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ABSTRACT

With the recent launch of the Global Development Lab in the Hunter and Stephanie Hunt Institute for Engineering and Humanity, fellows, faculty, and industry professionals have been working to create meaningful solutions to promote a resilient humanity, addressing the UN's Sustainable Development Goals and challenges. The Institute has taken on Dr. Ali Beskok's project, the development of a low cost, portable, Point-of-Care-Device for humanitarian and health applications. This paper provides a systematic review of current Point-of-Care-Devices using antibody antigen reactions. Additionally, it provides aspects of a market analysis and a literature review. Its overarching goal is to make recommendations regarding a disease the developing device could test for through antibody antigen reactions that would most positively affect global health. Traditionally, biomedical engineers have developed technologies in response to the needs of the developed world's medical community. These approaches often do not address the needs of the majority of the world's peoples afflicted with both communicable and non-communicable diseases as the developments are far too costly and those with most need have, at best, limited access to supporting clinical laboratory infrastructure in developing countries. A gap in care has emerged as a result of these conditions. As a result, Drs. Beskok and Koklu have developed a Lab-on-a-Chip technology that can test for a chosen disease with a turnaround time of just a few seconds and a detection limit of 1 ng of antigen per 1 mL of sample fluid. In contrast to other commonly used PoCD's, this technology can be adapted for detection of various diseases in various settings. This is a great improvement to current devices on the market in specificity, sensitivity, and ease of use, therefore making it particularly useful in high-throughput, low-skill staffing environments.[18] In creating disease selection criteria for Dr. Beskok and his team's device, several factors were taken into consideration. Generally, the selection criteria consists of diseases that result in a high DALY value, are communicable, identifiable with antibody-antigen reactions, and can be tested for using urine. The diseases that were identified with this criterion were Tuberculosis and Malaria. Various antibody-antigen recommendations for diagnosis, advantages, and limitations of the proposed Point-of-Care-Device are discussed in the Proposed Disease section. In addition, current funding for each disease is overviewed. In order to make the greatest impact, deployment of the PoCD in the Sub-Saharan region, most specifically the Democratic Republic of Congo and Sierra Leone are recommended. Furthermore, children under the age of 5 who suffer from malnutrition should be given special attention. Focusing in these locations and populations will best aid in accomplishing the third Sustainable Development Goal set out by the UN: to ensure healthy lives and promote wellbeing for all at all ages.

1. INTRODUCTION

Inequalities in health between population groups exist in all countries and furthermore between countries. Conversations regarding global health equity have recently become more prominent as decentralized medical systems and universal health coverage have become more commonly practiced. Equity is defined by the World Health Organization as "the absence of avoidable, unfair, or remediable differences among groups of people, whether those groups are defined socially, economically, demographically or geographically or by other means of stratification."[1] When you apply this term to health care, it implies that all people should have the opportunity to attain their full health potential and that no one should be disadvantaged from achieving this potential. This being said, it is widely accepted that as various social determinants affect heath determinants, variance in wealth, power, and prestige create systematic differences. These differences can be seen along socioeconomic, political, ethnic, cultural and even gender lines. Often times, the causes of such inequalities vary between developing countries and developed countries. Additionally, access to medical care often varies with different infrastructure, education, and engineering practices.

Traditionally, biomedical engineers have worked to develop technologies in response to the needs of the world's developed community. However, these devices are often out of reach and inadequate to support the needs of the majority of the world's people who are afflicted with infectious diseases. These groups often only have access to poorly resourced health care facilities with little to no supporting clinical laboratory infrastructure. A gap in care has emerged as a result of these conditions. A major challenge for the biomedical engineering community is to develop diagnostic tests to meet the needs of these people, the majority of whom are in the developing world. In order

1 In cooperation with The Hunt Institute for Engineering and Humanity.

to address this challenge, our team of multidisciplinary students and subject matter experts have been working to improve the current immunoassay diagnostic standard through Lab-on-a-Chip technology by developing a portable Point-of-Care-Device.

The remainder of the paper will provide a systematic review on current and emerging Point-of-Care-Devices currently being used in countries with developing and developed economies. Next, it provides an overview of the diagnostic capabilities of the device being designed by Dr. Ali Beskok and his team on the Southern Methodist University campus. Selected diseases that have the biggest global impact on DALY measures and are appropriate for Point-of-Care diagnosis will be reviewed. Lastly, locations, populations, and people who will benefit most are identified.

1.1. Mission Statements

1.1.1 The Hunt Institute for Engineering and Humanity

Our mission is to serve as a national and international hub to partner with leaders in business, academia, NGOs, and government, in order to develop and scale sustainable and affordable technologies and solutions to the challenges facing people locally and globally. Our hub connects students, industry professionals, community members, faculty, and staff. Our innovation is evidenced through faculty-lead student research and demonstration projects that are focused on solving issues that impact humanity.

1.1.2 The Global Development Lab

The goal of the Hunt Institute Global Development Lab (GDL) is to foster innovative solutions for a resilient humanity addressing the UN's Sustainable Development Goals. The GDL selects innovative ideas with potential for transformational impact and provides programmatic support, in-kind and financial, to help develop them from the idea to proof of concept stage.

1.2 Sustainable Development Goals

In 1945, the United Nations (UN), an intergovernmental organization responsible for maintaining international peace and security, developing friendly relations among nations, achieving international cooperation, and harmonizing the actions of nations, was formed with the ratification of the United Nations Charter. In doing so, various nations signaled their commitment to battle issues confronting humanity in the 21st century, such as peace and security, climate change, sustainable development, human rights, disarmament, terrorism, humanitarian and health emergencies, gender equality, governance, food production, and more.[2]

In 2000, 191 UN member states met to develop 8 goals concerning direct improvements in well-being, all of which were set to be achieved by 2015. Known as the Millennium Development Goals (MDGs), these goals set to (1) eradicate extreme poverty and hunger; (2) achieve universal primary education; (3) promote gender equality and empower women; (4) reduce child mortality; (5) improve maternal health; (6) combat HIV/AIDS, malaria, and other diseases; (7) ensure environmental sustainability; and (8) develop a global partnership for development.[2]

By 2015, the end of the MDG period, the world had seen remarkable gains in the anti-poverty movement. With much more work to be done, a bold new agenda was developed to transform the world to better meet the human needs and the requirements of economic transformation, while protecting the environment, ensuring peace, and realizing human rights.[1] On January 1st, 2016, the 17 Sustainable Development Goals of the 2030 Agenda for Sustainable Development were created. With 169 targets, these 17 goals aim to address—in a manner that works for all people—the root causes of poverty and the universal need for development. Broadly, these goals are: (1) to end poverty in all its forms everywhere; (2) to end hunger, achieve food security and improved nutrition, and promote sustainable agriculture; (3) to ensure healthy lives and promote well-being for all at all ages; (4) to ensure inclusive and quality education for all and promote lifelong learning; (5) to achieve gender equality and empower women and girls; (6) to ensure access to water and sanitation for all; (7) to ensure access to affordable, reliable, sustainable, and modern energy for all; (8) to promote inclusive and sustainable economic growth, employment, and decent work for all; (9) to build resilient infrastructure, promote sustainable industrialization, and foster innovation; (10) to reduce inequality within and among countries; (11) to make cities inclusive, safe, resilient, and sustainable; (12) to ensure sustainable consumption and production patterns; (13) to take urgent action to combat climate change and its impacts; (14) to conserve and sustainably use the oceans, seas, and marine resources; (15) to sustainably manage forests, combat deforestation, halt and reverse land degradation, and halt biodiversity loss; (16) to promote just, peaceful, and inclusive societies; (17) and to revitalize the global partnership for sustainable development.

1.3 SDG 3: Ensure Healthy Lives and Promote Wellbeing for All at All Ages

The Third Sustainable Development Goal aims to ensure healthy lives by promoting the well-being of those at all ages.[3] Broken down into nine subsections, this ambitious goal looks to both address structural issues within the health care sector while also finding new cures to better address high mortality rates. Although significant strides have been made in this sector, there are still great distances to go in order in order to accomplish the newly identified targets by 2030. Primarily due to the interconnected relationship of health care and macroeconomic performance, progress towards meeting this goal has been uneven both between and within countries.[1] Although many important milestones have been reached, national averages hide the fact that many are still being left behind. Ensuring healthy lives and well-being for all at all ages is also contingent upon completing other goals set out by the United Nations such as water and sanitation, gender equality, climate change, and peace and stability. In order to accomplish these goals, the United Nations works in conglomeration with entities in the private and public sectors such as non-governmental agencies, non-profits, and government entities. Together, these bodies work to make positive impacts on the lives of humans around the world.

Countries are often put into the three categories of developed economies, economies in transition, and developing economies.[1] As technology and means of transportation have advanced, maintaining and supporting the articles laid out in the Universal Declaration of Human Rights become more complex and stratified than ever. These articles dictate human rights that are claimed to be unalienable for all in all places, but reality does not always align with these claims. Many of the Sustainable Development Goals directly relate to attaining these rights. For example, relating to Goal 3, Article 25 states: "(1) Everyone has the right to a standard of living adequate for the health and well-being of himself and of his family, including food, clothing, housing and medical care and necessary social services, and the right to security in the event of unemployment, sickness, disability, widowhood, old age or other lack of livelihood in circumstances beyond his control" and, "(2) motherhood and childhood are entitled to special care and assistance."[3] Although there have been many debates and conversations regarding the broad wording of these statements, the world still sees grave inequalities in access to and the distribution of health care. Many of these disparities exist between majority and minority, rich and poor, and urban and rural populations. In addition to this, across the world, children are at a higher risk of dying than the poor.[3] That being said, nearly all child deaths occur in countries with developing economies. The average under 5 mortality rate for those living in developed countries was 5 deaths per 1,000 live births.[1] For those living in low income countries, the average under 5 mortality rate was 68 deaths per 1,000 live births.1 In order to accomplish the SDG's set out by the UN, efforts must be focused on the locations and populations with the highest need. Common terminology in sustainable development refers to this concept as creating the "greatest impact." Regionally, of the countries with the 20 highest child mortality rates, the vast majority are located in Africa with the exceptions being Haiti ranked at number 7 and Afghanistan ranked at number 11.[1]

2. DIAGNOSTIC PRACTICES

2.1 Technical Aspects

2.1.1 Antibody-Antigen Reactions

When pathogens enter the body, they can trigger humoral immunity. This occurs when an antigen, a foreign substance that elicits an immune response, binds to its specific immunoglobulin (Ig) receptor on B cells. These B cells are then stimulated to proliferate and produce antibodies. Antibodies are Ig proteins that either directly kill their specific antigen or flag it for destruction. Antibodies can be monoclonal, meaning one antibody binds to only one antigen, or polyclonal, meaning one antibody can recognize and bind to multiple antigens. In either instance, the binding between antibodies and antigens is extremely specific.[4]

2.1.2 Immunoassays

Immunoassays utilize the reactions between antibodies and antigens to detect and quantify the presence of a pathogen. Immunoassays are highly beneficial because of their inherent extreme specificity and sensitivity. The most common immunoassays include radioimmunoassay, chemiluminescenceimmunoassay, counting immunoassay, enzyme linked immunosorbent assay, and fluoroimmunoassay.[4]

ELISA, or enzyme linked immunosorbent assay, is the most common immunoassay that utilizes competitive, direct, indirect, or sandwich assay methods, as well as enzyme conjugated antibodies to produce a quantified result.[5] ELISA tests have a high throughput and are highly specific and sensitive.[4] But ELISA, and many other conventional immunoassays, are limited by their high cost, lack of portability, slow analysis time, utilization of additional sophisticated instruments, and requirement of a skilled user.[6] Therefore, currently used immunoassay techniques are not necessarily suitable for diagnosing diseases in developing communities.

2.1.3 Point of Care Technologies

Lab-on-a-Chip, or LoC, devices aim to be "chip sized" recreations of an entire biological laboratory. Because of their small size, LoC's are portable, simple to use, low cost, low in energy consumption, require only small biofluid sample sizes, and rapidly produce results.[7] LoC technologies align with the ASSURED guidelines of Point-of-Care-Testing and are therefore better diagnostic options for less developed areas.

Point-of-Care-Testing (PoCT) or Point-of-Care-Devices (PoCD) are those that can provide diagnostic results while the user is nearby or with the patient. This diagnostic method should be rapid, low-cost, and simple. Point-of-Care-Testing can be separated into two categories: small, handheld devices, such as Lab-on-a-Chip technologies, or desktop instruments that are simplified versions of pre-existing technologies.[8] Many PoCDs use immunoassays to detect the presence of pathogenic substances and are recommended for use in low-resource areas as long as they follow the ASSURED guidelines. These ASSURED guidelines are set by the World Health Organization and are discussed in Section 2.3.2.

The Point-of-Care-Device developed by Drs. Beskok and Koklu utilizes Lab-on-a-Chip technology and contains nanorods coated with an antibody which has a specific and strong affinity for a particular antigen. When a biofluid sample is run through the device, only the antigen of interest, or target molecule, will bind to the antibodies covering the nanorods. The binding will then generate an impedance value that can be read as a quantified value of antigen present in the sample. Information on the PoCD is continued in Section 3.0.

2.2 Developed Countries Current and Developing Diagnostic Practices

Diagnostic technologies and trends tend to emerge first in developed countries. Though some technologies primarily in use in developed countries are reviewed in this report, there are many diagnostic technologies that continue to change the medical field but are not currently feasible to implement outside of stable, resource-rich environments. To better combat trends, such as overgeneralized treatment with antibiotics – leading to increased rates of antibiotic-resistant strains, the medical community is shifting the way they provide treatment. Specifically, the medical community is

putting an emphasis on developing faster, high-specificity, high-sensitivity diagnostics to pin-point exact strains before pharmaceutical treatment.

A majority of research and development (R&D) takes place in developed countries because such locations are both hubs for R&D funding and allow for sooner availability to patients. However, developed countries often have different diagnostic goals that are not always directly applicable to the developing world. While a shift has begun to occur in developing countries, the highest burdens of diseases remain from communicable diseases.[9] In contrast, noncommunicable and chronic diseases elicit higher burdens of disease in developed countries.

2.2.1 Current Diagnostic Practices in Developed Countries

Centralized laboratories are facilities dedicated to lab testing and assessment, localizing diagnostic services to medical institutions as a whole. Laboratory tests which were once outsourced to facilities miles away for processing now have the ability to be done on site. The in-house capability greatly reduces time from initial sample to diagnostic result and therefore time to treatment. In addition, faster result turnaround reduces the potential for a patient with a communicable disease to expose non-infected persons. The centralized laboratory has also allowed off-site labs to become more specialized in their testing and reduce cost to hospital per test. Typically, laboratory-based-testing has few limitations, which are often outweighed by the benefits of specificity, timeliness, and information provided. Because of large size, high cost, and specific skillset required, immunoassay systems such as ELISA and other diagnostic instruments are commonly a part of these centralized laboratory facilities.

In state-sponsored healthcare systems, such as that of the U.K., there is a growing trend towards the laboratory network. A laboratory network allows for smaller laboratories to specialize in a specific set of medical tests, distributing fixed-initial cost over the system for testing that requires specialized equipment. This process allows for reduced per-test-cost and reduces redundancies, but is only viable where transportation challenges can be overcome and transmission of results is efficient and reliable.

Digital Diagnostic medicine, as defined by the Digital Therapeutics Alliance, seeks to "deliver evidence-based therapeutic interventions to patients that are driven by high quality software programs to prevent, manage, or treat a broad spectrum of physical, mental, and behavioral conditions."[10] PricewaterhouseCoopers (PwC) outlines a trend in developed countries towards Digital Diagnostic medicine as well as digital vital sign tracking, which allow for the implementation of personalized medicine. In developed countries, greater levels of resources allow for more expensive digital Point-of-Care-Devices that can interface with smart watches, smart phones, and more. There has been an observable shift in developed countries towards decentralized medical care that focuses on keeping patients out of hospital and treated by satellite health partners when needs arise.[11]

Although there is still a need for highly skilled workers to produce diagnostic data for patients, some of this need is being lessened in developed countries. Patient and market comfortability with telemedicine—allowing for physicians to view data remotely, conference with patients via mobile apps, and opt for attachments to smart phones that enable Point-of-Care applications—is on the rise.[10] In developed countries, issues with internet connection, power usage, and access to highly specialized labs are significantly reduced, but some of the factors that allow for the rise of highly specialized central care, decentralized general care, and "smart" medicine are not as readily or consistently available in the developing countries.[10]

Within current diagnostic markets, PoCT and immunoassays hold strong market-share, at near 35% and 25% respectively, and continue to trend with higher than average market growth (see Figure 1)[2]. Although globally beneficial, developed countries possess strong enough infrastructure for significant R&D and deployment. POC devices serve as a way to incorporate personalized medicine and increase safety in infectious disease diagnostics for patients in the U.S., U.K., and Europe.[9]

2.2.2 Emerging Diagnostic Practices of Developed Countries

Within developed countries, there are several immunoassay or near-patient devices of interest on the rise. In particular, Curetis has launched a class of devices that can act as a diagnostic near-patient tool for any number of chronic and infectious diseases that commonly affect developed countries. This class of devices all use a prepared cartridge-style table-top system to diagnose strains of Pneumonia; detect implant, tissue, intraabdominal, and Urinary Tract infections; and perform blood culture. The company has plans to continue increasing the range of cartridges available.[12]

For example, Unyvero is a Curetis device that can perform the processes described above. One study compared the abilities of Unyvero and the PLY-ELISA immunoassay to diagnose Pneumonia.[13] The PLY-ELISA immunoassay is a well-accepted method for pneumonia strain-specific detection, but it takes substantially longer to process than Unyvero. With an increasing global need for better diagnostics in order to combat antibiotic-resistance, technology like the Unyvero device not only fills this gap, but does so in a near-to-patient, quick, simple way. In independent studies testing the efficacy of Unyvero's pneumonia specific cartridge, it performed as well as traditional PLY-ELISA in sensitivity, although not specificity (see Figure 2). However, it is important to note the particular strain tested is not common in developed countries, and therefore Unyvero is still considered a very viable option. In more common strains, Unyvero performed consistently near or above 90% in specificity.[12, 13]

2 All figures appear in Appendix 3, starting on page 15.

2.3 Developing Countries: Current and Developing Diagnostic Practices

Diagnostic technologies are equally as important for health care within the developing world. Proper diagnostic methods are critical to accurately and effectively treat patients and ultimately save lives. Current issues such as antibiotic resistance, cost of care, and communicable disease pandemics have all ignited a growing interest in investing in diagnostic technology across the globe.[11] With that being said, many of the tools currently used to diagnose disease in low-resource settings are not suitable for the communities they are meant to serve. Many of the diagnostic practices used in developing countries are modeled off practices used in developed countries. Developed countries are focused on tackling noncommunicable health problems, and despite a rise in these ailments within the developing world, infectious disease remains most prevalent within many developing countries.[14] As a result, the mass-processing automated machines and lab tests that are common within the developed world are often too expensive, resource-dependent, and time consuming for developing areas.

2.3.1 Current Diagnostic Practices in Developing Countries

While a centralized laboratory is often considered the gold standard within developed countries, the standard and quantity of these labs are often lower in developing countries. The centralized lab allows for complex tests, incredibly specific and sensitive diagnoses, and advanced technologies. Often, lack of support for qualified personnel, limited funding, geographic barriers, and quantity of centralized labs and instruments result in various limitations. These include but are not limited to: a high ratio of samples to laboratories and qualified personnel, lack of proper resources and supplies, sample transportation time, and lengthy result turnaround.[14] Inconsistent and sparse access to electricity and water are another reason that hospital laboratories are insufficient within developing countries. In conclusion, "gold standard" laboratories are rare in the developing world and current labs do not produce similar results.

In an effort to mitigate the problems presented by centralized labs, the number of rural clinics has increased. With hopes of being lower-cost and more geographically accessible, local clinics are similar to the developed world's physician's office.[14, 15] These clinics run quick diagnostic tests, often utilizing the less expensive and simpler lateral flow methods of diagnosis.[16] But, these tests have poor sensitivity, around 31.3%, and rural clinics still do not possess the staff and resources necessary to be a complete solution.[16]

The most common way that a diagnosis is reached in developing countries is through symptomatic review.[14] In this case, treatment is given to patients dependent on the prevalence of a certain disease within a community and how well patient symptoms align with that disease. While diagnosing diseases in this way can be cost- and resource-effective, there are also downfalls. For example, deciding to treat for Malaria in Sub-Saharan Africa by review of symptoms is up to 88% specific but only up to 66% sensitive.[17] This means that only up to 66% of reviewed patients are correctly diagnosed as not having Malaria. Patients can be incorrectly treated for diseases that they do not have, while their true ailments go untreated.

In summary, the most common diagnostic issues within developing areas include staffing constraints, lack of quality lab and medical resources, lack of reliable water and electricity, geographic access to health care, and cost. Diagnostic practices currently being used within less developed countries are often based off of the methods of the developed world. This misalignment of diagnostic practices creates a need for the development of new diagnostic technologies.

2.3.2 Emerging Diagnostic Practices in Developing Countries

Point-of-Care diagnostic technologies seek to fill the gap created by current diagnostic practices, especially within developing countries. The WHO has outlined the ideal diagnostic technology for less developed communities. This technology should be ASSURED—affordable, sensitive, specific, user-friendly, rapid, equipment-free, and deliverable to patients.[18]

A POC test should be cost-effective for both providers and patients. Determining cost-effectiveness should include measurements of the price of the test, disease prevalence, cost of treatment, cost of mistreatment, and throughput. In areas where POC technology is most needed, health care providers and their patients often lack abundant budgets, funding, and resources. Therefore, new technologies must be cost-effective.

The device should also be sensitive and specific. The test should produce few false positives and few false negatives if it is going to achieve what other diagnostic methods do not. Ideally, the technology should also be multiplex, or capable of detecting multiple different antigens, in order to best come to a conclusive diagnosis. To mediate the misdiagnosis, and therefore mistreatment, of disease in underdeveloped countries, a PoCD must be both sensitive and specific.

Due to the limitations posed by the small number of skilled, qualified health care workers, a PoCD needs to be simple. The device should be simple enough that those without medical training should be able to perform the test. In addition, the biofluidic sample used should be easy to obtain and handle. Diagnostic results should be presented in a way that is easy to understand and record.

In addition, the developed diagnostic technology should be rapid in order to best prevent mistreatment, correctly treat patients, and also optimize efficacy of that treatment. The PoCD should output within a small enough time period that treatment can begin during that same visit—hence, "point of care."

A PoCD needs to be portable to truly live up to its name. This means that the device should be small, robust, and easy to move around. The device should not require large equipment or machinery to operate. These qualities ensure the portability of the device as well as successful operation without the need for extraneous supplies or resources.

Finally, POC technology needs to be deliverable to those who need it most. Whether in rural clinics or

emergency response settings, patients in need must be physically able to access diagnosis.

3. PROPOSED DEVICE

3.1 Current Device

The technology developed by Dr. Ali Beskok and Dr. Anil Koklu in Southern Methodist University's (SMU) Biomicrofluidics Lab utilizes Lab-on-a-Chip technology that aims to be a possible solution for diagnosis of disease, especially for implementation in developing communities. The appropriate development of this technology has been informed by the ASSURED criteria suggested by the WHO.[14] An overview is provided in the paragraphs below but for further technical explanations of the methodology and capabilities of the device, please refer to the article published in Analytical Chemistry titled: Rapid and Sensitive Detection of Nanomolecules by an AC Electrothermal Flow Facilitated Impedance Immunosensor.[18] To see a configuration of the device refer to Figure 3.

This technology fundamentally functions similarly to other LoC devices, it utilizes antibody-antigen reactions to detect the presence of pathogens. Conventional immunosensors are often limited by low sensitivity and long detection times as they typically rely on passive diffusion. Uniquely, this LoC utilizes a pumping loop and the system can analyze both high and low conductivity physiological fluids such as urine, saliva, and blood. As the sample fluid is pumped through channels, it interacts with nanorod electrodes coated with a certain antibody. The pumping loop utilizes the micro stirring effect of AC electrothermal flow and structural advantage of nanorod-covered interdigitated electrodes with impedance spectroscopy to analyze a physiological fluid.[18] The nanostructured electrodes of the LoC increase the surface area for binding interactions to occur, which widens the frequency range for impedance measurements. In addition, the hydrophilic surface of the LoC reduces nonspecific binding and lowers friction to improve pumping. These techniques allow for analytes to bind onto nanorod surfaces within a few seconds and enriches the number of binding molecules. The nanorods can be also be programmed, or coated in new antibodies, in order to attract different antigens, depending on which disease the user would like to test for. This allows the LoC to be flexible for target molecules detected, while the combination of the chip's surface and nanostructuring dramatically increase the sensitivity.[18] If the biofluidic sample contains the specific antigen that binds the selected antibody configured to the device, the antibody-antigen reaction will increase impedance within the circuit. The increased impedance value is then measured and used to quantify the amount of antigen within the sample.

In summary, Drs. Beskok and Koklu have developed a novel Lab-on-a-Chip technology that can test for a chosen disease with a turnaround time of just a few seconds and a detection limit of 1 ng of antigen per 1 mL of sample fluid. In contrast to other commonly used PoCD's, this technology can be adapted for detection of various diseases in various settings. This is a great improvement to current devices on the market in specificity, sensitivity, and ease of use, making it particularly useful in high-throughput, low-skill staffing environments.[18]

3.1.1 Limitations of Current Device

The current technology still faces a few limiting factors. As of now, the Lab-on-a-Chip requires two additional pieces of equipment: a function generator and an impedance analyzer (see Figure 3). The function generator is necessary for signal production, which is needed for the chip's functionality, and the impedance analyzer is needed to produce impedance values. These pieces of equipment are large and high-cost. Therefore, the current technology fails to be small, portable, and inexpensive. The current LoC technology is also only capable of testing for one antigen using one antibody, which means that diagnosis is not multiplex. Biosafety level restrictions and limited funds for further lab instruments have hindered further development of the device. Without an increase in the Lab's biosafety level, usage of pathogens and higher-level biofluids needed for device testing and development are unavailable. With further partnerships and grant-approval, further developments will be made. These goals are elaborated on in Section 3.2.

3.2 Goal Device

Although the current device is still in development, further benefits are expected with continued development. Although the current device already has high detection specificity and flexibility in target diseases, increased funding will lead to development of elliptical pumps that will allow for an increase in multiplex detection to 10 species.

Due to the antibody fixing method and single-use design of the chip, there is flexibility to fit various desired applications. Such increased flexibility allows for location specific and population specific diagnosis criteria. Further development also aims to allow for nanorods to be coated in multiple antigens, allowing for multiplex assays. Because of the impedance based detection design of multiple species as well as the single use structure of the LoC, it is expected that there will be little to no cross-contamination .

In addition, the goal LoC utilizes a cyclical pumping loop. By using a cyclically pumped loop system, with no external pump components, the device is able to reduce the amount of patient specimen needed for detection, past the 1ng/mL mentioned earlier, increasing sensitivity.

The nanostructured surfaces within the detection zone increase the binding surface area and allow wider frequency range for impedance measurements.

The hybrid surfaces further reduce nonspecific binding on the walls of the device and enable low friction to pump the fluid.

In terms of buffers and fluids used for the device, the microfluid transport is suitable for both low and high conductivity. For global usage, the breadth allowed in buffers increases the possible supplier market and allows for greater in-field flexibility while prepping samples.

The device can be developed as a handheld device or the Lab-on-a-Chip (LoC) can be incorporated with an external app through an impedance measurement interface. The final device will include components that allow impedance detection and function generation without traditional lab devices, such as the AFG1062 Tektronix

Arbitrary Waveform Generator – retail for $1,240 – or the Agilent / HP 4294A Precision Impedance Analyzer, 40 Hz to 110 MHz – retailing for $37,495.

3.2.1 Limitations of Goal Device

Though there are numerous discussed benefits, the device also presents limitations—even in an ideal condition.

Before the chip is used, the glass plates must be cautiously transported to avoid breaking the glass. These glass plates provide both the top and bottom portion of the current chip, with nanorods and more being sandwiched in-between. These nanostructures, situated between the glass plates, are vital that for greater surface area in detection. The technology to safely transport glass plates is widely on the market; however, transportation limitations must be noted as a possible concern.

The device also has limitations because of the need for a secondary display. The complete device needs to interface with a data output display for timely diagnostics, and currently, this is a fixed cost that may or may not incur variable cost of maintenance.

In addition, the secondary display which reads impedance and generates the signal required for the operation need to be portable. Furthermore, the device still needs power infrastructure to be usable. Once again, these extra parts are included in the fixed set-up cost incurred by organizations to operate the portable device, and will present a variable cost to the operation due to the need for future replacement or maintenance.

4. PROPOSED DISEASES

The World Health Organization defines health as a state of complete physical, mental and social well-being and not merely the absence of disease or infirmity. Human rights doctrines that call for adequate health are also often phrased broadly, making "good" hard to quantify. In attempts to best describe and measure health phenomena on a global scale, various health indicators have been developed and standardized. In general, they are categorized into proximal and distal indicators. Proximal indicators aim to provide direct measurements by looking at diseases, deaths, and use of services. Distal indicators aim to measure social development, education, and poverty indicators.[3]

Medical practices and forms of treatment often vary depending on access to resources. Quality of care is generally comprised by the three sections of structure, process, and outcomes. Structure refers to the material characteristics such as infrastructure, tools, and technology. Process refers to the interaction between caregivers and patients during which structural inputs from the health care system are transformed into health outcomes. Lastly, outcomes can be measured in terms of health status, deaths, or disability-adjusted life years—a measure that encompasses the morbidity and mortality of patients or groups of patients.[19]

Primarily through the practice of Epidemiology, which looks at the incidence, distribution, and possible control of diseases across the world, medical professionals developed mathematical techniques to measure the rates of death and disease. Certain measures quantify the risk of occurrence of contraction and mortality where other rates more broadly measure years of life that are spent in good or poor health. These measures help direct needs for policy reforms, action plans and the provision of services. The most comprehensive methods that do so are known as Disability Adjusted Life Years (DALY).[19] One DALY equates to one lost year of healthy life. When DALY's are added together across a population, we can see a measurement of the gap between the current health status and an ideal health situation where the entire population lives to an advanced age free of disease and disability. Quantifying the burden of disease assists doctors, nurses, public health workers, and other medical professionals as they work to measure the performance of health on a global, regional, national, and subnational scale. Because the highest DALY numbers result from diseases that result in the disability of children, these diseases are often prioritized on the global health scale.[19]

In creating disease selection criteria for Dr. Beskok and his team's device, several factors were taken into consideration. Generally, the selection criteria consists of diseases that result in a high DALY value, are communicable, identifiable with antibody-antigen reactions, and can be tested for using urine. Because the LoC microfluidic platforms are employed for POC diagnosis purposes, they have low cost, low power consumption, short analysis time, and use minute amounts of sample volumes. Therefore, they are useful in higher stress or emergency situations. Diseases with high rates of communicability are those which spread and replicate rapidly. Early detection in such cases allows for earlier treatment and decreased infection rates. Accurate detection also decreases the rate of misdiagnosis, maltreatment, and the possibility for the development of antibiotic resistant strains. The diseases that were identified with this criterion were Tuberculosis and Malaria. Together, these diseases accounted for 89,011,364 years of healthy life lost in 2016.[19]

4.1 Tuberculosis

A WHO re port on global and regional Tuberculosis (TB) incidents in Africa and South-East Asia indicates that such incidents are nearly 70% of all incidents world-wide. Annually, more than 95% of cases and deaths from TB occur within developing countries. Those undernourished are 3 times as likely to contract TB. In addition, 1.1 million children (0–14 years of age) fell ill with TB, and 230,000 children (including children with HIV associated TB) died from the disease in 2018.[20] These numbers are striking, and the DALY values for TB rank in the top 10 globally (see Figure 4).[19]

The treatment options for groups affected by TB make it a great option for initial deployment of a rapid-diagnosis PoCD. TB continues to have a significant effect on countries with developing economies and countries in transition, and based on 2018 published numbers, is off track for the UN Sustainability Goal 3, under which a target is to end the epidemics of AIDS, TB, malaria and neglected tropical diseases, and to combat hepatitis, water-borne diseases and other communicable diseases.[3]

4.1.1 Global Funding for Tuberculosis

Based on a report by the Treatment Action Group (TAG) published in December 2018, global funding for TB has remained steady since 2009, when adjusted for inflation.[21] These findings prove optimistic for the continued interest in TB research and the Global End TB campaign.

When funding for TB is broken down into 5 major categories, over the 2016 and 2017 funding periods, research in vaccines and diagnostics still has a wide gap left for the 5-year period. As funding organizations choose sector-specific funding targets in the coming years, diagnostics are high on the list with nearly $3.3 billion USD left to fund to meet the 5-year global funding target (see Figure 5). This monetarily signifies a need for continued development and deployment in the three areas of diagnostics, drugs, and vaccines.

The largest funder of TB world-wide has remained the National Institute of Health (NIH) and the National Institute of Allergy and Infectious Diseases (NIAID) under the NIH. With continued bipartisan support for the NIH and increased allocation over the years, support by the NIH to TB funding is not expected to drop.[22] In total, the United States remains the top funder of TB research globally at $312 million, followed by the Bill and Melinda Gates Foundation—the top philanthropic giver—at $126 million, and in third place the European Union, at $37 million. It is important to note, as a U.S. research initiative, the two top funding resources globally reside in the U.S. In 2017, 66% of funding has come from the public sector, with the private sector coming in at 11%.[22]

The TAG report also makes mention of the investment group, Unitaid.[21] Unitaid is focused on investing in innovations to prevent, diagnose and treat HIV/AIDS, tuberculosis and malaria more quickly, affordably and effectively."[23]

Within funding for diagnostics, the best areas to seek grant opportunities are from either public/government funding, the Bill and Melinda Gates foundation, or Unitaid. The U.S. remains a global leader in TB funding support and research.

4.1.2 Pathology of Tuberculosis

Tuberculosis is an infectious disease caused mainly by the aerobic, rod-shaped bacteria *M. tuberculosis*. Additional microbes, *M. bovis* and *M. africanum*, can also induce Tuberculosis, but often only in underdeveloped areas. *M. tuberculosis* is especially virulent because it is carried in airborne droplet nuclei and can remain in the environment for hours after a host has infected the space. The disease is transmitted to an uninfected host by inhalation of a droplet nuclei. Successful transmission of tuberculosis is impacted by the susceptibility of the exposed person, the level of exposure that an uninfected person experiences, the infectiousness of the pathogen, and any environmental factors that may increase or decrease virulence.[24]

If inhaled, *M. tuberculosis* can reach the alveoli of the lungs. There the bacteria are recognized by macrophage cells and targeted for destruction by lysosomes. *M. tuberculosis* can then release proteins that inhibit the hydrolytic functions of the lysosomes. While most bacterium may be expelled or destroyed before being able to cause an infection, often some successfully remain in the lungs. The body's immune response creates a covering, or granuloma, containing the bacteria. While the granuloma prevents the infection from spreading, the tissue within the containment becomes necrotic and develops into a harmful Gohn Complex.[25] If the infection is contained by the granuloma, it is latent and can later become activated. If Tuberculosis becomes active soon after infection, it is classified as Primary Tuberculosis.[26]

If the bacteria within the granulated tissue survives, it can successfully spread throughout the lungs and rest of the body, most commonly if the individual is immunocompromised or the granuloma fails to contain the bacterium.[24, 25] Those at highest risk of developing Active Tuberculosis from the latent form are children under five, those infected with HIV, and those in communities with increased incidence, especially medically underserved, low income areas.[26] Tuberculosis can cause further necrosis and even cavity formation within the lungs and can lead to complications in the pleura, larynx, bones, kidneys, lymph nodes, and the brain.[24] Active cases of Tuberculosis and its further systemic complications are fatal if not detected and treated properly.[25]

It is important to test for Tuberculosis as early as possible to prevent progression from latent to active versions and to provide accurate and efficient treatment in already active cases. Rapid and early detection of the disease is necessary to prevent these adverse outcomes. Individuals with latent Tuberculosis are not contagious and cannot spread the infection to others. But, because *M. tuberculosis* is slow-growing and persistent, it can easily develop drug resistance or later, further progress throughout the body. Therefore, it is still necessary to quickly detect even latent TB infections. Those with active TB are highly contagious and can be for up to weeks after starting treatment.[25] Due to the differences in latent, primary, and active Tuberculosis, there is no set critical period for the disease. Despite the complexities posed by various stages of TB, early diagnosis of the disease is one of the most effective ways to decrease its negative impact on those diagnosed and their surround communities.

4.1.3 Diagnostic Methods for Tuberculosis

The gold standard for diagnosing Tuberculosis is mycobacterial culturing using liquid media as this is the only test that is considered to actually confirm the diagnosis of Tuberculosis.[27] TB cultures most commonly utilize sputum as a biofluid sample, but other biofluids can also be used. Cultures can differentiate between active and latent forms of TB and can also determine if the sampled bacteria are drug resistant. Mycobacterial cultures are highly specific and sensitive and can diagnose TB with as little as 10-100 colony forming units per mL of sample.[28] While TB cultures are considered to be the "gold standard" of TB testing, they are not actually the most commonly used test. This is because culturing a sample of bacteria requires extensive lab equipment and biosafety clearance, has a turnaround time of 10 to 14 days, and is costly.[27] Therefore, other methods are often relied on to detect an individual's exposure to TB.

TB is typically tested for using a purified protein derivative (PPD) skin test. In this test, a purified protein of the Tuberculosis bacteria, tuberculin, is injected intradermally on the forearm.[28] If the individual has been previously exposed to *M. tuberculosis*, they will produce a

localized reaction at the site of injection. This reaction is then read by qualified personnel between 48 to 72 hours post-injection and is then measured in millimeters of the induration.[28] The Tuberculosis skin test is inexpensive and requires little to no sophisticated medical instruments or laboratory equipment, making it one of the most common forms of diagnosis. Despite this, there are limitations. The test requires skilled personnel to read the results and patients must be able and willing to return two to three days after initial injection of tuberculin. In addition, a 2011 WHO article stated that Tuberculosis skin tests have a pooled sensitivity of 83% and specificity of 58%.[29] Skin tests conducted on those who are vaccinated for TB often result in a false-positive.[29] Lastly, this diagnostic method also only informs if a patient has been exposed to TB and additional testing is required to determine if the TB infection is actually active.[30]

4.1.4 Recommendations for Dr. Beskok's Device

While cell-mediated immunity is much better understood in the body's response to TB, there is evidence that humoral immunity plays a role in protection from the disease.[30] Lipoarabinomannan, or LAM, is a lipopolysaccharide associated with the cell wall of *M. tuberculosis* and is specifically bound by certain antibodies. LAM can be detected in urine and is better for detecting TB in those co-infected with HIV. For those with HIV infections, it can be difficult to produce sputum, the biofluid used in gold standard TB testing, and difficult to detect the presence of an antigen due to low CD4+ cell counts.[31] Because 11% of HIV patients also have TB and those with HIV are at a 16-27 times greater risk to develop TB, it is important to prioritize diagnostic methods that can detect for tuberculosis in individuals that also have HIV.[31] This is why the LAM antigen is most highly recommended.

The only current commercialized rapid test for detecting LAM is the Alere LF-LAM test. This is a Point-of-Care lateral flow test that detects the presence of LAM in urine with 42% sensitivity and 91% specificity in symptomatic patients.[32] The Alere LF-LAM test has a limit of detection (LOD) of 100 cells/µl, which is the recommendation for PoCT made by the World Health Organization.[33] Because of the current test's low sensitivity, specific reagents with specific binding sites on LAM have been studied to improve assay results. One study showed that pairing capture antibody S4-20 with the A194-01 detection antibody increased sensitivity to 93%, specificity to 97%, and showed no cross-reactivity with other bacteria.[34] This pair of antibodies also detects Tuberculosis in those that are HIV-negative with a high sensitivity of 80%. Therefore, it is highly recommended that LAM antigen is detected using S4-20 and A194-01 antibodies.[34]

Not only can LAM be detected in urine, but utilizing this biofluid as a sample for PoCD testing presents other benefits. Urine based testing has advantages over sputum, blood, and other biofluids as it is easy to collect, handle, and store, and it lacks infection control risks that are often associated with the collection of other biofluids.[35] Urine enables a PoCD to be used by people with little to no medical training as blood will not have to be drawn, nor sputum collected. Furthermore, utilizing urine allows for a decrease in various anthropological cultural sensitivities.

4.1.4.1 Advantages and Limitations of Dr. Beskok's Device

In order to reach the Third SDG and End TB Strategy set for 2030, further research and the development of vaccines and/or biomedical devices is needed. The advantages of a Point-of-Care-Device that can test for TB would allow for a decreased time to diagnosis and therefore faster access to treatments. This would not only decrease the mortality rate but also decrease the rate of transmission from person to person. Currently, there are no single rapid, accurate and robust TB diagnostic test that recommended by the WHO for use at the Point-of-Care.[35]

The most commonly used test, the Tuberculosis skin test, requires the patient to return between 48 and 72 hours after the test is initiated. Dr. Beskok's proposed device would ideally allow for the patient to receive their results within minutes.

Recent studies have also shown that differences in antibody glycosylation can distinguish between active and latent forms of TB. Specifically, the digalactosylated (G2) Ab glycoform was found to be the best biomarker between Active TB and Latent TB.[36] With further research and implementation of this glycoform, in addition to LAM capturing, a PoCD could potentially detect TB in HIV positive or negative patients, vaccinated patients, and also differentiate between active and latent forms.

Alternatively, there are various limitations to take into consideration. The development of a new PoCD is likely to initially yield results that are less specific, less sensitive, and have a higher LOD than current diagnostic methods such as culturing and immunoassays. Currently, the device closest to producing the desired results is the Alere-LAM diagnostic test, costing only $3.50 per test.[33] In order to surpass current diagnostic methods, the proposed device must be more sensitive and specific, cheaper, or faster than those already on the market. In addition, further research needs to be conducted on the specific antibody antigen reactions recommended above for a TB PoCD.

4.2 Malaria

Malaria is currently a global epidemic. Within the Africa region, the WHO reported Malaria as the 4th highest DALYs percentage in 2016. At the start of the 20th century Malaria was also ranked 4th on the DALY measure in the African region. While the DALYs percentage has fallen from 9.0 in 2000 to 5.8 in 2016, significant progress in diagnostics and treatment still must be made in order to reach the subset of the third SDG: to end Malaria by 2030. Although Malaria persists an issue world-wide, the greatest WHO region affected continues to be Africa with a concentration in the Sub-Saharan region (see Figure 6).[19]

Other regions in need include Oceania and the subcontinent of India (see Figure 7). Malaria is primarily regionally transmittable and is a disease which will not affect most developed countries due to geography. The affected region conversation is important when talking about current research, current funding, and incentives to the End Malaria 2030 goal.[37]

The WHO notes "in sub-Saharan Africa, rapid-diagnostic tests (RDTs) are increasingly becoming the most used method to test for malaria diagnosis among suspected malaria patients in public health facilities. In 2017, an estimated 75% of malaria tests were conducted using rapid-diagnostic tests, up from 40% in 2010."[38] These findings help us to understand the readiness by the market for a POC diagnostic solution for Malaria. Currently however, most RDTs are only testing for one strain of Malaria—*P. falciparum*— leaving a big need gap in the diagnostic market for other strains tested.[38]

4.2.1 Global Funding for Malaria

Current funding efforts are concentrated within Sub-Saharan Africa, with consistent funding from 2010 to 2017. However, global funding still has yet to meet funding markers outlined by the SDG3. Three current areas of greatest need for Malaria funding support fall to Africa, a majority of South-East Asia, and areas of the Eastern Mediterranean. Four big players in the Malaria Funding sphere are Global Fund, PMI/USAID, World Bank and the UK (see Figure 8).[3]

As noted in the Tuberculosis funding section, Unitaid, an investment group focused on investing in innovations to prevent, diagnose and treat HIV/AIDS, tuberculosis, and malaria more quickly, affordably, and effectively is also a great source of private funding for Malaria diagnostic development and deployment.[23]

It is important to also realize that the Eastern Mediterranean, though not prominently seen on the CDC map of transmission, continues to be a high incidence area for Malaria (see Figure 8). The countries included in Figure 8's summary are: Afghanistan, Djibouti, Iran, Pakistan, Saudi Arabia, Somalia, Sudan, and Yemen.

Within funding for Malaria diagnostics, the best areas to seek grant opportunities are from public/government funding or Global Fund, PMI/USAID, World Bank, the UK, or Unitaid. The U.S. also remains a global leader in Malaria funding support and research.[23]

4.2.2 Pathology of Malaria

Malaria is caused by four different protozoa that are introduced by *Anopheles* mosquitoes. These four protozoans are of the genus *Plasmodium* and include *P. falciparum*, *P. malariae*, *P. ovale*, and *P. vivax*. It is important to note that only *Plasmodium falciparum* causes fatal disease. When feeding, female *Anopheles* inject sporozoites into a human host. The immature sporozoites then travel through the bloodstream and infect the liver, where they mature and reproduce. After maturation in the liver, cells rupture and release merozoites that invade red blood cells. During the erythrocytic phase, the merozoites reproduce by digesting hemoglobin and producing hemozoin. This is called a schizont, which can then rupture and release merozoites that will again undergo the erythrocytic phase. Some merozoites then enter the gametogony phase where the produced gametocytes can be ingested by an *Anopheles* mosquito and new, infectious sporozoites are formed that are passed along to a different human host.[39]

Most forms of malaria present with cyclic and unspecific symptoms such as fever, headaches, and nausea. Specifically, *P. vivax* and *P. ovale* can remain dormant in the liver and cause later relapses of Malaria symptoms.[40] This is termed uncomplicated malaria and is considered a more mild presentation of the disease, as it very rarely is fatal. Severe malaria is caused only by *P. falciparum* and can lead to organ failure and various complications including comas and even death.[38] Patients should be tested within seven days of their first possible exposure to *P. falciparum* to three months after their last possible exposure.[40] Although *P. falciparum* is inherently the most fatal form of Plasmodium species, lack of access to proper medical resources and treatment make the rapid diagnosis of any form of malaria imperative.[39] With simple and fast diagnosis through PoCD's, patients in underdeveloped communities are provided the opportunity to early and proper treatment of disease.

4.2.3 Diagnostic Methods for Malaria

The gold standard for diagnosing malaria is microscopic laboratory examination.[41] Microscopy is performed using both thick and thin smears of blood samples under light microscopes to determine the presence of Plasmodium parasites. Thick smears are used to confirm the presence of parasites and thin smears are used to determine parasite species.[42] Microscopy has a limit of detection of 50-500 parasites/μl of blood. Microscopy can be a fairly quick—if proper resources are readily available—and inexpensive way of diagnosing Malaria.[43] With that being said, microscopy can have very low sensitivity when performed by unqualified personnel, is heavily-resource dependent, is often limited by quality control, and can be delayed in producing results when under certain conditions.[42] Low microscopy sensitivity can especially result in a variance in diagnostic rates. This variance in diagnosis can lead to excessive use of anti-malarial drugs or negligent treatment, and therefore higher rates of morbidity and the development of resistant strains of pathogens. Therefore, rapid diagnostic tests are more commonly used in low-resource areas.[38]

RDTs are often used and more readily available in underdeveloped communities.[38] RDTs utilize immunoassay to detect the presence of varying Malaria pathogens. One study compared the 13 best, according to the WHO-FIND Product Testing Programme, Malaria rapid tests and found that the best-performing test for detecting *P. falciparum* had a LOD of 0.8 ng/mL. The best performing test for the other three *P.* species was found to have an LOD of 25 ng/mL. Overall, *P. falciparum* was better tested for than the other three *Plasmodium* species.[44] These RDTs present with the same benefits as previously mentioned Point-of-Care-Tests, such as low cost, simplicity, and portability. Despite the numerous advantages, there are limitations to the current rapid tests for Malaria. Current RDTs often only test for *P. falciparum* or for the other three *P.* species and do not possess very low limits of detection.[44]

4.2.4 Recommendations for Dr. Beskok's Device

Current immunoassays for detection of malaria infection typically target histidine-rich protein 2 (HRP2) or

Plasmodium lactate dehydrogenase (pLDH). HRP2 is specific only to *P. falciparum* while pLDH is conserved across all human malaria species.[45] pLDH also has *Plasmodium* species-specific epitopes, which allows for discrimination between different *Plasmodium* species. Pf-pLDH is an epitope specific to *P. falciparum*, Pv-pLDH to *P. vivax*, and Pvom-pLDH to *P. vivax*, *P. ovale*, and *P. Malariae*. In addition, C-reactive protein (CRP), a human inflammation marker, has been investigated as a biomarker to differentiate viral from nonviral Malaria infections.[44] One study described the recent development of a multiplex device capable of detecting HRP2, Pv-pLDH, Pvom-pLDH, and CRP, with extremely low LODs (0.2 pg/ml, 9.3 pg/ml, 1.5 pg/ml, and 26.6 ng/ml, respectively) and without cross-reactivity. While this study used blood as the biofluid sample, as well as techniques that combined chemiluminescence and ELISA, it does provide evidence for the eventual development of a PoCD that can discriminate between Malaria *Plasmodium* species, as well as viral versus nonviral infections.[45] In conclusion, it is recommended that a PoCD be multiplex in its assay in order to increase test sensitivity and specificity, as well as elicit the most effective treatment plans for diagnosed persons, based on species specific infection types.

Another recent rapid Malaria test of interest is the Urine Malaria Test™, as it shows promise for diagnosing Malaria with urine as the biofluid sample. This PoCT is a simple dipstick test that detects the presence of P. falciparum, through antibody specific binding to HRP2. This urine test at a parasite density less than ≤ 200 parasites/μl had a sensitivity of 50% and at density ≥ 201 parasites/μl was 89.71% sensitive and 83.48% specific.[46] So, while the development and commercialization of a simple, rapid urine PoCD for Malaria is encouraging, future PoCDs should possess lower LODs, higher sensitivities, and higher specificities.

It is also important to consider the specific WHO recommendations for Malaria rapid diagnostic tests. The WHO sets its standards for Malaria RDTs as having a limit of detection of 200 parasites /μL, false positive rates less than 10%, and invalid rates less than 5%.[38]

4.2.4.1 Advantages and Limitations of Dr. Beskok's Device

While numerous rapid tests have been produced and implemented for use in the diagnosis of Malaria, there is still a great need for continued development. Current diagnostic methods are limited by low sensitivity, human error, low limits of detection, detection of a singular *Plasmodium* species, and biofluid sample used. Unlike current RDTs for Malaria, Dr. Beskok's device has a low LOD compared to current commercial RDTs. Dr. Beskok's device also promises not to be just rapid, but also Point-of-Care, enabling fast result turnaround and appropriate use in low-medical resource settings. In addition, further research could potentially allow for a Point-of-Care-Test that is multiplex and uses urine samples, rather than other biofluids.

The proposed device could possibly be limited by cost-per-test as well as the already-high volume of rapid tests for Malaria. RDTs and light microscopy both have estimated costs of diagnosis that range from 1.0 to 2.0 US dollars, making them extremely low-cost options.[44] Also, the development of a multitude of Malaria RDTs, especially an extremely sensitive multiplex test and a simple urine test, possibly hinder the need for a new diagnostic device. With that being said, the developed multiplex test uses blood, which is more invasive and difficult to collect, handle and store than urine, and is similar to the bulky, resource-dependent ELISA method. The Urine Malaria Test™ uses an easier-to-handle biofluid and is extremely simple and portable, but has poor limit of detection and sensitivity.

5. GREATEST IMPACT

5.1 Location and People

Point-of-Care-Devices are designed to provide care quickly and simply, lessening the need for sophisticated laboratory equipment. Generating quick results offer health professionals the ability to provide faster access to appropriate treatments, therefore improving both clinical and economic outcomes. Although the need for the development of Point-of-Care-Devices exists globally, factors which constitute POC technologies, such as their ability to deliver less costly care and their transportability, also allow for these devices to better address the heavy burdens of diseases that disproportionately affect those in remote locations. Throughout this report, measures which quantify the need for new diagnostic technologies for TB and Malaria were explored. In order to generate the highest levels of impact, employing a PoCD in areas with high incidence rates of these diseases are of most interest.

Tuberculosis is caused by an airborne pathogen that thrives in densely populated environments, meaning it is highly communicable. Key high-impact populations that are at the greatest risk of initial TB infection are adults during their most productive years and undernourished children. Those at highest risk of progressing from Latent TB to Active TB are children under five, those infected with HIV, and those in communities with increased incidence.[26]

Malaria is a vector-borne disease, with climate sensitive infected vectors (i.e., ticks and mosquitoes that prefer warm temperatures with high humidity and high rainfall).[41] The most vulnerable group affected by malaria is children under the age of five. Secondarily, pregnant women are adversely affected.[39]

Globally, the region with the highest incidence rates of these diseases is Sub-Saharan Africa, with TB ranking at 237 per 100,000 people and Malaria ranking at 209.5 per 100,000 people.[46,47] The country with the highest incidence rate of Malaria is Rwanda with 505.6 cases per every 100,000 people.[48] The country with the highest incidence rate of TB is Lesotho with 665 cases per every 100,000 people.[47] The country that ranks highest for the incidence rate of both TB and Malaria together is the Democratic Republic of Congo. The country that ranks second for the incidence rate of both TB and Malaria together is Sierra Leone.[47, 48]

In 2014, the largest Ebola virus epidemic began in Sierra Leone resulting in major loss of life and socioeconomic disruption in the region. Furthermore, it greatly reduced access to health services for diagnosis and treatment for the major diseases that are endemic to the region: malaria, HIV/AIDS, and tuberculosis. In 2018, the 10th outbreak of Ebola virus disease in the Democratic Republic of Congo began, producing similar reductions in

access to health services. Deterioration of health infrastructure in these countries and fear of seeking care as a result of these epidemics has exacerbated the already intense need to rebuild health infrastructure and propose solutions to decentralize care.[49]

In order to make the greatest impact, deployment of the PoCD in the Sub-Saharan region, most specifically the Democratic Republic of Congo and Sierra Leone are recommended. Furthermore, children under the age of 5 who suffer from malnutrition should be given special attention. Focusing in these locations and populations will best aid in accomplishing the Third Sustainable Development Goal set out by the UN: to ensure healthy lives and promote wellbeing for all at all ages.

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7. APPENDIX 1: TABLE OF FIGURES

8. APPENDIX 2: ACRONYMS

DALY:	Disability-Adjusted Life Years
ELISA:	Enzyme-Linked Immunosorbent Assay
Ig:	Immunoglobulin
PPD:	Purified Protein Derivative
PoCD:	Point–of–Care–Device
POC:	Point–of–Care
PoCT:	Point–of–Care–Testing
LoC:	Lab–on–a–Chip
LOD:	Limit Of Detection
MDG:	Millennium Development Goals
R&D:	Research and Development
RDT:	Rapid Diagnostic Test
SDG:	Sustainable Development Goals
SMU:	Southern Methodist University
TB:	Tuberculosis
TAG:	Treatment Action Group
UN:	United Nations
NIH:	National Institute of Health
NIAID:	National Institute of Allergy and Infectious Disease
WHO:	World Health Organization
RDT:	Rapid Diagnostic Test

9. APPENDIX 3: TABLES AND FIGURES

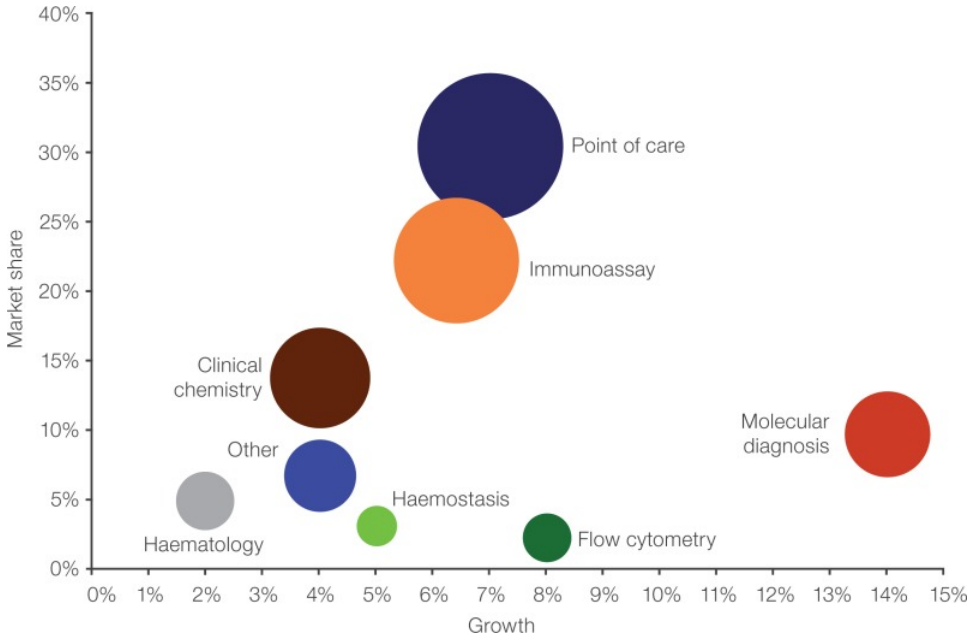

Figure 1: Market share and expected growth of in vitro diagnostics. In vitro diagnostic tests are the broader umbrella of diagnostic tests commonly used in the Global North. Immunoassay and POC devices fall within this market analysis.[9]

Device/ Technology	Unyvero™ Curetis	PLY-ELISA
Disease	Pneumonia	Pneumonia
Sensitivity	56.9%	56.6%
Specificity	63.2%	92.2%

Figure 2: Outlining sensitivity data collected from an analysis published in the Journal of Clinical Microbiology. The table directly compares found sensitivity.[12, 13]

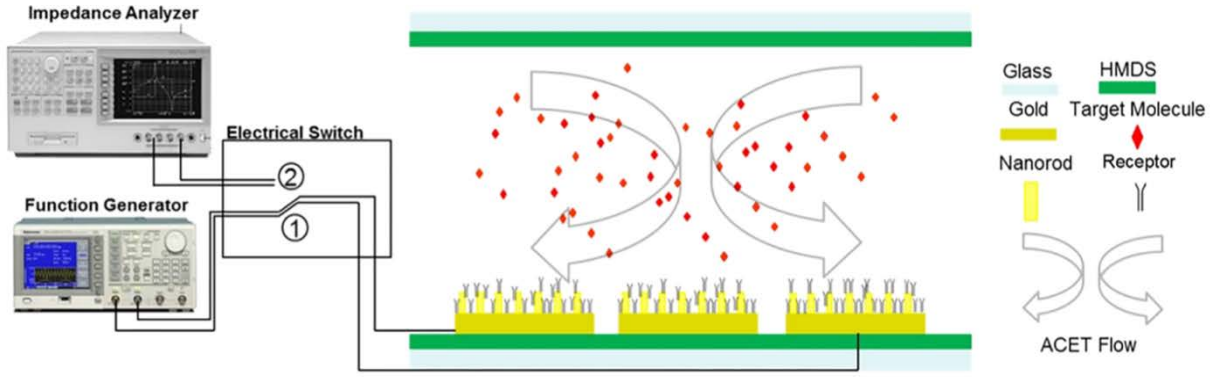

Figure 3: Configuration of the experimental setup. A function generator supplied an AC potential to induce ACET flow to concentrate target molecules onto the receptors-immobilized gold nanorod electrodes, and the binding was then measured using an impedance analyzer for the quantification of the analyte.[18]

Region	Global		Africa		South-East Asia	
	DALYs (000s)	% of total DALYs	DALYs (000s)	% of total DALYs	DALYs (000s)	% of total DALYs
Tuberculosis	51642.60	1.94	18393.29	3.07	23836.35	3.35
Malaria	37368.77	1.40	34679.77	5.79	1833.92	0.26

Figure 1: 2016 DALY for Global, the African region, and the South-East Asia region as provided by the WHO's report on leading causes of death from 2000 to 2016.[18]

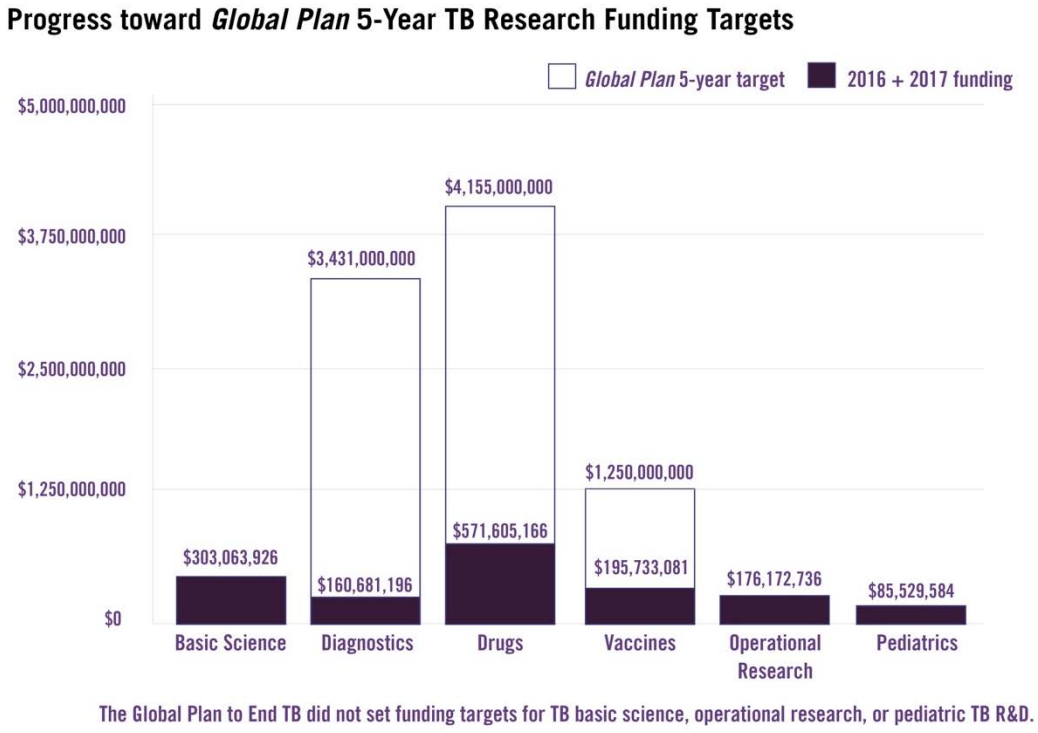

Figure 5: Graphic of compared funding from 2018 TAG report on 2005-2017 on Tuberculosis funding.[21]

Region	Global		Africa		South-East Asia	
	DALYs (000s)	% total of DALYs	DALYs (000s)	% total of DALYs	DALYs (000s)	% total of DALYs
Tuberculosis	51642.60	1.94	18393.29	3.07	23836.35	3.35
Malaria	37368.77	1.40	34679.77	5.79	1833.92	0.26

Figure 6: 2016 DALY for Global, the African region, and the South-East Asia region as provided by the WHO's report on leading causes of death from 2000 to 2016.[18]

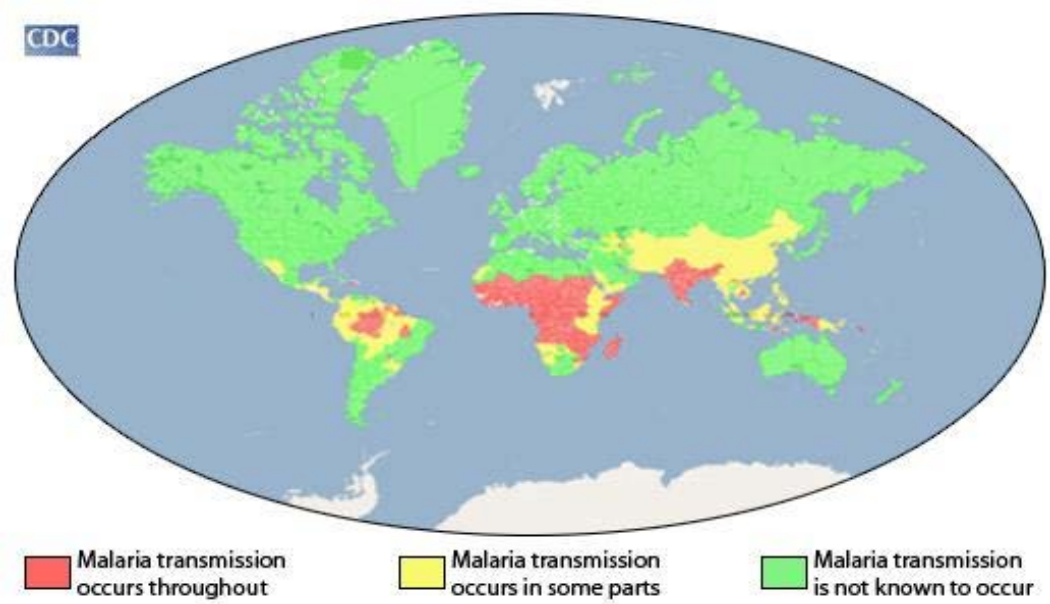

Figure 7: This map shows an approximation of the parts of the world where malaria transmission occurs, as supported by the CDC report of global Malaria.[37]

REGION	Year	Global Fund	PMI/USAID	World Bank	UK
AFRICA	2015	$738,786,118	$605,640,000	$9,229,381	$66,200,701
	2016	$904,764,475	$599,760,000	$30,269,301	$74,107,591
	2017	$1,076,502,060	$671,000,000	$30,269,301	$26,835,158
SOUTH-EAST ASIA	2015	$97,611,096	$9,270,000	$0	$5,199,619
	2016	$75,297,356	$10,200,000	$0	$13,022,911
	2017	$162,335,788	$10,000,000	$0	$4,715,737
EASTERN MEDITERRANEAN	2015	$78,596,307	$0	$-383,193	$0
	2016	$5,820,274	$5,820,274	$5,820,274	$5,820,274
	2017	$6,888,183	$6,888,183	$6,888,183	$6,888,183

Figure 8: Contributions (USD) reported by donors for Malaria Funding in 3 main effected WHO geographical regions.[23]