MAYO CLINIC LABORATORIES

Test Definition: TBSP

Mycobacterium tuberculosis Complex Species Identification, PCR, Varies

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

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 pinnepedii*. 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		*M tuberculosis*	*M bovis*	*M bovis* BCG	*M africanium*
	M tuberculosis	53			
	M bovis		4		
	M bovis BCG			14	
	M africanium				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
Mycobacterium tuberculosis	+	+	+	+	+
Mycobacterium bovis	+	-	-	-	+
Mycobacterium bovis BCG	-	-	-	-	+
Mycobacterium africanum	+	+	-	+	+
Mycobacterium canettii	+	+	+	-	+
Mycobacterium caprae	+	+	-	-	+
Mycobacterium microti	-	+	-	+	+
Mycobacterium mungi	+	+	-	+	+
Mycobacterium pinnepedii	+	+	-	-	+

PDF Report

No

Day(s) Performed

Monday through Friday

Report Available

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