



NOMADS 2: Expanding Nanopore Sequencing of Malaria

Malaria is caused by a parasite and spread from person to person by the bite of an infected mosquito. The deadliest form of the malaria parasite, *Plasmodium falciparum*, is most common in Africa, where an estimated 90 percent of all malaria deaths occur. Tools to prevent and treat malaria are growing more advanced and easier to access, but just as our tools evolve, so too does the malaria parasite. As countries push to reduce and ultimately eliminate malaria, surveillance of the parasite will be critical to responding appropriately.

Most surveillance involves identifying the location and number of malaria cases. However, **genomic surveillance** provides much more information on the parasite causing the infection, which helps us to understand where the parasite came from. For example, during the SARS-CoV-2 pandemic, genomic surveillance was crucial in identifying new variants of the virus that were driving waves of infections.

For malaria, genomic surveillance can detect mutations (changes in the parasite's DNA sequence) that make the parasite resistant to antimalarial drugs or invisible to malaria tests. It can also assess how diverse the parasite population is and even help understand how the parasite is spreading. This DNA sequencing can be performed on easily collected dried blood spots (DBS) and provide an early warning signal to the threat of malaria drug resistance.

NOMADS 2

With support from the Bill & Melinda Gates Foundation (Gates Foundation), and working on-site with colleagues at Zambia's National Malaria Elimination Centre (NMEC), part of the Ministry of Health, our NOMADS team developed a cost-effective and scalable testing material (known as an assay) for antimalarial drug resistance in *P. falciparum* that uses a technique called nanopore sequencing. When we tested the assay at the Zambian Ministry of Health's National



Genomic sequencing takes place in the molecular lab on the grounds of Zambia's National Malaria Elimination Center. The NOMADS assay will be deployed on samples from across the country to routinely assess levels of drug resistance and diagnostic escape.

Malaria Elimination Centre (NMEC), we demonstrated that rapid in-country sequencing to identify antimalarial drug resistance was not only possible, but also required minimal infrastructure, allowing for the detection of drug resistance to happen closer to where policies and decisions are made. The NOMADS team also reduced sequencing costs from approximately US\$90 to US\$25 per sample.

Building on the success of NOMADS, renewed investment from the Gates Foundation, NOMADS 2 is focused on strengthening and expanding nanopore sequencing of malaria.

The partnership

NOMADS 2 is implemented in close collaboration with the NMEC and the following key partners:

- Max Planck Institute for Infection Biology (MPIIB): Under MPIIB's leadership, we will continuously improve the interpretation and analysis of sequencing outcomes for communication with policy stakeholders.

- Africa CDC: Aligning our activities with Africa's key public health body will allow us to leverage existing sequencing capabilities and streamline procurement and shipping across Africa.
- Ministries of health and national malaria programs in the new geographies.

Areas of focus

1. Integrate nanopore sequencing into routine surveillance in Zambia.

Drug resistance is a threat to malaria control efforts. Without an early warning system, drug-resistant parasites could become more common, leading to untreatable infections. We will work with the Ministry of Health to establish a genomic surveillance unit on the NMEC campus that will deploy the NOMADS assay on samples from across Zambia to routinely assess levels of drug resistance and diagnostic escape.

In collaboration with the NMEC, we will combine existing epidemiological and entomological data streams into sampling strategies. By regularly connecting researchers with decision-makers and other specialists, NOMADS 2 will provide data needed by malaria programs to make decisions on control efforts, for example, on identifying effective drugs.

2. Expand nanopore sequencing to new geographies through collaboration and capacity building.

NOMADS 2 aims to create a malaria nanopore sequencing community by building sequencing capacity across several countries. Through equipment provision, training, and remote support, we will empower in-country collaborators in at least three new target geographies to establish in-country malaria genomic surveillance. We will also develop training materials that will enable even more geographies to adopt the assay in their setting.

3. Optimize existing and develop novel nanopore sequencing assays for malaria molecular surveillance.

We will continue to optimize the existing NOMADS assay that focuses on drug resistance and diagnostic escape to reduce cost and increase performance. Simultaneously, in response to the needs of our collaborators, we will develop new approaches, such as enabling mosquito genomic surveillance.



The NOMADS assay takes approximately two days to go from dried blood spot to sequence data. This quickly provides decision makers with information on malaria control and elimination efforts.

NANOPORE EXPLAINER

Sequencing DNA is a very complex process. Historically, it required expensive equipment, massive computing power, and a host of experts to perform



the work. However, nanopore sequencing makes this technology affordable and maintenance-free, and uses a tool called the MinION that can fit into your pocket.

It works by threading individual DNA strands through extremely tiny openings (nanopores). As DNA moves through the pore, an electrical signal is created that can be decoded into a string of bases (A, C, G, or T). Nanopore can read very long fragments of DNA, and the analysis can be performed on a laptop in real time. By comparing the resulting strings of bases with a reference tool (the assay), mutations like drug resistance can be identified.

