Advances in Infectious Diseases, 2022, 12, 799-812
https://www.scirp.org/journal/aid
ISSN Online: 2164-2656
ISSN Print: 2164-2648

Outcome of Symptom-Based RT-PCR Testing for SARS-CoV-2: Experience from a Large Public Testing Centre in Nigeria

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How to cite this paper: Ohihoin, A.G., Osuolale, K.A., Okwuraiwe, P.A., Musa, A.Z., James, A.B., Onwuamah, C.K., Amoo, O.S., Shaibu, J.O., Ige, F., Ohihoin, E.N., Chukwu, E., Otuonye, N.M., Bamidele, T., Olukosi, A., Ajibaye, O., Raheem, Y., Oladele, D., David, A.N., Adewale, B., Audu, R. and Salako, R.L. (2022) Outcome of Symptom-Based RT-PCR Testing for SARS-CoV-2: Experience from a Large Public Testing Centre in Nigeria. *Advances in Infectious Diseases*, **12**, 799-812.
https://doi.org/10.4236/aid.2022.124057

Received: March 26, 2022
Accepted: December 6, 2022
Published: December 9, 2022

Abstract

Background: Access to testing for SARS-CoV-2 in Nigeria is still highly restricted. Only patients who are symptomatic for SARS-CoV-2 are selected for testing. This pattern of testing will miss a large proportion of individuals with the infection who are asymptomatic and presymptomatic. This study reports the experience of a symptom-based study from a large testing centre in Nigeria. **Methods:** A cross-sectional study, reviewing data collected from respondents presenting at the Nigerian Institute of Medical Research (NIMR) modified drive-through center for COVID-19 test between the period March 31st and August 31st, 2020 were included in the analysis. **Results:** A total of 9891 participants were recruited into the study and 2465 participants (24.9%) had a positive PCR result for SARS-CoV-2. The majority of the respondents were above 18 years old, n = 9163 (93.4%). The average age of the respondents was 36.7 years (sd 13.8 years). The age of the participants has a significant effect on SARS-CoV-2 status (AOR = 1.009, CI, 1.005 to 1.012, $p = 0.0001$). There was a male preponderance, n = 5652 (57.6%). The odds of having a positive status for SARS-CoV-2 were 0.9 times lower for female participants. The majority of the respondents had a history of travel, n = 6788 (68.6%). Cough was the commonest symptom, n = 1062 (10.7%) followed by fever, n = 979 (9.9%). The mortality rate was 0.1%. Among the participants with comorbidity, 31 (51.7%) had positive results for SARS-CoV-2 while 29 (48.3%) had negative results. **Conclusion:** Symptom-based approach to testing for SARS-CoV-2 in Nigeria revealed a high positivity rate, while mortality from those tested in the study is low. Age and gender appear to play a significant role in the dynamics of the disease.

Keywords

SARS-CoV-2, COVID-19, Symptoms, RT-PCR, Nigeria

1. Introduction

One of the identified cornerstones for the control of SARS-CoV-2 infection has been upscaling testing for efficient case identification and isolation of infected individuals [1]. This measure alongside other public health measures such as hand hygiene, use of face masks, and physical distancing has been the major intervention promoted by the World Health Organization for the control of the COVID-19 pandemic [2]. Although the recent vaccine breakthrough has offered renewed hope towards stemming the tide of the pandemic, there are concerns that acceptability and logistics involved with the widespread distribution of the vaccine could constitute a challenge to harnessing the full potential of the vaccines [3] [4] [5]. It is therefore imperative to maintain established public health measures of active case search via testing and isolation of cases found to be positive for the virus.

Active case searching depends largely on rapid diagnosis via scaling up testing opportunities. Although access to testing has consistently been on the rise globally, there is still a huge gap in this regard as a large proportion of individuals in resource-constrained settings do not have access to testing facilities [6] [7]. Testing for SARS-CoV-2 in these parts of the world is still restricted to those who satisfy certain eligibility criteria as only patients who are symptomatic for SARS-CoV-2 are selected for testing. This pattern of testing will miss a large proportion of individuals with the infection who are asymptomatic and presymptomatic as there is evidence to suggest that as much as 30% of individuals with SARS-CoV-2 are asymptomatic [8] [9] [10].

Asymptomatic and presymptomatic individuals with SARS-CoV-2 infection who are not tested and hence not identified and isolated will continually be a reservoir for the spread of the virus thus making efforts to control the virus a big challenge. Furthermore, the disease dynamics in Nigeria and many parts of sub-Saharan Africa have taken a different trajectory from that observed in Europe and America [3] [5] [11]. Morbidity and mortality figures appear to be lower than that experienced in the Northern hemisphere, although specific data to corroborate this speculation is scarce. The Nigerian Institute of Medical Research, an agency of the Federal Ministry of health-based in Lagos is one of the pioneer public health sites for testing for COVID-19. The Institute makes use of symptom-based testing as recommended by the Nation's Centre for Disease Control (NCDC) [12]. Symptom-based testing involves testing only individuals who are symptomatic for the disease or with a significant history of exposure. The Institute was the first to initiate a modified drive-through sampling technique in the country to facilitate symptom-based testing for COVID-19 [1]. The rationale for symptom-based testing is to ensure the efficient use of scarce resources for those with the greatest need. The challenge, however, is that it does not reflect the epidemiology of the disease in the general population, particularly for a disease that a significant section of those affected is either asymptomatic or presymptomatic [6] [13]. This scenario implies that it will be difficult to track the affected patients thus compromising active case searching and contact tracing.

This study reviewed the outcome of 10,000 participants who met NCDC criteria for testing and presented to the modified drive-through sampling centre to test for SARS-CoV-2 at NIMR, a large public health testing centre in Lagos, Nigeria.

2. Methods

2.1. Study Design

A cross sectional study, reviewing data collected from respondents presenting at the NIMR modified drive-through centre for the COVID-19 test. The institute made use of predesigned Case Report Forms (CRF) from NCDC used to collect information from respondents presenting at COVID-19 testing sites as the data collection tool at the modified drive-through centre. These data were abstracted from the NIMR data based where the data collected were entered, validated, and, cleaned for statistical analyses. Consecutive consenting participants who presented for testing at the study centre and satisfied the inclusion criteria were recruited for analysis. The duration was between March 31st and August 31st, 2020.

2.2. Study Setting

NIMR, the apex medical research institute in the country, is an agency of the Federal Ministry of Health (FMOH) with the mandate to conduct research into diseases of public health importance. NIMR is at the forefront of the COVID-19 disease response in the country as one of the designated centres for SARS-CoV-2 testing and thus has access to the study population. NIMR is located in Lagos Mainland and easily accessible from all parts of the state. Sample collection was done via the institute's drive-through centre while laboratory analysis was done at the institute's Centre for Human Virology and Genomics (CHVG). CHVG is a recognized WHO prequalification evaluation laboratory for the validation of molecular and serological test kits [2].

2.3. Study Population

The study population consisted of participants who presented to the drive-through sample collection centre of the institute. Information on the availability of testing at the centre was disseminated through the website and other social media platforms inviting persons with suspicion of COVID-19 infection to register. An online form was completed and the study team reviewed the eligibility of the participants from the back end. Text messages or emails were sent to eligible persons inviting them for the test. The messages had a uniquely coded identifier [example-NCV-00111] with a bar code stating the location of the centre, the test date, and the time. Those who did not meet eligibility criteria were given feedback that they were not eligible. Eligibility was based on some epidemiological parameters such as a history of travel, and on symptomatology such as the presence of fever, cough, difficulty with breathing, gastrointestinal, and other constitutional symptoms linked to acute respiratory disease. After the collection of surveillance and laboratory data, eligible participants were given their sample collection kit and directed to the sample collection station. At this station, both Naso- and Oropharyngeal samples were collected.

2.4. Data Collection Tool

A structured questionnaire used in the country at COVID-19 testing sites adapted from WHO by NCDC was used. The tool had sections on demographics, COVID-19 symptoms, sample collection details, clinical complications, exposure in the last 14 days, and travel history. Once collected, this information was entered into an electronic web-based platform called Research Electronic Data Capture (REDCap).

3. Laboratory Testing of SARS-CoV-2

3.1. Sample Collection

Personal protective equipment (PPE) was completely donned before collecting samples from each suspected person. Oropharyngeal (OP) and nasopharyngeal (NP) swabs were collected. The swab samples were collected by inserting swabs to the posterior end of the nasal and throat regions respectively, swirling the area for 10 seconds, and immediately immersing swabs into the same tube of 2 ml of Viral Transport Medium (VTM). All samples were transported in a cold chain (2˚C - 8˚C) to the CHVG for laboratory analysis.

3.2. Testing of Samples

Viral RNA was manually extracted from oral and nasal swabs using the QIAamp viral RNA Mini Kit (Qiagen, Hilden, Germany), under a biosafety level 2 laminar flow cabinet. The viral RNA was subsequently amplified and detected using the Quant Studio 3 real-time PCR (Thermo Fisher Scientific Inc, USA) and the BGI real-time fluorescent RT-PCR kit (BGI, Shenzhen, China) according to the manufacturer's instructions. The BGI assay detects the SARS-CoV-2 open reading frame 1 (ORF1) region, with an internal control detecting a human housekeeping gene [9]. The internal control verifies that sampling was collected properly and checks for false negatives or inhibition. Assay validation included ensuring curves are S-shaped, no cycle threshold (CT) values for the negative control, and both targets detected for the positive control with CT ≤ 32. All samples tested with internal control detected at CT ≤ 32 were accepted as valid. The assay has a limit of detection of 100 RNA copies/ml[9].

3.3. Data Management and Analysis

Data was validated and cleaned, abstracted from REDCap, and exported to SPSS version 26.0 for statistical analysis. Descriptive statistics was used to summarize the data using counts, percentages, and means. Inferential statistics was done to determine the predictors of SARS-CoV-2 positivity using the logistic regression model and its odd ratios for interpretation at 95% Confidence Interval which were statistically significant at $p < 0.05$. The χ^2 test using **Omnibus tests of Model Coefficient for SARS-CoV-2 Status** was done with a two-sided α level of 0·05. The main interest is the SARS-CoV-2 status to determine which of the explanatory variables are predictive of the response, positive or negative to SARS-CoV-2. The response variable is the final RT-PCR result of SARS-CoV-2 which is binary and therefore a logistic regression approach was used. For a binary response taking the values 0 and 1 (negative and positive), the expected value is simply the probability, p, that the variable takes the value one, *i.e.*, the probability of positive status. This p is modelled indirectly via logit transformation of p, *i.e.*, $\ln[p/(1-p)]$ and this leads to the logistic regression model employed in this study. The estimated regression coefficients in this model gave the estimated change in the log-odds corresponding to a unit change in the corresponding explanatory variable conditional on the other explanatory variables remaining constant. We selected variables that were representative of different potential modes of SARS-CoV-2 transmission for the multiple logistic regression analysis and included variables such as gender, age, history of exposure and occupation. The main interest here about the SARS-CoV-2 status is to determine which of the explanatory variables are predictive of the response, positive or negative to SARS-CoV-2.

4. Results

A total of 9891 participants were involved in the study and, 2465 participants (24.9%) had a positive PCR result for SARS-CoV-2. The majority of respondents were above 18 years old, n = 9163 (93.4%). The average age of the respondents was 36.7 years (sd 13.8 years). The age of the participants has a significant effect on SARS-CoV-2 status (AOR = 1.009, CI, 1.005 to 1.012, $p = 0.0001$). There was a male preponderance, n = 5652 (57.6%). The odds of having a positive status for SARS-CoV-2 were 0.9 times lower for female participants. The majority of the respondents had a history of travel, n = 6788 (68.6%). The mortality rate was 0.1%. Among the participants with comorbidity, 31 (51.7%) had positive results for SARS-CoV-2 while 29 (48.3%) had negative results.

In **Table 1**, the male gender accounted for the majority of participants (57.6%) while female participants accounted for 42.4%. The mean age of participants was 36.7 ±13.8 years. **Table 2** revealed that most of the patients resided in Africa (63.2%, n = 6247), with both international and domestic travel histories. In **Table 3**, fever was the commonest symptom as seen in 43% of study participants (n—421). In **Table 4**, the odds of having a positive status for SARS-CoV-2 was 0.9 times lower for female participants (CI, 0.79 to 0.97). The age of the participants has a significant effect on SARS-CoV-2 status (AOR = 1.009, CI, 1.005 to 1.012, $p < 0.01$). History of exposure (for participants with no exposure) does not contribute significantly to explaining SARS-CoV-2 probabilities once the other variables are accounted for (AOR = 1.012, CI, 0.896 to 1.144, $p = 0.843$) unlike participants that had a history of exposure in the reference group (AOR = 1.480, CI, 1.281 to 1.710, $p < 0.01$). History of exposure (for participants with unknown exposure) does contribute significantly to explaining SARS-CoV-2 probabilities once the other variables are accounted for (AOR = 1.424, CI, 1.251 to 1.261, $p < 0.01$) like participants that had a history of exposure in the reference group. Occupation of the participants also has no significant effect on SARS-CoV-2 status (AOR = 0.970, CI, 0.679 to 1.387, $p = 0.868$). SARS-CoV-2

Table 1. Socio-demographic characteristics of respondents.

Variables	Frequency (%)
Age group	
0 - 9	390 (4.0)
10 - 17	260 (2.6)
18 and above	9163 (93.4)
Total	**9813**
Mean Age: 36.7 ± 13.8	
Gender	
Female	4161 (42.4)
Male	5652 (57.6)
Occupation	
Employed	3866 (39.1)
Housewife	44 (0.4)
Student	37 (0.4)
Unemployed	170 (1.7)

Table 2. Travel history.

Variables	Frequency (%)	Variable	Frequency (%)
Country of residence by Continents	n (%)	Exposure to the person with similar illness	n (%)
Africa	6247 (63.2)	Yes	2205 (22.3)
America	1(0.01)	Unknown	1712 (17.3)
Asia	2 (0.02)	No	2840 (28.7)
Europe	7 (0.1)		

Table 3. Symptomatology.

Clinical Symptoms	Total n (%)	Positive n (%)
Fever	979 (9.9)	421 (43)
Sore Throat	742 (7.5)	245 (33)
Runny nose	576 (5.8)	216 (37.5)
Cough	1062 (10.7)	443 (41.7)
Shortness of Breath	509 (5.1)	171 (33.6)
Vomiting	110 (1.1)	58 (52.7)
Nausea	198 (2.0)	87 (43.9)
Diarrhea	253 (2.6)	91 (36)

Table 4. The results of factors found to be independently associated with a positive status for SARS-CoV-2 after controlling for potential confounding variables.

Variable	Coefficient	AOR	95% C1	*p*-value	χ^2 test
Gender (1)	−0.133	0.876	0.794 - 0.966	<0.01	168.6; $p < 0.01$
Age (in years)	0.009	1.009	1.005 - 1.012	<0.01	
History of exposure (No exposure)	0.012	1.012	0.896 - 1.144	0.843	
History of exposure (unknown exposure)	0.353	1.424	1.251 - 1.621	<0.01	
Employed	−0.030	0.970	0.679 - 1.387	0.868	
Housewife	0.014	1.014	0.708 - 1.451	0.941	
Student	0.285	1.330	0.643 - 2.753	0.442	
Unemployed	0.269	1.309	0.587 - 2.916	0.510	

Status is significantly related to the explanatory variables: Gender, Age, and History of Exposure (Score test: $\chi^2(2) = 168.6$, $p < 0.01$).

Figure 1 and Figure 2 showed the responses to both domestic and international travel histories. Figure 1 showed that 97% (6785) of the respondents presented at the NIMR modified drive-through centre for COVID-19 test had international travel history, 1% (98) had no travel history while information on travel history

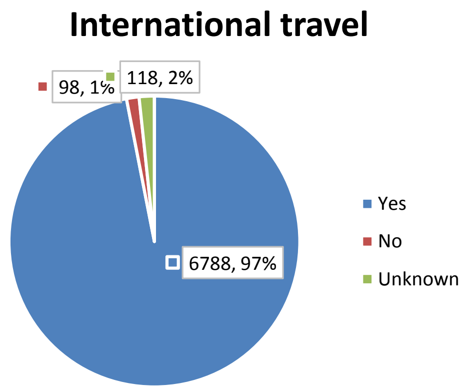

Figure 1. International travel history.

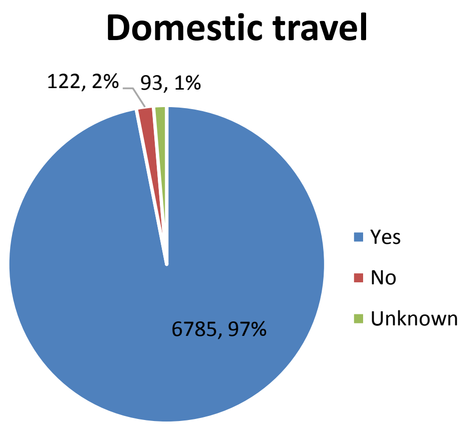

Figure 2. Domestic travel history.

was unknown for 2% (118) of the respondents. Figure 2 showed that 97% (6788) of the respondents had domestic travel history, 2% (122) had no travel history and information on travel history was unknown for only 1% (93) of the respondents. Figure 3 showed various symptoms reported by the respondents with cough (1062) having the highest, followed by fever (979) and vomiting (110) having the least.

The results presented in Figure 4 showed that among the positive participants, 99.9% were alive while 0.1% of the participants died. In Figure 5, among the participants with co-morbidity, 31 (51.7%) had positive results for SARS-CoV-2 while 29 (48.3%) of the participants had negative results. In Figure 6, 32.32% of the participants with positive results visited the hospital (n = .2381)

The prevalence in the cohort of the respondents is as given below:

$$\text{Prevalence of COVID-19 Infection}$$
$$= \frac{\text{number of participants infected}}{\text{total number of participants}} \times 100\%$$
$$= \frac{2465}{9891} \times 100\%$$
$$= 24.9\%$$

Out of 9891 participants, the overall prevalence of COVID-19 infection is 24.9%.

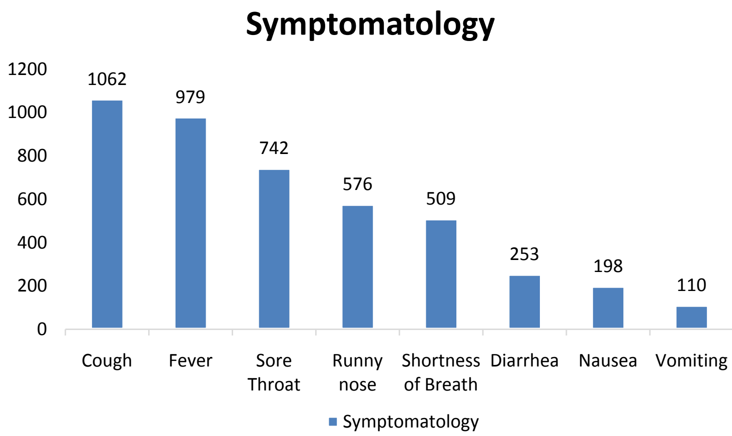

Figure 3. Symptoms reported by various respondents.

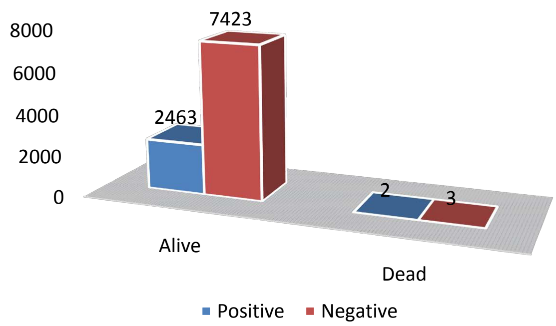

Figure 4. SARS-CoV-2 results with status alive or dead.

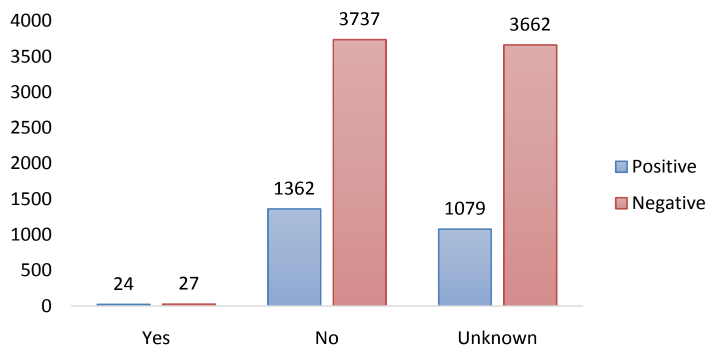

Figure 5. SARS-CoV-2 results of respondents with comorbidities.

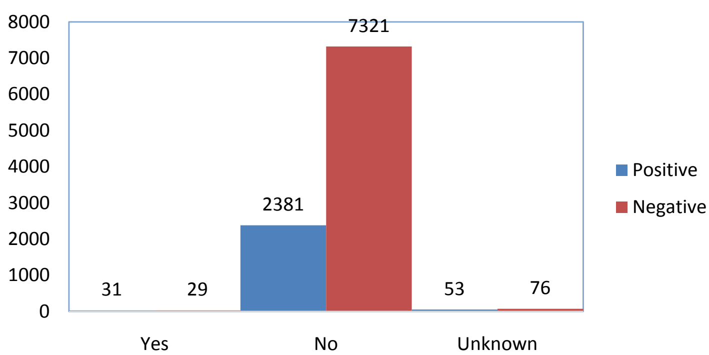

Figure 6. SARS-CoV-2 positive respondents that visited the hospital.

5. Discussion

The study reviewed the outcome of the SARS-CoV-2 RT-PCR test conducted for 9891 participants at the Nigerian Institute of Medical Research. A total of 2465 participants had a positive result for SARS-CoV-2 constituting 24.9% of the participants. This rate of 24.9% should be viewed against the background of the eligibility criteria used for the selection of the participants by selecting only participants with symptoms or clear risk factors for being positive for SARS-CoV-2.

The positivity rate for SARS-CoV-2 is influenced by various factors that include the timing of the testing in the pandemic, eligibility criteria for testing, and access to testing services [6] [14]. Positivity rate from the published literature ranges from 8.2% to 32% [8] [9] [15]. Restricting testing for SARS-CoV-2 to individuals with clear symptoms and obvious risk factors could lead to a missed opportunity for asymptomatic and presymptomatic individuals who have contracted the virus. Furthermore, testing performed during the peak of the pandemic in any region is expected to be associated with a high rate of positivity. This study was conducted during the peak of the pandemic in Nigeria. During the initial stage of the pandemic, NIMR was one of four laboratories with the capacity to conduct a molecular test for SARS-CoV-2 and as such received the bulk of the samples being tested for SARS-CoV-2. This could also account for the relatively high rate of positivity seen in this study. It is refreshing to know that currently, the number of laboratories in the country with the capacity to perform a molecular test for SARS-CoV-2 stands at seventy-eight. The majority of the respondents were above 18 years old, n = 9163 (93.4%). The average age of the respondents was 36.7 years (sd 13.8 years). The role of age on the transmission and dynamics of the disease has been the subject of widespread discussion and research. There is evidence to suggest that the severity of SARS-CoV-2 tends to be more in the elderly [16]. The risk of a positive status for SARS-CoV-2 tends to increase with advancing age [17]. There is evidence to suggest that children and adolescents demonstrate a lower risk of becoming positive for SARS-CoV-2 when compared to the adult population [11] [14] [18]. This study also corroborates other studies because a large proportion of the participants fell into the adult age range. Only 6.6% of the respondents fell below the adult age range. Recognizing that the younger age group demonstrates less tendency to contracting the disease could be a piece of useful public health information in identifying the role of younger people in disease transmission and disease dynamics. This will be useful in instituting guidelines for prioritizing the closure and opening of public places during the pandemic. There was a preponderance of male respondents among the participants. n = 5652 (57.6%). The odds of having a positive status for SARS-CoV-2 were 0.9 times lower for female participants. Gender difference in susceptibility to SARS-CoV-2 has been interrogated by some researchers. The available evidence tends to suggest a male predisposition to SARS-CoV-2 [4] [19] [20]. Some studies have also suggested that severity appears to be more in males when compared to females [4] [10]. Our study however did not interrogate the issue of gender bias as it relates to severity. In this study, we also established a male susceptibility to SARS-CoV-2 when compared to females (n = 5652 for M, and n = 4161 for F). The reason for increased male susceptibility has been postulated to include the tendency for males to be involved with activities that take them from outside the confines of the home, a greater predisposition to being more adventurous [10]. Other workers have however debunked this theory and postulated that genetic differences in the way male and female individuals respond to the virus could actually explain the increased predisposition of males to the disease [14] [16]. A significant proportion of the respondents had a history of travel n = 6788 (68.6%). The majority of those that travelled made trips to countries within the continent n = 6242 (63.2%). The SARS-CoV-2 transmission has been facilitated by the movement of people across borders. The index case in Nigeria came from a visiting European national [6]. This study revealed that about two-thirds of those who presented to the centre volunteered a history of travel. 22.3% (n = 2205) of the respondents had a history of exposure to people with similar symptoms. Patients with fever had the highest percentage of a positive SARS-CoV-2 status (43%), followed by those with cough (41.7%). History of exposure to people with similar symptoms is one of the eligibility criteria of the nation's CDC used to select members of the public for testing [12]. COVID-19 symptomatology is highly heterogeneous and bears similarity to symptoms of other relatively common diseases such as malaria fever and other common viral illnesses [21]. Fever and cough seem to be the most consistent symptoms across the board. This is not surprising as COVID-19 being basically a viral respiratory illness is expected to create an inflammatory response within the respiratory system hence manifesting as fever and cough. It is worthy of note that participants with fever tend to come out with a positive result for SARS-CoV-2 when compared to other symptoms. This finding supports the rationale for temperature checks in public places. There are genuine concerns about the use of a symptom-based approach for screening because a significant proportion of individuals with a positive outcome for SARS-CoV-2 are asymptomatic thus compromising the use of a symptom-based approach as an effective tool for active case identification [18] [22] [23].

The mortality rate among the participants in the study was 0.1%. This finding should however be viewed against the background that most of the participants in this study were ambulant and not severely ill. This finding does not necessarily reflect the mortality picture for SARS-CoV-2 in the general population. The mortality picture in Nigeria however appears to be relatively low when compared to findings from other parts of Europe and America [6]. This finding underlines the need to have a better understanding of the disease dynamics and prognosis in this part of the world.

Among the participants with comorbidity, 31 (51.7%) had positive results for SARS-CoV-2 while 29 (48.3%) of the participants had negative results. This outcome tends to suggest that individuals with co-morbidity are at higher risk of being positive for SARS-CoV-2. This may however not be the case as those individuals with co-morbidity may have a lower threshold to present for testing because SARS-CoV-2 generally runs a more aggressive course in patients with co-morbidities as mortality is known to be higher in positive patients with co-morbidities [7] [19] [24].

6. Conclusion

In conclusion, this study demonstrated that a symptom-based approach to testing for SARS-CoV-2 in Nigeria revealed a high positivity rate, while mortality from those tested in the study is low. A limitation of this study is that this pattern of symptom-based testing does not lead to a better understanding of the role of presymptomatic and asymptomatic persons in disease dynamics. The study only observes a snap-shot of the situation at best. A large-scale epidemiological study within the community is needed to understand the disease dynamics at the community level. This information is urgently needed to understand the trajectory of this pandemic in the most populous nation in Africa.

Acknowledgements

I wish to acknowledge Mercy Mayowa Ojetunde for her secretariat input.

Ethical Approval and Consent to Participate

Ethical approval was obtained for the conduct of this study from the Institutional Review Board (IRB) of the Nigerian Institute of Medical Research (NIMR). Written informed consent was obtained from the participants.

Consent for Publishing

All authors gave their consent for publishing.

Availability of Supporting Data

Supporting data are available on request.

Funding

The project is an institutional project of the Nigerian Institute of Medical Research (NIMR), an agency of the Federal Ministry of Health of the federal government of Nigeria. The federal government of Nigeria funds the ministry of health and research institutions to respond to the COVID-19 pandemic. Hence, there was no special funding for this project.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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