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Molecular diagnosis of COVID-19 in Burkina Faso: successful challenge

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ABSTRACT

COVID-19 has worsened the health situation in Burkina Faso. In fact, the country has known a peak of the second wave, which began in November, and ended around January 2021. Biological diagnosis has played a key role in the management of COVID-19. The aim of this review paper is to address the practical aspects that laboratories have faced in order to meet the challenge of SARS-CoV-2 diagnosis in Burkina Faso. According to international requirements, Burkina Faso has used real-time Reverse Transcription Polymerase Chain Reaction (rRT-PCR) as the "gold standard" for the diagnosis of COVID-19. From March 9, 2020 to July 31, 2021, in Burkina Faso, laboratories involved in COVID-19 diagnosis analyzed 226,189 samples by molecular tests and 2, 352 samples by rapid antigenic tests, whose peak was in January 2021 with 35,984 samples analyzed. The daily average rate of samples analysis was 456.02 tests. The majority of the individuals requesting COVID-19 tests were travelers (62.00%), followed by contact cases (18.42%), suspected cases (7.95%), voluntary screening (7.57%), and 4.06% of other applicants consisting of health care personnel and at-risk patients. In terms of prevention, vaccines are being administered to the general population. However, some efforts must be made to provide automated sample analysis equipment and complete sequencing of SARS-CoV-2 remains among the challenges.

Keywords: Laboratory capacity, COVID-19, Burkina Faso.

INTRODUCTION

Coronavirus disease 2019 (COVID-19), which pathogen is SARS-CoV-2, caused a major health crisis as an emerging infectious disease which started in December 2019. The first cases of atypical acute lung disease have been reported in China (Chan et al., 2020). The disease has spread quickly worldwide. On

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January 30, 2020, the World Health Organization (WHO) declared covid-19 as a "public health emergency of international concern" and a pandemic on March 11th, 2020. Between December 2019 and August 31, 2021, there were estimates 197,667,583-reported cases of COVID-19 and over 4,214,100 deaths and 4,076,942,148 vaccine doses administered worldwide (CSSE, 2021). At the same time, Africa recorded 6,686,692 diagnosed cases including 169,608 deaths (Africa CDC, 2021) and Burkina Faso recorded 13,588 cases including 169 deaths and 37,329 vaccinated persons (CORUS, 2021; SIG, 2021). In Burkina Faso, the response to the COVID-19 is provided by the Emergency Response Operations Center (CORUS). In terms of vaccination, despite the low impact of the disease, on behalf of the Covax mechanism, the country received on May 30, 2021, 115,200 doses of AstraZeneca vaccine and on July 20, 2021, about 151,200 doses of Johnson&Johnson vaccine. Vaccination started on June 1st, 2021 (Ministère de la Santé, 2021).

Since the appearance of COVID-19, between rumors and denials, Burkina finally recorded its first two confirmed cases on March 9, 2020. Nationwide, COVID-19 spread from March 9, 2020 onward, making the country the sixth (6th) most affected in sub-Saharan Africa after Cameroon, Nigeria, Senegal, South Africa, and Togo, and fourth in West Africa. In light of the rapid evolution of the virus around the world, Burkina Faso authorities quickly activated their mechanism for managing epidemics of this type, which had been put in place during the Ebola epidemic in West Africa in 2013-2014 (CORUS, 2021).

Diagnostic tests have played a critical role in the management of COVID-19. These tests allow for confirmation of infection in patients, assist in the rapid triage of suspected cases (particularly in the community setting). They also contribute to the overall understanding of this new virus by determining exposure (current and past) to the virus and mapping the pandemic in different countries. In terms of laboratory capacity, Burkina Faso has also mobilized resources to ensure diagnosis and follow-up of suspected/contacted cases and travelers. Thus, from a single laboratory at the beginning of the pandemic, the country currently has about thirty laboratories involved in the molecular diagnosis of SARS-CoV-2. Efforts have been made by the government, laboratory stakeholders and partners of Burkina Faso to improve diagnosis performance. Thus, the aim of this study is to address the practical aspects that laboratories have faced in order to meet the challenge of SARS-CoV-2 diagnosis in Burkina Faso.

MOLECULAR AND GENOMIC FEATURES OF THE SARS-COV-2

Coronaviruses (CoVs) are a large group of viruses common among many animals, including humans. Before 2003, human CoVs were not considered deadly viruses. A virus called severe acute respiratory syndrome coronavirus 2 (SARSCoV-2) causes the coronavirus disease 2019 (COVID-19). The SARS-CoV-2 virus is classified by the International Committee on Viral Taxonomy (ICTV) and belongs to the family of Coronaviridae (subtype Coronavirinae and genus βetacoronavirus) (Gorbalenya et al., 2020 ; Wu et al., 2020). SARS-CoV-2 is the seventh (7th) coronavirus known to infect humans; SARS-CoV, MERSCoV, and SARS-CoV-2 can cause severe disease, while HKU1, NL63, OC43, and 229E are associated with mild symptoms (Andersen et al., 2020; Cheepsattayakorn and Cheepsattayakorn, 2020).

As a single-stranded and positive-sense RNA virus, SARS-CoV-2 is the most closely related (70% nucleotide similarity) with 2003 SARS-CoV and Middle East respiratory syndrome coronavirus (MERS-CoV) (Wu et al., 2020 ; Cao et al., 2021). Genomic analysis suggests that in December of 2019, the SARS-CoV-2 Wuhan NC_045512.2 (GenBank reference genome for 2019-nCoV), originated in bats and transmitted to humans through humans probably by pangolin in a seafood market in Wuhan, Hainan Wuhan, Hubei province, China (Goldsmith et al., 2004; Ahn et al., 2020). However, it is not clear whether the spread also involved a different intermediate animal host (Lai et al., 2020).

SARS-CoV-2 carries one of the largest RNA genomes (~30 kilobases, kb) and encodes about 29 proteins (Cao et al., 2021). This genome encodes the structural proteins, spike (S), envelope (E), membrane (M), and nucleocapsid (N). From genomic organization, sequence, and function, there is a large gene encoding for a polyprotein open reading frame (ORF1ab) at the 5′ end about two-thirds of the whole genome length, and at least six other accessory proteins (ORF3a; ORF6, ORF7a, ORF7b, ORF8, ORF10) are unique to SARS-CoV-2. Indeed, the two large genes ORF1a, ORF1b, encode 16 non-structural proteins (NSP1– NSP16). These NSPs are processed to form a replication–transcription complex (RTC) that is involved in genome transcription and replication. For example, NSP12 encodes for RNA-dependent RNA polymerase (RdRp). NSP3 and NSP5 encode for Papain-like protease (PLP) and 3CL-protease, respectively. Both proteins function in polypeptides cleaving and blocking the host innate immune response. NSP15 encodes for RNA helicase (Alanagreh et al., 2020; Abu et al., 2020; Yuan et al., 2020).

BRIEF OVERVIEW OF SARS-COV-2 DIAGNOSTIC TECHNIQUES

"The most effective way to prevent infections and save lives is to break the transmission chain. In addition, to do so, one has to test and be isolated. In fact, one cannot fight a fire blindfolded. Moreover, we cannot stop this pandemic if we do not know who is infected. We have a simple message for countries: test, test, test" says Dr. Tedros Ghebreyesus, WHO Director-General.

According to the Koch's postulates, the "gold standard" remains virus isolation from clinical samples in diagnosing viral infections (Falzone et al., 2021).

In the diagnosis of COVID-19, there are three (3) main methods of different but complementary importance and utility. These are nucleic acid amplification tests (NAATs), SARS-CoV-2 antigen detection tests and antibody detection tests. However, many other nucleic acid-based techniques such as Loop-mediated isothermal amplification (LAMP) and CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) are promising options. For SARS-CoV-2 diagnosis needs nasopharyngeal swab (NPS), oropharyngeal swab (OPS), bronchoalveolar lavage fluid (BALF), sputum, and stool samples collected from suspicious cases. Sample requirements are:

- Applicable sample types: upper respiratory tract samples (including throat swabs, nasal swabs, nasopharyngeal extracts, deep cough sputum); lower respiratory tract samples (including respiratory tract extracts, bronchial lavage fluid, lung tissue biopsy samples).
- Sample collection: Collect according to the conventional sample collection method.
- Sample storage and transportation: the collected specimens should be submitted for analysis in time, or stored at 4°C for 24 hours; it is best to store at -70°C for more than 24 hours and avoid repeated freeze-thaw cycles (WHO, 2020).

Molecular testing: rRT-PCR, next generation sequencing (NGS) and Xpert® Xpress SARS-CoV-2 (GeneXpert)

Real-time Reverse Transcription Polymerase Chain Reaction (rRT-PCR) and Xpert® Xpress SARS-CoV-2 (GeneXpert) are the major two frequently used NAAT for COVID-19 detection (Ilhan and Uysal, 2020; GeneXpert, 2020). Due to the relatively low costs of the entire viral RNA extraction, reverse transcription and amplification procedure, and the availability of RT‑PCR thermal cyclers, RT‑PCR‑based molecular tests are considered the optimal diagnostic option for wide surveillance strategies in hospitals, research institutes and private laboratories (Falzone et al., 2021). These rRT-PCR tests are used for the diagnosis, screening and elimination of SARS-Cov-2 infection. Molecular diagnosis of SARS-COV-2 targets viral genes (N for Nucleocapsid, E for Envelope, S for Spike, M for Matrix/Membrane and RdRP for RNA-dependent RNA polymerase) which constitute the basis of differences between diagnostic kits. As already mentioned, the European Medicines Agency (EMA) has approved 192

PCR based methods while the FDA (Food and Drug Administration) in the United States has approved 235 different molecular tests for both RT‑PCR and the rapid detection of SARS‑CoV‑2 RNA (EC, 2020).

The Xpert Xpress SARS-CoV-2 test performed on GeneXpert systems is a real-time RT-PCR test for the qualitative detection of SARS-CoV-2 nucleic acid. Positive results indicate the presence of SARS-CoV-2 RNA. Positive results do not rule out bacterial infection or co-infection with other viruses. Negative results do not exclude infection with SARS-CoV-2 and should not be used as the exclusive criterion for treatment or management decisions (GeneXpert, 2020).

In clinical diagnosis, the use of NGS platform (Ion AmpliSeq SARS-CoV-2 Research Panel (ThermoFisher), Scientic MinION (Oxford Nanopore Technologies) and IDbyDNA (Illumina)) is limited because of its equipment dependency and high cost.

Antigen detection test: rapid diagnostic test (RDT), FIA

Antigenic tests detect Sars-CoV-2 specific proteins. These tests can be performed on nasopharyngeal swabs; lower respiratory tract swabs are not yet recommended for diagnosis but only for monitoring COVID-19. However, due to their low performance, especially in the case of low viral load, these antigenic tests are not yet validated for the diagnosis of COVID-19. Rapid antigen detection tests (Ag-RDT) using immunochromatographic (ICT) or fluorescence immunoassays (FIAs) have recently become available. For example, the PanbioTM COVID-19 Ag Rapid Test (Abbott) is a lateral flow immunochromatographic test. It is a rapid *in vitro* diagnostic test for the qualitative detection of SARS-CoV-2 antigen (Ag) in human nasopharyngeal swab specimens from individuals meeting the clinical and/or epidemiological criteria for COVID-19. STANDARD F COVID-19 Ag FIA (SD Biosensor is a rapid fluorescence immunoassays (FIA) which use an automated reader.

Antibody detection test: rapid diagnostic test (RDT), ELISA

Serological tests allow the detection of specific antibodies (Ac) (immunoglobulins: Ig) produced by the organism and directed against SARS-COV-2. Immunoglobulins M and G (IgM and IgG) are the most frequently biomarkers used for the SARS-COV-2 serological revelation. Within viral infections, IgM antibodies are the first line of defense and indicate that the patient has recently been infected/re-infected, while COVID-19 IgG antibodies appear later and last longer and signalize exposure to the virus some time ago. These tests are only for monitoring and seroprevalence studies of COVID-19. For example, the Biosensor Standard™ COVID-19 IgG/IgM Rapid Test Device is an immunochromatographic test for the qualitative detection of IgG and IgM antibodies to SARS-CoV-2. The use of this method may be limited as it is less probable to find out cases in the early stages of the disease, in addition, cross reactivity to other coronaviruses may be challenging (Jaddaoui et al., 2021).

To confirm the serological tests, it is often used enzyme-linked immunosorbent assay (ELISA). ELISA is a colorimetric, chemiluminescent, fluorescent microwell plate-based assay, with the availability of automated, or semi‑automated systems that allow a precise quantitation of human proteins, immunoglobulins, viral antigens and other peptides. SARS‑CoV‑2 ELISAs represent a good clinical option for large screening and surveillance campaigns mainly adopted for specific work categories due to the rapidity of this method, the possibility of analyzing multiple samples in one round and the availability of automated or semi‑automated systems that allow a precise quantitation of viral antigens or human antibodies (Falzone et al., 2021).

Rapid antigenic and rapid antibody tests are characterized by more rapid execution times of ~15‑30 min, a lower cost and an easier compared to RT‑PCR‑based methods. Their procedure that does not require the presence of highly trained personnel. These tests are mainly built on platforms based on the

principle of lateral flow immunoassay for the direct detection of viral proteins (rapid antigen tests) (Falzone et al., 2021).

Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR- Cas9)

Over the last 6 years, the CRISPR-Cas9 technology has expanded dramatically in many areas and become a powerful toolbox for genetic manipulation based on a small guide RNA (gRNA) recognition. Among the COVID-19, the US FDA-EUA (U.S. Food and Drug Administration-Emergency Use Authorization (EUA) have approved the Specific High Sensitivity Enzymatic Reporter UnLOCKing (Sherlock™ CRISPR SARS-CoV-2 assay (Sherlock Biosciences). This system based on Cas13a nuclease activity is composed of two different stages. It is a CRISPR/Cas13 used synthetic RNA fragments of the SARSCOV-2 virus, target S and ORF1ab regions can be revealed in a range between 10 and 100 copies per microliter of input. Hence, SHERLOCK approach can rapidly substitute rRT-PCR given the high demand for rapid diagnostic tests in the current epidemiological situation of COVID-19 (Zhang et al., 2020).

Likewise, the COVID-19 RT–PCR assay performed by the CDC-Atlanta could be replaced with this CRISPR-based DETECTR (DNA Endonuclease-Targeted CRISPR Trans Reporter) assay, as it presents 100% negative predictive agreement and 95% positive predictive agreement (Broughton et al., 2020a, 2020b).

RT-LAMP: Loop-mediated isothermal amplification

It is a rapid, sensitive and efficient visual amplification method for nucleic acids. Recently, this method has been widely used for the isolation of influenza virus, Middle East respiratory syndrome-CoV, West Nile virus, Ebola virus, Zika virus, yellow fever virus and a variety of other pathogens (Chotiwan et al., 2017). The scientists developed a Lamp a reverse transcription (RT-Lamp) assay to detect Sras-CoV-2 in people with COVID-19 (Amir et al., 2020). In a study revealed that the sensitivity of RT-PCR is identical and that of RT-Lamp (Li et al., 2015).

The Abbott ID NOW™ COVID‐19 assay (Abbott Laboratories) is the most commonly used in SARS-CoV-2 RT‐LAMP methods approved by US FDA-EUA. This RT‐LAMP‐based system ensures high‐sensitive results in ~5 min through the identification of the SARS‐CoV‐2 RdRp gene (Huang et al., 2020).

MOLECULAR DIAGNOSTIC: LABORATORY CAPACITY DURING COVID-19 IN BURKINA FASO

COVID-19 cases in Burkina Faso

Until July 31, 2021, Burkina Faso has only officially used molecular diagnostics and at times antigenic tests (PanbioTM COVID-19 Ag (Abbott) and STANDARD™ Q COVID-19 Ag (Biosensor)) in the city of Ouagadougou. All the suppliers who brought the antigenic and serological tests into the country gave samples to the Ministry of Health (Ministère de la Santé, 2021) for technical evaluation and authorization before sales. The Biomedical Research Laboratory of the Institut de Recherche en Sciences de la Santé (LaReBio/IRSS/CNRST) has evaluated these tests. The results are currently being validated.

Figure 1 shows the number of samples analyzed per day according to positive cases of COVID-19. From March 9, 2020 to July 31, 2021, in Burkina Faso, laboratories involved in COVID-19 diagnosis analyzed 226,189 samples by molecular tests (RT-PCR and GeneXpert) and 2352 samples by rapid antigenic tests ((PanbioTM COVID-19 Ag (Abbott) and STANDARD™ Q COVID-19 Ag (Biosensor)), whose peak was in January 2021 with 35,984 samples analyzed. The average daily rate of samples analysis was 45,602 tests, but other January 2021 and February 2021 recorded respectively an average of 1,160.77 and 917 tests per day (CORUS, 2021 ; SIG, 2021).

The majority of the individuals requesting a COVID-19 test were travelers (140,312 or 62.00%), followed by contact cases (41,680 or 18.42%), suspected cases (17,984 or 7.95%), voluntary screening

(17,130 or 7.57%), and 4.06% of other applicants consisting of health care personnel and at-risk patients including hemodialysis patients, hypertensives and diabetics. In January 2021, it was noted nationwide that almost 2 out of 3 tests were performed for travel reasons (iMMAP, 2021).

Figure 1 shows the second wave of COVID-19 in Burkina Faso between November 2020 and February 2021. During the months of December 2020 and January 2021, the average number of new cases was 123 and 127 per day respectively. Moreover, the highest daily number of samples (2,241) analyzed per day was on February 5, 2021. The new wave of COVID-19 cases observed over the last few months did not spare any administrative region of Burkina Faso. However, the two main epicenters remained the Centre and "Hauts-Bassins" regions. These two regions account for more than 85% of the cases recorded in the country. At the end of February 2021, the case fatality rate in Burkina Faso was 1.16% (iMMAP, 2021).

Increase in laboratory capacity during the pandemic

According to the Medical Biology Laboratories Department of the Ministry of Heath (DLBM), at the end of December 2019, the number of laboratories listed was 235, of which 104 were public (44%) and 131 private and religious (Ministère de la Santé, 2021; SIG, 2021). As soon as the first cases of COVID-19 were announced on March 09, 2020, the National Influenza Reference Laboratory (LNR-G), located in Bobo-Dioulasso, was put in charge of testing. Then the health authorities gradually increased the country's diagnostic capacity. According to Brice Wilfried Bicaba, the Corus coordinator: Burkina Faso has gone from one testing center to seven laboratories in the country's two main cities (Ouagadougou and Bobo-Dioulasso) in May 2020, each of which is capable of performing more than 190 tests per day. These centers are the armed wing of Burkina Faso's response and will allow the country to reach a capacity of 1,000 diagnostic tests per day, if necessary (Coulibaly, 2020).

Then on November 30, 2020, it was there were ten (10) laboratories involved in the molecular diagnosis by RT-PCR of COVID-19 disease in Ouagadougou and Bobo-Dioulasso (Sagna et al., 2021) . As of June 2021, the country has 36 facilities performing COVID-19 molecular testing (Ministère de la Santé, 2021) . Each administrative region has at least one laboratory involved in the diagnosis of COVID-19 (Figure 2). According to medical experts, the number of reported cases is an acute underestimation, and a high number of cases remain undetected.

Most African countries have in-adequate surveillance and laboratory capacity. Furthermore, the majority of the countries rely on donors aid to supplement public health budget, and some countries were only able to start COVID-19 testing after receiving donated testing kits from the Jack Ma Foundation (Dzinamarira et al., 2020).

In Burkina Faso, laboratory workers staff were present almost 24 hours a day, 7 days a week, especially during the second wave (November 2020-February 2021), to manage diagnostic emergencies for travelers, suspected cases and contact cases, while handling samples. This has made their job more difficult: shifting hours, night work and work on weekends and holidays. This prompted the French deputy Jean-Carles Grelier, in his address to the Minister of Health, to suggest that: "for all these legitimate reasons, laboratory staff deserve a fair recognition" (Dalmat, 2020).

Disparity of SARS-CoV-2 RNA extraction and amplification kits

In Burkina Faso, laboratories involved in the diagnosis of COVID-19 used different viral RNA extraction kits during the same working day (table 1). This could lead to mismatches in results for samples from the same person collected under the same conditions. The table 1 shows that there were as an equal number of manual and automatic extraction kits in Burkina Faso. Some laboratories already had all of the automated extractors listed in table 1 except for the KingFisher Flex Purification System 24 and

96* (ThermoFisher) which was acquired to strengthen the capacity of laboratories involved in COVID-19 diagnostics. However, fully automated diagnostic devices have a Limit of detection (LoD) of 125 to 1 × 105 copies/mL, which seems less sensitive than no fully automated diagnostic devices with a LoD of 10 to 5.5 × 104 copies/mL (Falzone *et al.,* 2021). However, a comparison of three automated extraction systems, found that the EZ1AdvancedXL system (Qiagen) demonstrated the best analytical sensitivity. The nucleic acids extracted by EZ1AdvancedXL showed higher positive rates for virus detection than MICROLAB Nimbus IVD (Hamilton, USA) and QIAcube (Qiagen, Germany). Meanwhile, the MICROLABNimbus IVD system was comprised of fully automated steps from nucleic extraction to PCR setup function that could reduce human errors. For the nucleic acids recovered from nasopharyngeal swab specimens, the QIAcube system showed the fewest false negative results and the best concordance rate, and it may be more suitable for detecting various viruses including RNA and DNA virus strains. Therefore, these factors should be considered when new nucleic acid extraction systems are introduced to the laboratory (Kim et al., 2014).

According to the WHO interim guidance, the confirmation of suspicious cases is by Nucleic Acid Amplification Tests (NAAT), in an area with no COVID-19 virus circulation, using at least two different targets on SARS-COV-2 genome. However, in areas where the pandemic is widely diffused, the case is determined to be positive by screening only a single distinctive target (WHO, 2020). For Burkina Faso, several amplification kits detecting one to three genes were used. BGI Real-Time Fluorescent RT-PCR Kit for Detecting SARS-CoV-2 kit is the unique kit with one gene. These kits had LoD of 10 copies/mL to 1,000 copies/mL (Liferiver Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit) (table 2). RT‑PCR methods ensure a low limit of detection (LoD) of SARS‑CoV‑2 RNA (Böger et al., 2021).

However, there are the possible reasons which can explain the "RT-PCR false negative". First, a mutation leads to the off-target mismatch between the primer and the target sequence. For example, the ORF1a, ORF8, and N gene contain hot-spot loci. Second, for asymptomatic, mildly symptomatic, or discharged patients, low viral loads may be insufficient for RT-PCR detection. Third, the presence of interfering substances may generate "false-positive" results (Falzone et al., 2021). Despite the importance given for RT-PCR, some pre-analytical and analytical issues may contribute to threatening its correctness such as inadequate procedures for collection, proper transportation and storage of samples, personnel qualifications, use of inadequate assays, as well as malfunctioning equipment. For all these reasons, the sensitivity and specificity of the real-time RT-PCR test is not 100% (Jaddaoui et al., 2021). In Burkina Faso, there is no study done to detect possible mutations, which could optimize the use of the kits listed in the table 2. Furthermore, at the beginning of the pandemic, the collection, storage and transport of samples was handled by (CORUS, 2021). Since January 2021, this process is the Regional Health Directorates of the Ministry of Health. Thus, the laboratories did not control the entire chain from the sampling to the PCR result.

Concerning the thermocyclers, Burkina used about ten different models. Before COVID-19, some laboratories already had (a limited number) of the types of equipment listed in the Table 3 except for the *Applied Biosystems™ QuantStudio 3 and 5 Real-Time PCR Systems which was acquired to strengthen the capacity of laboratories involved in COVID-19 diagnostics.

External evaluation of the quality (EEQ) of biological diagnosis of COVID-19

As part of the improvement of the quality of biological diagnosis of COVID-19, under the supervision of the Medical Biology Laboratories Department (DLBM-Ministry of Health), the Burkina Faso has benefited from

three (3) external quality control programs, one national and two international.

Firstly, in September 2020, the National Influenza Reference Laboratory (LNR-G) organized in collaboration with the Ministry of Health-Burkina Faso, an external evaluation of the quality of the diagnosis of COVID-19, which involved all 11 laboratories (07 in Ouagadougou and 04 in Bobo-Dioulasso) involved at that time in this diagnosis by RT-PCR technique. The results highlighted the disparity of the amplification kits used by the laboratories compared to the amplification kit used by the LNR-G for quality control.

Secondly, an assessment was organized by WHO-Burkina Faso, which involved 12 laboratories involved in this diagnosis by molecular biology technique (RT-PCR or GeneXpert) in the cities of Ouagadougou, Bobo Dioulasso, Gaoua, Dori and Tenkodogo.

Thirdly, Burkina Faso benefited from the financial support of Africa CDC (Centers for Disease Control and Prevention in Africa) and ASLM (African Society for Laboratory Medicine) through the RESOLVE SurgeCov19Testing project to participate in the External Quality Assessment (EQA) program organized by One World Accuracy (Vancouver, Canada). After first and second rounds organized in October 2020 and November 2020 in which Burkina Faso participated with 15 laboratories, a 3rd round with 30 laboratories was organized in July 2021 for which Burkina Faso still participated (list of laboratories in Table 1). Thus, One World Accuracy sent test samples to those medical laboratories performing the biological diagnosis of COVID 19 by molecular biology techniques (rRT-PCR or GeneXpert). The results were recorded by the laboratories on the "Oasis" platform (https://www.oneworldaccuracy.com/).
Burkina Faso has always performed well on all these assessments (Ministère de la Santé 2021).

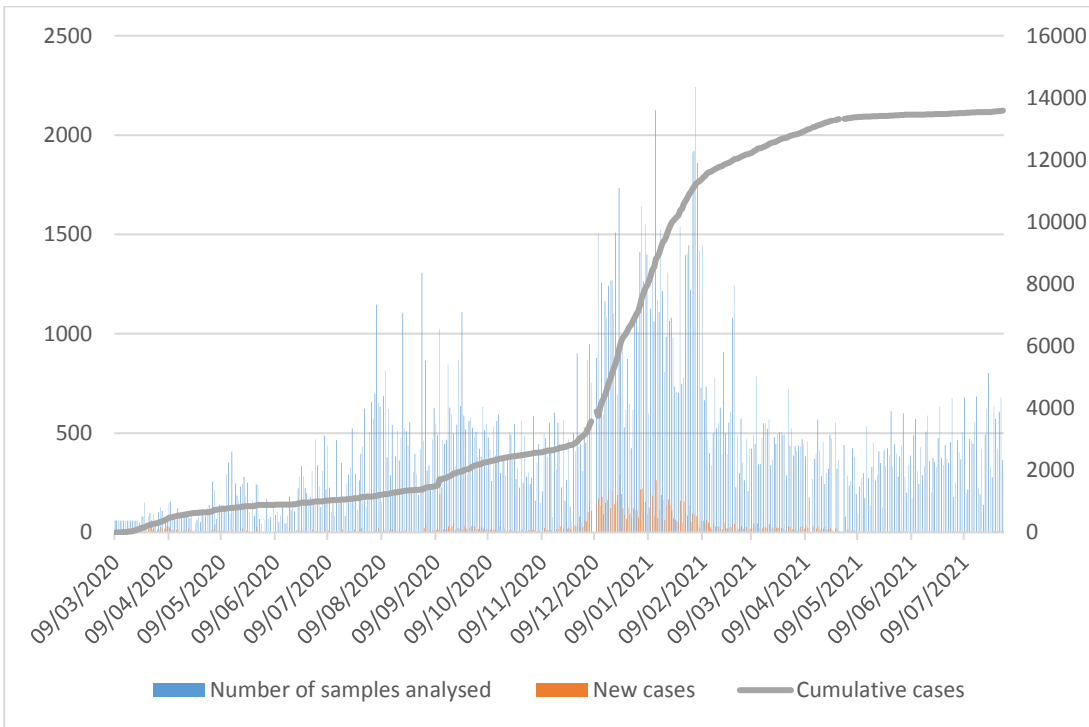
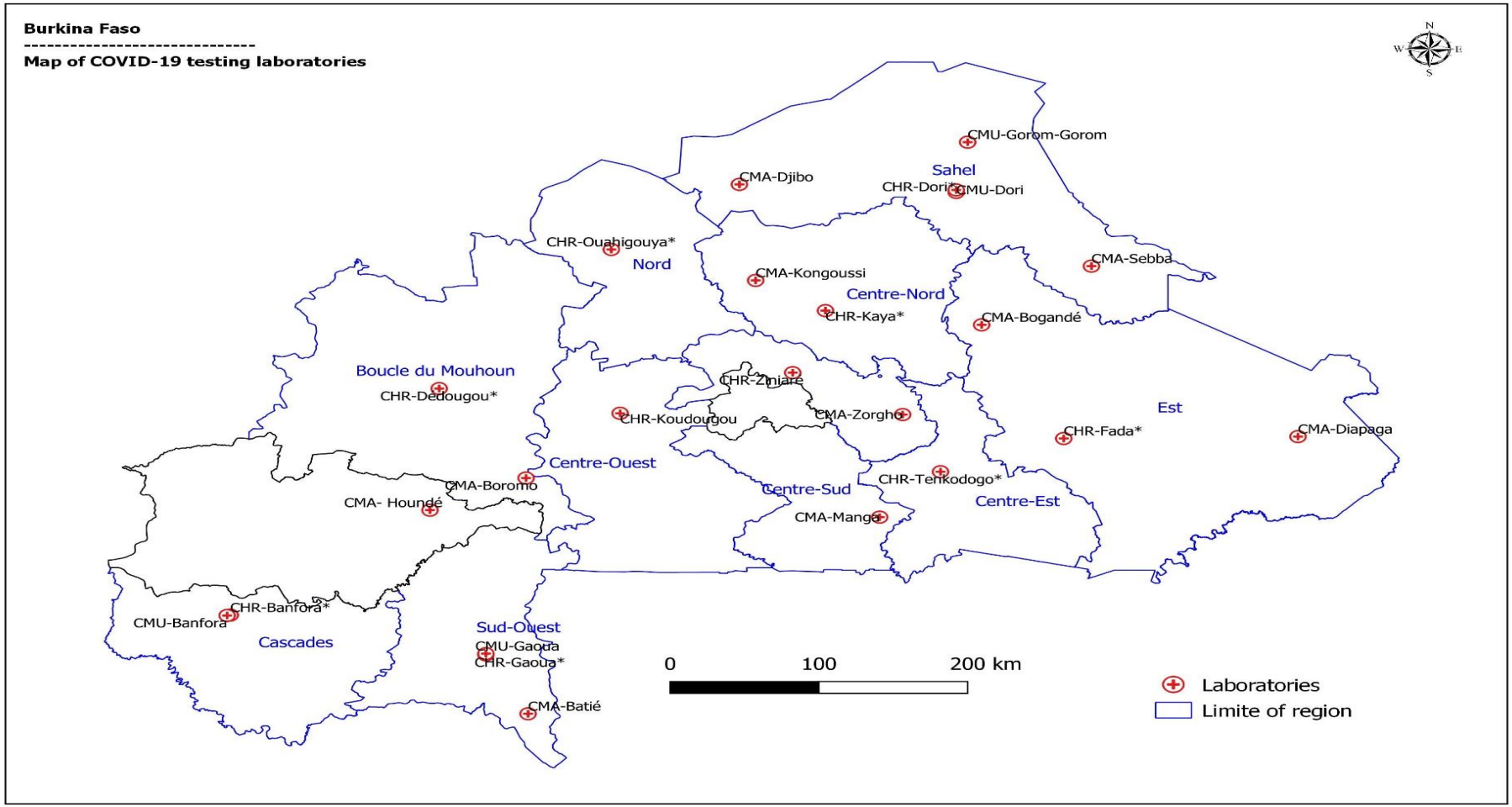
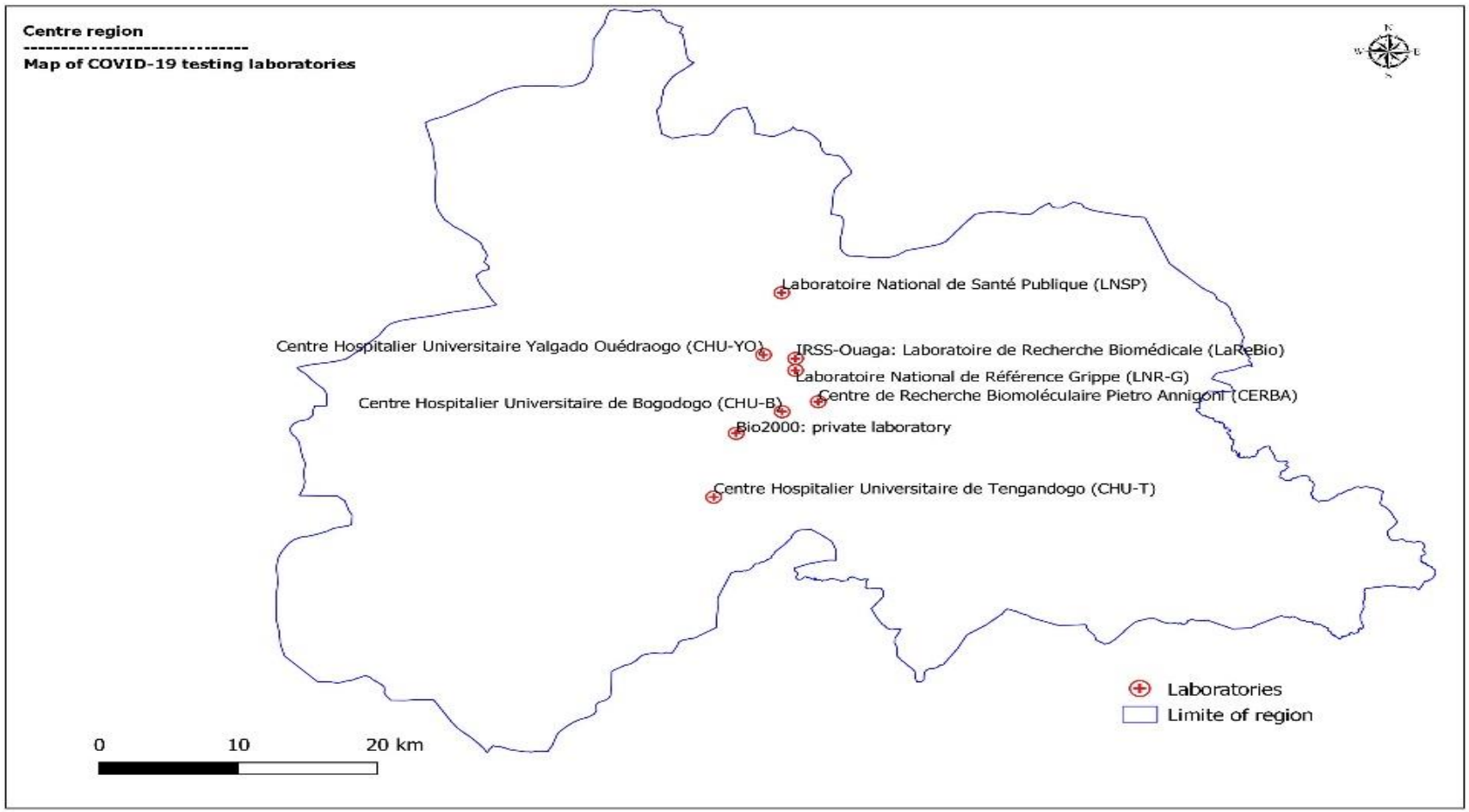


Figure 1: Evolution of COVID-19 cases in Burkina Faso. Legend: Ordinate axis: on the left is the number of cases or tests performed and on the right is the cumulative number of cases.


Burkina Faso
Map of COVID-19 testing laboratories
CMU-Gorom-Gorom
CMA-Djibo
Sahel
CHR-Dori*
CMU-Dori
CHR-Ouahigouya*
Nord
CMA-Kongoussi
CMA-Sebba
Centre-Nord
CHR-Kaya*
CMA-Bogandé
Boucle du Mouhoun
CHR-Dédougou*
CHR-Zniare
CHR-Koudougou
CMA-Zorgho
Est
CHR-Fada*
CMA-Diapaga
Centre-Ouest
CMA-Boromo
CMA- Houndé
CHR-Tenkodogo*
Centre-Sud
Centre-Est
CMA-Manga
CHR-Banfora*
CMU-Banfora
Cascades
Sud-Ouest
CMU-Gaoua
CHR-Gaoua*
CMA-Batié
0
100
200 km
Laboratories
Limite of region


Centre region
Map of COVID-19 testing laboratories
Laboratoire National de Santé Publique (LNSP)
Centre Hospitalier Universitaire Yalgado Ouédraogo (CHU-YO)
IRSS-Ouaga: Laboratoire de Recherche Biomédicale (LaReBio)
Laboratoire National de Référence Grippe (LNR-G)
Centre de Recherche Biomoléculaire Pietro Annigoni (CERBA)
Centre Hospitalier Universitaire de Bogodogo (CHU-B)
Bio2000: private laboratory
Centre Hospitalier Universitaire de Tengandogo (CHU-T)
Laboratories
Limite of region
0
10
20 km

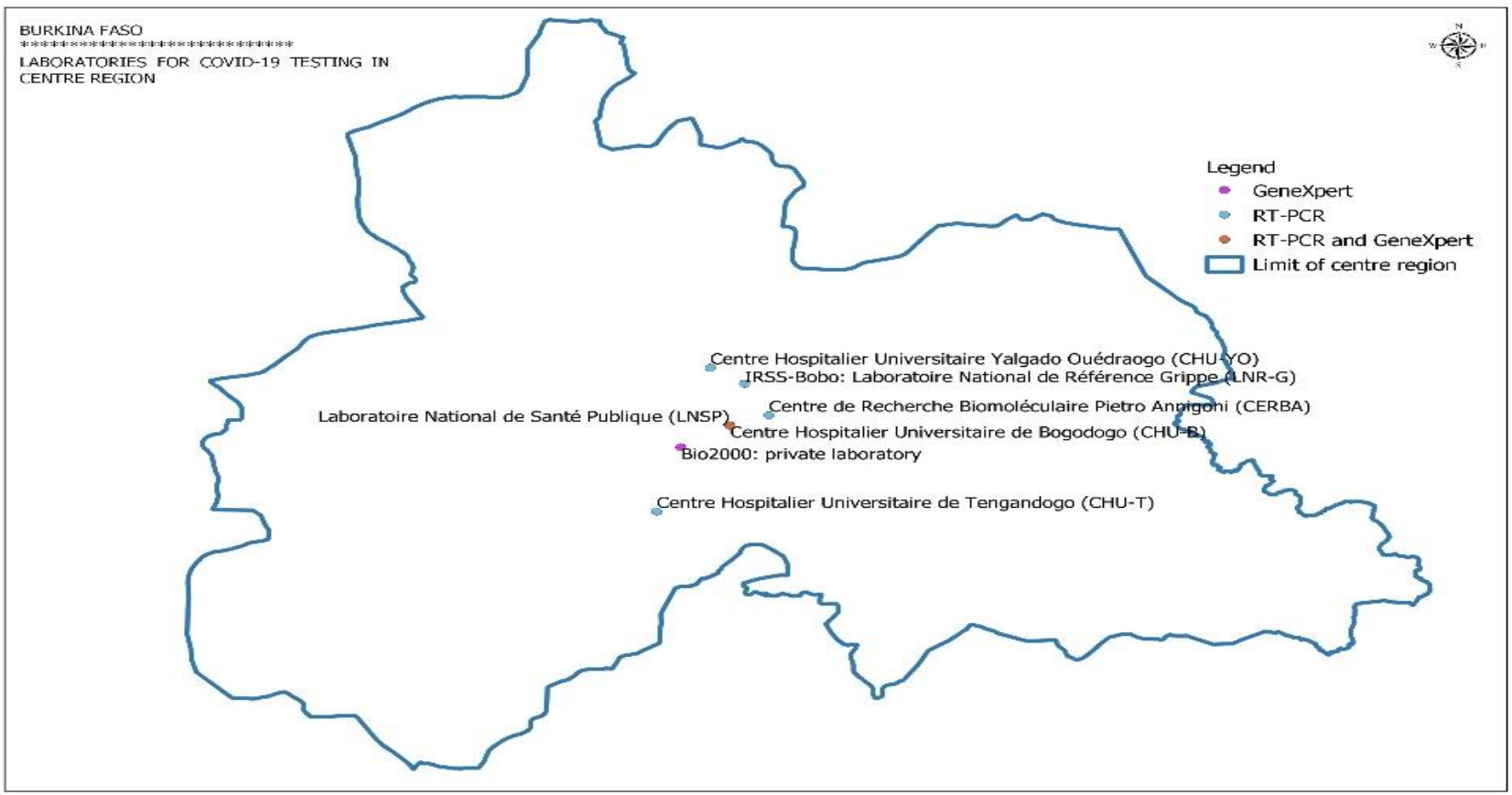


Figure 2: Laboratories involved in the molecular diagnosis of COVID-19 by RT-PCR.

* Regional laboratories equipped with QuantStudio 5 thermal cyclers (Applied Biosytems) during the pandemic. CMU : Centre Médical Urbain. ; CMA : Centre Médical avec antenne chirugicale ; CHR : Centre Hospitalier Régional

Table 1: RNA extraction kit used for SARS-CoV-2 diagnostic in Burkina Faso.

Manual extraction kit					
N°	**Platform(extraction equipment)**	**Reactions numbers**	**Principe**	**Sample**	**reference**
1	QIAamp Viral RNA Mini Kit (QIAGEN®)	50 or 250 preparations	-Manual or automated on QIAcube -silica-based membrane with the speed of microspin or vacuum technology	Viral RNA from plasma (treated with anticoagulants other than heparin), serum and other cell-free body fluids.	https://www.qiagen.com/us/products/diagnostics-and-clinical-research/sample-processing/qiaamp-viral-rna-kits/
2	MGIEasy Nucleic Acid Extraction Kit (MGI Tech Co., Ltd)	96 or 1728 preparations	-Manual or automated extraction on MGISP-960 - Magnetic Beads	viral DNA and RNA from throat swabs, BALF (bronchoalveolar lavage fluid)	https://en.mgi-tech.com/products/reagents_info/26/
3	MagMAX ™ Viral/PathogenNucleic Acid Isolation Kit (Applied Biosystems™)	100 preparations	- Manual or automated on KingFisher Flex -Magnetic Beads using a magnetic stand	RNA and DNA from virus and easy to lyse bacteria in biofluids and transport media samples.	https://www.thermofisher.com/order/catalog/product/A42352#/A42352
4	NUCLISENS® MINIMAG® Manual (Biomérieux)	24 preparations	-Manual nucleic acid extraction magnetic -Silica version of BOOM technology	RNA or DNA from plasma, CSF, Stool, Throat swab, Sputum, Whole blood, Lung biopsy	https://www.biomerieux-diagnostics.com/nuclisensr-minimagr
5	Abbott Sample Preparation	96 samples per run in 4 hours	Abbott mSample Preparation System (4 X 24 Preps) Manual nucleic acid extraction	Viral RNA and DNA, bacterial DNA, genomic DNA from serum/Plasma, urine, whole blood, Swabs, puncture Fluids, biopsy, semen, sputum, surgical fluids, stool	https://www.molecular.abbott/download-ifu?controlNumbers=51-608381R5
Automatic extraction kit					

N°	**Platform(extraction equipment)**	**Reactions numbers**	**Principe**	**Sample**	**reference**
1	MagNA PURE 96* and 24* (Roche)	96 samples in less than one hour	MagNA Pure DNA and Viral NA Small Volume Kit (Roche)	Viral DNA and RNA, and bacterial and fungal DNA from mammalian whole blood, serum, plasma, urine, sputum, swabs, stool, bronchoalveolar lavage (BAL), cerebrospinal fluid (CSF), and bacterial cultures (with or without external lysis option) and fresh tissue or single FFPE tissue slices (10 µm)	https://diagnostics.roche.com/global/en/products/instruments/magna-pure-96.html
2	NucliSENS®easyMAG® 24* (BioMérieux)	Hands-on time: <15 for 24 samples 24 extractions in 40 minutes	NUCLISENS Nucleic Acid Extraction Reagents (Biomérieux)	RNA or DNA from plasma, CSF, Stool, Throat swab, Sputum, Whole blood, Lung biopsy,	https://www.biomerieux-usa.com/clinical/nuclisens-easymag
3	**KingFisher Flex Purification System 24 and 96* (ThermoFisher)	96 in less than 20 minutes	MagMAX Viral/Pathogen Nucleic Acid Isolation Kit (Applied Biosystems™)	RNA and DNA from virus and easy to lyse bacteria in biofluids and transport media samples.	https://www.thermofisher.com/order/catalog/product/5400640#/5400640
4	Arrow 12* (NorDiag/DiaSorin)	12 samples in less than 45 minutes	Arrow Viral Nucleic Acid Kit (NorDiag /DiaSorin)	RNA and DNA isolation Stool, Blood, Urine, Serum, Plasma, Swabs, Tissue, Cells, FFPE, Sputum, Culture, Saliva	http://isogen.nl/arrow
5	Abbott m2000sp instrument 96* (Abbott)	96 samples per run in 4 hours	Abbott mSample Preparation System (4 X 24 Preps)	Viral RNA and DNA, bacterial DNA, genomic DNA from serum/Plasma, urine, whole blood, Swabs, puncture Fluids, biopsy, semen, sputum, surgical fluids, stool	https://www.molecular.abbott/int/fr/products/instrumentation/m2000-realtime-system
6	abGenix™ 32* Nucleid acid extractor (AIT Biotech)	32 samples per run in as short as 20 minutes	abGen Viral Nucleic Acid Extraction Kit Magnetic Pillar Rod Technology	Viral DNA and RNA from cell-free body fluids such as serum, plasma and cell culture supernatant	https://aitbiotech.com/abgenix/

*in this column, the numbers indicate the number of wells **automatic extractor acquired during the COVID-19 pandemic

Table 2: RT-PCR amplification kit used for Sars-Cov-2 diagnostic in Burkina Faso.

	RT-PCR amplification kits	**Manufacturer**	**Result interpretation**	**Detection target region/ Time of run**	**Specimen**	**Instrument**	**LoD or specificity**	**References**
1	BGI Real-Time Fluorescent RT-PCR Kit for Detecting SARS-CoV-2». 50 tests/kit	BGI Health (HK) Co. Ltd, China	Positive control Ct<35 **Positive**: VIC/HEX Ct <35 and FAM Ct <37. **Negative**: VIC/HEX Ct <35 and FAM Ct >37. **Invalid**: VIC/HEX Ct >35 and FAM Ct <37	1 gene (ORF1ab: FAM) and internal control (IC): VIC/HEX 1h54min16s*	Oropharyngeal swabs (OPS), nasopharyngeal swabs (NPS) and Broncho alveolar lavage fluid (BALF), anterior nasal swabs, mid-turbinate nasal swabs, nasal washes, and nasal aspirates	ABI 7500 Fast Real Time PCR, Roche LightCycler® 480, ABI QuantStudio 5 Real-Time PCR	100 copies/mL	(BGI 2020)
2	Liferiver Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit 25 tests/Kit	Shanghai ZJ Bio-Tech Co., Ltd. Shanghai, China	Positive Control ≤35 **Positive**: positive detection signal at least 2 genes with Ct≤41. **Negative**: no detection signal or Ct>41 with internal control Ct≤41 **Invalid**: all genes with Ct>41 Detection of Internal Control is not required if result positive in any of the other three detection channels.	3 genes (ORF1ab FAM, N : HEX/VIC/JOE, and E : Cal Red 610/ROX/TEXAS RED) and IC: Cy5 1h36min20s	deep cough sputum, NPS and BALF	ABI Prism®7500/7900; Bio-Rad CFX96; Rotor Gene™6000; SLAN	1,000 copies/ml	(Liferiver 2020)
3	DAAN Gene Detection Kit for 2019 Novel	Da An Gene (DAAN Gene	Positive control Ct≤ 32 **Positive**: FAM and VIC Ct value ≤ 40	2 genes (ORF1ab: VIC and N: FAM) and IC: Cy5	throat swabs, sputum, BALF, anus swab, blood	ABI 7500, LightCycler480, AGS4800,	500 copies/ml.	(DaAnGene 2020)

	Coronavirus (2019-nCoV) RNA (PCR-Fluorescence Probing) 24 tests/kit; 48 tests/kit	Co. Ltd, China	**Negative**: no signal or VIC and FAM Ct > 40 **Invalid**: Ct value of ≤ 40 in a single channel of FAM or VIC. When the IC Cy5 result is negative, the test tube is also negative	2h10min48s		Bio-Rad CFX96		
4	TIB MolBiol real-time RT-PCR assay 96 tests/kit	TIB MOLBIOL, GmbH, Germany	Positive control Ct≤ 36 **Positive**: FAM (E and RdRP) Ct ≤ 36 and CY5 negative or positive **Negative**: no signal or FAM (E and RdRP) Ct >36 and CY5 positive **Invalid**: FAM (E) Ct ≤ 36 and no signal of FAM (RdRP) and CY5 negative or positive	3 genes (E, RdRP : FAM) and IC: Cy5 Screening with E gene and Confirmation with RdRP gene assay to detect 0h56min39s	tracheal aspirates, BALF, throat and NPS	ABI 7500 Fast Real-Time PCR., BioRad CFX96 RotorGene, LightCycler	10 copies/mL:RdRP-gene assay 10 copies/ mL: E-gene assay	(TIBMolBiol 2020)
5	Novel Coronavirus(2019-nCoV) Nucleic Acid Diagnostic Kit (PCRFluorescence Probing) 24 tests/kit; 48 tests/kit	Sansure Biotech , Changsha, Hunan Province, China	Positive control Ct≤ 40 **Positive**: FAM or ROX Ct ≤ 40 and CY5 negative or positive **Negative**: no signal or FAM and ROX Ct >40 and CY5 positive **Invalid**: no signal of FAM, ROX and CY5	2 genes (ORF1ab: FAM and N: ROX) and IC: Cy5 2h45min30s	NPS, OPS and BALF, anterior nasal, and midturbinate swabs, nasal washes and nasal aspirates	7500 Fast Real-Time PCR., QuantStudio™5 , Roche cobas 4800, SLAN-69P, MA-6000PCR	200 copies/mL no cross reactivity	(Sansure 2020)

6	TaqPath™ COVID‑19 CE‑IVD RT‑PCR Kit 1,000 reactions/kit	ThermoFisher Scientific, Life Technologies Corporation, Kwartsweg,, The Netherlands	Interpretation of the results is performed by the Applied Biosystems™ COVID‑19 Interpretive Software Positive control MS2 Ct≤ 32 **Positive**: Two or more targets are positive Ct≤37 and MS2 negative or positive **Negative**: no signal of all targets and MS2 positive **Invalid**: no signal of all targets and MS2 **Inconclusive**: Only one target is positive Ct≤37 and MS2 negative or positive	3 genes (ORF1ab: FAM, N: VIC and S: ABY) and IC MS2: JUN 0h50 min00s	nasopharyngeal aspirate, NPS, OPS, and BALF	ABI 7500 and 7500 Fast, ABI QuantStudio™ 5 or 7	10 GCE/reaction no cross-reactivity = 10 copies/mL	(TaqPath 2020)
7	FastPlex™ Triplex SARS-CoV-2 Detection Kit (RT-PCR) 96 tests/Kit	Precigenome LLC | Ringwood, USA	Positive control Ct≤ 39 **Positive**: FAM or HEX Ct ≤ 39 and CY5 negative or positive **Negative**: no signal or FAM and HEX Ct >39 and CY5 positive **Invalid**: no signal of FAM, HEX and CY5	2 genes (ORF1ab: FAM and N: HEX) and IC: Cy5. 1h43mns58s	Broncheoalveolar lavage; Throat swab	ABI 7500™; Bio-Rad CFX96™PCR systems	285.7 copies/mL	(PreciGenome 2020)
8	STANDARD M nCoV Real-Time Detection kit	Sd Biosensor, Gyeonggi-do, Korea	Positive control Ct≤ 26 **Positive**: FAM or	2 genes (ORF1ab/RdRp:	NPS, OPS and Sputum	RocheLightCycler® 480, Bio-Rad CFX96™,	250 and 125 copies/mL for upper	(Biosensor 2020)

	96 tests/Kit		JOE/VIC/HEX Ct ≤ 36 and CY5 negative or positive **Negative**: no signal or FAM and JOE/VIC/HEX Ct >36 and CY5 positive **Invalid**: no signal of FAM, JOE/VIC/HEX and CY5 **Presumptive positive**: no signal of FAM with JOE/VIC/HEX Ct ≤ 36 and CY5 negative or positive	FAM and E: JOE (VIC or HEX)) and IC: Cy5 Within 90 min 1h45min50s		ABI 7500 Real-Time PCR	and lower respiratory specimens, respectively; cross-reacts with SARS-CoV-1.	
9	cobas® SARS-CoV-2 Test 192 tests/kit; 480 tests/kit	Roche Molecular Systems, Inc. Highway , USA	**Positive**: Target 1 and target 2 are positive; target 1 is positive and target 2 is negative **Negative**: Target 1 and target 2 are negative **Invalid**: Target 1 and target 2 are invalid **Presumptive positive**: Target 1 is negative and Target 2 is positive	2 genes (ORF-1a and E) SARS-CoV-2 (Target 1) and pan-Sarbecovirus (Target 2) 3.5 hours after loading the sample on the system	NPS and OPS	Cobas 6800/8800	0.007 and 0.004 **TCID50/ mL for SARS-CoV-2 and pan-Sarbecovirus respectively =28 copies/mL	(Roche 2020)
10	Abbott RealTime SARS-CoV-2 Assay 24 tests/kit , 48 tests/kit, 72 tests/kit, 96 tests/kit	Abbott molecular Inc. East Touhy, USA	The Abbott m2000rt instrument automatically reports the results and interpretations on the Abbott m2000rt workstation	2 genes (RdRp and N) 4h00min00s	Nasal swabs, NPS and OPS	Abbott m2000sp automatic extractor and Abbott m2000rt RealTime PCR	100 copies/mL with positive rates ≥ 95%; no cross-reactivity	(Abbott 2020)

11	Xpert® Xpress SARS-CoV-2 10 tests/kit	Cepheid, Sunnyvale, USA	**Positive**: positive N2 and E with negative or positive SPC. **Negative**: negative N2 and E with positive SPC. **Invalid**: negative N2 and E with negative SPC. **Presumptive positive**: negative N2 and positive E with negative or positive SPC	2 genes (N2 and E) and IC: Sample Processing Control (SPC) 0h45min00s	NPS and/or nasal wash/ aspirate specimens	GeneXpert Dx	250 copies/mL; no cross-reactivity	(GeneXpert. 2020)
12	Quick SARS-CoV-2 rRT-PCR Kit 100 tests/kit, 1 000 tests/kit, 10000 tests/kit	Zymo Research Corp, Murphy ,USA	Positive control : Ct of ≤ 40 Ct for all N targets; and Ct ≤ 30 for RNase P **Positive**: HEX Ct ≤ 40 and any Ct for Quasar® 670/Cy5 **Negative**: no signal for HEX and Ct < 40 for Quasar® 670/Cy5 **Invalid**: no signal for HEX and signal undetected or Ct value ≥ 40 for Quasar® 670/Cy5	1 gene (Target 1, 2, and 3 of gene N: HEX) IC: RNase P (Quasar® 670/Cy5) -------	upper respiratory specimens (such as nasal, NPS, mid-turbinate or OPS), and lower respiratory specimens (such as sputum, tracheal aspirates, BALF)	Bio-Rad CFX96, ABI QuantStudio 5 RealTime PCR	83 GEC/Ml (5***GEC/rxn), cross-reacts with some organisms =83 copies/ml	(Zymoresearch 2020)

RdRP: RNA-dependent RNA polymerase; Ct: Cycle threshold is amplification signals
*h=hours; min=minutes; s=seconds. ** TCID: Median Tissue Culture Infectious Dose. One **TCID50**/**mL** (corresponding to 4×10^3 **copies**/**mL**). ***GEC: Genomic Equivalents Copies. One GEC/**ml** is equal to one **copy**/**mL. 1 copy per reaction is equal 100 copies/mL**

Table 3: RT-PCR thermocyclers used for Sars-Cov-2 diagnostic in Burkina Faso.

N°	RT-PCR platform	Manufacturer	Sample capacity/run	Filters available	Thermal uniformity	Detection sensitivity
1	Applied Biosystems™ 7500 Fast Dx Real-Time PCR Instrument	Thermo Fisher Scientific™	Fast 96-well plates (100 µL) or 8-tube strips (100 µL) Optimized for 10 µL reactions	FAM™/SYBR™ Green, VIC™/JOE™, NED™/TAMRA™/Cy® 3, ROX™/Texas Red™ and Cy®5	±1°C	Distinguish between 5,000–10,000 genome equivalents (two-fold copy number difference) with 99.7% confidence
2	LightCycler® 480 System	Roche Molecular Diagnostic	96 or 384 wells	FAM, HEX/VIC, SYBR Green I, LightCycler® Red610, LightCycler® Red640, Cy5 and Cyan 500	±0.1°C No edge effect	detectable single copy gene
3	HUMACycler Real-Time PCR Cycler	HUMAN Gesellschaft für Biochemica und Diagnostica mbH	96-Well	FAM™, SYBR® HEX™, VIC®, TET™, TexRed™, JOE™, ROX™, and CY5™	Cooling/heating rate 4°C/sec	-
4	Abbott m2000 RealTime System (Abbott M2000RT)	Abbott Molecular Inc.	Abbott 96-Well Optical Reaction plate	FAM™, SYBR® Green, VIC®, NED™, TAMRA™, JOE™, ROX™, and CY™5	Thermal block temperature accuracy within ± 0.5°C from setpoint, specified in product requirement	-
5	Cobas® z 480 analyzer	Roche Molecular Diagnostic	96 or 384 wells			
6	AriaMx Real-time PCR System	Agilent Technologies	96 wells, 0.2 mL block: 10–30 µL	SYBR/FAM, HEX, ROX, Cy5, Cy3, ATTO425	Easily maintains within ± 0.4°C or less of target temperature.	Two-fold discrimination in a single cycle with 95% confidence over a wide range of copies.

7	CFX96 Touch Real-Time PCR Detection System	Bio-Rad	Standard 96-well plates, 0.2 ml 8-strip tubes or individual tubes	FAM, SYBR® Green I, VIC, HEX, TET, CAL Gold Fluor 540, ROX, Texas Red, CAL Fluor Red 610, Cy5, Quasar 670, Quasar 705	+/- 0.4°C well-to-well within 10 sec of arrival at 90°C	Detect 1 copy of target sequence in human genomic DNA
8	Stratagene MX3005P™ QPCR System	Agilent Technologies	Standard 96-well plates, 0.2 ml 8-strip tubes or individual tubes	Alexa Fluor® 350, FAM™/SYBR® Green I, TET™, HEX™/JOE™/VIC™, Cy™3, TAMRA™, ROX™/Texas Red®, Cy™5	+/- 0.25°C within 12 seconds at 72°C	-
9	*Applied Biosystems™ QuantStudio 3 and 5 Real-Time PCR Systems	Thermo Fisher Scientific™	96-well 0.1 mL block: 10–30 µL 96-well 0.2 mL block: 10–100 µL 384-well: 5–20 µL (S5)	FAM/SYBR Green, VIC/JOE/HEX/TET, ABY/NED/TAMRA/Cy3, JUN, ROX/Texas Red, Mustang Purple, Cy® 5/LIZ™, Cy®5.5	0.4°C	Detect 1 copy
10	TechnePrimpro 48 Prime Pro 48 Real-time qPCR machine	Techne	48-well block	SYBR®, FAM™, HEX™, ROX™, and Cy®5	±0.1°C across the block ±0.1°C uniformity across the whole block instantly after every temperature change means that any well	Detect 1 copy
11	Veri Q PCR 316	MiCo BioMed	16 wells using 3-5 ul of Sample	FAM, HEX, TEX, CY5	-	-
12	NucliSens Easy Q® Analyzer	Biomerieux	48 well	Nucleic Acid Sequence Based Amplification (NASBA) FAM™ and ROX™	the amplification reaction is isothermal and takes place at 41°C	-
13	GeneXpert	Cepheid	8 cartridges	-	-	-

*: cycler acquired during the COVID-19 pandemic

Conclusion

The COVID-19 has worsened the health situation in Burkina Faso. The peak of the second wave in Burkina Faso began in November 2020, is through January 2021. The case fatality rate contained at 1%. In accordance with international requirements, Burkina Faso has used RT-PCR as the reference technique for the diagnosis of COVID-19. The government and partners of Burkina Faso have made efforts to improve the technical level of laboratories. However, efforts are still needed to provide automated sample analysis equipment that can process a large number of samples daily. Finally, in terms of prevention, vaccines are being administered to the general population. The complete sequencing of the genome remains one of the challenges, because until today no SARS-CoV-2 sequence from Burkina Faso is found in the sequence databases. This could allow the detection of probable variants.

COMPETING INTERESTS

The authors have no conflicts of interest to declare.

AUTHOR'S CONTRIBUTIONS

AAZ conceived the idea for the study. KC carried out the maps; AAZ wrote the manuscript the draft of the manuscript; HGO, TS, TRC, STS, DK, OO, SZ, CD, DZ, BS, ATY and SK revised the manuscript. JS and HGO supervised the study. All authors have read and corrected the manuscript.

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REFERENCES

Abbott. 2020. Abbott RealTime SARS-CoV-2. https://www.molecular.abbott/us/en/products/infectious-disease/RealTime-SARS-CoV-2-Assay (accessed 31 July 2021)

Abu A, Naqvi T, Fatima K, Mohammad T, Fatima U, Singh IK. 2020. Insights into SARS-CoV-2 genome, structure, evolution, pathogenesis and therapies: Structural genomics approach. *Mol. Basis Dis.*, **1866**(10): 165878. DOI: https://doi.org/10.1016/j.bbadis.2020.165878.

Africa CDC. 2021 Coronavirus Disease 2019 (COVID-19) Latest updates on the COVID-19 crisis from Africa CDC. https://africacdc.org/covid-19/ (accessed 31 July 2021)

Ahn D, Shin H, Kim M, Lee S, Kim H, Myoung J, Kim S. 2020. Current Status of Epidemiology, Diagnosis, Therapeutics, and Vaccines for Novel Coronavirus Disease 2019 (COVID-19). *J. Microbiol. Biotechnol.*, **30**(3): 313–324. DOI: https://doi.org/10.4014/jmb.2003.03011

Alanagreh L, Alzoughool F, Atoum M. 2020. The Human Coronavirus Disease COVID-19 : Its Origin , Characteristics , and Insights into Potential Drugs and Its Mechanisms. *Pathogens*, **9**(331) : 1–11. DOI: https://doi.org/10.3390/pathogens9050331

Amir IJ, Lebar Z, Yahyaoui G, Mahmoud M. 2020. Covid-19 : virologie, épidémiologie et diagnostic biologique. *OptionBio*, **31**(619): 15–20 DOI : https://doi.org/10.1016/S0992-5945(20)30178-1.

Andersen KG, Rambaut A, Lipkin WI, Holmes EC, Garry RF. 2020. The proximal origin of SARS-CoV-2. *Nat. Med.*, **26**, 450–452. DOI: https://doi.org/10.1038/s41591-020-0820-9

BGI. 2020 Real-Time Fluorescent RT-PCR Kit for Detecting SARS-CoV-2. https://www.bgi.com/us/sars-cov-2-real-time-fluorescent-rt-pcr-kit-ivd/

Biosensor 2020 STANDARD M nCoV Real-Time Detection kit Instructions for Use. http://www.sdbiosensor.com/product/product_view?product_no=119

Böger B, Fachi MM, Vilhena RO, Cobre AF, Tonin FS, Pontarolo R. 2021. Systematic review with meta-analysis of the accuracy

of diagnostic tests for COVID-19. *Am. J. Infect. Control*, **49**(1): 21–29. DOI: https://doi.org/10.1016/j.ajic.2020.07.011

Broughton J, Deng X, Yu G, Fasching C, Singh Streithorst J, Granados A, Sotomayor-Gonzalez A, Zorn K, Gopez A, Hsu E, Gu W, M, Jasmeetiller S, Pan C-Y, Guevara H, Wadford D, Chen J, Chiu C. 2020. Rapid Detection of 2019 Novel Coronavirus SARS-CoV-2 Using a CRISPR-based DETECTR Lateral Flow Assay. *medRxiv Prepr. Serv. Heal.*, *Sci.*, (415): 1–28. DOI: https://doi.org/10.1101/2020.03.06.20032334

Broughton JP, Deng X, Yu G, Fasching CL, Servellita V, Singh J, Miao X, Streithorst JA, Granados A, Sotomayor-Gonzalez A, Zorn K, Gopez A, Hsu E, Gu W, Miller S, Pan CY, Guevara H, Wadford DA, Chen JS, Chiu CY. 2020. CRISPR–Cas12-based detection of SARS-CoV-2. *Nat. Biotechnol.*, **38**(7): 870–874. DOI: https://doi.org/10.1038/s41587-020-0513-4

Cao C, Cai Z, Xiao X, Rao J, Chen J, Hu N, Yang M, Xing X, Wang Y, Li M, Zhou B, Wang X, Wang J. 2021. The architecture of the SARS-CoV-2 RNA genome inside virion. *Nat. Commun.*, **12**: 3917. DOI: https://doi.org/10.1038/s41467-021-22785-x

Chan JFW, Yuan S, Kok KH, To KKW, Chu H, Yang J, Xing F, Liu J, Yip CCY, Poon RWS, Tsoi HW, Lo SKF, Chan KH, Poon VKM, Chan WM, Ip JD, Cai JP, Cheng VCC, Chen H, Hui CKM, Yuen KY. 2020. A familial cluster of pneumonia associated with the 2019 novel coronavirus indicating person-to-person transmission: a study of a family cluster. *Lancet*, **395**(10223): 514–523. DOI: https://doi.org/10.1016/S0140-6736(20)30154-9

Cheepsattayakorn A, Cheepsattayakorn R, Gorbalenya AE, Baker SC, Baric RS, Groot RJ De, Gulyaeva AA, Haagmans BL, Lauber C, Leontovich AM. 2020. Proximal Origin and Phylogenetic Analysis of COVID-19 (2019-nCoV or SARS-CoV-2). *EC Microbiol.*, **19**: 9–12.

Chotiwan N, Brewster CD, Magalhaes T, Weger-Lucarelli J, Duggal NK, Rückert C, Nguyen C, Luna SMG, Fauver JR, Andre B, Gray M, Iv WCB, Kading RC, Ebel GD, Kuan G, Balmaseda A, Jaenisch T, Marques ETA, Brault AC, Harris E, Foy BD, Quackenbush SL, Perera R, Rovnak J. 2017. Rapid and specific detection of Asian- and African-lineage Zika viruses. *Sci. Transl. Med.*, **9**(388). DOI: https://doi.org/10.1126/scitranslmed.aag0538

CORUS. 2021. Evolution de cas de covid au Burkina. http://www.corus.gov.bf/statistiques (accessed 31 January 2021)

Coulibaly N. 2020. Coronavirus au Burkina : pas de répit dans les labos. https://www.jeuneafrique.com/mag/989050/societe/coronavirus-au-burkina-pas-de-repit-dans-les-labos/ (accessed 31 July 2021)

CSSE. 2021. COVID-19 Dashboard by the Center for Systems Science and Engineering (CSSE) at Johns Hopkins University (JHU). https://www.arcgis.com/apps/dashboards/bda7594740fd40299423467b48e9ecf6 (accessed 31 July 2021)

DaAnGene. 2020. Instructions for Use of Detection Kit for Novel Coronavirus (2019-nCoV) RNA (PCR-Fluorescence Probing). http://www.daangene.com

Dalmat Y-M. 2020. Situation des techniciens de laboratoire au sein de l'hôpital public. *OptionBio* **31**(625): 7. DOI: https://doi.org/10.1016/S0992-5945(20)30249-X

Dzinamarira T, Dzobo M, Chitungo I. 2020. COVID-19: A perspective on Africa's capacity and response. *J. Med. Virol.*, **92**(11): 2465–2472. DOI: https://doi.org/10.1002/jmv.26159

EC. 2020. European Commission (EC): Current performance of COVID‐19 test methods and devices and proposed performance criteria.

https://ec.europa.eu/docsroom/documents/40805.

Falzone L, Gattuso G, Tsatsakis A, Spandidos DA, Libra M. 2021. Current and innovative methods for the diagnosis of COVID‐19 infection (Review). *Int. J. Mol. Med.*, **47**, 100. DOI: https://doi.org/10.3892/ijmm.2021.4933

GeneXpert. 2020. Xpert ® Xpress SARS-CoV-2. https://www.cepheid.com/Package Insert Files/Xpert Xpress SARS-CoV-2 Assay FRENCH Package Insert 302-3787-FR%2C Rev. B.pdf

Goldsmith CS, Tatti KM, Ksiazek TG, Rollin PE, Comer JA, Lee WW, Rota PA, Bankamp B, Bellini WJ, Zaki SR. 2004. Ultrastructural Characterization of SARS Coronavirus. *Emerg. Infect. Dis.*, **10**(2): 320–326. DOI: https://doi.org/10.3201/eid1002.030913

Gorbalenya AE, Baker SC, Baric RS, Groot RJ De, Gulyaeva AA, Haagmans BL, Lauber C, Leontovich AM. 2020. Severe acute respiratory syndrome-related coronavirus: The species and its viruses – a statement of the Coronavirus Study Group. *Biorxiv (Cold Spring Harb. Lab.*, 1–15. DOI: https://doi.org/10.1101/2020.02.07.937862

Huang WE, Lim B, Hsu CC, Xiong D, Wu W, Yu Y, Jia H, Wang Y, Zeng Y, Ji M, Chang H, Zhang X, Wang H, Cui Z. 2020. RT-LAMP for rapid diagnosis of coronavirus SARS-CoV-2. *Microb. Biotechnol.*, **13**(4): 950–961. DOI: https://doi.org/10.1111/1751-7915.13586

Ilhan E, Uysal E. 2020. Evaluation of current diagnostic methods for COVID-19. *APL Bioeng.*, **4**(41506–1). DOI: https://doi.org/10.1063/5.0021554

iMMAP. 2021. COVID-19 situation analysis. Washington, p. Retrieved from https://immap.org/

Jaddaoui I El, Allali M, Raoui S, Sehli S, Habib N, Chaouni B, Idrissi N Al, Benslima N, Maher W, Hamamouch N, Bissati K El, Kasmi S El, Hamdi S, Bakri Y, Nejjari C, Amzazi S. 2021. A Review on Current Diagnostic Techniques for COVID-19. *Expert Rev. Mol. Diagn.*, **21**(2): 141–160. DOI: https://doi.org/10.1080/14737159.2021.1886927

Kim Y, Han M, Kim J, Kwon A, Lee K. 2014. Evaluation of Three Automated Nucleic Acid Extraction Systems for Identification of Respiratory Viruses in Clinical Specimens by Multiplex Real-Time PCR. *Biomed Res. Int.*, **2014**(430650): 8 DOI: https://doi.org/10.1155/2014/430650.

Lai A, Bergna A, Acciarri C, Galli M, Zehender G. 2020. Early phylogenetic estimate of the effective reproduction number of SARS-CoV-2. *J. Med. Virol.*, (**February**): 14–18. DOI: https://doi.org/10.1002/jmv.25723

Li H, Wang X, Liu W, Wei X, Lin W, Li E, Li P, Dong D, Cui L, Hu X, Li B, Ma Y, Zhao X, Liu C, Yuan J. 2015. Survey and visual detection of Zaire ebolavirus in clinical samples targeting the nucleoprotein gene in Sierra Leone. *Front. Microbiol.*, **6**(1332): 1–7. DOI: https://doi.org/10.3389/fmicb.2015.01332

Liferiver. 2020. Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit (Detection for 3 Genes) Instructions for Use. http://www.liferiverbiotech.com/Pages/Product/InstrumentsList.aspx

Ministère de la Santé. 2021. Coronavirus-BF. https://www.sante.gov.bf/corona-virus (accessed 31 July 2021)

PreciGenome. 2020. FastPlex Triplex SARS-CoV-2 detection kit（RT-PCR）. : www.precigenome.com/coronavirus-covid19-pcr-assay Page

Roche. 2020. cobas® SARS-CoV-2 Manual., https://diagnostics.roche.com/global/en/products/params/cobas-sars-cov-2-test.html

Sagna T, Ouedraogo HG, Zouré AA, Zida S, Compaore TR, Kambire D, Soubeiga ST, Ouedraogo O, Zongo D, Tarnagda G, Valea D, Dabiré C, Nikiema AR, Camara M, Kagambega A, Ilboudo AK, Yonli AT, Kouanda S, Simpore J. 2021.

Laboratoire et la pandémie COVID Burkina Le Laboratoire à l'épreuve de la pandémie de la COVID-19 au Burkina Faso: Quels défis pour la régularité de l'offre de diagnostic. The COVID-19 pandemic-proof laboratory in Burkina Faso : What challenges for the. *Rev Mali Infect Microbiol*, **16**, 32.

Sansure. 2020. Novel Coronavirus (2019-nCoV) Nucleic Acid Diagnostic Kit (PCR-Fluorescence Probing). http://eng.sansure.com.cn/index.php?g=&m=article&a=index&id=81

SIG. 2021. INFOS COVID-19. https://www.sig.gov.bf/infos-covid-19 (accessed 31 July 2021)

TaqPath 2020 TaqPath™ COVID‑19 CE‑IVD RT‑PCR Kit INSTRUCTIONS FOR USE. https://assets.thermofisher.com/TFS-Assets/LSG/manuals/MAN0019215_TaqPathCOVID-19_CE-IVD_RT-PCR Kit_IFU.pdf

TIBMolBiol. 2020. TIB MolBiol real-time RT-PCR assay. https://www.tib-molbiol.de/covid-19

WHO. 2020. Laboratory testing for coronavirus disease (COVID-19) in suspected human cases: interim guidance, 11 Setember 2020 World Health Organization (WHO). Retrieved from https://apps.who.int/iris/handle/10665/334254

Wu F, Zhao S, Yu B, Chen YM, Wang W, Song ZG, Hu Y, Tao ZW, Tian JH, Pei YY, Yuan ML, Zhang YL, Dai FH, Liu Y, Wang QM, Zheng JJ, Xu L, Holmes EC, Zhang YZ. 2020. A new coronavirus associated with human respiratory disease in China. *Nature*, **579**(7798): 265–269. DOI: https://doi.org/10.1038/s41586-020-2008-3

Yuan X, Yang C, He Q, Chen J, Yu D, Li J, Zhai S, Qin Z, Du K, Chu Z, Qin P. 2020. Current and Perspective Diagnostic Techniques for COVID-19. *ACS Infect. Dis.*, **6**: 1998–2016. DOI: https://doi.org/10.1021/acsinfecdis.0c00365

Zhang F, Abudayyeh OO, Gootenberg JS, Sciences C, Mathers L. 2020. A protocol for detection of COVID-19 using CRISPR diagnostics. *Bioarchive*, 1–8.

Zymoresearch. 2020. Quick SARS-CoV-2 rRT-PCR Kit intruction manual., https://www.zymoresearch.com/products/quick-sars-cov-2-rrt-pcr-kit