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SARS-CoV-2 seroprevalence and associated factors among outpatient attendees at health facilities in different provinces in Chad

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Abstract

Background Chad with 7,698 confirmed cases of infection and 194 deaths since the beginning of the COVID-19 pandemic, is one of the African countries with the lowest reported case numbers. However, this figure likely underestimates the true spread of the virus due to the low rate of diagnosis. The high rate of asymptomatic infections reflects the reality of SARS-CoV-2 transmission in Chad. In this study, we estimated the seroprevalence and identified factors associated with SARS-CoV-2 infection.

Methods A cross-sectional study was conducted between September 2022 and February 2023. A total of 1,290 plasma samples were collected from outpatient attendees at Health Facilities located in 11 provinces of Chad and tested by ELISA method, for the presence of IgG antibodies to SARS-CoV2 nucleocapsid (N) protein. KoboToolbox was used to gather data from the participants and data were analyzed using STATA 16.

Results The overall seroprevalence was 83.0% [95% CI = 81.6%—85.5%], with variations between provinces, ranging from 99.2% [95% CI = 94.0%—100%] in Moundou (Southern Chad) to 46.8% [95% CI = 36.0% -57.1%] in Biltine (Eastern Chad). Factors associated with the seroprevalence included military occupation (OR = 0.37 CI [0.80–1.77] $p = 0.025$) and age group between 55–64 years (OR = 0.33 CI [0.15–0.72] $p = 0.005$). While, other factors, such as gender and age were not significantly associated with seroprevalence.

Conclusion Our results indicated that, the seroprevalence of COVID-19 in Chad is among the highest in Sub-Saharan Africa. These estimates could guide the response and public health policy decisions, enhancing the management of future outbreaks involving respiratory pathogens.

Keywords Seroprevalence, SARS-CoV-2, COVID-19, Associated factors, Chad

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Background

When the World Health Organization (WHO) declared COVID-19 a global pandemic, scientists anticipated that Africa would be the hardest hit in terms of incidence, prevalence, and mortality [1]. However, current data show that Africa has been relatively spared compared to the rest of the world, accounting for only 1.2% of global infections and 2.5% of deaths attributed to the virus (SARS-CoV-2) [2].

Recently, serological studies conducted across African countries have revealed elevated levels of viral circulation despite the relatively low number of reported cases and deaths [3–5].

A meta-analysis of 965 seroprevalence studies carried out between January 2020 and April 2022, with 41.0% of the studies conducted in low- and middle-income countries, found that the overall seroprevalence due to both infection and vaccination was 59.2% [56.1% to 62.2%] [6]. In the African region, seroprevalence surged dramatically, rising from 26.6% [24.6 to 28.8] in January 2021 to 86.7% [84.6% to 88.5%] in December 2021 [6]. In sub-Saharan Africa, despite low reported COVID-19 case and mortality rates, some studies have shown a high overall anti-SARS-CoV-2 seroprevalence [7, 8].

These findings clearly demonstrate that SARS-CoV-2 infections are far more widespread than indicated by epidemiological surveillance data [6]. The underestimation of the true scale of the outbreak in Africa is largely due to limited access to testing and the high percentage of asymptomatic individuals. Importantly, symptomatic infections and severe forms of the disease are rare among young people, a population segment strongly represented in Africa where 40.0% of the individuals are under 15 years old and an average age of around 20 years [9, 10].

However, widespread circulation of the virus increases the likelihood of new mutations that could alter its biological properties, such as transmissibility, severity, infectivity and antigenicity [11].

In this context, SARS-CoV-2 antibodies detection as markers of viral exposure is crucial for identifying the proportion of previously infected asymptomatic individuals. Two seroprevalence studies conducted in N'Djamena in October 2021 after the second wave of the pandemic, reported an overall seroprevalence of 69.5% [67.7 to 71.3] [12] and confirmed the presence of neutralizing SARS-CoV-2 antibodies in 59.0% of the analyzed samples [13].

Chad, a landlocked country in Central Africa with a population of 16.9 million and a median age of just 16.6 years, is one of the poorest countries in the world, with a fragile healthcare system. Universal health coverage remains significantly limited, and health indicators, though improved in recent years, still lag [5].

To monitor the extent and progression of the COVID-19 epidemic in Chad, it is crucial to measure the proportion of individuals who have developed antibodies against the virus, especially given the country's young population. Such data will allow for the implementation of targeted and context-specific prevention strategies, as well as an evaluation of the effectiveness of the measures in place. To our knowledge, this is the third study on anti-SARS-CoV-2 seroprevalence in Chad. Unlike previous studies, this survey, conducted between 19 September 2022 and 3 February 2023 (after the fourth wave of the pandemic), covered 11 provinces.

The study aimed to estimate seroprevalence and identify risk factors associated with SARS-CoV-2 infection in ambulatory patients at hospitals across 11 provinces of Chad.

Methods

Study design and setting

A cross-sectional study was conducted between September 2022 and February 2023 to expand both the temporal and geographical scope of a previous pilot study conducted by researchers from the Major Tropical Epidemics Laboratory (LAGET) and the Institute for Research on Livestock and Development (IRED). This earlier study had reported a high seroprevalence of anti-N SARS-CoV-2 antibodies in N'Djamena [12].

This current study is part of an initiative led by the National Coordination Committee for Health Crisis Management, established by the Republic of Chad, in response to the COVID-19 pandemic. Its primary goal is to assess community immunity to SARS CoV-2 across 10 provinces and the city of N'Djamena following the fourth wave of the pandemic. Provinces were selected based on several criteria: (i) higher incidence of infections or deaths; (ii) presence of healthcare facilities with the ability to process and transport samples according to required protocols, and (iii) specific territorial and geographical factors (e.g. Ouaddaï, where the main city, Abéché, serves as a key transit hub known as the "gateway to the desert"). N'Djamena was included to provide updated data in comparison to the previous survey. The study was made possible through collaboration with the main healthcare facilities in each of the 11 Chadian provinces: Chari-Baguirmi (N'Djamena), Ouaddaï (Abéché), Lake Chad (Bol), Logone Oriental (Doba), Waddi-Fira (Biltine), Logone Occidental (Moundou), Moyen Chari (Sarh), Batha (Ati), Mayo-Kebbi Est (Bongor), Mayo-Kebbi Ouest (Pala) and Guerra (Mongo).

Given the mixed levels of knowledge, attitudes and practices regarding COVID-19 within the Chadian population [14], healthcare facilities were the most suitable locations to recruit an adequate number of participants.

While enrolling individuals visiting hospitals for reasons unrelated to the serological study, may introduce selection bias and potentially influence the results, in the Chadian context, this was the only viable option for accessing biological samples and relevant data from individuals. This study, approved by the National Ethics Committee, was made possible thanks to the active involvement of doctors in healthcare facilities, who raised public awareness and emphasized the importance of participation. It should be noted that, outside this specific context, obtaining sufficient participation from the population would have been impossible.

Study population and sample size

During the data collection period, all individuals, regardless of age, who attended an outpatient appointment at the healthcare facilities and freely agreed to participate, were considered eligible for the study. There were no exclusion criteria at the enrollment stage.

Selection criteria were applied in the subsequent phase, based on the equality of the samples and completeness of the required information (e.g. participant with hemolyzed samples or incomplete data were excluded).

Of the 1,539 volunteers initially enrolled, 249 (16.1%) were excluded due to hemolyzed samples or incomplete data. As a result, the study was conducted on a final total number of 1,290 plasma samples.

Since population density varies across provinces [15], the number of participants at each site was determined by the attendance rate at the referral hospitals during the study period.

Data collection

After obtaining informed consent, an electronic questionnaire administrated through KoboToolbox was used to collect data on socio-demographic characteristics, COVID-19 vaccination status, and other relevant parameters. Participants were also asked to provide a venous blood sample.

Laboratory procedures

Whole blood was collected in BD VACUTAINER K3 EDTA tubes. Plasma was separated by centrifugation at 3000 rpm for 10 min at room temperature. The plasma was aliquoted into cryotubes and stored at -80 °C until use. Laboratory analyses were conducted between January and February 2023 at LAGET and IRED in N'Djamena.

Detection of Anti-SARS-CoV-2 nucleocapsid protein

The identification of plasma IgG antibodies against the SARS-CoV-2 Nucleocapsid (N) antigen, was performed using the indirect enzyme-linked immunosorbent assay (ELISA) technique, following the manufacturer's instructions (Diatheva COVID-19 NP IgG ELISA).

Briefly, the patient's anti-N SARS-CoV-2 IgG antibodies specifically bind to the N SARS-CoV-2 antigen immobilized on the plate. Secondary antibodies conjugated to horseradish peroxidase (HRP) then bind to the captured IgG antibodies. The resulting immune complex is visualized by adding the substrate ABTS (2,2'-Azino bis (3-ethylbenzothiazoline-6-sulfonic acid), producing a green reaction product. Absorbance at 405 nm was measured using an ELISA microplate reader (Multitaska FC Microplate Photometer). The presence of specific anti-SARS-CoV-2 IgG antibodies correlates with the color intensity of the sample. Results were calculated as the ratio of the sample absorbance (S) to the cut-off value (Co), with samples considered positive when $S/Co \geq 1.5$. The manufacturer reported a sensitivity of 98.8% and a specificity of 98.0%.

Data analysis

All data were analyzed using STATA version 16.0 (StataCorp, 2014). A descriptive analysis of the study population characteristics was performed.

Estimation of the seroprevalence of SARS-COV-2

Differences in seroprevalence among variables were analyzed using prevalence ratios and chi-square tests, with 95% confidence intervals (CIs) and significance levels estimated.

We conducted a binary logistic regression using a bottom-up stepwise approach, including independent variables associated with the dependent variable at the 20% threshold and those identified in the literature. The contribution of each variable was assessed using the parametric Wald test. The final model was evaluated for fit using the Hosmer–Lemeshow test, and model specification was verified by the link test. Missing data were addressed either by imputing values for certain variables, such as age, or by analyzing the data without imputation using the data deletion method, specifically through multivariate analysis of available cases (e.g., blood group/rhesus variable).

Results

Sociodemographic data

A total of 1,290 individuals were eligible and were included in our analysis. The most common age groups were 25–34 years (460/1290) and 15–24 years (353/1290), although the over-65 s were under-represented (22/1290).

Men were the most represented sex at (669/1290), with a sex ratio of 1.1. Of the 1290 people included, only 15 had previously formally been diagnosed as positive for COVID-19. In addition, a small percentage of

participants had pre-existing conditions: 0.7% for other infectious diseases and 4.0% for non-communicable diseases. These data enabled us to hypothesize that the health status of enrollees, had little influence on the statistical analysiscarried out to identify risk or protective factors for SARS-CoV-2 infection (Table 1).

Seroprevalence of anti-nucleocapsid IgG SARS-CoV-2 (anti-N) and associated factors

Data on seroprevalence indicated an overall rate of 83.0% [95% CI = 81.6% to 85.5%] (Table 1). Depending on the region, seroprevalence varied greatly from one part of Chad to another, with the higher rate 99.2% [95% CI = 94.7% to 100%] in Moundou (Southern Chad) and the lowest 46.8% [95% CI = 36.5% to 57.3%] in Biltine, the Eastern part of Chad. In the capital, N'Djamena, only one site was considered, and the rate was 92.1% [95% CI = 86.9% to 95.3%] (Table 2).

In the regression analysis, two variables were significantly associated with the event of interest: age group and occupation. Univariable analysis showed that the probability of developing anti-N after contact with the SARS-CoV2 virus was reduced by 44.0% in the 55–64 age group (OR = 0.56 CI [0.29–1.08] $p = 0.086$). Military personnel had a 63.0% lower probability of being seropositive for SARS-CoV-2 (OR = 0.37 CI [0.16–0.88] $p = 0.033$) compared with those who were not employed. However, in the multivariate analyses, this age group (55–64 years) showed a 67.0% reduction in the probability of seropositivity (OR = 0.33 CI [0.15–0.72] $p = 0.005$), while military personnel had a 61.0% lower probability of being HIV positive (OR = 0.39 CI [0.16–0.92] $p = 0.033$) (Table 1).

Discussion

This SARS-CoV-2 seroprevalence survey, conducted across ten provinces of Chad and the capital N'Djamena, revealed a significant discrepancy between the actual percentage of individuals infected and the officially reported cases in the country.

During the survey period (September 2022 to February 2023), 7.698 confirmed cases of COVID-19 were reported in Chad, including 193 deaths, representing a national mortality rate of 2.5% [16]. However, the true prevalence of the virus was considerably higher, as shown by seroprevalence data.

In the early stages of the COVID-19 pandemic, most seroprevalence studies were conducted in high-income countries, leaving a substantial knowledge gap in low- and middle-income (LMI) nations. For instance, only 23% of the 968 studies included in a systematic literature review of SARS-CoV-2 seroprevalence surveys conducted in 2020 were from LMI countries [17]. To address this gap, the. Centres for Disease Prevention and Control (CDC), along with the World Health Organization (WHO) and other international partners, began supporting seroprevalence studies in LMI countries starting in mid-2020. In line with CDC guidelines for international seroprevalence surveys, this study aimed to estimate the prevalence of SARS-CoV-2 antibodies in a specific regional population. These findings serve as a proxy for evaluating prior virus exposure and/or vaccination rates. The data have proven invaluable to policymakers, allowing them to estimate the actual burden of the disease, identify transmission hotspots, assess the effectiveness of prevention strategies, gauge population immunity, identify at-risk groups, and inform public health planning, particularly regarding vaccination needs.

This survey found an overall seroprevalence of 83.0% making a 19% increase compared to data from the 2021 survey in N'Djamena [12].

A review by Chisale et al. in 2022 [3] of 153 studies conducted in Africa, reported lower seroprevalence rates than those in our study.

This difference likely stems from the fact that earlier studies were conducted before our survey period. The African studies showed an overall prevalence of 22% (95% CI: 14–31), with country-specific seroprevalence rates ranging from 0 to 63% in 2021. Central Africa had the highest seroprevalence (41%, CI: 14–72), compared to Southern Africa (34%, CI: 13–59), West Africa (25%, CI: 13–39), North Africa (13%, CI: 2–32), and East Africa (12%, CI: 2–28). Similarly, a WHO meta-analysis of 151 studies from January 2020 to December 2021 [18] found that SARS-CoV-2 exposure in Africa surged from 3% (range: 1.0–9.2%) in June 2020 to 65% (range: 56.3–73%) by September 2021, equating to 800 million infections compared to 8.2 million reported cases at the time. This indicated that true infection rates were 97 times higher than reported cases, with a sharp rise in infections following the emergence of the Beta and Delta variants.

Our study, conducted in 2022 after Chad's fourth wave, coincided with the appearance of more contagious variants like Delta and Omicron in November 2021, which spread rapidly [19–21] as confirmed by genomic surveillance data.

The extension of the survey to different provinces, including reference hospital structures, was driven by the need to identify transmission hotspots and assess the diagnostic capacity of health facilities.

A recent study by Karampre et al. in Cameroon [22] show provincial differences, with seroprevalence ranging from 7.5% (95% CI: 5.9%–9.5%) in the East region to 12.4% (95% CI: 10.4%–14.8%) in the Far North and Northwest regions.

Table 1 Seroprevalence of COVID-19 and associated factors (*the reference corresponds to individuals without the specific disease)

					Univariable		*Multivariable*	
Factors	**Seropositive individuals**	**Seronegative individuals**	**Total**	**Seroprevalence %**	**Odds ratio 95%**	***P value***	***Odds ratio ajusted 95%***	***P Value***
Overall	1073	217	1290	83 (81–85)	–	–	–	–
Sex					–	–	–	–
Male	556	113	669	83 (80–85)	0.98(0.73–1.32)	*0.945*	–	–
Female	517	104	621	83 (80–85)	Reference		–	–
COVID–19 vaccine	323	54	377	85 (81–88)	1.29 [0.93–1.81]	*0.124*	–	–
Age group						–	–	–
< 15	12	4	16	75 (49–90)	0.53 [0.16–1.71]	*0.296*	0.95 [0.62–1.65]	0.840
15–24	292	61	353	82 (78–86)	0.85 [0.59–1.25]	*0.428*	1.09 [0.77–1.71]	0.654
25–34	390	70	460	84 (81–87)	Reference		1	
35–44	217	46	263	82 (77–86)	0.84 [0.56–1.26]	*0.411*	0.67 [0.42–1.13]	0.114
45–54	98	20	118	83 (75–88)	0.87 [0.51–1.51]	*0.644*	0.75 [0.69–1.81]	0.366
55–64	44	14	58	75 (63–85)	0.56 [0.29–1.08]	*0.086*	0.33*[0.15–0.72]	0.004
> 65	20	2	22	90 (70–97)	1.79 [0.41–7.85]	*0.437*	1.01 [0.34–4.26]	0.982
Profession					–	–		
Unemployed	372	79	451	82(74–88)	Reference		1	
Other profession	295	50	345	85 (77–89)	1.25 [0.85–1.84]	0.252	1.33 [0.89–1.98]	0.330
Employe	90	15	105	85 (76–89)	1.27 [0.70–2.51]	*0.427*	1.36[0.73–2.58]	0.405
Pupil/student	242	54	296	81(80–88)	0.95 [0.64–1.39]	*0.800*	0.91 [0.60–1.38]	0.327
Military	16	9	25	64 (43–80)	0.37 [0.16–0.88]	0.025	0.39* [0.16–0.92]	0.033
Health worker	58	10	68	85 (77–91)	1.23 [0.60–2.31]	*0.427*	*1.27 [0.61–2.62]*	0.739
Blood group					–	–		
A	139	16	155	89 (83–93)	Reference			
AB	20	6	26	76 (56–91)	0.38 [0.13–1.09]	*0.073*	–	–
B	148	25	173	85 (79–90)	0.68 [0.34–1.33]	*0.261*	–	–
O	282	50	332	85 (79–93)	0.64 [0.35–1.18]	*0.157*	–	–
Unknown group	484	120	604	80 (76–83)	0.46 [0.26–0.80]0.261	0.007		
Rhesus +	582	93	675	86 (83–88)	Reference		–	–
Rhesus-	7	4	11	63 (30–89)	0.27 [0.08–0.97]	*0.045*		
Chronic disease								
Diabetes	9	2	11	81 (49–95)	0.90 [0.19–4.23]	*0.904*	–	–
Hypertension	18	4	22	81 (60–93)	0.90 [0.30–2.71]	*0.863*	–	–
Asthma	7	1	8	87 (46–98)	1.41 [0.17–11.58]	*0.077*	–	–
Obesity	10	1	11	90 (56–98)	2.03 [0.25–15.95]	*0.500*	–	–
Symptoms (fever cough, cold)							–	–
Symptomatic	293	48	341	85 (81–89)	1.32 [0.93–1.87]	*0.115*	–	–
Asymptomatic	780	169	949	82 (79–84)	Reference		–	–
Contact with case	17	3	20	85 (62–95)	1.12[0.83–1.85]	*0.281*	–	–
Old COVID-19 case	11	4	15	73 (46–89)	0.55[0.17–1.74]	*0.312*	–	–
Vaccination								
Unvaccinated	754	164	918	82 (79–84)	Reference		1	
Johnson & Johnson	149	38	187	79 (73–85)	0.85[0.57–1.26]	*0.429*	0.90[0.59–1.36]	*0.634*
PFizer	170	15	185	91 (86–95)	2.46[1.41–4.29]	*0.001*	2.60[1.46–4.61]	*0.001*

Number of observations 1290

number of covariate patterns = 93

Pearson chi2(23) = 84.70

Prob > chi2 = 0.3384

Table 2 Distribution of COVID-19 seroprevalence according to 10 provincial hospitals in Chad and in N'Djamena (in the table are reported the names of the cities where the hospitals involved are located and the total population for each province)

Factors	Positive samples	Total samples	Seroprevalence (%) (95% CI)	Name of the provinces
Abéché	56	63	88.9 (78.8–94.5)	Ouaddai
Ati	77	99	77.8 (68.6–84.8)	Batha
Biltine	44	94	46.8 (36.5–57.3)	Wadi Fira
Bol	124	129	96.1 (90.5–98.7)	Lac
Bongor	137	139	98.6 (94.3–99.7)	Mayo-Kebbi East
Doba	80	103	77.7 (68.7–84.6)	Logone Oriental
Mongo	70	129	54.3 (45.7–62.6)	Guera
Moundou	128	129	99.2 (94.7–100)	Logone Occidental
N'Djamena	151	164	92.1 (86.9–95.3)	Capital
Pala	70	101	69.3 (59.2–77.9)	Mayo-Kebbi West
Sarh	136	140	97.1 (91.4–98.9)	Moyen Chari

In our study, seroprevalence varied significantly across the 10 provincial hospitals and the hospital in N'Djamena. It was highest in Moundou, in the southern Chad, at 99.2% [95% CI = 94.4% to 100%] and lowest in Biltine, eastern Chad, at 46.8% [95% CI = 36.5% to 57.3%]. This variation may only be partially due to the population density as in case of Moundou (77 inhabitants/km^2) compared to Biltine (9 inhabitants/km^2). Population density is, indeed, a well-known risk factor for airborne infections and even if our study sought to gather, within the limitations of available resources, information on possible risk factors among participants we consider fundamental to delve deeper into the number of factors that may have influenced these differences. In any case despite this limitation our findings, shared with Chad's Ministry of Health, have underscored several key challenges: a) ineffective surveillance systems failing to capture most cases, b) a significant gap in the public health system's ability to diagnose and report asymptomatic cases, c) a less severe clinical presentation nationwide, which may reduce healthcare-seeking behavior, and d) the need to prioritize strengthening diagnostic capacities and applying targeted (at provincial level) mitigation strategies in future epidemics.

Looking at other associated factors through the seroprevalence of IgG anti-nucleoprotein. The data show lower seroprevalence in the 55–64 age group (OR = 0.56 [0.29–1.08], $p = 0.086$). This aligns studies by Chisale et al. in 2022 [3], Pollàn et al. in Spain in 2020 [11] and Stringhini et al. in Switzerland in 2020 [23], all of which found lower seroprevalence among older adults, potentially due to immunosenescence and more cautious behavior in this group.

Older adults may have lower seroprevalence due to immunosenescence, which can lead to higher mortality [24] and, consequently, a lower proportion of people with evidence of previous infection. In addition, more cautious behavior may also lead to fewer infections in this age group. Regarding socio-professional category, in our study, military personnel were less likely to have positive anti-SARS-CoV2 antibodies compared with those not in employment (OR = 0.37 CI [0.16–0.88] $p = 0.025$). This may be explained by the fact that military personnel are in barracks and are not in contact with the community. In addition, discipline and strict obedience to rules and orders are mandatory in the military. So physical distancing measures are better applied, and the adopted mitigation strategies could be, in case of next epidemic also be implemented and disseminated to the entire population.

One major limitations of the study, despite the effort to standardize the proposed methodology, was the difficulty in obtaining a complete information sheet for each participant. This reduced the possibility of extending the multivariant analysis to all parameters considered relevant, such as blood group.

Indeed, susceptibility of viral infection has been found to be related to the blood group [25]. ABO blood group is long known to be an influencing factor for the susceptibility to infectious diseases, and many studies have been describing associations between ABO blood types and COVID-19 infection and severity, with conflicting findings [25]. Blood groups are frequent targets in epidemiological investigations since they are genetically determined traits with known polymorphic expression among individuals and populations. Several studies have been reporting significant associations between blood type A and higher susceptibility to SARS-CoV-2 infection [26] confirming the partial results obtained in this study. The data, although incomplete, on the suggested trend on the correlation between blood group and greater susceptibility to infection, leads us to support the importance of including this parameter in future studies.

Conclusions

Our findings indicate that, while the apparent health impact of the COVID-19 pandemic in Chad has been lower than in other parts of the world, the virus has spread extensively. The varying levels of antibodies detected suggest some degree of immune memory within the population. Identifying individuals potentially immune to SARS-CoV-2 could play a crucial role in determining when and how to ease social distancing restrictions and can assist health policymakers in shaping vaccination campaigns.

Accurately assessing the true scale of an outbreak is essential for developing effective public health measures and control strategies. Seroprevalence surveys enable public health officials and policymakers to identify hotspots and regions with reduced transmission, allowing for targeted responses. Serological data can also guide decision-makers in public health strategies, including vaccine procurement and prioritization, resource allocation (e.g., PPE, financial support), and the development of mitigation tactics for future epidemics.

Abbreviations

SARS-CoV-2	Severe Acute Respiratory Syndrome-Coronavirus-2
CNBT	Chad National Bioethics Committee
MAGIS	Italian Jesuit Movement and Action for Development
AICS	Italian Agency for Development Cooperation
LAGET	Major Tropical Epidemics Laboratory
CIRCB	Chantal Biya International Reference Center for research on HIV/AIDS prevention and management.
ELISA	Enzyme-linked immunosorbent assay
IgG	Immunoglobulin G
EDTA	Ethylenediaminetetraacetic Acid
IRED	Livestock Research Institute for Development
WHO	World Health Organization

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Authors' contributions

MAM, MFA and SA designed this survey. AMM helped design the survey. KD, and FAZ, ALDW, collected the original data. OD analyzed the data. KD, NN, and OD, contributed to the interpretation of the data. OD, KD, GC, FJDM, DC, and VC contributed to the writing of the article. AB, CAM, IM, NA, MIH, NH, KF, AAM, YMD, AMA, MG, contributed equally to this study. All the authors have read, edited and contributed to the content of this manuscript.

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Data availability

The data sets used and/or analyzed in this study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

The study protocol was approved by the Chad National Bioethics Committee (CNBT) on 08 April 2022 under number: 011CMT/PC/PMT/MESRI/SG/CNBT/2022 and authorized by the Chad Ministry of Public Health under number: N°2326/CMT/PC/PMT/MSPSN/SE/SG/DGPC/DPERO/SRO/2022. The protocol was presented to 11 focal points including 10 provinces and N'Djamena selected for the study. Training sessions on the data collection tool were organized before the start of data and sample collection. An information sheet was given to each voluntary participant as well as the signature of informed consent and assent for minors. The informed consent was obtained from all participants involved. For adult participants (aged ≥ 16 years), informed consent was obtained directly from each individual. For minor participants (aged < 16 years), informed consent was obtained from parents or the legally authorized representative (LAR). In both cases, consent was obtained in accordance with the ethical guidelines established by the approving ethics committee. Each participant was assigned a unique code, and the data was handled confidentially.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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