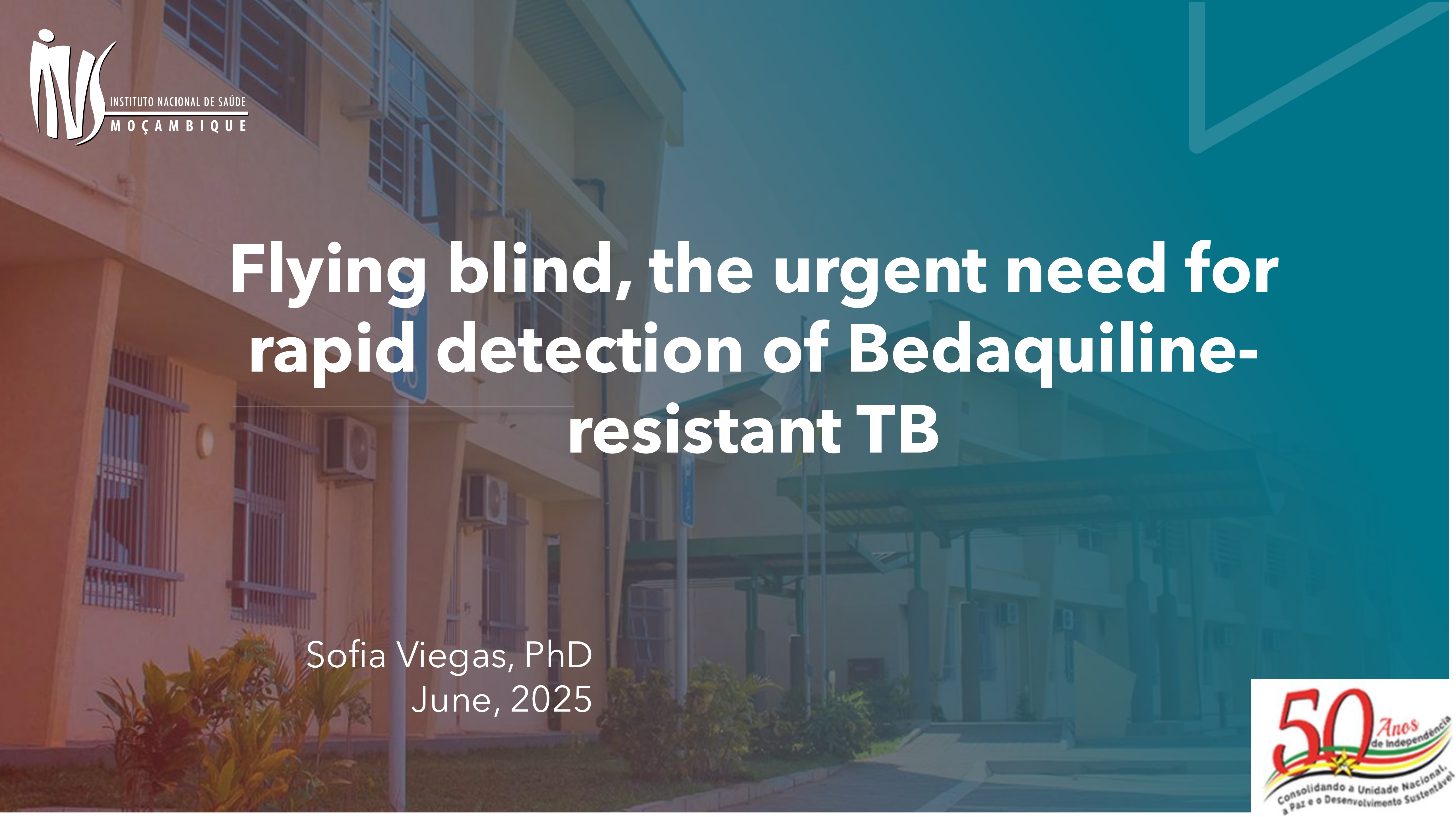
INSTITUTO NACIONAL DE SAÚDE
MOÇAMBIQUE



Flying blind, the urgent need for rapid detection of Bedaquiline-resistant TB

Sofia Viegas, PhD
June, 2025

50 Anos de Independência
Consolidando a Unidade Nacional, a Paz e o Desenvolvimento Sustentável

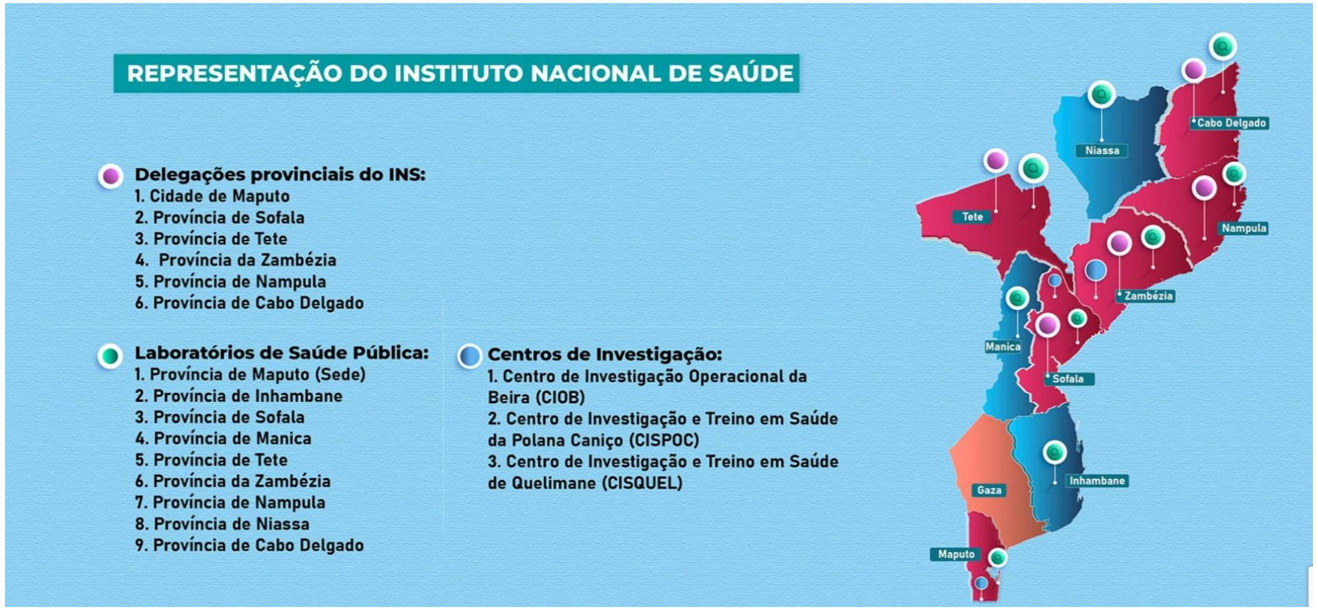
Presentation Outline

1. Mozambique and the *Instituto Nacional de Saúde*
2. TB epidemic in Mozambique
3. About Bedaquiline
4. The pathway to implement sequencing in Mozambique
5. Results of our research project
6. Next steps and concluding remarks

The *Instituto Nacional de Saúde* (INS) of Mozambique

- The main mandate is to generate evidence to inform policy-making decisions within the health sector in Mozambique.

- INS became an autonomous institution in 2017, turning into the national authority for activities related to the generation of evidence in health in Mozambique.



REPRESENTAÇÃO DO INSTITUTO NACIONAL DE SAÚDE

Delegações provinciais do INS:
1. Cidade de Maputo
2. Província de Sofala
3. Província de Tete
4. Província da Zambézia
5. Província de Nampula
6. Província de Cabo Delgado

Laboratórios de Saúde Pública:
1. Província de Maputo (Sede)
2. Província de Inhambane
3. Província de Sofala
4. Província de Manica
5. Província de Tete
6. Província da Zambézia
7. Província de Nampula
8. Província de Niassa
9. Província de Cabo Delgado

Centros de Investigação:
1. Centro de Investigação Operacional da Beira (CIOB)
2. Centro de Investigação e Treino em Saúde da Polana Caniço (CISPOC)
3. Centro de Investigação e Treino em Saúde de Quelimane (CISQUEL)

INS Responsibilities within the Health System

INSTITUTO NACIONAL DE SAÚDE
MOÇAMBIQUE

01 Clinical, biomedical, health system and socio-anthropological research

02 Laboratory Reference Services and Systems

03 Surveillance, Emergency Response and Health Observation

04 Health Communication

05 Training in Public Health

06 Partnerships with national and international institutions for research, education and public health activities

TB epidemic in Mozambique

- In 2011, TB was declared a National Emergency;
- TB; MDR-TB; TB-HIV high burden country;
- TB incidence: 361 / 100,000 population;
- Total cases notified (2023): 116 317;
- RR-TB cases: 4200
- New cases of MDR/RR-TB: 2.99%
- Previous treated cases of MDR/RR-TB: 12.47%
- Bacteriologically-confirmed cases tested for susceptibility to rifampicin: 85%
- 242 GeneXpert equipment, 34 Truenats, 36 of them are 10 colours and 5 culture laboratories.

About Bedaquiline...

1. Bedaquiline as a Critical Drug in DR-TB Treatment

- Bedaquiline (BDQ) is a core component of WHO-recommended regimens for MDR- and XDR-TB.
- Increasing global rollout: >150 countries have introduced BDQ since WHO endorsement in 2013.

2. Emerging Resistance: A Growing Threat

- BDQ resistance-associated mutations (e.g., in *Rv0678*, *atpE*, *pepQ*) increasingly reported.
- Cross-resistance with clofazimine due to *Rv0678* mutations complicates treatment options.

3. Diagnostic Gap

- No WHO-endorsed rapid molecular assay for BDQ resistance currently exists.
- WGS and MGIT-based DST are limited by infrastructure, cost, and long turnaround times.
- Absence of point-of-care diagnostics impairs timely resistance detection and regimen optimization.
- **Can TNGS be a solution?**

INSTITUTO NACIONAL DE SAÚDE
MOÇAMBIQUE

The pathway to implement sequencing in Mozambique

50 Anos de Independência
Consolidando a Unidade Nacional, a Paz e o Desenvolvimento Sustentável

Creating country capacity and strategic partnerships

2017

TB Sequel

2019

Conception of the project (tNGS and WGS)

2020

Capacity Building:

CTAB DNA extraction

2021-2022

WGS in RCB

In country Capacity builing for tNGS

2023

WGS and Bioinformatics capacity building

2023

Publication Lancet Infectious Disease

2024

Concept and discussion the tNGS Pilot study

2025

TNGS analysis/ paper in preparation

Pre and Post Pandemic sequencing capacity at INS

2016 HIV-1 sequencing

2019

TB sequencing

Sanger

2020
COVID
Pandemic

2021-2024

NGS

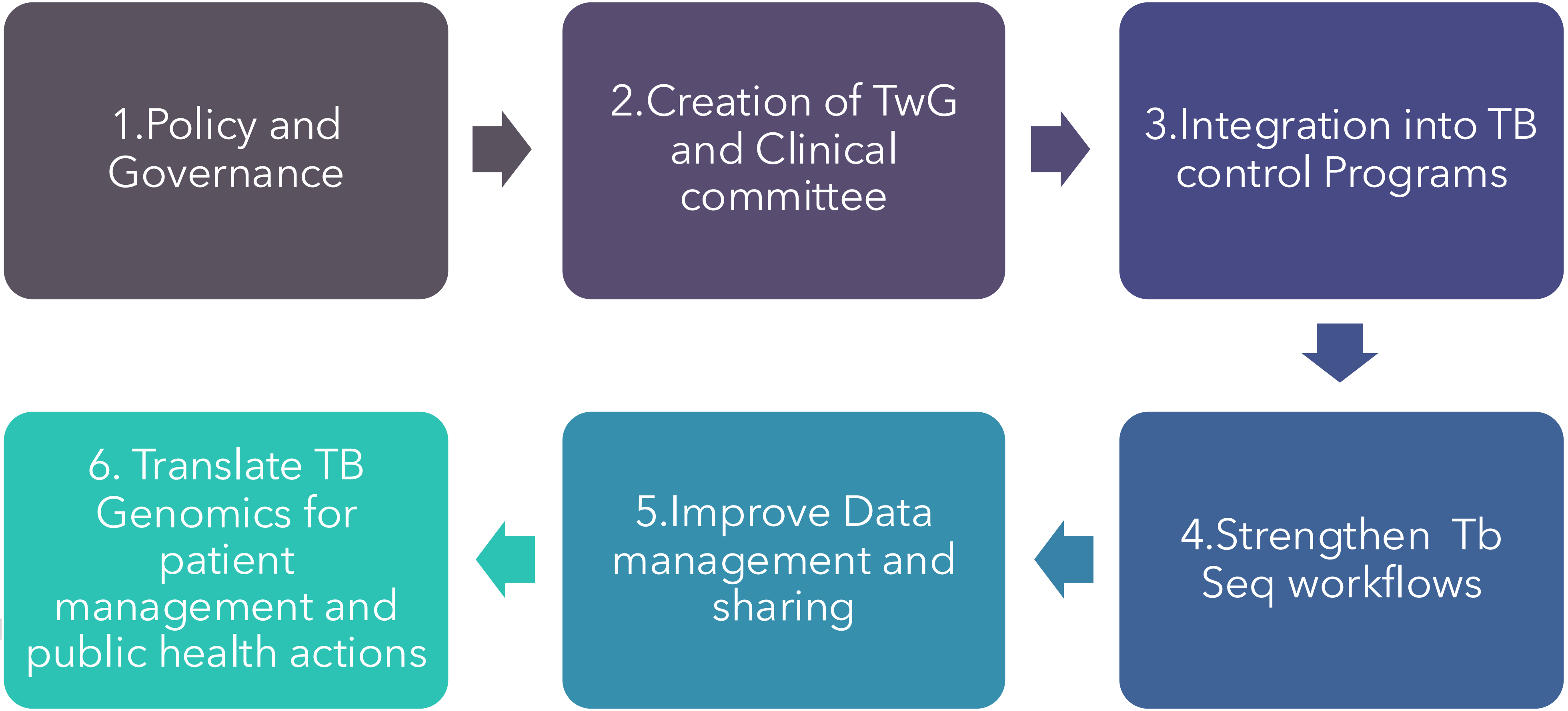
2021

2022

2023

2024

Framework to NGS implementation in the National Health System in Mozambique



1.Policy and Governance

2.Creation of TwG and Clinical committee

3.Integration into TB control Programs

4.Strengthen Tb Seq workflows

5.Improve Data management and sharing

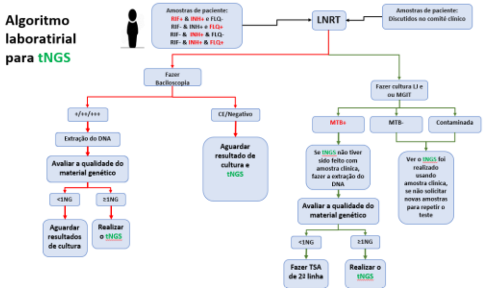
6. Translate TB Genomics for patient management and public health actions

Strategies that are being adopted currently to implementing tNGS in Mozambique

1. Capacity Building Installed for TB sequencing (2020-2021)

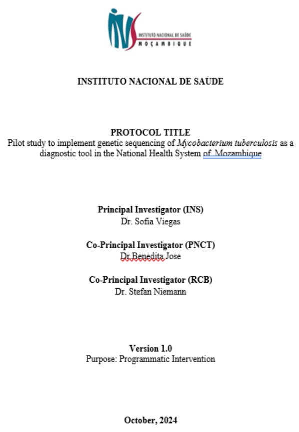
2.Advocacy of TB Genomics with other Stakeholders in Mozambique, NTP and partners
(2022)

3. I workshop to discuss TB Genomics surveillance strategy
(2023)



4.Training of the other TB culture laboratories
(2023)

5. Discussion of the implementation of TB sequencing into algorithm pilot study (2024)



INSTITUTO NACIONAL DE SAÚDE

PROTOCOL TITLE
Pilot study to implement genetic sequencing of *Mycobacterium tuberculosis* as a diagnostic tool in the National Health System of Mozambique

Principal Investigator (INS)
Dr. Sofia Viegas

Co-Principal Investigator (PNCT)
Dr.Benedita Jose

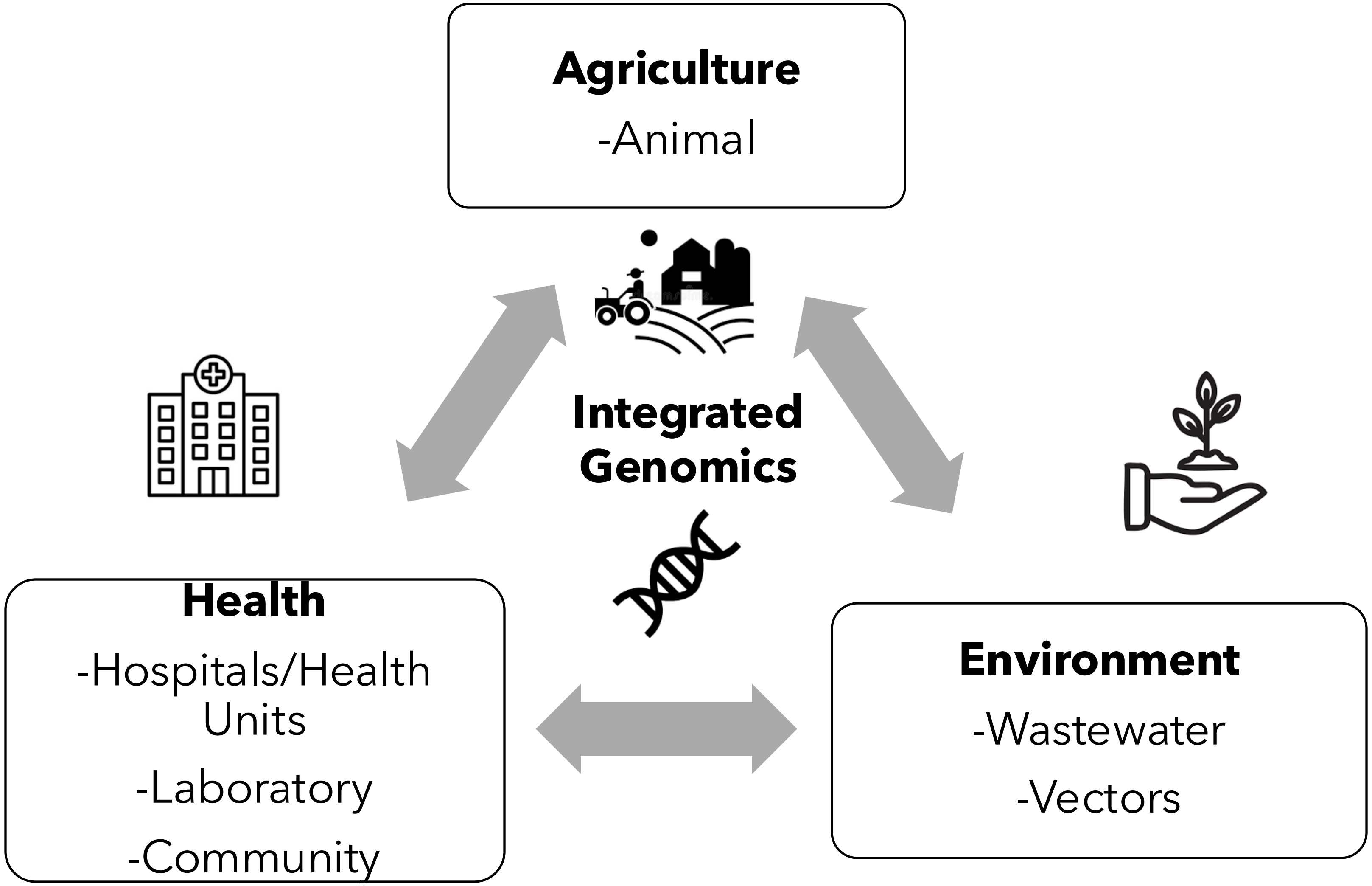
Co-Principal Investigator (RCB)
Dr. Stefan Niemann

Version 1.0
Purpose: Programmatic Intervention

October, 2024

6. Pilot to implement the tNGS in the HHS (2025)

Establish an integrated approach for genomic surveillance



Agriculture

-Animal

Integrated Genomics

Health

-Hospitals/Health Units

-Laboratory

-Community

Environment

-Wastewater

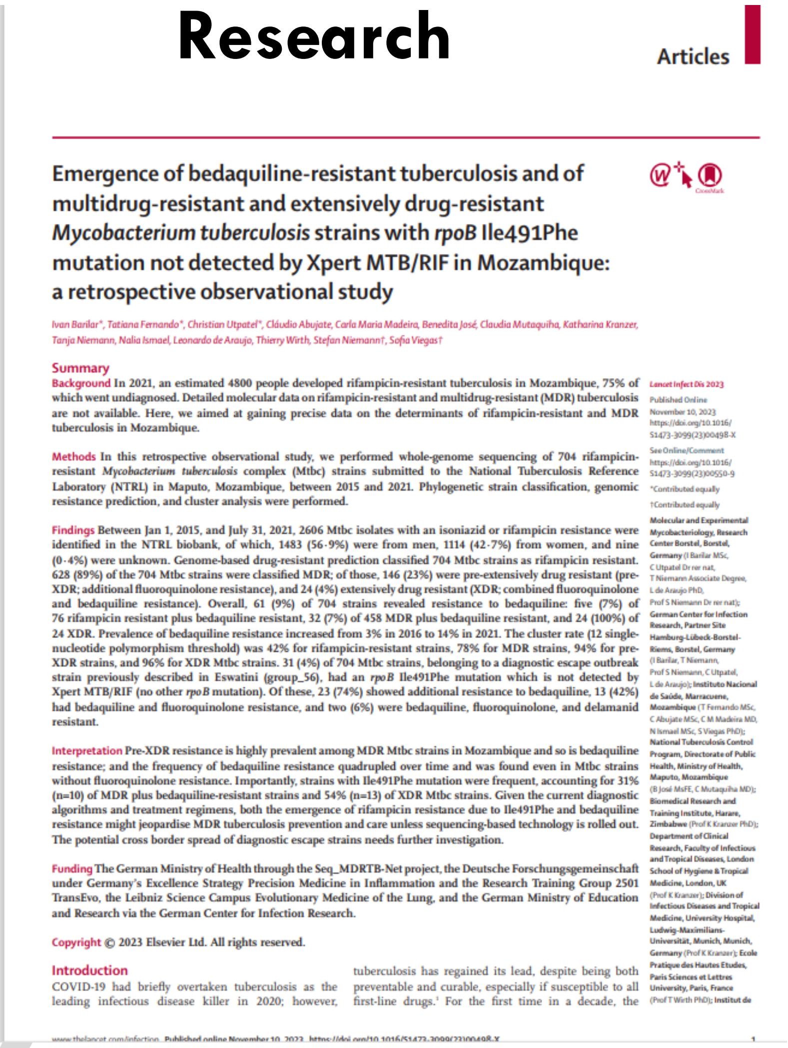
-Vectors

Areas of Interest:

1. HIV and STDs, TB and Malaria
2. AMR
3. VPDs
4. Emerging, zoonotic and respiratory diseases
5. Enteric diseases
6. Vector-borne disease
7. Non-communicable diseases

Evidence based implementation

Research



Articles

Emergence of bedaquiline-resistant tuberculosis and of multidrug-resistant and extensively drug-resistant *Mycobacterium tuberculosis* strains with *rpoB* Ile491Phe mutation not detected by Xpert MTB/RIF in Mozambique: a retrospective observational study

Ivan Barilar*, Tatiana Fernando*, Christian Utpatel*, Cláudio Abujate, Carla Maria Madeira, Benedita José, Claudia Mutaquiha, Katharina Kranzer, Tanja Niemann, Nalia Ismael, Leonardo de Araujo, Thierry Wirth, Stefan Niemann†, Sofia Viegas†

Summary

Background In 2021, an estimated 4800 people developed rifampicin-resistant tuberculosis in Mozambique, 75% of which went undiagnosed. Detailed molecular data on rifampicin-resistant and multidrug-resistant (MDR) tuberculosis are not available. Here, we aimed at gaining precise data on the determinants of rifampicin-resistant and MDR tuberculosis in Mozambique.

Methods In this retrospective observational study, we performed whole-genome sequencing of 704 rifampicin-resistant *Mycobacterium tuberculosis* complex (Mtbc) strains submitted to the National Tuberculosis Reference Laboratory (NTRL) in Maputo, Mozambique, between 2015 and 2021. Phylogenetic strain classification, genomic resistance prediction, and cluster analysis were performed.

Findings Between Jan 1, 2015, and July 31, 2021, 2606 Mtbc isolates with an isoniazid or rifampicin resistance were identified in the NTRL biobank, of which, 1483 (56·9%) were from men, 1114 (42·7%) from women, and nine (0·4%) were unknown. Genome-based drug-resistant prediction classified 704 Mtbc strains as rifampicin resistant. 628 (89%) of the 704 Mtbc strains were classified MDR; of those, 146 (23%) were pre-extensively drug resistant (pre-XDR; additional fluoroquinolone resistance), and 24 (4%) extensively drug resistant (XDR; combined fluoroquinolone and bedaquiline resistance). Overall, 61 (9%) of 704 strains revealed resistance to bedaquiline: five (7%) of 76 rifampicin resistant plus bedaquiline resistant, 32 (7%) of 458 MDR plus bedaquiline resistant, and 24 (100%) of 24 XDR. Prevalence of bedaquiline resistance increased from 3% in 2016 to 14% in 2021. The cluster rate (12 single-nucleotide polymorphism threshold) was 42% for rifampicin-resistant strains, 78% for MDR strains, 94% for pre-XDR strains, and 96% for XDR Mtbc strains. 31 (4%) of 704 Mtbc strains, belonging to a diagnostic escape outbreak strain previously described in Eswatini (group_56), had an *rpoB* Ile491Phe mutation which is not detected by Xpert MTB/RIF (no other *rpoB* mutation). Of these, 23 (74%) showed additional resistance to bedaquiline, 13 (42%) had bedaquiline and fluoroquinolone resistance, and two (6%) were bedaquiline, fluoroquinolone, and delamanid resistant.

Interpretation Pre-XDR resistance is highly prevalent among MDR Mtbc strains in Mozambique and so is bedaquiline resistance; and the frequency of bedaquiline resistance quadrupled over time and was found even in Mtbc strains without fluoroquinolone resistance. Importantly, strains with Ile491Phe mutation were frequent, accounting for 31% (n=10) of MDR plus bedaquiline-resistant strains and 54% (n=13) of XDR Mtbc strains. Given the current diagnostic algorithms and treatment regimens, both the emergence of rifampicin resistance due to Ile491Phe and bedaquiline resistance might jeopardise MDR tuberculosis prevention and care unless sequencing-based technology is rolled out. The potential cross border spread of diagnostic escape strains needs further investigation.

Funding The German Ministry of Health through the Seq_MDRTB-Net project, the Deutsche Forschungsgemeinschaft under Germany's Excellence Strategy Precision Medicine in Inflammation and the Research Training Group 2501 TransEvo, the Leibniz Science Campus Evolutionary Medicine of the Lung, and the German Ministry of Education and Research via the German Center for Infection Research.

Introduction

COVID-19 had briefly overtaken tuberculosis as the leading infectious disease killer in 2020; however, tuberculosis has regained its lead, despite being both preventable and curable, especially if susceptible to all first-line drugs.[1] For the first time in a decade, the

Lancet Infect Dis 2023

Published Online
November 10, 2023
https://doi.org/10.1016/S1473-3099(23)00498-X

See Online/Comment
https://doi.org/10.1016/S1473-3099(23)00550-9

*Contributed equally

†Contributed equally

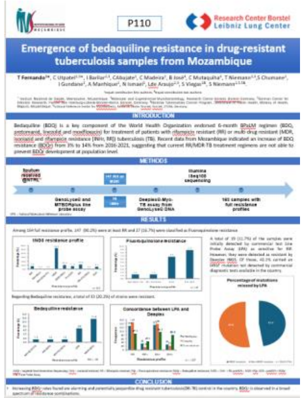
Molecular and Experimental Mycobacteriology, Research Center Borstel, Borstel, Germany (I Barilar MSc, C Utpatel Dr rer nat, T Niemann Associate Degree, L de Araujo PhD, Prof S Niemann Dr rer nat); **German Center for Infection Research, Partner Site Hamburg-Lübeck-Borstel-Riems, Borstel, Germany** (I Barilar, T Niemann, Prof S Niemann, C Utpatel, L de Araujo); **Instituto Nacional de Saúde, Marracuene, Mozambique** (T Fernando MSc, C Abujate MSc, C M Madeira MD, N Ismael MSc, S Viegas PhD); **National Tuberculosis Control Program, Directorate of Public Health, Ministry of Health, Maputo, Mozambique** (B José MsFE, C Mutaquiha MD); **Biomedical Research and Training Institute, Harare, Zimbabwe** (Prof K Kranzer PhD); **Department of Clinical Research, Faculty of Infectious and Tropical Diseases, London School of Hygiene & Tropical Medicine, London, UK** (Prof K Kranzer); **Division of Infectious Diseases and Tropical Medicine, University Hospital, Ludwig-Maximilians-Universität, Munich, Munich, Germany** (Prof K Kranzer); **Ecole Pratique des Hautes Etudes, Paris Sciences et Lettres University, Paris, France** (Prof T Wirth PhD); **Institut de**

Surveillance

Genetic sequencing for surveillance of drug resistance *Mycobacterium tuberculosis complex (Mtbc):Laboratory based surveillance*

-Whole Genome Sequencing

-Target Next Generation Sequencing



P110

Research Center Borstel
Leibniz Lung Center

Emergence of bedaquiline resistance in drug-resistant tuberculosis samples from Mozambique

INTRODUCTION

Bedaquiline (BDQ) is a key component of the World Health Organization endorsed 6-month BPaLM regimen (BDQ, pretomanid, linezolid and moxifloxacin) for treatment of patients with rifampicin resistant (RR) or multi drug resistant (MDR, isoniazid and rifampicin resistance [INHr, RR]) tuberculosis (TB). Recent data from Mozambique indicated an increase of BDQ resistance (BDQr) from 3% to 14% from 2016-2021, suggesting that current RR/MDR-TB treatment regimens are not able to prevent BDQr development at population level.

METHODS

RESULTS

CONCLUSION

Patient Care: Pilot

To assess the feasibility of integrating tNGS for TB as an additional diagnostic tool for detecting DR-TB in Maputo Province and Maputo City,

To evaluate the potential of incorporation tNGS into the national diagnostic algorithm.

Emergence of bedaquiline-resistant tuberculosis and of multidrug-resistant and extensively drug-resistant Mycobacterium tuberculosis strains with rpoB Ile491Phe mutation not detected by Xpert MTB/RIF in Mozambique: a retrospective observational study

Emergence of bedaquiline-resistant tuberculosis and of multidrug-resistant and extensively drug-resistant *Mycobacterium tuberculosis* strains with *rpoB* Ile491Phe mutation not detected by Xpert MTB/RIF in Mozambique: a retrospective observational study

Ivan Barilar*, Tatiana Fernando*, Christian Utpatel*, Cláudio Abujate, Carla Maria Madeira, Benedita José, Claudia Mutaquiha, Katharina Kranzer, Tanja Niemann, Nalia Ismael, Leonardo de Araujo, Thierry Wirth, Stefan Niemann†, Sofia Viegas†

Lancet Infect Dis 2023

https://doi.org/10.1016/S1473-3099(23)00498-X

See Online/Comment
https://doi.org/10.1016/S1473-3099(23)00550-9

*Contributed equally

†Contributed equally

Molecular and Experimental Mycobacteriology, Research Center Borstel, Borstel, Germany (I Barilar MSc, C Utpatel Dr rer nat, T Niemann Associate Degree, L de Araujo PhD, Prof S Niemann Dr rer nat); **German Center for Infection Research, Partner Site Hamburg-Lübeck-Borstel-Riems, Borstel, Germany** (I Barilar, T Niemann, Prof S Niemann, C Utpatel, L de Araujo); **Instituto Nacional de Saúde, Marracuene, Mozambique** (T Fernando MSc, C Abujate MSc, C M Madeira MD, N Ismael MSc, S Viegas PhD); **National Tuberculosis Control Program, Directorate of Public Health, Ministry of Health, Maputo, Mozambique** (B José MsFE, C Mutaquiha MD); **Biomedical Research and Training Institute, Harare, Zimbabwe** (Prof K Kranzer PhD); **Department of Clinical Research, Faculty of Infectious and Tropical Diseases, London School of Hygiene & Tropical Medicine, London, UK** (Prof K Kranzer); **Division of Infectious Diseases and Tropical Medicine, University Hospital, Ludwig-Maximilians-Universität, Munich, Munich, Germany** (Prof K Kranzer); **Ecole Pratique des Hautes Etudes, Paris Sciences et Lettres University, Paris, France** (Prof T Wirth PhD); **Institut de**

Summary

Background In 2021, an estimated 4800 people developed rifampicin-resistant tuberculosis in Mozambique, 75% of which went undiagnosed. Detailed molecular data on rifampicin-resistant and multidrug-resistant (MDR) tuberculosis are not available. Here, we aimed at gaining precise data on the determinants of rifampicin-resistant and MDR tuberculosis in Mozambique.

Methods In this retrospective observational study, we performed whole-genome sequencing of 704 rifampicin-resistant *Mycobacterium tuberculosis* complex (Mtbc) strains submitted to the National Tuberculosis Reference Laboratory (NTRL) in Maputo, Mozambique, between 2015 and 2021. Phylogenetic strain classification, genomic resistance prediction, and cluster analysis were performed.

Findings Between Jan 1, 2015, and July 31, 2021, 2606 Mtbc isolates with an isoniazid or rifampicin resistance were identified in the NTRL biobank, of which, 1483 (56·9%) were from men, 1114 (42·7%) from women, and nine (0·4%) were unknown. Genome-based drug-resistant prediction classified 704 Mtbc strains as rifampicin resistant. 628 (89%) of the 704 Mtbc strains were classified MDR; of those, 146 (23%) were pre-extensively drug resistant (pre-XDR; additional fluoroquinolone resistance), and 24 (4%) extensively drug resistant (XDR; combined fluoroquinolone and bedaquiline resistance). Overall, 61 (9%) of 704 strains revealed resistance to bedaquiline: five (7%) of 76 rifampicin resistant plus bedaquiline resistant, 32 (7%) of 458 MDR plus bedaquiline resistant, and 24 (100%) of 24 XDR. Prevalence of bedaquiline resistance increased from 3% in 2016 to 14% in 2021. The cluster rate (12 single-nucleotide polymorphism threshold) was 42% for rifampicin-resistant strains, 78% for MDR strains, 94% for pre-XDR strains, and 96% for XDR Mtbc strains. 31 (4%) of 704 Mtbc strains, belonging to a diagnostic escape outbreak strain previously described in Eswatini (group_56), had an *rpoB* Ile491Phe mutation which is not detected by Xpert MTB/RIF (no other *rpoB* mutation). Of these, 23 (74%) showed additional resistance to bedaquiline, 13 (42%) had bedaquiline and fluoroquinolone resistance, and two (6%) were bedaquiline, fluoroquinolone, and delamanid resistant.

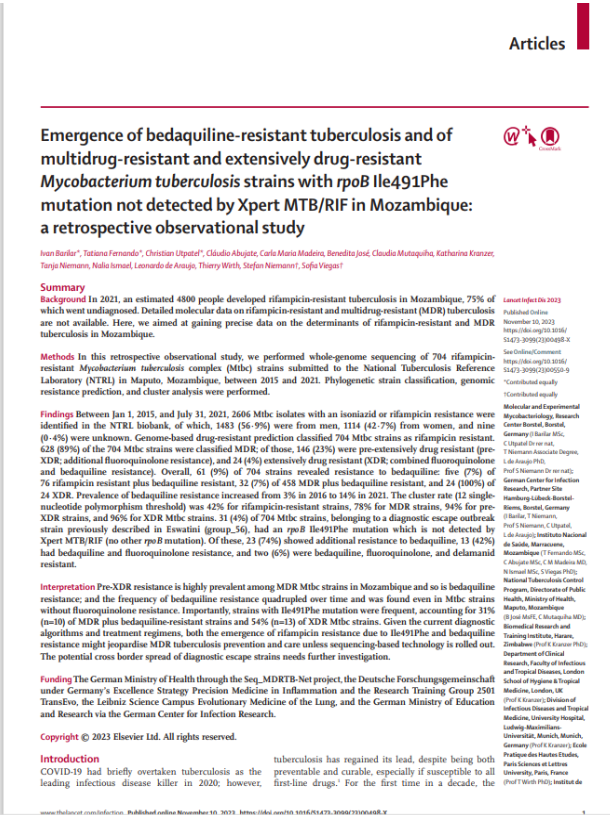
Interpretation Pre-XDR resistance is highly prevalent among MDR Mtbc strains in Mozambique and so is bedaquiline resistance; and the frequency of bedaquiline resistance quadrupled over time and was found even in Mtbc strains without fluoroquinolone resistance. Importantly, strains with Ile491Phe mutation were frequent, accounting for 31% (n=10) of MDR plus bedaquiline-resistant strains and 54% (n=13) of XDR Mtbc strains. Given the current diagnostic algorithms and treatment regimens, both the emergence of rifampicin resistance due to Ile491Phe and bedaquiline resistance might jeopardise MDR tuberculosis prevention and care unless sequencing-based technology is rolled out. The potential cross border spread of diagnostic escape strains needs further investigation.

Funding The German Ministry of Health through the Seq_MDRTB-Net project, the Deutsche Forschungsgemeinschaft under Germany's Excellence Strategy Precision Medicine in Inflammation and the Research Training Group 2501 TransEvo, the Leibniz Science Campus Evolutionary Medicine of the Lung, and the German Ministry of Education and Research via the German Center for Infection Research.

Introduction

COVID-19 had briefly overtaken tuberculosis as the leading infectious disease killer in 2020; however, tuberculosis has regained its lead, despite being both preventable and curable, especially if susceptible to all first-line drugs.[1] For the first time in a decade, the

Bedaquiline resistance rase from 3-14% and overall Fluoroquinolone resistance among MDRs were 23% between 2016-2021



Articles

Emergence of bedaquiline-resistant tuberculosis and of multidrug-resistant and extensively drug-resistant *Mycobacterium tuberculosis* strains with *rpoB* Ile491Phe mutation not detected by Xpert MTB/RIF in Mozambique: a retrospective observational study

Ivan Barilar, Tatiana Fernando*, Christian Utpatel*, Cláudio Abujate, Carla Maria Madeira, Benedita José, Claudia Mutaquiha, Katharina Kranzer, Tanja Niemann, Nalia Ismael, Leonardo de Araujo, Thierry Wirth, Stefan Niemann†, Sofia Viegas†*

Summary

Background In 2021, an estimated 4800 people developed rifampicin-resistant tuberculosis in Mozambique, 75% of which went undiagnosed. Detailed molecular data on rifampicin-resistant and multidrug-resistant (MDR) tuberculosis are not available. Here, we aimed at gaining precise data on the determinants of rifampicin-resistant and MDR tuberculosis in Mozambique.

Methods In this retrospective observational study, we performed whole-genome sequencing of 704 rifampicin-resistant *Mycobacterium tuberculosis* complex (Mtbc) strains submitted to the National Tuberculosis Reference Laboratory (NTRL) in Maputo, Mozambique, between 2015 and 2021. Phylogenetic strain classification, genomic resistance prediction, and cluster analysis were performed.

Findings Between Jan 1, 2015, and July 31, 2021, 2606 Mtbc isolates with an isoniazid or rifampicin resistance were identified in the NTRL biobank, of which, 1483 (56·9%) were from men, 1114 (42·7%) from women, and nine (0·4%) were unknown. Genome-based drug-resistant prediction classified 704 Mtbc strains as rifampicin resistant. 628 (89%) of the 704 Mtbc strains were classified MDR; of those, 146 (23%) were pre-extensively drug resistant (pre-XDR; additional fluoroquinolone resistance), and 24 (4%) extensively drug resistant (XDR; combined fluoroquinolone and bedaquiline resistance). Overall, 61 (9%) of 704 strains revealed resistance to bedaquiline: five (7%) of 76 rifampicin resistant plus bedaquiline resistant, 32 (7%) of 458 MDR plus bedaquiline resistant, and 24 (100%) of 24 XDR. Prevalence of bedaquiline resistance increased from 3% in 2016 to 14% in 2021. The cluster rate (12 single-nucleotide polymorphism threshold) was 42% for rifampicin-resistant strains, 78% for MDR strains, 94% for pre-XDR strains, and 96% for XDR Mtbc strains. 31 (4%) of 704 Mtbc strains, belonging to a diagnostic escape outbreak strain previously described in Eswatini (group_56), had an *rpoB* Ile491Phe mutation which is not detected by Xpert MTB/RIF (no other *rpoB* mutation). Of these, 23 (74%) showed additional resistance to bedaquiline, 13 (42%) had bedaquiline and fluoroquinolone resistance, and two (6%) were bedaquiline, fluoroquinolone, and delamanid resistant.

Interpretation Pre-XDR resistance is highly prevalent among MDR Mtbc strains in Mozambique and so is bedaquiline resistance; and the frequency of bedaquiline resistance quadrupled over time and was found even in Mtbc strains without fluoroquinolone resistance. Importantly, strains with Ile491Phe mutation were frequent, accounting for 31% (n=10) of MDR plus bedaquiline-resistant strains and 54% (n=13) of XDR Mtbc strains. Given the current diagnostic algorithms and treatment regimens, both the emergence of rifampicin resistance due to Ile491Phe and bedaquiline resistance might jeopardise MDR tuberculosis prevention and care unless sequencing-based technology is rolled out. The potential cross border spread of diagnostic escape strains needs further investigation.

Funding The German Ministry of Health through the Seq_MDRTB-Net project, the Deutsche Forschungsgemeinschaft under Germany's Excellence Strategy Precision Medicine in Inflammation and the Research Training Group 2501 TransEvo, the Leibniz Science Campus Evolutionary Medicine of the Lung, and the German Ministry of Education and Research via the German Center for Infection Research.

Introduction

COVID-19 had briefly overtaken tuberculosis as the leading infectious disease killer in 2020; however, tuberculosis has regained its lead, despite being both preventable and curable, especially if susceptible to all first-line drugs.[1] For the first time in a decade, the

Lancet Infect Dis 2023

Published Online
November 10, 2023
https://doi.org/10.1016/S1473-3099(23)00498-X

See Online/Comment
https://doi.org/10.1016/S1473-3099(23)00550-9

*Contributed equally

†Contributed equally

Molecular and Experimental Mycobacteriology, Research Center Borstel, Borstel, Germany (I Barilar MSc, C Utpatel Dr rer nat, T Niemann Associate Degree, L de Araujo PhD, Prof S Niemann Dr rer nat); **German Center for Infection Research, Partner Site Hamburg-Lübeck-Borstel-Riems, Borstel, Germany** (I Barilar, T Niemann, Prof S Niemann, C Utpatel, L de Araujo); **Instituto Nacional de Saúde, Marracuene, Mozambique** (T Fernando MSc, C Abujate MSc, C M Madeira MD, N Ismael MSc, S Viegas PhD); **National Tuberculosis Control Program, Directorate of Public Health, Ministry of Health, Maputo, Mozambique** (B José MsFE, C Mutaquiha MD); **Biomedical Research and Training Institute, Harare, Zimbabwe** (Prof K Kranzer PhD); **Department of Clinical Research, Faculty of Infectious and Tropical Diseases, London School of Hygiene & Tropical Medicine, London, UK** (Prof K Kranzer); **Division of Infectious Diseases and Tropical Medicine, University Hospital, Ludwig-Maximilians-Universität, Munich, Munich, Germany** (Prof K Kranzer); **Ecole Pratique des Hautes Etudes, Paris Sciences et Lettres University, Paris, France** (Prof T Wirth PhD); **Institut de**

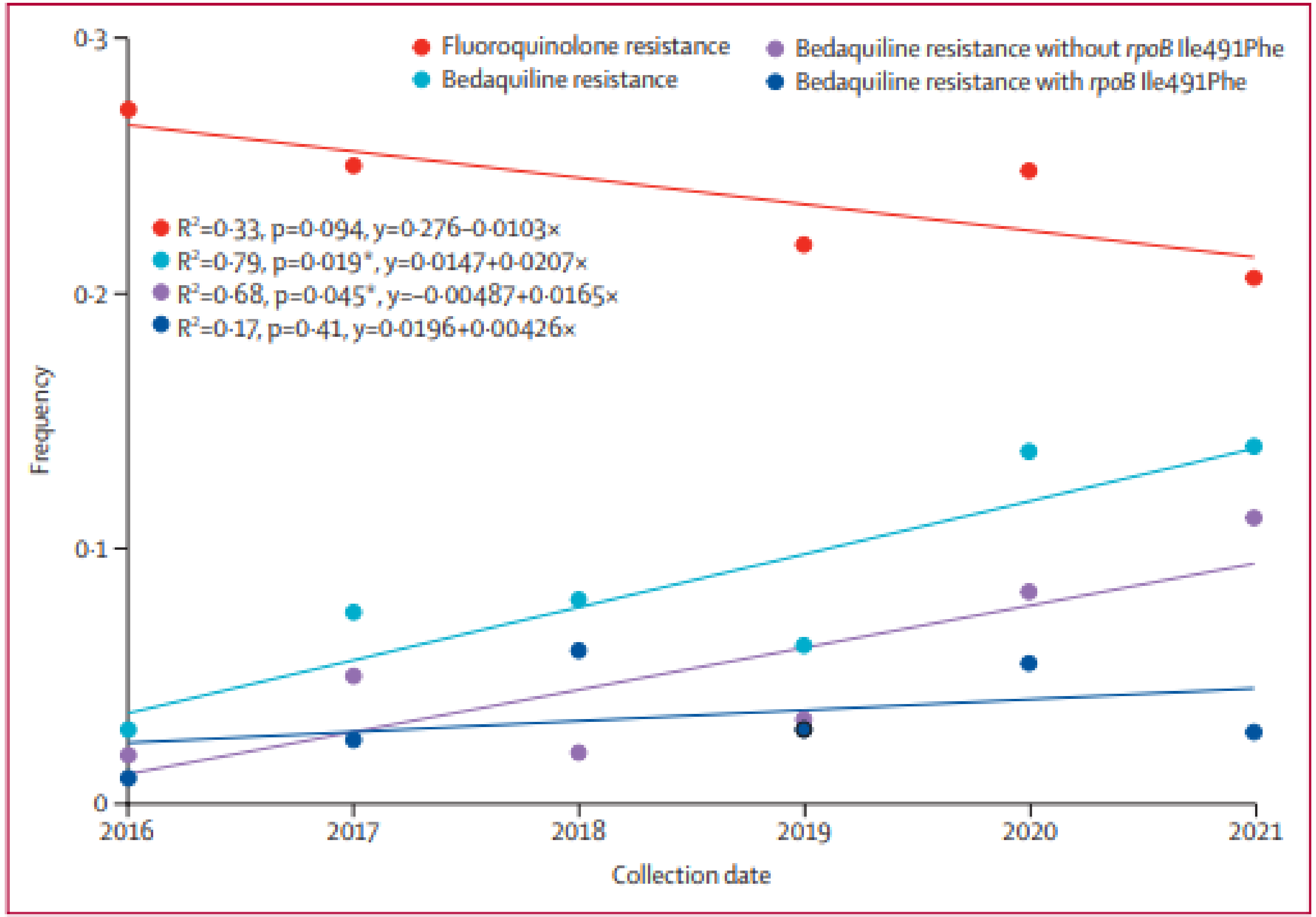
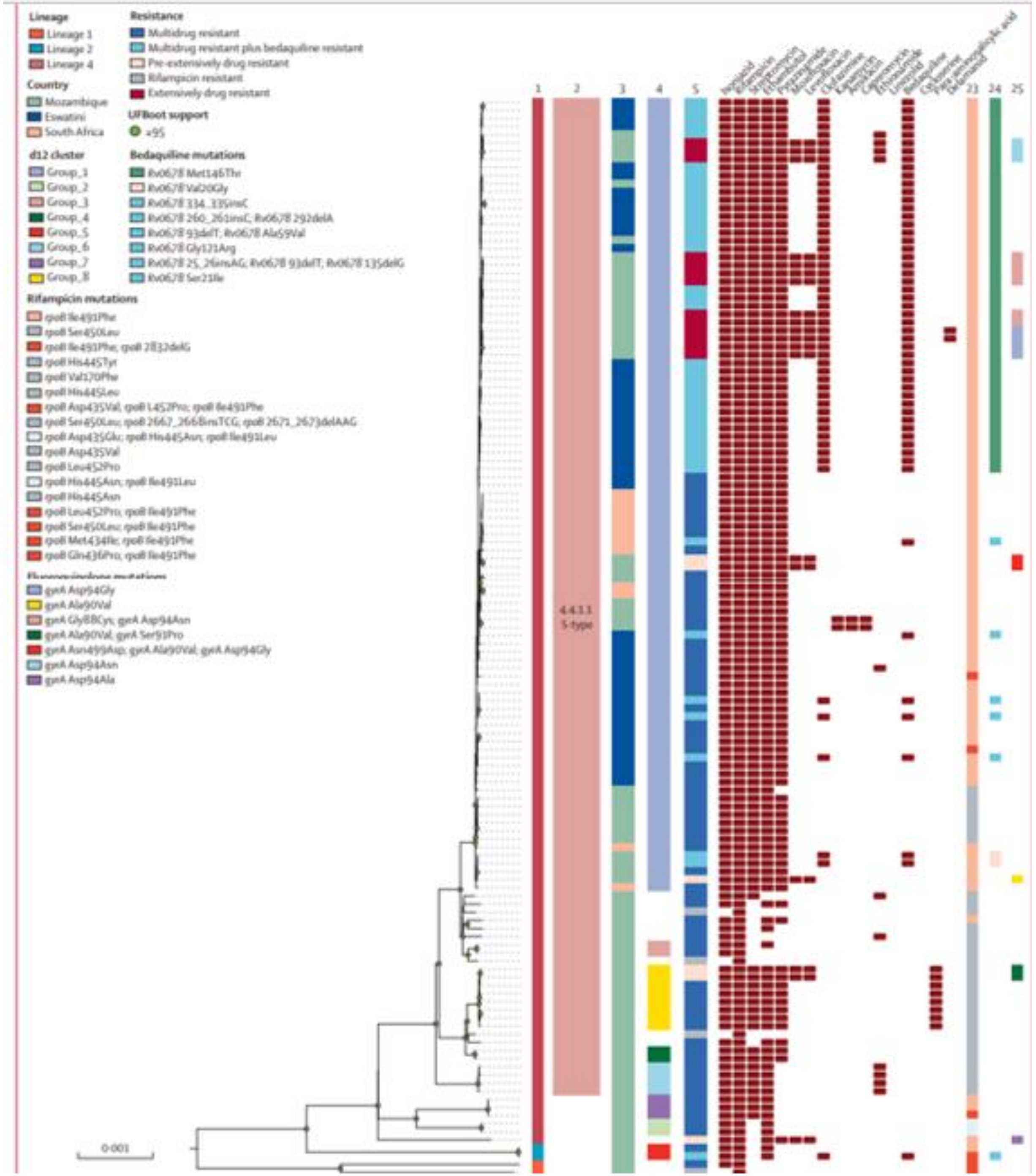


Figure 1: Frequency of bedaquiline and fluoroquinolone resistance in Mycobacterium tuberculosis complex strains over time

Source: Barilar *et al.*, 2024



Diagnostic outbreak strain I491F mutation

MDR+BDQr (31%) and XDR (54%) in Mozambique are due to strains with the I491F mutation

wrong diagnosis led to additional resistances over time

- Comparison of strains from 3 countries supports cross border spread
- By integrating tNGS in the algorithm I491F mutation spread can be prevented for better patient care

Preliminary Results - Whole Genome Sequencing (n=161)

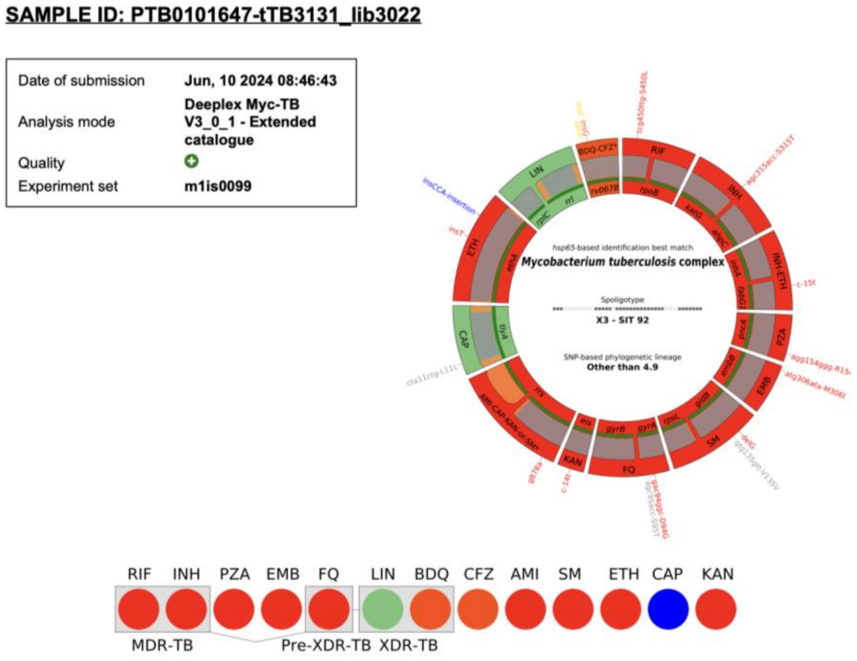
	Paper 2016-2021 N=704	**Recent 2022-2024 N=161**
Lineage 1	9%	19%
Lineage 2	22%	13%
Lineage 3	3%	5%
Lineage 4	66%	64%
rpoB Ile491Phe	4%	2%
BDQr	**9% (3%-14%)**	**19% (18%-22%)**

Bedaquiline resistance

	Paper 2016-2021	**Recent 2022-2024**
RR TB	5/76 (7%)	3/14 (14.3%)
MDR	32/458 (7%)	11/63 (17.5%)
XDR	24/24 (100%)	15/15 (100%)
rpoB Ile491Phe	24/61 (39.3%)	2/6 (33.3%)

WGS Preliminary Results (n=161) - BPaL M drugs

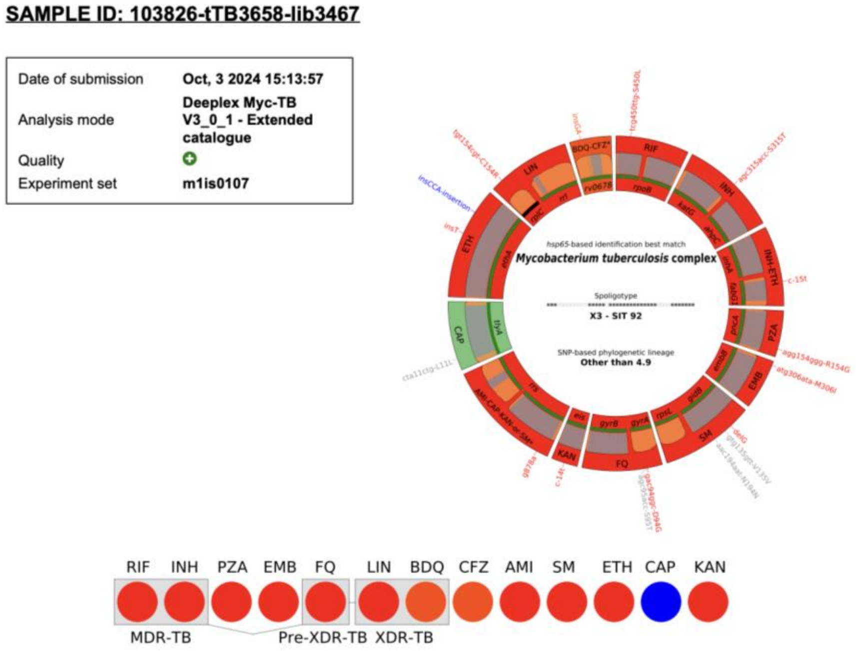
Examples of Deeplex shocking results from Mozambique



SAMPLE ID: PTB0101647-tTB3131_lib3022

Date of submission	**Jun, 10 2024 08:46:43**
Analysis mode	**Deeplex Myc-TB V3_0_1 - Extended catalogue**
Quality	⊕
Experiment set	**m1is0099**

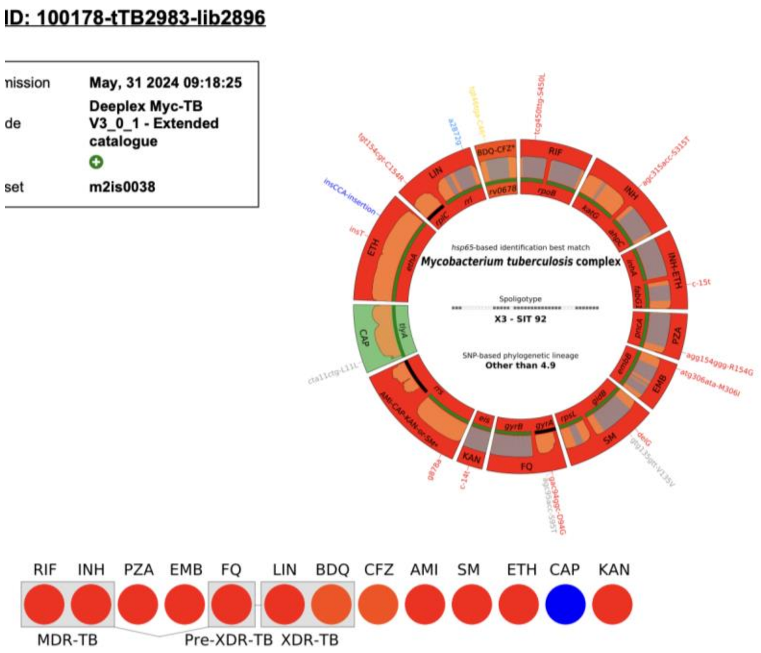
PTB101647
Inhambane
35Y
Female
Previous treated
Pre XDR-TB



SAMPLE ID: 103826-tTB3658-lib3467

Date of submission	**Oct, 3 2024 15:13:57**
Analysis mode	**Deeplex Myc-TB V3_0_1 - Extended catalogue**
Quality	⊕
Experiment set	**m1is0107**

PTB103826
Maputo Provincia
32Y
Male
New case
MDR-TB

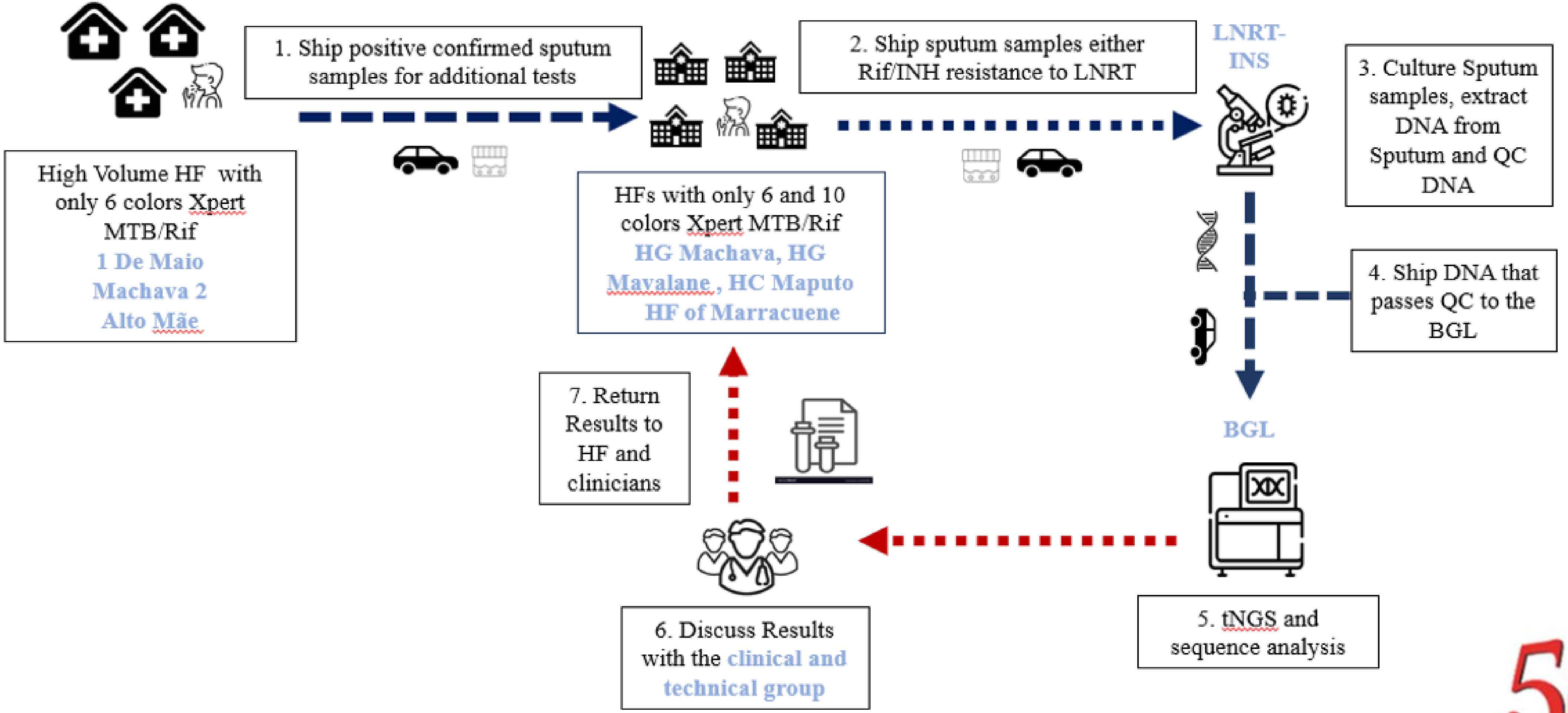


ID: 100178-tTB2983-lib2896

nission	**May, 31 2024 09:18:25**
de	**Deeplex Myc-TB V3_0_1 - Extended catalogue**
	⊕
set	**m2is0038**

PTB100178
Maputo Provincia
30Y
Male
Previous treated
Pre XDR-TB

Pilot study flowchart



1. Ship positive confirmed sputum samples for additional tests

2. Ship sputum samples either Rif/INH resistance to LNRT

3. Culture Sputum samples, extract DNA from Sputum and QC DNA

4. Ship DNA that passes QC to the BGL

5. tNGS and sequence analysis

6. Discuss Results with the **clinical and technical group**

7. Return Results to HF and clinicians

High Volume HF with only 6 colors Xpert MTB/Rif
1 De Maio
Machava 2
Alto Mãe

HFs with only 6 and 10 colors Xpert MTB/Rif
HG Machava, HG Mavalane, HC Maputo HF of Marracuene

Next Steps:

❑Implement tNGS pilot study in Maputo.

❑Strengthen current WGS analysis.

❑Expand sample selection and referencing to other TB culture laboratories.

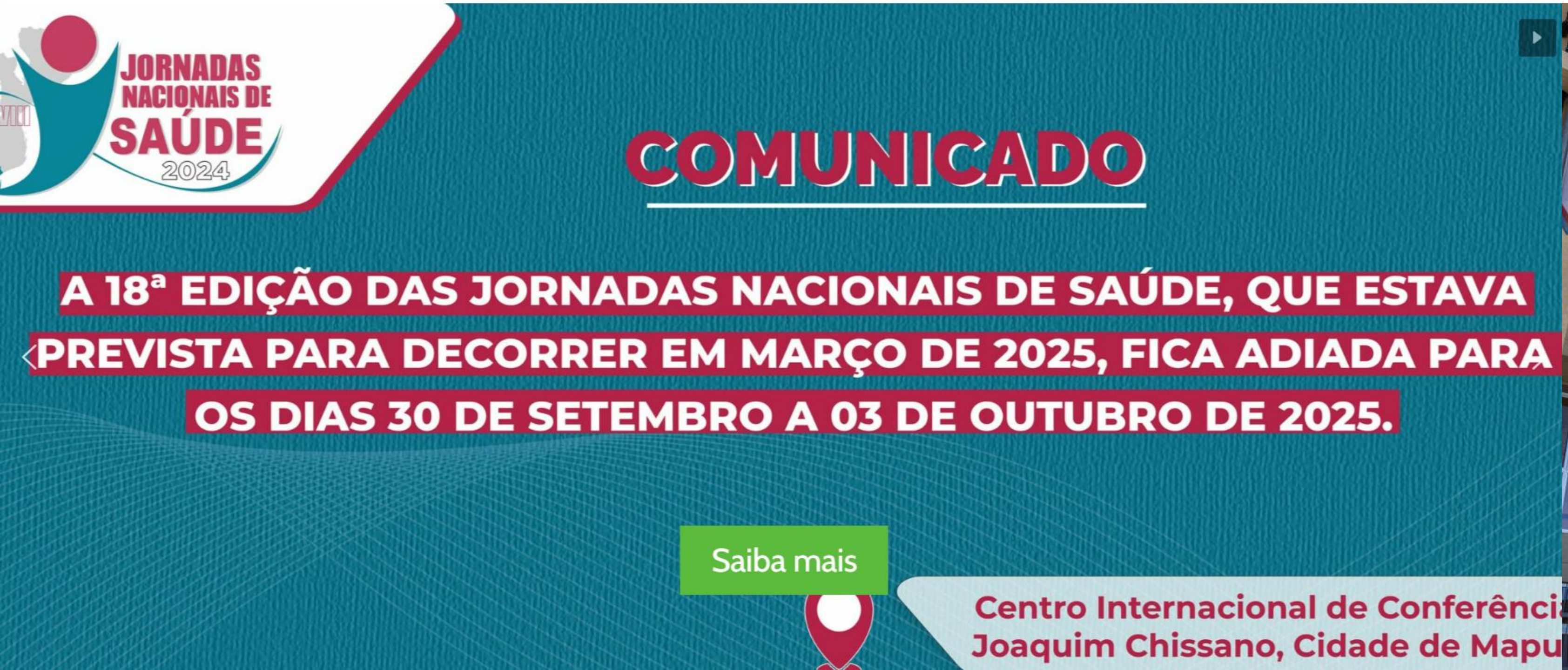
❑Finalize MTBC sequencing implementation framework for Mozambique.

❑Construction of the Bioinformatics Centre.

Concluding remarks for country NGS implementation:

- Building institutional capacity is a long but rewarding journey.
- Well-equipped sequencing laboratory with trained personnel is crucial.
- Strategic partnerships are essential for success.
- Understanding the country's epidemiology through scientific evidence is crucial.
- Engaging the NTP and key stakeholders throughout the process raise awareness and drive action.
- Adopting an integrated approach ensures long-term sustainability.

Parallel session: Advancements in Next Generation Sequencing for Tuberculosis Diagnosis and Management in Mozambique



JORNADAS NACIONAIS DE SAÚDE 2024

COMUNICADO

A 18ª EDIÇÃO DAS JORNADAS NACIONAIS DE SAÚDE, QUE ESTAVA PREVISTA PARA DECORRER EM MARÇO DE 2025, FICA ADIADA PARA OS DIAS 30 DE SETEMBRO A 03 DE OUTUBRO DE 2025.

Centro Internacional de Conferênci
Joaquim Chissano, Cidade de Mapu

INSTITUTO NACIONAL DE SAÚDE
MOÇAMBIQUE

Acknowledgments

Niemann group RCB

INS Team

PNCT
PROGRAMA NACIONAL DE CONTROLO DA TUBERCULOSE

República de Moçambiqu
Ministério da Saúde

Forschungszentrum Borstel
Leibniz Lungenzentrum

SeqMDR*TB*_NET

GHP Programme

PANGenS

Global Health
EDCTP3

Co-funded by the European Union

INSTITUTO NACIONAL DE SAÚDE
MOÇAMBIQUE

Obrigada,
kanimambu,
Thank you!

50 Anos de Independência
Consolidando a Unidade Nacional, a Paz e o Desenvolvimento Sustentável