Case Studies of Rifampin Resistance MTBC in Contra Costa County

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Abstract
Rapid detection of Mycobacterium tuberculosis complex (MTBC) is crucial in minimizing the spread of disease and improving patient outcomes. The Cepheid GeneXpert MTB/rif assay detects MTBC and potential rifampin resistance. Previous studies have shown that there are a multitude of reasons in which false-positive rifampin resistance may occur. We retrospectively analyzed five MTBC rifampin resistant results, including three falsely reported as resistant.

Introduction
The GeneXpert MTB/rif assay provides MTBC results, as well as the detection of mutations in the rpoB gene, indicating potential rifampin resistance. Drug regimens prescribed as a result of interpretation of erroneous lab results may result in worse patient outcomes such as increased costs, rates of drug failure, toxicities and death. We investigated five cases of reported rifampin resistance.

The rifampin resistance detection works in the “opposite” fashion of a traditional PCR assay. As a result, a false-resistant no binding occurs, and rifampin “resistance” is opposite fashion of a traditional PCR assay.

Methods
We retrospectively analyzed all Contra Costa County PHL AFB sputum specimens from 2017 to 2021. Specimens were included if:
- MTB/rif PCR ordered
- Were positive for MTB
- Were positive for rifampin resistance

Four specimens were found to match the selection criteria and an additional specimen was submitted by the San Francisco Department of Public Health (SFPDHL) (Sample E).

Confirmatory testing was performed using either the BD MGIT DST protocol or by CDPH’s Microbial Diseases Laboratory pyrosequencing assay.

Results
Five cases of rifampin resistance were identified (Table 1).

- 3 cases of false-rifampin resistance
- 2 cases of true-rifampin resistance.

Sample A
Patient A’s initial sample was identified by Xpert MTB/rif as rifampin susceptible, but a second specimen (Sample A) collected 11 days later resulted as rifampin resistant. PSQ and DST were also ordered to confirm rifampin resistance. Both showed the 2nd specimen was rifampin susceptible.

Sample B
Sample B was identified by Xpert MTB/rif as MTBC positive with rifampin resistance. Subsequent PSQ revealed a silent mutation at MTBC codon 514, TTC > TTT, with no change from the original amino acid phenylalanine. Because MTBC codon 514 is located in the middle of Probe B, no amplification occurred which ultimately resulted in reporting of a rifampin resistant result.

Sample C
Sample C was identified as MTBC positive with rifampin resistance by Xpert MTB/rif, followed by PSQ which showed a missense mutation at MTBC codon 450, TCG > TTG, and resulted in a change from serine to leucine. The MTBC codon 450 is also located in the middle of probe E and likely contributed to the lack of amplification. DST confirmed that the isolate was resistant to rifampin at 1.0 μg/mL. Of note, this mutation has previously been associated with rifampin and rifabutin resistance.

Sample D
A sample from Patient D was identified as MTBC positive with rifampin resistance by Xpert MTB/rif. Followed by PSQ which showed a silent mutation in the MTBC core region, with no change from the original amino acid phenylalanine.

Sample E
The isolate was reported as rifampin susceptible, but no rifampin DST was ordered. Because rifampin resistance is known to be associated with mutations in the MTBC core region, the isolate was subjected to DST and PSQ. DST confirmed the isolate was rifampin resistant, but PSQ results showed the same mutation as in sample C.

Results (Cont.)
We suspect that the discrepant 2nd result was due to the treatment for active MTBC at the time of specimen collection, it is possible that ongoing medical therapy may have contributed to the discrepant results.

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References

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