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Research Article

Public Health Laboratories Response to SARS-COV-2 Diagnostic Testing during COVID Pandemic in Pakistan

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Abstract: During COVID-19 Pandemic, diagnostic laboratories played a vital role in outbreak investigation, surveillance, patient monitoring, and therapeutic effectiveness, and hampered the transmission cycle globally. In Pakistan, the health department took an initiative to build BSL-III labs at divisional levels. A qualitative study was conducted among healthcare professionals from 13 major public health national-level laboratories through in-depth interviews with key informants to note down the challenges they faced during the COVID-19 pandemic during diagnostic testing. In this study, 77 % of public health laboratories faced sampling, administrative, and leadership issues. 53 % of laboratories have faced the unavailability of well-trained staff and human resources while both the biosafety and biosecurity protocols, and the lack of resources were compromised in 69 % of labs. Some lab staff (54 %) felt the wastage of resources in terms of excessive testing and fake sampling, while others (54 %) discussed a lack of training and work experience issues. As the majority of the technical lab staff was hired in temporary consultancy mode so 61 % of issues were related to late salaries. 38 % of issues were about fake reporting pressure from higher authorities. 69 % had issues with the continuous supply chain of kits, reagents, PPEs, etc. The work environment was not up to the mark of 69 %. High workload and mental health issues were faced by 92 %, while waste management was 23 %, shortage of lab space for massive testing by 38 %, and stigma and discrimination among healthcare workers and the general public due to involvement in COVID-19 testing were felt by 46 %.

Keywords: COVID-19, Pakistan, Healthcare system strengthening, Challenges of outbreak, Pandemic, Lesson Learned

1. INTRODUCTION

Human health remains at risk worldwide due to emerging infectious diseases. Scientists and researchers are always on the verge of war by coping with the multiple outbreaks of emerging infectious pathogens and their multiple serotypes like influenza, corona, dengue, chikungunya, cholera, tuberculosis, and many others right from the beginning of this world. In recent years, mortality rate percentages are declining due to good public health strategies, but still many new infectious diseases have been identified and registered, including Hantavirus pulmonary syndrome, AIDS, Ebola and Legionnaire virus [1].

The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) caused a recent novel coronavirus disease outbreak that emerged in Wuhan China and became a pandemic in no time. Due to health emergencies, several national health organizations faced public health crisis which was threatening to world. A total of 4820714 infected cases were reported on the 18th of May, and the mortality rate was more than 316998 (7 %) [2]. In the last 2 decades, the world has faced 3 outbreaks of coronavirus; SARS-CoV-1 in 2003, MERS-CoV in 2012, and SARS-CoV-2 pandemic in 2019. To fight against such outbreaks and pandemics, the most practical and sensible approach is a robust response in form of early diagnosis and preventive strategies. For this purpose, better advanced diagnostic

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laboratories and effective surveillance programs are needed of the hour. Diagnostic laboratories play a pivotal role in outbreaks investigations through rapid detection of virus infection, rapid serological assays, and advanced molecular diagnostic techniques [3, 4]. These laboratories reported quick results and played a very important role in concluding the outbreak.

It is very important to break the transmission chain of COVID-19 in the population in the first place by diagnosis, reducing the number of suspects, and reducing the basic reproductive number. The viral transmission is decreased by controlling various factors such as COVID-19-positive individuals, infection severity, and viral shedding [5]. Due to compromised facilities of vaccination and treatment, the only possible method to decrease transmission of COVID-19 is the isolation of infected individuals to prevent transmission of the disease. Diagnostic testing for COVID-19 has been varying differently around the globe. The COVID-19 pandemic outbreak exposed some major vulnerabilities, limitations, and gaps in the healthcare departments of many countries.

Few developed countries in Asia showed prompt responses to control the COVID-19 pandemic. Singapore performed a wide screening program on patients with influenza, pneumonia, patients in ICU, and deaths with possible infection [6]. South Korea controlled the COVID-19 outbreak through extraordinary efforts of national testing and managed to perform 3 lac tests in the first 9 weeks after they diagnosed the first case of COVID-19 [7]. Hong Kong and Taiwan did the same job [8]. They implemented resource-intensive strategies that promoted diagnostic testing and isolation strategies to prevent transmission [8]. In the pressure of rapid transmission of COVID-19 across borders, the diagnostic testing was dependent on the type of test available, resources, and time of test results. Because the suspected cases were on high priority to assign them isolation to prevent spread in the community. Different diagnostic tests for COVID-19 are available and many more getting approval every day [9, 10]. Different diagnostic tests were available in Pakistan to cope with COVID-19 diagnosis such as, R-T PCR, LAMP, Lateral flow and ELISA. To fight with pandemic public health labs have been in deep need of reliable, accurate and fast testing for COVID-19 as given in Table 1. Graphical representation of the total number of tests performed by public sector laboratories of Pakistan shown in figure 1.

During the COVID pandemic, all molecular testing labs have had multiple diagnostic challenges worldwide [11]. The diagnostic labs face challenges when not only their healthcare workers are required

Table 1. Public Sector Laboratories and number of COVID-19 tests performed by laboratories during the COVID-19 pandemic in Pakistan (Reference: COVID-19 PORTAL HISDU Primary and Secondary Healthcare Department P&SHD)

S. No.	Public Sector Labs of Punjab Province Pakistan	Location	Covid-19 Tests Performed
1	Provincial Public Health Reference Laboratory, Lahore, Punjab, Pakistan	Lahore	3800000
2	National Institute of Health Islamabad	Islamabad	248799
3	Lahore General Hospital Laboratory	Lahore	51927
4	Jinnah Hospital Laboratory	Lahore	23320
5	Tb Bsl-3 Laboratory Lahore	Lahore	277637
6	University of Veterinary and Animal Sciences (Bsl-3)	Lahore	341244
7	Institute Of Public Health Lahore	Lahore	235510
8	Benazir Bhutto Hospital (BBH) Rawalpindi	Rawalpindi	557188
9	Allied Hospital Faisalabad	Faisalabad	940212
10	Trauma Centre (Bsl-3) Wazirabad	Wazirabad	397578
11	Nishtar Medical College	Multan	220104
12	KSMC Sialkot	Sialkot	240407
13	SHAHEEN Bsl-3 Lab Sargodha	Sargodha	507854
	Total No. of Tests		**79,41,780**

to deal with a high workload due to the increasing number of patients due to high transmission of an infection but also their own life remains at risk due to dealing with infectious pathogens [12, 13]. Diagnostic laboratories' fragility is significantly magnified during this pandemic. Reliability and accuracy of results remained a matter of concern for health authorities as false positive and false negative results not only affect the patient's health but can bring damage to public health policies, quality testing, emergency plans efficiency and pandemic control preventive measures [14].

In the present study, we tried to figure out the challenges faced by molecular-based public health labs during the COVID-19 pandemic in resource-limited settings. These challenges and lessons learned can be of utmost importance for any new pandemic preparedness and response. The knowledge of current diagnostic technologies and associated gaps in public health labs of developing countries can have a serious impact on global outbreaks.

This knowledge gap can be filled at the national level by a thorough assessment of issues faced by different diagnostic laboratories to tackle the COVID-19 pandemic. Identification of these gaps can halt the hidden deficiencies of any healthcare system and can facilitate removing those by ensuring improvements in the fight against any new pandemics such as COVID-19 global emergency.

2. METHODOLOGY

To access Pakistan's health system response to COVID-19 a qualitative large study was conducted. The data was collected using open-ended questions among healthcare professionals from 13 major public health national-level laboratories followed by in-depth interviews (IDI) among professionals. Collectively these laboratories performed almost 8 million SARS-CoV-2 PCR tests till now (from 1st March 2020 till 20th September 2022) which is such a huge number and contribution to the healthcare system of Pakistan in terms of tackling this COVID-19 severe pandemic and infection risks among the population. So, these labs also faced issues regarding preparedness and responses which are discussed in this study.

This study was conducted in two phases. In the first phase, open-ended questions among professionals were conducted to find out gaps related to Punjab Pakistan's health system response to COVID-19. According to the Strategic Preparedness and Response Plan (SPRP) from WHO COVID-19, different questions about 8 important points of the healthcare system were asked which include Country coordination, monitoring and planning, Risk communications and management, Surveillance, Entry points, National laboratories, Control and Prevention, Management of cases and Support & logistics [15]. Questions regarding these points were asked among a few healthcare

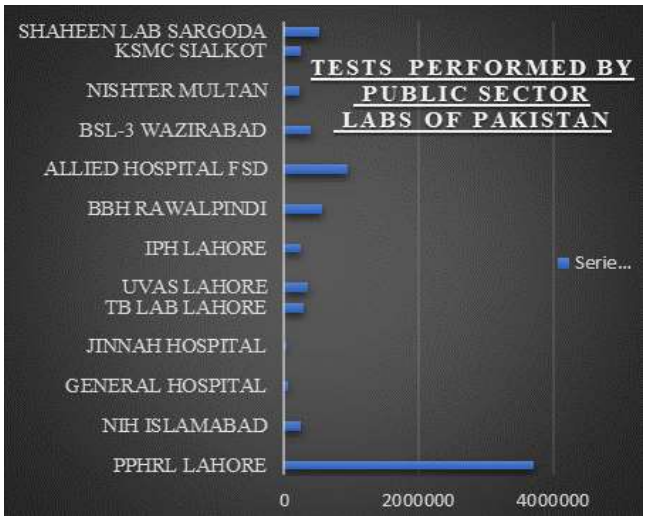

Fig. 1. Graphical representation of the total number of tests performed by public sector laboratories of Pakistan

professionals with various backgrounds to obtain their feedback for further improvement.

2.1 Data Collection

This study aimed to collect data from respondents from different laboratories with different experiences. The consent of respondents was taken before they participate in this study.

Respondents were allowed to share their experiences during the COVID-19 pandemic relevant to their background and expertise without any barrier. All responses were noted down in detail.

Few respondents based on their diverse backgrounds and expertise in the health system e.g. Health specialists, Lab in charge, public health specialists, focal persons, and health community representatives participated in this study. They also participated in the second phase of in-depth interviews.

2.2 Analysis of Data

The responses to open-ended questions were checked and analyzed. The content was arranged and content analysis was done independently by the research team followed by a series of group discussions. Gaps regarding health systems' preparedness and response during COVID-19 were marked, discussed, and noted.

3. RESULTS AND DISCUSSION

There are huge numbers of pre-analytical, analytical, and post-analytical challenges that are needed to be taken into consideration by public health authorities, medical lab technologists, and clinical microbiology laboratories to give accurate results shown in figure 2. It appears that the pre-analytical phase is the main source of errors in diagnostic laboratories accounting for approximately 46 % to 68.2 % of errors [16], It is observed despite continuous improvements during the whole testing process. Pre-analytical errors can result in compromised patient care, magnified financial burden, and unnecessary investigation in the healthcare system resulting in a compromised healthcare system [17].

3.1 COVID-19 Sample Collection Issues

During the Covid-19 pandemic, good sample collection materials and sample collection methods remained a hot topic as patients with COVID-19 had a high reservoir of the virus in their upper and lower respiratory tracks. Nasopharyngeal and oropharyngeal swabs are recommended for this purpose. According to the literature, nasopharyngeal swabs become a better option because they reach the exact area to be tested in the nasal cavity. But in China, oral swabs were mostly used instead of nasopharyngeal swabs frequently during the pandemic. The limit of detection accuracy from oral swabs is 30 % and from nasal swabs is 70 %. So, the COVID-19 infection detection with oral swabs was 32 %, and with nasal swabs was 63 %. Another reason to limit the sampling with nasal swabs was the unavailability of nasal swabs and viral transport media in Pakistan and also in those areas where extensive testing was required. Sampling issues which have been reported by different diagnostic labs were 77 % which contributed to compromised quality of diagnostic testing. These issues are an inappropriate and inadequate collection of the sample, sample mishandling, insufficient viral transport media in sampling vial, inappropriate conditions of sample transportation, prolonged sample storage, sample storage false temperature maintenance, presence of substances in the sample which can interfere with the such as cellular components due to additives and protocols issues occurring during sample preparation, manual sample preparation, pipetting issues, sample mismatch, inhibition and cross-contamination [18-20].

3.2 Leader Ship Issues

Governance and leadership are important components which influence the functioning of all main health systems. They depend on decisions made by the stakeholders in response to a pandemic [21-23]. There have been less effective coordination and strategic vision towards achieving a common goal, within and beyond the health systems among all the leadership. Seventy-seven percent of leadership and governance issues are reported from national labs of Pakistan because of high pressure from leadership, high workload, no appreciation, mismanagement, and less effective coordination.

Fig.2. Percentage of challenges in Laboratory Diagnosis faced by major public health laboratories of Pakistan

3.3 Biosafety, Biosecurity, and Lack of Resources Issues

Biosafety, biosecurity, and insufficient availability of resources issues are reported in 69 % of different national laboratories in Pakistan. This is a huge percentage level. Both of things are very important for qualitative working place. We should be prepared for coming pandemics to not repeat the same mistakes and should learn from the COVID-19 pandemic. Adding to this around 54 % of resources have been wasted. There must be a balance in the availability of resources in every lab to manage lack and waste of resources issues.

3.4 Mental Health, Salaries, and Workload Issues

Working under pressures in long duty shifts, getting exposed to COVID-19 infection, seeing people dying every day because of severe infection, leadership pressure, high workload, mismanagements, no salaries on time, short-term job contracts, fears of losing jobs, lack of resources and PPEs affected the mental health of health care workers to higher levels which are 92 %. This is a serious drawback of a pandemic. Mental health is very important for good well-being but in case, but if it affects a huge level the health of an individual is at risk. So, to prevent mental health issues a happy, motivated, and appreciative environment should be given to healthcare workers. Forty-six percent of the stigma of the population has been reported for healthcare workers who work in testing labs and 38 % of issues reported were related to late salaries.

Workload should be balanced between shifts and different labs working on the national level. Few labs receive very few samples and others receive thousands. The equal distribution of samples is very important in all the labs working to prevent workload. Ninety-two percent workload issue has been reported from national labs which is huge.

3.5 Assay Selection Issues

Amplification methods and deep sequencing methods played an important role in SARS-COV-2 diagnosis. Right after the SARS-CoV-2 outbreak, molecular diagnostic approaches were considered robust and primary diagnostic techniques for its detection but no one was sure what technique they should use for appropriate diagnosis. However, these approaches became a severe challenge for resources limited countries due to cost and shortages of diagnostic kits after its global outbreak. Thirty-eight percent of assay selection challenges were faced by different national laboratories in Pakistan. Molecular methods of deep sequencing e.g., next-generation sequencing and metagenomics now need

the hour for the identification of future variants of SARS-CoV-2. RT-PCR technique is mostly used to reduce time and cost. Multiple molecular methods are also in practice such as Multiplex isothermal amplification, loop-mediated isothermal amplification, microarray detection, and CRISPR (Clustered regularly interspaced short palindromic repeats) [18-20, 24].

3.6 Issues Related to False Results

In Pakistan, 38 % of labs reported the issue of false results. Molecular diagnostic techniques always remained the gold standard assays in the majority of infectious diseases diagnosis. RT-PCR method occasionally gives false positive and false negative results. But in the case of SARS-CoV-2, the situation remained twisted. Sometimes PCR results did not match with patients' signs and symptoms, leading to misunderstanding of false assay performance. On the other hand from a lab management perspective, quality test performance, kits reliability, and issues in sample collection are doubted. Nevertheless, these false results claim always lead to under or over-diagnosis of disease. A false positive result can have many consequences, it does not only lead to unnecessary treatment of patients but can also lead to social problems, as it makes the working of health professionals a question who are working in public laboratories facilities. In retrospect, a false negative result not only contributes to more spread of COVID-19 in the community but also to the patient's health remains at risk without any treatment. The precise and reliable results of diagnostic tests play a vital role in the diagnosis and management of the COVID-19 outbreak.

3.6.1 False Negative Results Issues

It is important to make precise and accurate approaches to the diagnosis of COVID-19 due to its high infection rate. False-negative results have harmful epidemiological effects to contain the outbreak and transmission of infection [25]. It is vital to reduce the number of false negative results for cohorts of patients in hospitals and determination of quarantine measures. Due to false negative results some patients who are hospitalized for other conditions unknowingly carry SARS-CoV-2. There is a need to differentiate recovered patients and silent carriers of SARS-CoV-2. This will help hospital management to sort out patients whom to discharge and whom to hospitalize.

3.7 Covid-19 Testing Results in Interpretation Issues

United States molecular diagnostic labs consider a sample confirm positive when initially both two targets N1 and N2 in CDC assay are present. A Cycle Threshold (CT) value less than 40 is defined as a positive test and 40 or more than 40 is considered as a negative test. CT value <40 of one of both targets, the test is considered as determinant and requires retesting for confirmation. In China, the assays for three targets, when there are two or more targets are positive, the test is considered positive. There have been some wrong practices during results interpretations due to a lack of experience, training, and CT value knowledge in laboratory staff. Low CT values for high viral load approaches should be used for results interpretation.

3.8 Laboratory Design Issues

Pakistan's government established BSL-3 laboratories at district levels due to a high number of patient samples during the COVID-19 pandemic. Each district designed its BSL-3 level lab in available resources and space. Healthcare workers reported many issues regarding lab design and space. Some labs were having huge spaces and some were very congested and inconvenient to work in them. Thirty-eight percent of labs reported this issue in Pakistan.

3.9 Test of Cure and Infectivity Issues

After recovering from COVID-19 disease and coming out from isolation the monitoring of patients with resolution of covid-19 pneumonia is not being done. It is a major concern as such patients enhance viral transmission. Even the discharged patients are shedding viable coronavirus and there are chances of transmission exist. It is recommended that the patient should remain isolated for at least 1 month. Two consecutive COVID-19 PCR tests with confirmed negative results should be conducted after cure and needs further investigation.

To increase diagnostic efficacy different approaches should be taken to overcome these

Table 2. Percentages of challenges and gaps reported by different healthcare laboratories.

Percentage of Labs	Gaps/Challenges
77 %	Sampling Issues
77 %	Administrative & Leadership Issues
53 %	Unavailability of Well-Trained Staff
53 %	Unavailability of Human Resource
69 %	Lack of Resources
69 %	Biosafety and Biosecurity issues
54 %	Wastage of Resources
92 %	Mental Health Issues
38 %	Shortage of Space
54 %	Fake Sampling
54 %	Lack of Training and Experience
61 %	Late Salaries
38 %	Fake Reporting Pressure
69 %	Supply chains of Kits, PPEs & Reagents
69 %	Work environment issues
92 %	High Work load
23 %	Waste Management
46 %	Stigma & Discrimination among HCW

challenges and gaps. Selecting the most appropriate source of sampling, as CDC recommends the nasopharyngeal swabs sampling which gives more accurate results [25-27]. Multiple diagnostic techniques should be used for confirmed results and to decrease the false negative results rate. Establishing a combined workflow of serological testing will help to achieve multidimensional, high-quality, reliable, and cost-effective diagnostic approaches for COVID-19 testing. The diagnostic testing should be done at different time points throughout the disease from hospitalization to weekly intervals [28] to decrease transmission. Governance and leadership are important components that expect to have effective coordination and strategic vision towards achieving a common goal, within and beyond the health systems among all the leadership. Strategic and timely coordination and communication is the main catalyst for resource optimization and distribution of workload. The expansion of laboratory testing capacity and enhancement in technology utilization through collaborative efforts between government agencies, universities, and industries would enable fast disease detection with diagnostic measures. With a centralized administrative system, COVID-19 responses such as health workforce mobilization and implementation of standardized operating procedures at all levels could be well-coordinated and synchronized effectively. However, such organized and managed commands could potentially restrict the local governments from making timely decisions for the best of their communities.

4. CONCLUSION

COVID-19 pandemic has dramatically highlighted the critical role of diagnostic technologies in the control of infectious diseases. The availability of established diagnostic technologies, which took decades to develop and optimize, has enabled scientists to plug and play in the design of SARS-CoV-2 diagnostics [29]. Different gaps and challenges have been faced by public health laboratories during the pandemic. Measures against the ongoing pandemic should be taken properly. Strategies should be established which are easier to administer and cover the challenges of pandemic diagnostic challenges. There is now a call for the development of ways to be rapidly implemented according to public health needs against any pandemic. Finally, the blinding speed with which SARS-CoV-2 has spread illustrates the need for preparedness and long-term investments in diagnostic testing.

5. CONFLICT OF INTEREST

No conflicts of interest between the authors and members of the potential conflicts of interest, counseling, expertise, working conditions, shareholding, and similar situations in any firm.

6. DECLARATION

The results of this manuscript are original. The same material is neither published nor under consideration elsewhere. The approval of all authors has been obtained before publication. In case the article is accepted for publication, its copyright will be assigned to the Pakistan Academy of Sciences. Authors must obtain permission to reproduce, where needed, copyrighted material from other sources and ensure that no copyrights are infringed upon.

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