Using a confidence graded mutation list to predict DR-TB phenotypes

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Webinar Series 2019 – Next-generation sequencing for drug-resistant TB
Webinar 2: Interpreting and reporting mutations
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Standardized regimens:
- efficacy in patients with drug-sensitive TB
- lower efficacy in patients with drug-resistant strains
- role of INH in management of TB cases

From a recent meta-analysis on 9,153 MDR patients:
- analysis restricted to cohorts of patients in whom DST was routinely performed;
- caution is warranted given the well-known limitations of DST for many of the drugs used;

MDR-TB treatment success and patients survival may be associated with the use of selected drugs, the use of a greater number of effective drugs, and longer treatment

Technical challenges in traditional DST

Standardization of procedures is still difficult to set up

The correct performance of DST requires the understanding of several steps such as:

- Origin of resistance and interpretative criteria
- Dosage and stability of the incorporated compounds
- Anti-mycobacterial activity of the incorporate drug
- Interpretation of results and data reporting
Phenotypic vs genotypic DST

• Molecular methods are less operator-dependent than conventional DST

• Sources of errors, such as sample mix up or clerical errors may occur with both molecular and microbiological methods, but factors like the critical inoculum size require experienced technicians to reduce variability for microbiological DST methods

• In general, molecular approaches have objective and measurable parameters that can be used for QC
Error rates varied substantially between drugs

- PZA was consistently the most problematic drug, followed by AMK, EMB and STR
Genotypic vs phenotypic DST

...not all the mutations affecting DR-associated genes are equal...

- Occurrence of discrepancies between DST performed on MGIT system and “no wild-type” molecular assays
- Occurrence of discrepancies between DST performed on liquid vs solid systems and molecular assays
- Not every genotypic modification of a DR-associated gene affects phenotypic resistance equally (MIC values, silent mutations...)
- Not every genotypic modification of a DR-associated gene affects phenotypic resistance equally to different drugs of the same class
- Borderline/low-level resistance has been strongly associated with treatment failure

The identification of the nature of the mutations is needed for accurate diagnosis of resistance (e.g. mutations not associated with DR)

The identification of the nature of mutations is needed for the interpretation of the resistance level (e.g. mutations causing low-level R, combination of mutations)

The type of mutation will be relevant for patient management
DR TB diagnosis by NGS technologies

In-house

Outsourcing

Case 1

Case 2

Case 3

Case 4
The Need

“A lack of user-friendly data analysis and interpretation tools has been frequently cited as a major barrier to routine use of WGS techniques.”

“Our experience with WGS in the clinical context of drug-resistant TB highlights the urgent need for an internationally recognized database for standardized genotype-phenotype correlation interpretation that can be added to and interrogated by clinicians and microbiologists who will be increasingly accessing WGS platforms at the local hospital level.”

Existing databases for TB

Table 1
Relevant SNP databases for MTBC. Databases containing SNP data of MTBC, and examples of SNP-databases of human variation, which could serve as examples for a future MTBC SNP-database.

<table>
<thead>
<tr>
<th>Name</th>
<th>Species</th>
<th># of SNPs (# of genomes)</th>
<th>Features</th>
<th>URLRef</th>
</tr>
</thead>
<tbody>
<tr>
<td>MTBC SNP-databases</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>dbSNP</td>
<td><em>M. tuberculosis</em></td>
<td>40,303 MTBC (3827 MTBC samples in SRA)</td>
<td>NCBI curated relational SNP-database for all organisms, MTBC SNPs are not annotated</td>
<td><a href="http://www.ncbi.nlm.nih.gov/projects/SNP93">http://www.ncbi.nlm.nih.gov/projects/SNP93</a></td>
</tr>
<tr>
<td>TBDB</td>
<td><em>M. tuberculosis</em></td>
<td>23,795 (25 MTBC)</td>
<td>Relational database with various MTBC data sets such as expression, diversity, proteins, ChiPseq, publications etc. SNPs are well annotated and interlinked with other tables, but not updated.</td>
<td><a href="http://www.tbdb.org/90">http://www.tbdb.org/90</a></td>
</tr>
<tr>
<td>PATRIC</td>
<td><em>M. tuberculosis</em></td>
<td>0 (75 MTBC)</td>
<td>Extensive relational database for various bacterial pathogens linking genomic data with NIH disease, epitopes etc. SNP database in preparation.</td>
<td><a href="http://www.patricbrc.org/portal/portal/patric/Home58">http://www.patricbrc.org/portal/portal/patric/Home58</a></td>
</tr>
<tr>
<td>MGDD</td>
<td><em>M. tuberculosis</em></td>
<td>n.a. (6 MTBC)</td>
<td>One-by-one comparison of 6 MTBC strains; not updated since 2008</td>
<td><a href="http://mirna.jnu.ac.in/mgdd91">http://mirna.jnu.ac.in/mgdd91</a></td>
</tr>
<tr>
<td>MTCID</td>
<td><em>M. tuberculosis</em></td>
<td>n.a.</td>
<td>List of mainly drug resistance-conferring mutations</td>
<td><a href="http://ccbb.jnu.ac.in/Tb92">http://ccbb.jnu.ac.in/Tb92</a></td>
</tr>
<tr>
<td>TDBReaMDB</td>
<td><em>M. tuberculosis</em></td>
<td>1447 (0)</td>
<td>Drug resistance-conferring mutations</td>
<td><a href="http://www.tbdreamdb.com65">http://www.tbdreamdb.com65</a></td>
</tr>
</tbody>
</table>

• Lack of a comprehensive, well-curated and user-friendly DB for SNP data
• Available DBs for “omics” data lack significant clinical metadata
Interpreting NGS data for DR TB


...Different approaches based on individual team expertise
Lack of studies providing high confidence genetic markers of R

Systematic review of allelic exchange experiments aimed at identifying mutations that confer drug resistance in MTB:

Studies considered: 922

Epidemiological studies $\rightarrow$ association of specific mutations with resistant phenotype $\quad 14\%$

Direct genetic manipulation $\rightarrow$ causality between the mutation and the resistant phenotype $\quad 2\%$

Clinical evidence $\rightarrow$ association of specific mutations with poor clinical outcome $\quad <1\%$ (?)
A centralized DB for TB

**Needs**

- Understand SNP-DST relationships (including MIC and cross-resistance)
- Understand SNP-clinical outcome relationships
- Understand frequency of mutations
- Improve statistical power for rare mutations
- Understand the geographical distribution of mutations
- Standardize interpretation of SNPs
- Guide the development of new molecular diagnostics and new policies
- Understand additional relationships (compensatory mutations, lineages...)

**Hurdles**

- Data sets scattered across a high number of studies
- Different studies use different methods (liquid/solid, CCs, SNP detection approach...)
- Few studies correlating SNP to MICs
- Few studies correlating SNP with clinical outcome
- Few studies on functional genetics
- Need to include ALL genes for ALL drugs
Relational Sequencing TB Data Platform (ReSeqTB)

A global partnership of academic institutions, public health agencies, and non-governmental organizations

- **To develop**
  - “One stop” source of curated genotypic, phenotypic and related metadata from MTB strains
  - A methodology for the categorizing of mutations to inform assay developers and provide the ability to objectively analysis in silico existing and planned assays

- **To ensure**
  - Patient data privacy
  - Standardized and validated WGS analysis pipeline for determining genetic variants and lineage
  - Quality and security of information
  - High quality of data governance (e.g. data ownership by the contributor)
ReSeqTB – Overview

Contributed Data

- WGS
  - Unified Pipeline
  - Genotypic data
  - Phenotypic data
  - Clinical trial data
  - DR study data
  - Surveillance data
  - Original WGS SNP reports
  - New SNP reports from Unified Pipeline

- SNP

RDST Consortium

- Expert Panel Review

CURATED AND AGGREGATED DATA
- Genotypic Data
- Phenotypic Data
- Clinical Data
- Drug Resistance Data
- SNP Reports

ANALYTICS TOOLS
- “R” Statistical Analysis
- Misc. Integrated Analysis

RECOGNITION
- Individual Recognition
- Institutional Recognition
- Global Impact

Resistance Associated Mutations: the grading system

• Resistance associated mutations validation
  – Binning; 5 bins based upon the associated data for a specific mutation
  – Bins are defined using a statistical approach and then revised according to the expert panels confidence of association with resistance of a mutation

• Grading criteria for confidence binning of mutations
  – Observed frequency of a mutation in phenotypically resistant or susceptible strains
  – Likelihood ratio (LR) and odd ratio (OR) were used for objectively evaluating whether or not mutations are positively associated with phenotypic resistance.
  – P-values and 95% confidence intervals associated with LR and OR have been also considered
**Statistical Significance**

**Likelihood ratio (LR)** represents “the probability that an isolate harbouring a given mutation is phenotypically resistant divided by the probability of an isolate with the mutation testing as susceptible”.

\[
LR^+ = \frac{\text{sensitivity}}{1 - \text{specificity}}
\]

\[
LR^+ = \frac{\Pr(T+ | D+)}{\Pr(T+ | D-)}
\]

**Odds ratio (OR)** represents the ratio of the odds of an event (in our case “the presence of a mutation”) occurring in one group (in our case “phenotypically drug resistant isolates”) to the odds of it occurring in another group (in our case “phenotypically drug susceptible isolates”). It is a measure of effect size, describing the strength of association or non-independence between two binary data values.

\[
OR = \frac{\frac{\text{sensitivity} \times \text{specificity}}{(1 - \text{sensitivity}) \times (1 - \text{specificity})}}{\frac{p_1/(1 - p_1)}{p_2/(1 - p_2)}} = \frac{p_1/q_1}{p_2/q_2} = \frac{p_1q_2}{p_2q_1}
\]
Why LR and OR?

- Reliance only on sensitivity and specificity frequently leads to exaggeration of the benefits of a test or strength of an association.

- LR and ORs represent best independent measures for assigning confidence to the association between a SNP and its associated phenotypic resistance.

- LR and ORs are universal measures of association in diagnostics and do not rely on the prevalence of the mutation in question.

- ORs are commonly used in the field of genomics – more familiar among researchers.

- LR and ORs are traditionally used to guide clinical decision making – more familiar among clinicians.
The interpretation of LR could be summarized as follow:

- LR=1 percentage of resistant and susceptible isolates with the mutation are the same
- LR>1 increases in the probability of a real association between the presence of a mutation and phenotypic resistance
- LR<1 decreases in the probability of a real association between the presence of a mutation and phenotypic resistance

The interpretation of OR could be summarized as follow:

- OR=1 presence of a mutation does not affect odds of phenotypic resistance
- OR>1 presence of a mutation associated with higher odds of phenotypic resistance
- OR<1 presence of a mutation associated with lower odds of phenotypic resistance
Thresholds

Using this rationale, thresholds used most commonly in evidence-based medicine have been adapted to grade *M. tuberculosis* mutations:

<table>
<thead>
<tr>
<th>Evidence-based medicine accepted thresholds</th>
<th>Adapted thresholds for the ReSeqTB</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>LR+</strong></td>
<td><strong>Interpretation</strong></td>
</tr>
<tr>
<td>≥ 10</td>
<td>Large and often conclusive increase in the likelihood of disease</td>
</tr>
<tr>
<td>5 ≤ ... &lt; 10</td>
<td>Moderate increase in the likelihood of disease</td>
</tr>
<tr>
<td>2 ≤ ... &lt; 5</td>
<td>Small increase in the likelihood of disease</td>
</tr>
<tr>
<td>1</td>
<td>No change in the likelihood of disease</td>
</tr>
<tr>
<td>0.5 &lt; ... &lt; 1</td>
<td>Minimal decrease in the likelihood of disease</td>
</tr>
<tr>
<td>0.2 &lt; ... ≤ 0.5</td>
<td>Small decrease in the likelihood of disease</td>
</tr>
<tr>
<td>0.1 &lt; ... ≤ 0.2</td>
<td>Moderate decrease in the likelihood of disease</td>
</tr>
<tr>
<td>≤ 0.1</td>
<td>Large and often conclusive decrease in the likelihood of disease</td>
</tr>
</tbody>
</table>

*Indeter* – no statistically significant threshold reached; additional data required.
### List of graded mutations

**Table 3: List of confidence-graded mutations associated with phenotypic drug resistance as determined by best confidence values**

<table>
<thead>
<tr>
<th>Drug (phenotypic testing)</th>
<th>Gene</th>
<th>High-confidence mutations</th>
<th>Moderate-confidence mutations</th>
<th>Minimal-confidence mutations</th>
<th>No association with resistance</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>First-line</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Isoniazid (H)</td>
<td>inhA-mabA</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>katG</td>
<td></td>
<td>c-15t</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>ftrA</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>mabA</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Second-line (group A)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ofloxacin (DXF)/Lefloxacin (LFX)</td>
<td>gyrB</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>E459K, A504V</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Second-line (group B)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Amikacin (AM)</td>
<td>rrs</td>
<td>a1401g, g1484t</td>
<td>c-14t, g-10a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kanamycin (KM)</td>
<td>eis</td>
<td>a514c, a1401g, c1402t, g1484t</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>rrs + eis</td>
<td></td>
<td>rs c517t + eis g-37t</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Capreomycin (CM)</td>
<td>rrs</td>
<td>a1401g, c1402t, g1484t</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Streptomycin (S)</td>
<td>eis</td>
<td>N236K, pooled frameshifts and premature stop codons</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>rpsL</td>
<td>K43R, K43T, K88Q, K88R, T40I</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>rps</td>
<td>a1401g, a514c, c462t, c513t, c517t</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Second-line (group C)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ethionamide and prothionamide (ETO/PTO)</td>
<td>inhA</td>
<td>c-15t + 1194T, c-15t + 549A</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>ethA</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Second-line (group D)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

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### Grading system – Identifying phenotypic testing problems

<table>
<thead>
<tr>
<th>Drug</th>
<th>Gene</th>
<th>Mutation</th>
<th>Found in R (TP)</th>
<th>Found in S (FP)</th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>Accuracy</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>% 95% Cis</td>
<td>% 95% Cis</td>
<td>% 95% Cis</td>
<td>% 95% Cis</td>
<td>% 95% Cis</td>
</tr>
<tr>
<td>RIF</td>
<td>rpoB</td>
<td>S531L</td>
<td>4388</td>
<td>20</td>
<td>54.25</td>
<td>99.59</td>
<td>71.46</td>
</tr>
<tr>
<td></td>
<td>katG</td>
<td>S315T</td>
<td>1548</td>
<td>1</td>
<td>63.39</td>
<td>99.59</td>
<td>66.70</td>
</tr>
<tr>
<td>INH</td>
<td>katG</td>
<td>S315T</td>
<td>397</td>
<td>4</td>
<td>10.87</td>
<td>99.89</td>
<td>55.57</td>
</tr>
<tr>
<td>OFX/LEV</td>
<td>gyrA</td>
<td>A90V</td>
<td>600</td>
<td>5</td>
<td>74.16</td>
<td>99.61</td>
<td>89.83</td>
</tr>
<tr>
<td>AMK</td>
<td>rrs</td>
<td>a1401g</td>
<td>265</td>
<td>1</td>
<td>47.49</td>
<td>99.84</td>
<td>75.76</td>
</tr>
<tr>
<td>KAN</td>
<td>rrs</td>
<td>a1401g</td>
<td>358</td>
<td>6</td>
<td>62.91</td>
<td>98.06</td>
<td>75.31</td>
</tr>
<tr>
<td>STR</td>
<td>rpsL</td>
<td>K43R</td>
<td>...</td>
<td>...</td>
<td>...</td>
<td>...</td>
<td>...</td>
</tr>
</tbody>
</table>

Wrong phenotypic DST?
Mistakes in reporting DST results?
### Grading system – Discrepancies among different phenotypic testing methods

<table>
<thead>
<tr>
<th>Drug</th>
<th>Gene</th>
<th>Mutation</th>
<th>Phenotypic testing</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Liquid</td>
</tr>
<tr>
<td>OFX/LEV</td>
<td>gyrA</td>
<td>D94A</td>
<td>High</td>
</tr>
<tr>
<td>OFX/LEV</td>
<td>gyrA</td>
<td>D94N</td>
<td>High</td>
</tr>
<tr>
<td>RIF</td>
<td>rpoB</td>
<td>Q513K</td>
<td>Moderate</td>
</tr>
<tr>
<td>RIF</td>
<td>rpoB</td>
<td>H526C</td>
<td>Moderate</td>
</tr>
<tr>
<td>RIF</td>
<td>rpoB</td>
<td>D516Y</td>
<td>indeter</td>
</tr>
<tr>
<td>RIF</td>
<td>rpoB</td>
<td>L533P</td>
<td>indeter</td>
</tr>
<tr>
<td>RIF</td>
<td>rpoB</td>
<td>L511P</td>
<td>NOT associated</td>
</tr>
<tr>
<td>MOX</td>
<td>gyrA</td>
<td>A90V</td>
<td>NOT associated</td>
</tr>
<tr>
<td>PZA</td>
<td>pncA</td>
<td>K48T</td>
<td>Moderate</td>
</tr>
</tbody>
</table>

Cases misclassified by liquid phenotypic testing

---

* PZAs assay
** MOX liquid CC: 2 mg/L
<table>
<thead>
<tr>
<th>Drug</th>
<th>NOT associated</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>inhA t-80g, g-47c, t-8g, T253A</td>
</tr>
<tr>
<td></td>
<td>mabA T4I</td>
</tr>
<tr>
<td></td>
<td>mshA N111S + A298A</td>
</tr>
<tr>
<td>MOX</td>
<td>gyrA E21Q, S95T, G668D, V712L</td>
</tr>
<tr>
<td>OFX/LEV</td>
<td>gyrA E21Q, T80A, S95T, G668D, V712L</td>
</tr>
<tr>
<td>KAN</td>
<td>rrs a1338c</td>
</tr>
<tr>
<td>STR</td>
<td>gidB V110G, E92D, L16R</td>
</tr>
<tr>
<td>ETH/PTH</td>
<td>ethA K448E</td>
</tr>
</tbody>
</table>
Advantages of the grading system

• Sensitivity

• The use of graded mutations allows for improvement in specificity with a relatively small decrease in sensitivity

• Reduction of Very Major Errors (DR reported as DS)

A standardised method for interpreting the association between mutations and phenotypic drug resistance in Mycobacterium tuberculosis


Technical Report on critical concentrations for drug susceptibility testing of medicines used in the treatment of drug-resistant tuberculosis

Genetic sequencing for surveillance of drug resistance in tuberculosis in highly endemic countries: a multi-country population-based surveillance study

World Health Organization 2018.

Emerging Bacterial Pathogens Unit, OSPEDALE SAN RAFFAELE

ReSeqTB: platform status

- Represents data sets from 39 countries
- 13 data sets (being mapped/curated)
- 4943 isolates sequenced
- Over 2700 isolates shipping
- Additional 1500 isolates in discussion
- >90 genetic loci considered for DR

**Legend**

- Data in platform or being curated
- Data/samples transferred or DCA completed.
- Contribution in discussion

**Courtesy of M Schito, CPTR**
ReSeqTB: public site

Address:
- Deidentification of patients
- Disclosure and publication rights
- Comply with IRB rules
- Protect IP rights of contributors
- Liability and indemnification

https://www.reseqtb.org/
ReSeqTB visual browser
ReSeqTB visual browser

ReSeqTB Drug Resistance Mutations List v1.0 – User Guide

Drug Resistance Reports

These statistics represent the relationship between ReSeqTB isolates’ laboratory testing results and variants in a specific list of genomic loci defined by a panel of collaborating scientists.

The predictive value of microbiological testing varies by testing methods, laboratory practices, and concentrations of antimicrobial agents in question.

Please refer to the Functional Specifications document for the criteria used to include and exclude phenotypic results.

Note that first-line drugs are routinely tested, but other antimicrobial agents will lack the sampling intensity to produce reliable statistics.

Because the included isolates represent valued contributions from willing scientists around the globe, and not from randomized sampling, there will be some inherent bias in the lineages and geographic regions represented.

For this reason, some variants will not have been identified by our pipeline often enough to produce expected statistics.

Continued contributions are adding clarity to many of these underrepresented drugs and variants.

Loci of Interest - v5

RSTB Functional Specifications

Resistance Report - Feb 2018
ReSeqTB visual browser

Courtesy of M Schito, CPTR
ReSeqTB visual browser

Courtesy of M Schito, CPTR
ReSeqTB visual browser

Public Data

The files below are a compilation of all publicly available data extracted from the ReSeqTB Platforms as of 1/24/2019.
Click the icon to start the download process.

- PFF Table
- MSF Table
- LowQual Table
- NoCov Table

Courtesy of M Schito, CPTR
ReSeqTB: a solution for global TB surveillance

Courtesy of M Schito, CPTR
Summary

• A total of 304 highly specific confidence mutations have been determined (286 associated with DR)
• Overall sensitivity between 80 and 90% for R, H, MFX/LEV/OFX, and AM

• Advantages of the grading system:
  – Identification of high-confidence markers of resistance
  – Identification of likely wrong phenotypic testing results
  – Identification of mutations with different confidence levels according to the different reference phenotypic testing method considered (and/or associated with MIC values close to the critical concentration/Clinical breakpoint)
  – Identification of polymorphisms NOT associated with phenotypic resistance
  – suitable for automated classification of mutations
  – LR is a universal measure of association which is not affected by local prevalence
  – unlike sensitivities and specificities, LRs do not lead to an exaggeration of the benefits of a test or strength of an association

• The validation process is driven by statistics based on frequency of mutations, this can be improved by integrating
  – Homoplasy data
  – MIC data
  – functional genetic studies

• The validation process will need to be evaluated periodically and refined as tools and our understanding increase
Thank you!

Webinar Series 2019
Next-generation sequencing for drug-resistant TB

World Health Organization
FIND
StopTB Partnership
ACCESS Campaign

New Diagnostics Working Group
Proficiency testing


Disputed mutations, mutations vs MICs/critical concentrations, mutations vs clinical outcomes...

Additional references

Disputed mutations, mutations vs MICs/critical concentrations, mutations vs clinical outcomes...


Additional references

Disputed mutations, mutations vs MICs/critical concentrations, mutations vs clinical outcomes...


- Treating tuberculosis with high doses of anti-TB drugs: mechanisms and outcomes.


- Clinical Outcomes of Patients With Drug-Resistant Tuberculous Meningitis Treated With an Intensified Antituberculosis Regimen.


Additional references

Application of WGS in TB diagnosis

- Mycobacterial DNA extraction for whole-genome sequencing from early positive liquid (MGIT) cultures.
- Report from the EUCAST Subcommittee on the Role of Whole Genome Sequencing (WGS) in Antimicrobial Susceptibility Testing of Bacteria. EUCAST WGS Subcommittee Report v1.0 April 2016