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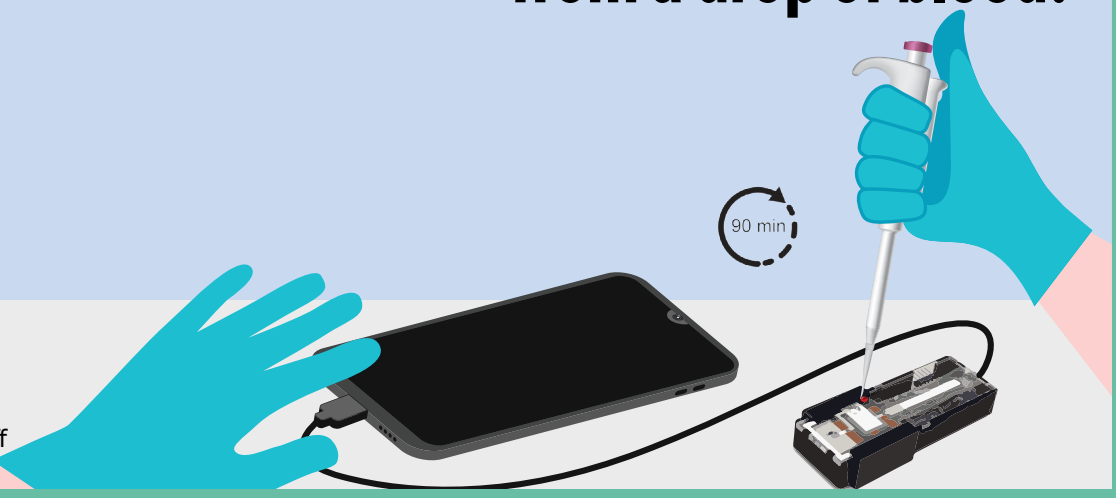
Can we detect HIV quickly and accurately from a drop of blood?



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Abstract

HIV stands for human immunodeficiency virus – a virus that makes your immune system weak so that it cannot fight off infection. It's not easy to detect HIV in the first few weeks after infection, but it's actually very important so the person can begin treatment and prevent transmission to another person.

This is why we wanted to create a cheap, portable, and reliable method to detect HIV in the early stages of infection. We were able to develop a small device that can rapidly detect the virus from blood. This low-power, automated device could be easily used by healthcare workers – especially in areas with limited resources.

Introduction

HIV (human immunodeficiency virus), as its name suggests, is a virus that makes the *immune system* weak and incapable of fighting off germs (Fig. 1). There are currently over 36 million people worldwide with HIV. Even though the virus can be deadly, there are medications that can help a person with HIV live a long and healthy life. It turns out that only half of the people with HIV receive the medications they need. One reason for this is the lack of cheap and reliable tests for early diagnosis. When people don't know they have HIV, they don't receive medication and therefore, can *transmit* the virus to others. There are cheap, rapid tests that healthcare workers can perform on-site, but they are not very accurate in the first weeks after infection. The highest quality methods detect the viral *nucleic acids* but these methods require:

1. expensive devices – to perform *amplification* or multiplication of the nucleic acids
2. refrigeration of samples and reagents
3. trained laboratory personnel
4. extensive sample preparation – to process the blood taken from the patient
5. a stable electrical supply

What if we could somehow develop a cheap, portable, and simple test that directly detects viral nucleic acids in a drop of *whole blood*?

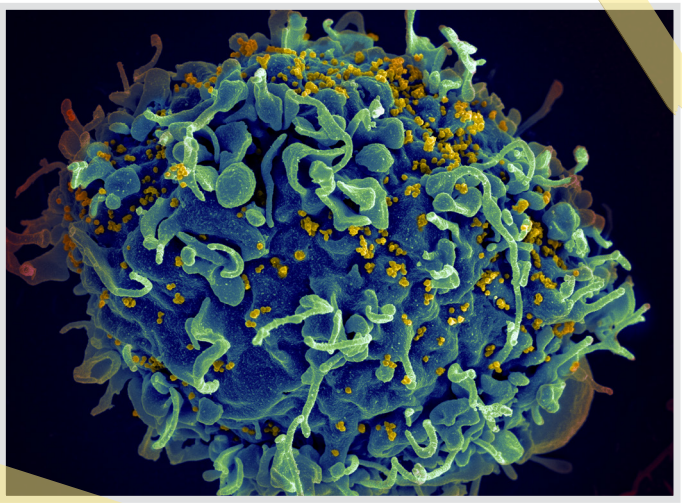

Figure 1:
A human T cell (blue) under attack by HIV (yellow), the virus that causes AIDS.
Source: National Institutes of Health

Methods

We wanted to design a small device that could easily and rapidly detect HIV (Fig. 2). Our device includes:

- A *membrane* with larger pores, which traps the blood cells in whole blood
- A membrane with smaller pores, which captures the virus

Viruses are really small! A lot smaller than blood cells.

- An amplification zone with dried reagents to multiply the viral nucleic acids through a test called *RT-LAMP*

This is one of the advantages of our methods – we can store dried reagents at room temperature!

- A small heater to heat the amplification zone
- Wax valves that melt when heated, allowing the sample to move through the different sections of the device
- A process called *LFIA* to detect the amplified viral nucleic acids in a simple, visual format

To make sure our device worked:

- We tested both membranes with colored particles which were the same size as red blood cells and the virus. This made it easy for us to see whether the membranes we chose were capturing the correct sizes of particles.
- We assembled the components of the device and tested fluid flow.
- We determined our test's *sensitivity* and *specificity* by running it on a *real-time PCR system*.
- We compared the HIV test results using dried reagents to results with fresh reagents.
- We tested the device using whole blood samples with HIV.

Detecting HIV with our device only requires four actions: deposit blood from a patient into the device, add a *buffer* into the device, seal the device with tape, and connect the device to a cell phone, computer, or battery to start heating the sample (so that amplification can occur). In 90 minutes the results are ready!

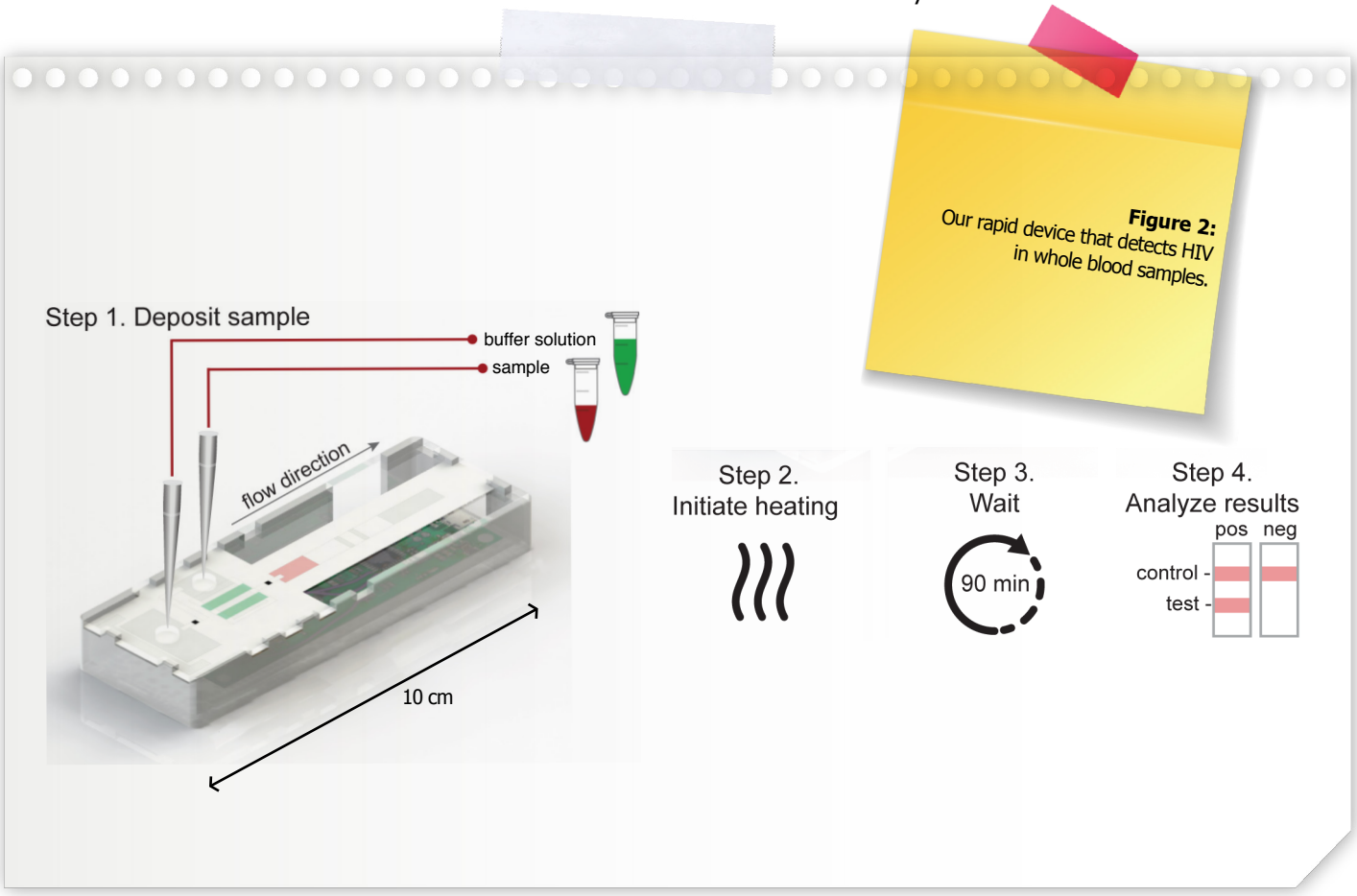

Figure 2: Our rapid device that detects HIV in whole blood samples.

Results

Our new, rapid device for detection of HIV from whole blood worked great!

- We found the device was sensitive and specific.
- Detection of HIV from whole blood samples using dried reagents showed accurate results even after the reagents had been stored for 21 days at room temperature.
- The device produced visual results 90 minutes after depositing the whole blood sample.
- The device is simple to use – there are only four operation steps.
- The device works just as well with virus in whole blood as it does with virus in water (Fig. 3).

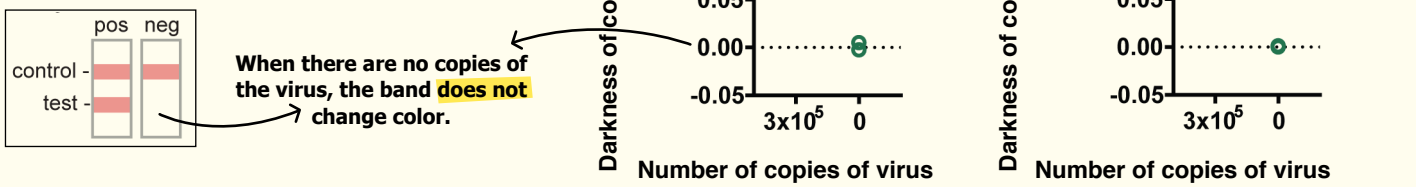

Figure 3:
Detection of HIV with our device with virus in water and in whole blood (4 samples in water, 3 in whole blood). If the device detected the virus, the test band changed color.

Discussion

Early HIV diagnosis is very important! The sooner the treatment begins, the better the chances for a long and normal life. And the earlier the diagnosis, the less likely transmission to someone else will occur.

Our newly created device turned out to be simple to use and very accurate for detecting HIV soon after infection. It has several advantages over laboratory-based tests. The device can be kept at room temperature so it can be used in the field where there are no refrigerators.

The device uses whole blood, so healthcare workers do not need to be trained to process blood samples. It's very fast too – it needs only 90 minutes to produce results!

Another factor is an economic one: it's cheap. The heater, which is reusable, costs around $70. The cost of each test is approximately $2. We think other scientists could even use this method and design to create a detection device for other viruses or bacteria.

Conclusion

There are a lot of myths about how HIV is transmitted. HIV spreads when infected blood or body fluids enter the body. This can happen through unprotected sex or sharing used needles. You cannot get HIV through sneezes, coughs, talking, or shaking hands.

Luckily, there are medications to treat HIV and the earlier treatment is started, the better. This makes early diagnosis very important. If you have questions or want to learn more about HIV, ask your doctor or other healthcare professional. There are also several resources listed on the next page.

Glossary of Key Terms

Amplification – a process used to multiply the molecules of RNA or DNA so they are easier to detect.

Buffer – a solution which maintains a stable environment for a chemical reaction.

HIV (human immunodeficiency virus) – a virus that attacks cells of the immune system. Over time, these cells become fewer and fewer and our immune system can no longer fight off common infections. Without proper treatment, HIV leads to AIDS - acquired immunodeficiency syndrome.

Immune system – the part of our body which fights off diseases, including infectious diseases.

RT-LAMP (reverse transcription loop-mediated isothermal amplification) – a laboratory test that amplifies/multiplies specific nucleic acids so that scientists can detect them if they are present. Unlike polymerase chain reaction (PCR), where cycles of temperature changes are needed (i.e. cycles of 95°C, 55°C and 72°C), LAMP tests require only one elevated temperature (usually between 60°C and 65°C).

LFIA (lateral-flow immunochromatographic assay) – a simple, visual test that tells us whether a molecule, such as viral RNA, is present or absent. The most famous example of LFIA is the home pregnancy test.

Membrane – a sheet of material that functions as a filter; it has pores (holes) of different sizes to separate and capture particles of different sizes.

Nucleic acids – viruses store their genetic information in either DNA - deoxyribo**nucleic** acid, or RNA - ribo**nucleic** acid. (For HIV it's RNA).

Real-time PCR system – a device that helps scientists perform PCR (or LAMP) and see the results as the reaction happens (in real time).

Sensitivity – if a test is highly sensitive it will find almost everyone who has the virus – the 'true positives' (it will almost never miss someone with the virus).

Specificity – if a test is highly specific it will correctly identify the people without the virus or with other diseases - the 'true negatives' (it will almost never detect the virus when it's not there).

Transmit/Transmission – to pass a virus or disease to another person.

Whole blood – blood that has not been separated into its components (e.g., red cells, white cells, and plasma). In laboratory HIV tests, red and white blood cells must be removed from whole blood before testing.

Check your understanding

1. What is the difference between HIV and AIDS?
2. Why do we need two different membranes (one for blood cells and another for the virus) in our device? Why can't we just use one membrane?
3. Why do we need to amplify/multiply the viral nucleic acids (RNA)?
4. What is the benefit of using dried reagents in our device?
5. Why is early HIV diagnosis important?

REFERENCES

Elizabeth Phillips, Taylor Moehling, Karin Ejendal, Orlando Hoilett, Kristin Byers, Laud Anthony Basing, Lauren Jankowski, Jackson Bennett, Li-Kai Lin, Lia Stanciu and Jacqueline Linnes (2019) *Microfluidic Rapid and Autonomous Analytical Device (MicroRAAD) to Detect HIV from Whole Blood Samples*. Lab on a Chip.
https://pubs.rsc.org/en/content/articlehtml/2019/lc/c9lc00506d

Seattle Children's Hospital: Questions Kids Ask About HIV (PDF)
https://www.seattlechildrens.org/pdf/PE1215.pdf

CDC: HIV
https://www.cdc.gov/hiv/default.html

KidsHealth: My Friend Has HIV. How Can I Help?
https://kidshealth.org/en/teens/friend-hiv.html