AFRICA UNIVERSITY
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DEVELOPING A MULTIPLEXED 3D-PRINTED PROTOTYPE POINT-OF-CARE TESTING DEVICE FOR HIV AND HEPATITIS C IN HIGH-RISK POPULATIONS

BY

STANFORD CHIGARO

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Abstract

The manufacture of point-of-care testing devices for key diseases such as HIV and Hepatitis C remains neglected in sub-Saharan Africa despite the critical need for the point-of-care devices. This research project provides initial steps in the development of a lateral flow immunoassay-based multiplexed point-of-care device (HIV/HCV multiplexed point-of-care device) that can detect co-occurrence of HIV and the hepatitis C virus. Additionally, the project provides insights on the perspectives of healthcare professionals on multiplexed point-of-care devices. The study further reports on the market analysis of the point-of-care devices industry, assessing the market size, the customer segments, competition, and barriers to market entry. Data for understanding the healthcare professionals' perspectives on multiplexed point-of-care was collected through a survey with 21 Bambanani Newstart Centre healthcare professionals. The market analysis was conducted using secondary data, and laboratory experiments were utilised in the development of the HIV/HCV multiplexed point-of-care device. Findings from the survey and the market analysis, along with a laboratory protocol manual guided the development of the HIV/HCV multiplexed point-of-care device. The study found that healthcare professionals at Bambanani Newstart Centre have an unmet need for multiplexed point-of-care devices. The respondents highlighted that multiplexed point-of-care devices improve patient management; ensure targeted treatment; offer a cheaper diagnostic option; ensure diagnostic certainty; save time for diagnosis and reduce the need to refer a patient to the laboratory. The respondents cited the shortage of multiplexed point-of-care devices as the biggest challenge in their routine work. The market analysis reviewed that multiplexed point-of-care devices have a ready market in sub-Saharan Africa. The global point-of-care device industry is expected to grow at a compound annual growth rate of 11.1% to reach US$93.2 billion by 2030, with Africa providing the largest market for infectious diseases testing devices. There is however a limited supply of HIV/HCV multiplexed point-of-care devices in sub-Saharan Africa. The available multiplexed point-of-care devices are manufactured outside the African region, mainly in the United States of America, Europe and China. The market analysis cited competition from imported point-of-care devices as a major barrier. Access to distribution channels is another barriers the HIV/HCV multiplexed point-of-care device may face. Despite the development of the HIV/HCV multiplexed point-of-care device for this project being still in progress, the mode of multiplexing, and the capture and detection antibody concentrations have been determined. The device is composed of multiple strips supplied by a single sample stream. The antibody concentration of 0.5 μg/ml was selected for HIV capture antibody, Hepatitis C capture antibody and Hepatitis C detection antibody. The concentration of 1 μg/ml was selected for HIV detection antibody. The researcher however recommends further research with a larger sample size to understand the perspectives of healthcare professionals. Additionally, collaboration among industry partners and healthcare professionals is essential in order to promote local innovation.

Keywords: Point-of-care devices; Sub-Saharan Africa; Lateral flow immunoassay; Healthcare professionals; Market analysis

Declaration Page

I declare that this dissertation is my original work except where sources have been cited and acknowledged. The work has never been submitted, nor will it be submitted to another university for the award of a degree.

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Student's Full Name	Student's Signature (Date)
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List of Acronyms and Abbreviations

ART	Antiretroviral Therapy
ASSURED	Affordable, Sensitive, Specific, User-friendly, Rapid and Robust, Equipment free, Delivered
CAGR	Compound Annual Growth Rate
COVID-19	Coronavirus Disease 2019
ELISA	Enzyme-linked Immunosorbent Assay
HBsAG	Hepatitis B Surface Antigen
HCV	Hepatitis C Virus
HIV	Human Immunodeficiency Virus
LFIA	Lateral Flow Immunoassay
Mlsi	Microfluidic Large-Scale Integrations
mPADs	Microfluidic Paper-Based Analytical Devices
PhD	Doctor of Philosophy
PLWHA	People Living With HIV/AIDS
POC	Point-of-Care
PT	Prototype
PrEP	Pre-exposure Prophylaxis
SSA	Sub-Saharan Africa

STI	Sexually Transmitted Infections
3D	3-Dimensional
TUT	Tshwane University of Technology
USA	United States of America
US$	United States Dollar
VMMC	Voluntary Medical Male Circumcision
WHO	World Health Organisation

Table of Contents

List of Tables

List of Figures

List of Appendices

CHAPTER 1: INTRODUCTION

1.1 Introduction

The purpose of this study was to develop a lateral flow immunoassay (LFIA)-based multiplexed point-of-care (POC) device that can detect co-occurrence of the human immunodeficiency virus (HIV) and hepatitis C virus (HCV). Sub-Saharan Africa (SSA) experiences a huge burden of both HIV and HCV among other infectious diseases. Although effective treatments have existed for a number of years, HCV causes 400,000 deaths a year in people living with HIV/AIDS (PLWHA) in SSA, due to low numbers diagnosed and put on treatment (Leidner *et al*., 2015). Limited access to diagnostic testing due to various reasons that include high cost and scarcity of the services has been blamed for the HCV public health problem in the sub-Saharan region (Sonderup *et al*., 2017).

The manufacture of POC devices for key diseases such as HIV and HCV remains neglected in SSA. Most POC devices for the detection of HIV and HCV currently in the African market are manufactured in the United States of America (USA), China, and Europe where the combined market size reached US$32.5 billion in 2020 (Nichols, 2021). The World Health Organisation [WHO] (2020) notes that only 20% of HCV-infected persons have been diagnosed. Given that the WHO Afro region has 69.6% of the global 36.9 million population living with HIV (Karoney & Siika, 2013), Africa has a ready market for HIV/HCV multiplexed POC devices. Multiplexed POC devices can therefore be expected to address the disparity in the diagnosis of HIV and HCV co-infection, improve mass HCV screening and set a platform for locally produced diagnostics for other diseases.

Multiplexed POC testing devices have recently gained increasing importance for clinical diagnostics, with emerging applications in limited-resource settings such as in the developing world. (Dincer, Bruch, Kling, Dittrich & Urban, 2017). Teebagy *et al*. (2022) discovered that healthcare professionals accept and have an unmet need for POC testing devices. Healthcare professionals agree that an early and accurate diagnosis of a specific disease plays a decisive role for its effective treatment, especially at the point-of-care (Jung, Han, Choi & Ahn, 2015). However, in many instances clinical evidence based on a single biomarker is not adequate for an appropriate diagnosis of a disease. Multiplexed POC testing devices close this gap by detecting multiple infections resulting in appropriate and effective treatment being offered.

The most popular multiplexed POC testing devices currently in the market are based on the LFIA technology (Kim, Chungu & Kang, 2019). LFIA-based POC devices detect multiple target analytes from a sample stream, using nano-sized particles immobilized on a single strip or multiple strips. Although there are various techniques that can be adopted, the use of multiple strips has proved to be more practical for the detection of multiple pathogens (Hanbi & Doo-Ryeon, 2019). There are however numerous gaps in the development of multiplexed POC devices which this study tried to address.

This chapter begins with the background that frames the study. Following this is the statement of the problem, objectives of the study and significance of performing this project. The chapter concludes with the delimitations and limitations of the study.

1.2 Background to the Study

Hepatitis C is a viral infection that causes liver inflammation and is known to share transmission routes with HIV (Leoni, Ustianowski, Farooq & Arendes, 2018). The severity of HCV ranges from mild, acute illness to lifelong disease that may lead to active liver disease, cirrhosis, liver cancer and death (Mora *et al*., 2016). WHO (2020) reported that SSA has the highest prevalence of both HCV and HIV in the world, and many people living with HCV in the region are unaware of their status. Kedar *et al.* (2021) further indicated that there is likely to be a high incidence of advanced liver disease in SSA due to the high prevalence of HCV.

Multiplexed POC testing technologies with capacity to simultaneously detect multiple infectious diseases, are needed in limited-resource SSA (Drain, & Rousseau, 2017). A study by Lanini, Easterbrook, Zumla, & Ippolito, (2016) revealed that nearly 70% of the people living with HIV globally are in the Afro region. The study further indicated that of the 71 million people infected with HCV worldwide, more than 80% of the burden is in low or middle-income countries. Despite HCV high prevalence, only 20% of HCV-infected persons have been diagnosed and only 7% have received treatment worldwide (Lohia, 2020). Affordable technologies for concurrent detection of these infectious diseases are therefore invaluable in eradicating infectious diseases.

Multiplexed POC devices based on various forms of LFIA are the most popular diagnostic tool for rapid diagnosis of multiple infections (Kim *et al*., 2019). Ideally, multiplexed LFIA are based on detecting multiple target analytes from a sample stream, using nano-sized particles immobilized on a single (single lateral flow immunoassay) or multiple

strips (multiplex lateral flow immunoassay). The latter technique of using multiple strips has proved to be more practical, allowing multiple conjugates for the detection of up to ten pathogens (Hanbi & Doo-Ryeon, 2019).

Multiplexed POC devices based on LFIA technology are suitable for limited-resource settings due to the low cost of production and simplicity in using the devices (Anfossi, Di Nardo, Cavalera, Giovannoli, & Baggiani, 2018). Lanini *et al*. (2016) noted that co-occurrence of infectious diseases, often affecting the effectiveness of health interventions, necessitates concerted efforts on multiplexed POC devices. However efforts to find effective multiplexed POC devices remains a technically challenging front, particularly because the devices have to be "Affordable, Sensitive, Specific, User-friendly, Rapid and Robust, Equipment free, Delivered" (ASSURED) as per the WHO guidelines on POC devices (Reipold *et al*., 2017).

This project develops a LFIA-based multiplexed POC prototype device that can detect co-occurrence of HIV and HCV, with the primary target population for HCV diagnosis being PLWHA. The development of the HIV/HCV multiplexed POC device was informed by the perspectives of healthcare professionals on multiplexed POC devices, and the results from the market analysis on the global and regional POC industry.

1.3 Statement of the Problem

Sub-Saharan Africa experiences a huge burden of both HIV and HCV. Although effective treatments have existed for a number of years, HCV causes 400,000 deaths a year in PLWHA in SSA, due to low numbers diagnosed and put on treatment (Leidner *et al.*, 2015). This is despite the fact that HCV is known to share transmission routes with HIV.

Risk-based screening for HCV infection, as recommended by WHO, is non-existent in most sub-Saharan countries with inaccessibility of diagnostic testing being the major challenge. (Sonderup *et al*., 2017). More affordable technologies like multiplexed POC devices that concurrently detect HIV and HCV co-infections are not readily available in SSA and this makes mass screening difficult, delaying diagnosis and treatment. This project therefore looks at developing an HIV/HCV multiplexed POC prototype device.

1.4 Objectives of the Study

1.4.1 General Objective

The purpose of this study was to understand the perspectives of healthcare professionals on multiplexed point-of-care testing; conduct a market analysis on the point-of-care industry; and to develop a multiplexed 3D-printed prototype point-of-care testing device for detecting co-occurrence of HIV and HCV in high-risk populations.

1.4.2 Specific Objectives

I. To determine healthcare professionals' perspectives on multiplexed point-of-care testing devices at a key population clinic in Bulawayo City, Zimbabwe.

II. To conduct a market analysis for HIV and hepatitis C point-of-care devices for the period 2020 to 2030, assessing size of the market, the customer segments, competition, and barriers to market entry.

III. To develop a multiplexed point-of-care prototype device based on lateral flow immunoassay technology that detects co-occurrence of HIV and Hepatitis C.

1.5 Research Questions

I. What are the current perspectives of healthcare professionals towards multiplexed point-of-care testing devices?

II. What is the demand and supply situation of HIV and hepatitis C point-of-care devices in sub-Saharan Africa?

III. What is the optimal design and specifications for the proposed multiplexed point-of-care device based on lateral flow immunoassay technology that can be adopted?

1.6 Significance of the Study

The HIV/HCV multiplexed POC technology seeks to address a gap in the simultaneous determination of HIV and HCV in poorly resourced settings in SSA. A huge social disparity exists in terms of access to testing technologies, as experienced during the current COVID-19 pandemic. Many people especially in poor environments are unable to access essential diagnostic testing. The full scale development of this innovation will therefore provide an affordable multiplex option for simultaneously diagnosing and monitoring HIV and HCV at a low cost in under-resourced settings. The device is also expected to provide a baseline platform for the local production of multifunctional diagnostic platforms for other diseases affecting the African continent.

1.7 Delimitation of the Study

Despite the availability of other technologies that can be adopted on POC devices, this project focused on developing a multiplexed POC device based on LFIA. The multiplexed POC device is developed according to the specifications of the grant. The primary target

market for the multiplexed POC device is limited-resource SSA where the full-scale device is expected to improve the screening and prevention of HIV and HCV.

1.8 Chapter Summary

In summary, SSA faces the burden of HIV and HCV, but the majority of the population has limited access to diagnostic testing due to various reasons that include high cost and scarcity of the services. Despite HIV testing having been ramped up in most sub-Saharan countries, HCV diagnosis still lags behind. A multiplexed POC device with capacity to detect HIV and HCV is required to reduce the disparity in the detection of these co-infections. While there are a few HIV/HCV multiplexed devices already available in the market, these are mostly expensive hence cannot be adopted for mass testing. Local manufacturing provides a more affordable option. Therefore this study explores the development of a HIV/HCV multiplexed POC device applicable in limited-resource SSA.

CHAPTER 2: REVIEW OF RELATED LITERATURE

2.1 Introduction

Heidt *et al*. (2020) noted that the inadequacy of POC testing devices for HCV infections in developing countries puts HCV on the list of neglected infectious diseases. Experts agree that the global POC industry, is set to grow, but the shortage of POC devices for HCV is likely to worsen (Nichols, 2021; Tonen-Wolyec *et al*., 2021) Efforts to innovate and manufacture multiplexed POC devices for HIV and HCV addresses a critical need in SSA and other regions with a similar problem.

Since the purpose of this project is to develop a multiplexed POC testing device that detects co-occurrence of HIV and HCV, and specifically targeting the African market, it is necessary to complete a critical review of current literature on the subject. This critical review discuss the perspectives of healthcare professionals towards POC devices; explores the global POC market, and the technology and concepts of multiplexed POC devices. Emphasis is put on the hurdles around the manufacture of different forms of LFIA-based multiplexed POC devices.

To conduct this selected literature review, the researchers used multiple information sources, including books, dissertations, internet resources, professional journals, and periodicals. Internet sources were accessed through PubMed central and google scholar. Throughout the review, the study attempted to point out important gaps and omissions in particular segments of the literature as and when they became apparent.

2.2 Perspectives of Healthcare Professionals on Multiplexed Point-of-Care Testing Devices

The COVID-19 pandemic, emergence of infections and lack of diagnostic resources in SSA led to the implementation of non-pharmaceutical healthcare interventions, such as telemedicine. This shift has created an interest in the use of POC testing devices. (Ding *et al*. 2019). Furthermore, even before the COVID-19 pandemic, the advantages of expanding the role of patients in the management of their own health through the use of POC devices were widely recognized. Of note in the United States of America (USA), a significant number of people care for the aging members of the community with the use of POC testing devices. POC devices provide an opportunity for a more patient-centered approach, and have the potential to expedite the diagnosis. Cost of patient management is also reduced through low transportation cost (Ding, Marcus & McManus, 2020).

While previous studies have highlighted the benefits of POC devices, little is known about the adoption of multiplexed POC devices by healthcare providers in Zimbabwe. Dunlap *et al*. (2021) argue that it is crucial to understand the perceived utility and adoption of the POC technology by healthcare professionals in order to increase the successful implementation and use. In a study conducted in the USA, Teebagy *et al*. (2022) perceived an unmet need for POC testing devices among the healthcare workers involved in the survey. Most respondents described POC testing devices as an efficient and cheaper approach to promote health and wellbeing. However Reipold *et al*., (2017) argue that only POC devices satisfying the ASSURED rigmarole are acceptable.

2.3 Current State of the Global Point-of-Care Devices Industry

Globally, the demand for POC testing devices continue to grow (Kuupiel, Bawontuo, Drain, Gwala & Mashamba-Thompson, 2019). Of note, as of 2021, the global POC industry was worth US$35 billion and is expected to reach US$81.4 billion in 2028 (Nichols, 2021). Despite the fact that the global POC industry, led by COVID-19 and HIV, is set for a lucrative growth phase in 2022, the gap for HIV/HCV POC devices in developing countries including SSA is likely to worsen. Efforts to innovate and manufacture multiplexed POC devices for HIV and HCV addresses a critical need in SSA and other regions with a similar problem.

The glucose monitoring kit segment currently dominates in the global POC diagnostics market (Di Nardo, Chiarello, Cavalera, Baggiani & Anfossi, 2021). However, the increase in prevalence of chronic and infectious diseases such as cardiac diseases, hepatitis and sexually transmitted infections like HIV has played a pivotal role in the growth of the global POC diagnostic market share. Changes in lifestyle patterns have also caused several medical conditions such as diabetes that require POC testing devices (Nichols, 2021).

Despite major companies initiating strategic product launches to strengthen their market presence, the manufacture of POC devices for key diseases such as HCV, remains neglected in SSA (Webster, Klenerman & Dusheiko, 2015). Companies such as Abbott and Cepheid are expanding their market share but with little focus on HCV testing devices. This is despite the fact that only 19 percent of the people infected with HCV know their status (Lohia, 2021). There is still a need to manufacture HCV POC testing devices especially for limited-resource settings like SSA.

2.4 Multiplexed Point-of-Care Devices: Current Technologies and Concepts

Apparently, to the developed world, the whole point of multiplexed point-of-care testing is personalised therapy whilst in limited-resource settings, improved access to *in vitro* diagnosis of the several endemic infectious diseases such as HIV, hepatitis, tuberculosis and malaria is the focal drive (Dincer *et al*., 2017). Arguably, there are opportunities to leverage the resources available in large-scale screening programmes such as in the 'Test and Treat' programmes for HIV in SSA (Kumar *et al*., 2020).

Dincer *et al.* (2017) indicated that multiplexed devices currently available in the market make use of the following approaches (i) spatial separation of detection sites on paper, (ii) channel network or arrays, (iii) the use of various labels, for example beads, and (iv) microfluidic channel technologies. The last two techniques have been referred to as lab-on-chip technologies due to their capacity to miniaturize sophisticated laboratory procedures onto a tiny chip (Schönberger & Hoffstetter, 2016). However, these technologies are still too expensive for wide use in limited-resource settings. Dincer *et al*. (2017) further noted that paper-based spatial separation technologies, for example in home pregnancy tests, remain suitable in low resource settings but the development of the technology based on LFIAs has been lagging behind in developing countries where they are needed most.

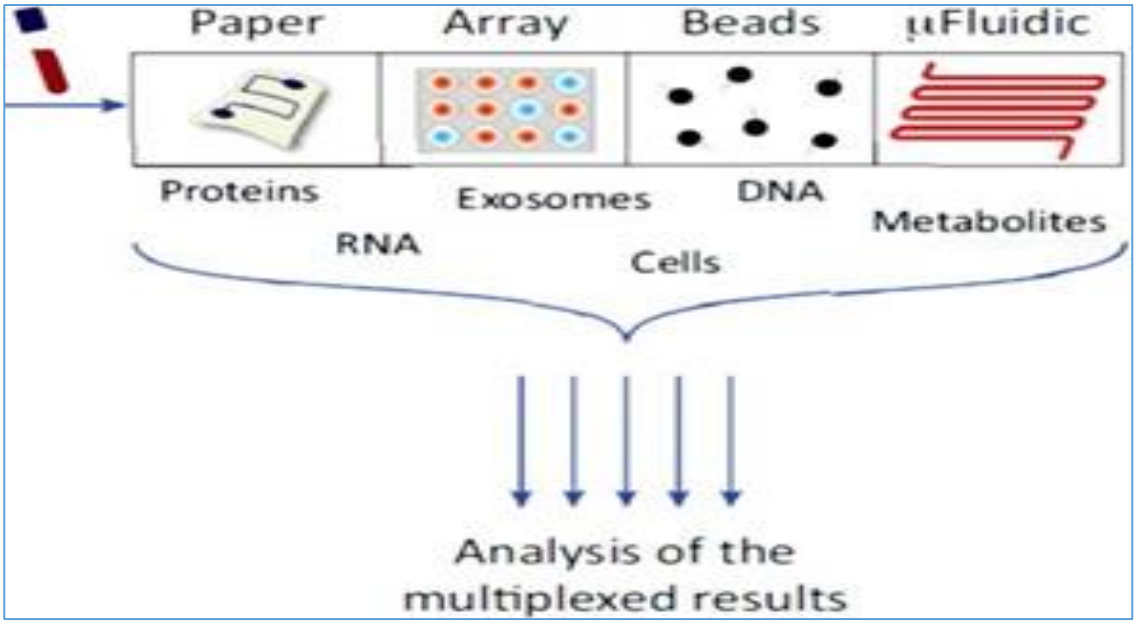


Figure 1 Major technologies for multiplexed point-of-care devices (Dincer *et al*., 2017).

Figure 2 below highlights the evolution of multiplexing technologies, depicting the simplicity of the paper technology despite their limitations regarding relative performance when compared to the other three technologies. With LFIAs continuing to be the pillar of POC diagnostics for infectious diseases in limited resource settings, this technology warrants a place for further research within the nanotechnology space. Thus, the following section analyses the types of multiplexed LFIAs, exposing current technology hurdles to be overcome in the making of the HIV/HCV multiplexed POC device.

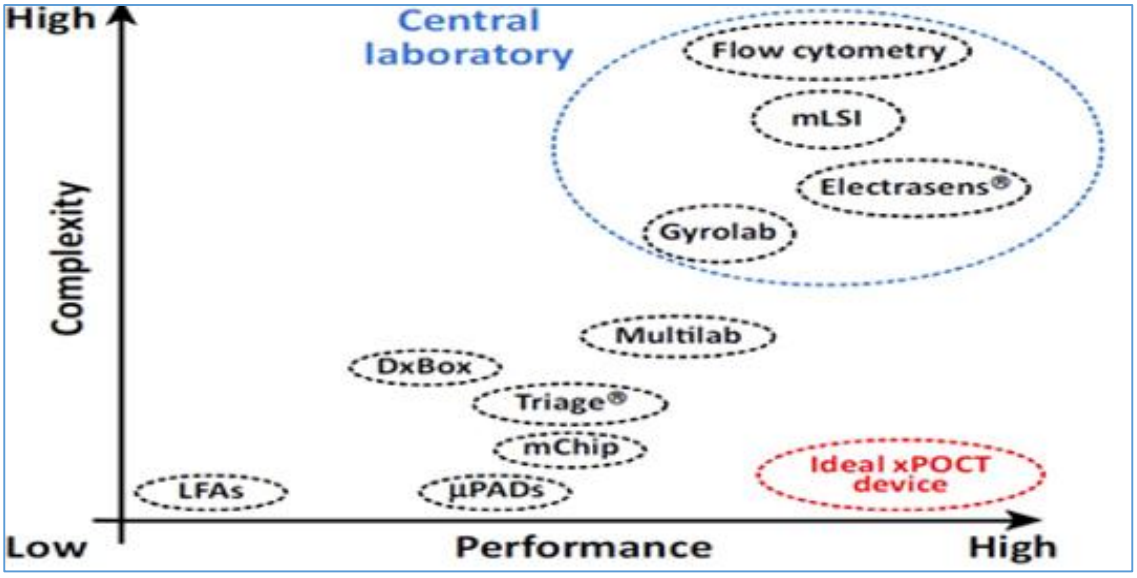


Figure 2 Evolutionary map for general multiplexed point-of-care devices

LFAs = lateral flow assays; mLSI = microfluidic large-scale integration;

mPADs = microfluidic paper-based analytical devices (Dincer *et al*., 2017).

2.5 Hurdles in the Manufacture of Multiplexed Point-of-Care Devices Based on Lateral Flow Immunoassay

Multiplexed POC testing devices based on various forms of LFIA are the most popular diagnostic tools for rapid diagnosis of multiple infections (Kim *et al*., 2019). LFIA-based POC devices detect multiple target analytes from a sample stream, using nano-sized particles immobilized on a single strip or multiple strips. The use of multiple strips has proved to be more practical for the detection of multiple pathogens (Hanbi & Doo-Ryeon, 2019). Dincer *et al*. (2017) reviewed the various multiplexed POC devices currently in the market and under development, and observed numerous gaps in the development of multiplexed POC devices addressing the co-occurrence of infectious diseases in limited resource settings.

2.5.1 Challenges in Simultaneous Detection of Multiple Target Analytes

The first hurdle in the making of multiplexed LFIA is the simultaneous detection of multiple target analytes from a single sample stream using single or multiple paper strips, or microarray technologies. According to Taranova *et al.* (2013), the paper strip is usually multilayered with three pads, that is, the topmost membrane for removal of debri in the sample, followed by sample stream pad allowing easy sample flow, with the conjugate/antigen pad layered afterwards, and lastly the absorbent pad to take up excess sample. Figure 3 illustrates the composition of a multiplexed POC device.

Major innovation differences lie in the conjugate pad as shown in figure 4. Dincer *et al*. (2017) indicated that putting multiple conjugates in separate lines on a single strip is affected by the possibility of cross-reactions across adjacent lines. Anfonsi *et al*. (2019) added that using several test lines on a single strip is also affected by the flow distance of the test sample, while using multiple strips has the disadvantage of space limitation. This leaves microarray technique as the most viable, although not without challenges, the most critical being complexity of the indicator system. This project adopts a LFIA sample separation technique.

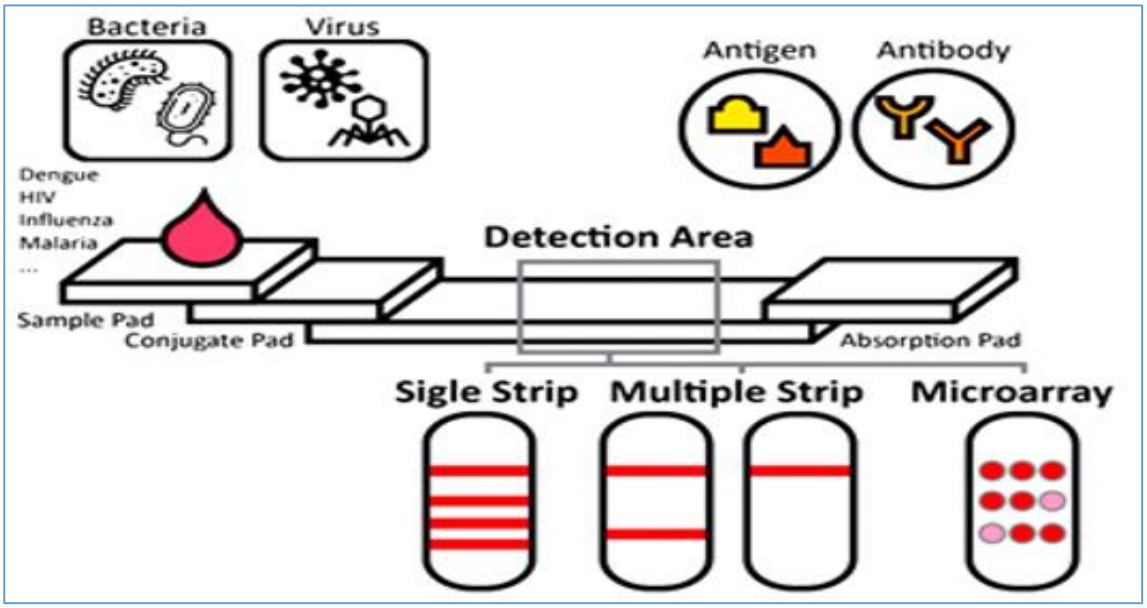


Figure 3 Simplified multiplex lateral flow immunoassay targeting antibodies to multiple infectious agents (Kim *et al*., 2019).

Various forms of multiplex LFIA platforms attempting to circumvent the hurdle of achieving simultaneous detection of multiple target analytes from a single sample stream based on multiple paper strips are at prototype stage. These include the 10-channel lateral flow assay in disc formation for the simultaneous detection of foodborne pathogens (Zhao et al., 2016). The assay was a major stride (Saylan, & Denizli, 2019), but the device could only detect four of the ten samples accurately mainly because the device made use of natural antigens and not 3D printed protein probes (Zhao et al., 2016). To minimize the unreliable detection system based on natural antigen probes, this project proposes the use of 3D printed viral antigens.

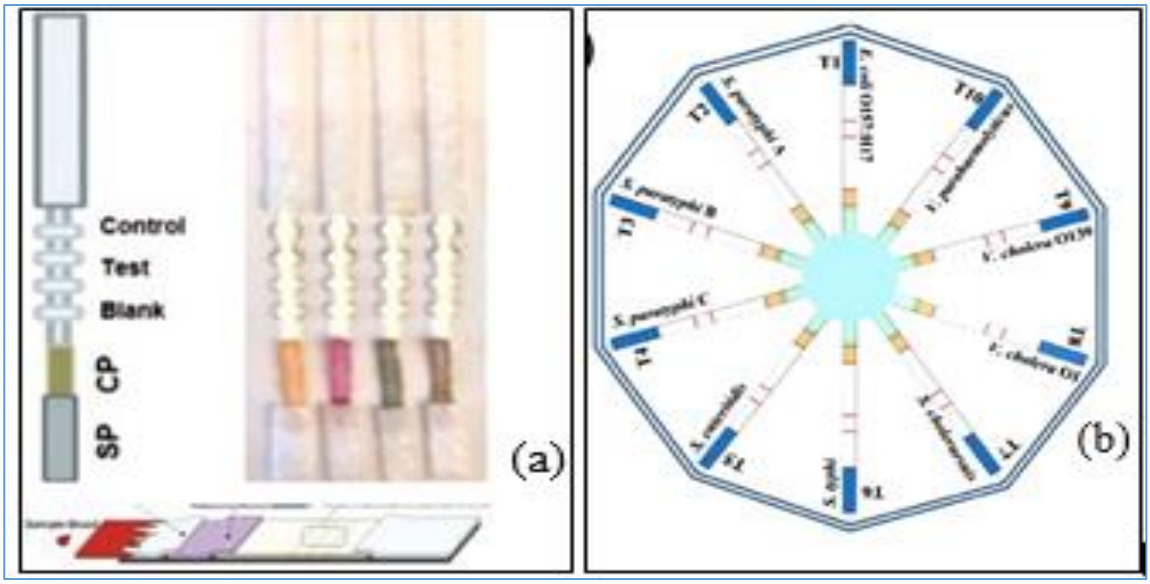


Figure 4 Modes of multiplexing lateral flow immunoassays (a) Multiple strips supplied by single stream (b) multiple strips in disc formation (Anfossi *et al*., 2018).

2.5.2 Challenges in Designing a Simplified Indicator System

The second hurdle in the making of multiplexed POC devices is the designing of a simplified indicator system. Kim *et al*. (2019) indicated that the simplest result indicator systems make use of colloidal gold or silver nanoparticles, although newer novel labels use paramagnetic nano-beads. Of note, fluorescent-based indicator systems have the major disadvantage of requiring expensive automated readers such as cameras, spectrometers, and magneto-inductive sensors. Sajid, Kawdea & Daud (2014) noted that some technologies such as combining microarray technique with lateral flow immunoassay technology have the potential to offset the limitations of paper strip technology while improving diagnostic validity. However, the technology has a complex indicator system barely useful in limited resource settings.

Opportunities exist to develop a new indicator system based on 3D-printed viral proteins conjugated with colloidal gold or silver nanoparticles and this project proposes a modified indicator system that leverages the use of 3D-printed viral proteins coupled with indicator molecules.

2.5.3 Challenges with Cross-reactivity of Natural Antigens

The third hurdle is to overcome challenges with the nature of nano-sized particle probes or antigens for detecting the antibodies. Despite global advancement in research on engineered 3D protein particle probes that detect antibodies in the sample, progress on the same in the developing world remains curtailed. Engineered probes are known to overcome cross-reactivity of natural antigens, and they have been used in devices that simultaneously detect multiple infectious diseases including HIV, HCV, and Hepatitis A viruses (Hanafiah *et al*., 2017). Investing in the making of these 3D printed probes is not only a potentially lucrative venture, but also an important field of nano-biotechnology with well-placed technology spinoffs in SSA (Liu *et al*., 2013).

2.6 Chapter Summary

In summary, the demand for POC devices is high but the gap for HIV/HCV POC devices in developing countries is worsening. The development of HCV diagnostic kits is lagging, and HCV remains neglected in burdened SSA. The manufacture of multiplexed POC devices that detect HIV and HCV remains low worldwide due to various reasons. A number of hurdles in the manufacture of multiplexed POC devices have been cited which include challenges in simultaneous detection of multiple target analytes on a single strip; challenges in designing a simplified indicator system; and challenges with cross-reactivity

of natural antigens. There is still room to develop new multiplexed POC devices. This project therefore looks at designing a prototype device for the simultaneous detection of HCV and HIV based on innovations that address the above hurdles

CHAPTER 3: METHODOLOGY

3.1 Introduction

This chapter discusses in detail the methodological choice and the research design process of the study. The chapter also includes the study setting, population and sampling procedure, instruments and the methods of data collection. The researcher gives explanations for the chosen methods. A section on data analysis, and ethical consideration conclude this chapter.

3.2 Research Design

3.2.1 Perspectives of Healthcare Professionals on Multiplexed Point-of-Care Testing Devices

A cross-sectional study of Bambanani Newstart Centre healthcare professionals was conducted. Participants were invited to complete a questionnaire between February and March 2022.The researcher chose this research design because it best serves to answer the purpose of the study. In cross-sectional study the researcher measures the outcome and the exposures in the study participants at the same time (Setia, 2016).

3.2.2 Conducting Market Analysis for Point-of-Care Devices

Market analysis for POC devices was performed using secondary data. Published information was analyzed to identify multiplexed POC devices currently available in SSA, to identify competitors, establish benchmarks and identify more target segments. The secondary data that was utilized include public information on Google Scholar, PubMed, Web of Science electronic databases, Researchgate, the WHO library, and universities repositories for grey literature such as dissertations, theses, and reports. POC devices

specifications and company marketing information were also utilised. Appraisal of the previous studies was performed using the 2018 version of the Mixed methods appraisal tool. Thematic content was used to extract emerging themes to present a narrative account of the findings.

3.2.3 Development of the HIV/HCV Multiplexed Point-of-Care Device

An experimental laboratory-based study design was adopted for the development of the multiplexed POC device based on LFIA to concurrently detect HIV and HCV using nano-sized particles immobilized on multiple strips on a single cartridge. Informed by the hurdles outlined in Chapter 2, four critical innovations were needed, that is, paper strips, protein-particle probes, indicator system and 3D-printing of the housing cartridge, with these innovations constituting three standalone sub-hubs of microfluidics, immunoassay and 3D-printing, all under the umbrella of nano-technology as depicted in figure 5. Protein-particle probes, indicator system and the 3D printing innovations were however not used at this stage. They will however be utilised in the next phase of the project.

The multiplexed POC device is based on LFIA technology, with the multiplexing achieved via multiple strips supplied by a single stream. Various pad arrangements and sample miniaturization (microfluidics) alternatives that circumvent the problems of sample volume, flow distance and space limitation were experimented with to achieve the simplest but effective paper strip technology.

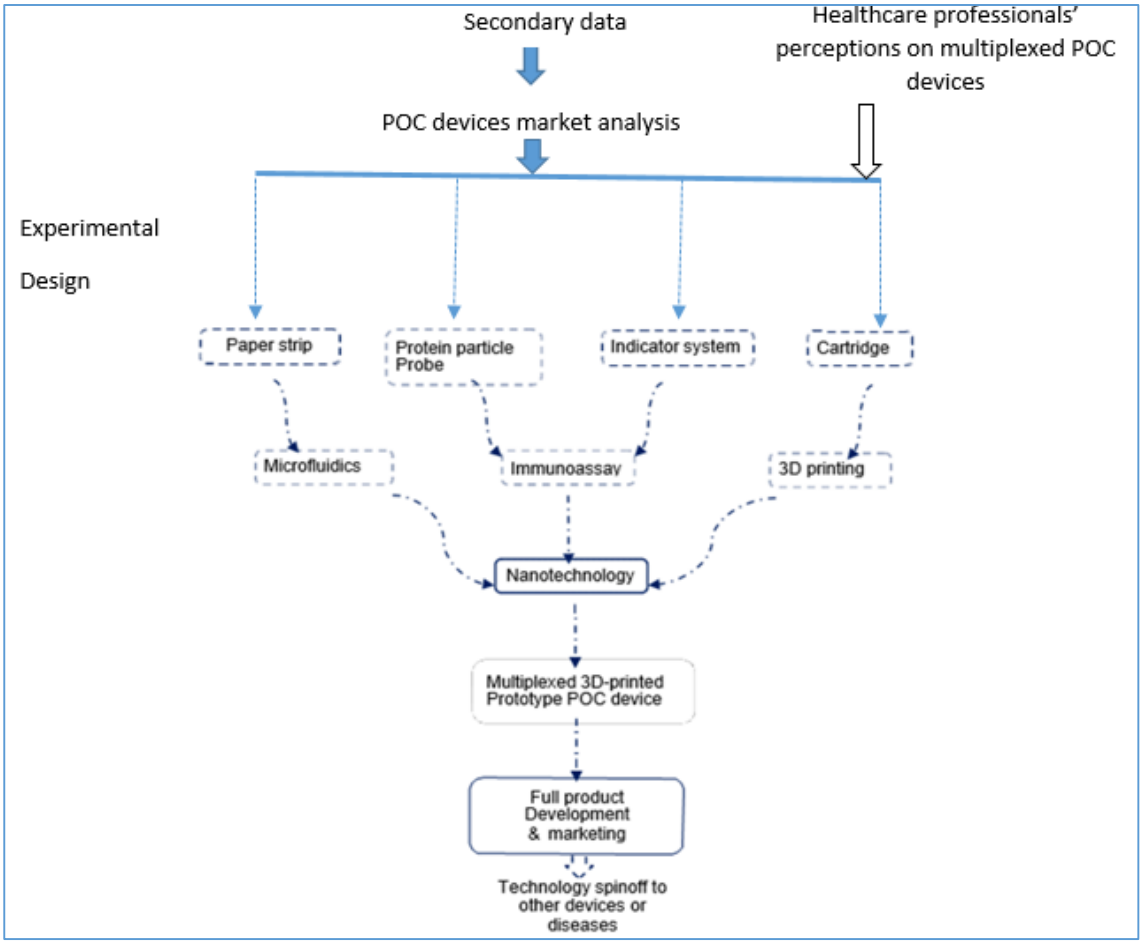


Figure 5 Conceptual framework of the study

3.2.4 The Test Principle of the Lateral Flow Immunoassay Strip

The test principle of the POC device is based on a direct sandwich assay, implying that the biotinylated antibody attached to the gold nanoparticles on the conjugate pad moves with the blood until it encounters the capture antibody with the specific HIV or HCV antigen attached to the membrane at the test lines. With the addition of the antigen and the second antibody, also known as the detection antibody, the detection antibody binds the antigen at a different epitope than the capture antibody. One of these antibodies is conjugated to the detection reagent and is held at the conjugate release pad, while the other

antibody is immobilized at the test line on the membrane. Any excess labeled antibody will be captured at the control line. The intensity of the color at the test line is indicative of the amount of analyte present in the sample.

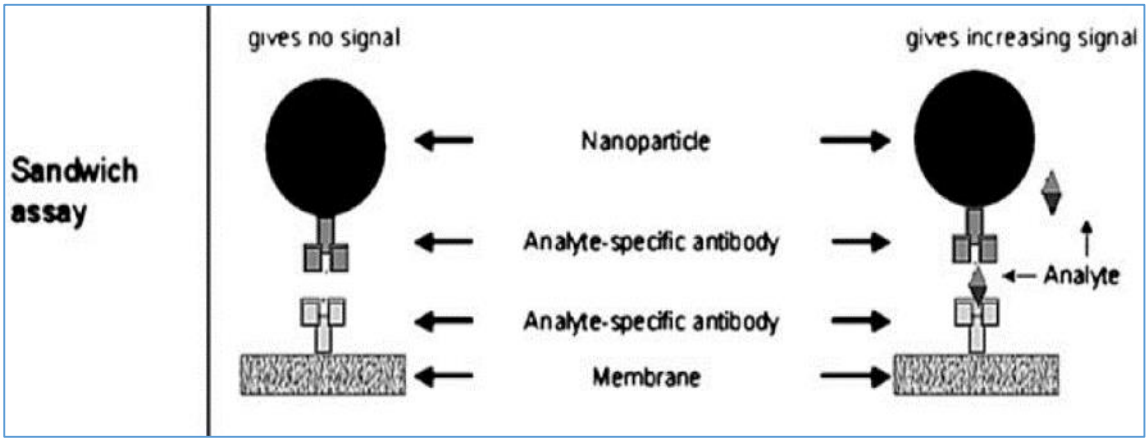


Figure 6 Direct sandwich assay format (Dincer *et al*., 2017).

An alternative double sandwich indirect assay that targets HIV and HCV antibodies was experimented with. The technique uses a two-step process for detection, whereby a primary antibody specific for the antigen binds to the target, and a labeled secondary antibody against the host species of the primary antibody binds to the primary antibody for detection. The antigen is immobilized on the membrane test line. This is shown in figure 7 below.

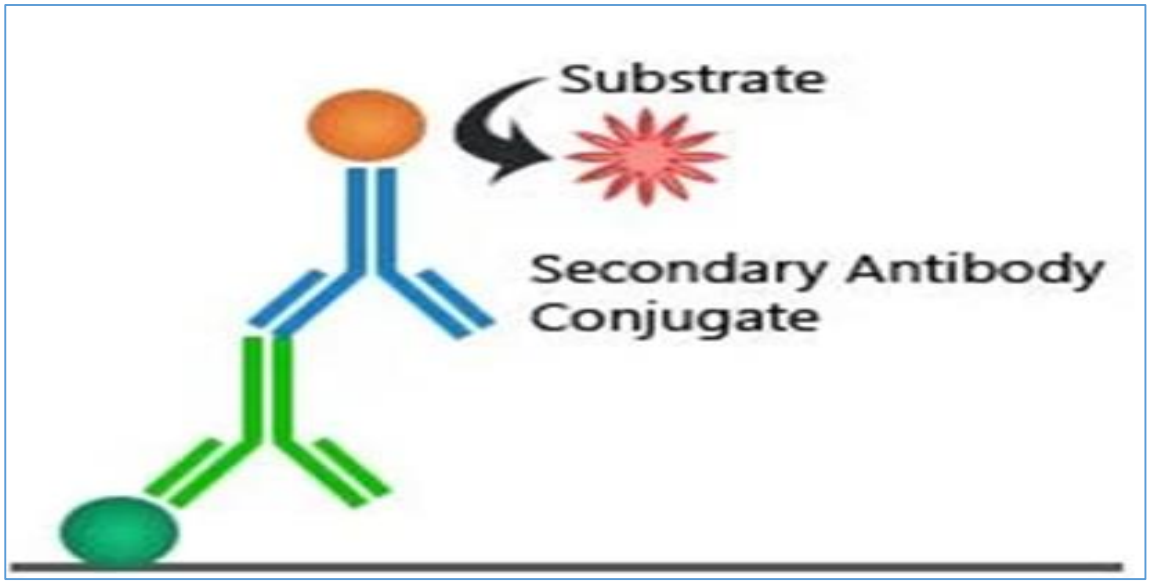


Figure 7 Indirect sandwich assay format (Kim *et al*., 2019)

3.3 Study Setting

3.3.1 Perspectives of Healthcare Professionals on Multiplexed Point-of-Care Testing Devices

The study was conducted at Bambanani Newstart Centre in Bulawayo City, Zimbabwe. The clinic is a key population designated centre providing TB screening and testing, cervical cancer screening, voluntary medical male circumcision (VMMC) HIV testing, antiretroviral therapy, (ART), HIV pre-exposure prophylaxis (PrEP) services, laboratory services, and family planning services. The clinic mainly serves people living with HIV/AIDS and key populations such as sex workers and men having sex with other men (MSM).

3.3.2 Development of the HIV/HCV Multiplexed Point-of-Care Device

To develop the HIV/HCV multiplexed POC device much of the work required laboratory equipment as this was a laboratory based experimental study. Therefore the project was conducted at Tshwane University of Technology (TUT) laboratories. TUT is a higher education institution in South Africa that came into being through a merger of three technikons: Technikon Northern Gauteng, Technikon North-west and Technikon Pretoria.

3.4 Population and Sampling

3.4.1 Perspectives of Healthcare Professionals on Multiplexed Point-of-Care Testing Devices

Bambanani Newstart Centre has 21 healthcare professionals. The study population of 21 healthcare professionals is small enough to warrant the inclusion of all of them in the study. Therefore the survey was conducted including all healthcare workers from February 2022 to March 2022.

3.4.2 Conducting Market Analysis for Point-of-Care Devices

Published data on electronic databases made up the population of the study. The search and selection of relevant literature on POC devices was guided by the search terms. Studies and published information on global POC industry and trends, POC devices development and POC devices utilsation in SSA made up the sample.

3.4.3 Development of the HIV/HCV Multiplexed Point-of-Care Device

Experimental methods were used to identify the suitable design for the POC device, the mode of multiplexing and the determination of suitable capture antibody and detection

antibody concentrations. The experiments were conducted in triplicates. Performing the experiments in triplicate control for validity of the method and enhance the statistical power of the study. The experimental method with the satisfactory technical reproducibility and satisfactory sensitivity and specificity was adopted.

3.5 Inclusion and Exclusion Criteria

3.5.1 Perspectives of Healthcare Professionals on Multiplexed Point-of-Care Testing Devices

The study included all Bambanani Newstart Centre healthcare professionals. These are nurses, laboratory scientists, nurse assistants and pharmacy technician. Non-healthcare professionals were excluded from the study.

3.5.2 Conducting Market Analysis for Point-of-Care Devices

Secondary data on the development of POC devices based on LFIA, utilization of POC devices in SSA, and global POC industry and trends was considered for market analysis. The data was extracted from internet sources. No primary data was collected at this stage.

3.5.3 Development of the HIV/HCV Multiplexed Point-of-Care Device

Laboratory techniques and methods for POC devices development, based on LFIA were considered in this study. These laboratory methods were determined from literature review. Any other laboratory methods not based on LFIA were excluded.

3.6 Data Collection Instruments and Procedure

3.6.1 Perspectives of Healthcare Professionals on Multiplexed Point-of-Care Testing Devices

Survey invitations were distributed via internal emails to all healthcare professionals. The total number of individuals reached was 21. A previously validated questionnaire was adopted from a study by Teebagy *et al*. (2022). The survey for this study was launched on 5 February 2022 and closed on 3 March 2022. If potential survey respondents did not respond in 10 days, a second reminder email was sent.

3.6.2 Conducting Market Analysis for Point-of-Care Devices

The study adopted a rapid review to search, identify, select and critically synthesise published literature from various electronic databases. Secondary data on Google Scholar, PubMed, Web of Science electronic databases, Researchgate, the WHO library, universities repositories, POC devices specifications and company marketing information was utilised. The search and selection of relevant literature was guided by the search terms. Appraisal of the secondary data was performed using the 2018 version of the Mixed methods appraisal tool. Thematic content was used to extract emerging themes to present a narrative account of the findings.

3.6.3 Development of the HIV/HCV Multiplexed Point-of-Care Device

The LFIA test strips were cut using a paper cutter as shown in figure 8. The LFIA strip consists of a nitrocellulose membrane with several pads: backing pad, absorbent pad, conjugate pad, and sample pad. The configuration is shown in Figure 9. Determination of the capture and detection antibodies for HIV and HCV was performed using the enzyme linked immunosorbent assay (ELISA) (figure 10) technique as described in the laboratory protocol manual.

Figure 8 Cutting the lateral flow immunoassay test strips

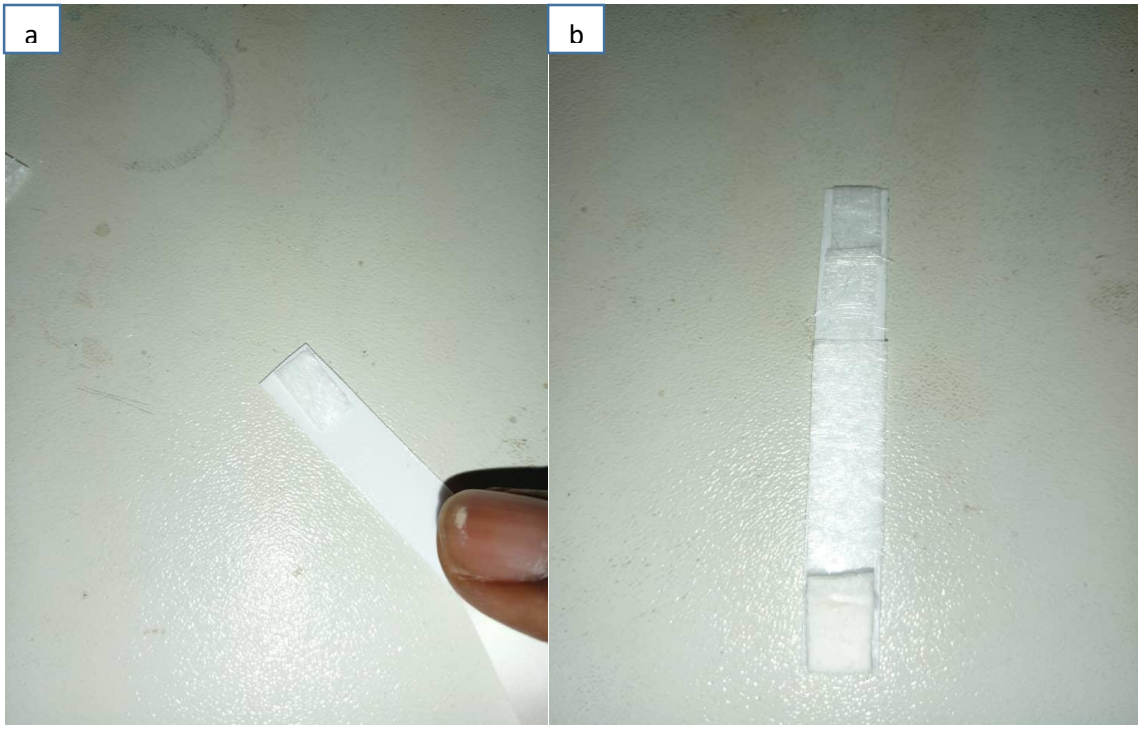


Figure 9 Attaching the test strips on backing pad

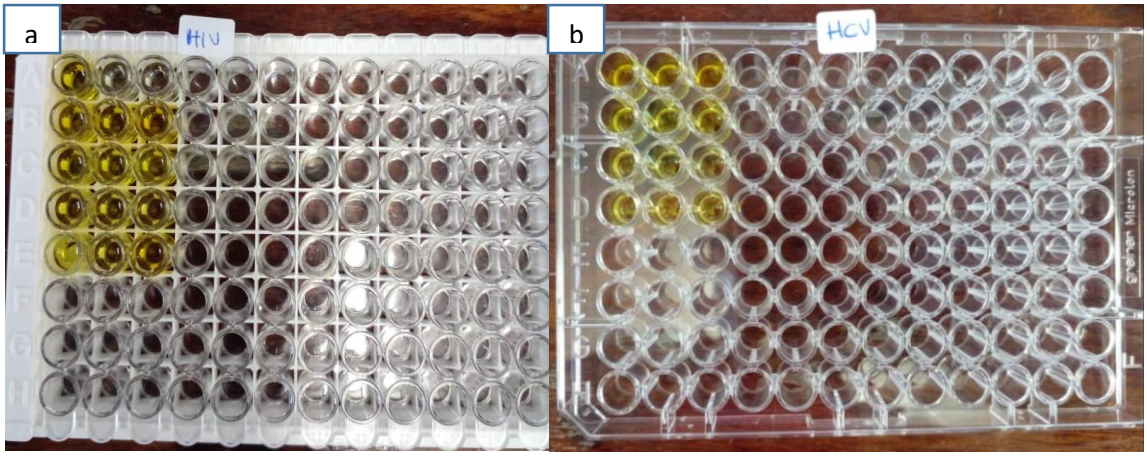


Figure 10 Determination of (a) HIV detection antibody concentration (b) Hepatitis C detection antibody concentration

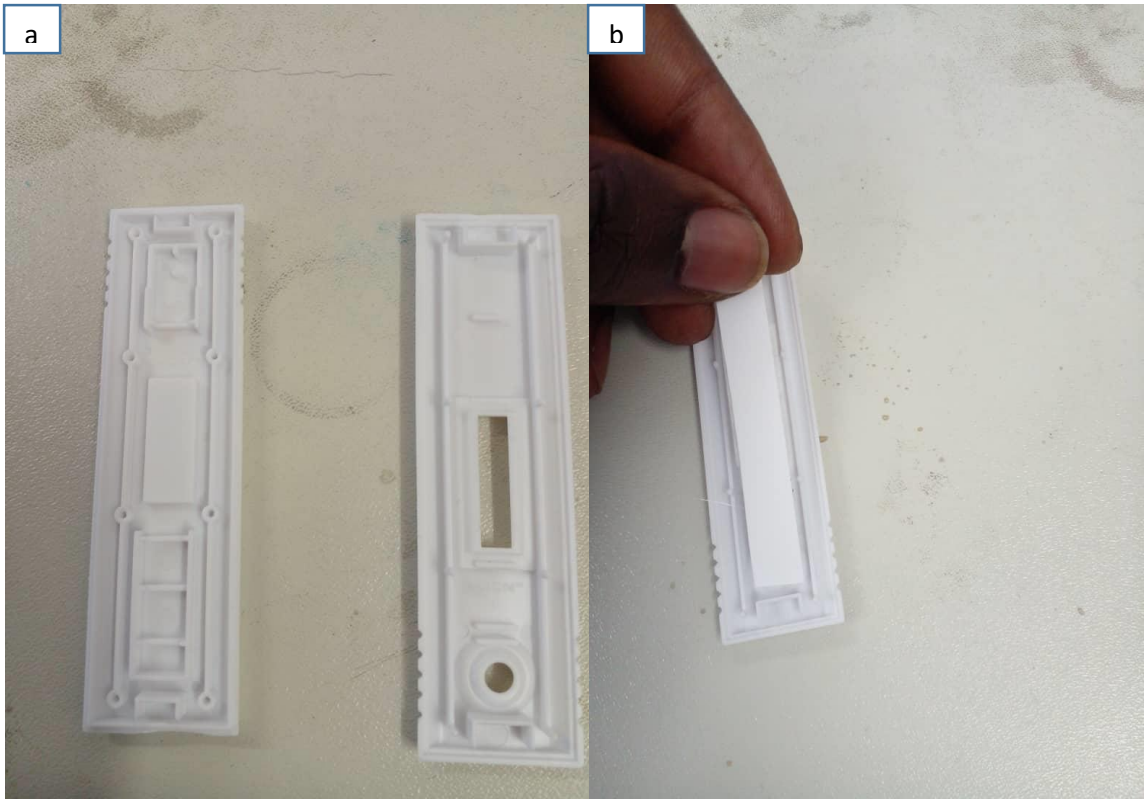


Figure 11 Inserting attached lateral flow immunoassay strips into the cassette

3.7 Analysis and Organization of Data

3.7.1 Perspectives of Healthcare Professionals on Multiplexed Point-of-Care Testing Devices

Two categories were created for variables from survey questions that allowed ranking of responses on the 5-point Likert scale. Responses were categorized as follows: (i) responses indicated “strongly agree” and “agree” were categorised as agreement, (ii) “strongly disagree” and “disagree” were designated as disagreement. Neutral/not sure responses were excluded from the analysis.

3.7.2 Conducting Market Analysis for Point-of-Care Devices

Data was abstracted from the included internet sources and studies. Content that was abstracted include author and title of article, size of the POC market, the customer segments, competition, and barriers to market entry. Thereafter, data was collated and summarized in a narrative format with figures and tables embedded where appropriate.

3.7.3 Development of the HIV/HCV Multiplexed Point-of-Care Device

A laboratory protocol was prepared to guide the development of the HIV/HCV multiplexed POC device. Results from the ELISA tests done were recorded on an excel sheet. The study will be continued at Doctor of Philosophy (PhD level) were the accuracy of the full-scale developed POC device to detect HIV and HCV in commercial standard simulative samples will be assessed by diagnostic sensitivity and specificity, and positive and negative likelihood ratios. Factors associated with the lack of result or false-negative results will be identified using Fisher’s exact test for categorical variables. All the variables found to be significantly associated with the lack of result or false negative in

the crude analysis ($P<0.05$) will further be assessed in the multivariable logistic regression. Sample size of the case–control study to determine sensitivity and specificity will be based on the WHO (2021) target product profile indicating the minimally acceptable sensitivity and specificity as 95% and 98%, respectively.

3.8 Ethical Consideration

Permission to carry out the study was obtained from the Bambanani Newstart Centre site manager, the Africa University Research Ethics Committee (Reference number: AU2358/22) and Tshwane University of Technology Faculty Committee for Research Ethics (Reference number: FCRE 2022/03/007 SCI 02). Furthermore, informed consent was obtained from all participants. The purpose of the study was well explained and confidentiality was assured. Emails of respondents were not collected and the collected data was be kept in confidence and was used for academic purposes only.

3.9 Chapter Summary

Market analysis for POC devices was performed to identify multiplexed POC devices currently available in SSA, to identify competitors, establish benchmarks and identify more target segments. An assessment of perspectives of healthcare professionals gave an insight on the ideal POC device. An experimental laboratory-based study was conducted at TUT laboratories for the period between March to April 2022 to develop the HIV/HCV multiplexed POC device. Experimental procedures were developed to identify a suitable design for the multiplexed POC device, addressing specificity and sensitivity requirements. Permission to conduct the study was sought from TUT, Bambanani Newstart Centre and the Africa University Research Ethics Committee.

CHAPTER 4: DATA PRESENTATION, ANALYSIS AND INTERPRETATION

4.1 Introduction

This chapter focuses on data analysis and the presentation of the study findings. Findings are presented with the aid of frequencies and percentages, tables, graphs and pie-charts. The purpose of this study was to develop and optimize a multiplexed POC device based on LFIA technology that detects co-occurrence of HIV and HCV. The study also evaluated healthcare professional's perspectives on multiplexed POC devices using a cross sectional study design. A market analysis for HIV and HCV POC devices in the sub-Saharan market was also conducted; assessing size of the market, the customer segments, competition, and barriers to market entry. Secondary data was used for the market analysis. An experimental laboratory based design was adopted for the development of the HIV/HCV multiplexed POC device.

4.2 Results of the Perspectives of Healthcare Professionals on Multiplexed Point-of-Care Testing Devices

4.2.1 Demographic Characteristics of Respondents

Participants were asked about their demographic information during the survey. Though not central to the study, demographic data helps to contextualise the findings and may assist in the formulation of appropriate recommendations. A total of 21 participants responded to the survey. Of those, 15 (71.4%) were females and 6 (28.6%) were males. In regards to race, all participants were black. Of the respondents, 16 (76.2%) had practiced for 5 years or more, while 5 (23.8) had less than 5 years of professional

experience. A majority of the survey responses (71.4%) were from nurses but other healthcare professionals were also represented in the sample, including laboratory scientists (19.0%), nurse assistants (4.8%) and pharmacy technicians (4.8%).

Table 1 Demographic information of survey respondents

Respondents demographics, n=21	**Number of respondents (%)**
Gender	
	6 (71.4)
Female	15 (28.6)
Race	
Black	21 (100)
White	0 (0)
Other	0 (0)
Years in practice	
0-5	5 (23.8)
6-10	7 (33.3)
11-15	5 (23.8)
16-20	4 (19.1)
Profession	
Nurse	15 (71.4)
Laboratory scientist	4 (19.0)
Nurse assistant	1 (4.8)
Pharmacy technician	1 (4.8)

4.2.2 Important Aspects of Point-of-Care Testing Devices

Participants were asked to select the most important characteristics of point-of-care testing devices they value in their routine work. Ease of use was the most selected response. The findings are summarised in table 2 below.

Table 2 Most important aspects of point-of-care testing devices

Characteristics	**Number of times listed in top 3 (% of respondents)**
Ease of use	21 (100)
Accuracy	21 (100)
Availability	15 (71.4)
Cost of testing	10 (47.6)

4.2.3 Benefits of Multiplexed Point-of-Care Devices

Participants were presented with 10 statements on the benefits of multiplexed POC devices and asked to rate the degree to which they agreed with the given beneficial statements. The choices were “strongly agreed”, “agreed”, “not sure”, “disagreed” or “strongly disagreed”. The percentage of respondents who agreed or strongly agreed is shown to the right of each statement. The statement “Multiplexed POC devices improve patient management” was the most agreed with, with 20 (95.2%) of the participants selecting “agree” or “strongly agree”. The findings are shown in figure 12.

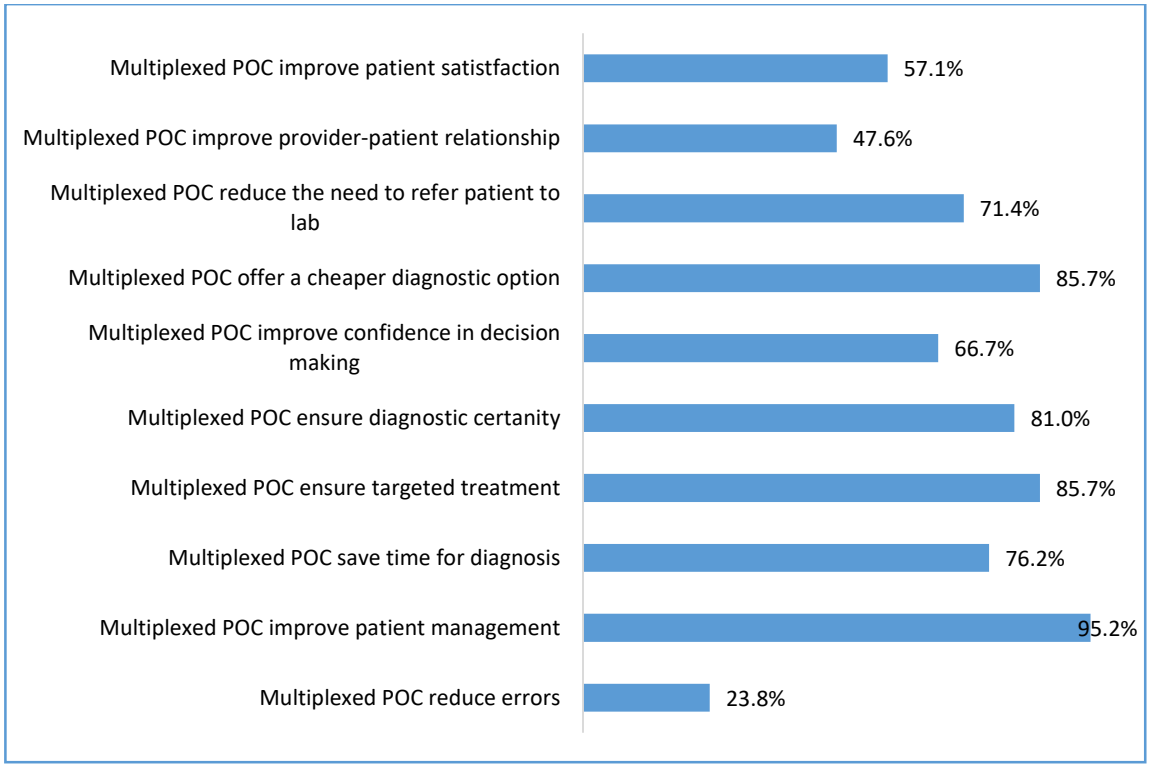


Figure 12 Participants responses to statements regarding benefits of multiplexed point-of-care devices

4.2.4 Concerns Over Multiplexed Point-of-Care Testing Devices

Participants were presented with a series of statements regarding possible concerns on multiplexed POC testing devices and the degree to which they agreed with the concerns. The percentage of respondents who agreed or strongly agreed is shown to the right of each statement. Of the respondents, 18 (85.7%) indicated that there is a shortage of multiplexed POC devices, a minority of respondents agreed or strongly agreed with the other statements, as shown in figure 13.

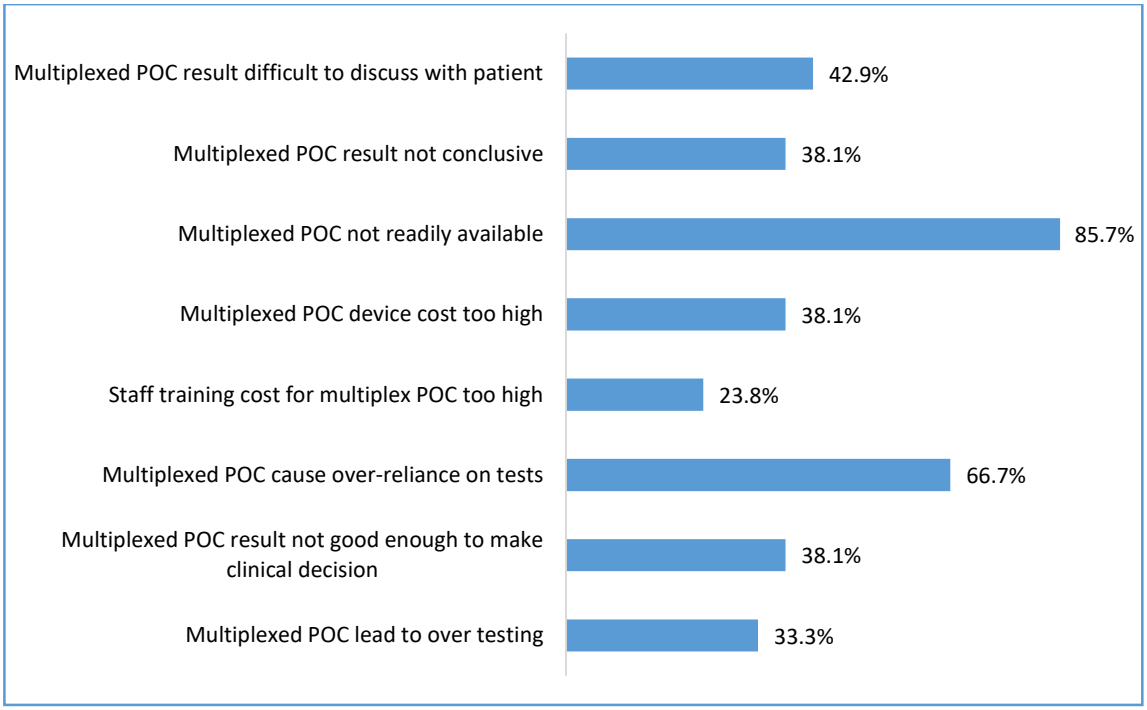


Figure 13 Participants responses regarding concerns over multiplexed point-of-care devices

4.3 Results of the Market Analysis for Point-of-Care Devices

4.3.1 Characteristics of the Articles Included in the Market Analysis Report

The initial POC articles search found 102 potentially eligible articles. Table 3 presents results from each database search. Following title screening, 49 articles were eligible for inclusion in abstract screening. Of the 49 articles, 5 duplicates were removed. Forty-four articles were eligible for full-text screening after abstract screening, and 10 articles were included in this market analysis report. Fifty three articles were excluded as not meeting the inclusion criteria.

Table 3 Results of keyword search

Search engine	Number of publications retrieved	Number of article eligible after title screening	Number of abstracts screened	Number of full-text articles screened	Number of articles included in report writing
Google Scholar	54	26	26	25	6
PubMed	32	18	18	17	3
Web of Science	8	3	3	2	1
Researchgate	4	1	1	0	0
WHO library	3	1	1	0	0
Universities repositories	1	0	0	0	0
Total	**102**	**49**	**49**	**44**	**10**

The characteristics of articles included in the market analysis report writing are shown in Table 4. All the included articles reported on the global, regional and international POC industry status, trends and demand; development of POC devices and POC testing devices utilisation. Of the 10 articles included in this study, 2 reported on the POC utilization in Africa and 1 on accessibility of POC devices in SSA.

Table 4 Characteristics of included studies

Author name	Title	Country/Region of study	Study design	Theme
Nichols (2021).	Utilising point-of-care testing to optimise patient care	United States of America	Narrative review	Utilisation of POC devices
Rajan & Glorikian, (2021).	Point-of-care diagnostics: market trends and growth drivers	United States of America	Observational	POC market, trends and growth drivers
Heidt *et al*., (2020).	Point-of-care diagnostics in resources-limited setting: A review of the present and future of POC in its most needed environment	Netherlands	Narrative review	POC market, trends and growth drivers
Nkengasong & Tessema, (2020)	Africa needs a new public health order to tackle infectious disease threat	Africa	Narrative review	Utilisation of POC devices
Falchetta, Hammad & Shayegh, (2020).	Planning universal accessibility to public health care in sub-Saharan Africa	Africa	Narrative review	Accessibility of healthcare
Schroeder & Amukele, (2014).	Medical laboratories in sub-Saharan Africa that meet international quality standards.	Africa	Narrative review	Medical laboratories in Africa
Clark (2002)	The triage cardiac panel	United States of America	Experimental	POC device development

Table 4 (Continued)

Author name	Title	Country/Region of study	Study design	Theme
Tonen-Wolyec *et al*. (2021)	Self-testing for HIV, HBV and HCV using finger-stick whole-blood multiplex Immunochromatographic rapid test: a pilot feasibility study in sub-Saharan Africa	Africa	Cross-sectional	POC device development
Kalla *et al*. (2018)	Mass campaigns for HIV, HBV and HCV screening by multiplexed rapid diagnostics test in sub-Saharan Africa using mobile units: the game changer	Africa	Cross-sectional	POC device development
Leman *et al*. (2018)	Analytical performanceof simulteneous detection of HIV-1, HIV-2 and hepatits C-specific antibodies and hepatitis B surface antigen (HBsAG) by multiplex immunochromatographic rapid test with serum samples: a cross-sectional study	Africa	Cross-sectional	POC device development

4.3.2 Current State of the Global Point-of-Care industry

Globally, the POC business is a multi-billion-dollar industry with intense competition and areas of high growth. As shown in figure 14, the global market for POC diagnostics was valued at US$32.5 billion in 2020 and is expected to grow at a compound annual growth rate (CAGR) of 11.1% to reach US$93.2 billion by 2030 (Nichols, 2021). The major factors contributing to the growth of the POC diagnostics market is the rising prevalence of infectious diseases.

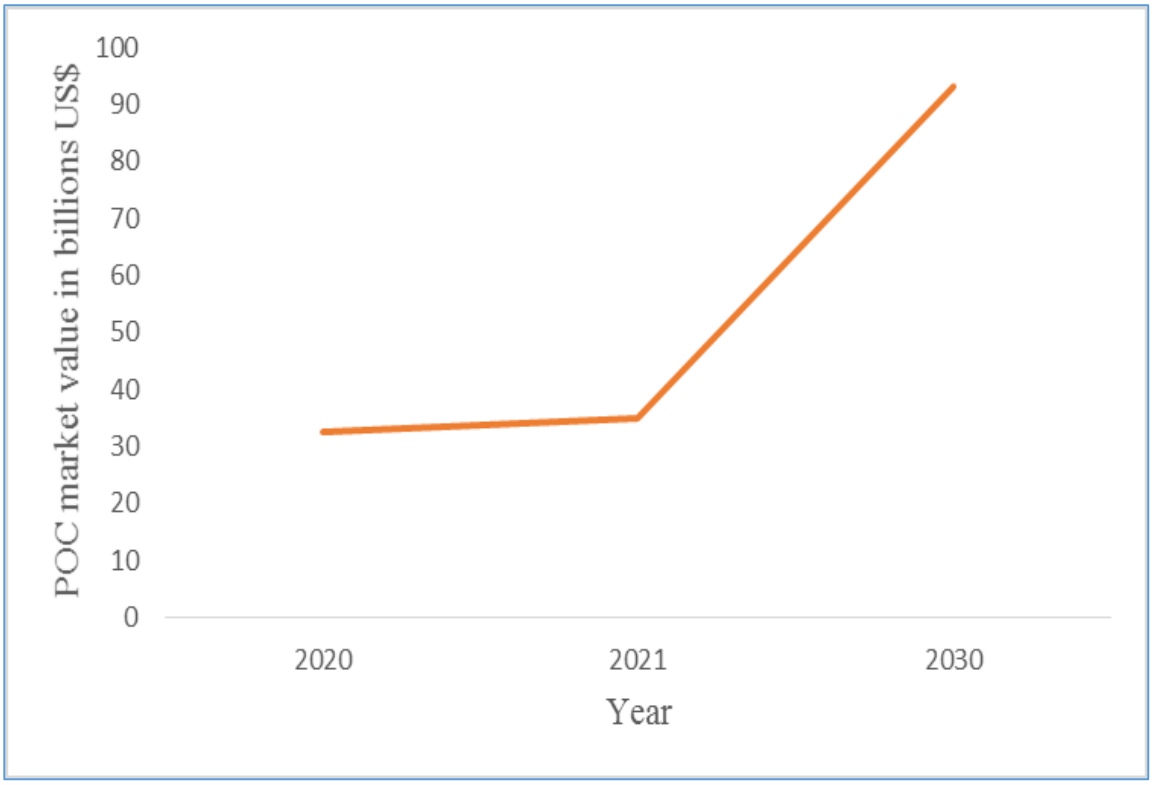


Figure 14 Global point-of-care devices industry market value

The United States of America is the world's largest manufacturer of POC testing devices, and Africa due to the rising burden of both communicable and non-communicable diseases, is providing a significant market for the diagnostic devices (Rajan & Glorikian,

2021). In Africa, POC testing is growing in both home use, laboratory settings and testing near-patient applications. The utilisation of POC testing devices in Africa and the Middle-east markets is expected to rapidly grow due to emerging diseases, increasing burden of infectious diseases, increasing disposable income, large-scale rising population and changing lifestyle (Nichols, 2021). Figure 15 shows the global utilisation of POC devices.

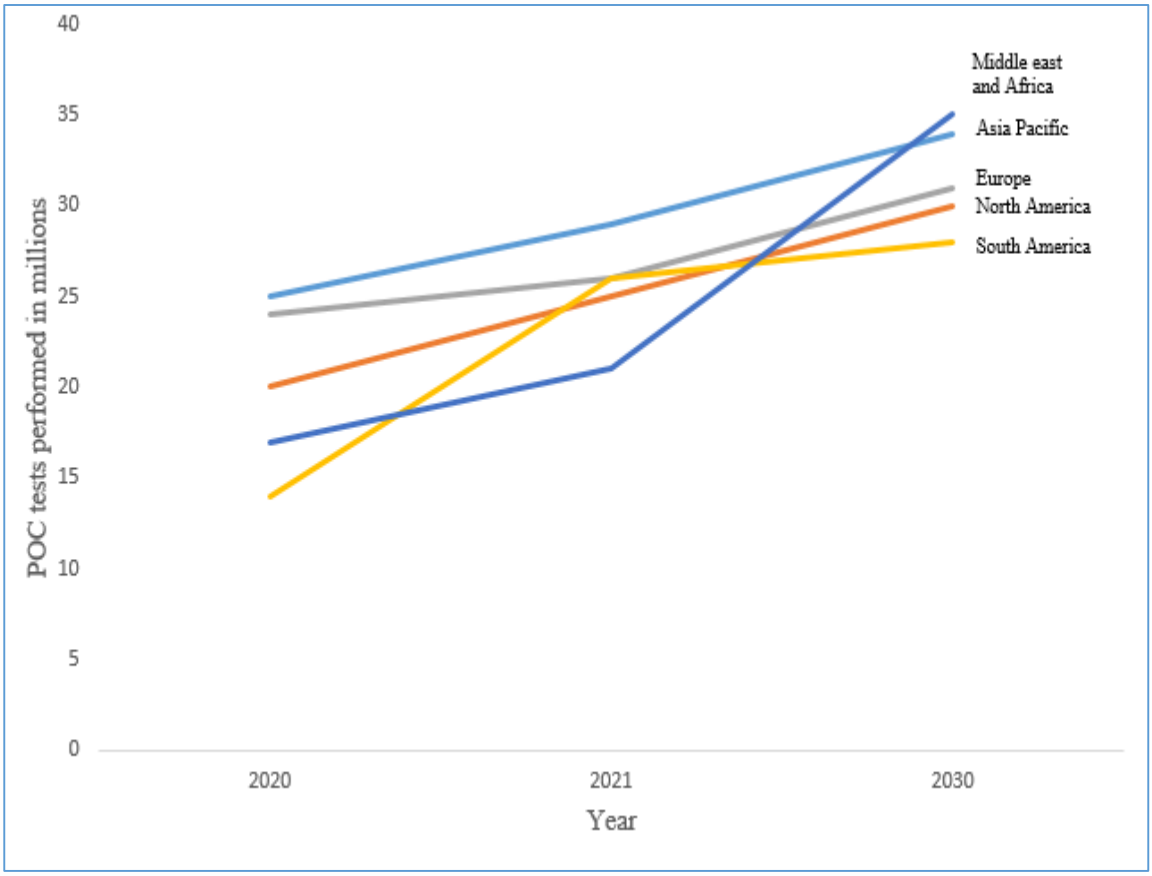


Figure 15 Point-of-care tests utilisation by region

Based on the product segment, the POC diagnostic market includes blood glucose monitoring, infectious diseases, cardiometabolic diseases, pregnancy and infertility testing, hematology testing and others (figure 16). Blood glucose and infectious diseases POC testing devices combined, account for over 80% of the market share. The blood

glucose monitoring segment is expected to hold the largest market share in the forthcoming years due to higher sales of diabetes rapid diagnostic kits (Heidt *et al*., 2020). The infectious disease segment is anticipated to show a significant growth due to rising prevalence of infectious disease like COVID-19, HCV and HIV. Africa is expected to provide one of the largest markets for infectious diseases POC testing devices (Nkengasong & Tessema, 2020).

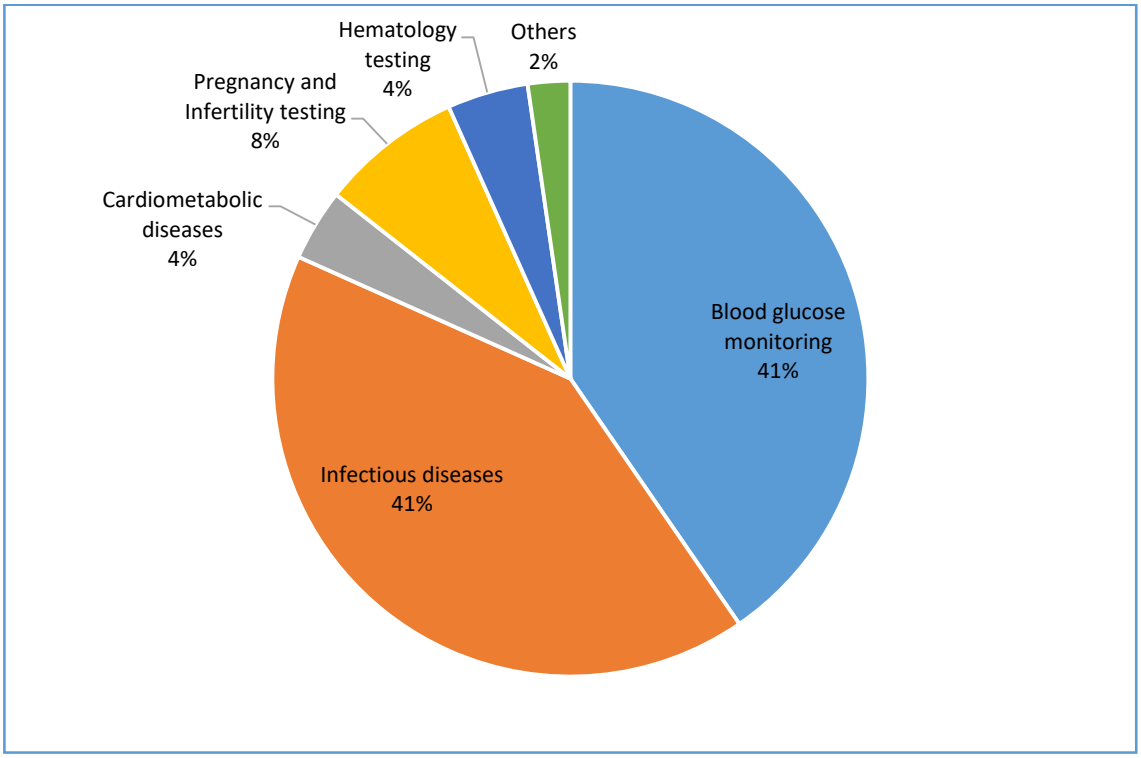


Figure 16 Global point-of-care diagnostics market share by products, 2021

4.3.3 Demand and Supply of HIV/HCV Multiplexed Point-of-Care Testing Devices in Sub-Saharan Africa

Sub-Saharan Africa faces the burden of communicable diseases such as tuberculosis, HIV and sexually transmitted infections that include syphilis and hepatitis B and C. Even though burdened with HIV and HCV co-infections, there is still low testing in SSA mainly

due to limited diagnostic tests. Figure 17 indicates the burden Zimbabwe is facing from HCV and HIV. Despite being on a downward trend since 2005, HIV prevalence is still high. Most sub-Saharan African countries show a similar picture of the HIV and HCV burden. Lack of access to good-quality laboratory diagnostic tests for HIV and HCV makes the demand for more affordable multiplexed POC testing devices very high in SSA. In addition, the limited supply of multiplexed POC devices increase the demand in SSA.

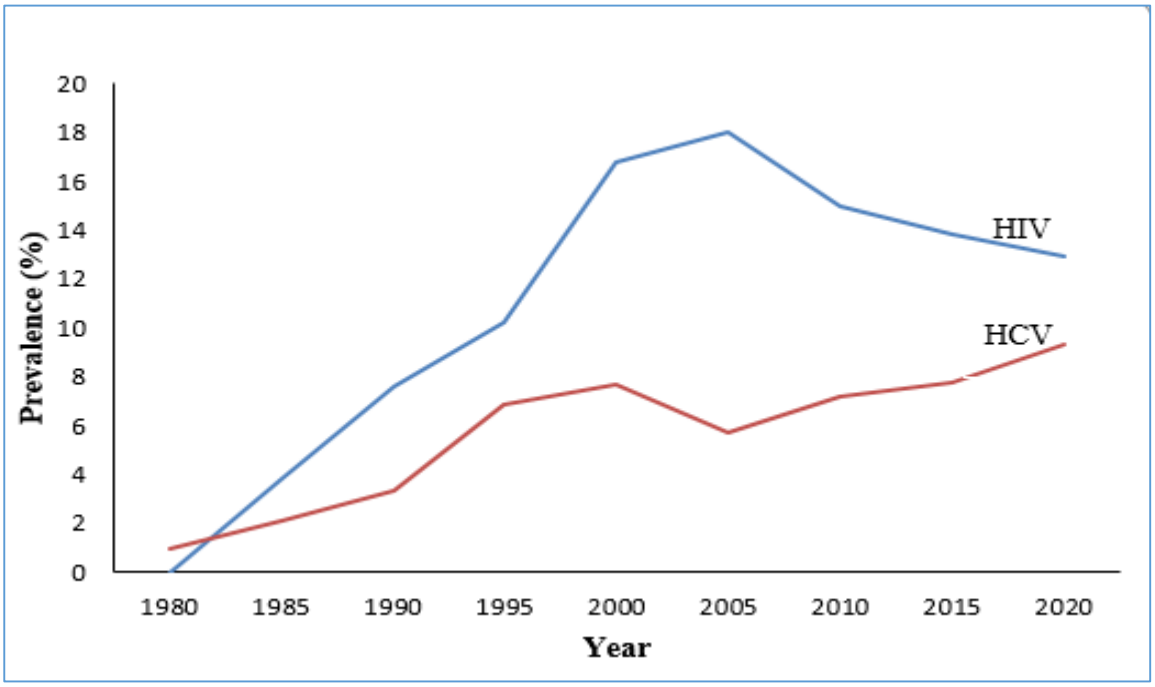


Figure 17 HIV and HCV prevalence in Zimbabwe

4.3.4 Potential Customer Segments for the HIV/HCV Multiplexed Point-of-Care Device

Potential market segments for the multiplexed POC testing device include pharmacies, laboratories (public, private, research, mission and council), clinics (public, private, mission and council), hospitals (public, private, mission and council) and private medical

practitioners. Hospitals and clinics provide the widest market for the multiplexed HIV/HCV device. Currently there are 5,723 hospitals in SSA (Falchetta, Hammad & Shayegh, 2020). Adopting HIV/HCV multiplexed POC testing devices in hospitals and clinics improves HIV/HCV screening, particularly among key populations and in antenatal care clinics.

Currently, private pharmacies are the most common target for HIV/HCV multiplexed POC devices in SSA and so, they should be the next logical target for the multiplexed POC devices after hospitals and clinics. South Africa has the highest numbers of pharmacies in SSA. The country has a total of 2,693 retail pharmacies (Schroeder & Amukele, 2014). This provides a wide market for the device.

Most laboratories in Africa also use POC devices in routine laboratory testing especially for infectious diseases like HIV, syphilis and hepatitis. Despite the fact that the majority of clinical laboratories in SSA are not accredited, they still offer HIV and HCV testing services to requesting doctors. National research and reference laboratories also use POC devices for surveillance purposes. HIV/HCV multiplexed POC devices will therefore provide a more cost-effective approach to the HIV/ HCV testing national and subnational algorithms.

4.3.5 Availability of Technologies for the Diagnosis of HIV/HCV in Sub-Saharan Africa

Technologies for the diagnosis of HIV and HCV available in SSA are listed in Table 5. Most POC devices for the detection of HIV and HCV and other related infectious diseases in SSA are mostly based on LFIA technologies and are manufactured by various

companies from USA, China, France and the United Kingdom (Table 5) and none from SSA. Three POC devices are still at the prototype stage. The lowest price is for the HIV, HBsAg, HCV and Syphilis Combo Rapid Test devices and this represents the highest risk of competition in cost terms.

Table 5 HIV and HCV diagnostic point-of-care technologies in sub-Saharan Africa

Device name	Manufacturer	Device target	Technology used	Development stage: Prototype (PT), Market (M)	Local (SSA) manufacture (Y/N)	Projected / Cost (US$)	Competition risk level: Low (L), Medium (M), High (H)	Reference
HIV, HBsAg, HCV, & Syphilis Combo Rapid Test	Biopanda, UK	HIV, HBsAG, HCV, Syphilis	LFIA	M	N	2.05	H*	(Biopanda, 2021)
Alere Determine HIV-1/2	Abbott/Alere Inc., USA	HIV	LFIA	M	N	5.30	M	(Clark, 2002);
Alere Determine HCV	Abbott/Alere Inc., USA	HCV	LFIA	M	N	5.30	M	(Clark, 2002)
Multiplo Rapid HBc/HIV/HCV Antibody Test	MedMira, Canada	HIV, HBsAg, HCV	Vertical flow technology	M	N	9.52	L	(Medmira, 2021)
HCV/HBSAG/HIV Combo Rapid Test Cassette [ITHD-C43]	Biotest Biotech Inc., China	HIV/HCV/HBsAg	LFIA	PT	N	(-)	L	(Leman *et al*., 2018)
Multiplex HIV/HCV/HBsAg	Biosynex, France	HIV/HCV/HBsAg	LFIA	PT	N	(-)	L	(Kalla *et al*., 2018)
Triplex HIV/HCV/HBsAg self-test	Biosynex, France	HIV/HCV/HBsAG	LFIA	PT	N	(-)	L	(Tonen-Wolyec *et al*., 2021)

(-) denotes that the POC device is still in prototype stage;

LFIA=lateral flow immunoassay;

H* denotes ranking based on price competition. The devices with prices less than US$3.00 were ranked high, those between US$3.00 and US$6.00 were ranked medium whilst the devices costing more than US$6.00 or are in prototype stage were ranked low.

4.3.6 Competitors' Strengths

Most of the competitors for the HIV/HCV multiplexed POC device are from developed countries such as the USA, United Kingdom, France and China as shown in figure 18. Notable examples include Alere Inc. (USA), Biosynex (France), Biopanda (United Kingdom), Cepheid (USA), and Biotest Biotech Inc. (China). The competitors have the financial resources which allow faster development of new devices. Furthermore, the competitors are technologically advanced and have better access to human resources. This provides the first mover advantage to competitors.

Manufacturers such as Alere Inc. already have a big market share in SSA, providing quality products and enjoying a good reputation. The current manufacturers enjoy a good brand recognition, hence have a better bargaining power.

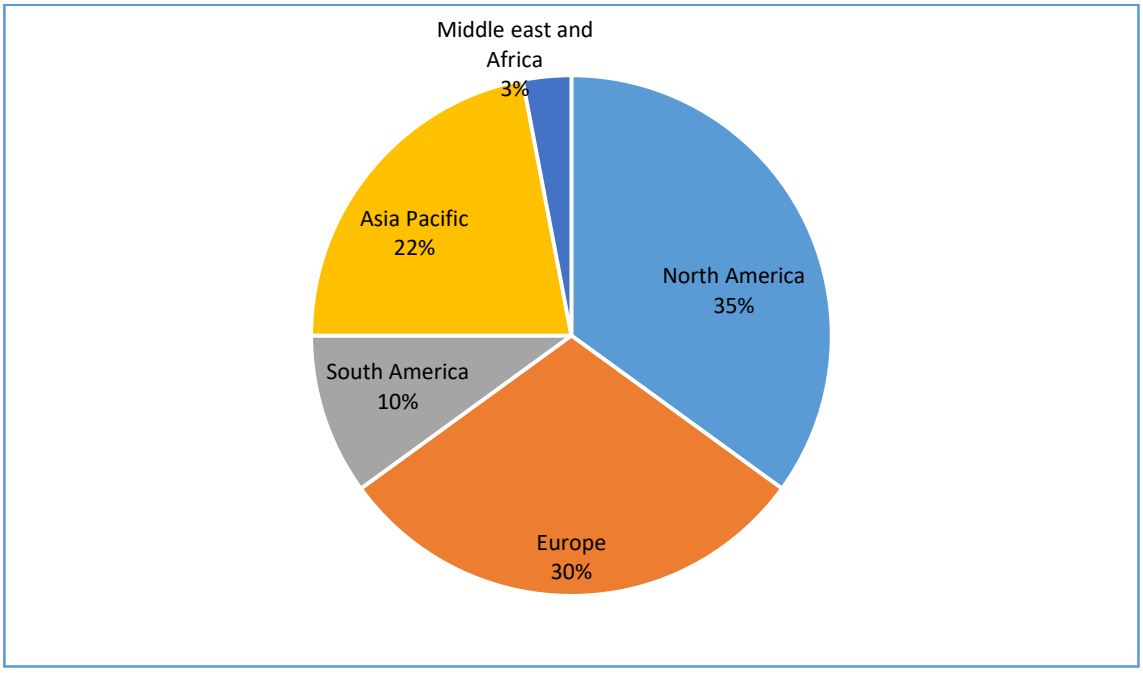


Figure 18 Global point-of-care diagnostics market revenue by region, 2021

Well-resourced international POC devices manufacturers have the ability to innovate. These well-established manufacturers have the ability to generate ideas that create value and improve processes, from inventing new POC devices based on better technologies to finding an effective strategy to monopolise the market. This is one of the greatest competitors' strengths, this market analysis report identified.

4.3.7 Competitors' Weaknesses

I. High cost of POC kits – As shown in table 1, the average cost of the HIV/HCV multiplexed POC devices currently produced for SSA is US$7.20. Despite the cost of production being on average US$1, shipping fee inflate the price. Local manufacturing has less production costs hence may produce relatively affordable POC devices that could rival imported devices.

II. Limited product availability - SSA countries such as Zimbabwe frequently face shortages of HIV and HCV POC kits. Donor reliance and lack of foreign currency are blamed for the temporary shortages. International POC manufacturing companies usually do not have flexible payment methods and platforms, and this creates temporary shortages. A local manufacturer can utilise the opportunity, offering flexible payment methods using local currency, and rival competitors.

III. Confusing service policies and procedures - POC devices from non-english speaking countries such as China may have unclear policies and procedures. Notable examples include the Diagret POC kit that comes with package inserts in the Chinese language. Package inserts for AccuBioTech Co. another Chinese firm, contains procedures in non-standard english. Unlike local manufacturer that can

offer products in vernacular languages, unclear procedures can be another weakness to the competitors.

IV. Competitors are foreign - The current competitors are not from Africa. The competitors are not experiencing first-hand the extent of the need and urgency for the HIV/HCV multiplexed POC devices. There is inadequate definition of customers for the POC devices.

4.3.8 Barriers to Market Entry

I. Product differentiation - Existing POC manufacturing companies supplying devices in SSA enjoy brand identification and customer loyalties. Of note Alere Inc. is the sole supplier of HIV POC kits adopted on the national HIV testing algorithms across sub-Saharan African countries. Substituting existing HIV and HCV POC devices requires clear communication to the target customer, which increases advertising costs.

II. Capital requirements - Infrastructure, machinery, research and development and advertising requires financial resources. Additional anticipated costs include patent and registration fees. These present a major challenge to penetrating the market.

III. Access to distribution channels – Approval of the POC device for use in SSA healthcare systems presents one of the most notable challenges. Some logical distribution channels have already been locked up by competitors in SSA. Switching costs, by consumers may delay adoption of the HIV/HCV multiplexed POC device for routine use.

4.4 Development of the HIV/HCV Multiplexed Point-of-Care Device

4.4.1 Laboratory Protocols Manual

A laboratory protocol was developed to guide in the development of the HIV/HCV multiplexed POC device. The laboratory protocol contains instructions, experiment procedures, safety protocols and required equipment that enabled the researchers to carry out experiments. To identify the capture and the detection antibody, determination of the optimum antibody concentrations for both the capture and detection antibody were conducted by following the steps below. Multiple antibodies to the analyte may exist, so it was important to examine all possible pairs of the antibodies.

4.4.2 Results of the Experiment

The ELISA plate was coated with several dilutions of each antibody that was used as part of the sandwich assay. The analyte to be measured was added at a high, medium and low concentration. The different analyte concentrations were used to identify the detection antibody and capture antibody.

Table 6 illustrates results of the determination of the capture antibody concentration for HIV. Indicated on the table are absorbance readings for the ELISA tests performed. Microtiter plate wells were coated with antibodies concentrations of 0.5 μg/ml, 1 μg/ml, 2 μg/ml and 5 μg/ml. Different target analyte concentrations were added as shown in the table. The 0.5 μg/ml antibody concentration, showed the highest absorbance readings indicating the best antibody concentration to be adopted.

Table 6 Absorbance readings for HIV capture antibody determination

HIV capture antibody concentrations (µg/ml)	Absorbance readings		
	Target analyte (low concentration)	Target analyte (medium concentration)	Target analyte (high concentration)
0.5	1.523	1.260	1.763
1	0.913	0.741	0.739
2	0.589	0.499	0.416
5	0.505	0.455	0.563

The absorbance readings for HIV detection antibody concentrations are shown in table 7. Concentrations of 0.5 µg/ml, 1 µg/ml, 2 µg/ml and 5 µg/ml of the detection antibody were experimented with against different concentrations of the target analyte. The detection antibody concentration of 1 µg/ml produced consistent results.

Table 7 Absorbance readings for HIV detection antibody determination

HIV detection antibody concentrations (µg/ml)	Absorbance readings		
	Target analyte (low concentration)	Target analyte (medium concentration)	Target analyte (high concentration)
0.5	1.726	0.174	0.198
1	1.921	1.838	1.925
2	1.590	1.359	1.317
5	1.081	0.918	0.733

The HCV capture antibody determination is shown in table 8. As in HIV antibody concentrations determination, 0.5 μg/ml, 1 μg/ml, 2 μg/ml and 5 μg/ml of the capture antibody were experimented with, against different analyte concentrations. The 0.5 μg/ml antibody concentration gave the highest reading and was adopted.

Table 8 Absorbance readings for HCV capture antibody determination

HCV capture antibody concentrations (μg/ml)	**Absorbance readings**		
	Target analyte (low concentration)	**Target analyte (medium concentration)**	**Target analyte (high concentration)**
0.5	1.487	1.234	1.781
1	0.874	0.739	0.743
2	0.550	0.488	0.404
5	0.518	0.493	0.566

HCV detection antibody determination is illustrated in table 9. HCV detection antibody concentrations of 0.5 μg/ml, 1 μg/ml, 2 μg/ml and 5 μg/ml were experimented with. The experiment also showed that 0.5 μg/ml was ideal for the HCV detection antibody.

Table 9 Absorbance readings for HCV detection antibody determination

HCV detection antibody concentrations (μg/ml)	**Absorbance readings**		
	Target analyte (low concentration)	**Target analyte (medium concentration)**	**Target analyte (high concentration)**
0.5	1.748	1.554	1.582
1	1.074	1.139	1.243
2	0.850	1.088	0.704
5	0.418	0.593	0.566

4.4.3 Assembling of the Lateral Flow Immunoassay Strip

Assembling of the LFIA-based HIV/HCV multiplexed POC device consists of a sample pad, conjugate pad, immobilized nitrocellulose membrane, and absorbent pad pasted, in sequence, with 1-2 mm of overlap. The configuration is shown in figure 19. The assembled pads are inserted in a plastic cassette as shown in figure 20. However the capture antibody, the detection antibody and the indicator system are sprayed onto the strip before insertion into the cassette. The spraying was however not done at this stage, and will be performed at the PhD level.

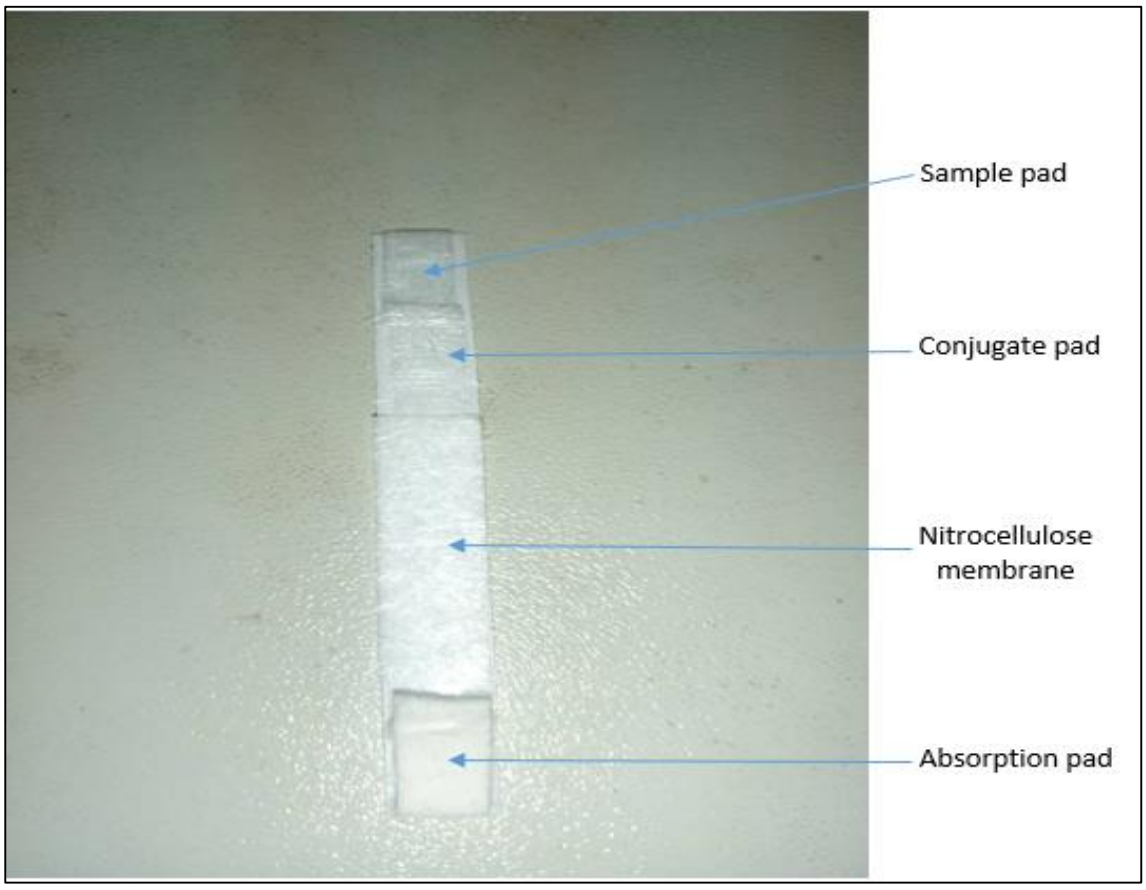


Figure 19 Assembled lateral flow immunoassay strip

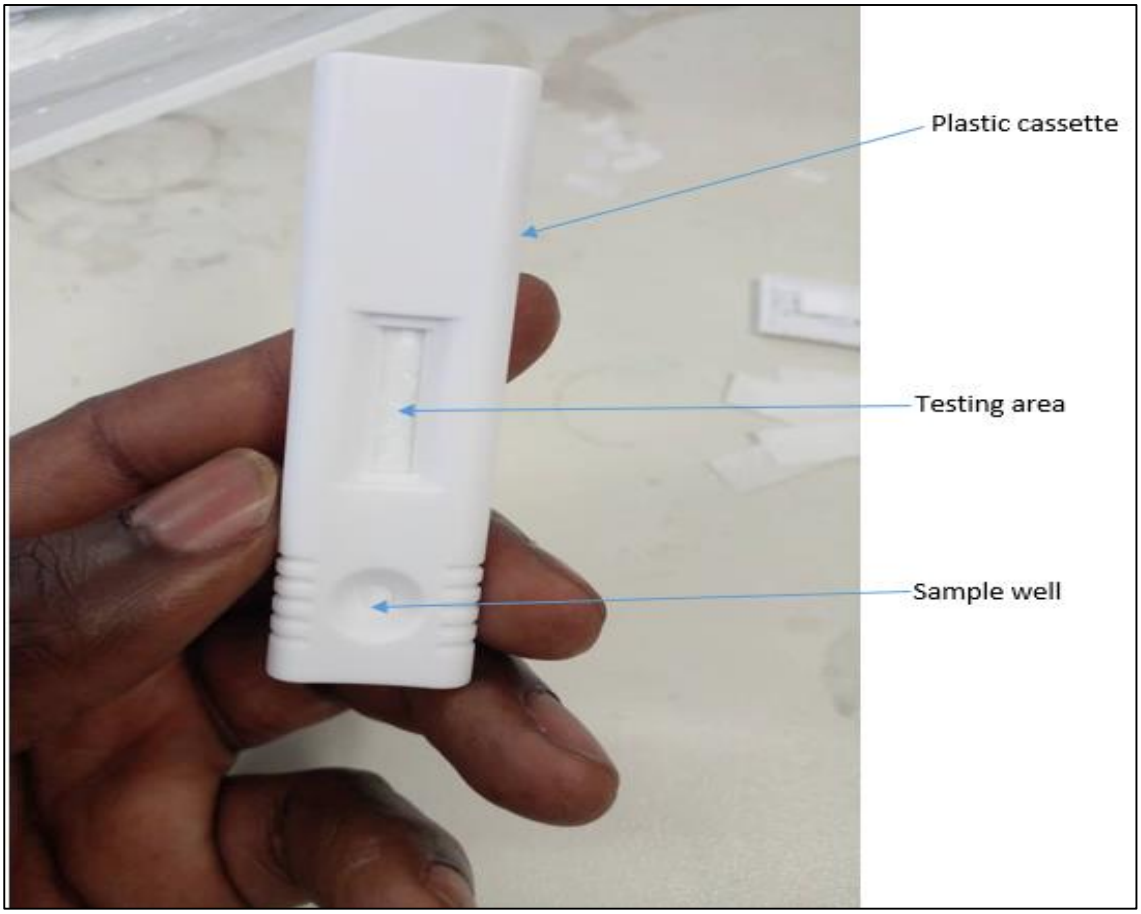


Figure 20 3D HIV/HCV multiplexed point-of-care prototype device

4.5 Chapter Summary

The majority of the participants who responded to the survey on determining the healthcare professionals' perspectives on multiplexed POC devices had more than 5 years of working experience. Accuracy of multiplexed POC devices and ease of use were the aspects considered the most important. Despite the unavailability of multiplexed POC devices, a number of benefits on their use were cited. The market analysis further highlighted the scarcity of multiplexed POC devices in SSA, and indicated that the demand for POC devices for infectious diseases will continue to rise in the future. Local production of multiplexed POC devices is therefore essential to promote mass screening

in limited-resource SSA. This project assembled the lateral flow immunoassay strip and determined the antibody concentrations. The prototype device was assembled.

CHAPTER 5: SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

5.1 Introduction

The purpose of this study was to develop a multiplexed POC device prototype based on LFIA that detects co-occurrence of HIV and HCV; conduct a market analysis for POC devices in SSA; and assess the perspectives of healthcare professionals on multiplexed POC devices at a key population clinic in Bulawayo, Zimbabwe. This chapter includes a discussion of major study findings as related to the literature. The chapter concludes with recommendations and suggestion on areas of further study.

5.2 Perspectives of Healthcare Professionals on Multiplexed Point-of-Care Testing Devices

This study revealed that healthcare professionals at Bambanani Newstart Centre accept the use of POC devices in the diagnosis and management of patients. Most participants agreed with statements relating to benefits of POC. Only a minority of participants registered possible concerns.

As expected, a larger proportion of participants agreed or strongly agreed that multiplexed POC devices are not readily available at Bambanani Newstart Centre. This is in tandem with findings by Teebagy *et al*. (2022) who perceived an unmet need for POC testing devices among the healthcare workers in their study. Furthermore, as expected the cost of POC tests was not a concern in this study. This is in agreement with findings by Dincer *et al*. (2017) who argued that POC testing devices are an invaluable cheaper option in limited-resource settings that can improve access to diagnostic testing.

Despite the positive perspectives from the healthcare professionals, the participants cited over-reliance on tests as a concern in the adoption of multiplexed POC devices. It was also evident in the findings that factors such as accuracy and ease of use are critical for a multiplexed POC device. This agrees with findings by Reipold *et al*., (2017) who indicated that an acceptable POC device has to meet the ASSURED rigmarole.

5.3 Market Analysis on Point-of-Care Devices

The study findings came out as anticipated in terms of global and regional demand for POC devices. For the included studies, high burden of infectious disease in SSA and the emergence of non-communicable diseases raise the demand for POC devices. The COVID-19 pandemic has undoubtedly played a paramount role in the size of the global POC industry. Furthermore, Africa is expected to provide the largest market for POC devices for infectious diseases by 2030.

Despite major companies initiating strategic product launches to strengthen their market presence, the manufacture of POC devices that detect HCV lags behind and that puts HCV on the list of neglected diseases in SSA. The increase in prevalence of chronic diseases, and the existing burden of HIV and HCV require the availability of POC devices that provide a more affordable diagnostic alternative. Technologies for the diagnosis of HIV and HCV are available but still too expensive for wide use in SSA due to cost constraints. However, to ensure an effective public health approach that tackles HIV and HCV in SSA, an increase in the supply of multiplexed POC devices through local manufacturing is key

Despite SSA offering wide market segments for HIV/HCV multiplexed POC devices, barriers that hinder local manufacturing need to be eliminated. The most serious threat for

the device is competition from imported devices. However, a local POC device can compete well given that the cost of the device will exclude shipping costs. Other barriers mainly revolves around capital requirements to scale-up the manufacturing due to scarcity of local funding. In addition, access to distribution channels is restricted for new entries but with adequate funding and advertisement, the new product may be competitive.

5.4 Development of the HIV/HCV Multiplexed POC Device

A presentation of the LFIA membrane strip and cassette holder of the HIV/HCV multiplexed POC device was successfully developed. The mode of multiplexing that involves using multiple strips supplied by a single sample well was adopted. The concentration of the capture and detection antibody was successfully determined using serial dilutions in ELISA. This approach could contribute to a new generation of multiplexed field friendly technology for improving the diagnostic precision for infectious diseases and their applicability in low-resources settings.

5.5 Limitation of the Study

The study exceeds Africa University's time schedule for a dissertation due to the volume of work involved. Additionally, the study focuses on the LFIA technology only, and does not experiment with other technologies that can be adopted on multiplexed POC devices. Despite the limitations, this project offers an opportunity for further study and will be carried on to the PhD Level.

5.6 Conclusion

This study aimed to develop a HIV/HCV multiplexed 3D- printed prototype POC device, understand healthcare professionals' perspectives on multiplexed POC devices; and

perform a market analysis on the POC industry. Based on the quantitative and qualitative analysis performed it can be concluded that healthcare professionals have an unmet need for multiplexed POC testing devices. Despite a few concerns such as unavailability of multiplexed POC devices and potential over-reliance on tests for clinicians, a significant number of benefits were cited in the survey. Healthcare professionals highlighted that multiplexed POC devices improve patient management, ensure targeted treatment and offer a cheaper diagnostic option.

The results of the market analysis indicate that there is low supply of multiplexed POC devices for mass testing in SSA especially for HIV and HCV. The multiplexed POC devices available in the African market are manufactured outside SSA and are expensive for wide use. A ready market was identified for cheaper technologies in SSA such as the HIV/HCV multiplexed POC devices this project is developing. In this study major progress was made in the development of the HIV/HCV multiplexed POC device, especially on identifying mode of multiplexing and antibody concentrations. The study is however still ongoing.

5.7 Recommendations

The researcher recommends a multi-sectoral approach and greater collaboration among industry partners and healthcare professionals. Synergy of efforts among the Ministry of industry and commerce, universities and private organisations such as Datlabs, and CAPS pharmaceuticals is crucial. This promotes innovation by providing a conducive environment and the necessary support.

The researcher recommends local universities to facilitate scientific advancement through creation of innovation hubs. Availability of necessary laboratory equipment and funding eliminate barriers to innovation. Additionally, constant revision of the education curriculum to keep up with current scientific trends is a necessity.

Mobilisation of funds by the Ministry of finance and a significant budget allocation also supports local innovation. State universities and other tertiary institution are underfunded resulting in poorly equipped laboratories that fail to support innovation.

The researcher also recommends the implementation of restrictions on imported POC devices so as to minimise competition with local products. Importantly entry of cheap and sub-standard POC devices into the country should not be tolerated. Lastly creation of a friendly environment without corruption and exorbitant fees is equally crucial.

REFERENCES

Anfossi, L., Di Nardo, F., Cavalera, S., Giovannoli, C., & Baggiani, C. (2018). Multiplexed lateral flow immunoassay: an overview of strategies towards high-throughput point-of-need testing. *Biosensors, 9*(1), 2-4.

Araz, M. K., Tentori, A. T., & Herr, A. E. (2013). Microfluidic multiplexing in bioanalyses. *Journal of Laboratory Automation, 18*(5), 350-366.

Biopanda. (2021). *HIV, HBsAg, HCV & Syphilis Combo Rapid Test*. Retrieved from https://www.biopanda.co.uk/php/products/rapid/infectious_diseases/hhhs_combo.php

Clark, T. J. (2002). The triage cardiac panel. *Point Care Journal of Near-Patient Testing and Technology, 1*(1), 42-46.

Daniel, E. (2016). The Usefulness of qualitative and quantitative approaches and methods in researching problem-solving ability in science education curriculum. *Journal of Education and Practice, 7*(15), 91-100.

Di Nardo, F., Chiarello, M., Cavalera, S., Baggiani, C., & Anfossi, L. (2021). Ten Years of Lateral Flow Immunoassay Technique Applications: Trends, Challenges and Future Perspectives. *Sensors, 21*(15), 5185.

Dincer, C., Bruch, R., Kling, K., Dittrich, P., & Urban, G. (2017). Multiplexed Pointof-Care Testing-xPOCT. *Trends in Biotechnology, 7*(15), 728-742.

Ding, E., Ensom, E., Hafer, N., Buchholz, B., Picard, M., Dunlap, D., . . . Lilly, C. (2019). Point-of-care technologies in heart, lung, blood, and sleep disorders from the Centre for advanced point-of-care technologies. *Current Opinion in Biomedical Engineering, 11*(5), 58-67.

Ding, E., Marcus, G., & McManus, D. (2020). Emerging technologies for identifying atrial fibrillation. *127*(1), 128-142.

Drain, P. K., & Garrett, N. J. (2015). The arrival of a true point-of-care molecular assay-ready for global implementation. *Lancet Global Health, 3*(11), 663-664.

Drain, P. K., & Rousseau, C. (2017). Point-of-care diagnostics extending the laboratory network to reach the last mile. *HIV and Diagnostics, 12*(2), 175-181.

Dunlap, D., Ding, E., Abramo, K., Wang, Z., Lilly, C., Haffer, N., . . . McManus, D. (2021). Point-of-care testing, your cardiologist and affairs of the heart. *Cardiovascular Digital Health Journal, 2*(10), 331-335.

Falchetta, G., Hammad, A. T., & Shayegh, S. (2020). Planning universal accessibility to public health care in sub-Saharan Africa. *Proceedings of the Natitional Academy of Sciences, 117*(15), 31760-31769.

Hanafiah, K. M., Arifin, N., Butsami, Y., Noordin, R., Garcia, M., & Anderson, D. (2017). Development of multiplexed infectious diseases lateral flow assay: challenges and opportunities. *Diagnostics, 7*(3), 51.

Hanbi, K., & Doo-Ryeon, C. (2019). A new point-of-care test for the diagnosis of infectious diseases based on multiple lateral flow immunoassays. *Analyst, 144*(8), 2460-2466.

Heidt, B., Siqueira, W. F., Eersels, K., Dilien, H., Van Grinsven, B., Fujiwara, R. T., & Cleij, T. J. (2020). Point of care diagnostics in resources-limited setting: A review the present and future of POC in its most needed environment. *Biosensors, 10*(10).

Jung, W., Han, J., Choi, J., & Ahn, C. H. (2015). Point-of-care testing (POCT) diagnostics systems using microfluidic lab-on-a-chip technologies. *Microelectronic Enginering, 132*(10), 46-57.

Kalla, G. C., Voundi, E. V., Guiadem, R., Lii, A. F., Belec, L., & Mbopi-Keou, F. X. (2018). Mass campaigns for HIV, HBV and HCV screening by multiplexed rapid diagnostics test in sub-Saharan Africa using mobile units: the game changer. *International Jornal of Infectious Diseases, 79*(1), 256.

Karoney, M. J., & Siika, A. M. (2013). Hepatitis C Virus infections in Africa: a review. *Pan Africa Medical Journal, 14*(44), 2199.

Kedar, V., Sewram, V., Ndlovu, N., Kimani, S., Abdelaziz, A. O., Chiao, E. Y., & Abou-Alfa, G. K. (2021). Hepatocellular carcinoma in sub-Saharan Africa. *JCO Global Oncology, 7*(4), 756-766.

Kim, H., Chungu, D., & Kang, M. (2019). A new point-of-care test for the diagnosis of infectious diseases based on multiplex lateral flow immunoassay. *Analyst, 144*(8), 2460-2466.

Kumar, S., Nehra, M., Khurana, S., Dilbaghi, N., Kumar, V., Kaushik, A., & Kim, K. H. (2020). Aspects of point-of-care diagnostics for personalised health wellness. *Done Press, 16*(1), 383-402.

Kuupiel, D., Bawontuo, V., Drain, P. K., Gwala, N., & Mahamba-Thompson, T. P. (2019). Supply chain management and accessibility to point-of-care testing in resources-limited settings: a systematic scoping review. *BMC Health Services Research, 19*(1), 519.

Lanini, S., Easterbrook, P., Zumla, A., & Ippolito, G. (2016). Hepatitis C: global epidemiology and strategies for control. *Clinical Microbiology and Infection, 22*(10), 833-838.

Leidner, A. J., Chesson, H. W., Xu, F., Ward, J. W., Spradling, P. R., & Holmberg, S. D. (2015). Cost-effectiveness of hepatitis C treatment for patients in early stages of liver disease. *Hepatology, 61*(6), 1860-1869.

Leman, R., Ralph-Sydney, M. B., Zita, A. N., Laura, C., Mathieu, M., Sylvie, S., . . . Laurent, B. (2018). Analytical performanceof simulteneous detection of HIV-1, HIV-2 and hepatits C-specific antibodies and hepatitis B surface antigen (HBsAG) by multiplex immunochromatographic rapid test with serum samples: a cross-sectional study. *Journal of Virological Methods, 253*(2), 1-4.

Leoni, M. C., Ustianowski, A., Farooq, H., & Arendes, J. E. (2018). HIV, HCV and HBV: A review of parallels and differences. *Infectious Diseases and Therapy, 7*(8), 407-419.

Liu, W., Li, Y., Liu, J., Niu, X., Wang, Y., & Li, D. (2013). Application and performance of 3D printing in nanobiomaterials. *Journal of Nanomaterials, 14*(13), 21-23.

Lohia, V;. (2020). *World hepatitis day 2020: finding the missing millions*. Retrieved from, https://www.clintonhealthaccess.org/world-hepatitis-day-2020-finding-the-missing-millions/

Luppa, P. B. (2016). Clinical relevant analytical techniques, organisational concepts for application and future perspectives of point-of-care testing. *Biotechnology Advances, 34*(3), 139-160.

Medmira;. (2021). *Multiplo HBc/HIV/HCV product sheet*. Retrieved from https://pdf.medicalexpo.com/pdf/medmira/multiplo-hbc-hiv-hcv-product-sheet/108366-151951.html

Mora, N., Adam, W. H., Kliethermes, S., Dugas, L., Balasubramanian, N., Sandhu, J., . . . Layden, J. E. (2016). A synthesis of hepatitis C prevalence estimates in sub-Suharan Africa. *BMC Infectious Diseases, 16*(2), 283.

Nichols, J. H. (2021). Utilising point-of-care testing to optimise patient care. *Journal of the International Federation of Clinical Chemistry and Laboratory Medicine, 32*(2), 140-144.

Nkengasong, J. N., & Tessema, S. K. (2020). Africa needs a new public health order to tackle infectious disease threat. *Elsevier Public Health Emergency Collection, 183*(2), 296-300.

Rajan, A., & Glorikian, H. (2021). Point-of-care diagnostics: market trends and growth drivers. *Expert Opinion on Medical Diagnostics, 3*(1), 1-4.

Reipold, E. I., Easterbrook, P., Trianni, A., Panneer, N., Krakower, D., Ongarello, S., & Denkinger, C. (2017). Optimising diagnosis of viraemic hepatitis C infection: the development of a target product profile. *BMC Infectious Diseases, 17*(1), 707.

Sajid, M., Kawdea, A. N., & Daud, M. (2014). Designs, formats and applications of lateral flow assay: a literature review. *Journal of Saudi Chemical Society, 19*(6), 689-705.

Saylan, Y., & Denizli, A. (2019). Molecular imprinted polymer-based microfluidic system for point-of-care applications. *Micromachines, 10*(11), 766.

Schönberger, M., & Hoffstetter, M. (2016). *Emerging trends in medical plastic engeneering and manufacturing.* Waldachtal, Germany: Elsevier Science and Technology.

Schroeder, L. F., & Amukele, T. (2014). Medical laboratories in sub-Saharan Africa that meet international quality standards. *America Journal of Clinical Pathology, 141*(6), 791-795.

Setia, M. (2016). Methodology series module 3: cross-sectional studies. *Indian Journal of Dermatology, 61*(3), 261-264.

Sonderup, M. W., Afihene, M., Ally, R., Apica, B., Awuku, Y., & Cunha, L. (2017). Hepatitis C in sub-Saharan Africa: the current status and recommendations for achieveing elimination by 2030. *Gastroenterology and Hepatology, 2*(12), 910-919.

Taranova, A. N., Byzova, N., Zaiko, V. V., Starovoitova, T. A., Vengerov, Y. Y., Zherdev, A. V., & Dzantiev, B. B. (2013). Integration of lateral flow and microarray technologies for multiplex immunoassay: application to the determination of drugs abuse. *Microchimica Acta, 180*(1), 11-12.

Teebagy, S., Wang, Z., Dunlap, D., Saleeba, C., DiMezza, D., Crain, J., . . . Hafer, N. (2022). Understanding healthcare professionals' perspectives on point-of-care testing. *Diagnostics, 12*(2), 533.

Tonen-Wolyec, S., Djang'eing'a, R. M., Batina-Agasa, S., Kayembe-Tshilumba, C., Muwonga-Masidi, J., Hayette, M. P., & Belec, L. (2021). Self-testing for HIV, HBV and HCV using whole-blood multiplex immunochromatographic rapid test: a pilot feasibility study in sub-Saharan Africa. *PLOSONE, 16*(4), 14-19.

Webster, D. P., Klenerman, P., & Dusheiko, G. M. (2015). Hepatis C. *Lancet, 385*(9973), 1124-1135.

World Health Organisation. (2020). *Hepatitis C*. Retrieved from https://www.who.int/news-room/ fact-sheets/detail/hepatitis-c.

World Health Organisation. (2021). *Target product profile*. Retrieved from https://www.who.int/observatories/global-observatory-on-health-research-and-development/analyses-and-syntheses/target-product-profile/who-target-product-profiles

Zhao, Y., Wang, H., Zhang, P., Sun, C., Wang, X., Wang, X., . . . Zhou, L. (2016). Rapid multiplex detection of 10 foodborne pathogens with an upcovering phosphor technology-based 10-channel lateral flow assay. *Scientific Reports, 6*(2), 21342.

APPENDICES

APPENDIX 1: Budget for the Development of HIV/HCV Multiplexed Point-of-Care Device

#	Category	Description	Quantity	Freq	Rate (US$)	Total (US$)
1	Equipment	Colorimeter	1	1	1000.00	1000.00
		3D printer	1	1	2000.00	2000.00
		Spectrophotometer	1	1	667.00	667.00
2	Consumables	Pad paper	1	1	1333	1333.00
		Conjugate protein kit	1	1	667.00	667.00
		HCV/HIV viral antigens	2	1	333.50	667.00
		Gold/silver colloids	4	1	166.75	667.00
		HCV/HIV positive and negative sera	2	1	333.50	667.00
		Immunoassay reagents	5	1	133.40	667.00
3	Consultations	Paper strip design	1	1	1000.00	1000.00
		Current industrial trends on Viral protein probes	1	1	333.00	333.00
		3D-printing of cartridge	5	1	267.00	1335.00
		Review technologies on multiplexed POC devices	1	1	667.00	667.00
5	Business plan	Business plan for the scale up phase	1	1	2133.00	2133.00
Sub-total						**13535.00**
	Vat @ 15%					**2030.35**
TOTAL COST OF WORKS						**15565.25**

APPENDIX 2: Work Plan for the Project

	YEAR											
	2021							2022				
ACTIVITY	**Jun**	**Jul**	**Aug**	**Sep**	**Oct**	**Nov**	**Dec**	**Jan**	**Feb**	**Mar**	**Apr**	**May**
Identify Research area												
Formulate research Topic												
Literature review												
Proposal writing												
Proposal defense												
Submission of Proposal to AUREC												
Market analysis												
POC device design												
Work on patent ideas												
Final Report Submission and publication												

APPENDIX 3: Questionnaire for the Study

Welcome!

Please answer the questions below honestly. We want your expert opinion about what qualities of multiplexed point-of-care devices (POC) that are most important to healthcare providers. Moreover, we would like to better understand the decision-making process of implementing these new technologies into everyday practice. Multiplexed POC are tests that detect more than one infections and can be done onsite with results available during the visit and before the patient leaves. The information collected below will not be linked to your confidential personal information. When we publish the results of this study, your confidential personal information will not be shown. Our hope is that the results of this survey will identify areas of need and encourage new research and development.

SECTION A

1. What is your gender?

◡ Male ◡ Female ◡ Other (Specify)……

2. What is your race?

◡ Black ◡ White ◡ Other (Specify)……

3. What is your profession?

◡ Nurse ◡ Lab Scientist ◡ Nurse assistant ◡ Pharmacy technician

4. How many years have you practiced after completing your terminal training/degree?

◡ 0-5 ◡ 6-10 ◡ 11-15 ◡ 16-20

SECTION B

1. Which characteristic of a multiplexed POC technology is the most (1st) important when incorporating it into your regular?

- Availability
- Ease of use
- Sample type
- Accuracy
- Cost
- Does not affect workflow.
- Reimbursement for testing

2. Which characteristic of a multiplexed POC technology is the second most important when incorporating it into your regular practice?

- Availability
- Ease of use
- Sample type
- Accuracy
- Cost
- Does not affect workflow.
- Reimbursement for testing

3. Which characteristic of a multiplexed POC technology is the third most important when incorporating it into your regular practice?

- Availability
- Ease of use
- Sample type
- Accuracy

◡ Cost

◡ Does not affect workflow.

◡ Reimbursement for testing

SECTION C

Do you agree with the following benefits of multiplexed POC devices? Please select one response.

1. Multiplexed POC testing improve patient satisfaction.

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

2. Multiplexed POC ensure diagnostic certainty.

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

3. Multiplexed POC reduce the need to refer patient to lab

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

4. Multiplexed POC improve clinician confidence in decision making.

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

5. Multiplexed POC improve patient management.

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

6. Multiplexed POC offer a cheaper diagnostic option

Strongly agree Agree Neutral Disagree Strongly Disagree

7. Multiplexed POC ensure targeted treatment

Strongly agree Agree Neutral Disagree Strongly Disagree

8. Multiplexed POC save time for diagnosis

Strongly agree Agree Neutral Disagree Strongly Disagree

9. Multiplexed POC reduce error

Strongly agree Agree Neutral Disagree Strongly Disagree

10. Multiplexed POC improve provider-patient relationship

Strongly agree Agree Neutral Disagree Strongly Disagree

SECTION D

1. Multiplexed POC result is difficult to discuss with patient

Strongly agree Agree Neutral Disagree Strongly Disagree

2. Multiplexed POC result not conclusive

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

3. Multiplexed POC not readily available

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

4. Multiplexed POC device cost too high

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

5. Staff training cost for multiplexed POC too high

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

6. Multiplexed POC cause over-reliance on tests

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

7. Multiplexed POC result not good enough to make clinical decision

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

8. Multiplexed POC lead to over-testing.

◡ Strongly agree ◡ Agree ◡ Neutral ◡ Disagree ◡ Strongly ◡ Disagree

APPENDIX 4: Consent Form

Study title: Developing a multiplexed 3D-printed prototype point-of-care testing device for HIV and hepatitis C in high-risk populations

My name is Stanford Chigaro, and I am a Master of Public Health student at Africa University. As part of my dissertation, I am researching to understand the perspectives of healthcare professionals on multiplexed point-of-care devices. I am kindly asking you to participate in this study by responding to my questionnaire.

Purpose of the study

The purpose of the study is to identify the benefits that multiplexed point-of-care devices have, from the healthcare professional's perspective. The study also seek to identify the concerns, healthcare professionals have over the use of multiplexed point-of-care devices, and the aspects they consider crucial about the devices. This will guide the development of the HIV/HCV multiplexed point-of-care device this dissertation proposes to develop.

Procedures and duration

The eligible participants for this study are healthcare professionals at Bambanani newstart centre. You have been selected as a possible participant because you meet the stated selection criteria. If you decide to participate, you will be asked to respond to the survey. The survey will take about 10 minutes.

Risks and discomforts

We anticipate no harm/risk/discomfort to occur during the survey. Privacy and confidentiality will be observed and protected. The survey will take place in private.

Benefits

There are no costs or direct benefits to you for participating in this study. You are free to ask for further clarification as need be.

Confidentiality

If you participate in the study, you will be assigned a participant identity to be used on the questionnaire as no personal details will appear on the questionnaire. Any information that is obtained in connection with this study that can be identified with you will remain confidential and will be disclosed only with your permission. All study records will be kept secure, separate from any information that identifies you personally like this consent form. Your name will not be used in any reports or publications that may arise from this study. Under some circumstances, the University or Medical Research Council of Zimbabwe may need to review records for compliance audits only.

Voluntary participation

Participation in this study is voluntary. If you decide not to participate in this study, your decision will not affect your future relationship with the researcher. If you chose to participate, you are free to withdraw your consent and to discontinue participation without penalty at any time.

Questions

Before you sign this form, please ask any questions on any aspect of this study that is unclear to you.

Authorisation

If you have decided to participate in this study, please sign this form in the space provide below as an indication that you have read and understood the information provided above and have agreed to participate.

---------- ----------

Name of Research Participant (please print) Date

APPENDIX 5: Mixed Methods Appraisal Tool (Hong *et al*., 2018)

Part I: Mixed Methods Appraisal Tool (MMAT), version 2018					
Category of study designs	**Methodological quality criteria**	**Responses**			
		Yes	**No**	**Can't tell**	**Comments**
Screening questions (for all types)	S1. Are there clear research questions? S2. Do the collected data allow to address the research questions? Further appraisal may not be feasible or appropriate when the answer is 'No' or 'Can't tell' to one or both screening questions				
1. Qualitative	1.1. Is the qualitative approach appropriate to answer the research question? 1.2. Are the qualitative data collection methods adequate to address the research question? 1.3. Are the findings adequately derived from the data? 1.4. Is the interpretation of results sufficiently substantiated by data? 1.5. Is there coherence between qualitative data sources, collection, analysis and interpretation?				
2. Quantitative randomized controlled trials	2.1. Is randomization appropriately performed? 2.2. Are the groups comparable at baseline? 2.3. Are there complete outcome data? 2.5 Did the participants adhere to the assigned intervention?				
3. Quantitative non-randomized	3.1. Are the participants representative of the target population? 3.2. Are measurements appropriate regarding both the outcome and intervention (or exposure)? 3.3. Are there complete outcome data? 3.4. Are the confounders accounted for in the design and analysis?				

APPENDIX 5 (Continued)

Category of study designs	Methodological quality criteria	Responses			
		Yes	No	Can't tell	Comments
4. Quantitative descriptive	4.1. Is the sampling strategy relevant to address the research question? 4.2. Is the sample representative of the target population? 4.3. Are the measurements appropriate? 4.4. Is the risk of nonresponse bias low? 4.5. Is the statistical analysis appropriate to answer the research question				
5. Mixed methods	5.1. Is there an adequate rationale for using a mixed methods design to address the research question? 5.2. Are the different components of the study effectively integrated to answer the research question? 5.3. Are the outputs of the integration of qualitative and quantitative components adequately interpreted? 5.4. Are divergences and inconsistencies between quantitative and qualitative results adequately addressed? 5.5. Do the different components of the study adhere to the quality criteria of each tradition of the methods involved				

APPENDIX 5 (Continued)

<table>
<tr><th colspan="2">Part II: Explanations</th></tr>
<tr><td>1. Qualitative studies</td><td>Methodological quality criteria</td></tr>
<tr><td rowspan="2">Qualitative research is an approach for exploring and understanding the meaning individuals or groups ascribe to a social or human problem.

Common qualitative research approaches include (this list if not exhaustive):

Ethnography
The aim of the study is to describe and interpret the shared cultural behaviour of a group of individuals.

Phenomenology
The study focuses on the subjective experiences and interpretations of a phenomenon encountered by individuals

Narrative research
The study analyzes life experiences of an individual or a group.</td><td>1.1. Is the qualitative approach appropriate to answer the research question?

Explanations
The qualitative approach used in a study (see non-exhaustive list on the left side of this table) should be appropriate for the research question and problem. For example, the use of a grounded theory approach should address the development of a theory and ethnography should study human cultures and societies.

This criterion was considered important to add in the MMAT since there is only one category of criteria for qualitative studies (compared to three for quantitative studies).</td></tr>
<tr><td>1.2. Are the qualitative data collection methods adequate to address the research question?

Explanations
This criterion is related to data collection method, including data sources (e.g., archives, documents), used to address the research question. To judge this criterion, consider whether the method of data collection (e.g., in depth interviews and/or group interviews, and/or observations) and the form of the data (e.g., tape recording, video material, diary, photo, and/or field notes) are adequate. Also, clear justifications are needed when data collection methods are modified during the study.</td></tr>
<tr><td></td><td>1.3. Are the findings adequately derived from the data?</td></tr>
</table>

APPENDIX 5 (Continued)

Part II	
1. Qualitative studies	**Methodological quality criteria**
Grounded theory Generation of theory from data in the process of conducting research (data collection occurs first).	Explanations This criterion is related to the data analysis used. Several data analysis methods have been developed and their use depends on the research question and qualitative approach. For example, open, axial and selective coding is often associated with grounded theory, and within- and cross-case analysis is often seen in case study.
Case study In-depth exploration and/or explanation of issues intrinsic to a particular case. A case can be anything from a decision-making process, to a person, an organization, or a country.	1.4. Is the interpretation of results sufficiently substantiated by data? Explanations The interpretation of results should be supported by the data collected. For example, the quotes provided to justify the themes should be adequate.
Qualitative description There is no specific methodology, but a qualitative data collection and analysis, e.g., in-depth interviews or focus groups, and hybrid thematic analysis (inductive and deductive).	1.5. Is the interpretation of results sufficiciently substantiated by data? Explanations There should be clear links between data sources, collection, analysis and interpretation.

APPENDIX 5 (Continued)

2. Quantitative randomized controlled trials	**Methodological quality criteria**
Randomized controlled clinical trial: A clinical study in which individual participants are allocated to intervention or control groups by randomization (intervention assigned by researchers).	2.1. Is randomization appropriately performed? Explanations In a randomized controlled trial, the allocation of a participant into the intervention or control group is based solely on chance. Researchers should describe how the randomization schedule was generated. A simple statement such as 'we randomly allocated' or 'using a randomized design' is insufficient to judge if randomization was appropriately performed. Also, assignment that is predictable such as using odd and even record numbers or dates is not appropriate. At minimum, a simple allocation should be performed by following a predetermined plan/sequence. It is usually achieved by referring to a published list of random numbers, or to a list of random assignments generated by a computer. Also, restricted allocation can be performed such as blocked randomization, stratified randomization, or minimization. Another important characteristic to judge if randomization was appropriately performed is allocation concealment that protects assignment sequence until allocation. Researchers and participants should be unaware of the assignment sequence up to the point of allocation. Several strategies can be used to ensure allocation concealment such relying on a central randomization by a third party, or the use of sequentially numbered, opaque, sealed envelopes
	2.4. Are outcome assessors blinded to the intervention provided? Explanations Outcome assessors should be unaware of who is receiving which interventions. The assessors can be the participants if using participant reported outcome, the intervention provider, or other persons not involved in the intervention.

APPENDIX 5 (Continued)

<table>
<tr><th>3. Quantitative non-randomized studies</th><th>Methodological quality criteria</th></tr>
<tr><td rowspan="3">Non-randomized studies are defined as any quantitative studies estimating the effectiveness of an intervention or studying other exposures that do not use randomization to allocate units to comparison groups

Common designs include (this list if not exhaustive):

Non-randomized controlled trials
The intervention is assigned by researchers, but there is no randomization, e.g., a pseudo-randomization. A non- random method of allocation is not reliable in producing
alone similar groups.

Cohort study
Subsets of a defined population are assessed as exposed, not exposed, or exposed at different degrees to factors of interest. Participants are followed over time to determine if
an outcome occurs (prospective longitudinal).</td><td>3.1. Are the participants representative of the target population?

Explanations
Indicators of representativeness include: clear description of the target population and of the sample (inclusion and exclusion criteria), reasons why certain eligible individuals chose not to participate, and any attempts to achieve a sample of participants that represents the target</td></tr>
<tr><td>3.2. Are measurements appropriate regarding both the outcome and intervention (or exposure)?

Explanations
Indicators of appropriate measurements include: the variables are clearly defined and accurately measured; the measurements are justified and appropriate for answering the research question; the measurements reflect what they are supposed to measure; validated and reliability tested measures of the intervention/exposure and outcome of interest are used, or variables are measured using ‘gold standard’.</td></tr>
<tr><td>3.3. Are there complete outcome data?

Explanations
Almost all the participants contributed to almost all measures. There is no absolute and standard cut-off value for acceptable complete outcome data. Agree among your team what is considered complete outcome data in your field (and based on the targeted journal) and apply this uniformly across all the included studies. For example, in the literature, acceptable complete data value ranged from 80% to 95%. Similarly, different acceptable withdrawal/dropouts rates have been suggested: 5%, 20% and 30% for follow-up of more than one year.</td></tr>
</table>

APPENDIX 5 (Continued)

3. Quantitative non-randomized studies	**Methodological quality criteria**
Case-control study Cases, e.g., patients, associated with a certain outcome are selected, alongside a corresponding group of controls. Data is collected on whether cases and controls were exposed to the factor under study (retrospective). **Cross-sectional analytic study** At one particular time, the relationship between health- related characteristics (outcome) and other factors (intervention/exposure) is examined. E.g., the frequency of outcomes is compared in different population subgroups according to the presence/absence (or level) of the intervention/exposure.	3.4. Are the confounders accounted for in the design and analysis? Explanations Confounders are factors that predict both the outcome of interest and the intervention received/exposure at baseline. They can distort the interpretation of findings and need to be considered in the design and analysis of a non-randomized study. Confounding bias is low if there is no confounding expected, or appropriate methods to control for confounders are used (such as stratification, regression, matching, standardization, and inverse probability weighting). 3.5. During the study period, is the intervention administered (or exposure occurred) as intended? Explanations For intervention studies, consider whether the participants were treated in a way that is consistent with the planned intervention. Since the intervention is assigned by researchers, consider whether there was a presence of contamination (e.g., the control group may be indirectly exposed to the intervention) or whether unplanned co-interventions were present in one group. For observational studies, consider whether changes occurred in the exposure status among the participants. If yes, check if these changes are likely to influence the outcome of interest, were adjusted for, or whether unplanned co-exposures were present in one group.

APPENDIX 5 (Continued)

4. Quantitative descriptive studies	**Methodological quality criteria**
Quantitative descriptive studies are concerned with and designed only to describe the existing distribution of variables without much regard to causal relationships or other hypotheses. They are used to monitoring the population, planning, and generating hypothesis. Common designs include the following single-group studies (this list if not exhaustive):	4.1. Is the sampling strategy relevant to address the research question? Explanations Sampling strategy refers to the way the sample was selected. There are two main categories of sampling strategies: probability sampling (involve random selection) and non-probability sampling. Depending on the research question, probability sampling might be preferable. Non- probability sampling does not provide equal chance of being selected. To judge this criterion, consider whether the source of sample is relevant to the target population; a clear justification of the sample frame used is provided; or the sampling procedure is adequate.
Incidence or prevalence study without comparison group In a defined population at one particular time, what is happening in a population, e.g., frequencies of factors (importance of problems), is described (portrayed).	4.2. Is the sample representative of the target population? Explanations There should be a match between respondents and the target population. Indicators of representativeness include: clear description of the target population and of the sample (such as respective sizes and inclusion and exclusion criteria), reasons why certain eligible individuals chose not to participate, and any attempts to achieve a sample of participants that represents the target population.
Survey Research method by which information is gathered by asking people questions on a specific topic and the data collection procedure is standardized and well defined.	4.3. Are the measurements appropriate? Explanations Indicators of appropriate measurements include: the variables are clearly defined and accurately measured, the measurements are justified and appropriate for answering the research question; the measurements reflect what they are supposed to measure; validated and reliability tested measures of the outcome of interest are used, variables are measured using ‘gold standard’, or questionnaires are pre-tested prior to data collection.

APPENDIX 5 (Continued)

4. Quantitative descriptive studies	**Methodological quality criteria**
Case series A collection of individuals with similar characteristics are used to describe an outcome. **Case report** An individual or a group with a unique/unusual outcome is described in detail.	4.4. Is the risk of nonresponse bias low? Explanations Nonresponse bias consists of an error of nonobservation reflecting an unsuccessful attempt to obtain the desired information from an eligible unit. To judge this criterion, consider whether the respondents and non- respondents are different on the variable of interest. This information might not always be reported in a paper. Some indicators of low nonresponse bias can be considered such as a low nonresponse rate, reasons for nonresponse (e.g., noncontacts vs. refusals), and statistical compensation for nonresponse (e.g., imputation). The nonresponse bias is might not be pertinent for case series and case report. This criterion could be adapted. For instance, complete data on the cases might be important to consider in these designs.
	4.5. Is the statistical analysis appropriate to answer the research question? Explanations The statistical analyses used should be clearly stated and justified in order to judge if they are appropriate for the design and research question, and if any problems with data analysis limited the interpretation of the results.

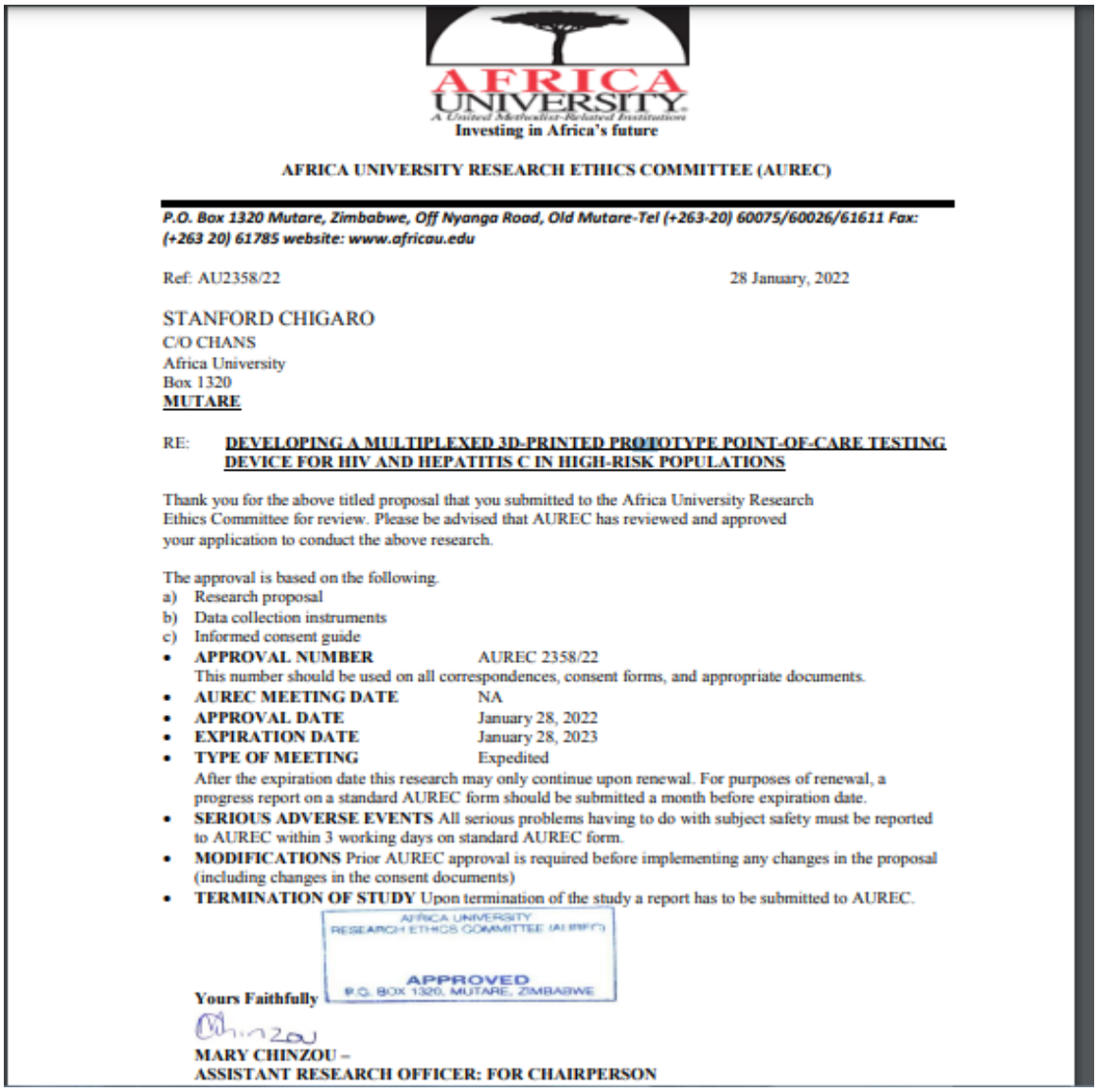
APPENDIX 5 (Continued)

5. Mixed methods studies	**Methodological quality criteria**
Mixed methods (MM) research involves combining qualitative (QUAL) and quantitative (QUAN) methods. In this tool, to be considered MM, studies have to meet the following criteria (Creswell and Plano Clark, 2017): (a) at least one QUAL method and one QUAN method are combined; (b) each method is used rigorously in accordance to the generally accepted criteria in the area (or tradition) of research invoked; and (c) the combination of the methods is carried out at the minimum through a MM design (defined *a priori*, or emerging) and the integration of the QUAL and QUAN phases, results, and data. Common designs include (this list if not exhaustive): **Convergent design** The QUAL and QUAN components are usually (but not necessarily) concomitant. The purpose is to examine the same phenomenon by interpreting QUAL and QUAN results (bringing data analysis together at the interpretation stage), or by integrating QUAL and QUAN datasets (e.g., data on same cases), or by transforming data (e.g., quantization of qualitative data).	5.1. Is there an adequate rationale for using a mixed methods design to address the research question? Explanations The reasons for conducting a mixed methods study should be clearly explained. Several reasons can be invoked such as to enhance or build upon qualitative findings with quantitative results and vice versa; to provide a comprehensive and complete understanding of a phenomenon or to develop and test instruments. 5.2. Are the different components of the study effectively integrated to answer the research question? Explanations Integration is a core component of mixed methods research and is defined as the explicit interrelatin of the quantitative and qualitative component in a mixed methods study. Look for information on how qualitative and quantitative phases, results, and data were integrated. For instance, how data gathered by both research methods was brought together to form a complete picture (e.g., joint displays) and when integration occurred (e.g., during the data collection-analysis or/and during the interpretation of qualitative and quantitative results). 5.3. Are the outputs of the integration of qualitative and quantitative components adequately interpreted? Explanations This criterion is related to meta-inference, which is defined as the overall interpretations derived from integrating qualitative and quantitative findings. Meta-inference occurs during the interpretation of the findings from the integration of the qualitative and quantitative components, and shows the added value of conducting a mixed methods study rather than having two separate studies.

APPENDIX 5 (Continued)

5. Mixed methods studies	**Methodological quality criteria**
Sequential explanatory design Results of the phase 1 - QUAN component inform the phase 2 – QUAL component. The purpose is to explain QUAN results using QUAL findings. E.g., the QUAN results guide the selection of QUAL data sources and data collection, and the QUAL findings contribute to the interpretation of QUAN results.	5.4. Are divergences and inconsistencies between quantitative and qualitative results adequately addressed? Explanations When integrating the findings from the qualitative and quantitative components, divergences and inconsistencies (also called conflicts, contradictions, discordances, discrepancies, and dissonances) can be found. It is not sufficient to only report the divergences; they need to be explained. Different strategies to address the divergences have been suggested such as reconciliation, initiation, bracketing and exclusion. Rate this criterion 'Yes' if there is no divergence.
Sequential exploratory design Results of the phase 1 - QUAL component inform the phase 2 - QUAN component. The purpose is to explore, develop and test an instrument (or taxonomy), or a conceptual framework (or theoretical model). E.g., the QUAL findings inform the QUAN data collection, and the QUAN results allow a statistical generalization of the QUAL findings.	5.5. Do the different components of the study adhere to the quality criteria of each tradition of the methods involved? Explanations The quality of the qualitative and quantitative components should be individually appraised to ensure that no important threats to trustworthiness are present. To appraise 5.5, use criteria for the qualitative component (1.1 to 1.5), and the appropriate criteria for the quantitative component (2.1 to 2.5, or 3.1 to 3.5, or 4.1 to 4.5). The quality of both components should be high for the mixed methods study to be considered of good quality. The premise is that the overall quality of a mixed methods study cannot exceed the quality of its weakest component. For example, if thequantitative component is rated high quality and the qualitative component is rated low quality, the overall rating for this criterion will be of low quality.

APPENDIX 6: Africa University Research Ethics Committee Approval letter



AFRICA UNIVERSITY
A United Methodist-Related Institution
Investing in Africa's future

AFRICA UNIVERSITY RESEARCH ETHICS COMMITTEE (AUREC)

P.O. Box 1320 Mutare, Zimbabwe, Off Nyanga Road, Old Mutare-Tel (+263-20) 60075/60026/61611 Fax: (+263 20) 61785 website: www.africau.edu

Ref: AU2358/22 28 January, 2022

STANFORD CHIGARO
C/O CHANS
Africa University
Box 1320
MUTARE

RE: **DEVELOPING A MULTIPLEXED 3D-PRINTED PROTOTYPE POINT-OF-CARE TESTING DEVICE FOR HIV AND HEPATITIS C IN HIGH-RISK POPULATIONS**

Thank you for the above titled proposal that you submitted to the Africa University Research Ethics Committee for review. Please be advised that AUREC has reviewed and approved your application to conduct the above research.

The approval is based on the following.
a) Research proposal
b) Data collection instruments
c) Informed consent guide

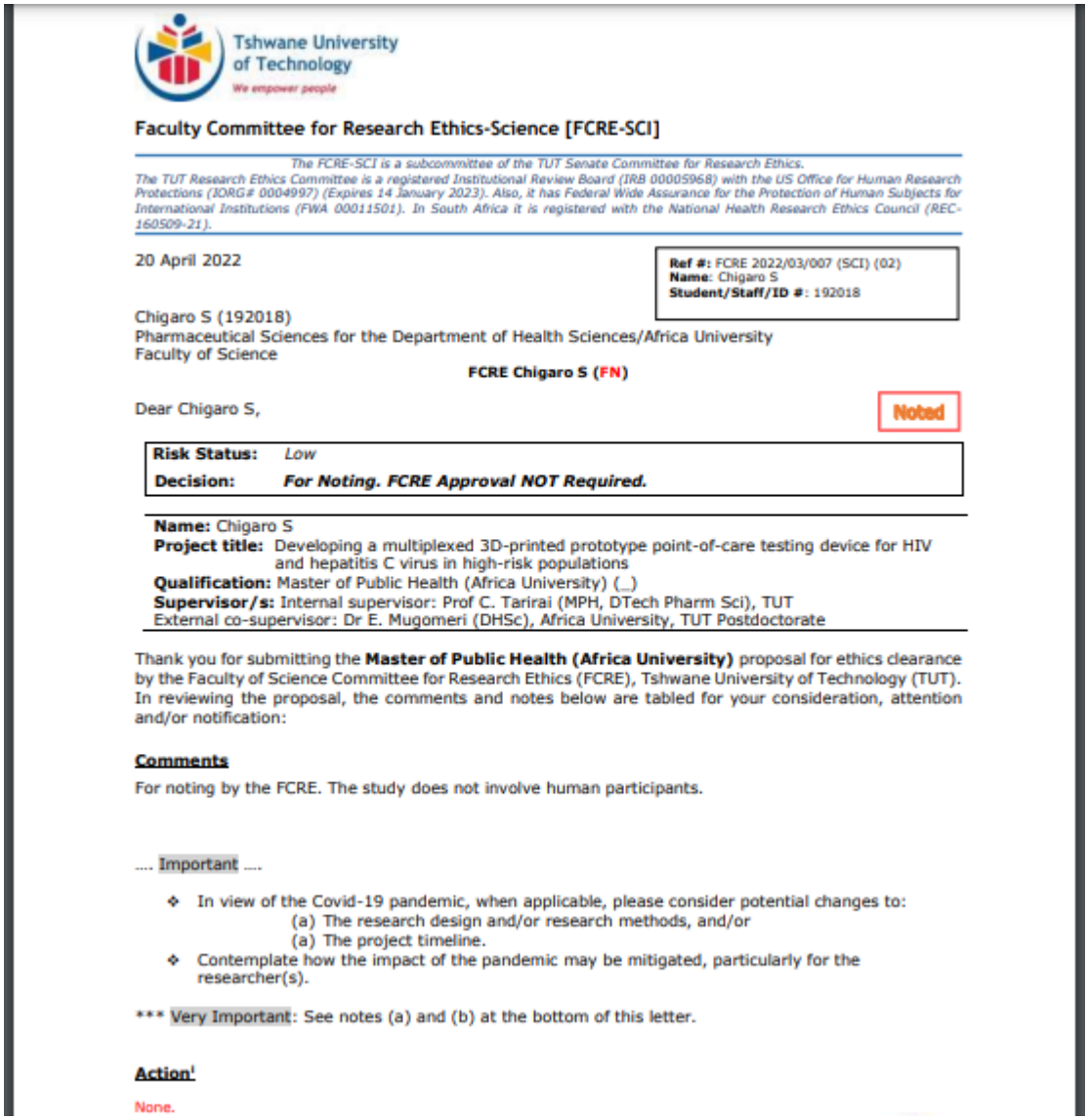
- **APPROVAL NUMBER** AUREC 2358/22
  This number should be used on all correspondences, consent forms, and appropriate documents.
- **AUREC MEETING DATE** NA
- **APPROVAL DATE** January 28, 2022
- **EXPIRATION DATE** January 28, 2023
- **TYPE OF MEETING** Expedited
  After the expiration date this research may only continue upon renewal. For purposes of renewal, a progress report on a standard AUREC form should be submitted a month before expiration date.
- **SERIOUS ADVERSE EVENTS** All serious problems having to do with subject safety must be reported to AUREC within 3 working days on standard AUREC form.
- **MODIFICATIONS** Prior AUREC approval is required before implementing any changes in the proposal (including changes in the consent documents)
- **TERMINATION OF STUDY** Upon termination of the study a report has to be submitted to AUREC.

AFRICA UNIVERSITY RESEARCH ETHICS COMMITTEE (AUREC) APPROVED P.O. BOX 1320, MUTARE, ZIMBABWE

Yours Faithfully

MARY CHINZOU –
ASSISTANT RESEARCH OFFICER: FOR CHAIRPERSON

APPENDIX 7: Tshwane University of Technology, Research Ethics Approval letter



Tshwane University of Technology
We empower people

Faculty Committee for Research Ethics-Science [FCRE-SCI]

The FCRE-SCI is a subcommittee of the TUT Senate Committee for Research Ethics.
The TUT Research Ethics Committee is a registered Institutional Review Board (IRB 00005968) with the US Office for Human Research Protections (IORG# 0004997) (Expires 14 January 2023). Also, it has Federal Wide Assurance for the Protection of Human Subjects for International Institutions (FWA 00011501). In South Africa it is registered with the National Health Research Ethics Council (REC-160509-21).

20 April 2022

Ref #: FCRE 2022/03/007 (SCI) (02)
Name: Chigaro S
Student/Staff/ID #: 192018

Chigaro S (192018)
Pharmaceutical Sciences for the Department of Health Sciences/Africa University
Faculty of Science

FCRE Chigaro S (FN)

Dear Chigaro S,

Noted

Risk Status: *Low*
Decision: ***For Noting. FCRE Approval NOT Required.***

Name: Chigaro S
Project title: Developing a multiplexed 3D-printed prototype point-of-care testing device for HIV and hepatitis C virus in high-risk populations
Qualification: Master of Public Health (Africa University) (_)
Supervisor/s: Internal supervisor: Prof C. Tarirai (MPH, DTech Pharm Sci), TUT
External co-supervisor: Dr E. Mugomeri (DHSc), Africa University, TUT Postdoctorate

Thank you for submitting the **Master of Public Health (Africa University)** proposal for ethics clearance by the Faculty of Science Committee for Research Ethics (FCRE), Tshwane University of Technology (TUT). In reviewing the proposal, the comments and notes below are tabled for your consideration, attention and/or notification:

Comments

For noting by the FCRE. The study does not involve human participants.

.... Important ....

- In view of the Covid-19 pandemic, when applicable, please consider potential changes to:
  (a) The research design and/or research methods, and/or
  (a) The project timeline.
- Contemplate how the impact of the pandemic may be mitigated, particularly for the researcher(s).

*** Very Important: See notes (a) and (b) at the bottom of this letter.

Action[1]

None.

APPENDIX 7 (Continued)

Yours sincerely,

pp Prof Y Havenga
Chairperson: Faculty Committee for Research Ethics
Email: repsoldl@tut.ac.za Telephone: +27 12 382-6169

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[1] *The **reference number** [top right corner of this communiqué] should be clearly indicated on all forms of communication.*

A copy of this letter must be included in all correspondence.

PLEASE NOTE:

a) ***Time schedule**: Participant recruitment and/or data collection must not commence until ethics **Final Approval (FA)** has been obtained.*

b) ***Gatekeeper permission letter, where applicable**: When your research is conducted (i) within an institution/ organisation/workplace or public place, (ii) involves health and social care professionals working with participants such as patients, children or the elderly, and (iii) involves community leaders and/or family members of the participants, you need to obtain and submit a dated and signed permission letter from the relevant gatekeeper/s that allows you to undertake the planned research. This letter must be submitted to the FCRE prior to the commencement of the study.*