High prevalence of SARS-CoV-2 detected by reverse transcription polymerase chain reaction at the *Laboratoire des Grandes Épidémies Tropicales* in N'Djamena, Chad

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Abstract

The COVID-19 pandemic, caused by SARS-CoV-2, posed a significant challenge to global public health systems, particularly in resource-limited settings. Accurate molecular diagnosis is critical for effective disease surveillance and control. This study aimed to determine the prevalence and associated factors of SARS-CoV-2 infection among individuals tested at the *Laboratoire des Grandes Épidémies Tropicales* (LAGET) in N'Djamena, Chad. A retrospective cross-sectional study was conducted from January 2021 to June 2022. Nasopharyngeal swabs were collected from symptomatic and asymptomatic individuals and tested for SARS-CoV-2 using reverse transcription polymerase chain reaction (RT-PCR). Associations between test positivity and demographic/clinical factors were assessed using odds ratios (OR) and chi-square tests. Among 625 participants (mean age 37±14 years; 29.3% female), the overall SARS-CoV-2 positivity rate was 31.5% (197/625). The viral load distribution among positive cases was as follows: 20.3% high (cycle threshold [Ct] <20), 51.3% moderate (Ct 21-30), and 21.3% low (Ct 31-36). Being symptomatic (OR=1.68, 95% confidence interval [CI]: 1.18-2.38, p=0.003) and having a sample collected at an external health facility (OR=0.28, 95% CI: 0.19-0.40, p<0.0001) were significantly associated with a higher likelihood of testing positive. No significant associations were found with age or sex. The high prevalence of SARS-CoV-2 detected at LAGET, far exceeding national figures, indicates substantial community transmission and underreporting in Chad. These findings underscore the critical need for strengthened molecular diagnostic capacity and decentralized testing services. Implementing an integrated One Health surveillance approach is essential for managing the current pandemic and preparing for future epidemic threats.

Introduction

The emergence of the novel coronavirus SARS-CoV-2 in late 2019 marked the beginning of an unprecedented global health crisis. On March 11, 2020, the World Health Organization (WHO) officially declared COVID-19 a pandemic.[1] As of August 2022, more than 584 million confirmed cases and over 6.4 million deaths had been reported globally.[2]

Contrary to early projections that anticipated devastating

impacts on the African continent, reported incidence and mortality rates have remained comparatively lower. This paradox may reflect a combination of younger demographics, limited diagnostic capacity, underreporting, and differences in surveillance strategies.[3]

In Somalia, the first case of COVID-19 was confirmed in March 2020.[4] As of July 2025, approximately 27,334 confirmed cases and 1,361 deaths had been officially reported.[5] By March 2024, nearly 50% of Somalia's population was fully vaccinated, represented over 12 million doses administrated through the COVID-19 Emergency Vaccination Project coordinated by the World Bank and the WHO.[6]

Somalia and Chad share several contextual similarities that justify a comparative public health perspective. Both countries have vast territories with low population density, a predominantly pastoral economy centered on camel herding, and large nomadic or semi-nomadic populations. These factors influence mobility, access to healthcare, and disease surveillance. Furthermore, both nations face similar challenges in diagnostic decentralization, vaccination coverage, and epidemic preparedness.[6-8]

In Chad, the first case of COVID-19 was confirmed on March 19, 2020.[9] By January 2022, official figures reported 7,211 cases and 190 deaths, suggesting a relatively low national prevalence.[10]

Accurate and timely diagnosis remains a cornerstone of epidemic response. Reverse transcription polymerase chain reaction (RT-PCR) is widely considered the gold standard for SARS-CoV-2 detection due to its high sensitivity and specificity.[11] However, in resource-limited countries such as Chad, diagnostic infrastructure is often centralized, restricting access to testing outside major health facilities.

The *Laboratoire des Grandes Épidémies Tropicales* (LAGET), located within the Bon Samaritain University Hospital (CHU-BS) in N'Djamena, plays a critical role in national surveillance. Monitoring positivity rates and identifying associated demographic and clinical factors can inform public health interventions and help bridge gaps in epidemic preparedness.

This study aimed to determine the prevalence of SARS-CoV-2 RT-PCR positivity at LAGET and to identify demographic and clinical factors associated with infection. The findings are intended to support evidence-based strategies for strengthening diagnostic capacity and to highlight the importance of integrated surveillance within a One Health framework that recognizes the interconnectedness of human, animal, and environmental health.[12]

Materials and Methods

Study design and setting

This was a retrospective cross-sectional study conducted at LAGET, located within the CHU-BS in N'Djamena, Chad. LAGET functions as a national reference laboratory for molecular diagnostics and epidemic surveillance. The study period extended from January 2021 to June 2022, covering multiple epidemic waves in Chad.

Study population and inclusion criteria

The study included all individuals referred to LAGET for SARS-CoV-2 RT-PCR testing during the study period. Both symptomatic and asymptomatic individuals were tested. Inclusion criteria were: i) submission of a nasopharyngeal swab for SARS-CoV-2 RT-PCR testing, and ii) availability of complete demographic and clinical data (age, sex, clinical status, and collection site). Specimens with incomplete information or invalid RT-PCR results were excluded.

Sample size calculation

Based on the Lorentz formula, with an expected prevalence of 0.04% (Ministry of Health data), a 95% confidence interval (CI), and a 5% margin of error, the minimum required sample size was estimated at 59. The final analysis included 625 individuals, providing substantial statistical power.

Sample collection and transport

Nasopharyngeal swabs were collected using sterile flocked swabs and transported in viral transport medium (VTM) according to the WHO guidelines.[13]

Samples collected at external health facilities were transported under a cold chain system and processed at LAGET within 24 hours of collection.

Laboratory procedures

Viral RNA was extracted using the QIAamp Viral RNA Mini Kit (Qiagen, Hilden, Germany), according to the manufacturer's instructions. RT-PCR amplification was performed with the Maccura™ SARS-CoV-2 Fluorescent PCR Kit (Maccura Biotechnology, Chengdu, China), targeting the *ORF1ab*, *N*, and *E* genes. Amplification and detection were performed on an ABI 7500 Real-Time PCR System (Applied Biosystems, Foster City, USA).

Thermal cycling conditions were as follows: reverse transcription at 50°C for 15 minutes, initial denaturation at 95°C for 15 minutes, followed by 45 cycles of denaturation at 94°C for 15 seconds and annealing/extension at 55°C for 45 seconds. A sample was considered positive if at least two viral targets were detected with a cycle threshold (Ct) value <37, in accordance with the WHO recommendations,[14] and manufacturer's instructions. Similarly, in Somalia, RT-PCR has been the reference method for SARS-CoV-2 diagnosis since the onset of the pandemic. With support from the WHO and international partners, the National Public Health Reference Laboratory (NPHRL) in Mogadishu and regional laboratories in Hargeisa, Garowe, and other state capitals were rapidly equipped with RT-PCR systems between 2020 and 2022.[4,15] By 2023, seven public health laboratories were performing molecular testing nationwide, complemented by the repurposing of GeneXpert platforms for remote areas.[15] Serological assays and rapid antigen tests have also been used to support epidemiological surveillance in hard-to-reach communities, in line with the strategic guidance of the WHO Eastern Mediterranean Regional Office (EMRO) and the Food and Agriculture Organization (FAO) regarding laboratories decentralization and One Health integration in fragile settings.[16,17]

Positive samples were stratified into three viral load categories based on Ct values: high viral load (Ct<20), moderate viral load (Ct 21-30), low viral load (Ct 31-36).

Data collection and variables

Demographic variables (age, sex), clinical status (symptomatic or asymptomatic), and site of sample collection (CHU-BS *vs.* external facilities) were recorded. Test results and Ct values were extracted from laboratory registers.

Statistical analysis

Data were entered and analyzed using SPSS version 25.0 and Epi Info™ version 7.2. Categorical variables were summarized as frequencies and percentages, while continuous variables were expressed as means ± standard deviation (SD). Associations between SARS-CoV-2 positivity and independent variables (sex, age, clinical status, collection site) were assessed using chi-square tests. Odds ratios (OR) with 95% CI were calculated. A p-value <0.05 was considered statistically significant.

Results

Sociodemographic characteristics

A total of 625 participants were included in this study. The mean age was 37±14 years (range: 1-85 years). The largest age group was 20-39 years (43.2%). The majority of participants were male (70.7%), with a male-to-female ratio of 2.4:1 (Table 1).

SARS-CoV-2 positivity and viral load distribution

The overall SARS-CoV-2 RT-PCR positivity rate was 31.5% (197/625). Among positive cases, 20.3% had high viral loads (Ct<20), 51.3% had moderate viral loads (Ct 21-30), and 21.3% had low viral loads (Ct 31-36).

These distributions are presented in Table 2 and illustrated in Figure 1, which highlights the predominance of moderate viral loads among positive individuals.

Factors associated with SARS-CoV-2 positivity

No significant association was found between SARS-CoV-2 positivity and sex or age group. However, symptomatic individuals and those sampled at external health facilities were significantly more likely to test positive (Tables 3 and 4).

Symptomatic individuals were significantly more likely to test positive compared to asymptomatic individuals (OR=1.68, 95% CI: 1.18-2.38, p=0.003). Samples collected at external facilities had more than twice the odds of being positive compared to those collected at CHU-BS (OR=2.16, 95% CI:1.52.3.06, p<0.001).

Discussion

This study revealed a high SARS-CoV-2 positivity rate of 31.5% among individuals tested at LAGET between January 2021 and June 2022. This prevalence is considerably higher than the national figures officially reported during the same period, which indicated fewer than 10,000 cumulative cases in Chad.[10] The discrepancy suggests a significant level of underreporting and highlights the limitations of surveillance systems in resource-limited settings.

The predominance of moderate-to-high viral loads (71.6%) among positive individuals emphasizes the potential role of asymptomatic and mildly symptomatic carriers in sustaining community transmission. Similar observations have been made in other African countries, where silent spread was facilitated by limited testing coverage and centralized laboratory systems.[18]

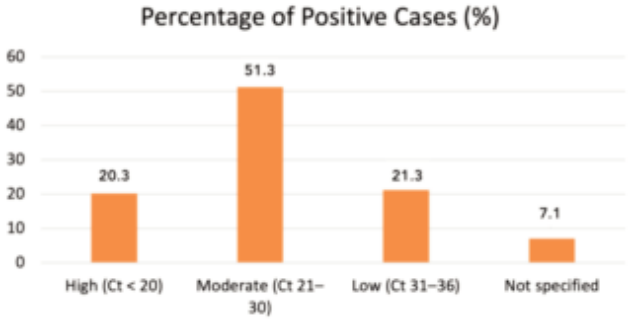

Figure 1. Distribution of SARS-CoV-2 viral load among positive cases at LAGET (n=197).

Table 1. Participants' sociodemographic characteristics.

Variable	Category	Frequency (n)	Percentage (%)
Sex	Male	442	70.7
	Female	183	29.3
Age group (years)	<20	102	16.3
	20-39	270	43.2
	40-59	185	29.6
	≥60	68	10.9
Mean age (years)	37±14	-	-

Table 2. SARS-CoV-2 positivity rate and viral load distribution.

Status	Category	Number (n)	Percentage (%)
Overall result	Negative	428	68.5
	Positive	197	31.5
Viral load (Ct) among positives (n=197)	High (Ct<20)	40	20.3
	Moderate (Ct 21-30)	101	51.3
	Low (Ct 31-36)	42	21.3
	Not specified	14	7.1

Ct, cycle threshold.

Table 3. Association between SARS-CoV-2 positivity and sociodemographic factors.

Factor	Category	Total tested (n)	Positive, n (%)	OR	95% CI	p
Sex	Male	442	136 (30.8)	1.12	0.77-1.62	0.53
	Female	183	61 (33.3)	Ref		
Age group (years)	<37	303	96 (31.7)	1.01	0.70-1.40	0.93
	≥37	322	101 (31.4)	Ref		

OR, odds ratio; CI, confidence interval.

Table 4. Association between SARS-CoV-2 positivity and clinical/contextual factors.

Factor	Category	Total tested (n)	Positive, n (%)	OR	95% CI	p
Symptoms	Symptomatic	221	86 (38.9)	1.68	1.18-2.38	0.003
	Asymptomatic	404	111 (27.5)	Ref		
Sampling Site	External facility	202	87 (43.1)	2.16	1.52-3.06	<0.0001
	CHU-BS	423	110 (26.0)	Ref		

OR, odds ratio; CI, confidence interval; CHU-BS, Bon Samaritain University Hospital.

Our results also demonstrated a significant association between clinical symptoms and positivity (OR=1.68, p=0.003). This is consistent with reports showing that symptomatic individuals have higher viral loads and increased transmissibility.[19] However, the detection of nearly one-quarter of positive cases among asymptomatic individuals underscores the necessity of expanded testing beyond symptom-based screening.

Importantly, samples collected at external sites were more than twice as likely to test positive compared to those collected at CHU-BS (OR=2.16, p<0.0001). This finding may reflect delayed presentation to external facilities, differences in case selection criteria, or possible clustering of outbreaks in communities distant from centralized testing hubs. Similar geographic disparities in positivity have been reported in other low-resource countries, where access to timely diagnosis is unevenly distributed.[3]

No significant associations were observed between age or sex and SARS-CoV-2 positivity, although global evidence indicates that older age and male sex are risk factors for severe disease rather than for infection itself.

From Chad to Somalia

The inclusion of this work in the *Somali Journal of Science and Technology Studies* (SJSTS) is particularly relevant, given the strong contextual parallels between Chad and Somalia. Both nations face structural limitations in health infrastructure, diagnostic decentralization, and surveillance coverage, especially among pastoral and nomadic populations. The lessons learned from the LAGET experience in Chad could thus inform Somalia's ongoing efforts to strengthen molecular testing capacity, improve vaccination equity, and enhance epidemic preparedness under a One Health framework.[16,17]

The comparative lens between Chad and Somalia highlights how decentralized molecular diagnostics, combined with One Health surveillance, can improve epidemic preparedness and resilience in fragile contexts across the Sahel and Horn of Africa. This shared experience underscores the value of cross-country collaboration, knowledge transfer, and adaptive laboratory systems to address future pandemics and zoonotic threats.

Limitations

This study has several limitations. First, it was based on individuals who presented for testing or were referred to LAGET, which may not represent the general population. Second, detailed clinical outcomes were not available, limiting the ability to correlate viral load with severity. Third, the cross-sectional design precludes assessment of temporal dynamics or causal relationships. Finally, the reliance on centralized testing facilities may have introduced referral bias, especially for samples from external sites.

Conclusions

This study demonstrated a high prevalence of SARS-CoV-2 infection (31.5%) among individuals tested at LAGET between January 2021 and June 2022, far exceeding the national figures officially reported during the same period. The strong association between clinical symptoms and positivity, together with the increased odds of infection in samples collected at external sites, highlights persistent gaps in access to timely diagnosis and surveillance in Chad. These findings underscore the crucial role of molecular testing in capturing the true burden of COVID-19 and reveal the underestimation of community transmission.

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