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IMPACT AND PREVALENCE OF NEWCASTLE DISEASE AND ASSOCIATED RISK FACTORS IN VILLAGE CHICKENS IN SOUTHERN NIGER

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*Supporting Information

ABSTRACT: The present study was conducted to determine the prevalence of Newcastle disease and to identify potential risk factors in village chickens in Niger. A total of 1,627 serum samples were collected using a stratified random sampling method with proportional allocation. Samples were collected from village breedings in the departments of the Maradi region (Guidan Roumdji, Madarounfa, Aguié, Gazaoua, and Tessaoua), departments of Zinder region (Takeita, Kantché, Magaria, Dungass, and Mirriah), and cities of Maradi and Zinder. Data on risk factors were collected through an interview with the herders. All collected sera were subjected to competitive enzyme-linked immunosorbent assay (cELISA) to detect Newcastle disease virus-induced antibodies. The findings were indicative of 302 positive sera, representing an overall seroprevalence of 18.6%. The Student's t-test at p<0.05 revealed a significant difference between regions and among some departments. Furthermore, the logistic regression test identified the agroecological zone, type of breeding, species mix, and the origin of the animals as risk factors associated with seropositivity to Newcastle disease virus. The present results confirmed the exposure of village chickens to the Newcastle disease virus, emphasizing the need to intensify vaccination campaigns and educate poultry farmers on adopting biosecurity measures.



Keywords: Newcastle disease, Risk factors, Vaccination, Village chickens, Niger.

INTRODUCTION

Niger is one of the countries with an agricultural vocation, so its economy is mainly based on the rural sector, including agriculture, breeding, and fishing. In 2020, the rural sector employed 80% of the active population and contributed 37.7% of the Gross Domestic Product (HCI3N, 2021). Poultry production is one of the most important economic activities, 98% of which is carried out by rural populations (HCI3N, 2021). Based on recorded poultry sales statistics, national poultry meat production was estimated at 3,592 tons of carcass equivalent in 2017 (MAG/EL, 2020) .These data do not take into account the total meat production of traditional and modern poultry farming due to the lack of a reliable information system on the sector. The production of eggs for consumption was estimated at 596,717,980 eggs in 2019. In addition, the poultry capital was estimated at more than 20 billion FCFA in 2019 (MAG/EL, 2020). As in other developing countries, rural Niger residents depend significantly on poultry for their household income and dietary protein intake (Amoia et al., 2021). However, poultry practices involve few biosecurity measures and a high risk of infectious diseases (Getabalew et al., 2019; Chowdhury et al., 2020), including Newcastle disease (Otte et al., 2021). In many low-income, food-deficit countries, Newcastle disease causes significant economic losses in poultry farming. These losses are related to high poultry mortality, vaccination costs, drop in production, cost of treating animals, as well as expenses related to the implementation of biosecurity measures (Ipara et al., 2021). In traditional poultry farming, Newcastle disease kills an average of 55% of poultry per year (Antipas et al., 2012). It affects the potential source of nutrition for rural households, their source of income, and discourages them from investing in poultry breeding (de Bruyn et al., 2017; Waweru et al., 2023).

Newcastle disease is a highly contagious infectious disease of poultry caused by virulent strains of avian paramyxovirus type 1 (OIE, 2022). It is clinically characterized by respiratory impairment, nervous system impairment, gastrointestinal disorders, and reproductive impairment (Caroline, 2022). Many species of birds, both domestic and wild, are susceptible to Newcastle disease, but chickens are particularly susceptible. The pathogenicity of Newcastle disease in chickens is primarily determined by the virus strain, viral dose, and age of the chicken, environmental conditions, administration route, and transmission mode (Getabalew et al., 2019).

Newcastle disease is endemic in Niger as it is in many developing countries (Dimitrov et al., 2016; Absalón et al., 2019; Toroghi et al., 2020; Amoia et al., 2021) and negatively impacts farm household economies, dietary diversity, and consumption of animal-based foods (Knueppel et al., 2010; McElwain and Thumbi, 2017). Due to its severe nature and

disastrous consequences, Newcastle disease is included in the list of notifiable diseases in Niger (MEL, 2011) and the World Organization for Animal Health (OIE, 2022). As such, its control represents a major challenge for national veterinary services.

The objective of the present study is to contribute to a better knowledge of the epidemiology of Newcastle disease in Niger, more specifically, to determine the frequency of Newcastle disease, its distribution, and the risk factors associated with its occurrence. The findings can provide those in charge of the national animal health policy with a guiding tool in surveillance and control.

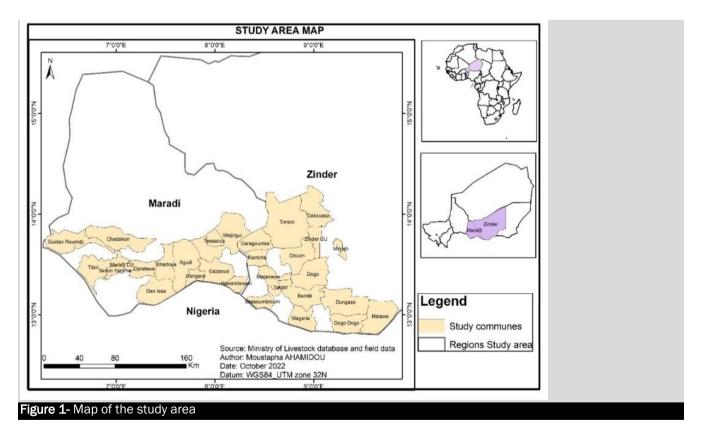
MATERIALS AND METHODS

Ethical regulation

Due to the absence of an Animal Care Committee available at the University of Lomé at the time of this research, the research was conducted under the supervision of the research team leader following the guidelines of the Canadian Council on Animal Care (2009).

Study area

This study was conducted in the southern part of the Maradi and Zinder regions bordering the Federal Republic of Nigeria. These include the departments of Guidan Roumdji, Madarounfa, Aguié, Gazaoua, and Tessaoua in the Maradi region and the departments of Takeita, Kantché, Magaria, Dungass and Mirriah in the Zinder region, as well as the cities of Maradi and Zinder. The study area is covered by the Sahelian agroecological zone and the Sahelo-Sudanese zone. The Sahelian zone receives an average annual rainfall of 350-600 mm, and the Sahelo-Sudanese zone accumulates more than 600 mm of rain per year (Wata et al., 2012). The two study regions have an area of 197,574 km² and an estimated population of 9,584,421 in 2021, or 40.6% of the total population (INS, 2019). The investigated sites have a large poultry flock estimated at 6,205,390 birds in 2017, or 31.82% of the national flock (MAG/EL, 2020). Figure 1 illustrates the study area.



Sampling method

Samples were selected using a stratified random sampling method with proportional allocation to the size of agricultural households. First, the agricultural departments were selected from each of the two regions along the borders with Nigeria and the cities of Maradi and Zinder. In the second step, a maximum of three communes or boroughs were selected per department or city. In the next step, a maximum of three villages were selected within each commune or city. Using the national directory of localities, the number of farm households surveyed per commune was determined in proportion to the total number of farm households. Then, the farm households to be surveyed were drawn from the list of households drawn up by the village chief. Finally, a maximum of three unvaccinated chickens were collected from each surveyed household after the owners' permission.

Data and sample collection

Data were collected from November 2021 to January 2022 using a survey questionnaire with semi-open and closedended questions. The questionnaire was administered face-to-face using the KoBoCollect v2021.2.4 collection tool. Information collected included the geographic location of sites and data on poultry practices.

For sample collection, 1.5 to 2 ml of blood was collected from the chickens' wing veins in tubes without any anticoagulant. Serum samples were separated from clotted blood samples by centrifugation at 1500 rpm for 5 minutes. Serum samples were then decanted, aliquoted into microtubes, and frozen at -20°C at the Central Livestock Laboratory of Niamey, Niger, until testing.

Methods of analysis and interpretation of results

The competitive enzyme-linked immunosorbent assay (ELISA) was used to detect antibodies to the Newcastle disease virus in the collected sera. The diagnostic kits (Reference NDVC-12P LOT K34) were obtained from the Innovative Diagnostics IDVet Laboratory in Marseille, France. The procedure used in the laboratory was conducted according to the manufacturer's instructions. The reading of the plates was done with the Skanlt Software v.3.1 at an optical density (OD) of 450 nm. The assay is validated if: the mean OD value of the negative control (ODNC) is greater than 0.6 (ODNC > 0.6) and the percentage inhibition (PI%) of the positive control (mean ODPC value) is greater than 40% (PIPC > 40%). For each sample tested, the percentage inhibition value was calculated (PI%) with the following formula: PI% = (ODNC - ODsample / ODNC)*100. Samples with a PI% greater than 40% were recorded as positive, those with a PI of 30-40% were considered doubtful, and samples with a PI less than 30% were categorized as negative.

Statistical analysis

The data obtained were analyzed using STATA 13.0 software. Student's t-test was used to compare the prevalences of Newcastle disease between departments and regions. A logistic regression model was adopted to determine the risk factors associated with Newcastle disease. The variable seropositivity to Newcastle disease was explained through the explanatory variables, including the agroecological zone, chicken sex, breeding type, species mix, and origin of the animals. Individuals (chickens) were then classified into two groups with regard to their seropositivity (those with and those without seropositivity to Newcastle disease). This variable was dichotomous or binary, and the candidate explanatory variables were all categorical.

RESULTS

Prevalence of Newcastle disease

Tables 1 and 2 tabulate the distribution of Newcastle disease prevalence in village chickens by commune and department. Of 1627 sera tested, 302 were positive for Newcastle disease virus, with an overall prevalence of 18.6%. All study communes had a prevalence of 3.3-46.7%, except for the commune of Kantché in the Zinder region, where the prevalence was 0.00%. The highest prevalence was obtained in the city of Maradi (46.7%) and the lowest in the commune of Yaouri (3.3%). Figure 2 shows the distribution map of Newcastle disease prevalence by commune and study department.

The proportion of Newcastle disease prevalence of each department or city was compared to that of all other departments or cities using the Student's t-test at a significance level of 0.05. The results of the comparison tests between the prevalence proportions of the departments or cities took two by two showed a significant difference between the departments of Takeita and Guidan Roumdji, Takeita and Madarounfa, Takeita and Aguié, Kantché and Guidan Roumdji, Kantché and Madarounfa, Kantché and Aguié, Dungass and Guidan Roumdji, Dungass and Madarounfa, Dungass and Aguié, Mirriah and Guidan Roumdji, Mirriah and Madarounfa, Mirriah and Aguié, Maradi city and Takeita department, Maradi city and Kantché department, Maradi city and Dungass department, Maradi city and Mirriah department, Zinder city and Guidan Roumdji department, Zinder city and Madarounfa department, Zinder city and Aguié department, and Maradi city and Zinder city (p < 0.05). However, no significant difference was found between the Gazaoua and Madarounfa departments, the Gazaoua department and Maradi city, the Magaria department and Zinder city regarding prevalence proportions (p > 0.05).

The Student's t-test revealed a significant difference between the Maradi and Zinder regions in terms of Newcastle disease prevalence (p < 0.05). The highest prevalence was recorded in the Maradi region (Table 3).

According to the results of the model, seropositivity was significantly explained by the agroecological zone, the type of breeding practised, the composition of the flock, and the origin of the poultry (Figure 3).

The risk of Newcastle disease was not the same in the two agroecological zones. It was 1.45 times higher in the Sahelo-Sudanese zone than in the Sahelian zone. Newcastle disease was more prevalent among herders who practiced divagation than those who practiced semi-divagation. The risk of a chicken being seropositive in a mid-scavenging system was 0.42 times lower than that of a scavenging system. Of note, the effect of confinement could not make a significant difference between the two systems. Poultry flocks of different species indicated different exposure rates to Newcastle disease. There was a higher risk of Newcastle disease in a breeding system composed of several avian species, compared to one containing only one particular species. The risk is 1.5 times higher for poultry in multi-species breeding than for a breeding group composed of only one avian species. Finally, when live poultry is bought on the local market, the risk of

Newcastle disease was 2.19 times higher than when he used other sources of supply, such as the neighboring village, traditional and modern hatcheries, and imports (Tables 4 and 5).

Regions	Departments/Cities	Communes/Cities	Samples tested	Positive samples	Prevalence (%)
	Guidan Roumdji	Chadakori	66	17	25.8
		Tibiri	79	23	29.1
		Guidan Roumdji	55	24	43.6
	Madarounfa	Serkin Yamma	23	7	30.4
		Djiratawa	48	17	35.4
		Dan Issa	37	6	16.2
	Aguié	Tchadoua	50	13	26
Maradi		Aguié	60	23	38.3
	Gazaoua	Gazaoua	50	11	22
	Gazaoua	Gangara	$\begin{array}{c cccccc} 48 & 17 \\ 37 & 6 \\ 50 & 13 \\ 60 & 23 \\ \hline 50 & 11 \\ 32 & 9 \\ \hline 75 & 17 \\ 42 & 7 \\ 35 & 8 \\ \hline 15 & 7 \\ \hline 53 & 10 \\ 80 & 7 \\ \hline 36 & 2 \\ \hline 32 & 0 \\ 34 & 7 \\ \hline 30 & 1 \\ \hline 54 & 5 \\ 87 & 20 \\ \end{array}$	28.1	
	Tessaoua	Tessaoua	75	17	22.7
		Maijirgui	42	7	16.7
		Hawandawaki	35	8	22.9
	Maradi city	Maradi city	15	7	46.7
	Dakoussa	Dakoussa	53	10	18.9
		Tirmini		7	8.8
		Garagoumssa	36		5.6
	Kantché	Kantché	32	0	0
		Matamey	34	7	20.6
		Yaouri			3.3
		Sassoumbroum	54		9.3
	Magaria	Magaria	87	20	24.1
Zinder		Bandé	32	3	9.4
		Malawa	87	9	10.3
	Dungass	Dungass	83	12	14.5
		Dogo dogo	49	5	10.2
		Dogo	96	11	11.5
	Mirriah	Droum	97	9	9.3
		Mirriah	60	7	11.7
	Zinder city	Zinder city	50	5	12
Total			1627	302	18.6

Table 1 - Distribution of Newcastl	e disease prevalence in vill	lage chickens by commune or city
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Table 2- Distribution of Newcastle disease prevalence in village chickens by department or city

Departments/Cities	Samples tested	Positive samples	Prevalence (%)	P-Value
Guidan Roumdji	200	64	32.0a	
Madarounfa	108	30	27.8a	
Aguié	110	36	32.7a	
Gazaoua	82	20	24.4a	
Tessaoua	152	32	21.1 b	
Maradi city	15	7	46.7a	< 0.05
Takeita	169	19	11.2 b	
Kantché	96	8	8.3b	
Magaria	173	28	16.2b	
Dungass	219	26	11.9 b	
Mirriah	253	27	10.7b	
Zinder city	50	5	10.0b	
Total	1627	302	18.6	

Table 3- Prevalence of Newcastle disease by region				
Regions	Samples tested	Positive samples	Prevalence (%)	P-Value
Maradi	667	189	28.3a	< 0.001
Zinder	960	113	11.8 b	< 0.001
Total	1627	302	18.6	
The proportions of Newcastle disease prevalence of the departments, cities and regions with a different letter (a, b) are significantly different $a_1 p < 0.05$.				

212

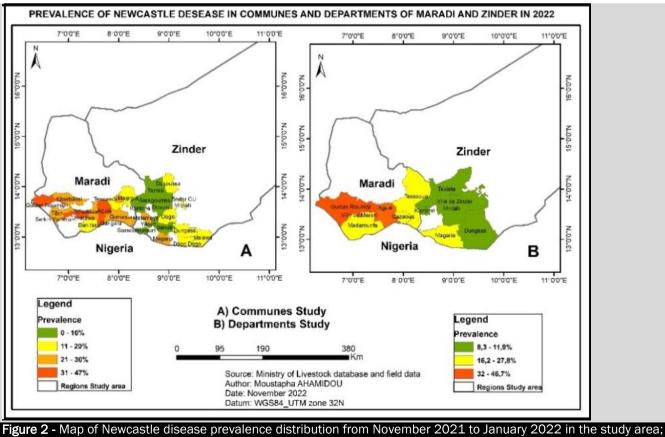
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Table 4- Distribution of prevalence according to potential risk factors

Variables	Modalities	Samples tested	Positive samples	Prevalence (%)
Agro opplogical zono	Sahelian	638	83	13.0
Agro-ecological zone	Sahelo-sudanian	989	219	22.1
Gender of chicken	Male	362	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	18.5
Gender of chicken	Female	1265	235	18.6
	Scavenging system	1277	262	20.5
Breeding methods	Mid-scavenging system	338	38	11.2
	Claustration 11		2	18.2
Composition of the herd	Monoespecies	m 1277 262 20.5 ystem 338 38 11.2 11 2 18.2 982 157 16.0 645 145 22.5 1033 218 21.1	16.0	
composition of the neru	Polyespecies	645	145	22.5
	Local market	1033	218	21.1
	Neighboring village	501	70	14.0
Origin of poultry	Traditional hatchery	66	10	15.2
	Modern hatchery	17	3	17.6
	Importation	10	1	10.0

Table 5- Risk factors associated with Newcastle disease in village chickens in the study area

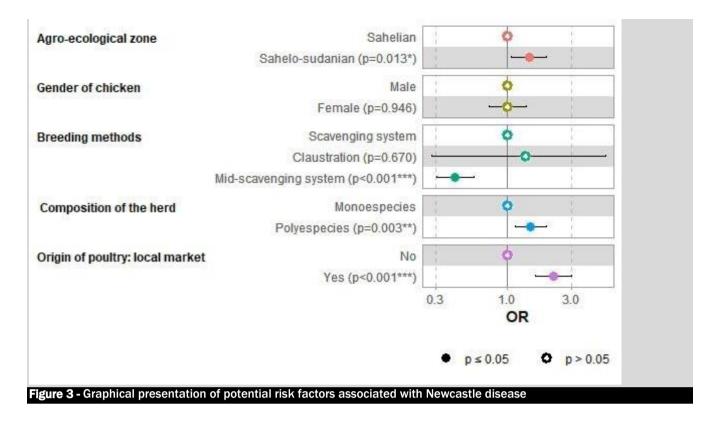
Factor	Categories	Odd_ratio	IC (odd_ratio)	P-value
	Sahelian		Reference	
Agro-ecological zone	Sahelo-Sudanian	1.45	1.08 - 1.95	0.013*
Gender of chicken	Male	Reference 1.45 1.08 - 1.95 Reference 1.01 0.75 - 1.38 Reference 1.36 0.28 - 5.28 1.36 0.28 - 5.28 1.5 1.15 - 1.95 Reference 1.02 1.5 1.15 - 1.95 Reference 2.19		
dender of chicken	Female		0.9	
	Scavenging system		Reference	
Breeding methods	Mid-scavenging system	0.42	0.28 - 5.28	< 0.001**
	Claustration	1.36	Reference 1.08 - 1.95 Reference 0.75 - 1.38 Reference 0.28 - 5.28 0.28 - 5.28 Reference 1.15 - 1.95 Reference 1.62 - 2.99	0.7
Composition of the hard	Monoespecies	1.01 0.75 - Refere ystem 0.42 0.28 - 1.36 0.28 - Refere 1.36 1.15 - Refere 1.5 1.15 - Refere 1.5 1.15 -	Reference	
Composition of the herd	Polyespecies	1.5	1.15 - 1.95	0.003**
Origin of noultry local market	No		Reference	
Origin of poultry: local market	Yes	2.19	1.62 - 2.99	< 0.001**
* Significant, and ** Very significant. Ref category.	erence: the comparison between the categ	ories of the factor	was made with respe	ct to a chosen



A: Communes in the study area; B: Departments in the study area; the prevalence distribution is divided into four different colored classes: Low prevalence, medium prevalence, high prevalence, and very high prevalence.

213

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DISCUSSION

The present serological study revealed the presence of antibodies against the Newcastle disease virus in village chickens sampled in all communes of the study area except Kantché. These results consequently showed the presence of the Newcastle disease virus in village chickens in the entire study area. In both study areas, an overall seroprevalence of 18.6% was obtained using ELISA. These results are comparable to several other previous studies. Abraham-Oyiguh et al. (2014), and Meher et al. (2020) reported a prevalence of 17%, and 19.83% in their study conducted in Nigeria, and Bangladesh, respectively. The prevalence of Newcastle disease in the present study is higher than in previous reports, where Mai et al. (2014) in Cameroon and Sahoo et al. (2022) in India revealed a prevalence of 10.5% and 11.7%, respectively. However, the overall prevalence in the present study is lower, compared to those previously reported by Channa et al. (2020) and Aliye et al. (2022). These authors reported a prevalence of Newcastle disease at 48.7% and 50% in Ethiopia and Pakistan, respectively.

The logistic regression test revealed that the Newcastle prevalence was significantly higher (p < 0.05) in the Sahelo-Sudanian agro-ecological zone (22.1%) than in the Sahelian agro-ecological zone (13%). Therefore, the risk of seropositivity to the Newcastle disease virus was higher in the Sahelo-Sudanian zone than in the Sahelian zone. The reports of the present study are in agreement with those of a Malian study, where Molia et al. (2017) reported that Newcastle disease is more likely to be present in the Sudanian agro-ecological zone than in the Sahelian agro-ecological zone. Njagi et al. (2010) reported climate as a risk factor in the occurrence of Newcastle disease in indigenous chickens of Kenya. However, these authors found that the prevalence of Newcastle disease was significantly higher in the hot dry zone (17.8%) than in the humid cool zone (9.9%).

Poultry raised in a free-range breeding system (20.5%) were significantly different from those raised in a semi-freerange system (11.2%) regarding the prevalence of Newcastle disease virus antibodies (p < 0.05). The Newcastle disease seropositivity of a bird kept in a scavenging system was higher than that of a bird kept in a semi-divisional breeding system. The present observations corroborate those of a previous study conducted in Nigeria, where Lawal et al. (2015) reported exposure of animals to infectious diseases, such as Newcastle disease, in the extensive breeding system.

The prevalence of Newcastle disease virus seropositivity was significantly higher in multi-species avian breeding (22.5%) than in single-species avian breeding (16%, p < 0.05). The results of the present study are in agreement with those of a serological study conducted in Nigeria on Newcastle disease virus antibodies in local chickens, ducks, and pigeons, where Abah et al. (2020) revealed that the prevalence of Newcastle disease virus antibodies was higher in ducks (20.5%) than in chickens (10%) and pigeons (7.5%). These authors reported the presence of the Newcastle disease virus in the avian population, and the epidemiological role that ducks, chickens, and pigeons may play in transmitting the Newcastle disease virus to other susceptible poultry when kept in close proximity.

There was a significant difference between chickens purchased from the local live poultry market (21.1%) and those purchased from other poultry supply sources (14.1%) regarding Newcastle disease seroprevalence (p < 0.05). The logistic regression test showed that chickens purchased from the local live poultry market were more likely to be seropositive for

Newcastle disease virus than those purchased from other poultry sources. These observations are consistent with a study by Ipara et al. (2019), reporting the positive effects of complex chicken trade channels on the frequency of Newcastle disease outbreaks in Kenya. Lawal et al. (2015) found that 65.5% of farmers usually took their animals to market for sale to reduce economic losses during outbreaks, which could amplify the transmission of Newcastle disease among poultry of different species present in the same market.

CONCLUSION

The present study established that Newcastle disease is endemic in the southern part of the Maradi and Zinder regions of Niger. A higher prevalence was observed in the Maradi region (28.3%), compared to the Zinder region (11.8%) with a significant difference. The agroecological zone, type of breeding practised, species mix, and source of poultry supply were statistically identified as the main risk factors for Newcastle disease seropositivity. Apart from health issues, Newcastle disease is of significant nutritional and socioeconomic importance due to the reduced production and high mortality it causes. To minimize such losses, efforts must be focused on intensifying vaccination campaigns and educating poultry farmers to adopt biosecurity measures. Finally, further studies should be considered to effectively control this disease to determine the circulating strains to match the control strategies with the epidemiological situation.

DECLARATIONS

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Authors' Contribution

This work was carried out with the contribution of all authors. Ahamidou Moustapha designed the protocol, collected and analyzed data, and drafted the manuscript. Essodina Talaki and Adamou Akourki validated the protocol, supervised the data collection, and revised the manuscript. Haladou Gagara participated in the analysis of the samples. All authors read and approved the final version of the manuscript.

Conflict of interests

The authors declare that they have no competing interests.

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