



MYCF

Mycobacteria DNA by PCR - FFPE

GA Test Code	250 FFPE - Formalin-fixed Paraffin-embedded
Method	Real-Time Polymerase Chain Reaction (rPCR) – Qualitative
PCR/Probe Targets	Mycobacteria DNA (<i>16s rRNA</i> gene) Mycobacteria DNA (<i>hsp65</i> gene) <i>Mycobacterium tuberculosis</i> complex (<i>IS6110</i> gene) Human DNA (QC indicator) Note: Order GA Test #1000 Reflex to Mycobacteria DNA Sequencing to identify atypical species (e.g. <i>Mycobacterium gordonae</i>).
Specimens	FFPE (Formalin-fixed, Paraffin-embedded) Tissue: submit 6 shavings in 3-micron sections, sterile container, ambient. <i>Please do not submit entire FFPE tissue block, unless you are unable to produce shavings.</i> Slides: provide tissue on 5-6 slides, unstained, without cover slips, ambient. Note: Submitting fewer than the recommended number of shavings or slides is acceptable if adequate tissue/cellular material is present. Note: The preferred fixative for subsequent PCR analysis is 10% neutral buffered formalin. The least desirable are Carnoy's, Zenker's, and Boulin's fixatives, which severely degrade DNA. Optimal results are achieved with tissues that have been fixed in 10% neutral buffered formalin for less than 24 hours. Fixation has a more adverse effect on the detection of mycobacteria other than tuberculosis since only one copy of the 16S rRNA gene is present per cell and it is twice as large as the <i>M. tuberculosis</i> complex specific IS6110 gene.
Causes for Rejection	Quantity not sufficient (QNS) for analysis
Reference Range	Not Detected
Turnaround Time	Performed once per week on specimens received by Tuesday. Lysis takes 24 hours (overnight on Tuesday). Test performed on Wednesday. Preliminary results on Thursday. If necessary, results confirmed and reported on Friday.
CPT Codes	87551 (x2), 87556, 87798

Description

This assay involves testing by real-time polymerase chain reaction (rPCR) amplification of the *16s rRNA*, *hsp65*, and *IS6110* genes. The human *beta-globin* gene is used as a quality control indicator for formalin-fixed tissue by verifying that the sample DNA is of adequate quality and quantity for analysis. If human DNA is not detected, the sample will be rejected. The *16S rRNA* gene is a highly conserved mycobacteria target of 1000 bases and offers broad range coverage of all mycobacteria species. The *hsp65* gene is less conserved, but the amplicon is much smaller in size (134 bases), allowing greater sensitivity from formalin-fixed tissue where DNA fragmentation has occurred. The *IS6110* gene (70 base amplicon) is a multi-copy gene found only in *M. tb* complex mycobacteria. The assay is sensitive down to approximately 5 cells/sample for species in the *M. tuberculosis* complex (*M. tuberculosis*, *M. bovis*, *M. bovis BCG*, *M. africanum*, *M. microti*, and *M. canettii*) and 50 cells/sample for atypical mycobacteria.

Clinical Utility

Conventional methods for detection and identification of mycobacteria species based on biochemical tests need several weeks and may remain inconclusive. Rapid and accurate identification of mycobacteria species is important in order to administer timely and appropriate antibiotic therapy.

Genetic Assays, Inc.