

## Overview

### Useful For

Determining the species of a *Mycobacterium tuberculosis* complex culture isolate

### Additional Tests

Test Id	Reporting Name	Available Separately	Always Performed
RTBSP	Id, Mtb Speciation, PCR	No, (Bill Only)	Yes

### Testing Algorithm

When this test is ordered, species identification will always be performed at an additional charge.

### Special Instructions

- [Infectious Specimen Shipping Guidelines](#)

### Method Name

Real-Time Polymerase Chain Reaction (PCR)

### NY State Available

Yes

## Specimen

### Specimen Type

Varies

### Ordering Guidance

This test should be used to identify the species within the *Mycobacterium tuberculosis* complex from a known *M tuberculosis* complex isolate.

For identification of *M tuberculosis* complex from isolate growth, order CTBID / Culture Referred for Identification, *Mycobacterium* and *Nocardia*, Varies.

For rapid identification of *M tuberculosis* complex directly from a specimen, order MTBRP / *Mycobacterium tuberculosis* Complex, Molecular Detection, PCR, Varies or MTBXS / *Mycobacterium tuberculosis* Complex, Molecular Detection and Rifampin Resistance, PCR, Sputum.

### Shipping Instructions

1. For shipping information see [Infectious Specimen Shipping Guidelines](#).

2. Place specimen in a large infectious container and label as an etiologic agent/infectious substance.

### Necessary Information

Specimen source and suspected organism identification are required.

### Specimen Required

**Specimen Type:** *Mycobacterium tuberculosis* complex isolate growing in pure culture

**Supplies:** Infectious Container, Large (T146)

**Container/Tube:** Growth on solid slant media, eg, Middlebrook 7H10, 7H11 and Lowenstein Jensen; growth in broth medium, eg, Mycobacteria Growth Indicator Tube, 7H9 broth BACT/ALERT MP or VersaTREK

**Specimen Volume:** Isolate with visible growth on solid media; if broth is sent, 3 mL or more of broth culture

#### Collection Instructions:

1. Bacterial organism must be submitted in pure culture, actively growing. **Do not submit mixed cultures.**
2. Place specimen in a large infectious container and label as an etiologic agent/infectious substance.

**Additional Information:** If subculture to Middlebrook agar medium is needed to ensure purity, turnaround time for results may be delayed.

### Reject Due To

Agar plate Mixed culture	Reject
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### Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Ambient (preferred)		
	Refrigerated		

## Clinical & Interpretive

### Clinical Information

This assay provides a species-level identification of microbiologic culture isolates previously identified to be a member of the *Mycobacterium tuberculosis* complex. Species level identification can be important for patient care or for epidemiologic investigations. For example, the species-level identification of *Mycobacterium bovis* bacillus Calmette-Guerin (BCG) can assist with identification of disseminated infections following use of the vaccine as an adjuvant during chemotherapy.

### Reference Values

Not applicable

### Interpretation

This assay can differentiate the most common species within the *Mycobacterium tuberculosis* complex, which are, *M tuberculosis*, *Mycobacterium bovis*, *Mycobacterium bovis* bacillus Calmette-Guerin (BCG; the vaccine strain),

*Mycobacterium canettii*, *Mycobacterium caprae*, *Mycobacterium microti*, and *Mycobacterium pinnipedii*. This assay cannot distinguish *Mycobacterium africanum* from *Mycobacterium mungi* so if that result is obtained, the organism will be reported as *M africanum*/*M mungi*.

### Cautions

Only isolates of *Mycobacterium tuberculosis* complex should be submitted and they must be in pure culture. Nontuberculous mycobacteria should not be submitted. Mixed cultures will result in a delay because the *M tuberculosis* complex organism must be isolated prior to performing the polymerase chain reaction assay.

This assay has not been verified for the direct detection of *M tuberculosis* complex from clinical specimens. It is intended for use on microbiologic culture isolates already identified as *M tuberculosis* complex.

### Supportive Data

Type strains of *Mycobacterium tuberculosis* complex members were tested using the species identification polymerase chain reaction (PCR) assay and all were identified correctly. Type strains tested were *M tuberculosis* ATCC 27294, *Mycobacterium bovis* ATCC 19210, *M bovis* BCG ATCC 101472, *Mycobacterium africanum* ATCC 25240, *Mycobacterium microti* ATCC 19422, *Mycobacterium caprae* ATCC BAA 824, *Mycobacterium pinnipedii* ATCC BAA 688.

In addition, a clinical isolate of *Mycobacterium canettii*, identified by whole genome sequencing at the New York State Department of Health Wadsworth Center, was tested and confirmed to be *M canettii* by the *M tuberculosis* complex species identification PCR assay.

As part of the verification of this assay, 78 *M tuberculosis* complex isolates with the species identified at a reference laboratory were tested using the species identification PCR assay. All 78 isolates were correctly identified to the species level.

Table. Species reported by reference laboratory

LC 480 PCR results		<i>M tuberculosis</i>	<i>M bovis</i>	<i>M bovis</i> BCG	<i>M africanum</i>
	<i>M tuberculosis</i>	53			
	<i>M bovis</i>		4		
	<i>M bovis</i> BCG			14	
	<i>M africanum</i>				7

Although the species identification test can be used only for mycobacterial isolates already identified as *M tuberculosis* complex, 159 other *Mycobacterium* species isolates were tested to determine whether any nontuberculous mycobacteria would be positive in the test. No nontuberculous mycobacteria were positive in the *M tuberculosis* complex species identification PCR assay.

### Clinical Reference

Fitzgerald DW, Sterling TR, Haas DW. Mycobacterium tuberculosis. In: Mandell GL, Bennett JE, Dolin R, eds. Mandell, Douglas and Bennett's Principles and Practice of Infectious Diseases. 9th ed. Elsevier; 2020:2985-3021

### Performance

## Method Description

The method uses real-time polymerase chain reaction on the LightCycler 480 platform with *Mycobacterium tuberculosis* complex-specific primers coupled with fluorescence resonance energy transfer probes to differentiate the members of the complex to the species level. The probes target specific regions of difference (RD) within the *M tuberculosis* complex genome. Detection of the presence or absence of these RD allows for differentiation of the species within the *M tuberculosis* complex. (Halse TA, Escuyer VE, Musser KA. Evaluation of a single tube multiplex real-time PCR for differentiation of members of the *Mycobacterium tuberculosis* complex in clinical specimens. J Clin Microbiol. 2011;49:2562-2567; Warshauer DM, Salfinger M, Desmond E, and Lin S-Y G. *Mycobacterium tuberculosis* complex. In: Carroll KC, Pfaller MA, Landry ML, et al, eds. Manual of Clinical Microbiology. 12th edition, ASM Press; 2019:576-594)

The RD's expected for each species within the *M tuberculosis* complex are shown in the table below. "+" indicates the region is present and "-" indicates the region is absent.

Expected RD signature patterns	RD1	RD4	RD9	RD12	RD9-2
<i>Mycobacterium tuberculosis</i>	+	+	+	+	+
<i>Mycobacterium bovis</i>	+	-	-	-	+
<i>Mycobacterium bovis BCG</i>	-	-	-	-	+
<i>Mycobacterium africanum</i>	+	+	-	+	+
<i>Mycobacterium canettii</i>	+	+	+	-	+
<i>Mycobacterium caprae</i>	+	+	-	-	+
<i>Mycobacterium microti</i>	-	+	-	+	+
<i>Mycobacterium mungi</i>	+	+	-	+	+
<i>Mycobacterium pinnepedii</i>	+	+	-	-	+

## PDF Report

No

## Day(s) Performed

Monday through Friday

## Report Available

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7 to 14 days

**Specimen Retention Time**

Subculture: 1 year

**Performing Laboratory Location**

Rochester

**Fees & Codes****Fees**

- Authorized users can sign in to [Test Prices](#) for detailed fee information.
- Clients without access to Test Prices can contact [Customer Service](#) 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact [Customer Service](#).

**Test Classification**

This test has been cleared, approved, or is exempt by the US Food and Drug Administration and is used per manufacturer's instructions. Performance characteristics were verified by Mayo Clinic in a manner consistent with CLIA requirements.

**CPT Code Information**

87150

**LOINC® Information**

Test ID	Test Order Name	Order LOINC® Value
TBSP	M tuberculosis species ID, PCR	94576-6

Result ID	Test Result Name	Result LOINC® Value
TBSP	M tuberculosis species ID, PCR	94576-6