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Biobanking: Strengthening Uganda's Rapid Response to COVID-19 and Other Epidemics

Rogers Kamulegeya,[1–3,i,*] David Patrick Kateete,[1,2,*] Bernard S. Bagaya,[1,2,*] Emmanuel Nasinghe,[1,2,*] Winters Muttamba,[4] Gideon Nsubuga,[1,2,*] Edgar Kigozi,[1,2] Fred Ashaba Katabazi,[1,2] Fred Nakwagala,[5] Sam Kalungi,[5] Josaphat Byamugisha,[3] William Worodria,[5] Rose Magala,[1] Bruce Kirenga,[4] and Moses L. Joloba[1,2]

Introduction: SARS-CoV-2 is a fatal disease of global public health concern. Measures to reduce its spread critically depend on timely and accurate diagnosis of virus-infected individuals. Biobanks can have a pivotal role in elucidating disease etiology, translation, and advancing public health. In this article, we show how a biobank has been a critical resource in the rapid response to coronavirus disease of 2019 (COVID-19) in Uganda.
Materials and Methods: The Integrated Biorepository of H3Africa Uganda established a COVID-19 biobank. Standard Operating Procedures for sample and data collection, sample processing, and storage were developed. An *e-questionnaire* data tool was used to collect sociodemographic factors. Samples were collected at 7-day intervals from patients, analyzed for key parameters, processed, annotated, characterized, and stored at appropriate temperatures.
Results: Stored samples have been used in validation of 17 diagnostic kits, the Cepheid Xpert Xpress SARS-CoV-2 assay, as well as a sample pooling technique for mass screening and polymerase chain reaction assay validation. Kits that passed validation were deployed for mass screening boosting early detection, isolation, and treatment of COVID-19 cases. Also, 10 applications from researchers and biotech companies have been received and approved and 4 grants have been awarded
Conclusion: The CoV-Bank has proven to be an invaluable resource in the fight against the COVID-19 pandemic in Uganda, as samples have been resources in the validation and development of COVID-19 diagnostic tools, which are important in tracing and isolation of infected cases to confront, delay, and stop the spread of the SARS-CoV-2 virus.

Keywords: biobanking, COVID-19, pandemic, diagnostics, rapid response

Background

Biobanking is a rapidly growing field of science that involves collection, processing, storage, and distribution of biospecimens and the associated policies and procedures.[1] Biobanks form an ''unsung'' yet critical component of the substructures for scientific research, industry, and conservation, without which much of the current scientific activity would be impossible.[2] Biobanks, among other applications, can have a pivotal role in elucidating disease etiology, translation, and advancing public health. However, consistency and standardization of methods for collection of biospecimens and associated data must be ensured to minimize false analytical results that can lead to invalid findings.[1]

The coronavirus disease of 2019 (COVID-19) caused by SARS-CoV-2 is a disease of global public health concern, and can be fatal. Being a substantial cause of morbidity and

[1]Department of Immunology and Molecular Biology, School of Biomedical Sciences, Makerere University College of Health Sciences, Kampala, Uganda.
[2]Integrated Biorepository of H3Africa Uganda (IBRH3AU), Department of Immunology and Molecular Biology, School of Biomedical Sciences, Makerere University College of Health Sciences, Kampala, Uganda.
[3]Department of Medicine, Makerere University Hospital, Kampala, Uganda.
[4]Makerere Lung Institute, Makerere University College of Health Sciences, Kampala, Uganda.
[5]Department of Pathology, Mulago National Referral Hospital, Kampala, Uganda.
[i]ORCID ID (https://orcid.org/0000-0001-7972-3647).
*These authors contributed equally to this work.

mortality and increasing the threat of infectious diseases that the world is already battling, COVID-19 has also affected many other sectors, including but not limited to the economy and education. Measures to reduce its spread depend on timely and accurate diagnosis of virus-infected individuals.[3] In addition, researchers are working hard toward the development of therapeutics, vaccines, and diagnostics so as to reduce the spread of the virus and its effects on the operations of the world.

In absence of therapeutics against SARS-CoV-2 infection, behavioral change has been and remains crucial to preventing transmission. Isolation and social distancing measures, including edicts to stay at home, have been implemented across the globe to reduce transmission of the virus, although at a huge cost to individuals and society.[4] Additionally, diagnostics are playing an important role in the containment of COVID-19, enabling the implementation of control measures that limit the spread through case identification, isolation, and contact tracing (i.e., identifying people that may have come into contact with an infected patient).

The trend has not been any different in Uganda since the identification of its first COVID-19 case on March 21st, 2020.[5] Researchers in Uganda have joined the rest of the world in a battle to find fast solutions to the COVID-19 pandemic by undertaking research aiming at understanding the pathogenesis of the virus, immunological responses of the human host, transmission dynamics, development of cheaper and easy-to-use diagnostic assays with shorter turnaround time, vaccines, and development of therapeutics. Biospecimens collected from COVID-19 confirmed cases are a precious and nonrenewable resource for such translational research.

The Integrated Biorepository of H3Africa Uganda (IBRH3AU) is a biobank under the H3Africa Biorepository initiative with the ultimate goal of storing well-characterized and annotated quality biospecimens for use in training and research.[6] In this study, we describe how IBRH3AU responded to the COVID-19 pandemic by setting up a biobank (CoV-Bank) in Uganda for archiving of well-annotated samples from confirmed COVID-19 cases. We describe how the samples turned out to be an invaluable resource in strengthening Uganda's rapid response to COVID-19.

Materials and Methods

CoV-Bank activities involve the collection of extensive questionnaire information, clinical phenotyping, and biological samples from confirmed COVID-19 cases admitted at Mulago National Specialized Hospital in Kampala city, Entebbe Grade B Hospital in Entebbe and Masaka Regional Referral Hospital in Masaka city.

Biosafety

Before inception of the CoV-Bank, the safety of staff had to be ensured. To ensure this, training of staff on safety precautions and development of standard operating procedures (SOPs) on collection, transportation, processing, and storage of COVID-19 samples were developed There are numerous potential biosafety hazards that may be encountered when working with biospecimens. All IBRH3AU personnel were trained on the use of appropriate protective gear, such as hand/arm protection, laboratory coats, and closed-toe shoes, as well as knowledge of decontamination procedures to remove and minimize the effect of biohazardous materials. All training was in line with biosafety guidelines provided by the World Health Organization (WHO) and U.S. Centers for Disease Control and Prevention (CDC) on dealing with COVID-19 patients and SARS-CoV-2 samples.[7,8] Biosafety information documents for all materials used in the biobank were made readily available to all biobank personnel. These include the Material Safety Data Sheets (MSDS), which contain health hazard information for chemicals and infectious agents, and requirements for when to use chemical fume hoods and biosafety cabinets.

Standard operating procedures

A variety of SOPs were developed following the International Society for Biological and Environmental Repositories (ISBER) best practices for repositories,[9] WHO and CDC guidelines for laboratories testing for SARS-CoV-2 infection.[7,8] SOPs developed include: COVID-19 sample collection and transportation, isolation of plasma and serum from blood of COVID-19 case, isolation of peripheral blood mononuclear cells from blood of COVID-19 cases, urine processing, stool processing, saliva processing, and nasopharyngeal and oropharyngeal swab processing.

Data collection

We developed data collection tools on REDCAP, a computer-based system to facilitate data collection, tracked participant data (and linked samples) throughout their stay at the hospital, and minimized manual data entry. The collected data were consolidated in the study database at IBRH3AU. Data were collected using an *e-questionnaire*, which captured sociodemographic factors, occupational information, marital status, education background, travel history, and current and past health conditions. Interviews were administered by a trained nurse.

Biological material collected

Participants provided samples of nasopharyngeal and/or oropharyngeal swabs, saliva, stool, blood, and urine at different visits (Table 1). A proportion of the blood and urine collected was used for hematologic and chemistry analyses at Makerere University Hospital (Table 2). The remainder, plus the urine, saliva, swabs, and stool were processed, aliquoted, and stored in the biobank. Table 3 summarizes derivatives from different sample types and aliquots in storage.

Biospecimen processing and quality assurance

Processing and analysis of the biospecimens was standardized and centralized in a Biosafety Level 3 (BSL-3)-processing laboratory at IBRH3AU with the aim of increasing robustness of the data trail, reducing costs and increasing achievable throughput and accuracy of sample handling by avoiding nondetectable systematic errors that may be introduced by processing at multiple sites. This limited the impact of analytical variability and improved the power of subsequent analyses in which data derived from samples were used.[10] In addition, the laboratory carries

Table 1. Summary of Sample Types, Volumes, and Collection Schedule

Visit	*Visit 1*	*Visit 2*	*Visit 3*	*Visit 4*	*Visit 5*	*Visit 6*	
Week	0	1	2	3	4		
Day	0	7	14	21	Discharge	14 days after discharge	Post-mortem
Blood (mL)	30	10	30	10	30	30	
Saliva (mL)	≤20	≤20	≤20	≤20	≤20	≤20	
Urine (mL)	≤20	≤20			≤20	≤20	
Swab	2	2	2	2	2	2	
Stool (g)	≤10				≤10	≤10	
Lung and liver tissue (g)							~50

out interlaboratory comparisons for all routine tests, including polymerase chain reaction (PCR) tests once every month as a quality assurance measure. Quality assurance for SARS COV-2 PCR tests has also been ensured through participation in the External Quality Assurance programs of the Central Public Health Laboratory (CPHL) of Uganda, World Health Organization (2020 WHO International PTP for the Detection of SARS-CoV-2 by PCR), and One World Accuracy (https://results.oneworldaccuracy.com/results/catalog/browse?cm=CEQAL&y=2021&l=en)

All biospecimens are backed up internally by maintaining running freezer space and by splitting aliquots stored in other freezers, and the biobank maintains an up-to-date agreement with an off-site laboratory.

The entire facility runs on the national electricity grid, backed up by three well-maintained 250 KVA generators connected in series that are monitored for fuel reserves to run for up to 72 hours in case of power failure. All equipments are covered by a facility and equipment maintenance and service contract.

Table 2. Routine and Nonroutine Tests Done on Blood and Urine Samples

Group	*Variable*	*Group*	*Variable*	*Group*	*Variable*
Routine tests					
Differential white cell count	Basophil	Electrolytes and renal function tests	Chloride	Full blood count	Hematocrit
	Basophil %		Serum creatinine		Hemoglobin
	Eosinophils		Bicarbonate		Mean corpuscular hemoglobin
	Eosinophils %		Potassium		Mean corpuscular HGB concentration
	Lymphocytes		Sodium		Mean corpuscular volume
	Lymphocytes %		Urea nitrogen		Mean platelet volume
	Monocyte	Liver function tests	Albumin		Platelets
	Monocyte %		Alkaline phosphatase		Red blood cell
	Neutrophils		Alanine transaminase		C-Peptide
	Neutrophils %		Aspartate transaminase		
	White blood cell		Gamma glutamyl transferase		
			Total bilirubin		
			Total protein		
Nonroutine tests					
Diabetes-related tests	Glucose	Lipid profile	High-density lipoprotein		
	Glycated Hemoglobin A1c %		Low-density lipoprotein		
	Insulin		Triglycerides		
Thyroid function tests	Free triiodothyronine		Cholesterol		
	Free thyroxine	Other tests	Procalcitonin		
	Thyroid-stimulating hormone		D-Dimer		

HGB, hemoglobin.

Table 3. Products of Sample Processing and Aliquots Archived from Different Sample Types

Specimen/specimen collection tube	*Fraction*	*No. of aliquots*	
		–80°C	*Liquid N_2*
EDTA tube	Plasma	6	—
EDTA tube	Hematology (Immediate)	—	—
Silica clot activator (SST)	Serum	6	—
Silica clot activator (SST)	Chemistry (Immediate)	—	—
Lithium heparin (PST)	PBMCs	—	6
Blood RNA PAXgene tube	Whole blood (RNA)	1	—
Saliva	Mixed saliva sample	2	—
Stool	Stool	2 or 3	—
Nasopharyngeal swab	Nasopharyngeal swab	2	—
Urine	Urine	4	—
Total aliquots		21	6

EDTA, ethylenediamine tetraacetic acid; PBMCs, peripheral blood mononuclear cells; PST, plasma separator tube; SST, serum separator tube.

Results

Baseline characteristics of the participants

Participants were recruited from three COVID-19 treatment centers in referral hospitals in Kampala, Entebbe, and Masaka. The participants' age ranged from 7 to 74 years with a median age of 33 years; 83.4% of the participants were men, 60.23% symptomatic, and 14.02% asymptomatic. Fourteen percent of the participants had comorbidities that included HIV infection, diabetes mellitus, asthma, and hypertension, as shown in Table 4.

Table 4. Baseline Sociodemographic Characteristics of Participants in the COV-BANK Study (March 2020 to June 2020, *N*=435)

Characteristics	*Male (N=363) (%)*	*Female (N=72) (%)*	p
Age			0.069
<25	66 (15.2)	22 (5.1)	
25–34	121 (27.8)	25 (5.8)	
35–44	106 (24.4)	14 (3.2)	
≥45	69 (15.9)	11 (2.5)	
Marital status			0.000
Single	86 (20.1)	27 (6.3)	
Married	251 (58.6)	32 (7.5)	
Separated	10 (2.3)	7 (1.6)	
Level of education			0.004
Primary education or less	126 (31.2)	24 (6.0)	
Secondary level education	143 (35.4)	18(4.5)	
University or tertiary education	69 (17.1)	24 (6.0)	
Comorbidities			
Diabetes	6 (1.4)	3 (2.5)	0.171
HIV	15 (3.5)	5 (1.2)	0.308
Asthma	3 (0.7)	2 (0.46)	0.154
Hypertension	23 (5.5)	4 (0.96)	0.913
Presenting symptom			
Fever	44 (10.25)	11 (2.6)	0.67
Cough	66 (15.35)	16 (3.7)	0.59
Runny nose	61 (14.19)	17 (3.95)	0.01
Sore throat	15 (3.5)	9 (2.10)	0.35
Chest pain	12 (2.79)	6 (1.40)	0.15
Short-breath	3 (0.7)	2 (0.5)	0.23

Biospecimen utilization

Validation of Xpert Xpress SARS-CoV-2 test for use in Uganda. The Xpert Xpress SARS-CoV-2 test by Cepheid is a rapid, real-time reverse transcription (RT)-PCR test for the qualitative detection of SARS-CoV-2 ribonucleic acids in specimens like nasopharyngeal/oropharyngeal, nasal, or mid-turbinate swabs, and/or nasal wash/aspirates collected from individuals suspected of having COVID-19.[11] Emergency use authorization was issued to Cepheid for the Xpert Xpress SARS-CoV-2 test for use in high- and moderate-complexity CLIA-certified laboratories as well as in certain patient care settings.[12]

The Xpert Xpress SARS-CoV-2 was evaluated against the commercial real-time RT-PCR assays using nasopharyngeal swabs of COVID-19 confirmed cases archived in the IBRH3AU. The results from the evaluation informed the deployment of Xpert Xpress SARS-CoV-2 for use as point-of-care (POC) test in Uganda.

Validation of serology and antigen test kits. The Ministry of Health in Uganda received a donation of over 7000 SARS-CoV-2 Antibody and Antigen Rapid Diagnostic Testing Kits.[13] These kits were validated using serum and nasopharyngeal swab samples in the CoV-Bank. The performance of the kits is summarized in Tables 5. These data were made available to the Ministry of Health and will guide the deployment of serology tests to complement COVID-19 testing in Uganda. Notably, the antigen kit

Table 5. Sensitivity, Specificity, Positive and Negative Predictive Values of Serology Kits

RDT Kit Brand	*Sensitivity (%)*	*Specificity (%)*	*PPV (%)*	*NPV (%)*
KIT A	56.67	90.00	85.00	67.50
KIT B	52.00	91.00	30.77	55.00
KIT C	66.67	53.33	58.80	61.53
KIT D	40.00	80.00	66.70	57.14
KIT E	90.00	93.33	93.00	90.32
KIT F	76.67	93.33	92.00	80
KIT G	50	86.67	78.95	63.41
KIT I	13.33	100	100	53.57
KIT J	87.50	92.90	93.30	86.70
KIT K	56.67	100.00	100.00	69.77

NPV, negative predictive value; PPV, positive predictive value.

showed a sensitivity of 84% and a specificity of 100% and was authorized for use in Uganda by the Ministry of Health following validation of the results by the CoV-Bank.

Serology tests detect the presence of antibodies in blood when the body is responding to a specific infection, like SARS-CoV-2. These tests could play a role in the fight against COVID-19 by guiding health care professionals in identifying individuals who may have developed an immune response to SARS-CoV-2 infection. Additionally, these test results could aid in determining individuals who qualify to donate a part of their blood called convalescent plasma, which may serve as a treatment option for severe COVID-19 cases. However, proper use of serology tests depends on an understanding of their performance characteristics and limitations, assessed from their ''sensitivity,'' that is, the ability of the test to identify individuals with antibodies to SARS-CoV-2 (true positives), and their ''specificity,'' that is, the ability of the test to identify individuals without antibodies to SARS-CoV-2 (true negatives).

COVID-19 antigen/antibody test development. Molecular tests detecting viral RNA are the main diagnostics for coronaviruses like SARS-CoV-2 and are an essential part of contact tracing. However, in many low-income settings, molecular testing is challenging due to limited access to specialized testing laboratories and an inadequate supply of reagents due to huge demand worldwide. In such cases, rapid serology tests offer a needed additional option.[14] As such, medical diagnostic companies and laboratories are battling to produce antibody and antigen testing kits, to assess not only current viral infection, but also immunity to severe SARS-CoV-2 infection. These tests require knowledge of the proteins that form the viral coat—specifically, those proteins to which the immune system responds, triggering the production of antibodies that flag or neutralize the virus.[15]

In Uganda, three biotech companies have shown interest in the development of SARS-CoV-2 antibody testing kits. These companies requested us to validate their products, and the CoV-Bank provided access to swab samples from individuals who were positive for SARS-CoV-2 infection. It is noteworthy that in developing countries like Uganda, close collaboration between industry and the academia is lacking, yet it is necessary to expedite translation of research findings into diagnostics, especially POC tools. Therefore, by rapidly supporting biotechnology companies, IBRH3AU, through the CoV-Bank, has demonstrated the capacity to work closely with the local biotech sector in the development of technologies to combat COVID-19 and other epidemics in the future.[16]

Discussion

The CoV-Bank has proved to be an invaluable resource in the fight against the COVID-19 pandemic in Uganda as samples in storage have been significant resources in the validation and development of COVID-19 diagnostic tools, which are critical for tracing and isolation of infected cases to confront, delay, and stop the spread of the SARS-CoV-2 virus. Further still, it is important to recognize that we still have a long way to go regarding our understanding of the SARS COV-2 virus, and proper sample storage can expedite this path to discovery. This approach however, comes with challenges, beginning from participant recruitment, sample collection, analysis, and proper storage for advanced use. This major challenge/difficulty that this study explored was participant recruitment. Being a new disease, COVID-19 has been surrounded with a lot of stigma. Worse still, most of the first SAR COV-2-positive people in Uganda were asymptomatic. This made it difficult for the study to recruit participants as most of them doubted the diagnosis. As such, the partnering medical facilities, where participants were recruited from, offered social workers and medical doctors who explained the diagnosis to the patients, since these facilities were also eager to support any solution.

Conclusion

Disease control and prevention over the years have depended on data sharing and commitment toward collaborative research. However, the sharing of well-characterized specimens remains a major challenge for accelerating test development and evaluation.

During epidemics and pandemics like COVID-19, establishment of a sustainable biobanking mechanism for the standardized collection, characterization, and archiving of specimens, and sharing of these specimens to facilitate diagnostic testing, therapeutics development, and knowledge discovery is necessary. As the number of COVID-19 cases and fatalities continue to mount in Uganda, CoV-Bank will continue to support local and international researchers as they respond to the pandemic.

Ethical Approval

IBRH3AU has ethics approval from Makerere University School of Biomedical Sciences Research and Ethics Committee (SBS-REC) and from the Uganda National Council for Science and Technology (UNCST) to collect, process, store, and share biospecimens. Further still, IBRH3AU obtained ethics approval from the Mulago National Hospital REC and UNCST to collect biospecimens from all COVID-19 cases receiving care from various treatment centers under the Uganda's Ministry of Health. Participants consented to sample storage and use of their samples and data in current and future studies related to SARS-CoV-2 infection.

Consent for Publication

Not applicable.

Availability of Data and Materials

The datasets used and/or analyzed during the current study are available from the corresponding authors on reasonable request.

Authors' Contributions

R.K., E.N., and G.N. worked with medical teams at the three hospitals to ensure ethical guidelines are followed during sample collection and that quality samples are collected (under supervision of D.P.K., M.L.J., B.S.B., F.N., J.B., and B.K.) and wrote the first draft of the article. R.K, E.N., J.B., and G.N. did sample and data analysis. W.M., B.K., and W.W. provided e-tools for data collection. F.N. and M.L.J. gave ethical guidance. All authors read and approved the final article.

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Author Disclosure Statement

No conflicting financial interests exist.

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Address correspondence to:
Rogers Kamulegeya, BSc, MSc
Department of Immunology and Molecular Biology
School of Biomedical Sciences
Makerere University College of Health Sciences
Mulago Hill Road
Pathology building, Level 3, Rm 1
Box 7072
Kampala
Uganda

E-mail: vicrogerbre@gmail.com

David Patrick Kateete, BVM, MSc, PhD
Department of Immunology and Molecular Biology
School of Biomedical Sciences
Makerere University College of Health Sciences
Mulago Hill Road
Pathology building, Level 3, Rm 1
Box 7072
Kampala
Uganda

E-mail: dkateete@chs.mak.ac.ug

Moses L. Joloba, MD, MSc, PhD
Department of Immunology and Molecular Biology
School of Biomedical Sciences
Makerere University College of Health Sciences
Mulago Hill Road
Pathology building, Level 3, Rm 1
Box 7072
Kampala
Uganda

E-mail: moses.joloba@case.edu