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Full Length Research Paper

Foot-and-mouth disease (FMD) prevalence and exposure factors associated with seropositivity of cattle in north-central, Nigeria

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This study was designed to determine the seroprevalence and risk factors associated with foot-and-mouth disease (FMD) seropositivity in north central, Nigeria. A cross-sectional study was undertaken from February 2013 to April 2014 using 1206 sera from 150 herds collected by multi-stage and random sampling methods. Pre-tested questionnaire were also administered to participating farmers to collect information on the animal herd structure, movement pattern, management system and herds contact at watering points. Samples collected were tested for evidence of FMD antibodies using the 3ABC non-structural antibodies enzyme-linked immunosorbent assay (ELISA). The overall seroprevalence of FMD in North-Central Nigeria was found to be 70.98% (95% CI: 68.37-73.49). FMD seroprevalence was found to be higher in Niger State 85.4% (95% CI: 83.46-88.03) relative to Plateau State 54.2% (95% CI: 50.12-58.16), which was statistically associated with FMD seropositivity (P<0.05). Risk factors such as sex, management system, trans-boundary crossing and herd mixing at the watering point were found to be statistically associated with FMD seropositivity (p<0.05). This confirms that FMD is enzootic in the study area and control of foot and mouth disease in Nigeria using animal movement control and vaccination is therefore advocated.

Key words: Foot-and-mouth disease (FMD), prevalence, endemic, serotypes, enzyme-linked immunosorbent assay (ELISA), antibodies.

INTRODUCTION

Foot and mouth disease (FMD) has been recognized as an important trans-boundary animal disease impacting negatively on the cattle industry since the sixteenth century (Mahy, 2005). FMD is caused by foot and mouth disease virus (FMDV) of the genus *Aphthovirus,* family *Picornaviridae.* Seven distinct serotypes namely: A, O, C, Asia-1, SAT-1, SAT-2, and SAT-3 have been identified. It is known that infection with one serotype does not confer

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immune protection against another serotype. Different subtypes can be identified within a serotype by biochemical and immunological tests (OIE, 2012). The disease is known to exhibit high fever, loss of appetite, salivation, and vesicular eruptions on the feet, mouth and teats of lactating cows (Thomson, 1995a). FMD has a broad host range, high degree of infectivity, rapid replication rate and multiple transmission routes, which makes it very difficult and expensive to control and eradicate (Alexandersen and Mowat, 2005). The disease has a high morbidity although mortality is low in adult animals. However, myocarditis may occur in young animals resulting in death. The recovered animals may remain in poor physical condition over long periods of time leading to economic losses for livestock industries (Molla et al., 2010).FMD is endemic in most of sub-Saharan Africa, except in a few countries in southern Africa, where efforts were made to control the disease by the separation of wildlife from susceptible livestock using barrier veterinary cordon fencing in combination with prophylactic vaccination (Vosloo et al., 2002). Furthermore, due to the endemicity of the disease, and the fact that FMD does not normally cause high mortality in adult animals, FMD outbreaks are not often perceived as important and are poorly reported or investigated further to determine the causative serotypes. However, this is now changing, a number of countries are now recognizing FMD as one of the most important trans-boundary animal diseases that should be controlled in order to access profitable international markets for livestock and livestock by-products as well as to maximize the full genetic potential of the animals (Ayelet et al., 2009).

There is a dearth of information on the actual situation of FMD in Nigeria and the neighbouring countries. There are regular outbreaks, no national control strategy, no enforcement of legislation for disease reporting to veterinary authorities, and animal movement control are poor. Since most of the cattle populations in Nigeria are from the neighboring countries of West and Central Africa, the animals are at perpetual risk of infection from the endemic strains as well as antigenic variants prevalent in neighboring countries. Studies have shown that, FMD serotypes O, A, SAT 1 and SAT 2 have circulated in Nigeria between 1924 and 2009 (Lazarus et al., 2012; Fasina et al., 2013; Olabode et al., 2013; Nawathe and Goni, 1976; Owoludun, 1971). However, recent sampling conducted between 2007 and 2009 have indicated that despite the endemicity of FMD in Nigeria with its attendant production losses in livestock, very little is known about the epidemiology of FMD in Nigeria (Fasina et al., 2013). The prevalence of FMD can be determined serologically by measuring the antibody level to the 3ABC nonstructural protein (NSP) (De Diego et al., 1997). The objectives of this study were to determine the seroprevalence of FMD and to identify the risk factors associated with seropositivity of FMD in cattle from the north-central Nigeria.

MATERIALS AND METHODS

Study area

The North Central Nigeria is located at the central point of Nigeria (Middle belt). The zone is populated by mostly minority ethnic groups. It is characterized by Guinea savannah and marked by crystalline rock outcroppings and gently rolling hills such as the Jos Plateau. The major or notable rivers in Nigeria, River Niger and Benue meet at the region precisely at Lokoja town in Kogi State. The two major seasons are the raining season from the mouth of April through October and a dry season from November through March. The temperature is also relative from state to state as it is relatively cold weather in Jos Plateau while other states have predominantly hot weather condition. The zone has six states namely: Plateau, Niger, Nassarawa, Kogi, Benue and Kwara states. The geo-political zone has human population of 20,266,256 (Anon, 2013) and cattle population of 2,363,369 (Kogi 367,754, Kwara 66,905, Nassarawa 88,532, Niger 803,013, Plateau 976,029 and Benue 61,136) (GLIPHA, 2011).

The predominant economic activities are farming and fishing as a result of their fertile nature of land and the presence of river Niger and Benue around Kogi, Benue and part of Niger and some other related areas near the riverine environs, mining amongst Jos Plateau people. Because of the abundance of grassland in the zone, it supports a massive population of livestock and serves as the major cattle treks routes to the Eastern and Southern Nigeria. The region also shares International boundaries to the West with Benin Republic through Niger and Kwara states (Felix, 2009).

Study animals and sampling technique for serum collection

Study animals were selected from the animal population in two states of the region, namely, Plateau and Niger states. The states were selected based on their geographical location, proximity with the livestock market, ruminant population density, movement pattern, as well as cattle trek route and International boundary (Figure 1). Individual animals were randomly selected so that about 10% of animals from each herd were sampled to represent the herd, in total 150 cattle herds were sampled by multi-stage and random sampling method in two states of the North-central Nigeria.

The sample size for the seroprevalence study was determined by assuming a prevalence of 56.3% based on a previous study (Ishola et al., 2011). The sample size was determined using a simple random sampling method of Thrusfield (2005) with 95% confidence interval and desired precision of 0.05. The calculated sample size was 378. However, to improve precision, the sample size was increased by 3-fold and a total of 1250 cattle were sampled in this study.

The potential risks factors for FMD in the study area were assessed by a pre-tested structured questionnaire in all the states. The questionnaire was designed to assess the most important factors that could be associated with FMD such as animal location, management system, mixing at the watering point, animal movement pattern and international boundary crossing.

Whole blood was collected from the jugular vein using a 10 ml sterile plain vacutainer tubes and stored overnight at room temperature for serum separation. Each serum was transferred into a sterile cryovials, bearing the age and sex of sampled animal and was transported in a cold box to FMD Research Laboratory, National Veterinary Research Institute, Vom Nigeria, and stored at 20°C until use. FMD seroprevalence was estimated using 3ABC ELISA (Bronsvoort et al., 2006).

Ethical approval

For this type of study, formal consent is not required. All applicable

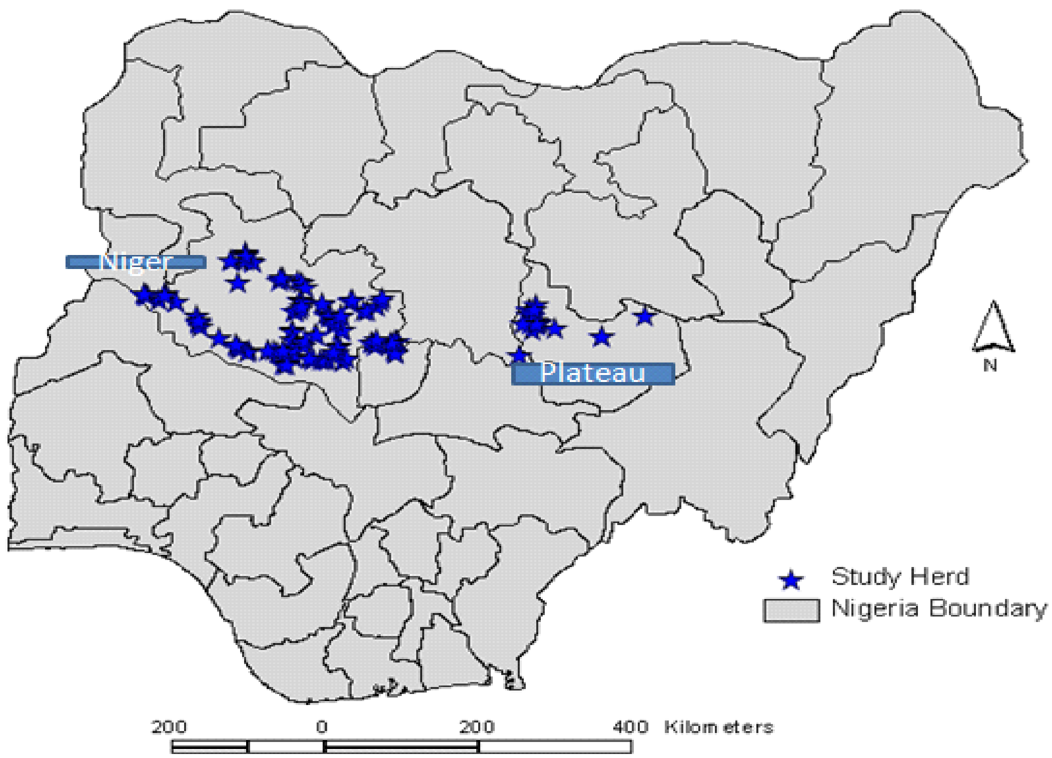

Figure 1. Map of Nigeria showing the distribution of the sampled herds in the North Central Nigeria.

international and national guidelines for the care and use of animals where followed.

Study design

A cross-sectional study was undertaken from February 2013 to April 2014, during which a total of over 1250 blood samples were collected, however, 1206 sera were used for laboratory analysis using 3ABC Non-structural protein ELISA. The questionnaire was designed to collect information on the animal movement pattern, management system and mixing at watering points.

Serum sample collection

Samples were stored overnight at room temperature for serum separation. Each serum was transferred into a sterile cryovial, bearing the age and sex of sampled animal and was transported in an icebox to National Veterinary Research Institute, Vom, Nigeria (NVRI), and stored in freeze at -20°C until analyses. The assay was conducted at the Foot and Mouth Research Centre, National Veterinary Research Institute, Vom, Nigeria.

Detection of antibodies against FMDV non-structural proteins (NSPs) ELISA

All the 1206 sera were subjected to FMD screening using the PRIOCHECK FMD-3ABC NS protein ELISA (NSP-ELISA) (PrioCHECKS® PrionicsLelystad Netherland). The PRIOCHECK FMD-3ABC NS protein ELISA kit is designed to detect FMDV specific antibodies in bovine serum (Sørensen et al., 1998). The test was useful because it was able to discriminate animals that had been infected (wild virus induced antibodies) from those that had been vaccinated with purified vaccine (vaccine induced antibodies).

The ELISA serology was performed according to the manufacturer's instructions for (PrioCHECKS® Prionics Lelystad Netherland) (Sørensen et al., 1998). Briefly described, 80 µl of the ELISA buffer and 20 µl of the test sera were added to the 3ABC-antigen coated test plates. Negative, weak positive and strong positive control sera were added to designated wells on each test plate, gently shook and incubated overnight (18 h) at 22°C. The plates were then emptied and washed six times with 200 µl of washing solution and 100 µl of diluted conjugate were added to all the wells. The test plates were sealed and incubated for 60 min at 22°C. The plates were then further washed six times with 200 µl of the washing solution and 100 µl of the chromogen (Tetra-Methyl Benzidine) substrate was dispensed to all wells of the plates and incubated for 20 min at 22°C following which 100 µl of stop solution was added to all the wells and mixed gently. Readings were taken on a spectrophotometer Multiskan® ELISA reader (Thermo Scientific, USA) at 450 nm and the OD450 values of all samples was expressed as percentage inhibition (PI) relative to the OD450 max using the following formula PI = 100 – [OD450 test sample/OD450 max] × 100. Samples with PI = ≥ 50% were classified as positive while those with PI < 50% were declared negative. Since the 3-ABC ELISA for FMD was = 100% specific and > 99% sensitive, the percentage prevalence was taken as true prevalence (Sørensen et al., 1998; Bronsvoort et al., 2006).

Data collection and analysis

The data were stored in Microsoft Excel ® and coded for analysis.

Table 1. Seroprevalence of FMD based on state distribution.

State	Number of sera tested	Number of sera positive	Prevalence (%) (95%CI)	
Niger	617	537	85.4	(83.46-88.03)
Plateau	589	319	54.2	(50.12-58.16)
Total	1206	856	70.98	

X^2=156.4; p-value= < 0.05.

Table 2. Seroprevalence of FMD based on geographical zones

Geographical zones	Number of sera tested	Seropositivity 3ABC ELISA	Prevalence (%) (95%CI)	
Plateau North	215	94	43.12	(37.2-50.4)
Plateau Central	107	59	55.14	(45.64-64.36)
Plateau South	267	166	62.17	(56.24-67.84)
Niger South	195	162	83.08	(77.35-87.86)
Niger East	240	204	85	(80.06-89.1)
Niger North	182	171	93.9	(89.74-96.8)
Total	1206	856		

X^2=184; df = 5; p < 0.05.

Seroprevalence was calculated on the basis of 3ABC ELISA test results. Serological data was subjected to statistical analysis using SPSS (version 13) and Open Epi (Version 2.3.1). Chi-square (χ^2) was used to assess the existence of association with FMD seropositivity. The associations of individual categories of each exposure factor with seropositivity of FMD were analyzed using univariable logistic regression. This univariable analysis assumed all other factors were constant and one category was used as a reference. In all the statistical analysis, confidence interval was set at 95%.

RESULTS

The overall seroprevalence of FMD in north-central Nigeria was found to be 70.98% (95% CI: 68.37-73.49). This was found to be higher in Niger State 85.4% (95% CI: 83.46-88.03) relative to Plateau state 54.2% (95% CI: 50.12-58.16) (Table 1) and the difference in prevalence was statistically associated with FMD seropositivity (P<0.05). Seroprevalence based on different geographical zones revealed that Niger North recorded the highest seroprevalence of 93.9% (95% CI: 89.74-96.8), followed by Niger East 85% (95% CI: 80.06-89.1), Niger South 83.08% (95% CI: 77.35-87.86), Plateau South, 62.17% (95% CI: 56.24-67.84), Plateau Central, 55.14% (95% CI: 45.64-64.36), and the lowest prevalence was recorded in Plateau North, 43.12% (95% CI:37.2-50.4).

The difference in prevalence across geographical zones was statistically associated with seropositivity (χ^2=184, df=5,p-value< 0.05).

Risk factors such as sex, management system, trans-boundary crossing and herd mixing at the watering point were found to be positively associated with FMD seropositivity (p<0.05).

Seroprevalence across geographical zones

Seroprevalence across the six geographical zones are presented in Table 2 and Figure 2 with a higher sero-prevalence of 93.9% (95%CI: 89.74-96.8) for Niger North, while Plateau North recorded the lowest prevalence of 43.12% (95%CI: 37.2-50.4), which was statistically significant (p<0.05).

Seroprevalence based on age category

Age of animals sampled were analyzed in two categories < 2years (Young) and >2 years (Adult). The sero-prevalence in cattle aged >2 years was higher (70.01%) than in cattle aged <2 years (67.7%). The difference in seroprevalence was, however, not statistically associated with the age of the cattle (p<0.05). The odd of FMD seropositivity was relatively more in cattle aged >2 years (1.14) than in cattle aged <2 years old (Table 3).

Seroprevalence based on sex category

Higher disease prevalence was observed more in females (71.9%) relative to the males (30.4%). The difference in prevalence between the two sex groups was found to be statistically significant (χ^2 =129.1; p>0.05). The odd ratio of FMD was 4.78% (3.6-6.54) times in females than males (Table 4)

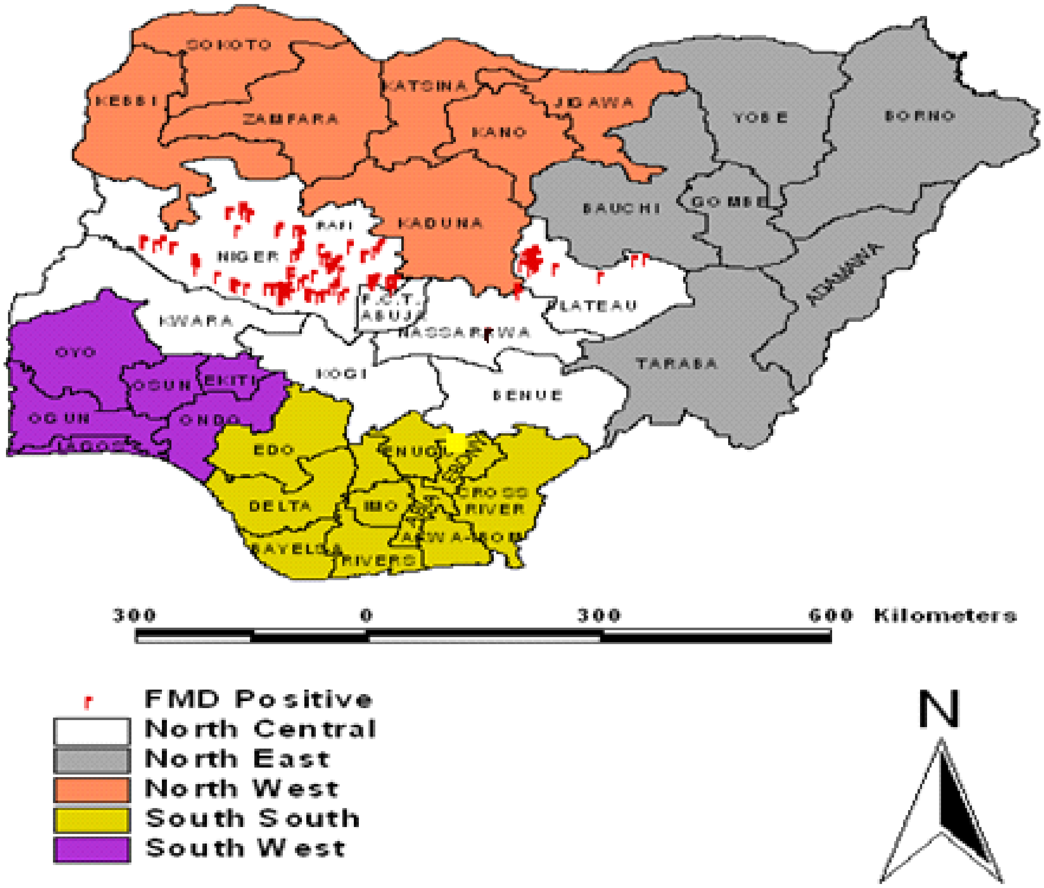

Figure 2. Distribution of herds positives for FMD in the study area.

Table 3. Seroprevalence of FMD based on age category.

Age	Number	%	Serological status		Prevalence (%) (95%CI)	OR
			+ve	-Ve		
Adult	767	72.5	537	230	70.01 (66.7-73.2)	1.14 (0.833-1.489)
Young	291	27.5	197	94	67.7 (62.2-72.9)	1
Total	1058	100				

X^2=0.5324; p>0.05.

Seroprevalence based on management systems

The study revealed higher disease prevalence in nomadic management system (75.8%) followed by sedentary management system (66.3%) and a lower prevalence was observed in the intensive management system (1.8%). The difference in prevalence between the nomadic and sedentary management system was not statistically significant (χ^2=10.79; p>0.05). However, the difference in disease prevalence among the three management systems was statistically associated with FMD sero-positivity (χ^2=123; p<0.05).

The odd of FMD in nomadic and sedentary husbandry systems was 171.9 (23.54-12.56) times more than intensive management system (Table 5). Management system showed a positive association with FMD seropositivity as risk factor.

Seroprevalence based on cattle movement

The study showed a higher prevalence of FMD sero-positivity in cattle that cross national boundary (88.75%) than those that move within the country (57.7%) and the

Table 4. Seroprevalence of FMD based on sex distribution.

Sex	Number	%	Serological status		Prevalence % (95%CI)	OR
			+Ve	- Ve		
Male	322	30.4	112	210	34.8(29.7-40.1)	1
Female	736	69.6	529	207	71.9 (66.5-75)	47.8(3.6-6.54)
Total	1058	100				

χ^2=129.1; p <0.05.

Table 5. Seroprevalence of FMD based on management system.

Management system	Number	%	Serological status		Prevalence % (95%CI)	OR
			+Ve	-Ve		
Sedentary	511	48.3	339	172	66.3 (62.2-70.3)	9.3 (4.885-19.02)
Nomadic	491	46.4	372	119	75.8 (71.8-79.4)	1
Intensive	56	5.3	1	55	1.8 (0.089-8.50)	
Total	1058	100				

χ^2=125.4; Df=2 p-value <0.05.

Table 6. Seroprevalence of FMD based on trans-boundary border crossing.

Trans-boundary crossing	Number	%	Serological status			OR
			+Ve	-Ve	Prevalence % (95% CI)	
Yes	417	39.4	370	47	88.7 (85.4-91.5)	5.184(3.696-7.369)
No	641	60.6	370	244	57.7(53.8-61.5)	1
Total	1058	100				

χ^2= 99.35; p< 0.05.

difference in FMD seropositivity was statistically significant (χ^2=99.35; P<0.05). The odd of FMD in cattle crossing the national boundary is 5.184 (3.699-7.369) times greater than those that move within the country (Table 6).

Seroprevalence based on herd mixing at the watering point

The study revealed higher FMD seropositivity in herds that indicated mixing at the watering points (75.8%) than those that do not mix at watering point (1.8%). The difference in seropositivity was statistically significant (χ^2=143.9; p<0.05). The odd of FMD is 171.8 (23.8-1253) times greater in herds that mixed at water points than those that do not (Table 7).

A survey to determine the seroprevalence of FMD in abattoir and cattle market as FMD hot spots was also conducted. The prevalence in abattoir samples was 65.7% and for the cattle market sample was 69.1%, respectively. The findings revealed that seroprevalence of FMD in these study areas were statistically insignificant. This could be attributed to the fact that most of the cattle population slaughtered in Nigeria abattoirs are directly purchased from the local cattle markets (Table 8).

DISCUSSION

FMD is one important trans-boundary animal disease (TAD) that limits prospects in local livestock production in Nigeria, with outbreaks occurring seasonally. In this study, nomadic and sedentary cattle in North-Central Nigeria were investigated for antibodies to FMD and risk factors for seropositivity evaluated. The overall seroprevalence of the disease was found to be 70.98% (95% CI: 68.37-73.49). This is consistent with the results of previous surveys conducted in Nigeria, in which a seroprevalence of 75.11% was reported by Olabode et al. (2013) in a study conducted in Kwara State, 64.7% in a

Table 7. Seroprevalence of FMD based on cattle herd mixing a watering point.

Mixing at the watering point	Number	%	Serological status		Prevalence (%) (95% CI)	OR
			+Ve	-Ve		
Yes	1001	94.7	759	242	75.8 (73.1-78.5)	171.8(23.8-1253)
No	56	5.3	1	55	1.8 (0.09-8.5)	
Total	1057			100		

χ^2=143.9; p-value <0.05.

Table 8. Seroprevalence of FMD hot spots (abattoir and cattle markets).

FMD hot spots	Number	Serological status		Prevalence % (95% CI)
		+Ve	-Ve	
Cattle Market	81	43	11	69.1 (58.5-78.5)
Abattoir	67	44	13	65.7 (51.4-76.3)
Total	148			

χ^2= 1.088; p-value > 0.05.

study conducted at the border states in Nigeria (Lazarus et al., 2012), 64.3 and 56.3% (Ehizibolo et al., 2010; Ishola et al., 2011), respectively, in studies carried out in Plateau State. The consistence of these findings confirmed that FMD is still an enzootic disease in Nigeria and this could be attributed to the fact that there has been no complementing vaccination campaign programme in the region, there is unrestricted herds mobility, continuous contact and intermingling of different herds at water points, communal grazing areas and porous borders. In addition, clinical diseases are usually underreported. This prevalence represents a higher prevalence than the 55% national prevalence reported by Abegunde et al. (1988).

Higher seroprevalence was recorded in Niger State (85.4%) than in Plateau State (54.2%). This could be attributed to the fact that many of the herds sampled indicated trans-boundary animal movement between Nigeria and the Republic of Benin. Niger State shares international boundary with the Republic of Benin, consequently, the animal population moves freely across the border in search of feed and drinking water. In most parts of West and Central Africa, the role of wildlife in the epidemiology of FMD has not been fully studied (Hedger and Condy, 1985; Thomson, 1995b; Alexandersen et al., 2002). However, the presence of wildlife population along the national park in Borgu might be a probable exposure factor that may have contributed to high FMD sero-positivity observed in this area. It has been established that countries like Nigeria with less developed livestock industries; the presence of many species of cloven-hoofed animals provides a possibility of reservoirs of the infectious viruses being established. It is believed that these free roaming species may normally come in contact with domesticated livestock, providing an opportunity for disease transmission. In comparison with the high seroprevalence observed in Niger North, Plateau North had the lowest seroprevalence which might be attributable to the fact that most of the cattle sampled in this area strictly practice intensive and sedentary management system contrary to the nomadism and extensive systems observed in most parts of Niger North.

Age category seropositivity revealed a higher sero-prevalence in cattle aged >2 years than in young cattle aged <2 years old. However, there was no association in seropositivity to age groups. The relative low seropositivity in young animals might be due to low exposure to risk factors. This is as a result of the practice of keeping young animals around the homestead and around areas separate from adult animals. Radostits et al. (2000), has indicated that young animals are relatively more susceptible than the adults, even though the present study showed that seroprevalence of FMD in adult cattle is slightly higher than that of the young cattle. This might be due to the fact that, adult cattle have repeated exposure and close contacts with other animals due to free animal movement. Generally, mortality is higher in young animals (over 20%) compared to 2% in adults. It has been observed that during outbreaks, morbidity rate in cattle can be up to 100% while mortality in young animals is up to 40% (Fiebre, 2015).

Furthermore, exposure factor to FMD seropositivity indicated both age groups had equal odds of FMD infection. Age association with FMD seropositivity was consistent with the previous study by Olabode et al. (2013) and Ishola et al. (2011) which reported higher prevalence of FMD in adult cattle than in young ones.

The higher seropositivity observed in female cattle was consistent with the findings of Olabode et al. (2013), who reported a risk difference in association with sex in Kwara State, Nigeria. Also, Mazengia et al. (2010) reported higher incidence of FMD in females in Northwest Ethiopia. However, more of the animals sampled were female as oppose to male cattle, therefore, the significant association in seropositivity in sex could be attributed to a small number of males sampled as both male and female animals are equally at risk.

Nomadic and sedentary management systems revealed a higher prevalence respectively, whereas, a lower seropositivity was recorded in the intensive management system. The higher seroprevalence recorded in nomadic and sedentary management systems might be as a result of unrestricted cattle movement, contact with the different herd and mixing at watering point, whereas the lowest prevalence recorded in the intensive management system could be attributed to restricted movement, less contacts with other herds and mixing at watering points. The study further revealed that the odd of FMD infection is 171.9% times more in nomadic and sedentary management than in intensive management system. This finding is in agreement with a study conducted in Southern Ethiopia by Megersa et al. (2009) where pastoral system was identified as one of the major risk factors for FMD transmission.

The seropositivity due to herd movement had indicated that the herds that reported movement across national borders recorded higher seropositivity relative to herds that reported movement within the country. This might be attributed to contacts with wildlife reservoirs which are continuous source of infection, as well as contact with different herds and different locations. All the herds that indicated national border crossing were in Niger State.

Cattle herd mixing at watering point had higher likelihood of being classified as FMD seropositive than those that do not mix at watering points, infection was observed to be 5.2 times higher in animals crossing national borders than those that do not. This study is in agreement with other studies which reported that the movement of herds in search of pasture and water from one area to another is a significant risk factor for the occurrence of FMD (Habiela et al., 2010; Molla et al., 2009; Megersa et al., 2009).Herds that reported mixing at the watering point with other herds recorded the highest seroprevalence relative to those that do not mix with other herds. Watering point was observed to be a common place where cattle of different herds meet in search of water, thereby serving as foci of FMD transmission. The odd of FMD infection at watering point was observed to be 17.8 times in herds mixing than those that do not mix at all. A similar observation was made in Thailand by Cleland et al. (1996) where the odds of FMD increased by 1.6 for every additional village that shared a water source (and village equates with the herd in our study).

This correlation might be due to either an increase in potential for transmission or from higher virus survival in a more humid microclimate around water sources (Donaldson and Ferris, 1975; Dawe et al., 1994).

The equal distribution of prevalence in abattoir and cattle market samples from study area is insignificant, which could be attributable to the fact that most of the cattle population being slaughtered in Nigeria abattoirs are directly purchased from the local cattle markets.

Conclusion

Identifying the risk factors of FMD is the first step towards progressive control pathway for FMD control. This study has established that FMD is enzootic in north central Nigeria, and it has also been able to identify some of the risk factors associated with FMD seropositivity in the study area. Further study to determine the possible role of wildlife and small ruminants in the epidemiology of FMD in the study area is strongly recommended. This will help in the implementation of the effective control programme.

Conflict of Interests

The authors have not declared any conflict of interests.

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