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Risk factors for farm-level African swine fever infection in major pig-producing areas in Nigeria, 1997–2011

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ABSTRACT

African swine fever (ASF) is an economically devastating disease for the pig industry, especially in Africa. Identifying what supports infection on pig farms in this region remains the key component in developing a risk-based approach to understanding the epidemiology of ASF and controlling the disease. Nigeria was used for this matched case–control study, because there is perpetual infection in some areas, while contiguous areas are intermittently infected. Risk factors and biosecurity practices in pig farms were evaluated in association with ASF infection. Subsets of farms located in high-density pig population areas and high-risk areas for ASF infection were randomly selected for analysis. Most plausible risk factor variables from the univariable analysis included in the multivariable analysis include: owner of farm had regular contact with infected farms and other farmers, untested pigs were routinely purchased into the farm in the course of outbreaks, there was an infected neighbourhood, other livestock were kept alongside pigs, there was a presence of an abattoir/slaughter slab in pig communities, wild birds had free access to pig pens, tools and implements were routinely shared by pig farmers, there was free access to feed stores by rats, and feed was purchased from a commercial source.

Only the presence of an abattoir in a pig farming community (OR = 8.20; $CI_{95\%}$ = 2.73, 24.63; $P < 0.001$) and the presence of an infected pig farm in the neighbourhood (OR = 3.26; $CI_{95\%}$ = 1.20, 8.83; $P = 0.02$) were significant. There was a marginally significant negative association (protective) between risk of ASF infection and sharing farm tools and equipment (OR = 0.35; $CI_{95\%}$ = 0.12, 1.01; $P = 0.05$).

Of the 28 biosecurity measures evaluated, food and water control (OR = 0.14; $CI_{95\%}$ = 0.04, 0.46; $P < 0.001$), separation/isolation of sick pigs (OR = 0.14; $CI_{95\%}$ = 0.04, 0.53; $P = 0.004$) and washing and disinfection of farm equipment and tools (OR = 0.27; $CI_{95\%}$ = 0.10, 0.78; $P = 0.02$) were negatively associated (protective) with ASF infection. Consultation and visits by veterinarian/paraveterinarians when animals were sick (OR = 8.11; $CI_{95\%}$ = 2.13, 30.90; $P = 0.002$), and pest and rodent control were positively associated with ASF infection of Nigerian farms (OR = 4.94; $CI_{95\%}$ = 1.84, 13.29; $P = 0.002$).

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The presentation of sick and unthrifty pigs for slaughter at abattoirs, farmers' inadvertent role, an infected neighbourhood, a pig to pig contact, rodents and wild birds may contribute to infections of farms, whereas washing, disinfection of tools, food and water control, and separation of sick pigs reduces the likelihood of infections. Underlying reasons for these observations and strategies for control are discussed.

1. Introduction

The African swine fever virus (ASFV), an *Asfivirus* of the *Asfarviridae* family, continues to spread across Nigerian farms, causing sporadic outbreaks of African swine fever (ASF), with associated mortalities. Historically, the virus first made an apparently unsustained incursion into Nigeria in 1973, wiping out the infected pig herd and then becoming extinct (Owolodun et al., 2010). However, there was a resurgence of outbreaks in West Africa from 1996 onwards, with the virus entering Nigeria in August/September 1997. ASF has remained a problem in Nigerian piggeries since then (see Fig. 1a and b). Persistent infections with ASFV appear to recur in the core pig-producing areas of the country. A theory of geographical contiguity (in other words, infection in one state is highly likely to cause an outbreak in the neighbouring state(s)) has been proposed and is supported by the incidence of infections (see Fig. 1a and b).

Nigeria, like many West and Central African countries where ASF is endemic, experiences intermittent infections. Very recently, in 2011, the Republics of Chad, Kenya, Cameroun, Tanzania and Malawi were infected, with huge fatalities in pigs and consequently significant loss of income and employment opportunities (OIE, 2011). Indeed, such infections have an overall effect on the pig industry worldwide, because they limit opportunities to explore external markets and because of the potential spread of the disease and the increasing rapidity/possibilities of inter-continental contamination. Some of the previously ASF-free areas of the world, including parts of Russia and the Caucasian region, are now experiencing repeated infections of ASF (Rowlands et al., 2008; OIE, 2011).

Intensive efforts have been made in the use of genetic epidemiology to analyse the ASF viruses circulating in different parts of Africa in order to gain an significant understanding of the relation between and geographic spread of each circulating genotype (Bastos et al., 2003, 2004; Lubisi et al., 2005, 2007; Boshoff et al., 2007; Gallardo et al., 2009, 2011; Owolodun et al., 2010). However, the causes/factors that support the continued circulation of ASF viruses in pig herds in various parts of Africa in general and in Nigerian pig populations in particular remain poorly understood or at best hypothetical.

Only three types of epidemiological cycles have been described for ASFV to date:

(i) an ancient sylvatic cycle that primarily involves warthogs (*Phacochoerus africanus*) and argasid ticks of the genus *Ornithodoros*, with occasional spill-over to domestic pigs;
(ii) a cycle in domestic pigs that involves *Ornithodoros* ticks inhabiting pig sties; and
(iii) a cycle in domestic pigs which occurs without the involvement of sylvatic hosts or vectors (Penrith et al., 2004).

ASFV has previously been detected as a spill-over infection via *Ornithodoros sonrai* ticks in Senegal (Vial et al., 2007) and from wild suids in Nigeria (Luther et al., 2007a), but the first two cycles have not been widely linked to the epidemiology of ASF in West Africa. Thus, a greater understanding of the factors responsible for the continued presence and maintenance of the virus in domestic pig populations (without the agency of sylvatic hosts and tick vectors) is vital for achieving regional control and eradication.

In order to investigate the risk factors for ASF in Nigerian pig herds and to identify high-risk farms, we carried out a case–control study that focused on environmental risks and biosecurity in pig herds under various farming conditions in the hope that the results of this analysis will inform the formulation of policies to support ASF control efforts and reduce the burden of ASF in West Africa.

2. Materials and methods

2.1. Study locations and mapping of the spread of ASF, 1997–2009

Samples from suspected ASF outbreaks submitted to the National Veterinary Research Institute (NVRI) in Vom, Nigeria, as well as samples obtained through two active surveillance programmes in which both suspect and apparently healthy pigs were evaluated between 2006 and 2009, form the basis of this study. A total of 1279 sera and 1332 pooled tissues (767 tissues from the 2006 to 2007 active surveillance; 269 tissues from the 2002 to 2007 passive surveillance; and 296 tissues from the 2006 to 2009 active surveillance) were collected from 19 states (see Fig. 1a and b) and were analysed at the NVRI diagnostic laboratories. Duplicate samples of selected tissues and all sera were dispatched to the Centro de Investigación en Sanidad Animal (CISA-INIA) in Valdeolmos, Madrid, Spain, for quality control and duplicate confirmation of positive and negative samples. These results were supplemented by data from peer-reviewed literature and commissioned reports on ASF outbreaks in Nigeria between 1997 and 2009 (El-Hicheri, 1998; Luther et al., 2007b; Owolodun et al., 2007, 2010; Fasina et al., 2010). The data were first filtered to exclude duplications and were then combined for the purposes of the spatio-temporal mapping of suspected and confirmed outbreaks (see Fig. 1a and b).

(a)

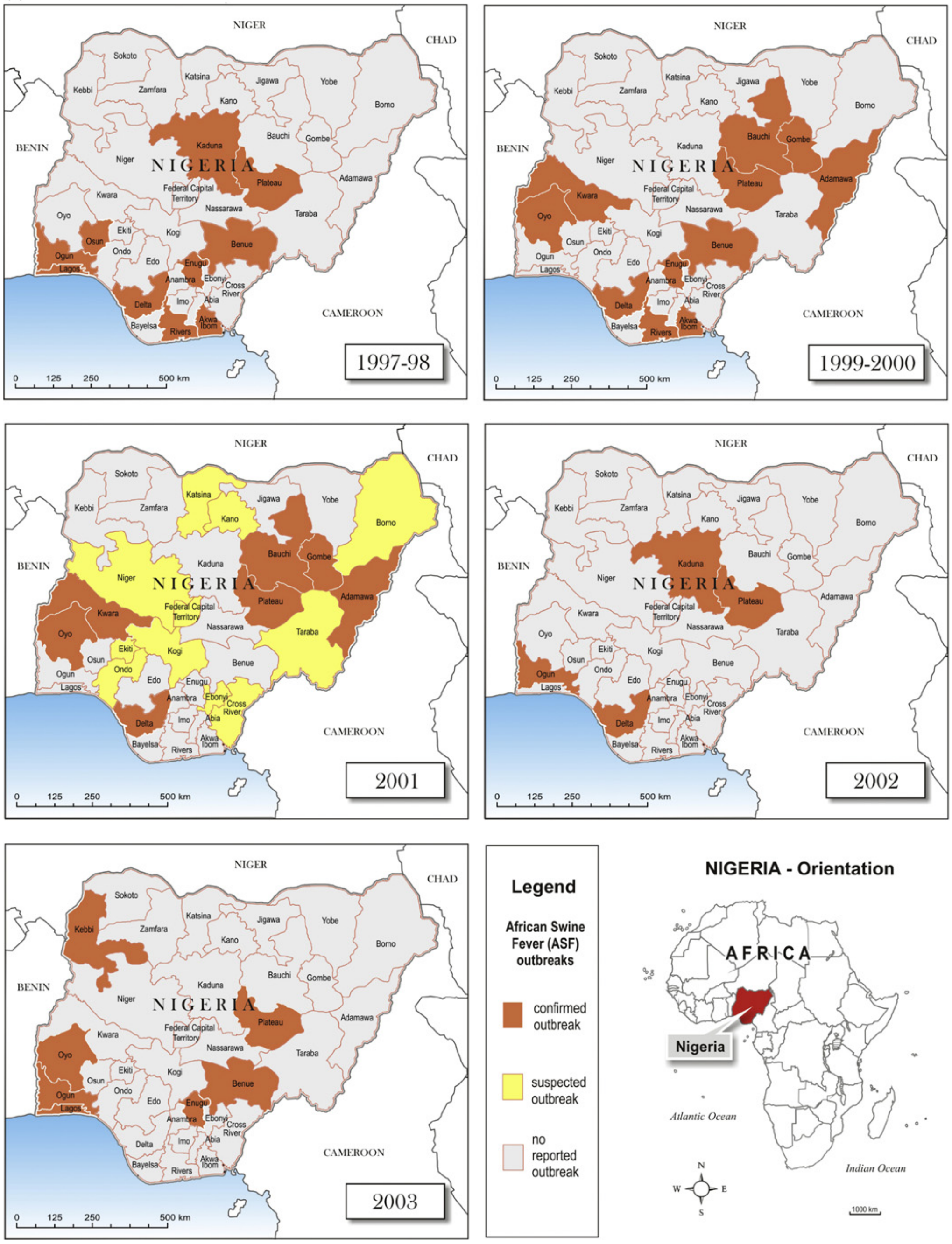

Fig. 1. (a) Spatio-temporal representation of reported and confirmed outbreaks of ASF outbreaks in Nigerian states, 1997–2003. The index outbreak occurred in the Ogun/Lagos axis in September 1997 and spread to other locations, eventually covering 9 states. Since these initial outbreaks, ASF has been reported (yellow) and confirmed (red) annually in Nigeria. It should be noted that several farmers may slaughter their sick pigs without reporting the outbreak. In that case, the true spatial prevalence of yearly intermittent and sporadic outbreaks may cover larger areas than those represented on the map. (b) Spatio-temporal representation of reported and confirmed outbreaks of ASF outbreaks in Nigerian states, 2004–2009. Outbreaks were confirmed by means of combinations of clinico-pathological findings and laboratory analyses (iELISA, immunoblotting assay, immunofluorescence, PCR and virus isolation). Maps were drawn based on the reports of El-Hicheri (1998), Luther et al. (2007b), Owolodun et al. (2007, 2010), Fasina et al. (2010) and the Annual Reports of the National Veterinary Research Institute, Nigeria.

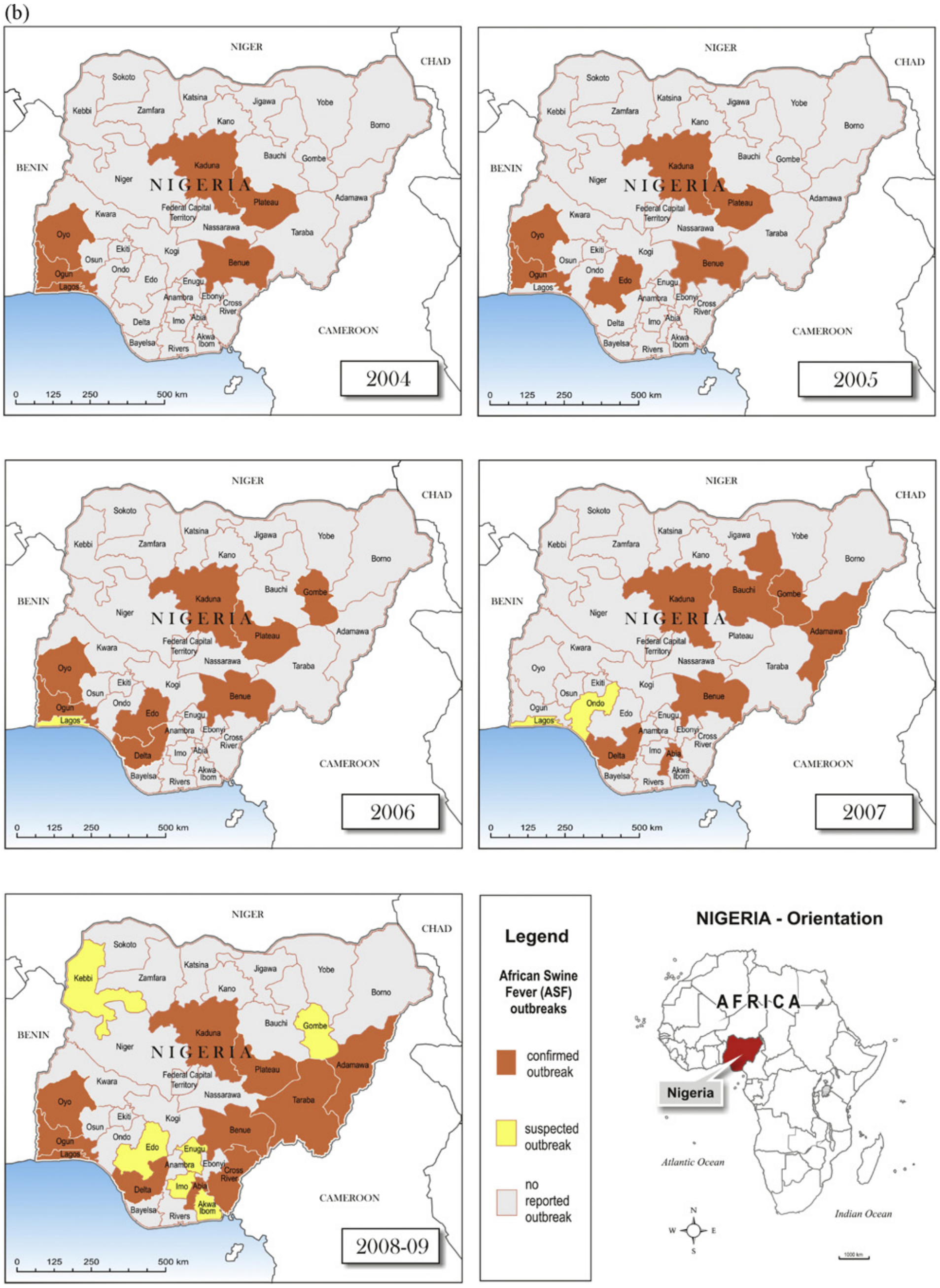

Fig. 1. (*Continued*).

From the list of high-risk locations investigated, four states were selected for inclusion in a matched case–control study. These included Imo (in south-east Nigeria), Kebbi (in north-west Nigeria), Lagos (in south-west Nigeria) and Taraba (in north-east Nigeria). The chosen states are representative of the distributions of outbreak locations and pig populations in Nigeria, and they were chosen as subsets of high-risk locations and high density pig areas. Within the selected states, case and control farms were subsequently selected as indicated below.

2.2. *Case farm definition*

Cases were defined in accordance with the international regulations for confirming ASF (OIE, 2008). Briefly, a farm is considered a potential case farm if it meets the following criteria:

(i) clinical signs consistent with ASF infection – high fever, depression, loss of appetite, heightened abortion, sudden death and loss of body condition;

(ii) pathological signs – extensive haemorrhage of the visceral organs, including the lymph nodes, spleen and kidneys; and
(iii) one or more animals from the farm being diagnosed positive for the presence of the ASF viral genome by polymerase chain reaction (PCR), in combination with at least one of the four diagnostic tests: indirect ELISA, immunoblotting assay, immunofluorescence assay and virus isolation.

Some of the case farms reported repeated outbreaks of ASF between 1997 and 2010. Detailed results of the tests have been reported by Fasina et al. (2010). All the case farms included in the study were selected randomly from among the farms confirmed positive for the presence of the ASF genome, antibodies or virus between 2008 and 2011 ($n = 343$). They originated from four states (Imo, Kebbi, Lagos or Taraba). A total of 120 questionnaires were sent out to collect data from case farms, but only 72 farms finally qualified for inclusion as case farms. Reasons for the elimination of the responses from the remaining 48 farms included inconsistent reports and double entries and/or incomplete entries on the questionnaires. A further three case farms were eliminated because no matched control farms (see below) were available for them, leaving 69 case farms for the analysis.

2.3. Control farm description

Control farms were matched with case farms on the basis of farm location (Imo, Kebbi, Lagos or Taraba) and farm population size (<50 pigs; 51–100 pigs or >100 pigs). Eligible controls were farms which were within the infected or surveillance zones of ASF-infected farms and which were at risk of infection due to close proximity (within 500 m and up to a 5 km perimeter) to an infected farm. These farms had similar population characteristics to the case farms (see Table 1) and were clustered geographically, like the case farms. Samples from these farms were collected at the same time as those from the case farms, but the samples tested negative for the ASF genome, antibodies or virus, using a combination of the clinico-pathological and laboratory diagnostic tests mentioned above.

A control farm was a pig farm under the management of one farmer with one or more pigs managed together as a group, where animals were at risk of infection with ASF, but consistently tested negative to ASF both serologically and virologically for the duration of the study period. Additional qualifications for eligibility included the presence of pigs on the farm between 2008 and 2011, when the case farms were sampled, and confirmation of the independent management of the control and case farms. Of the 120 questionnaires sent out, 86 were returned, but missing data rendered 26 unusable, leaving 60 control farms for the analysis.

2.4. Data collection: the questionnaire

Epidemiological data were gathered by means of a self-rated closed-ended questionnaire completed by farmers. Prior to the administration of the closed-ended questionnaire, farmers met in groups and the purpose of the questionnaire was discussed. Each farmer was then asked to fill in the questionnaire, without interference, at his/her individual farm to avoid personal and diplomatic biases. Matched variables (based on farm size and location) were collected and grouped in categories:

(i) farm characteristics;
(ii) farm operations; and
(iii) self-reported biosecurity measures (see Appendix A).

2.5. Statistical analysis

Each potential risk factor and biosecurity measure was coded as a dichotomous independent variable. The odds of being an ASF case based on serology and virology was then modelled as a function of the dichotomous risk factors and biosecurity measures, using conditional logistic regression models, as suggested by Hosmer and Lemeshow (1989). The initial screening of potential risk factors for ASF infection and biosecurity measures to prevent infection was performed using univariable conditional logistic regression.

Variables associated with the outcome (ASF virus infection) at $P \leq 0.2$ were considered for inclusion in the multivariable conditional logistic regression models. Independent variables were tested for pairwise associations, using a two-tailed chi-square test. Two multivariable conditional logistic regression models were developed: one for the risk factors and one for the biosecurity measures. A backward selection procedure was applied using a selection threshold of $P \leq 0.05$ to reduce the number of variables in the model. All the excluded variables were then individually re-tested in the model and retained if they were significant.

Farm population size was then entered into each model as a continuous variable to test for residual confounding effects and was retained if it resulted in more than a 10% change to the coefficient for any of the other remaining predictors. Interactions between farm size and each of the remaining predictors were also tested and retained in the model if they were significant.

The fit of the final models was assessed using the Akaike information criterion (AIC) and the Bayesian information criterion (BIC), since the Hosmer–Lemeshow goodness-of-fit test is inappropriate for conditional logistic regression models, and the m:n matching precluded the use of leverage and influence statistics. In the final models, the odds ratios (ORs), P-values and 95% confidence interval were reported. All statistical analyses were done using Stata 11 (StataCorp, College Station, Texas, USA).

3. Results

3.1. Spatial and temporal patterns

The mapping of laboratory-confirmed cases of ASF revealed that some locations were perpetually infected, while states contiguous to those locations were intermittently infected (see Fig. 1a and b). The localities defined as "perpetually infected" for

Table 1
Population characteristics of the case and control farms participating in the study of AFS in Nigeria, 2008–2011.

Designation	Counts	Herd size							Locations and descriptions
	Ratio: 1.15:1	Mean ± SD	Min.	Max.	25th percentile	50th percentile	75th percentile	95th percentile	
Case	69	98.48 ± 51.77	11	306	65	90	135	180	Selected from a lists of clusters of farms in Imo, Kebbi, Lagos and Taraba
Control	60	95.43 ± 47.96	12	195	52.5	89.5	134	177	Selected from a lists of clusters of farms in Imo, Kebbi, Lagos and Taraba

Data for the risk assessment periods were collected between October 2008 and April 2009 (39 datasets), and February and June 2011 (93 datasets). Several of the large communities of pig farms in Nigeria are grouped in clusters/cooperatives for the purposes of accessing services and marketing facilities jointly.

the period of study coincided roughly with high-density pig producing and marketing areas.

3.2. Case–control study

The matching pattern in the final dataset used for analysis was *m*:*n*, in other words, one or more case farms were matched with one or more control farms. There were 11 matched groups, with between 1 and 14 case farms and between 2 and 10 control farms per group. The population characteristics of the case and control farms are shown in Table 1.

The results of the univariable analysis of risk factors are set out in Table 2. The following variables were selected for inclusion in the multivariable model: the owner of the selected farm has regular contact with infected farms and other farmers on such farms, routine purchase of mostly untested pigs which are brought to the farm in the course of outbreaks, an infected neighbourhood, the keeping of other livestock alongside pigs, the presence of an abattoir/slaughter slab in pig communities, wild birds having free access to pig pens, tools and implements routinely being shared by pig farmers, free access to feed stores by rats, and the purchasing of feed from a commercial source (Table 2).

The final conditional logistic regression model for the risk factors is shown in Table 3.

The presence of an abattoir in the pig farm area was strongly associated with increased odds of ASF infection (OR = 8.20; $CI_{95\%}$ = 2.73, 24.63; P < 0.001). In addition, pig farms were at higher risk of infection if there was an infected pig farm present in the neighbourhood (OR = 3.26; $CI_{95\%}$ = 1.20, 8.83; P = 0.02). However, there was a marginally significant negative (protective) association between the risk of ASF infection in pig communities and the sharing of farm tools and equipment (OR = 0.35; $CI_{95\%}$ = 0.12, 1.01; P = 0.05).

For the self-reported biosecurity measures, based on the univariable analysis, several factors were selected for inclusion in the multivariable model (see Tables 4 and 5). The final conditional multivariable analysis (see Table 5) shows that only food and water control (OR = 0.14; $CI_{95\%}$ = 0.04, 0.46; P < 0.001), separation or isolation of sick pigs (OR = 0.14; $CI_{95\%}$ = 0.04, 0.53; P = 0.004) and washing and disinfection of equipment and tools (OR = 0.27; $CI_{95\%}$ = 0.10, 0.78; P = 0.02) showed negative (protective) associations with ASF infection. Consultation with and visits by veterinarians or paraveterinarians when animals were sick (OR = 8.11; $CI_{95\%}$ = 2.13, 30.90; P = 0.002), as well as pest and rodent control measures (OR = 4.94; $CI_{95\%}$ = 1.84, 13.29; P = 0.002), were positively associated with ASF infection of farms.

4. Discussion

In the current study, two sets of factors were studied with regard to the risk of ASF infection on pig farms in Nigeria, namely [A] farm environment and management practices, and [B] self-reported biosecurity practices. The former are contributory factors which may predispose farms to a higher risk of infection with the ASF virus while the latter are practices (hygiene and good management) that farmers reported to have taken to reduce the risk of these infections (FAO/OIE, 2010).

[A1]. The presence of an abattoir in pig communities was the risk factor that influenced ASF infection the most (OR = 8.20; $CI_{95\%}$ = 2.73, 24.63; P < 0.001). This observation can probably be ascribed to a number of factors, including the following:

1. The farmers tend to present sick and unthrifty pigs for slaughter at abattoirs first, without determining the cause of sickness, some of which may be ASF (Randriamparany et al., 2005; Fasina et al., 2010). Since the ASF virus is present in the tissues and body fluids of slaughtered sick pigs, massive environmental contamination and possible farm infection may result.
2. Rats and wild birds are usually observed near an open abattoir environment. When intestinal content and viscera, which are sometimes infectious, are indiscriminately disposed of, they may be carried to nearby pig farms by these scavengers, thereby facilitating the infection of naïve pigs.
3. Farmers often participate in various processes on abattoir floors with the consequent risk of farm infection.

[A2]. The presence of an infected farm in a neighbourhood was also significantly associated with the infection of farms (OR = 3.26; $CI_{95\%}$ = 1.20, 8.83; P = 0.02). This is related directly to a local spread between and within pig farms and may occur through direct pig-to-pig contact, especially in scavenging populations, by spreading through fomites, and possibly by tick vectors (although no tick vector has been associated with ASF in Nigeria to date). Mannelli et al.

Table 2
Univariable conditional logistic regression analysis of risk factors associated with presence of ASF outbreaks on pig farms, Nigeria, 2008–2011.

Variable/risk factor	Category	Cases *n* (%)	Controls *n* (%)	OR	95% CI	*P*-value
Sale method	Community/abattoir	34 (49.3)	28 (46.7)	1.00	–	–
	Market	35 (50.7)	32 (53.3)	0.82	0.40, 1.65	0.57
Contact infected farm	No	39 (56.5)	27 (45.0)	1.00	–	–
	Yes	30 (43.5)	33 (55.0)	0.64	0.32, 1.27	0.20
Purchased pigs routinely without testing during outbreaks	No	41 (59.42)	43 (71.67)	1.00	–	–
	Yes	28 (40.58)	17 (28.33)	1.72	0.81, 3.66	0.16
Infected neighbourhood	No	22 (31.88)	50 (83.33)	1.00	–	–
	Yes	47 (68.12)	10 (16.67)	8.52	3.81, 19.05	<0.001
Keep other animals on the farm	No	55 (79.71)	53 (88.33)	1.00	–	–
	Yes	14 (20.29)	7 (11.67)	2.18	0.77, 6.19	0.14
Abattoir/slaughter slabs within pig communes	No	14 (20.29)	50 (83.33)	1.00	–	–
	Yes	55 (79.71)	10 (16.67)	20.85	7.80, 55.75	<0.001
Visceral and intestinal contents disposed of indiscriminately	No	23 (33.33)	17 (28.33)	1.00	–	–
	Yes	46 (66.67)	43 (71.67)	1.26	0.59, 2.71	0.55
Wild bird enter pig pens	No	33 (47.83)	46 (76.67)	1.00	–	–
	Yes	36 (52.17)	14 (23.33)	3.57	1.64, 7.76	0.001
Ticks observed on pigs/premises	No	67 (97.10)	55 (91.67)	1.00	–	–
	Yes	2 (2.90)	5 (8.33)	0.39	0.07, 2.23	0.29
Share farm tools with other farms	No	49 (71.01)	13 (21.67)	1.00	–	–
	Yes	20 (28.99)	47 (78.33)	0.11	0.05, 0.26	<0.001
Use treated water	No	44 (63.77)	37 (61.67)	1.00	–	–
	Yes	25 (36.23)	23 (38.33)	0.99	0.48, 2.07	0.99
Rats have access to feed store and pig pens	No	11 (15.94)	30 (50.00)	1.00	–	–
	Yes	58 (84.06)	30 (50.00)	4.77	2.07, 10.97	<0.001
Buy commercial feed/compound	Commercial	14 (20.29)	39 (65.00)	1.00	–	–
	Self milling	55 (79.71)	21 (35.00)	0.14	0.06, 0.31	<0.001

(1997) and Costard et al. (2009a) have similarly reported that free-range pigs and local pig movement were associated with the spread of ASF in previous studies. Effort must therefore be made to reduce the networks, connectivity and neighbourhood-mediated spread of ASF (Rivas et al., 2010; Firestone et al., 2011).

[A3]. In our analysis, the sharing of tools was marginally negatively associated with the spread of infection ($OR = 0.35$; $CI_{95\%} = 0.12, 1.00$; $P = 0.05$). Since it is logical that tool-sharing may exacerbate the spread of the disease from one location to another, the reason for this observation was not immediately clear. However, in the analysis of the biosecurity measures, the washing/disinfection of tools was also negatively associated with the spread of ASF. It is possible that tools shared between farms are washed and disinfected more often, hence, the negative association (protection) observed. The practice of sharing tools and equipment will continue for the foreseeable future amongst small scale farmers who may not afford some of the farm equipment.

We evaluated 28 self-reported biosecurity measures. Only five had some association with ASF infection in the final multivariable model. These were food and water control, the separation or isolation of sick pigs, and the washing and disinfection of farm tools and equipment, all of which were negatively associated with ASF seropositivity. Consultation with and visits by veterinarians or paraveterinarians when animals are sick, and pest and rodent control were all positively associated with the risk of seropositivity of pig farms.

[B1]. Food and water control significantly reduced the risk of ASF in this analysis ($OR = 0.14$; $CI_{95\%} = 0.04, 0.46$; $P < 0.001$). Since the introduction of food and swill is an important means of transmission of pig diseases (Horst et al., 1997; El-Hicheri, 1998), a carefully planned and isolated feed store and covered water storage remain important parts of a comprehensive biosecurity programme. Such storage facilities also have the advantage of excluding contamination by rodents and wild birds. Contaminated feed and water have played role in the spread of ASF in West Africa in the past (El-Hicheri, 1998).

[B2]. Separation or isolation of sick pigs from healthy ones ($OR = 0.14$; $CI_{95\%} = 0.04, 0.53$; $P = 0.004$) was found to be equally important in the prevention of ASF. Infected pigs can shed a large amount of ASF virus, especially naso-pharyngeally, and these viruses may remain in the environment for a long time (FAO, 2009). Hence, the

Table 3
Multivariable conditional logistic regression analysis of risk factors associated with ASF virus infection in a matched case–control study of pig farms, Nigeria, 2008–2011.

Variable/risk factor	Category	OR	95% CI	*P*-value
Infected neighbourhood	No	1.00	–	–
	Yes	3.26	1.20, 8.83	0.02
Abattoir/slaughter slabs in pig communes	No	1.00	–	–
	Yes	8.20	2.73, 24.63	<0.001
Share farm tools with other farms	No	1.00	–	–
	Yes	0.35	0.12, 1.01	0.05

Table 4
Univariable conditional logistic regression analysis of the association between self-reported biosecurity practices and the presence of ASF outbreaks on pig farms, Nigeria, 2008–2011.

Variable/biosecurity measure	Category	Case (%)	Control (%)	OR	95% CI	*P*-value
Restricted access	No	24 (34.78)	14 (23.33)	1.00	–	–
	Yes	45 (65.22)	46 (76.67)	0.48	0.20, 1.11	0.09
Fence around premises	No	23 (33.33)	18 (30.00)	1.00	–	–
	Yes	46 (66.67)	42 (70.00)	0.79	0.37, 1.69	0.54
Gate at entrance	No	25 (36.23)	18 (30.00)	1.00	–	–
	Yes	44 (63.77)	42 (70.00)	0.66	0.30, 1.45	0.30
Foot bath/dips present	No	23 (33.33)	10 (16.67)	1.00	–	–
	Yes	46 (66.67)	50 (83.33)	0.35	0.15, 0.84	0.02
Change solution in foot pans regularly[a]	No	20 (28.99)	11 (18.33)	1.00	–	–
	Yes	49 (71.01)	49 (81.67)	0.57	0.25, 1.29	0.18
Records kept	No	18 (26.09)	10 (16.67)	1.00	–	–
	Yes	51 (73.91)	50 (83.33)	0.54	0.22, 1.29	0.16
Food and water control	No	28 (40.58)	12 (20.00)	1.00	–	–
	Yes	41 (59.42)	48 (80.00)	0.30	0.13, 0.71	<0.01
Quarantine of newly purchased pigs	No	30 (43.48)	32 (53.33)	1.00	–	–
	Yes	39 (56.52)	28 (46.67)	1.38	0.68, 2.79	0.37
Terminal cleaning (end-of-cycle cleaning)	No	33 (47.83)	20 (33.33)	1.00	–	–
	Yes	36 (52.17)	40 (66.67)	0.52	0.25, 1.08	0.08
Routine cleaning	No	13 (18.84)	12 (20.00)	1.00	–	–
	Yes	56 (81.16)	48 (80.00)	1.08	0.45, 2.59	0.87
Cleaning and disinfection of drinkers and feeders	No	23 (33.33)	28 (46.67)	1.00	–	–
	Yes	46 (66.67)	32 (53.33)	1.60	0.79, 3.23	0.19
Wash/disinfect equipment and tools	No	29 (42.03)	19 (31.67)	1.00	–	–
	Yes	40 (57.97)	41 (68.33)	0.61	0.29, 1.28	0.19
Remove manure and litter routinely	No	15 (21.74)	8 (13.33)	1.00	–	–
	Yes	54 (78.26)	52 (86.67)	0.55	0.22, 1.41	0.22
Prompt disposal of dead animals	No	25 (36.23)	15 (25.00)	1.00	–	–
	Yes	44 (63.77)	45 (75.00)	0.52	0.23, 1.16	0.11
Safe disposal of faeces and carcasses	No	17 (25.00)	8 (13.33)	1.00	–	–
	Yes	51 (75.00)	52 (86.67)	0.42	0.16, 1.10	0.08
Sufficient feeding and drinking space	No	11 (15.94)	9 (15.00)	1.00	–	–
	Yes	58 (84.06)	51 (85.00)	0.81	0.31, 2.12	0.67
Sufficient space for pigs (prevent overcrowding)	No	12 (17.39)	7 (11.67)	1.00	–	–
	Yes	57 (82.61)	53 (88.33)	0.61	0.23, 1.64	0.33
Use disinfectants	No	31 (44.93)	35 (58.33)	1.00	–	–
	Yes	38 (55.07)	25 (41.67)	1.63	0.81, 3.28	0.17
Do not mix pigs of different ages	No	26 (37.68)	11 (18.33)	1.00	–	–
	Yes	43 (62.32)	49 (81.67)	0.32	0.14, 0.75	<0.01
All-in all-out system	No	55 (79.71)	50 (83.33)	1.00	–	–
	Yes	14 (20.29)	10 (16.67)	1.18	0.48, 2.90	0.72
Move from young to old pigs	No	33 (47.83)	18 (30.00)	1.00	–	–
	Yes	36 (52.17)	42 (70.00)	0.48	0.23, 0.98	0.04
Change rubber boots	No	21 (30.43)	14 (23.33)	1.00	–	–
	Yes	48 (69.57)	46 (76.67)	0.58	0.25, 1.32	0.20
Change clothes to go in and out	No	37 (53.62)	29 (48.33)	1.00	–	–
	Yes	32 (46.38)	31 (51.67)	0.78	0.38, 1.58	0.48
Separate/isolate sick pigs	No	22 (31.88)	10 (16.67)	1.00	–	–
	Yes	47 (68.12)	50 (83.33)	0.40	0.17, 0.95	0.04
Consultation and visits of veterinarian/paraveterinarians when animals were sick	No	18 (26.09)	25 (41.67)	1.00	–	–
	Yes	51 (73.91)	35 (58.33)	2.02	0.91, 4.50	0.08
Downtime of >2 weeks	No	41(59.42)	47 (78.33)	1.00	–	–
	Yes	28 (40.58)	13 (21.67)	2.55	1.15, 5.65	0.02
Pest and rodent control	No	33 (47.83)	43 (71.67)	1.00	–	–
	Yes	36 (52.17)	17 (28.33)	2.50	1.20, 5.20	0.02
Evaluate and audit biosecurity measures periodically	No	42 (60.87)	40 (66.67)	1.00	–	–
	Yes	27 (39.13)	20 (33.33)	1.25	0.61, 2.59	0.54

[a] 46 case farms had a foot bath/dip and an additional three farmers used improvised pans in place of a foot dip, making a total of 49.

contamination of other pigs is highly likely if an infected pig is retained in the pig herd. Since domestic pig-to-pig contact remains the only proved means of transmission of ASF in Nigeria and West Africa, it is desirable to remove all uninfected pigs from infected/sick pigs to cut off the continued infection of farms.

[B3]. Washing and disinfection of farm equipment and tools was also negatively associated with ASF infection and seropositivity (OR = 0.27; $CI_{95\%}$ = 0.10, 0.78; P = 0.02). Some farm implements are shared between abattoirs and farms, especially in situations where an abattoir is sited inside a pig facility. These implements include shovels, knives, cutlasses, brooms, waste bins, wheelbarrows, etc. Heavily contaminated tools may be returned to the farm without disinfection, and these become sources of infection to naïve pigs.

Table 5
Multivariable conditional logistic regression analysis of self-reported biosecurity practices against ASF outbreaks on pig farms, Nigeria, 2008–2011.

Variable/biosecurity measure	Category	OR	95% CI	*P*-value
Food and water control	No	1.00	–	–
	Yes	0.14	0.04, 0.46	<0.001
Separate/isolate sick pigs	No	1.00	–	–
	Yes	0.14	0.04, 0.53	0.004
Consultation and visits of veterinarians/paraveterinarians	No	1.00	–	–
when animals are sick	Yes	8.11	2.13, 30.90	0.002
Wash/disinfect equipment and	No	1.00	–	–
tools	Yes	0.27	0.10, 0.78	0.02
Pest and rodent control	No	1.00	–	–
	Yes	4.94	1.84, 13.29	0.002

[B4]. The consultation with and visits of veterinarians or paraveterinarians to farms when animals are sick was positively associated with ASF infection of farms (OR = 8.11; $CI_{95\%}$ = 2.13, 30.90; P = 0.002). There are two possible explanations for this observation. Firstly, farmers usually only call in veterinarians or paraveterinarians when everything else (management procedures and the administration of antibiotics) has failed. The visit of a veterinarian/paraveterinarian is therefore more likely to be a consequence of an ASF outbreak than it is a predisposing factor. It is possible that, during a visit, more animals than those observed as clinically sick are already infected, and these animals will continue to spread infection after the visit unless they are removed alongside the sick animals. Secondly, the course of outbreaks of a disease such as ASF is often a crisis period and a veterinarian/paraveterinarian visiting a cluster of pig farms is likely to visit more than one farm/day in such situation. Tools for the administration of drugs and sampling may be shared, and clothing and shoes may not be changed in-between farms. This inadvertent error may therefore also spread infection to other farms subsequently visited in the course of disease management. Strict observance of biosecurity in-between movement to farms is encouraged by professionals.

[B5]. Finally, pest and rodent control were positively associated with ASF infection on farms in Nigeria (OR = 4.94; $CI_{95\%}$ = 1.84, 13.29; P = 0.002). Farmers do not usually implement/intensify rodent or pest control programmes unless they have problems with these vectors. These rodents/pests may contaminate feed and water, including the pig premises, with remnants taken from abattoir floors, which may predispose farms to ASF infection. Intermittent implementation of pest control programmes may also lead to abnormal local fluctuations in pest populations, which will in turn lead to increased pest movement between farms and a resulting increased risk of disease transmission. Finally, if farmers perceive rodents to be a risk factor for disease, then farmers may implement a rodent control programme in response to an outbreak on or near their farm; therefore the control programme is a consequence of the outbreak rather than of the fact that the presence of rodents is a risk factor.

4.1. Spatial and temporal patterns of ASF outbreaks in Nigeria

ASF appears to infect pig farms in Nigeria in a pig trade-related pattern. The outbreaks that started in the Ogun–Lagos axis in 1997–98 were linked to Benin, due to commercial pig-related activities along the border between Nigeria and Benin (El-Hicheri, 1998). Prior to this outbreak, Côte d'Ivoire (1996) and Benin (1997) were infected, and a regional early warning was sent to neighbouring countries to prevent the further spread of infection. However, a porous border, poor veterinary services, legal and illegal trade in pig products across the border, poor disease reporting systems and poor preparation, supported infections and subsequent outbreaks of ASF in Nigeria.

Infection rapidly spread from the Lagos–Ogun axis to some parts of south-west, south–south, south-east and north-central Nigeria, strictly following the trade routes of pigs in the country (El-Hicheri, 1998; Fasina et al., 2009). To date, periodic outbreaks have been found in these locations and pig movement continues to follow the same pattern (see Fig. 1a and b). Etter et al. (2011) has previously established a similar pattern of infection between Guinea Bissau, Senegal and Gambia – finished pigs are moved north-west to Dakar (a major consumption area), but these pigs are raised adjacent to the enzootic locations of Guinea Bissau and Gambia. Hence, high seroprevalence was obtained in pigs. Thus these trade movements played a critical role in the epidemiology of ASF in that part of West Africa.

Since infection in one area appears to have a contiguous effect on neighbouring areas (see Fig. 1a and b), it will be important to use a region-based approach to control the spread of infectious diseases such as ASF, in addition to farm-based biosecurity. Such approach will benefit the control of African swine fever and other infectious diseases.

This study was based on the serological and virological results obtained in past surveillance (Fasina et al., 2010). However, because a disease such as ASF is a dynamic system, it will be necessary to determine changes in the epidemiological picture regularly and also to check whether or not the proposed methods are having an impact, using a sustained surveillance system that should itself be evaluated periodically. The continuing surveillance and evaluation of risk factors supporting infection of pig farms in West Africa remains the key component in the development of a risk-based approach to understanding the epidemiology of ASF in the sub-region (Etter et al., 2011).

Biosecurity is a set of measures that are interlinked with one another and with good husbandry/management practices. Husbandry practices and management styles used on farms should be evaluated to determine good hygiene practices that will suit Nigerian-type piggeries (Costard et al., 2009b).

Our study was subject to a number of limitations, including the possibility of some types of bias. Every effort was made to reduce confounding bias by

- matching for farm population size and locations;
- restricting the study period and area to reduce model bias and confounding factors; and
- using a multivariable conditional logistic regression model to control for confounding between the measured predictors.

Self-report bias was another potential source of error in the study. We are aware that farmers may have wanted to give "socially acceptable" responses to the questions and that the level of self-reported biosecurity may have been at variance with the actual implementation, as reported in past studies (Nespeca et al., 1997; Casal et al., 2007). However, where possible, we observed farm management and biosecurity practices which were static and straightforward (such as fences, restricted access, records, disposal pits, abattoirs), and used them as a check against questionnaire responses, as recommended by Stärk et al. (1998).

In addition, the concept of biosecurity may have different interpretations to different farmers in terms of its comprehensiveness and content, but this lack of precision was addressed and reduced through the open fora and large group discussions held before the administration of questionnaires, where some agreement was reached. In this study, the problem of recall bias was considered to be negligible, since ASF was an ongoing infection in almost all the case farms selected for the study, and the control farms were fully aware of its presence. Spatial bias was also managed further by matching location. The selection of control farms around the case farms was random.

This study avoided the use of face-to-face interviews, because we wanted to eliminate professional bias, a situation where the interviewer's own concept of biosecurity (because of professional training) might be passed on and could influence the farmers' answers. The problems that may be associated with unsupervised questionnaire administration were minimized by the use of simple and unambiguous questions, pre-administration discussions and closed-ended questions.

5. Conclusions

The following conclusions have been reached in this study:

It is likely that the presence of an infected pig farm in the same neighbourhood and the presence of abattoirs and associated practices will increase the likelihood of ASF infection of farms. This also applies to the presence of vermin and wild birds in the pig farm community. However, strict food and water control, the immediate separation (isolation) of sick pigs from healthy pigs, and the washing and/or disinfection of farm equipment will assist in reducing the chances of infection. Region-based control of infectious diseases, together with farm-based biosecurity, will assist in controlling future outbreaks of ASF in Nigeria and West Africa.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.prevetmed.2012.05.011.

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