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Health

So Many Numbers: How to Interpret QFT Results

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*Clinical Microbiology Laboratory
Supervisor*

OUR MISSION:

To promote, protect, and improve the health and safety of all Hoosiers.

OUR VISION:

Every Hoosier reaches optimal health regardless of where they live, learn, work, or play.



Objectives

1. NAAT Fact Sheet
2. Online Ordering for Containers
3. Updated Lab Q&A
4. QFT Overview
5. QFT Results Interpretation



NEW—NAAT Fact Sheet

- Describes the importance of NAAT in rapid TB diagnosis
- Includes information on results interpretation
- Can be distributed to physicians, IPs, and other external partners as needed

Nucleic Acid Amplification Testing (NAAT)



Feb

What is a NAAT for TB?

A nucleic acid amplification test, or NAAT, for tuberculosis (TB) is a molecular test used to detect the DNA of *Mycobacterium tuberculosis* complex (MTBC) in a sputum or other respiratory specimen. As the quantity of DNA in a specimen may be very small, NAA testing includes a step that amplifies, or copies, the genetic material. Polymerase chain reaction (PCR) is a common form of NAAT performed in many TB laboratories. In some situations, NAAT may also be performed on extrapulmonary specimens.

When should a NAAT be ordered?

A NAAT should be ordered on at least one respiratory specimen from a patient with signs and symptoms of pulmonary TB when a diagnosis of TB is considered but not yet confirmed.

Source: Diagnosis of Tuberculosis in Children and Adults Clinical Infectious Diseases, Volume 64, Issue 1, January 2017, Pages e1–e33, <https://doi.org/10.1093/cid/ciw694>

How to order a NAAT at a laboratory

Please refer to the IDOH lab *Mycobacterium tuberculosis* Submission Guidelines

NEW: Ordering Containers on LimsNet

Containers can now be ordered through the Test drop down menu in LimsNet:

1. Check the box next to the type to be ordered
2. Enter the quantity of whole kits or parts
3. Click "Submit"

Request the needed test types from the LimsNet helpdesk: LimsAppSupport@isdh.in.gov

IDOHL Clinical Container Request					
Shipping Information					
Site*	<input type="text" value="ABC TESTING SITE"/>				
Contact Name*	<input type="text" value="JESSICA GENTRY"/>				
Phone	<input type="text" value="(317)921-5500"/>				
Email ID*	<input type="text" value="jgentry@isdh.in.gov"/>				
Street Address*	<input type="text" value="550 W. 16th Street (LAB)"/>				
City*	<input type="text" value="Indianapolis"/>	State*	<input type="text" value="IN"/>	Zip*	<input type="text" value="46202"/>
Kit Request Information					
Email Questions to: Containers@health.in.gov				Questions call: 317-921-5875	
Kit #	Kit Type	# Whole Kits*	Extra or Replacement Kit Components Only	Quantity	
<input type="checkbox"/> 1B	HIV (32/pkg). Max: 3	<input type="text"/>	Mailing Labels Max: 96	<input type="text"/>	
<input type="checkbox"/> 5B	Syphilis (32/pkg). Max: 3	<input type="text"/>	Mailing Labels Max: 96	<input type="text"/>	
<input type="checkbox"/> 11B	Hepatitis (32/pkg). Max: 3	<input type="text"/>	Mailing Labels Max: 96	<input type="text"/>	
<input type="checkbox"/> 6A	TB Min: 3, Max: 24	<input type="text"/>	Conical Tube Max: 24	<input type="text"/>	
			UPS Bags and RS Labels (except Marion County) Max: 24	<input type="text"/>	
<input type="checkbox"/> 6A	TB QFT Max: 10 * TB QFT boxes include 1 QFT tube. Additional tubes can be ordered under the "Extra or Replacement Kit Components Only" section on the right (up to six tubes per box).	<input type="text"/>	QFT Tubes Max: 60	<input type="text"/>	
			UPS Bags and RS Labels (except Marion County) Max: 10	<input type="text"/>	
			Transport Media (only) Max: 15	<input type="text"/>	

Updated Lab Q&A

- Will be posted to TB section of IDOH website
- Handy reference material for new TB nurses and for those who rarely handle a TB case
- Biggest change was the addition of QFT info



QFT Specimen Collection



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Collection Methods Available

Single Tube:
Minimum 5 mL



4 Tube:
1 mL in each tube

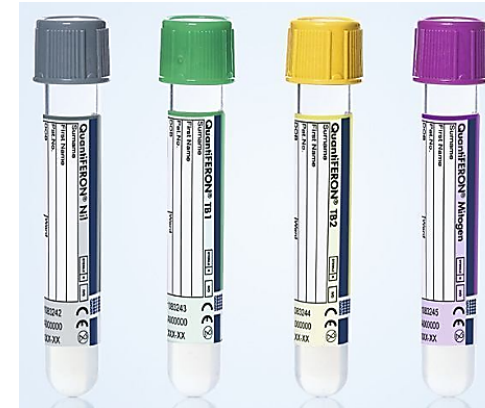


Blood Collection Methods

Option 1: Draw at least 5 mL of blood into a single lithium heparin collection tube. Blood may be stored at room temperature for up to 24 hours or refrigerated for up to 53 hours prior to testing.



Option 2: Collect 1 mL of whole blood directly into each of four QFT-Plus blood collection tubes and hold at room temperature for up to 16 hours prior to incubation.



Hold Time: Option #1

Draw into lithium-heparin tube and hold at room temperature

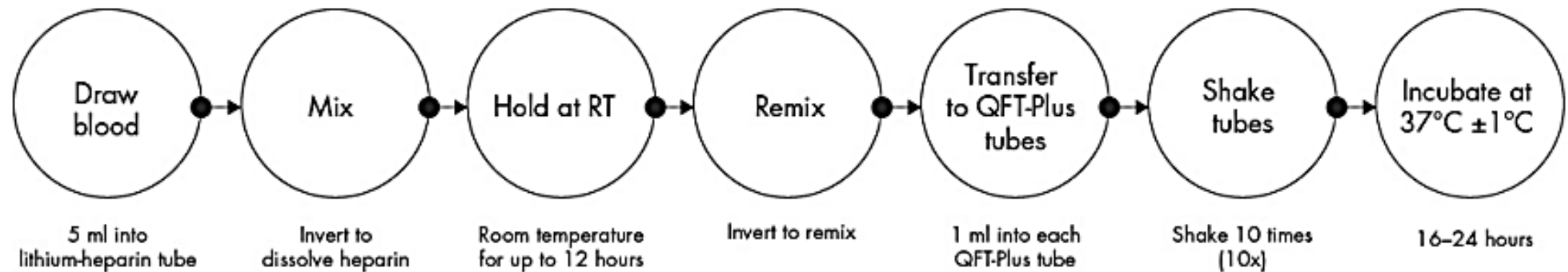
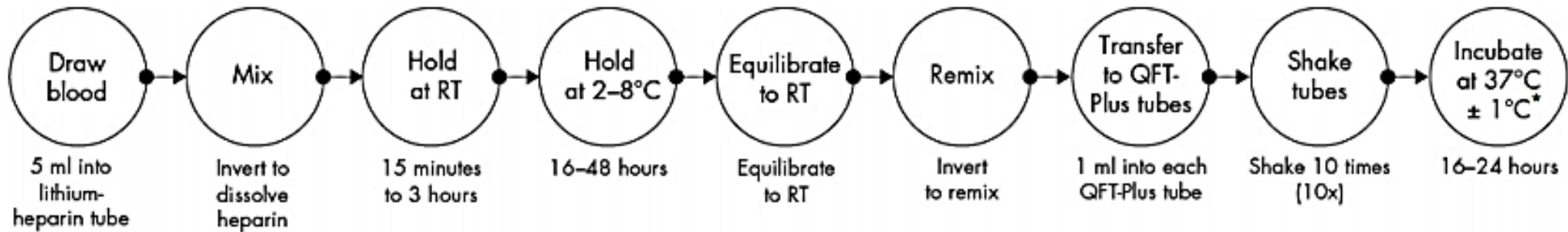


Figure 2. Blood collection option: Draw into lithium-heparin tube and hold at room temperature. The total time from blood draw in lithium-heparin tube to 37°C incubation must not exceed 12 hours.

Hold Time: Option #2

Draw into lithium-heparin tubes and hold at 2–8°C



* Aliquotted QFT-Plus Blood Collection Tubes should be placed in a 37°C incubator within 2 hours of blood transfer to QFT-Plus Blood Collection Tubes.

Figure 3. Blood collection option: Draw into lithium-heparin tube and hold at 2–8°C. The total time from blood draw in lithium-heparin tube to 37°C incubation must not exceed 53 hours.

Hold Time: Option #3

Draw into QFT-Plus Blood Collection Tubes and hold at room temperature

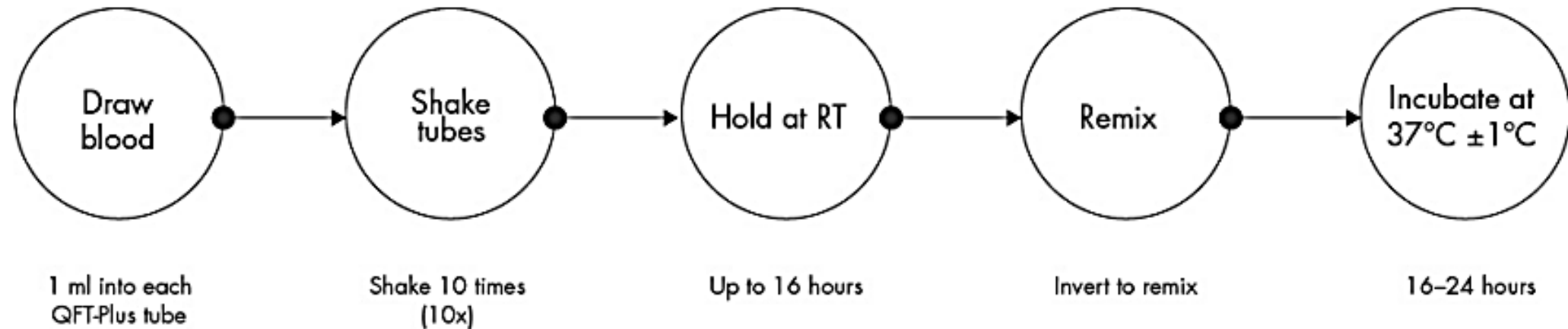


Figure 1. Blood collection option: Direct draw into QFT-Plus Blood Collection Tubes and hold at room temperature. The total time from blood draw in QFT-Plus Blood Collection Tubes to 37°C incubation must not exceed 16 hours.

Which Collection Method is Better?

Single Tube

- It's one tube!
- More accurate results--4 tube draw requires specific and detailed pre-analytical processing that can impact the results

4 Tube

- More flexible hold times

Whichever option is used, it is important to strictly follow the sample handling instructions exactly. Improper handling could lead to inaccurate results.



Packaging and Shipping



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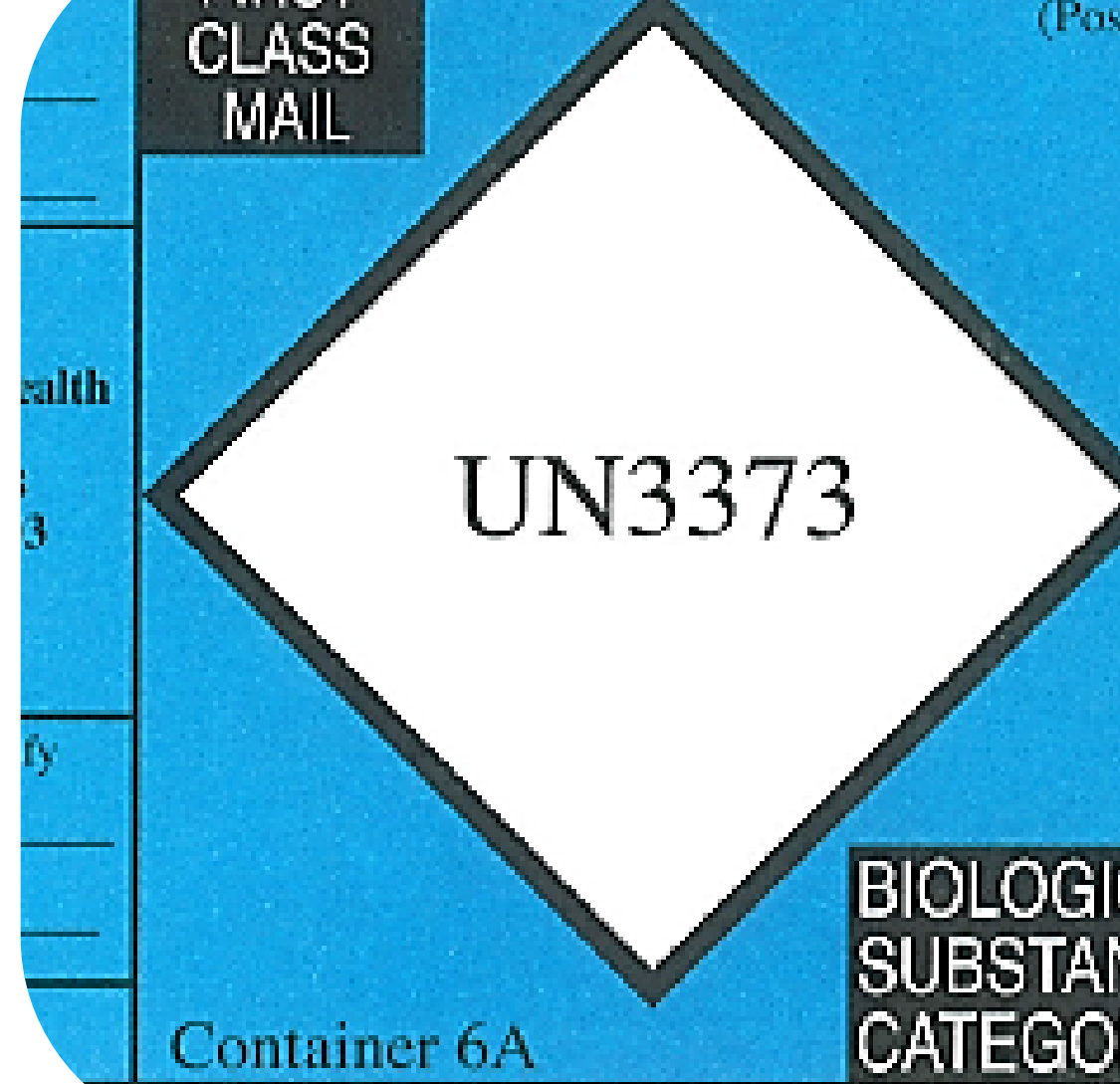
Specimen Packaging and Shipping

- All blood specimens must be packaged as a Category B, Biological Substance
- IDOH provides collection tubes and shipping supplies free for local health departments

To order containers:

Email: containers@isdh.in.gov

Phone: 317-921-5875



Refrigerated Shipping Supplies

1. Blood tubes should be placed in a biohazard bag with absorbent material
2. Bag and ice packs should be placed in cooler box
3. Cooler box is placed in outer box
4. Outer box is placed in the UPS bag





LimsNet



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Ordering QFT in LimsNet

- The same test name *Tuberculosis* is used for both sputum collections and QFT tests
- Test types:
 1. Specimen (AFB Smear and Culture)
 2. Mycobacteriology Isolate
 3. **QuantiFERON-TB**
 - ❖ When one of these is selected, it will gray out the non-applicable fields for each test type
- Reason for test
- Specimen information

Billing and Pricing

QFT is a billable test for IDOH:

- LHDs may provide valid insurance billing information for each patient tested
OR
- LHDs will be billed \$20 per test

This billing strategy will allow IDOH to recover the cost of the QFT tubes/ELISA test kits



Nicholas S. Tenorio, Health Communication Specialist

QFT Laboratory Testing

QFT Lab Testing

- Blood from lithium heparin tubes is transferred into QFT tubes after arrival at IDOH
- QFT tubes are shaken 10 times and incubated at 37 °C overnight
- After incubation, tubes are centrifuged to harvest plasma.



QFT Lab Testing

- Plasma is tested by ELISA
- ELISA gives a numerical result for each tube, in international units/mL (IU/mL)
- An interpretation is generated from the combination of results for the 4 tubes



→ Nil
→ TB 1 Ag
→ TB 2 Ag
→ Mitogen

QFT Stats

- Started accepting specimens in November 2019 from one county
- Rolled out the testing service to the entire state in early 2020
- Very few sites enrolled initially, due to COVID-19 pandemic response
- Through January 2022, IDOH has received 1569 specimens from 9 different LHDs

QFT Stats

Of 1569 specimens received:

- 88% Negative
- 10% Positive
- 1% Indeterminate
- 1.6% Rejected

Very low Indeterminate rate!

Reasons for Rejection

Only 25 of 1569 specimens have been rejected—1.6%

- 14 — Not enough blood in the tube—lab needs a minimum of 5 mL
- 6 — Extended transit times—shipped the day before a state holiday or UPS delayed delivery
- 3 — Lab error
- 2 — Collection error



James Gathany

Reporting and Results Interpretation



QuantiFERON Lab Report

Average time for results:
2-3 days



INDIANA STATE DEPARTMENT OF HEALTH LABORATORIES
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Judith C. Lovchik, Ph.D, D(ABMM)
Laboratory Director

CLIA Certification - 15D0662599

Laboratory Report

Submitter: ABC Labs
IN

Patient Name:	QFT, Valid 31	ISDH Lab Number:	C19000057
Patient ID:		Date Collected:	02/21/2019
Birth Date:	02/21/1954	Date Received:	02/21/2019
Sample Type:	Other		

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	2.19 IU/ml
QuantiFERON TB2 Ag	3.92 IU/ml
Mitogen	9.27 IU/ml
Nil	0.01 IU/ml
QuantiFERON-TB Gold Plus	POSITIVE

Result Interpretation

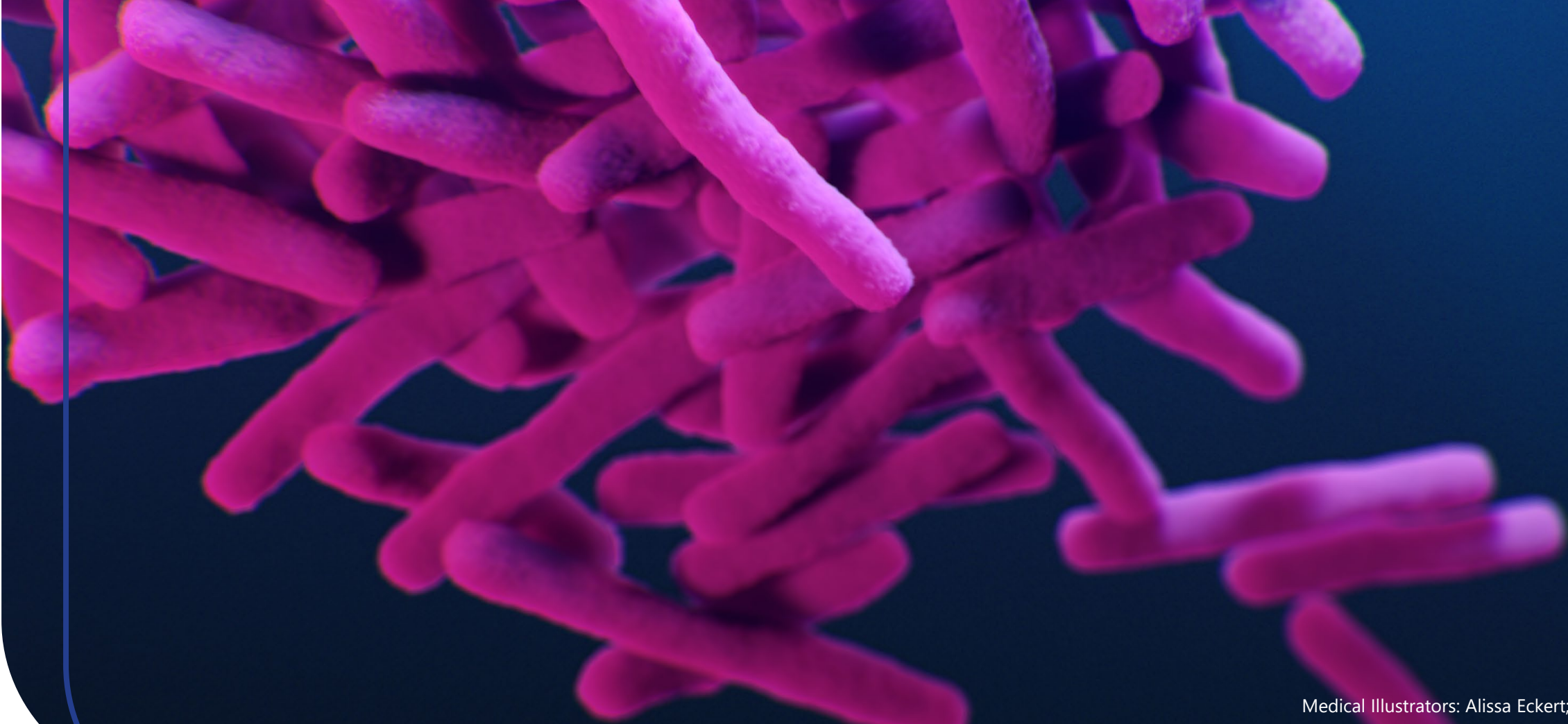
M. tuberculosis infection likely



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Results Interpretation

Nil (IU/ml)	TB1 minus Nil (IU/ml)	TB2 minus Nil (IU/ml)	Mitogen minus Nil (IU/ml)*	QFT-Plus Result	Report/interpretation
≤8.0	≥0.35 and ≥25% of Nil	Any	Any	Positive [†]	<i>M. tuberculosis</i> infection likely
	Any	≥0.35 and ≥25% of Nil			
	<0.35 or ≥0.35 and <25% of Nil	<0.35 or ≥0.35 and <25% of Nil	≥0.50	Negative	<i>M. tuberculosis</i> infection NOT likely
	<0.35 or ≥0.35 and <25% of Nil	<0.35 or ≥0.35 and <25% of Nil	<0.50	Indeterminate [‡]	Likelihood of <i>M. tuberculosis</i> infection cannot be determined
>8.0 [§]	Any				



Medical Illustrators: Alissa Eckert; James Arche

QFT Case Studies



QFT Patient #1

Patient history: 22-year-old female day care employee born in U.S.

Reason for test: annual screening requirement

Symptoms: none

Lab Report:

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	0.07 IU/mL
QuantiFERON TB2 Ag	0.07 IU/mL
Mitogen	> 10 IU/mL
Nil	0.06 IU/mL

QFT Patient #1

Nil: 0.06 IU/mL

TB1-Nil: 0.01 IU/mL

TB2-Nil: 0.01 IU/mL

Mitogen-Nil: 9.94 IU/mL

TB1-Nil and TB2-Nil are both
<0.35

Mitogen-Nil is >0.5

Result: **Negative**

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	0.07 IU/mL
QuantiFERON TB2 Ag	0.07 IU/mL
Mitogen	> 10 IU/mL
Nil	0.06 IU/mL

QFT Patient #2

Patient history: 48-year-old male born in the Philippines

Reason for test: contact investigation (known household contact)

Symptoms: productive cough, fever, malaise

Lab Report:

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	4.54 IU/mL
QuantiFERON TB2 Ag	5.27 IU/mL
Mitogen	> 10 IU/mL
Nil	0.20 IU/mL

QFT Patient #2

Nil: 0.20 IU/mL

TB1-Nil: 4.34 IU/mL

TB2-Nil: 5.07 IU/mL

Mitogen-Nil: 9.80 IU/mL

TB1-Nil and TB2-Nil are both >0.35

Mitogen-Nil is >0.5

Result: **Positive**

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	4.54 IU/mL
QuantiFERON TB2 Ag	5.27 IU/mL
Mitogen	> 10 IU/mL
Nil	0.20 IU/mL

QFT Patient #3

Patient history: 68-year-old being treated for cancer

Reason for test: abnormal CXR

Symptoms: non-productive cough

Lab Report:

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	0.01 IU/mL
QuantiFERON TB2 Ag	0.01 IU/mL
Mitogen	0.45 IU/mL
Nil	0.02 IU/mL

QFT Patient #3

Nil: 0.02 IU/mL

TB1-Nil: 0.0 IU/mL

TB2-Nil: 0.0 IU/mL

Mitogen-Nil: 0.43 IU/mL

TB1-Nil and TB2-Nil are both <0.35

Mitogen-Nil is <0.5

Result: **Indeterminate**

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	0.01 IU/mL
QuantiFERON TB2 Ag	0.01 IU/mL
Mitogen	0.45 IU/mL
Nil	0.02 IU/mL

QFT Patient #4

Patient history: 40-year-old, US born

Lab Report:

Reason for test: starting biologic treatment

Symptoms: none

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	2.22 IU/mL
QuantiFERON TB2 Ag	2.19 IU/mL
Mitogen	> 10 IU/mL
Nil	0.11 IU/mL

QFT Patient #4

Nil: 0.11 IU/mL

TB1-Nil: 2.11 IU/mL

TB2-Nil: 2.08 IU/mL

Mitogen-Nil: 9.89 IU/mL

TB1-Nil and TB2-Nil are both >0.35

Mitogen-Nil is >0.5

Result: **Positive**—likely LTBI

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	2.22 IU/mL
QuantiFERON TB2 Ag	2.19 IU/mL
Mitogen	> 10 IU/mL
Nil	0.11 IU/mL

QFT Patient #5

Patient history: 55-year-old born in India

Reason for test: abnormal CSX, cavitory lesion (right upper lobe)

Symptoms: productive cough, fever, weight loss

Lab Report:

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	0.50 IU/mL
QuantiFERON TB2 Ag	0.48 IU/mL
Mitogen	> 10 IU/mL
Nil	0.23 IU/mL

QFT Patient #5

Nil: 0.23 IU/mL

TB1-Nil: 0.27 IU/mL

TB2-Nil: 0.25 IU/mL

Mitogen-Nil: 9.77 IU/mL

TB1-Nil and TB2-Nil are both <0.35

Mitogen-Nil is >0.5

Result: **Negative**

<u>Test</u>	<u>Result</u>
QuantiFERON TB1 Ag	0.50 IU/mL
QuantiFERON TB2 Ag	0.48 IU/mL
Mitogen	> 10 IU/mL
Nil	0.23 IU/mL

QFT Patient #5

QFT collected 10/7

Sputum collected 10/13

Mycobacteriology Laboratory	
<u>Test</u>	<u>Results</u>
Microscopy	Auramine O-phenol Acid Fast Bacteria Found >50/field
Specimen PCR	M. avium complex: Not Detected
Specimen PCR	M. tuberculosis complex: Detected

Bottom Line...

QFT is just one piece of the puzzle. It ***MUST*** be considered in context with the patient history, radiological findings and other laboratory results.



Questions?

CONTACT:

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Thank you!

