 1960; Cox & Hess, 1962). Most African isolates are therefore believed to be very virulent, and in these circumstances, with very high mortality, the disease is generally self-limiting.

Most of the early investigators of ASF in domestic pigs in Africa occasionally encountered prolonged infections both in natural and experimental circumstances (Montgomery, 1921; Steyn, 1932; DeKock, Robinson & Keppel, 1940; DeTray, 1957). Instead of death occurring within a few days of the appearance of symptoms (as with the peracute and acute forms of disease), disease lingered in some animals, with death occurring in 3-4 weeks (subacute form). In others, appearance of clinical signs alternated with apparent periods of remission, and infection persisted for several months (chronic form), or pigs became carriers without first showing clinical signs (subclinical form) (DeTray, 1963). The number of surviving pigs may increase and act as potential sources of infection if the disease becomes established in the domestic pig population (Scott, 1965*a*; Pini & Hurter, 1975). Wild pigs can be carriers of ASF virus but do not show signs of disease (Plowright, Parker & Pierce, 1969).

The first report of ASF as an enzootic disease of domestic pigs was from Angola (Mendes & Daskalos, 1955). The disease had occurred in epizootic form in Angola

in the 1930s (Mendes, 1961) but an increase in the number of prolonged infections was later noted and attributed to modification of the virus resulting from continuous association with domestic pigs (Leite Velho, 1956; reviewed by Hess, 1971, 1981). More recently a strain from South Africa (Pini, 1977) and a strain from Zaire (Thomson, Gainaru & Van Dellen, 1979) have been shown to have reduced virulence.

In Malawi, reported field cases of ASF and hence cases which are confirmed by laboratory tests are usually from outbreaks in which mortality is high. However, in rural areas of Malawi, as in neighbouring countries as well as in Angola, most domestic pigs are kept under a free-range management system, feeding largely by scavenging and being fenced only at night. Therefore if carrier domestic pigs are present, transmission of the virus may be perpetuated (DeTray, 1960; Scott, 1965*b*; Hess, 1981). There have been no documented cases of subacute, chronic or subclinical infections in Malawi, but village surveys undertaken during 1981 and 1982 indicated that the disease is more widespread than laboratory statistics imply (Haresnape, 1984). It is rarely possible to reach an affected area in time to observe the infected pigs because most outbreaks are not reported until they are over. Therefore the only way to establish how many pigs survive primary infection is by serological testing.

A serological survey of the free-ranging domestic pigs in the Central and Southern Regions of Malawi is presented in this paper together with laboratory data on confirmed cases of ASF and information obtained from interviews with village pig owners. The intention was to identify the enzootic areas.

MATERIALS AND METHODS

Definition of localities

The localities studied were assigned letters and numbers according to district, as shown in Table 1 and Figs. 1 and 2. Studies were made in all 9 districts in the Central Region and in 7 of the 10 districts in the Southern Region. Mangochi, Machinga and Mwanza districts and the Northern Region were omitted because few pigs are kept there. Mangochi and Machinga are predominantly Moslem areas.

Laboratory diagnosis of clinical cases

ASF was routinely diagnosed by examination of spleen smears by a fluorescent antibody (FA) test using fluorescein-conjugated anti-ASF IgG. The diagnosis was confirmed in three cases (all from locality MC6 in Mchinji district) by infecting pig leucocyte cultures with an extract of suspect tissue and observing haemadsorption. The haemadsorption test was based on that of Malmquist & Hay (1960), except that erythrocytes were allowed to settle out of heparinized blood under gravity instead of by centrifugation, nutrient medium was composed of 60% RPMI 1640 (Flow Laboratories, Irvine, Scotland) and 40% pig serum, and leucocytes were added to nutrient medium together with plasma. In one case (from ZO2 in Zomba district) the diagnosis was confirmed both by observing haemadsorption in leucocyte cultures prepared from blood of a sick animal and by inoculation of a susceptible pig with an extract of suspect spleen tissue. The inoculated pig died of ASF 8 days later. In another case (from LL10 in Lilongwe district) ASF was

Table 1. Localities in Malawi in which the ASF situation was studied

District	Localities studied
Region: Central	
Mchinji	Sitolo (MC1), Ndawambe (MC2), Kondoole (MC3), Tikoliwe (MC4), Menyani (MC5), Bongera (MC6), Likasi (MC7), Kankhowo (MC8), Chalaswa (MC9), Chawala (MC10)
Lilongwe	Mfuti (LL1), Namitete (LL2), Mkoko (LL3), Mtsilo (LL4), Chisikwa (LL5), Chilinda (LL6), Sinyala (LL7), Malimbwe (LL8), Mbabzi (LL9), Malandi (LL10), Katola (LL11), Chikuse (LL12)
Kasungu	Kaluluma (KS1), Kasikidzi (KS2), Rusa (KS3), Mkhota (KS4), Kavizinde (KS5)
Ntchisi	Mkanda (NT1), Mwera Hills (NT2), Khuuwi (NT3), Malomo (NT4)
Dowa	Chisepo (DO1), Dzoole (DO2), Simakuni (DO3), Makalani (DO4)
Dedza	Ndebvu (DE1), Kafotokoza (DE2), Chamangwana (DE3), Maonde (DE4), Nadulu (DE5), Chiphazi (DE6), Magunditsa (DE7), Bembeke (DE8)
Nkhotakota	Mwansambo (NK1)
Salima	Thavite (SA1), Chipunza (SA2)
Ntcheu	Mpamadzi (NC1)
Region: Southern	
Zomba	Namiwawa (ZO1), Thondwe (ZO2)
Blantyre	Matindi (BL1)
Chiradzulu	Thumbwe (CD1)
Thyolo	Bvumbwe (TH1)
Mulanje	Mphonde (ML1), Mpassa (ML2)
Chikwawa	Kanjedza (CK1), Chimkole (CK2), Tomali (CK3), Ngabu (CK4)
Nsanje	Bangula (NS1), Nsanje (NS2)

confirmed by inoculation of a susceptible pig with blood from a sick animal. In this case the inoculated pig died of ASF 5 days later. ASF in the inoculated pigs was diagnosed by clinical observation and confirmed by the FA test.

Collection of interview data

Pig owners were interviewed between 1981 and 1984 in the Central and Southern Regions of Malawi in localities where ASF outbreaks were reported during the period, and also in some localities where no disease was reported but in which free-range pigs were numerous. Outbreaks of ASF which occurred in the Central Region during 1981 and 1982 have already been described (Haresnape, 1984).

Individual pig owners were interviewed and asked how many pigs they owned, whether any had died and, if so, details of the numbers which died, with dates, clinical signs and duration of illness. From the information obtained those deaths which seemed likely to be the result of ASF were identified. Pigs in which the cause of death was not clear were excluded. A presumptive diagnosis was based on assessment of clinical details given by the pig owner. Owners generally described sudden onset of fever, shivering, weakness, loss of appetite, loss of balance or weakness in the hindquarters and refusal to walk. Some described loss of body condition, diarrhoea, vomiting or emaciation. The length of illness varied from 1 day to 2 weeks and many owners reported the sudden death of all their pigs within a few days. In some affected villages, every khola (pig pen) was affected and all pigs lost within about a month. It was usually difficult to obtain a detailed

description of post-mortem findings, but many owners reported that some organs, particularly the spleen, were unusually dark in colour. In those cases where tissue samples were submitted to the laboratory, the diagnosis was confirmed as described above.

Serological survey

Blood samples were taken from village pigs in 42 localities in the Central Region and 13 localities in the Southern Region. Samples were taken from the ear vein. The owner, village and date were recorded, together with information on any deaths that had occurred within the same khola. At least 10 sera were collected in each locality, and more than 20 in most.

Each serum was tested for the presence of anti-ASF antibodies by an enzyme-linked immunosorbent assay (ELISA). A limited number of samples were also tested by indirect immunofluorescence (IIF).

In six localities in the Central Region (MC3, MC6 and MC9 in Mchinji district, LL1 in Lilongwe district and DE6 and DE7 in Dedza district) pig owners were interviewed shortly after the start of an outbreak. These areas were then revisited on a regular basis in order to monitor further deaths and to identify surviving pigs from affected kholas.

Enzyme-linked immunosorbent assay (ELISA)

The ELISA test was chosen for detecting antibodies because it is sensitive, requires no sophisticated equipment and is suitable for testing large numbers of serum samples (Wardley *et al.* 1979; Hamdy *et al.* 1981; Sanchez-Vizcaino, Crowther & Wardley, 1983; Wardley *et al.* 1983).

Microtitre plastic plates (Falcon flexible assay plates, Becton Dickinson & Co., Oxnard, U.S.A. or microelisa plates M129A, Dynatech AG, Denkendorf, West Germany) were sensitized with 0.1 ml/well ASF antigen, prepared in monkey kidney cells and supplied by Dr Thomson, Onderstepoort, South Africa, in carbonate/bicarbonate buffer, pH 9.6, overnight at room temperature. After washing, 0.05 ml test serum at 1:40 in PBS/Tween (0.05% Tween 20, 0.5% bovine serum albumin in phosphate buffered saline, pH 7.4) was added per well and plates incubated for 1 h at 37 °C. After washing, 0.05 ml peroxidase-conjugated rabbit anti-swine immunoglobulin (Mercia Brocades Limited, Weybridge, UK) at 1:100 in PBS/Tween was added per well and plates incubated for 1½ h at room temperature. After final washing, 0.02 ml 30% H₂O₂ was added to 25 ml 5-amino salicylic acid at 0.8 mg/ml adjusted to pH 6.0, and 0.2 ml of this mixture added to each well. After 30 min each well in which a significant colour reaction could easily be seen by eye was scored as positive. Each serum was tested in at least two wells. Positive and negative control sera were included on each plate (at least four wells each). Each test was performed in parallel with one in which negative control antigen prepared from uninfected cells was used. In general, little or no colour appeared with negative control antigen but sera were only considered positive if the intensity of colour was greater using ASF antigen than using negative control antigen. All washing stages consisted of five washes with 0.5% Tween 20 in phosphate buffered saline (PBS), pH 7.4.

Some sera were tested using purified virus protein VP73 as antigen, in parallel

with the ASF antigen, to eliminate the possibility of non-specific reactions with ASF antigen leading to false positive results. VP73 prepared by the method of Tabares *et al.* (1981) was supplied by Dr Wilkinson, Pirbright, UK.

Indirect immunofluorescence (IIF)

Each test serum, diluted 1:10 in PBS, pH 7.4, was applied to a smear made from ASF-infected cells in addition to positive and negative control sera, and incubated at 37 °C for 30 min. Smears were washed three times in PBS to remove unbound antibody, and then stained with fluorescein-conjugated anti-ASF IgG. Sera which inhibited production of specific fluorescence in smears were scored as positive.

RESULTS

The diagnosis of ASF was confirmed by laboratory demonstration of the presence of ASF virus in tissue or blood samples from 21 localities. In a further 21 localities there were outbreaks of disease which was considered to be ASF on the basis of interviews. There are therefore 42 localities in which ASF is believed to have occurred between 1981 and 1984 (Fig. 1). These are 37 localities in the Central Region and 5 localities in the Southern Region, and in all but one (locality ZO2 in Zomba district) the disease affected free-ranging village pigs. There have never been any laboratory-confirmed cases of ASF in the Northern Region.

The results of the serological survey (Fig. 2) are based on results obtained by ELISA using ASF antigen. Thirty-four sera were tested in parallel with both antigens; 9 were positive and 25 were negative in both tests, but with 1 positive serum the colour reaction was more intense using ASF antigen than VP73. Fifty-four sera were also tested by IIF and there was good general agreement between the two tests, but ELISA was more sensitive. Of 34 sera which were positive by ELISA, only 13 were positive by IIF, and in addition 2 sera were positive by IIF but negative by ELISA.

As shown in Fig. 2, a high proportion of pigs tested were seropositive in some areas; for example, most localities in Mchinji district and certain localities in Ntchisi, Dowa and Lilongwe districts. In contrast, in many localities elsewhere no seropositive pigs were identified despite recent occurrence of ASF (shown in Fig. 1).

Central Region

Forty-seven localities in the Central Region were studied, and 37 are believed to have had cases of ASF between 1981 and 1984. Laboratory confirmation was only obtained in 20 of these but there was clear evidence from interviews that ASF had occurred in a further 17 localities (Fig. 1). In 4 localities the disease was said to have occurred but the reports were somewhat confused. In the remaining 6 localities no disease was reported (Tables 2-5).

In all localities in Mchinji district which were visited, interviews provided evidence of ASF. In some localities a high proportion of pigs tested were seropositive, indicating that some ASF-infected pigs had survived and hence that mortality had been less than 100%. In addition, the interview data revealed that some pigs in affected kholas had survived, which also indicates that mortality was

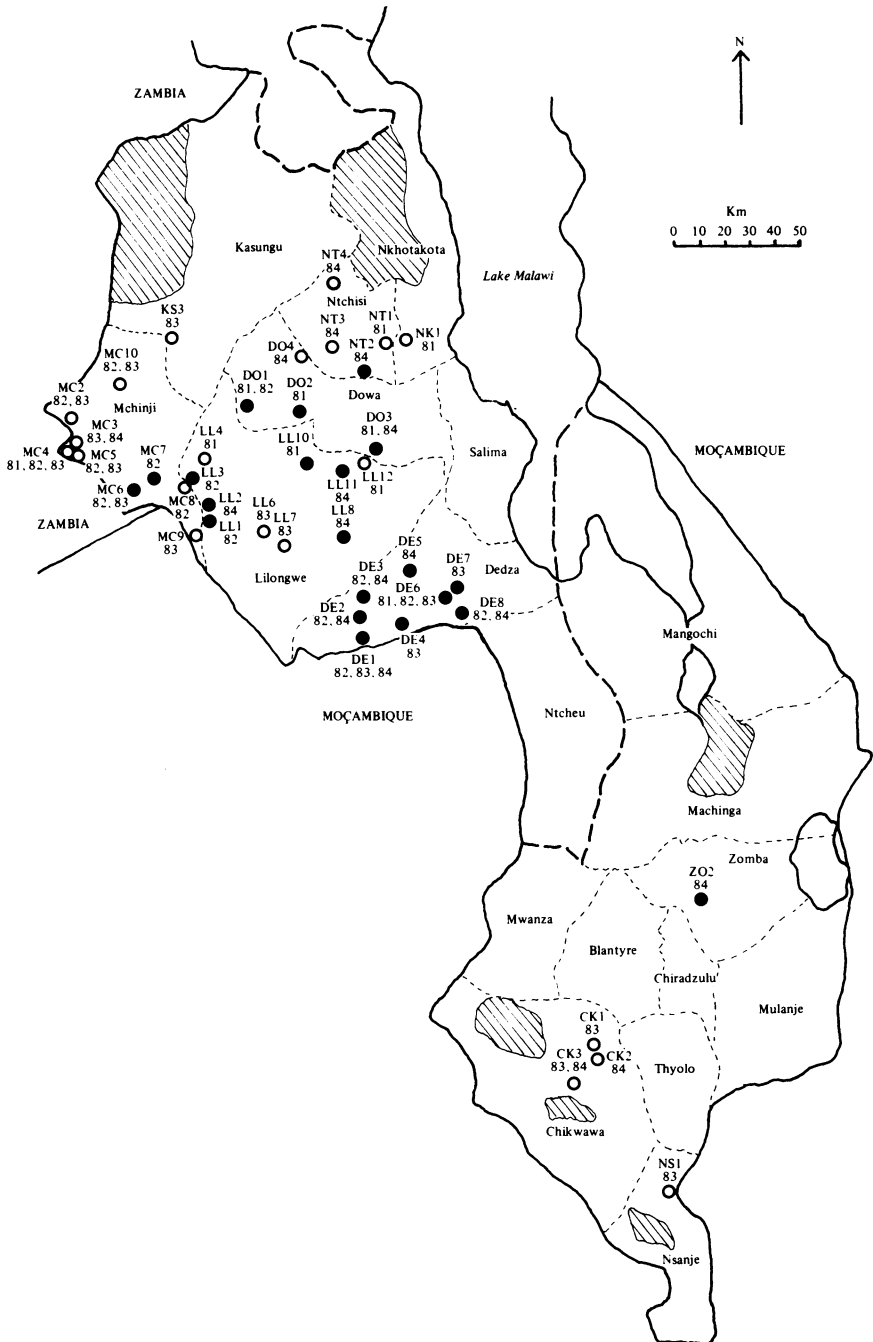


Fig. 1. African swine fever outbreaks in Central and Southern Regions of Malawi, 1981-4. ● LL1 82, Laboratory-confirmed outbreak with locality and year. ○ NT1 81, Other outbreak locality and year. ---, Regional boundary. -----, District boundary. ▨, National Park or Game Reserve.

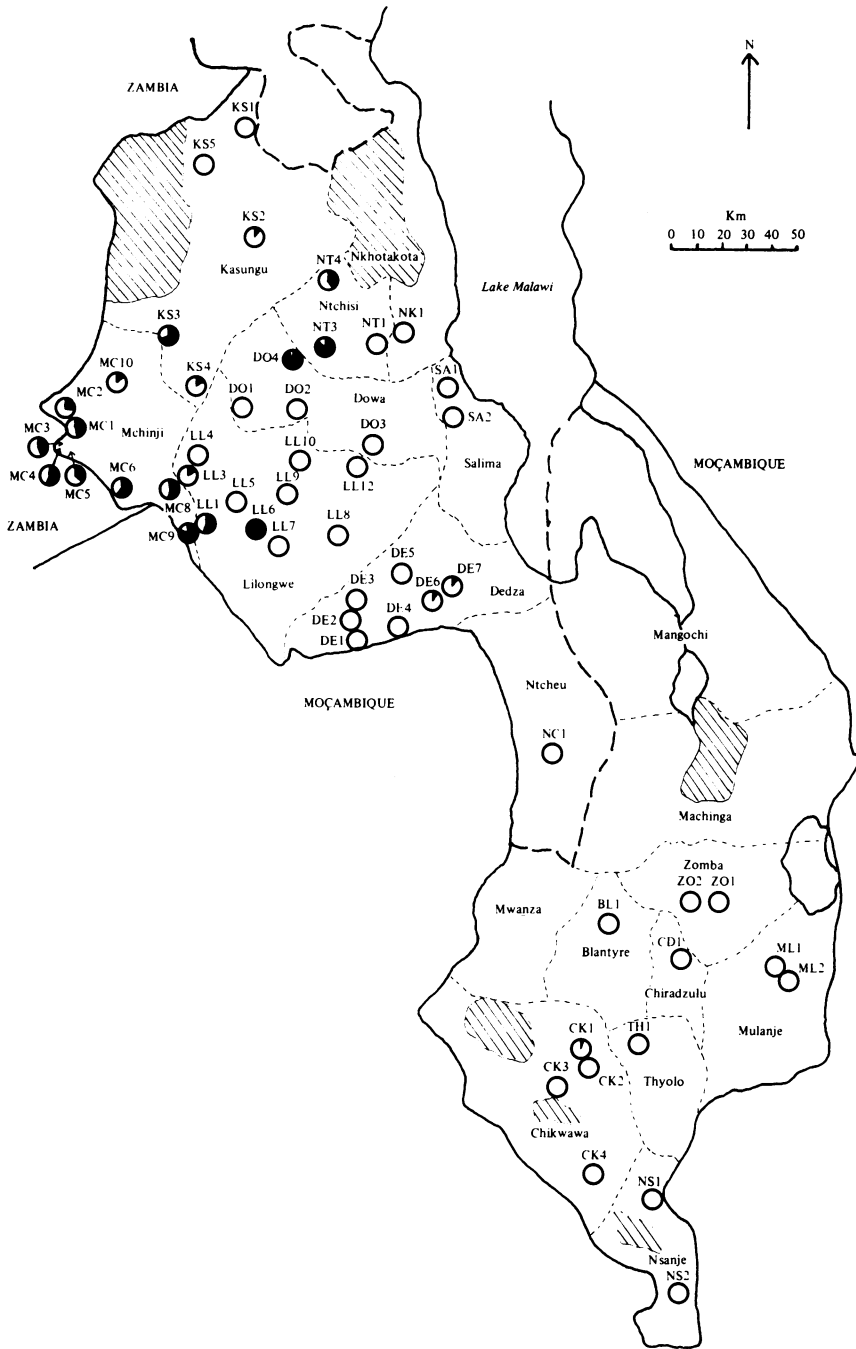


Fig. 2. Serological survey of free-ranging domestic pigs in the Central and Southern Regions of Malawi. ● LL1, Locality included in serological survey; shaded area shows proportion of sera positive. ---, Regional boundary. ·····, District boundary. ▨, National Park or Game Reserve.

Table 2. *ASF data from Mchinji district*

Locality	No. sera positive		Date collected	Interview data
	No. tested			
MC1	11/23 (48%)		Aug.-Sept. 82	No convincing reports of ASF outbreak Oct. 82 to Jan. 83. Mortality substantially less than 100%
MC2	5/21 (24%)		Sept. 82	
MC3	20/50 (40%)		June-Nov. 83	ASF outbreaks Jan.-June 83, Mar. 84. Mortality estimated 47-83% (see text)
MC4	6/7 (64%)		July 83	ASF outbreaks Oct. 81, Mar. 82, Mar. 83. Mortality substantially less than 100%
	1/4		May 84	
MC5	5/15 (33%)		June 83	ASF outbreaks Nov. 82, Mar. 83
MC6	12/23		Aug. 83	ASF outbreaks Oct. 82, Sept. 83*. > 130 pigs lost in 1983 outbreak
	15/16 (70%)		Oct.-Dec. 83	
	11/15		May 84	
MC7		No sera collected		ASF outbreak Oct. 82*
MC8	7/12 (58%)		June 83	ASF outbreak Oct. 82
MC9	1/12		July 83	ASF outbreak Nov. 83, 46 pigs lost. Mortality estimated 77-92% (see text)
	4/4		Nov. 83	
	35/41 (74%)		Jan.-Apr. 84	
	9/9		Aug. 84	Many seropositive pigs in apparently unaffected kholas
MC10	7/34 (21%)		July 84	ASF outbreaks Nov. 82, Oct. 83

* Laboratory-confirmed outbreak (other outbreaks were considered to be ASF on the basis of interview data only).

less than 100% (Table 2). In one village in MC3, 28 pigs died of ASF between January and June 1983. Thirty-two pigs were reported present in June but presumed infected since they were in affected kholas. In November 1983 only 10 of the 32 could be identified but all 10 were seropositive, indicating that at least 10 had survived primary infection. Had the other 22 all survived, the mortality would have been 47%; if they all died, mortality was 83%. ASF occurred again in March 1984 and among those which died were 2 which had survived the 1983 outbreak and been seropositive in November. In MC9, 46 pigs died of ASF in November 1983, and 14 from affected kholas were still alive at the end of November. Sera were collected from 5 of the 14 in January and were positive, indicating that mortality had been between 77% and 92%. Many of the other seropositive pigs were from apparently unaffected kholas. In some cases pigs had been sick but recovered. For example, in MC8 one of the seropositive pigs was reported by its owner to have been sick during the October 1982 outbreak but to have recovered; two pigs in the same khola died.

In Lilongwe district, seropositive pigs were identified in LL1, LL3 and LL6 (Table 3), all in the western part of the district. The material submitted to the laboratory from the confirmed outbreak in LL1 was from a khola in which 11 out of 16 pigs died. Sera collected from two of the remaining pigs approximately 4 months later were positive. If it is assumed that all five had been infected, the estimated mortality is 69%. In LL3 the interview data indicated less than 100% mortality but no detailed estimates were made. In LL6, a village survey conducted

Table 3. ASF data from Lilongwe district

Locality	No. sera positive		Interview data
	No. tested	Date collected	
LL1	8/14 (57%)	Mar.–June 83	ASF outbreak Nov. 82*, mortality estimated 69% (see text)
LL2	No sera collected		ASF outbreak June 84*
LL3	4/25 (16%)	Apr.–June 83	ASF outbreak Oct.–Dec. 82*, > 300 pigs lost (Haresnape, 1984). Mortality substantially less than 100%
LL4	0/21 (0%)	June–July 83	ASF outbreak Aug.–Nov. 81, > 200 pigs lost (Haresnape, 1984)
LL5	0/24 (0%)	Sept. 83	No reports of ASF
LL6	10/10 (100%)	Jan.–Apr. 84	ASF outbreak Aug.–Sept. 83, mortality estimated 85–90% (see text)
LL7	0/22 (0%)	Jan. 84	ASF outbreaks Oct. 83, Dec. 83
LL8	0/16 (0%)	Sept. 84	ASF outbreak Feb. 84*†
LL9	0/17 (0%)	Aug. 83	No reports of ASF
LL10	0/10 (0%)	Oct. 82	ASF outbreak July–Oct. 81*†, 96 pigs lost (Haresnape, 1984)
LL11	No sera collected		ASF outbreak July 84*
LL12	0/13 (0%)	Aug. 83	ASF outbreak Aug.–Nov. 81†, > 100 pigs lost (Haresnape, 1984)

* Laboratory-confirmed outbreak (other outbreaks were considered to be ASF on the basis of interview data only).

† Mortality estimated 100%.

Table 4. ASF data from Kasungu, Ntchisi and Dowa districts

Locality	No. sera positive		Interview data
	No. tested	Date collected	
KS1	0/24 (0%)	Aug. 83	No reports of ASF
KS2	3/25 (12%)	Aug. 83	No convincing reports of ASF
KS3	11/16 (69%)	Feb. 84	ASF outbreak Oct. 83, 129 pigs lost, mortality estimated 91–93% (see text)
KS4	2/14 (14%)	Aug. 83	No convincing reports of ASF
KS5	0/24 (0%)	Feb. 85	No reports of ASF
NT1	0/12 (0%)	Apr. 83	ASF outbreak July–Dec. 81†, > 600 pigs lost (Haresnape, 1984)
NT2	No sera collected		ASF outbreak Aug. 84*
NT3	19/23 (83%)	Aug. 84	ASF outbreak July 84
NT4	11/27 (41%)	Jan. 85	ASF outbreak Sept. 84
DO1	0/25 (0%)	Aug. 84	ASF outbreaks Aug.–Sept. 81*, Oct. 82
DO2	0/26 (0%)	Sept. 83	ASF outbreak Apr. 81*
DO3	0/17 (0%)	Aug. 84	ASF outbreaks June–July 81*, July–Aug. 84
DO4	20/21 (95%)	Aug. 84	ASF outbreak May 84, mortality substantially less than 100%

* Laboratory-confirmed outbreak (other outbreaks were considered to be ASF on the basis of interview data only).

† Mortality estimated 100%.

Table 5. ASF data from Dedza, Nkhotakota, Salima and Ntcheu districts

Locality	No. sera positive		Date collected	Interview data
	No. tested			
DE1	0/17 (0%) 0/23		Nov. 82 Aug. 84	ASF outbreaks Aug.–Nov. 82*†, 300 pigs lost (Haresnape, 1984), Mar. 83*, Mar. 84*†
DE2	0/26 (0%)		Aug. 84	ASF outbreaks Aug.–Sept. 82*†, July 84
DE3	0/19 (0%)		Aug. 84	ASF outbreaks Sept. 82*†, July–Aug. 84*
DE4	0/30 (0%)		Sept. 83	ASF outbreak Feb. 83*†, > 100 pigs lost
DE5	0/18 (0%)		Feb. 84	ASF outbreak Feb. 84*†, > 30 pigs lost
DE6	3/27 (11%)		Sept.–Oct. 83	ASF outbreaks Apr. 81*, Aug.–Oct. 82, Aug.–Sept. 83*†, > 60 pigs lost in 1983
DE7	2/15 (13%)		Nov. 83	ASF outbreak Oct. 83*†
DE8	No sera collected			ASF outbreaks Nov. 82*, May 84*
NK1	0/25 (0%)		Sept. 83	ASF outbreak 1981
SA1	0/21 (0%)		Sept. 83	No reports of ASF
SA2	0/25 (0%)		Sept. 83	No convincing reports of ASF
NC1	0/17 (0%)		July 84	No reports of ASF

* Laboratory-confirmed outbreak (other outbreaks were considered to be ASF on the basis of interview data only).

† Mortality estimated 100%.

in January 1984 revealed that 84 pigs had died between August and September 1983 and that 15 pigs in affected kholas were still surviving. Sera were collected from 10 of these and all 10 were positive. Therefore the estimated mortality is between 85% and 90%. In outbreaks in the eastern part of Lilongwe district, such as those in 1981 in LL10 and LL12 and in 1984 in LL8, the interview data indicated that mortality had been 100% in most affected kholas. In these localities no seropositive pigs were identified.

In Kasungu district, the only convincing reports of ASF were in KS3 (Table 4), where 129 pigs died in October 1983. Thirteen pigs in affected kholas survived and in February 1984 10 of these were seropositive and 3 seronegative, indicating that mortality was between 91% and 93%. Some seropositive pigs were identified in neighbouring localities KS2 and KS4, where there were only confused reports of disease which may not have been ASF. Seropositive pigs were also identified in two localities in Ntchisi district and one locality in Dowa district (Table 4).

In Dedza district, ASF was confirmed in the laboratory in all localities visited. Detailed interviews were conducted in most of these and all indicated that mortality had been 100% in affected kholas (Table 5). Three seropositive pigs were identified in DE6; 2 weeks later one of these had died of ASF and the other two had been slaughtered. Two more seropositive pigs were identified in DE7 but these were also slaughtered shortly afterwards. Since it is common practice in rural Malawi to slaughter pigs as soon as symptoms appear rather than wait for them to die naturally, it is possible that these pigs were suffering from ASF when they

Table 6. ASF data from Southern Region (Zomba, Blantyre, Chiradzulu, Thyolo, Mulanje, Chikwawa and Nsanje districts)

Locality	No. sera positive		Date collected	Interview data
	No. tested			
ZO1	0/22 (0%)		Sept. 83	No reports of ASF
ZO2	0/28 (0%)		Aug. 84	No reports of ASF in village pigs but an outbreak on an estate, Aug. 84* (see text)
BL1	0/18 (0%)		Sept. 83	Last laboratory-confirmed cases in Blantyre, Chiradzulu and Thyolo districts were 1979. No reports of ASF since
CD1	0/26 (0%)		Aug. 83	
TH1	0/23 (0%)		Aug. 83	
ML1	0/26 (0%)		Aug. 83	
ML2	0/15 (0%)		July 84	Last laboratory-confirmed case in Mulanje district was 1977. No reports of ASF since
CK1	1/9		Sept. 83	
	0/13 (5%)		July 84	ASF outbreak July–Aug. 83†, > 50 pigs lost
CK2	0/12 (0%)		Feb. 84	ASF outbreak Jan. 84†, > 80 pigs lost
CK3	0/23 (0%)		July 84	ASF outbreaks Aug.–Sept. 83†, > 90 pigs lost, Jan.–Mar. 84
CK4	0/26 (0%)		July 84	No reports of ASF
NS1	0/20 (0%)		July 84	ASF outbreak Oct. 83†
NS2	0/17 (0%)		July 84	No reports of ASF

* Laboratory-confirmed outbreak (other outbreaks were considered to be ASF on the basis of interview data only).

† Mortality estimated 100%.

were slaughtered. No seropositive pigs were identified in Nkhotakota, Salima or Ntcheu districts.

Southern Region

Of the 13 localities in the Southern Region which were studied between 1981 and 1984, only one (ZO2 in Zomba district) had a laboratory-confirmed outbreak of ASF, and this was on a commercial estate. In a further 4 localities (3 in Chikwawa and 1 in Nsanje district), there was evidence from interviews that ASF had occurred among free-ranging village pigs. In the other 8 localities there were no reports of disease.

Table 6 summarizes the data from the Southern Region. There were no reports of ASF in village pigs in Zomba, Blantyre, Chiradzulu, Mulanje or Thyolo districts. In the confirmed outbreak in ZO2 on a commercial estate the entire herd was slaughtered after 12 pigs died and ASF was confirmed in the laboratory. Four pigs had been sick and recovered, and apart from slight paralysis of the hindquarters appeared healthy at the time they were slaughtered. In addition to the 28 negative sera collected from free-ranging village pigs in this locality, a further 40 sera from pigs on two neighbouring estates were also negative.

In Chikwawa and Nsanje districts interviews provided evidence of ASF (Table 6). In one of the two villages in CK1 known to be affected, 32 pigs died between

July and August 1983, and 3 from affected kholas were still alive in September; of these 1 was seropositive and 2 seronegative. This was the only seropositive pig identified in the Southern Region.

There have never been any laboratory-confirmed cases of ASF in Mwanza, Mangochi or Machinga districts.

DISCUSSION

Most of the cases of African swine fever in Malawi between 1981 and 1984 were in the Central Region. Some outbreaks, particularly in the southern and eastern parts of the Region, have had similar characteristics to those reported previously in Africa, with mortality approaching 100% (DeTray, 1963; Hess, 1971). This is evident from field surveys in which pig owners in many affected localities in Dedza, Lilongwe and Ntchisi districts were interviewed following outbreaks of ASF. In some of these localities (DE1, DE2, DE3, DE4, DE5, DE6, DE7, LL8, LL10, LL12 and NT1) owners reported losing all their pigs within a short time, and most reported death within 2 or 3 days of the onset of symptoms. Nearly all sera collected in these and neighbouring localities following outbreaks of ASF were negative, implying that the pigs that were still present were those which had escaped infection altogether, and were not recovered animals. The only seropositive pigs identified in the localities listed above were five from Dedza district (from DE6 and DE7). These sera were collected within a few weeks of the first deaths in the area. One of the seropositive pigs died of ASF shortly afterwards, and the other four were apparently slaughtered. It is possible that these started showing symptoms of disease shortly before they were slaughtered and hence had been suffering from a subacute form of the disease. In other localities the interval between outbreak and serum collection was longer and pigs with the subacute form of disease, which usually causes death within about a month, would not have been present when serum samples were collected. Most cases of ASF which occurred in the southern and eastern parts of the Central Region between 1981 and 1984 thus appear to have been predominantly of the peracute and acute forms, with only a few pigs showing the subacute form of disease.

In other parts of the Central Region, particularly in the west, a different situation was encountered. Many pig owners reported that some pigs in affected kholas had survived, and many village pigs, selected at random in certain villages, were seropositive. It seems unlikely that a pig in the same khola as an affected animal could escape infection altogether, and assuming this to be the case, estimates of mortality were made from the interview data. These estimates in some localities (MC3, MC9, LL1, LL6 and KS3) were substantially less than 100%. In other localities (MC2, MC4, LL3, DO4, NT3 and NT4) it was clear that mortality was less than 100% although no detailed estimates were made. In all these localities many village pigs were seropositive. In some localities such as MC8 a pig owner identified an animal which had shown clinical signs of ASF such as fever, loss of balance and emaciation, but had recovered and was seropositive. This animal presumably had a chronic form of the disease. In other localities such as MC1, MC2 and MC9 pigs which had apparently not been sick were seropositive, and may therefore have had a subclinical form of ASF. Alternatively there may have been clinical signs which were ignored and followed by recovery. In localities MC1 and

MC2 seropositive pigs were identified although there had been no recent reports of disease at the time the sera were collected. Thus both the interview data and the serological survey showed that prolonged infections of ASF are common in Mchinji district and in parts of Kasungu, Ntchisi, Dowa and Lilongwe districts.

In the Southern Region there were fewer cases of ASF during the period of the study. This contrasts with the situation in previous years when more ASF was reported in the Southern than Central Region. There was insufficient data available to make any estimates of mortality from the outbreaks reported in Chikawa and Nsanje districts but only one seropositive pig was identified and this was later slaughtered. There is therefore no evidence for any chronic or subclinical forms of ASF in these districts. Of the pigs affected by the 1984 outbreak in Zomba district, four had recovered but were still slightly lame when slaughtered. These may have had a chronic form of the disease. The source of infection in this case is not clear, but since there was no evidence of ASF in neighbouring villages, and since the estate is situated close to the main road running north to south, the virus may have been introduced from a distant area.

The data presented here show that ASF has become enzootic throughout Mchinji district and in parts of Kasungu, Ntchisi, Dowa and Lilongwe districts. The occurrence of large numbers of seropositive pigs in these areas indicates that strains of ASF virus of reduced virulence are present. It is possible that the virus has become modified as a result of continuous association with domestic pigs, as was suggested by Leite Velho (1956) to explain the situation in Angola where Portuguese settlers kept their pigs in free-ranging herds (Scott, 1965*b*; Hess, 1981). A free-range system of management is common in Malawi; pigs feed by scavenging and are often only confined at night. Although this system is used throughout the country, it is only in the western part of the Central Region that a truly enzootic situation has developed. Elsewhere, it appears that pigs do not generally survive infection and that the percentage mortality in ASF outbreaks is higher. However, because of the free-range management system, the enzootic area could become larger in future. Once ASF virus strains of reduced virulence become widespread, as is now the case in parts of Europe, the disease is much more difficult to control because of the presence of apparently healthy carrier pigs which are potential sources of infection (Hess, 1981). It would be interesting to know if large numbers of seropositive pigs are present in neighbouring African countries such as Zambia and Mozambique where ASF is known to occur and where pigs are kept under free-range management (Anon, 1983; Scott, 1965*a*).

It has been considered likely that most ASF outbreaks in Malawi start following the introduction of infected meat from an affected area, since many pigs which suffer from the disease are slaughtered and sold to people from surrounding villages (Haresnape, 1984). However, it is also possible that some outbreaks represent a new incursion of virus from a natural reservoir.

Wild pigs (warthog, *Phacochoerus aethiopicus*, and bush pig, *Potamochoerus porcus*) and soft ticks, *Ornithodoros moubata* from warthog burrows are known to be reservoirs of ASF virus in East and South Africa (Plowright *et al.* 1969; Thomson *et al.* 1983). The possible role of these as reservoirs of virus in Malawi is currently being investigated. Bush pigs are more widely distributed in Malawi than warthogs and are common even outside protected areas (Hough, 1982). They often wander into settlements at night, and could therefore come into occasional contact with

domestic pigs. Lymph nodes from 3 bush pigs and sera and spleen from 11 bush pigs from various parts of the Central Region have been examined so far, but no evidence has been found of ASF infection in any of them. It will be necessary to examine many more animals before they can be ruled out as a possible reservoir. Warthogs are less widespread, and are rarely seen outside National Parks and Game Reserves (Hough, 1982). Sera were collected from 5 warthogs in Lengwe National Park in Chikwawa district in July 1984 and 4 were positive, indicating that warthogs are important in the epizootiology of ASF in Malawi. This is particularly interesting since Lengwe National Park is just south of locality CK3 where ASF was reported in 1983 and 1984 (Fig. 1 and Table 6).

There are many published records of occurrence of the soft tick *Ornithodoros moubata* in Malawi (Hoogstraal, 1956) and since this is a known vector of ASF, investigations are currently underway to assess its possible importance in the epizootiology of the disease in Malawi. ASF virus isolations have been made at the Animal Virus Research Institute, Pirbright, from ticks collected in pig kholas in localities MC2, MC3, MC4 and MC9, and this will be the subject of a later publication. Since all are in Mchinji district, it is interesting to speculate that there may be a connexion between the occurrence of infected ticks and the large proportion of seropositive pigs found in the same area. Investigations are currently underway to assess the extent to which *O. moubata* occur in pig kholas in Malawi and the virulence of the tick isolates.

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REFERENCES

- ANONYMOUS (1983). Zoo-sanitary situation in 1982. Reports presented during the 51st General Session of the OIE International Committee. Paris. 51.SG/12, item 4, pp. 271-275 (Moçambique) and pp. 375-380 (Zambia).
- COX, B. F. & HESS, W. R. (1962). Note on an African swine fever investigation in Nyasaland. *Bulletin of Epizootic Diseases of Africa* **10**, 439-440.
- DEKOCK, G., ROBINSON, E. M. & KEPPEL, J. J. G. (1940). Swine fever in South Africa. *Onderstepoort Journal of Veterinary Science and Animal Industry* **14**, 31-93.
- DETRAY, D. E. (1957). Persistence of viremia and immunity in African swine fever. *American Journal of Veterinary Research* **18**, 811-816.
- DETRAY, D. E. (1960). African swine fever - an interim report. *Bulletin of Epizootic Diseases of Africa* **8**, 217-223.
- DETRAY, D. E. (1963). African swine fever. *Advances in Veterinary Science* **8**, 299-333.
- HAMDY, F. M., COLGROVE, G. S., DERODRIGUEZ, E. M., SNYDER, M. L. & STEWART, W. C. (1981).

- Field evaluation of enzyme-linked immunosorbent assay for detection of antibody to African swine fever virus. *American Journal of Veterinary Research* **42**, 1441-1443.
- HARESNAPE, J. M. (1984). African swine fever in Malawi. *Tropical Animal Health and Production* **16**, 123-125.
- HESS, W. R. (1971). African swine fever virus. *Virology Monographs* **9**, 1-33.
- HESS, W. R. (1981). African swine fever: a reassessment. *Advances in Veterinary Science and Comparative Medicine* **25**, 39-69.
- HOOGSTRAAL, H. (1956). African Ixodidea. I. Ticks of the Sudan. *United States Naval Medical Research Report* N.M.005.050.29.07, p. 125.
- HOUGH, J. (Ed.) (1982). *Mammals of Malawi*, pp. 27, 30. Lilongwe: Environmental Unit, Department of National Parks and Wildlife.
- LEITE VELHO, E. (1956). Observations sur la peste porcine en Angola. *Bulletin de l'Office international des Epizooties* **46**, 335-340.
- MALMQUIST, W. A. & HAY, D. (1960). Hemadsorption and cytopathic effect produced by African swine fever virus in swine bone marrow and buffy coat cultures. *American Journal of Veterinary Research* **21**, 104-108.
- MATSON, B. A. (1960). An outbreak of African swine fever in Nyasaland. *Bulletin of Epizootic Diseases of Africa* **8**, 305-308.
- MENDES, A. M. (1961). Considerations sur le diagnostic et la prophylaxie de la peste porcine africaine. *Bulletin de l'Office internationale des Epizooties* **56**, 408-417.
- MENDES, A. M. & DASKALOS, A. M. (1955). Studies on the lapinisation of swine fever virus in Angola. *Bulletin of Epizootic Diseases of Africa* **3**, 9-14.
- MONTGOMERY, R. E. (1921). On a form of swine fever occurring in British East Africa (Kenya Colony). *Journal of Comparative Pathology* **34**, 159-191 and 243-262.
- PINI, A. (1977). African swine fever: some observations and considerations. *South African Journal of Science* **73**, 133-134.
- PINI, A. & HURTER, L. R. (1975). African swine fever: an epizootiological review with special reference to the South African situation. *Journal of the South African Veterinary Association* **46**, 227-232.
- PLOWRIGHT, W., PARKER, J. & PIERCE, M. A. (1969). The epizootiology of African swine fever in Africa. *Veterinary Record* **85**, 668-674.
- SANCHEZ-VIZCAINO, J. M., CROWTHER, J. R. & WARDLEY, R. C. (1983). A collaborative study on the use of the ELISA in the diagnosis of African swine fever. In *African Swine Fever*, CEC/FAO Research Seminar, Sardinia, 1981 (ed. P. J. Wilkinson), pp. 297-325. CEC Publication EUR 8466EN.
- SCOTT, G. R. (1965a). Prevention, control and eradication of African swine fever. *Bulletin de l'Office international des Epizooties* **63**, 751-764.
- SCOTT, G. R. (1965b). The smallest stowaways. I. African swine fever. *Veterinary Record* **48**, 1421-1427.
- STEYN, D. G. (1932). East African virus disease in pigs. *Eighteenth Report by the Director of Veterinary Services and Animal Industry*, vol. 1, pp. 99-109. Onderstepoort (South Africa).
- TABARES, E., FERNANDEZ, M., SALVADOR, E., CARNERO, M. E. & SANCHEZ BOTLJA, C. (1981). A reliable enzyme-linked immunosorbent assay for African swine fever using the major structural protein as antigenic reagent. *Archives of Virology* **70**, 297-300.
- THOMSON, G. R., GAINARU, M. D. & VAN DELLEN, A. F. (1979). African swine fever: pathogenicity and immunogenicity of two non-haemadsorbing viruses. *Onderstepoort Journal of Veterinary Research* **46**, 149-154.
- THOMSON, G., GAINARU, M., LEWIS, A., BIGGS, H., NEVILLE, E., VAN DER PYPEKAMP, H., GERBER, L., ESTERHUYSEN, J., BENGIS, R., BEZUIDENHOUT, D. & CONDY, J. (1983). The relationship between African swine fever virus, the warthog and *Ornithodoros* species in Southern Africa. In *African Swine Fever*, CEC-FAO Research Seminar, Sardinia, 1981 (ed. P. J. Wilkinson), pp. 85-100. CEC Publication EUR 8466EN.
- WARDLEY, R. C., ABU ELZEIN, E. M. E., CROWTHER, J. R. & WILKINSON, P. J. (1979). A solid-phase enzyme-linked immunosorbent assay for the detection of African swine fever antigen and antibody. *Journal of Hygiene* **83**, 363-369.
- WARDLEY, R. C., ANDRADE, C. M., BLACK, D. N., DE CASTRO PORTUGAL, F. L., ENJUANES, L., HESS, W. R., MEBUS, C., ORDAS, A., RUTILI, D., SANCHEZ-VIZCAINO, J. D. & WILKINSON, P. J. (1983). African swine fever virus: brief review. *Archives of Virology* **76**, 73-90.