

Supplementary Material to “Gyrase Mutations are Associated with Variable Levels of Fluoroquinolone Resistance in *Mycobacterium tuberculosis*”

Supplementary Table 1: **Sequence and coordinates of the molecular inversion probes used for target sequence capture.**

Some probes are duplicates except for one SNP in one of the arms.

BRIDGE = AGATCGGAAGAGCGTCTGTAGGGAAAGCTGAGCAAATGTTATCGAGGTC

# Probe name	Start	End Sequence
gyrB-gyrA_W_1_1	4973	5112 CAGGAAGAAAGATGTCCGACGCACG-(BRIDGE)-GGTCCGCGCGACACCTACG
gyrB-gyrA_W_2_2	5113	5252 TTCGGATCTGTGGCTGCCAGAG-(BRIDGE)-GCGCGTTAGATGGGTAAAAACGAGG
gyrB-gyrA_W_3_1	5253	5392 ACCATCTCATTGGGAGGTGGTCGAC-(BRIDGE)-AAAAGAAGGCCAAGACGAATACGGCG
gyrB-gyrA_W_4_2	5393	5532 CGGCATACCGACCGTCGACGT-(BRIDGE)-AACGCGGTGACGAGGCGAT
gyrB-gyrA_W_4_2a	5393	5532 CGGCATACCGACCGTCGACGT-(BRIDGE)-AACGCGGTGACGAGGCGAT
gyrB-gyrA_W_5_1	5533	5672 CGAGATCAAGCGCGACGGTACG-(BRIDGE)-GGTGTGACACAACATACATGCCGGC
gyrB-gyrA_W_6_2	5673	5812 GGAATACGACTTCGAAACCGTCGCC-(BRIDGE)-AGTGGTCTCAGTTTATGAGAAGTCGGAACC
gyrB-gyrA_W_7_1	5813	5952 TCGGCAAGTGAACGCGCAGC-(BRIDGE)-CGCCGGCTGCAAGAGATGGC
gyrB-gyrA_W_8_2	5953	6092 TTCGCGCAAGGGCACCGGGC-(BRIDGE)-CGAATCCACTGCACCCACAAAAGT
gyrB-gyrA_W_9_1	6093	6232 GCTGACGTCGGTGGTGAACAAGTAC-(BRIDGE)-ACGAGGTGGAGATCGCGATGCA
gyrB-gyrA_W_10_2	6233	6372 CAGACCAAGACCAAGTTGGGCAACAC-(BRIDGE)-GCCAAGGACCGCAAGCTACTGAAG
gyrB-gyrA_W_10_2a	6233	6372 CAGACCAAGACCAAGTTGGGCAACAC-(BRIDGE)-GCCAAGGACCGCAAGCTACTGAAG
gyrB-gyrA_W_11_1	6373	6512 CGCGGCACGTAAGGCACGAG-(BRIDGE)-CGAGGTCAAATCGTTTGTGAGAAAGGTC
gyrB-gyrA_W_12_2	6513	6652 CGGTTCTGCAAAAAGCGGTGCG-(BRIDGE)-AGTTGGTGCAGCGTAAAGAGCG
gyrB-gyrA_W_13_1	6653	6792 CCGGATCCACGACGAGTTCTGA-(BRIDGE)-GATTCTGATTTCCAGGGGACTTCCGC
gyrB-gyrA_W_14_2	6793	6932 CGAGAACGGGATGTGTTTTGGCAC-(BRIDGE)-TATCGGCAAGCTGCGTACCACA
gyrB-gyrA_W_15_1	6933	7072 GGAAGACGGCATTGAGCGTACAAG-(BRIDGE)-AACCGCGCTGTACAAACTCAAGTG
gyrB-gyrA_W_16_2	7073	7212 ATGGGCCAGGACGTCGACGC-(BRIDGE)-GGTCTAGTGAAATGGACGCTAAGGAGTTG
gyrB-gyrA_W_17_1	7213	7352 TCGCTCGACCGGATCGAACCG-(BRIDGE)-GCGGCGCAGTTTATCACCCG
gyrB-gyrA_W_17_1a	7213	7352 TCGCTCGACCGGATCGAACCG-(BRIDGE)-GCGGCGCAGTTTATCACCCG
gyrB-gyrA_W_18_2	7353	7492 TGCTCTATGCAATGTTGATTCGGCTT-(BRIDGE)-GTTGACATCGAGCAGGAGATGCAGC
gyrB-gyrA_W_18_2a	7353	7492 TGCTCTATGCAATGTTGATTCGGCTT-(BRIDGE)-GTTGACATCGAGCAGGAGATGCAGC
gyrB-gyrA_W_19_1	7493	7632 GCTGCGCTACCGCTGGTGG-(BRIDGE)-CCGCCGACCGCAGCCAGC
gyrB-gyrA_W_20_2	7633	7772 AGGAGACAGTCGATTTTCCTAAGTACGAC-(BRIDGE)-ACGCCAGGGCAACTTCGGC
gyrB-gyrA_W_21_1	7773	7912 GCCGACGCGTGTCTGGGC-(BRIDGE)-GGCCGGTGAAGAGCCGAC
gyrB-gyrA_W_22_2	7913	8052 CAAAAGTGGCCGCGCTCCA-(BRIDGE)-GCTGAGAATCAGCAGCCGAC
gyrB-gyrA_W_22_2a	7913	8052 CAAAAGTGGCCGCGCTCCA-(BRIDGE)-GCTGAGAATCAGCAGCCGAC
gyrB-gyrA_W_23_1	8053	8192 TCCGAGACGGCAAGCTGGCC-(BRIDGE)-TTCGAATGCGCGAGTTGTTGAGG
gyrB-gyrA_W_23_1a	8053	8192 TCCGAGACGGCAAGCTGGCC-(BRIDGE)-TTCGAATGCGCGAGTTGTTGAGG
gyrB-gyrA_W_24_2	8193	8332 GCAGACAGCTTTGGCCCAA-(BRIDGE)-GGCATTCCAACATTGAGGACCACTAGC
gyrB-gyrA_W_24_2a	8193	8332 GCAGACAGCTTTGGCCCAA-(BRIDGE)-GGCATTCCAACATTGAGGACCACTAGC
gyrB-gyrA_W_25_1	8333	8472 GGCAAAAGCAGCGACGCCACATTC-(BRIDGE)-CATGCTAGCGATCTGACGCGGG
gyrB-gyrA_W_26_2	8473	8612 CCAGGCCAGGCAATCCTGGAC-(BRIDGE)-TGCAGCGCTGTTAAAGCG
gyrB-gyrA_W_27_1	8613	8752 ATCGTGCAGCAGCAACTCGC-(BRIDGE)-ATGAGTTGCGGCGCTGGC
gyrB-gyrA_W_28_2	8753	8892 GGGATACGCCAAGCGCACCAAGA-(BRIDGE)-CGAAATCGTGGACGGCACGGC
gyrB-gyrA_W_29_1	8893	9032 CTTACACCCAGGGACGGGTTTAT-(BRIDGE)-CCGATCTGTATCGCAGCCAGAAACG
gyrB-gyrA_W_29_1a	8893	9032 CTTACACCCAGGGACGGGTTTAT-(BRIDGE)-CCGATCTGTATCGCAGCCAGAAACG
gyrB-gyrA_W_30_2	9033	9172 ACCGACGCCCTACCTGGT-(BRIDGE)-CGGGCCAAGGCTACGACTTGC
gyrB-gyrA_W_31_1	9173	9312 GTGTTCCGCGCGCAGCAGC-(BRIDGE)-GCTGGCACTCGCAACGGGC
gyrB-gyrA_W_31_1a	9173	9312 GTGTTCCGCGCGCAGCAGC-(BRIDGE)-GCTGGCACTCGCAACGGGC
gyrB-gyrA_W_32_2	9313	9452 GCTGCTGCTGTAACGCTGCTG-(BRIDGE)-TGCTGCTGCTCGGCCAACG
gyrB-gyrA_W_33_1	9453	9592 CGGGCGGCGCAGGTTGGTTGG-(BRIDGE)-CGTGAAGGCACCTATCTGCTGGTGG
gyrB-gyrA_W_34_2	9593	9732 TGAATCTGGCGAGGGCGACA-(BRIDGE)-GGCGTTGATGTCGACGACGACA
gyrB-gyrA_W_35_1	9733	9872 CGTAGCGATCTGAGGAGGAATCGGG-(BRIDGE)-CACTGTTGGCCATCGCGCGC
gyrB-gyrA_W_36_2	9873	10012 CACATCGGGCAGCGACCGGG-(BRIDGE)-CAGTAGGCTCGGACGGCG
gyrB-gyrA_C_1_2	5023	5162 TCTCAGCCGGGACCGGAGC-(BRIDGE)-CGGGCCAGGGCCGATCTT
gyrB-gyrA_C_2_1	5163	5302 GGTGCGACACCACTGTACCGTAT-(BRIDGE)-TTGAGAATGGTATAGACGACGCGC
gyrB-gyrA_C_3_2	5303	5442 TTGCGGACGGCTCCAGCCC-(BRIDGE)-ACTACGTTCACTGTGGTTGCATAACCGG
gyrB-gyrA_C_4_1	5443	5582 GACACGCCATCTCAAGCAGC-(BRIDGE)-CATACGCTCGGAGTCGAACTTGC
gyrB-gyrA_C_5_2	5583	5722 CGCCGTGACAGCACCAGATATCG-(BRIDGE)-CCCTTGCTTGAAGCCAGGGG
gyrB-gyrA_C_6_1	5723	5862 CCCGCTCTTGGTTCGGCGC-(BRIDGE)-AGGTTGATGGTCAGCCCTTGTGA
gyrB-gyrA_C_7_2	5863	6002 CTTGGGTCACCCCTCTGTCGGTC-(BRIDGE)-CCGGATAGTGAAGGTTGCGGCTCT
gyrB-gyrA_C_8_1	6003	6142 GTTTCACGAAGTCCACAGGCCAC-(BRIDGE)-CACCGACTCCGAATACCCGGCG
gyrB-gyrA_C_9_2	6143	6282 TGTGATGGTGTGGCGAAGGTGTG-(BRIDGE)-TCACCGGTGAGGTTGGGTTCC

# Probe name	Start	End Sequence
gyrB-gyrA_C_10_1	6283	6422 GGCCAGGCCTTCCCGGATATCG-(BRIDGE)-AGTGGGTACAGCTGTTTCGTTACAGACC
gyrB-gyrA_C_11_2	6423	6562 GTCGGTGGGGTTGGCTTCAAACC-(BRIDGE)-CTTGCCGGGCAATCCACCGATG
gyrB-gyrA_C_12_1	6563	6702 CGTGGAACGGCAATCGGCCAG-(BRIDGE)-ACATTGATGATCTTGCCGCGCAGC
gyrB-gyrA_C_13_2	6703	6842 CGTTCGATGCGCGCTTCTCC-(BRIDGE)-CATCGCGTCGGCCATCAGC
gyrB-gyrA_C_14_1	6843	6982 CAGCGTGAAATATGTTGGCCGTCAG-(BRIDGE)-TGCGAATTCGGGTCACTGCG
gyrB-gyrA_C_14_1a	6843	6982 CAGCGTGAAATATGTTGGCCGTCAG-(BRIDGE)-TGCGAATTCGGGTCACTGCG
gyrB-gyrA_C_15_2	6983	7122 TCGCGCTCGCGGTGCGAGTA-(BRIDGE)-GGATCCATGGTGGTCTCCCAACTC
gyrB-gyrA_C_16_1	7123	7262 ACTTGACGCAACACACGAACCGAG-(BRIDGE)-CATCCAGGAACCGAATCCTTGGCG
gyrB-gyrA_C_17_2	7263	7402 CAATCGAACGAGGTTGCGTTAGA-(BRIDGE)-CTCATCGCATAGTCGATGTAGCTGCGC
gyrB-gyrA_C_18_1	7403	7542 GCGCGCGCCGACGATCAGC-(BRIDGE)-TGGTCTCGGCAACCGACCGG
gyrB-gyrA_C_19_2	7543	7682 CGTGC GG GTGGTAGTTGCCA-(BRIDGE)-CATCGCCGCGGTGGGTCACT
gyrB-gyrA_C_19_2a	7543	7682 CGTGC GG GTGGTAGTTGCCA-(BRIDGE)-CATCGCCGCGGTGGGTCACT
gyrB-gyrA_C_19_2b	7543	7682 CGTGC GG GTGGTAGTTGCCA-(BRIDGE)-CATCGTCGCGGTGGGTCACT
gyrB-gyrA_C_20_1	7683	7822 CAGCCGGCTTCGGGTACCT-(BRIDGE)-AGCAGTTGGGGAACCGGCT
gyrB-gyrA_C_21_2	7823	7962 TGCCGCTGACCCGTTGGCC-(BRIDGE)-CCATGACCGCGCCAGGGTC
gyrB-gyrA_C_22_1	7963	8102 GTCCGGGCTTTAACCCGCC-(BRIDGE)-GGTACGACCGCGGGAATCCTCT
gyrB-gyrA_C_23_2	8103	8242 GGCAACTCGGTGATCACCAGCGA-(BRIDGE)-ATGCGTAAACCGACCCGATCGCT
gyrB-gyrA_C_24_1	8243	8382 GCATCGCGCTTGATCTCGATGACG-(BRIDGE)-GCTGGTCCAGCCGACCGTG
gyrB-gyrA_C_25_2	8383	8522 AGTTGGTGGTCAACGTAATAGCGGATCA-(BRIDGE)-CAGTGAATGACCTCGTCCAGCG
gyrB-gyrA_C_26_1	8523	8662 ACGTCTCCGACGCCCGGAT-(BRIDGE)-TCGTGATGATGCGCTGGCG
gyrB-gyrA_C_27_2	8663	8802 CTCGGCCTCGATTTTGCCAGG-(BRIDGE)-CCGCGATGATCCGGGTACGC
gyrB-gyrA_C_28_1	8803	8942 CGTCGCTGACGCTCCGTCGG-(BRIDGE)-CGCACCTGACGCGCCTTGC
gyrB-gyrA_C_28_1a	8803	8942 CGTCGCTGACGCTCCGTCGG-(BRIDGE)-CGCACCTGACGCGCCTTGC
gyrB-gyrA_C_29_2	8943	9082 CGACGATGTCGCTCTGCTCAACCC-(BRIDGE)-TGCCCGCGCGCGTCCGGGA
gyrB-gyrA_C_30_1	9083	9222 GGCTAACAGTTGGCCACGTGC-(BRIDGE)-CGAAGTCGCTGAGCTTGGACTTTTTCAC
gyrB-gyrA_C_31_2	9223	9362 TCCGCCGAGCGATTGGAGT-(BRIDGE)-GTCGGTCCGCGAGAACCCTGATGG
gyrB-gyrA_C_32_1	9363	9502 CCCATTGGCCGACGCGCTC-(BRIDGE)-CGTTTCGATAGCCCCCTGACG
gyrB-gyrA_C_33_2	9503	9642 ACCGGGTATTCTCGATCGCGGTA-(BRIDGE)-CGCCGGAAGTGACGGCATAACG
gyrB-gyrA_C_34_1	9643	9782 CGCGGTGCGGATCAGCCAC-(BRIDGE)-GGCATTATCGTCCCACTTTCTTCGG
gyrB-gyrA_C_35_2	9783	9922 TCTGCGCGTTGGCGTCCAC-(BRIDGE)-TGCAGTACCCCGACTCTAACAC

Supplementary Table 2: Parameters used to call variants from MIP read data.

Program	Version	Parameters
megablast (used by MIPCleaner)	2.2.14	-p 90 -W 10 -e 1e-1 -X16 -JF -m8 -FF -v9000000 -b9000000
Bowtie	0.12.7	-q -k 2 -l 20 -m 5 --best --strata (with the additional filtering out of alignments with NM:i:x tags having x>5)
samtools	0.1.18	mpileup -u -R -B -q0 -Q0 -C0 -d1000000
bcftools	0.1.18	view -vg -p0.05
vcfutils.pl	0.1.18	varFilter -d 10 -a 5 -D1000000 -W10 -10 -20 -30 -40 -e0
Stampy	1.0.23	Default
Platypus	0.5.2	Default. Filtered variants with platypusfilter='badreads' or quality≤30 or purity <40%.

Supplementary Table 3: Isolate status at lineage defining SNPs listed in Coll *et. al.* (PMID 25176035) or Feuerriegel *et. al.* (PMID 24458512). Additionally the Euro-American X-type SNP Q139H\_embB was not present in any isolate.



















2493	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	2
2496	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
2497	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
2506	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
2507	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	2
2510	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
2519	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
2578	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
2592	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
SUM	5	68	87	45	27	4	0	7	26	0	0	0	0	0	0	0	0	0	0	0	0	

Supplementary Table 4: Isolates with more than one *gyr* mutation and their moxifloxacin and ofloxacin

MICs. \*These two mutations were exactly co-linear

ID	Mutation number	MXF MIC	OFX MIC	mutations
181	2	0.125	<1	<i>gyrB_I310M*</i> , <i>gyrB_S700T*</i>
1167	2	1	4	<i>gyrA_D94Y</i> , <i>gyrA_P102S</i>
1370	2	1	4	<i>gyrA_D94Y</i> , <i>gyrA_T267I</i>
1578	2	4	4	<i>gyrA_A90V</i> , <i>gyrB_V340L</i>
1914	2	4	6	<i>gyrA_D94G</i> , <i>gyrB_V340L</i>
2029	3	8	>10	<i>gyrA_D94Y</i> , <i>gyrB_I310M</i>
2044	2	>8	>10	<i>gyrA_D94G</i> , <i>gyrB_N538T</i>
2049	2	>8	>10	<i>gyrA_D94N</i> , <i>gyrB_N538T</i>
2600	3	1	<1	<i>gyrB_I310M*</i> , <i>gyrB_S700T*</i> , <i>gyrB_R485H</i>
2772	2	<0.125	<1	<i>gyrB_I310M*</i> , <i>gyrB_S700T*</i>
3139	2	<0.125	<1	<i>gyrB_I310M*</i> , <i>gyrB_S700T*</i>
150	2	4	2	<i>gyrA_S91P</i> , <i>gyrA_D94G</i>
272	2	0.25	<1	<i>gyrB_I310M*</i> , <i>gyrB_S700T*</i>
492	2	0.25	<1	<i>gyrB_V340L</i> , <i>gyrB_A453S</i>
949	3	1	2	<i>gyrA_D94A</i> , <i>gyrB_I310M*</i> , <i>gyrB_S700T*</i>
950	2	4	2	<i>gyrA_D94G</i> , <i>gyrB_V340L</i>
951	5	<0.125	2	<i>gyrA_A90V</i> , <i>gyrB_I310M*</i> , <i>gyrB_S700T*</i> , <i>gyrB_R485C</i> , <i>gyrB_E78K</i>
1279	2	1	4	<i>gyrA_A90V</i> , <i>gyrB_V340L</i>
1280	2	1	4	<i>gyrA_A90V</i> , <i>gyrB_V340L</i>
1281	2	1	4	<i>gyrA_D94A</i> , <i>gyrA_T267I</i>
1293	3	1	4	<i>gyrA_A90V</i> , <i>gyrB_I310M*</i> , <i>gyrB_S700T*</i>
1316	3	1	4	<i>gyrA_A90V</i> , <i>gyrB_I310M*</i> , <i>gyrB_S700T*</i>
1444	2	1	4	<i>gyrA_A90V</i> , <i>gyrB_V340L</i>
1570	2	4	4	<i>gyrA_A90V</i> , <i>gyrB_V340L</i>

<b>1770</b>	2	4	6	gyrB_I310M*, gyrB_S700T*
<b>1811</b>	2	4	6	gyrA_D94G, gyrA_R392C
<b>2017</b>	2	4	>10	gyrA_D94G, gyrB_V340L
<b>2028</b>	2	8	>10	gyrA_D94G, gyrB_N538T
<b>2135</b>	2	0.25	<1	gyrB_I310M*, gyrB_S700T*
<b>3092</b>	2	<0.125	<1	gyrB_I310M*, gyrB_S700T*
<b>3202</b>	2	<0.125	<1	gyrB_I310M*, gyrB_S700T*
<b>3431</b>	2	<0.125	<1	gyrB_I310M*, gyrB_S700T*
<b>2170</b>	2	0.25	<1	gyrB_I310M*, gyrB_S700T*
<b>899</b>	2	0.5	2	gyrA_D94A, gyrB_V340L

Supplementary Figure 1: Isolate neighbor joining tree. Subtree branches were colored if a neighbor had a fingerprint from MIRU or spoligotyping as follows: LAM: olive, T1: purple, X: red, Beijing: dark blue, H: grey, T2: light blue, unknown: black.

