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Serological Prevalence of Foot and Mouth Disease in Parts of Keffi Local Government Area in Nassarawa State Nigeria

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ABSTRACT

About 80% of screened cattle were found to have been infected at least once with one of the seven serotypes of Foot-And- Mouth-Disease virus in areas of Keffi Local Government Area (LGA) in Nassarawa state. A total number of 108 bovine serum samples was collected from Maygaka, Angwa Ninzo and Kofar Hausa areas in Keffi LGA , with age, breed and sex of the animals recorded. Samples were collected base on previous history of foot And Mouth disease in the herd commonly called "Boro" by the herdsmen. Screening procedure was based on antibodies detection for the non structural protein mainly 3ABC protein in bovine serum regardless of the serotype of FMD virus involved using Chekit-FMD-3ABC ELISA (Bommeli Diagnostics, South Africa). Sample was said to be positive when its percentage inhibition (PI) calculated based on the kit manufacturer's recommendation was ≥ 30 %. Categorical variables (sex, age and location) were considered. Out of the total sampled animal, 77.08% was found to have been infected at least once with FMD virus with 33.33% of this infection in Angwa Ninzo, 8.33% in Kofar Hausa and 35.42% in Maygaka. There was no sex or breed predisposition to this disease. Prevalence was calculated by dividing the number of 3ABC ELISA positive animals by the total number of animals tested. Chi-square test was used for comparison of variables and tests were considered as significant at $P < 0.05$.Using double reciprocal simple linear model of analysis, it was observed that there is a significant relationship between and percentage inhibition and age of animal at 95% confidence interval.

Key Words: Seroprevalence**,** Foot-And-Mouth Disease, Nassarawa

INTRODUCTION

Over the years the FMD has caused tremendous losses in cattle in Sub-Saharan Africa[3]. The deep seated endemic nature of this disease in the sub-region could be attributed to the negligence of both the governments and herdsmen since case fatality in adult animal is usually not enormous or may be because of many other diseases such as Trypanosomosis, Rinderpest [16], Contagious Bovine Pleuropneumonia and others which achieve their economic importance through high mortalities. The wide host range, highly contagious nature of the disease has directly and indirectly hindered cattle production[14] which represent and important form of financial security and banking beside being an important source of draught power and food, being the major source of protein for human population in the areas concerned. Concurrently, the economic losses incurred from the disease through decreased milk production, poor performance in work animals and wasting have contributed immensely to the poverty level of the rural populations depending mainly on cattle production [3]. The endemicity and common occurrence of FMD among herds of cattle and less in other species [2] should be of serious concern with regards to control and eradication.

Foot and mouth disease (FMD) is a highly contagious viral disease affecting over 70 species of domestic and wild cloven-hoofed animals [11]. It affects cattle, sheep, goats, pigs and wild ruminants, causing significant production losses in adult animals and death in young stock. It is the single most important disease influencing global trade in live animals and animal products [6] and is on the List A of the Office International des Epizooties.

FMDV is the type species of the *Aphthovirus* genus of the *Picornaviridae* family. There are seven serotypes of FMD virus namely A, O, C, SAT 1, SAT 2, SAT 3, Asian 1 that have been identified serologically, and multiple subtypes occur within each serotype[1]. *Infection with one serotype does not confer immunity against another*.

Non structural protein which are identical in all the seven serotypes of FMDV have been characterized and are used for development of serological tests[13], that have been very useful in the epidemiological studies for differentiation between vaccinated and infected animals with FMDV. In many Sub- Saharan African countries where vaccination is not carried on, the test for Non-Structural FMD virus proteins is very useful for rapid diagnosis and confirmation of the presence of the disease [9] in the areas of study. After an animal is infected a number of FMDV non structural protein are produced during the replication cycle of the virus in the infected cell. Some of which are been shown to be highly immunogenic especially the 2C, 3A, 3D and the poly protein, 3ABC which are specific protein of the non structural protein complex of the FMDV. Most Serological test such as the liquid phase blocking ELISA measure antibodies produced against structural capsid protein of the FMDV which are produced in both infected and vaccinated animals and are serotype specific.

Vaccine are being formulated by inactivation of FMDV in tissue culture some level of purification of the inactivated virus from cellular and NSP

components residuals in the cell and addition of adjuvant to increase antigenicity[15]. Vaccination is greatly complicated by existence of seven immunologically distinct types of virus [8], many widely different subtypes and the continuous emergence of new types. Vaccination programmes require man power to vaccinate very large numbers of animals two or three times a year and for post vaccinal surveillance. Again there is disease implications for administrative (administration of multiple or cumulative doses to achieve prophylactic protection) and surveillance farm visits. Localization of highly dynamic (foci) spots of infection perpetuation will limit the risk of disease propagation and as such reduced need of repeated vaccination exercises [18] and of course the cost attached especially in countries where test and slaughter is unthinkable.

The onset of clinical disease is heralded by precipitate fall in milk yield and high fever, severe depression and anorexia, followed by appearance of an acute painful stomatitis. There is salivation smacking of the lips, formation of vesicles/bullae on the buccal mucosae and the dental pad. This rupture within 24hrs leaving raw painful surfaces which heals in about a week (Fig: 1). Concurrently with the oral lesion vesicles appear on the feet, particularly on the cleft and the coronets. Rupture cause acute discomfort and gross lameness, animal often remain recumbent (Fig: 2). Secondary bacterial invasion may interfere with healing and lead to involvement of deep structures of the feet. The vesicles may appear on the teat, severe mastitis may ensue. Abortion and infertility are common sequels of the disease. In outbreaks heavy mortalities are recorded in young calves as a result of severe myocardial damage while typical vesicular lesions in the mouth and feet may be absent.

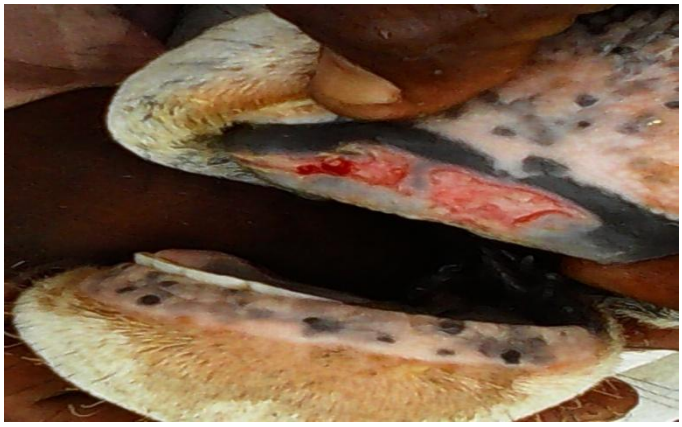


FIGURE1: Oral lesions involving the dental pad in Foot-And-Mouth Disease

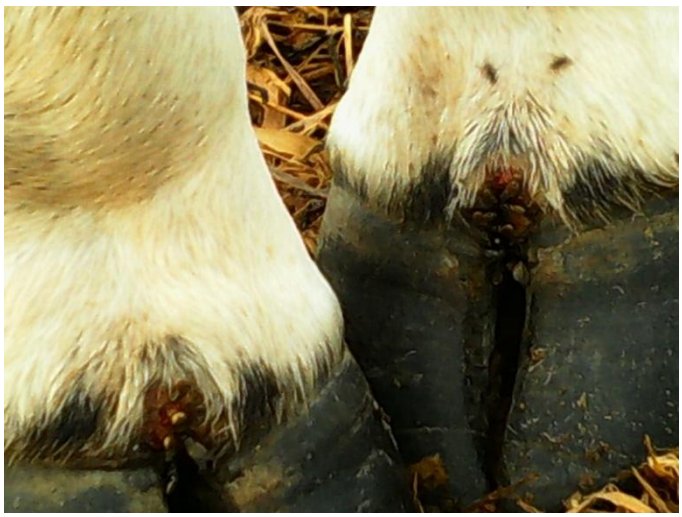


FIGURE 2: Interdigital lesions in the feet of cattle with foot and mouth disease

MATERIALS AND METHODS

Blood Samples were collected by jugular puncture without anti coagulant using vacuutainer needle and tubes. The blood was then spung within 12 hrs of collection at 1500 rpm in a centrifuge and serum sample harvest from each specimen. The serum was then preserved at -30^0 C till the time it was used.

The CHEKIT-FMD 3ABC bovine ELISA kit (Bommeli Diagnostics, South Africa) was used and this was indicated to be rapid, simple, sensitive and specific method for detecting antibodies against pathogen of FMDV [17] in serum samples of bovine origin. The test detects antibodies against 3ABC protein independent of the serotype of FMD virus [9] . In the kit, the entire necessary reagents for the standard indirect ELISA technique were included with polystyrene microtiter plate pre-coated recombinant FMD 3ABC protein. Dilutions of samples to be tested were incubated in the wells. Any antibody specific for 3ABC protein binds to the antigen in the wells. A peroxidase labeled anti-IgG-conjugate was added which binds to antibody of sample complexes with antigen. The TMB-containing substrate was added to the wells. In this assay, adequate washing procedure were undertaken in order to remove unbound reagent at each step of the testing procedure. The degree of color development measured by spectrophotometer is directly proportional to the amount of antibody in the sample serum specific to the antigen. The result was read by microplate spectrophotometer, where the optical density (OD) was measured at 405 nm within 15 min after addition of stop solution. The OD in wells coated with non structural proteins (NSP) 3ABC were corrected by subtraction of the corresponding wells containing the control antigens. This value called Percentage Inhibition (PI) was obtained base on the following formula (given by manufacturer) PI= [OD_{sample}- $OD_{negative}$]/[$OD_{positive}$- $OD_{negative}$] x100.

OD_{sample}= Optical Density of the test serum

$OD_{negative}$= Optical Density of the negative control

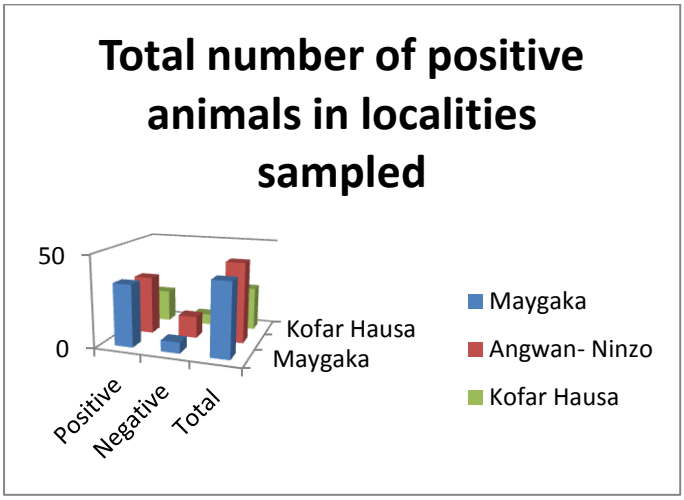
$OD_{positive}$ = Optical Density of the positive control

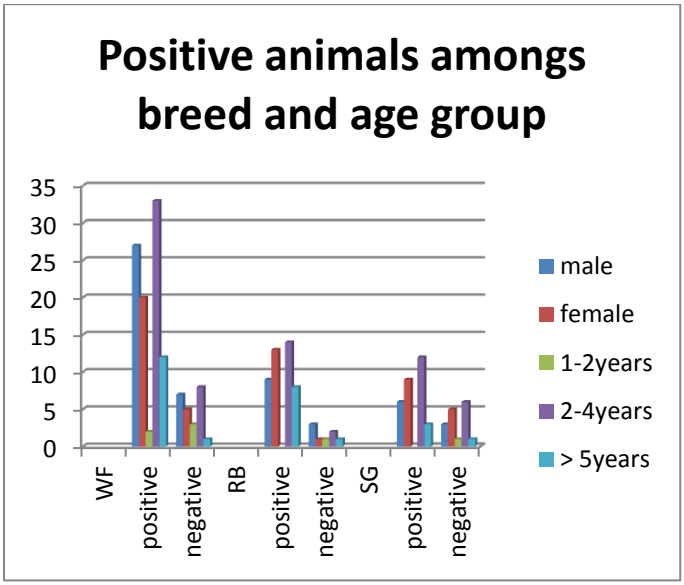
A test sample was then said to be positive of FMD when its PI ≥ 30 %. Positive Sera were then grouped into three different categories using graded anti body response into low (30-50%), medium (51-70%) and highly (71-90%) positive sera and the significance of multiple infections on the response was estimated using multivariate logistic regression.

Results and Interpretation

Origin	Frequency	Percent (%)
Angwan Ninzo	44	40.7
Kofar Hausa	24	22.2
Maygaka	40	37.0
Total	108	100.0
Breed		
Mixed	1	.9
RB	3	2.8
SG	10	9.3
WF	94	87.0
Total	108	100.0
Sex		
F	56	51.9
M	52	48.1
Total	108	100.0

This table shows the analysis of the data collected with respect to origin, breed and sex. it reveals that 40.7% of the data were collected in Angwan Ninzo, follow by Maygaka (37.0%) and the remaining (22.2%) were collected in Kofar Hausa. it also shows that the majority (87.0%) of the breed selected were WF follow by very small portion of SG, RB and Mixed breed with 9.3%, 2.8% and 0.9% respectively. the analysis with regard to sex shows that 51.9% are female while 48.1% are male.





Hypothesis: There is no predisposition of FMD with breed and sex.

Table 2: Chi-Square Tests analysis on predisposition of FMD with Breed and Sex

Sex		Value	df	P-value
F	Pearson Chi-Square	1.253	2	.535
	Likelihood Ratio	1.903	2	.386
M	Pearson Chi-Square	1.484	2	.476
	Likelihood Ratio	2.516	2	.284

Table 2 shows chi-square statistics for examining the hypothesis: there is no predisposition of FMD with breed and sex. Since X^2 (0.535) for sex female and the different types of breed and X^2 (0.476) for sex male and the different type of breed are all greater than 0.05 we do not reject the Null hypothesis at 5% significance level and therefore conclude that there is not predisposition of FMD with breed and sex

Simple Regression - PI vs. Age

Dependent variable: PI (Percentage Inhibition)

Independent variable: Age

Double reciprocal model: Y = 1/(a + b/X)

The output shows the results of fitting a double reciprocal model to describe the relationship between PI and Age. The equation of the fitted model is

A.PI = 1/(0.00871559 + 0.0698559/A.Age)

Since the P-value in the ANOVA table is less than 0.05, there is a statistically significant relationship between PI and Age at the 95.0% confidence level.

DISCUSSION

Construction of a two-way 2 by 3 contingency table was made to show the frequency of occurrence of Foot and Mouth Disease unique pairs of values for Results base on origin, breed, sex and age of the animals. From Chi-square results there was neither sex, breed, or age predisposition to FMD in this area. This study shows that FMD revolves in this area given that more than 70% of screened animals had experienced a clinical infection. The endemic nature of FMD in this region is well known but the complexity of its control remains an important challenge if this has to be successful. The multivariate logistic regression in this study was used to evaluate the effect of multiple infection on

percentage inhibition on assumption that and older animal in endemic area will have had more chances of exposure to the field virus than a younger one. The test for antibodies against non structural protein is not a direct quantitative test but the color change is based on the proportion of antigen/antibody complexes formed in test serum. Percentage Inhibition (PI) is a proportion of the optical density of test serum measured. There was a strong positive correlation with the magnitude of antibody response and the age of animals. FMD has multiple serotypes that circulate indiscriminately, and various types and subtypes that does not confer protective immunity. In this study it is considered that antibodies against NSP will be higher after several exposures since these proteins are identical in all the seven serotype.

This area can be identified as a region of priority in vaccination procedure whenever is started. This will reduce persistent distribution of the disease to other localities. It will equally help reduction of disease occurrence.

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