

# MYCOBACTERIUM TUBERCULOSIS SEQUENCING REPORT

## Sample Details

Patient Name	JOHN DOE	Patient ID	12345678910
Birth Date	2000-JAN-01	Location	SOMEPLACE
Sample Type	SPUTUM	Sample Collection Date	2016-DEC-25
Sample Source	PULMONARY	Sequenced From	CULTURED ISOLATE (LJ)
Sample ID	A12345678	Sample Received Date/Time	2017-JAN-02, 12:22
Laboratory Technician	TECHNICIAN NAME	Report Date/Time	2017-JAN-05, 11:45
Requested By	REQUESTER NAME	Requester Contact	REQUESTER@EMAIL.COM

## Assay Details

Sequencer	ILLUMINA HISEQ 2500	Method	WHOLE GENOME SEQUENCING
Pipeline	RESEQTBV.3.2C ( <a href="https://platform.reseqtb.org">https://platform.reseqtb.org</a> )	Reference	H37RV (NC_000962.3)

## Final Result

The sample was positive for *Mycobacterium tuberculosis*. It is resistant to isoniazid, rifampin, capreomycin, and kanamycin. It has intermediate resistance to ofloxacin and moxifloxacin. Expert consultation advised for pyrazinamide.

## Lineage

*Mycobacterium tuberculosis*, lineage 2.2.1 (East-Asian Beijing).

## Drug Susceptibility

Resistance is reported when a resistance-conferring mutation is detected in: *embB*, *gidB*, *gyrA*, *gyrB*, *inhA*, *katG*, *mshA*, *pncA*, *rpoB*, *rpsL*, *rrs*, or *tlyA*.<sup>1</sup> **No mutation detected does not exclude the possibility of resistance.**

- No mutations detected
- Multi-drug resistance predicted
- Extensive drug resistance predicted**

## Resistance Predictions for First Line TB Drugs

Interpretation	Drug	Gene Target (Codon Change, Amino Acid Change, Allele %)	Confidence in Resistance Association	Comments
Resistant	Isoniazid	<i>katG</i> (G944C, Ser315Thr, 100%)	High	
		<i>inhA</i>		No mutation detected
		<i>mshA</i>		No mutation detected
	Rifampin	<i>rpoB</i> (C1349T, Ser450Leu, 100%)	High	Rifabutin resistance likely
Susceptible	Ethambutol	<i>embB</i>		No mutation detected
Expert Consultation Advised	Pyrazinamide	<i>pncA</i> (T416C, Val139Ala, 98%)	Minimal	Mutation known to disrupt enzymatic activity and functional genetics in vitro. Insufficient data to determine clinical impact of this mutation.

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## Resistance Predictions for Second Line TB Drugs

Interpretation	Drug	Gene Target (Codon Change, Amino Acid Change, Allele %)	Confidence in Resistance Association	Comments
Resistant	Capreomycin	<i>rrs</i> (C1402T, NA, 97%)	High	100% of isolates with this mutation were capreomycin resistant in the ReSeqTB database.
		<i>tlyA</i>		No mutation detected
Intermediate	Ofloxacin	<i>gyrA</i> (C269T, Ala90Val, 14%)	High	Ofloxacin and levofloxacin resistance predicted. At least low-level resistance predicted ( $\geq 0.5 \mu\text{g/mL}$ in MGIT Culture)
		<i>gyrB</i>		No mutation detected
	Moxifloxacin	<i>gyrA</i> (C269T, Ala90Val, 14%)	High	At least low-level resistance predicted ( $\geq 0.5 \mu\text{g/mL}$ in MGIT Culture)
		<i>gyrB</i>		No mutation detected
Susceptible	Amikacin	<i>rrs</i>		The C1402T mutation is associated with resistance to other injectable drugs including kanamycin and capreomycin. This mutation has not been associated with resistance to amikacin in the ReSeqTB database.
	Ethionamide	<i>inhA</i>		No mutation detected
		<i>gidB</i>		No mutation detected
		<i>rpsL</i>		No mutation detected
Streptomycin	<i>rrs</i>		The C1402T mutation is associated with resistance to other injectable drugs including kanamycin and capreomycin. This mutation has not been associated with resistance to streptomycin in the ReSeqTB database.	

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## Resistance Predictions for Other TB Drugs

Interpretation	Drug	Gene Target (Codon Change, Amino Acid Change, Allele %)	Confidence in Resistance Association	Comments	
Insufficient Data for Resistance Prediction	Bedaquiline	<i>atpE</i>		No mutation detected	
		<i>mmpR</i> (G198Insertion, Frameshift, 98%)	Insufficient Data	Insertion of duplicate G: CGGGGGGG Gene target has shown <i>in vitro</i> resistance to bedaquiline and clofazimine in the literature.	
	Clofazimine	<i>mmpR</i> (G198Insertion, Frameshift, 98%)	Insufficient Data	Insertion of duplicate G: CGGGGGGG Gene target has shown <i>in vitro</i> resistance to bedaquiline and clofazimine in the literature.	
	Delamanid		<i>ddn</i>		No mutation detected
			<i>fbiA</i>		No mutation detected
			<i>fbiB</i>		No mutation detected
			<i>fbiC</i>		No mutation detected
			<i>fgd1</i>		No mutation detected
	Linezolid		<i>rplC</i> (T460C, Cys154Arg, 97%)	Insufficient Data	Gene target has shown <i>in vitro</i> resistance to linezolid in the literature.
			<i>rrl</i>		No mutation detected

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## Depth and Coverage Details

Locus of Interest <sup>1</sup>	# Reads Mapped (Depth of Coverage)	Proportion Covered (Coverage Width %)	Mutation Frequency (% Alternate Allele)
<i>atpE</i>	79	100	-
<i>ddn</i>	97	99.9	-
<i>embB</i>	88	98.8	-
<i>fbiA</i>	87	100	-
<i>fbiB</i>	91	100	-
<i>fbiC</i>	75	100	-
<i>fgd1</i>	88	100	-
<i>gyrA</i>	73	100	Ala90Val (13.6%*)
<i>gyrB</i>	76	100	-
<i>inhA</i>	91	100	-
<i>katG</i>	90	99.9	Ser315Thr (100%)
<i>mmpR</i>	62	100	G193Insertion (98.4%)
<i>pncA</i>	62	100	Val139Ala (98.4%)
<i>rplC</i>	78	99.9	Cys154Arg (97.4%)
<i>rpoB</i>	83	100	Ser450Leu (100%)
<i>rrl</i>	82	100	-
<i>rrs</i>	59	99.9	C1402I (96.6%)
<i>tlyA</i>	58	98.5	-

\*Hetero-resistance is defined in this report as a proportion of reads for a given allele between 10% and 90%.

<sup>1</sup>Loci of interest derived from ReSeqTB Data Platform and from Miotto P, et al. Eur Respir J. 2017 PMID: 29284687

## Disclaimer

Low frequency hetero-resistance below the limit of detection by sequencing may affect typing results. The interpretation provided is based on the current understanding of genotype-phenotype relationships. All results reference the *M. tuberculosis* mutation numbering system which differs from the *E. coli* numbering system. For *rpoB* add 81 to amino acid position to calculate the equivalent *E. coli* position. For *gyrB* subtract 7 amino acid positions to calculate the equivalent *E. coli* position.

Confidence is reported based on likelihood ratio value relating the mutation to resistance in the ReSeqTB database as follows:

**High** – LR ≥ 10, high confidence that the mutation confers or is associated with resistance

**Moderate** – LR ≥ 5 and <10, additional data desirable to improve evidence that the mutation confers or is associated with resistance

**Low** – LR ≥ 1 and < 5, inconclusive evidence that the mutation confers or is associated with resistance. Additional data required.

**Insufficient Data** – LR < 1, No evidence of association between mutation and resistance

## Authorized By

Name	AUTHORIZER NAME	Position	LAB SUPERVISOR
Signature		Date	2017-JAN-05
Reporting Laboratory	LAB NAME	LAB ADDRESS	LAB PHONE NUMBER