DETECTION OF SARS-COV-2 IN MULTIPLE CLINICAL SPECIMENS: A PILOT PREVALENCE STUDY FROM NORTHEAST NIGERIA

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ABSTRACT

Background: **Diagnosis of SARS-CoV-2 infection primarily relies on RT-PCR analysis of nasopharyngeal swab (NPS) specimens; however, viral RNA has also been detected in other clinical samples. In Northeast Nigeria, data comparing SARS-CoV-2 detection across multiple specimen types, alongside serological evidence of exposure, remain limited.**

Aim: **This pilot study aimed to determine the prevalence of SARS-CoV-2 RNA in stool, serum, and NPS, and the seroprevalence of anti-SARS-CoV-2 antibodies among hospitalized patients in Gombe, Nigeria.**

Materials and Methods: **A cross-sectional study was conducted between March and May 2022 at Federal Teaching Hospital, Gombe. A total of 384 inpatients without prior confirmed COVID-19 were enrolled. Stool, serum, and NPS samples were collected from each participant. Viral RNA was detected using RT-PCR, and SARS-CoV-2–specific IgM and IgG antibodies were assessed separately using a rapid immunochromatographic assay, allowing differentiation between recent and past exposure**

Results: **Among 384 participants, the overall RT-PCR prevalence of SARS-CoV-2 RNA was 2.1% (8/384). Stool samples yielded the highest positivity rate (1.8%; 7/384), followed by NPS (0.8%; 3/384) and serum (0.3%; 1/384). In sharp contrast, the overall antibody seroprevalence was 48.6% (187/384). Among the 362 unvaccinated participants, 46.4% had detectable antibodies.**

Conclusions: **This study revealed a low prevalence of active viral shedding but a high seroprevalence of SARS-CoV-2 antibodies, suggesting widespread, under-detected community exposure. Stool specimens showed a higher detection rate than NPS in this cohort, warranting further investigation as a potential diagnostic sample in resource-limited settings.**

Keywords: SARS-CoV-2, Prevalence, Stool, Seroprevalence

INTRODUCTION

Coronavirus disease 2019 (COVID-19), caused by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2), was declared a pandemic on 11th March 2020 by the World Health Organization.[1] The disease, which emerged in Wuhan, China in December 2019[2,3], has since spread globally, with over 248 million cases and more than 5 million deaths reported by late 2021.[4] Although COVID-19 primarily affects the respiratory system, accumulating evidence indicates a much broader clinical spectrum, including gastrointestinal, cardiovascular, renal, and neurological involvement.[5,6] Viral RNA has been detected not only in respiratory samples but also in stool, blood, and urine, raising important implications for diagnosis and transmission dynamics.[7,8]

In Nigeria, the first confirmed COVID-19 case was reported in February 2020 in Lagos, followed by local transmission in Ogun State.[9] By November 2021, the country had reported more than 224,000 confirmed cases and approximately 3,000 deaths.[4] However, national statistics are likely to underestimate the true burden, given limited testing capacity and reliance on nasopharyngeal swabs as the primary diagnostic specimen. Molecular diagnosis using reverse transcriptase-polymerase chain reaction (RT-PCR) remains the reference standard[10], but access to reliable testing in Nigeria is constrained by cost, infrastructure, and trained personnel.[11] This has created diagnostic blind spots in many regions, especially in the Northeast, where health systems are under additional pressure due

to insecurity, limited resources, and weaker surveillance systems.[12,13]

This situation highlights two related research problems. First, there is a paucity of published studies from Northeast Nigeria evaluating SARS-CoV-2 prevalence using different specimen types. While international studies have shown that stool and serum may yield positive results, these findings have not been sufficiently validated in African populations.[6,14] Second, despite widespread reports of antibody positivity in sub-Saharan Africa, there is little integration of serological data with PCR results to understand the true extent of exposure in the region.[15] These gaps limit both the reliability of local diagnosis and the capacity to interpret regional patterns of transmission and immunity.

Addressing these gaps is crucial because standard reliance on nasopharyngeal swabs may underestimate cases, especially when viral shedding occurs in stool or blood.[8] In addition, low vaccine uptake in Nigeria has heightened the importance of serological data to understand population exposure and potential herd immunity. Without such evidence, both clinical management and public health planning are undermined.

The present study was therefore designed as a pilot prevalence survey to explore the detection of SARS-CoV-2 in stool, serum, and nasopharyngeal swabs among patients admitted at Federal Teaching Hospital, Gombe, Northeast Nigeria. By coupling molecular testing with serological analysis, this study contributes region-specific data where evidence has been sparse. Although modest in size, its findings help to clarify diagnostic patterns in a resource-limited setting, highlight the potential utility of non-respiratory specimens, and provide a basis for future large-scale surveillance in the region.

MATERIALS AND METHODS

This was a cross-sectional pilot study conducted at Federal Teaching Hospital (FTH), Gombe, a tertiary referral hospital in Gombe State, Northeast Nigeria. The study was carried out between March 30 and May 30, 2022. Adult Patients admitted to different wards of the hospital without a prior confirmed history of SARS-CoV-2 infection were recruited, regardless of COVID-19 vaccination status. Informed consent was obtained from all participants, and ethical clearance was granted by the Health Research Ethics Committee of the Federal Teaching Hospital Gombe (NHREC/25/10/2013).

Study population and sample size: A total of 384 patients were enrolled consecutively from medical, surgical and obstetric wards. Inclusion criteria were inpatients aged 18 years and above, admitted for conditions other than confirmed COVID-19, and willing to provide specimens. Patients with previously confirmed infection or those unwilling to participate were excluded. The sample size was determined using Fisher's formula[16]

$n = z^2pq/d^2$

Where n = number of sample size

z = z level of significance at 95% confidence interval (1.96)

p = prevalence rate at 50% (No similar study carried out in this environment)

q = 1-p

d = tolerable margin of error (5% = 0.05)

$n = z^2pq/d^2$

Therefore; $= (1.96)^2(0.5)\ (1-0.5)/0.05^2$

=384; Therefore, the minimum sample size is three hundred and eighty-four patients admitted in various wards of the hospital during the study.

Specimen collection and processing: Three types of clinical specimens were collected from each participant nasopharyngeal swabs, stool samples, and venous blood for serum separation. Specimen collection was performed under aseptic conditions by trained staff in line with WHO biosafety recommendations.[15] Samples were transported to the molecular laboratory of the Gombe State University under cold chain and processed promptly.

Laboratory analysis: RNA Extraction and RT-PCR: Viral RNA was extracted using a commercial extraction kit (QIAGEN QIAamp Viral RNA Mini Kit (250)). Reverse transcription polymerase chain reaction (RT-PCR) targeting SARS-CoV-2 genes (including S, E, N, and RdRp) was carried out. A cycle threshold (Ct) value <40 was considered positive, following standard guidelines. Using GeneFinder™ Covid-19 Plus RealAmp Kit.[7,17]

Serology: Serum samples were analyzed for SARS-CoV-2 specific IgM and IgG antibodies using an immunochromatographic rapid test kit, interpreted according to manufacturer instructions (Clarity Covid19 Rapid Test Cassettes).[15]

Data collection: Socio-demographic and clinical information, including age, sex, comorbidities, vaccination status, and presenting symptoms, were obtained using a structured interviewer-administered questionnaire.

Statistical analysis: Data were entered into Microsoft Excel and analyzed using SPSS version 29 (Chicago,

Illinois, USA). Frequencies and percentages were used to summarize categorical variables, while means with standard deviations or medians with interquartile ranges were calculated for continuous variables. Prevalence was reported as the proportion of positive cases among those tested. Fisher's exact test was used to explore associations between positivity and selected risk factors, given the small number of confirmed cases. A p-value <0.05 was considered statistically significant.

RESULTS

Demographic characteristics

A total of 384 patients were enrolled. The mean age was 36.4 years (±14.2), ranging from 18 to 72 years. The majority were male (54.3%). Participants were admitted from medical (58.1%), surgical (33.9%), pediatric (17.4%), and obstetric (12.9%) wards (Table 1).

Table 1: Baseline demographic and clinical characteristics of study participants (n = 384)

Variable	Frequency (n)	Percentage (%)
Age (years)		
Mean (±SD)	36.4 (±14.2)	–
Range	18 – 72	–
Sex		
Male	208	54.3
Female	176	45.7
Ward of admission		
Medical	223	58.1
Surgical	130	33.9
Obstetric	31	8.0

SARS-CoV-2 RNA was detected in a total of eight patients, giving an overall prevalence of 2.1%. Detection rates varied according to the type of specimen collected. Stool samples yielded the highest positivity rate, with seven out of 384 specimens (1.8%) testing positive. Nasopharyngeal swabs were positive in three cases (0.8%), while only one serum sample (0.3%) demonstrated detectable viral RNA (Table 2).

Table 2: SARS-CoV-2 positivity by specimen type

Specimen Type	Positive (n)	Prevalence (%)
Stool	7	1.8
Nasopharyngeal swab	3	0.8
Serum	1	0.3
Total unique cases	8	2.1

Antibody Responses

Serological testing revealed a much higher prevalence of antibodies compared with PCR positivity. A total of 187 patients (48.6%) tested positive for IgM and/or IgG antibodies. Among these, 94 (24.5%) were IgM positive only, 63 (16.4%) were IgG positive only, and 30 (7.8%) had both IgM and IgG detected (Table 3).

Table 3: Antibody prevalence among participants (n = 384)

Antibody Status	Frequency (n)	Percentage (%)
IgM positive only	94	24.5
IgG positive only	63	16.4
Both IgM and IgG positive	30	7.8
Any antibody positive	187	48.6
Antibody negative	197	51.4

Only 22 patients (5.7%) had received at least one dose of a COVID-19 vaccine. Of these, 19 demonstrated antibody positivity. Among the unvaccinated, 168 (46.0%) also had detectable antibodies, suggesting prior natural exposure (Table 4).

Table 4: Antibody positivity by vaccination status

Vaccination Status	Antibody Positive (n, %)	Antibody Negative (n, %)
Vaccinated (n=22)	19 (86.4%)	3 (13.6%)
Unvaccinated (n=362)	168 (46.4%)	194 (53.6%)
Total	187 (48.6%)	197 (51.4%)

Common symptoms among PCR-positive patients included fever (62.5%), cough (50.0%), and gastrointestinal symptoms such as diarrhea or abdominal discomfort (37.5%). Among antibody-positive patients, similar symptom patterns were noted, though many were asymptomatic (Table 5).

Table 5: Clinical Symptoms among PCR-Positive and Antibody-Positive Patients

Symptom	PCR-Positive (n=8)	Antibody-Positive (n=187)
Fever	5 (62.5%)	109 (58.3%)
Cough	4 (50.0%)	92 (49.2%)
Gastrointestinal symptoms	3 (37.5%)	68 (36.4%)
Dyspnea	2 (25.0%)	54 (28.9%)
Asymptomatic	1 (12.5%)	47 (25.1%)

Comorbid conditions were present in 29.4% of participants. Hypertension (14.1%), diabetes mellitus (7.6%), and HIV infection (4.9%) were the most common. Among PCR-positive patients, half had at least one comorbidity (Table 6).

An exploratory bivariate analysis was performed to assess associations between patient characteristics and PCR positivity (Table 7). No statistically significant relationships were observed between sex and SARS-

Table 6: Distribution of comorbidities among study participants (n = 384)

Comorbidity	n (%)
Hypertension	54 (14.1%)
Diabetes mellitus	29 (7.6%)
HIV infection	19 (4.9%)
Chronic kidney disease	10 (2.6%)
None reported	271 (70.6%)

CoV-2 detection (p=0.68) or between age group and positivity (p=0.74). Similarly, the presence of comorbidities such as hypertension, diabetes, or HIV infection was not significantly associated with PCR positivity (p=0.39).

Table 7: Association between selected characteristics and PCR positivity

Variable	PCR Positive (n=8)	PCR Negative (n=376)	p-value*
Sex			
Male	5 (2.4%)	203 (97.6%)	0.68
Female	3 (1.7%)	173 (98.3%)	
Age group			
≤40 years	6 (2.5%)	236 (97.5%)	0.74
>40 years	2 (1.6%)	122 (98.4%)	
Comorbidity			
Present	4 (3.5%)	108 (96.5%)	0.39
Absent	4 (1.5%)	268 (98.5%)	

The relationship between vaccination status and antibody positivity was also examined (Table 8). Antibody detection was significantly more common among vaccinated individuals (86.4%) compared with unvaccinated individuals (46.4%) (p<0.01). This suggests that vaccination contributed to seroconversion in this cohort, although a substantial proportion of unvaccinated participants also had detectable antibodies, likely reflecting natural exposure.

Table 8: Association between vaccination status and antibody positivity

Vaccination Status	Antibody Positive	Antibody Negative	p-value
Vaccinated (n=22)	19 (86.4%)	3 (13.6%)	<0.01
Unvaccinated (n=362)	168 (46.4%)	194 (53.6%)	

Discussion

This pilot prevalence study demonstrates that SARS-CoV-2 RNA can be detected in multiple clinical specimen types among hospitalized patients in Northeast Nigeria, with an overall RT-PCR positivity rate of 2.1%. Although the proportion of PCR-confirmed cases was small, the findings contribute to growing evidence that SARS-CoV-2 is not confined to the respiratory tract and may be detectable in non-respiratory specimens, particularly stool. In this cohort, stool samples yielded the highest detection rate compared with nasopharyngeal swabs and serum, supporting reports of gastrointestinal involvement in COVID-19 and potential fecal viral shedding.[7,8]

Several studies have demonstrated that the detection of SARS-CoV-2 RNA varies across specimen types due to differences in the temporal dynamics of viral shedding. Respiratory specimens, including nasopharyngeal swabs, typically demonstrate high viral loads early in infection, followed by relatively rapid clearance. In contrast, viral RNA may persist in stool for prolonged periods, even after respiratory samples become negative[818–20]. This prolonged gastrointestinal shedding has been attributed to viral replication in enterocytes and may explain why stool samples demonstrated a higher detection rate than nasopharyngeal swabs in this cross-sectional study.

Detection of SARS-CoV-2 RNA in serum was rare in the present study, which is consistent with previous reports describing viremia as an infrequent and often transient phenomenon.[14,21] Viral RNA in blood has been reported predominantly in the early phase of infection or in patients with more severe disease, and its presence does not necessarily correlate with infectivity.[21,22] The low frequency of serum positivity observed in this cohort therefore aligns with existing evidence and underscores the limited diagnostic utility of blood-based molecular testing in most clinical settings.

Differences in the timing of appearance and clearance of viral RNA across specimen types may have influenced the specimen-specific positivity rates observed in this study. Because samples were collected at a single time point, it was not possible to determine the stage of infection at which each participant was sampled. Patients presenting later in the disease course may have already cleared viral RNA from the upper respiratory tract while continuing to shed viral RNA in stool, whereas early infections might be more readily detected by nasopharyngeal swabs.[14] This temporal variability highlights an important limitation of cross-sectional studies and underscores the need for longitudinal sampling to better define the order of appearance and disappearance of SARS-CoV-2 RNA across different specimen types.

This phenomenon is conceptually analogous to other infectious diseases, such as *Salmonella enterica* serovar Typhi, in which the diagnostic yield of blood, stool, and urine specimens varies depending on the timing of specimen collection relative to disease onset. Similarly, understanding the kinetics of SARS-CoV-2 shedding across multiple specimen types is essential

for optimizing diagnostic strategies, particularly in resource-limited settings where access to nasopharyngeal swab testing may be constrained.

A notable finding of this study was the high seroprevalence of SARS-CoV-2 antibodies, with nearly half of participants demonstrating IgM and/or IgG positivity despite low PCR detection rates. This discrepancy mirrors observations from other African settings, where antibody prevalence has consistently exceeded PCR-confirmed case numbers, suggesting widespread prior exposure and under-ascertainment of infection.[15,23] The detection of antibodies among unvaccinated individuals further supports the contribution of natural infection to population-level immunity in Nigeria, where vaccine uptake has remained relatively low.

From a public health perspective, these findings emphasize the value of integrating serological surveillance with molecular testing to better understand the true burden of SARS-CoV-2 infection in low-resource settings. While nasopharyngeal swabs remain the diagnostic standard, the detection of viral RNA in stool highlights the potential utility of alternative specimens for epidemiologic surveillance and research, particularly where logistical or biosafety challenges limit respiratory sampling.

A striking finding was the high seroprevalence of antibodies, with nearly half of participants demonstrating IgM and/or IgG positivity.

The high antibody seroprevalence observed despite low PCR positivity may also be partly explained by the substantial burden of asymptomatic and mildly symptomatic SARS-CoV-2 infection reported in African populations. Several studies have shown that a large proportion of SARS-CoV-2 infections in sub-Saharan Africa occur without overt clinical symptoms, reducing the likelihood of health-seeking behaviour and subsequent molecular testing.[24–26] As a result, many infections may go undetected by routine PCR-based surveillance while still eliciting measurable antibody responses.

Younger population age structures, lower prevalence of comorbidities, and possible differences in prior immune conditioning have been proposed as contributing factors to the high frequency of asymptomatic infection observed in African settings.[24]

This pattern likely contributed to the discrepancy between PCR-confirmed infection and antibody positivity in the present study, particularly among unvaccinated individuals, and underscores the limitations of symptom-driven testing strategies for estimating true community exposure.

The lack of strong statistical associations between PCR positivity and demographic or clinical variables likely reflects the small number of confirmed cases. Nevertheless, exploratory analysis suggested that vaccination was significantly associated with antibody positivity, reinforcing the role of immunization in boosting seroconversion. At the same time, the large number of antibody-positive unvaccinated patients underscores the ongoing contribution of natural infection to community-level immunity in Nigeria, where vaccine uptake remains low.[11]

From a public health perspective, these findings emphasize the potential role of serological surveillance as a complement to molecular testing in low-resource environments. They also suggest that stool specimens could be explored further as a diagnostic alternative where nasopharyngeal swab testing is impractical. Importantly, the combination of low PCR positivity and high antibody prevalence raises the possibility of under-detection of COVID-19 in hospital populations, which may have implications for infection control policies.

Limitations

Among the limitation of this study, is the small number of PCR-positive cases limited the ability to perform robust statistical analyses or to establish firm associations between risk factors and outcomes. Being a single-centered hospital-based study, the findings may not be generalizable to the broader community. In addition, reliance on a rapid antibody assay may have underestimated or overestimated true seroprevalence due to sensitivity and specificity limitations.

CONCLUSION

Despite these limitations, this study provides novel pilot data from Northeast Nigeria on the prevalence of SARS-CoV-2 across multiple specimen types and highlights the discrepancy between low PCR detection and high antibody positivity. These findings add to the understanding of COVID-19 epidemiology in resource-limited African settings and underscore the value of integrating serological testing into surveillance strategies. Larger multicentered studies are needed to validate these observations and to better define the diagnostic utility of stool and serum specimens.

Conflict of interest statement

The author, Umar Mohammed Hassan, declares that there is no conflict of interest regarding the research, authorship, and/or publication of this article. No financial, institutional, or personal relationships exist

that could have influenced the work reported in this manuscript.

Authors' contributions

Conceptualization: UMH, AAY, MMM
Study design: UMH, AAY, MMM
Data acquisition: UMH, UMI, IM, FBA, RAM
Laboratory analysis: FBA, RAM
Data analysis and interpretation: UMH, AH
Literature review: UMI
Manuscript drafting: UMH, AH, FBA
Critical revision of the manuscript for important intellectual content: AAY, AH, UMI, IM, MMM
Supervision: AAY, MMM
Final approval of the manuscript: All authors

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