Training Report on Sequencing in a Mobile Lab

Location and Duration

Location: Dar es Salaam
Duration: 15–21 September 2025

1. Background and Rationale

Genomic surveillance is a key tool for detecting and monitoring emerging pathogens. Metagenomic analysis of wastewater enables simultaneous detection of multiple pathogens circulating in a population, regardless of symptoms. Rapid barcoding sequencing (Rapid Barcoding, Oxford Nanopore) permits targeted, rapid identification of viruses such as Mpox (MPXV). Deploying a mobile laboratory for these analyses enables prompt field interventions and brings sequencing capability closer to epidemic contexts.

2. Training Objectives

- **Introduce** participants to the operation of a mobile sequencing laboratory.
- **Train** participants on protocols adapted for environmental (wastewater) and clinical samples.
- **Apply** a metagenomic protocol to DNA extracts from wastewater.
- **Apply** the Rapid Barcoding protocol to MPXV-positive extracts.
- **Strengthen** bioinformatics skills for analysis of generated data.

3. Training Activities and Procedures

Samples Submitted

- **20 DNA extracts** from wastewater samples.
- **5 DNA extracts** positive for MPXV from human clinical samples.

Protocols Applied

Metagenomics (20 wastewater extracts)

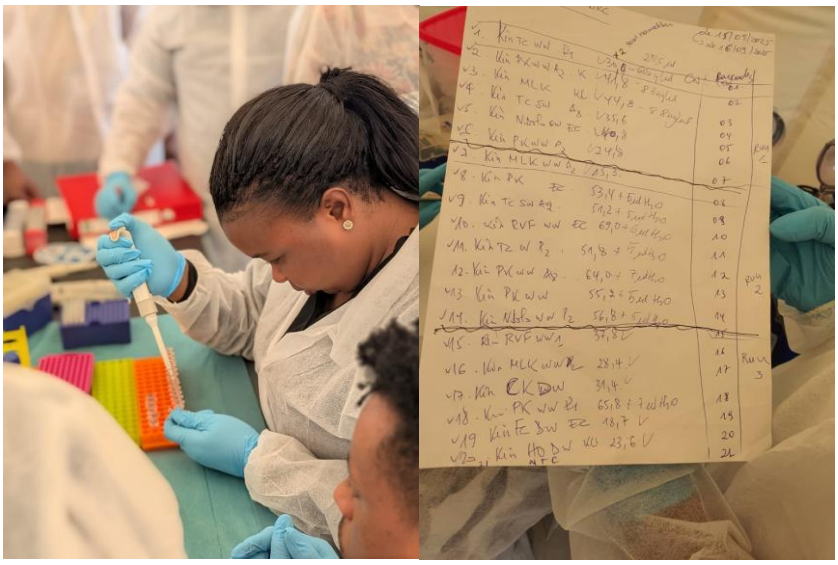
- DNA extraction and quality control.
- Preparation of metagenomic libraries.
- Sequencing using Oxford Nanopore (MinION, R10 flow cell).
- Bioinformatics analysis: read filtering, taxonomic and functional annotation.

Additional Workflows

- For ODIN/RDC DNA extracts: quantification → amplification → library preparation → sequencing → bioinformatics analyses → visualization and interpretation.
- For raw samples (drinking water and wastewater): filtration (pump and syringe) → extraction → nucleic acid purification → quantification → amplification → qPCR analyses.

Image 1. Presentation of the mobile lab

Images 2. Presentation of Protocols



Images 3. Library Preparation

Images 4. Loading of the first run of ODIN extracts.

Rapid Barcoding 5 MPXV-positive Extracts

- **Kit used:** SQK-RBK.
- **Multiplexing:** Five samples multiplexed on a single flow cell.
- **Sequencing platform:** Real-time rapid sequencing on MinION Mk1C.
- **Bioinformatics analysis:** Basecalling; demultiplexing; mapping to the MPXV reference sequence; consensus generation.

Image 5 Loading of the MPX library into the flow cell

Images 6. Syringe filtration of refrigerated samples collected 6 September — extraction of nucleic acids, NA, from the filters, followed by preservation of the filters for qPCR analysis on day 3.

Image7. DNA Extraction

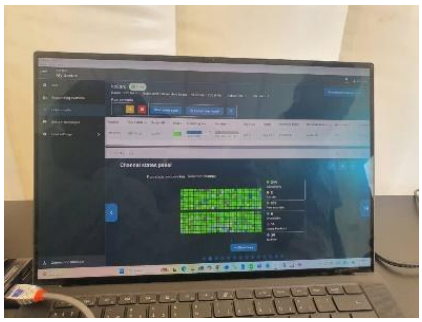
4. Results and Observations

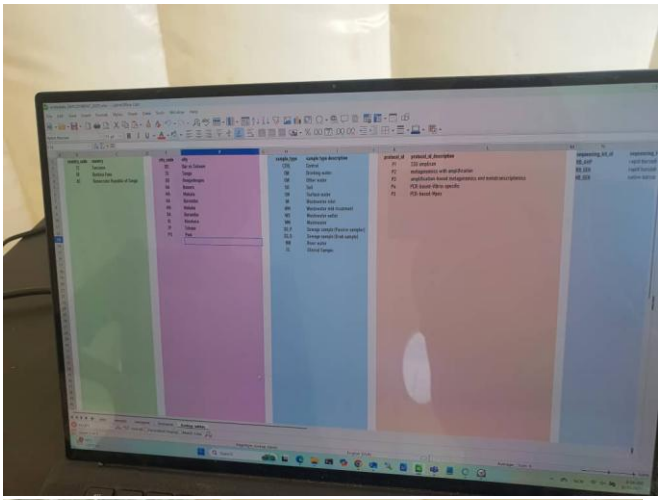
Metagenomics 20 wastewater extracts

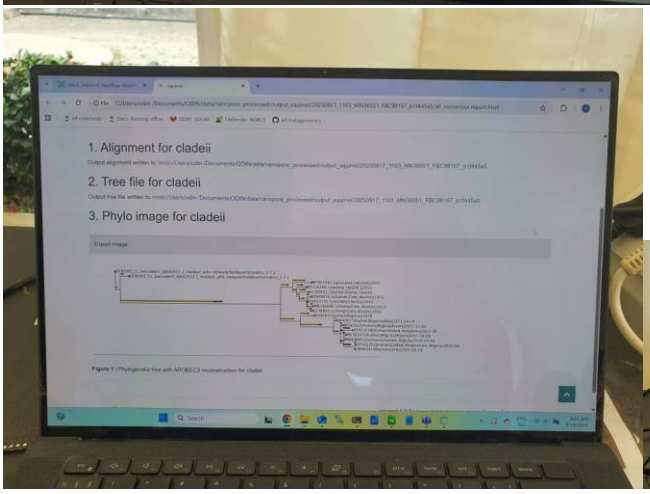
- **Detection:** A diverse spectrum of microorganisms detected including bacteria.
- **Significance:** Concrete demonstration of the role of environmental surveillance.

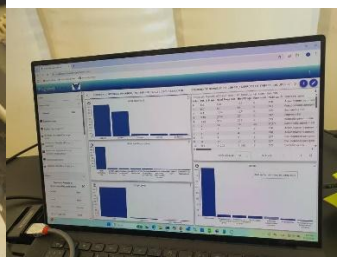
Rapid Barcoding 5 clinical MPXV extracts

- **Barcode read rate:** > 90% for two samples.
- **Genome coverage:** Good genomic coverage obtained for two of the five samples.
- **Confirmation:** MPXV confirmed in all five clinical extracts.









5. Challenges Encountered

- **Time constraints:** Insufficient time to complete the bioinformatics component; trainers performed the bioinformatics analyses themselves.
- **Connectivity:** Poor internet connection.

6. Recommendations

- **Integrate** environmental metagenomics as an early epidemiological surveillance tool.
- **Expand** use of Rapid Barcoding for targeted sequencing of other emerging pathogens.
- **Develop** a regional network of mobile laboratories to strengthen health surveillance.
- **Provide** ongoing training in bioinformatics and data management.

7. Conclusion

The training held in Dar es Salaam demonstrated the complementarity of two sequencing approaches in a mobile laboratory.

- Metagenomics on 20 wastewater DNA extracts illustrated environmental pathogen surveillance.
- Rapid Barcoding on five clinical MPXV samples demonstrated the speed and effectiveness of targeted sequencing.