



Seroprevalence of African Swine Fever in Apparently Healthy Pig Farming in Chad

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Abstract

Introduction: First described in Kenya, ASF is an infectious disease caused by a virus belonging to the *Asfarviridae* family and *Asfivirus* genus. Reported in eastern and southern Africa where the virus is known to be present in wildlife as well as incursions into central Africa. Chad first experienced ASF in October 2010. In less than four months, it was reported in four provinces. Failures in biosecurity, poor disease management, uncontrolled movement of animals, marketing and processing would have been the first factors for the introduction of the ASF virus in these different provinces of Chad.

Purpose: The objective of this work is to determine the seroprevalence of ASF virus in apparently healthy pig farms in Chad.

Material and method: The study was conducted from September 2021 to September 2022 in the provinces of Chari Baguirmi, Mayo Kebbi East and the city of Ndjamen. Started with is census of 492 farmers who had already registered ASF. Among them 198 consented to participate in this study. A total of 345 sera were collected and analyzed by the competition ELISA technical for the detection of antibodies directed against the ASF virus. The data from the interviews as well as the serological results were analyzed with the R Studio software.

Results: The seroprevalences were 12.10% and 33.33% respectively in Mayo Kebbi East and Chari Baguirmi. In the cities, the seroprevalence varied from 10.10% to 33.33%. In the cantons and boroughs, it reached 61.53%. The seroprevalence was above 30% in eight districts, notably in Darda, Digo, Dogoré, Kabalaye, Malam-Sadi I, Malam-Sadi II, Tchinvogo and Toukra-Massa, respectively 44%, 61.53%, 50%, 33.33%, 45.45%, 50% and 100%. The variations in seroprevalence were related to the rainy season, the types of pig farms (16.78%) and the floor of the pigsty and the absence of litter in the pigsties. Seroprevalence was 19.23% in males; the age group between 0 and 4 months was the most affected with a rate of 78.52%.

Conclusion: This study revealed that the ASF virus circulates in pig farms in Chad, despite the absence of visible clinical signs. The presence of this virus in farms would be maintained by the inadequacy of biosecurity measures and poor farming practices. The study provides proof that ASF actually circulates in pig farms in Chad. Absence of vaccine against disease is at the origin of the increase variations in seroprevalence in the different study sites, hence the need for an in-depth study of the factors influencing the circulation of the virus of the disease ASF.

Keywords: Seroprevalence; Virus; African Swine Fever; Competitive Elisa; Pig Farming; Chad

Abbreviations

ASF: African Swine Fever; ELISA: Enzyme Linked ImmunoSorbent Assay; DNA: Dideoxyribonucleic Acid; IRED: Livestock Research Institute for Development.

Introduction

African Swine Fever (ASF) is caused by a DNA virus belonging to the *Asfarviridae* family in which the ASF virus is the sole representative and included in the genus *Asfivirus* [1]. ASF viral infection was first diagnosed in Kenya in 1910 by Montgomery; it is manifested by an acute hemorrhagic fever with a mortality rate of 100% in domestic pigs [2]. Reports were reported from South Africa in 1928, Malawi in 1931, and Angola in 1932 [3]. Following the first descriptions, ASF was reported in most countries in eastern and southern Africa where the virus is known to have been present in wildlife for a very long time. In Central Africa, incursions into Sao Tome and Principe in 1979 and Cameroon in 1982 have been officially reported [4,5]. In October 2010, Chad experienced ASF for the first time. Despite emergency health measures taken by Chad's veterinary services, the disease was reported in less than four months in four provinces, namely Mayo-Kebbi East and West, Tandjilé, Logone Occidental and Chari Baguirmi [6]. In 2012, a first outbreak of ASF reappeared in Mandoul and Batha with cases of infection in warthogs [7]. In 2018, ASF was once again reported in Ndjamen and affecting the first, seventh and ninth arrondissements with a mortality rate of 89.72%. All these periods of epidemics were followed by sanitary culling of a large number of animals. Such an event could contribute to the extinction of local breeds already well adapted to environmental conditions [6,8,9]. Deficiencies in biosecurity and disease management as well as the pork value chain (type of breeding, state of the pigsty, movement of animals, marketing and processing) would be the first factors of introduction of the ASF virus in the different provinces of Chad and its persistence in the country. Failures were also observed in the practice of "stamping out". According to [6,9] and many other authors, the risk of ASF endemicity is real after the passage of the virus. The purpose of this work is to determine the seroprevalence of ASF in apparently healthy pig farms in Chad and to propose an appropriate approach for its eradication in Chad.

Material and Methods

The study was carried out in four areas, namely

- The sub-prefecture of Rigaza, located 50 km from the city of Bongor. The geographical coordinates of this area produced by Global Position System (GPS) are 10°91'604" of the North attitude and 15°19'604" of East-West longitude.

- The city of Bongor and its surroundings, capital of the province of Mayo Kebbi East, located 240 km from Ndjamen, capital of Chad. The geographical coordinates realized by GPS: are 10°16'29" North latitude and 15°22'39" East longitude.
- The city of Ndjamen whose geographical coordinates realized by GPS are: 12°6'47" North latitude and 15°2'57" East longitude.
- The towns of Mandalia and Mailao, respectively prefecture and sub-prefecture of Chari Baguirmi province. The geographical coordinates of this area produced by GPS are: 11°43'37" North latitude and 15°14'52" East longitude for the prefecture of Mandalia; 8°31'0" North latitude and 15°46'0" East longitude for the sub-prefecture of Mailao. These four areas correspond to pig farming areas, the most important in Chad, and are often hit by strong waves of ASF.

The sampling was done on a voluntary basis. It started with a census of 492 farmers who had already registered ASF. Among them 198 consented to participate in this study, 106 breeders refused to participate in the study and 188 breeders no longer have pigs in their piggeries when the team returns to take the samples. This is to increase the chances of detecting the disease. Then, targeted sampling was carried out to choose the areas explored. The samples were taken from pigs aged 0 to 48 months depending on the size of the herd.

The collected blood is left at least 30 to 45 minutes at room temperature for clot formation. The serums collected are transferred to dry tubes and then placed in a cooler containing an "ice-pack", then sent to the virology laboratory at IRED to be stored at -20°C for later use. Serum antibodies were detected using competitive enzyme-linked immunosorbent ELISA (c-ELISA) technical. Antibodies against ASF virus were detected by the c-ELISA test. The Kit used is marketed by Innovative Diagnostics veterinary (IDvet). The method used is that recommended by the manufacturer. The data from the interviews as well as the result of the sera tested were entered into a Microsoft Office 2007 Excel spreadsheet then converted to CSV then exported to R Studio software version 4.0.4.2021 for analysis. The Q.GisR 3.18 and Arc.Gis 10.5 software enabled the creation of the map of the study areas. Regarding the analytical statistics, the Chi-square test and the Fisher's Exact test were used to compare the proportions (department, towns and sub-prefectures) and for their significance. The significance threshold was set at 0.05 and the p-value calculated using Fisher's Exact Test.

Results

At the end of the laboratory work, the results below were obtained:

Study area

Provinces	N	ASF +	SR in %	CI at 95%	P-value	Interpretation
Chari Baguirmi	12	4	33,3	[2,85 ; 63,81]	4,18 e-10	Very significant
Mayo Kebbi-East	256	31	12,10	[11,81 ; 12,39]		

Table 1: Seroprevalence by province.

Legend: 4.18e-10: 0.00000000418; N: Number of Pig, ASF+: Serum samples tested positive for African Swine Fever, SR: Seroprevalence, CI: Confidence Interval, %: Percentage

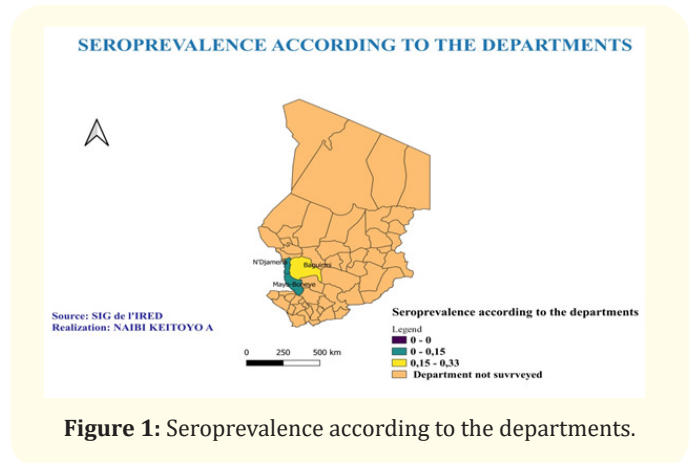


Figure 1: Seroprevalence according to the departments.

Terms		N	PPA +	SR in %	CI at 95%	P-value	Interpretation
Cities	Bongor	178	19	15,38	[13,97 ; 16,79]	4,18e-8	Very significant
	Ndjamena	78	12	12,03	[11,47 ; 12,59]		
Prefecture and Sub-prefecture	Bongor	188	19	10,10	[9,80 ; 10,40]	6,639e-10	Very significant
	Mandalia	12	4	33,33	[2,85 ; 63,81]		
	Rigaza	70	13	18,57	[16,45 ; 20,68]		

Table 2: Seroprevalence by town, prefecture and sub-prefecture.

Legend: 4,18e-8 = 0,000000418 et 6,639e-10 = 0,000000006639

Cantons/Dis.	N	ASF +	SR in %	CI at 95%	p-value	Interpretation
7 ^{ième} Dis.	13	8	61,53	[44,61 ; 78,45]		
9 ^{ième} Dis.	63	3	4,76	[4,51 ; 5,01]		
Bongor	133	16	12,03	[11,47 ; 12,59]	4.568e-16	Very significant
Darda	9	4	44,44	[-18,62 ; 107,5]		
Koumi	70	13	18,57	[16,46 ; 20,68]		
Mailao	2	0	0	0		
Telmè	55	3	5,45	[5,08 ; 5,82]		

Table 3: Seroprevalence by canton and district

Legend: Dis: District and 4.568e-16 = 0.000000000000004568

Villages/Neighborhoods	N	ASF +	SR in %	CI at 95%	P-value
Bariam-Dogom (V)	51	2	3,92	[3,70; 4,14]	7,484e-14
Biliam-Oursi (V)	10	2	20	[3,35; 36,65]	
Darda (V)	9	4	44,44	[-18,62 ; 107,44]	
Digo (N)	13	8	61,53	[-10,05 ; 132,53]	
Djoh-Bongor (N)	40	4	10	[8,61 ; 11,39]	
Dogoré (V)	2	1	50	[289,55 ; 389,55]	
Dongui-Baha (V)	8	2	25	[-4,40 ; 54,40]	
Ferme (N)	7	2	28,57	[-12,69 ; 69,83]	
Fort-Damba (N)	17	1	5,88	[4,53 ; 7,24]	
Goulmoune Révé (V)	10	3	30	[-1,13 ; 61,13]	
Gouneida (V)	2	0	0	0	
Kabalaye (N)	12	4	33,33	[2,85; 63,81]	
Lama-lama (N)	34	1	2,94	[2,75; 3,18]	
Mailao (V)	2	0	0	0	
Malam-Sadi I (V)	11	5	45,45	[7,94; 98,94]	
Malam-Sadi II (V)	2	1	50	[289,55 ; 389,55]	
Malam-Sadi III (V)	9	1	11,11	[3,78 ; 18,44]	
Malam-Sadi IV (V)	5	0	0	0	
Mandalia (V)	1	0	0	0	
Ngonbah (N)	53	0	0	0	
Siéké I (N)	14	3	21,42	[8,19; 34,65]	
Siéké II (N)	16	3	18,75	[9,34; 28,16]	
Tchinvigo (V)	2	1	50	[289,55; 389,55]	
Tiyor-Révé (V)	8	0	0	0	
Toukra-Massa (N)	8	8	100	[142,55; 342,55]	

Table 4: Seroprevalence in villages and neighborhoods.

Legend: v = village, N = neighborhoods and P-value 7,484e-14 = 0,000000000000007484

Discussion

In the two study provinces, the seroprevalence was highly significant and varied: 33.33% in the province of Chari Baguirmi and 12.10% in Mayo kebbi East. These results are similar to those obtained in the Kabare district in the Democratic Republic of Congo by [10] 34.3%, are much lower than those published in Uganda by [11] 52.96%. This increase in seroprevalence between the two provinces is explained by the fact that the province of Chari Baguirmi is cosmopolitan with a high population density in N'Djamena, and a very high need for pork consumption in this city. The pigs sold in the capital come from other producing provinces in southern Chad and the Cameroonian borders, notably Kousserie. These animals with unknown health status are transported to the capital without any control and with derisory means of transport. Animal

pressure, higher in the suburbs and neighboring towns close to the capital, is proof of the importance that this breeding offers to a large part of the Chadian population.

In traditional and modern pig farms in Cameroon, seroprevalence was 15.2% and 23.8%; those of Senegal and Burkina Faso were respectively 16.9% and 19.6% [12-14]. The results obtained in Bongor (15.38%) are close to those obtained in Cameroon, but lower than those of Senegal and Burkina Faso. On the other hand, those of the city of Mandalia are higher than those obtained in these countries. Our results show an increase in seroprevalence as we get closer to the city of N'Djamena. It was 10.10%; 18.57% and 33.33%, respectively in Bongor 233 km, Rigaza 153 km, Mandalia 60 km and Toukra-Massa in the city of Ndjamen 100%. This

increase in seroprevalence is explained by the importance of demand and supply, exchanges with a derisory commercial circuit, ignorance of biosecurity measures and pig farming technical.

The seroprevalences in the cantons and districts are 5.45%, 12.03%, 18.57% and 44.44% respectively in the Telmè, Bongor, Koumi and Darda cantons. In the district seroprevalence is 4.76% in the 9th district and 61.53% in the 7th district. The high seroprevalence rates in the district and Cantons can be explained by two reasons.

Conclusion

The aim of this study was to determine the seroprevalence of ASF in apparently healthy pig farms in Chad. Seroprevalence varied overall from 10.10% to 33.33% from one province to another, from one department to another, from one city to another. For villages or neighborhoods, it reached 61.53% in Digo and 100% in Toukra-Massa. Highlighting the seroprevalence of ASF in the provinces, departments, towns and villages demonstrates the presence and circulation of this virus on farms. This presence is maintained by the lack of knowledge of biosecurity. The lack of mastery of pig farming practices and the absence of control have greatly contributed to the circulation of the virus. Knowing the seroprevalence is a control tool for ASF.

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Author's Contributions

NKA contributed to the design of the protocol, to the writing of the first draft of the manuscript, BBA, AAI, NBNR and RLG in the development of the study protocol and its critical review. AD, BBA, RLG, SL, TA and NBNR supervised data collection and sample sorting. NBNR supervised the laboratory analyzes. NKA and RLG performed the statistical analyzes of the data. All the authors proceeded to the proofreading, correction and validation of the manuscript.

Conflict of Interest

All the authors declared that there are no known conflicts of interest associated with this publication.

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