Laboratory Services

CONTENTS

Introduction

Purpose

Use this section to

- get contact information for laboratories;
- determine which tests are available and the tests' turnaround times; and
- identify which laboratory can perform a specific test.

The diagnosis of tuberculosis (TB), management of patients with the disease, and public health TB control services rely on accurate laboratory tests. Laboratory services are an essential component of effective TB control, providing key information to clinicians (for patient care) and public health agencies (for control services).[1]

Policy

Public health laboratories should ensure that clinicians and public health agencies within their jurisdictions have ready access to reliable laboratory tests for diagnosis and treatment of TB.[2]

Effective TB control requires timely, complete, and accurate communication among the laboratory system, TB control program, and healthcare provider.[3]

For roles and responsibilities, refer to the "Roles, Responsibilities, and Contact Information" topic in the Introduction **1.11**.

State Laws and Regulations

Laboratories must report *Mycobacterium tuberculosis.* Alaska Statutes and Regulations pertaining to the control of tuberculosis in Alaska are available in the Statutes and Regulations section of the manual **19.1**.

Laboratory Contact Information

To locate and contact a laboratory, refer to Table 1: **Laboratory Contact Information**.

For the list of the tests performed at each laboratory, refer to Table 2: **Available Laboratory Tests**.

Table 1: **LABORATORY CONTACT INFORMATION**

Roles and Responsibilities	Contact Information
State Laboratory • Receives and performs primary specimen AFB concentrated smear and culture testing for mycobacteria • Susceptibility testing of first-line drugs on *Mycobacterium tuberculosis* complex isolates	**Alaska State Public Health Laboratory (ASPHL)** Mycobacteriology Laboratory 5455 Martin Luther King Jr Ave. P.O. Box 196093 Anchorage, AK 99507 Tel: (907) 334-2100 Fax: (907) 334-2161
National PHL DST Reference Center • Pyrosequencing (PSQ) for the molecular detection of drug resistance • Confirmation of first-line drug testing on *Mycobacterium tuberculosis* complex isolates • Second-line drug testing on *Mycobacterium tuberculosis* complex isolates	**California Department of Public Health** National PHL DST Reference Center 850 Marina Bay Parkway, E164 Richmond, CA 94804 Tel: (510) 412-3929 Fax: (510) 412-3704
Centers for Disease Control and Prevention • DNA sequencing for the molecular detection of drug resistance (MDDR Program) • Confirmation of first-line drug testing on *Mycobacterium tuberculosis* complex isolates • Second-line drug testing on *Mycobacterium tuberculosis* complex isolates	**CDC STAT Lab** TB Laboratory, Unit 29 1600 Clifton Road, NE Atlanta, Ga. 30333 Tel: (404) 639-2455 Fax: (404) 639-5491
Private Laboratory • Non-tuberculosis *mycobacterium* susceptibility testing • Confirmation of first-line drug testing on *Mycobacterium tuberculosis* complex isolates • Second-line drug testing on *Mycobacterium tuberculosis* complex isolates	**National Jewish Health** Mycobacteriology Reference Laboratory 1400 Jackson St. Denver, CO 80206 Tel: 800-550-6227 Fax: (303) 398-1953

Available Laboratory Tests

Table 2: **LABORATORY TESTS AVAILABLE IN ALASKA**

Test	Laboratory	Turnaround Time
Diagnosis		
Acid-fast bacilli (AFB) smear (fluorochrome)	Alaska State Public Health Laboratory	Within 24 hours from receipt of specimen in the laboratory Monday thru Friday only (closed on national and state holidays)
Culture	Alaska State Public Health Laboratory	Cultures are incubated for 6 weeks before reported as negative. Time to detection of Mycobacterial growth is dependent upon growth rate and quality of specimen. Identification of cultured mycobacteria is usually within 14-21 days from date of receipt (this can be dependent on factors such as overgrowth by other organisms).
Drug susceptibility (first-line drugs only)	Alaska State Public Health Laboratory	Within 15 days from identification
Nucleic acid amplification (NAA) test Please see NAAT section **12.5**	Alaska State Public Health Laboratory	**GeneXpert® Xpert® MTB/RIF Assay:** within 24 hours from receipt of specimen in the laboratory.
QuantiFERON®-TB Gold In-Tube (QFT-GIT)	Testing is only available at private reference labs in Alaska	Varies with laboratory
Epidemiologic Monitoring		
Genotyping*	Michigan Department of Community Health – TB Lab 927 Terminal Rd. Lansing, MI 48906 Tel: (517) 335-8395	Within 14 days of receipt of *Mycobacterium tuberculosis* complex isolates

*All isolates of *M. tuberculosis* identified at the ASPHL are sent to the Michigan Department of Community Health Laboratory for genotyping. On the rare occasion that *M. tuberculosis* is identified at another laboratory, arrangements must be made for isolates to be sent to the ASPHL so that genotyping can be done.

Laboratories should report positive smears, NAA or positive cultures, and primary healthcare providers should report suspected or confirmed cases of TB to the health department, as specified in the "Reporting Tuberculosis" topic in the Surveillance section **(2.6)**. Prompt reporting allows the health department to organize treatment and case management services and to initiate a contact investigation as quickly as possible.[4]

For information on reporting, see the "Reporting Tuberculosis" topic in the Surveillance section **2.6**.

For laboratory services available in Alaska, contact the Mycobacteriology Department at 907-334-2139.

TB Nucleic Acid Amplification Testing (NAAT):

The ASPHL will perform the Xpert® MTB/RIF Assay on **initial smear-positive specimens**. Additionally, ASPHL will perform the Xpert® MTB/RIF Assay on **smear-negative specimens** from patients considered to be TB suspects upon provider request and **pre-approval** from the Alaska Tuberculosis Program. The *TB NAA Testing Authorization Form* must be completed prior to testing.

Patient Criteria

- Patient must have signs and symptoms of pulmonary TB
- Patient must be reported to the Alaska Tuberculosis Program as a TB suspect or TB case (907-269-8000)
- Patient must not have been diagnosed with TB or a nontuberculous mycobacterial infection or received treatment within the last 12 months

The link to the most current version of the *TB NAA Testing Authorization Form* is available in the Forms Section of the Manual **18.1.**

Refer to Tables 3 and 4: **NAA Testing Algorithm and Result Interpretation** for information about interpreting the results.

Table 3: **GENEXPERT® XPERT® MTB/RIF ASSAY RESULT INTERPRETATION**

Smear Result	MTB/RIF Assay Result	Interpretation
Smear Positive for AFB	**MTB DETECTED**	MTB target is detected within the sample. Use clinical judgment to determine whether to begin therapy while awaiting culture results. A positive NAA test does not necessarily indicate the presence of viable organisms.
	MTB Not Detected	MTB target is not detected within the sample. Use clinical judgment to determine whether to begin therapy while awaiting culture results. A patient is presumed to have an infection with nontuberculous mycobacteria, pending culture results. A negative MTB result on the Xpert MTB/RIF assay does not rule out pulmonary TB.
Smear Negative for AFB	**MTB DETECTED**	MTB target is detected within the sample. Use clinical judgment to determine whether to begin therapy while awaiting culture results. A positive NAA test does not necessarily indicate the presence of viable organisms.
	MTB Not Detected	Use clinical judgment to determine whether to begin therapy while awaiting results of culture and other diagnostic tests. A negative MTB result on the Xpert MTB/RIF assay does not rule out pulmonary TB.

Table 4: **GENEXPERT® XPERT® RIFAMPIN RESULT INTERPRETATION**

Rifampin Result	Interpretation
RIF Resistance NOT DETECTED	No *rpoB* mutation detected; likely rifampin susceptible.
RIF Resistance DETECTED	*rpoB* mutation detected; likely rifampin resistant. Confirmatory testing in progress.
RIF Resistance INDETERMINATE	Insufficient MTB in the sample to allow determination of the *rpoB* mutation result.

Sputum is phlegm from deep in the lungs. The important characteristics needed in sputum specimens are freshness and actual sputum, rather than saliva. An early morning specimen is best, so when collecting a set of three sputum specimens, at least one of them should be an early morning specimen.

To isolate mycobacteria from clinical materials successfully, handle specimens carefully after collection. For optimal results, collect specimens in clean, sterile containers and keep them in conditions that inhibit the growth of contaminating organisms, since most specimens will contain bacteria other than mycobacteria.[5]

Refer to Table 5 to review the methods used to collect various specimens and the type of specimens obtained for pulmonary tuberculosis (TB).

During procedures in which aerosols may be produced, use appropriate respiratory protection and environmental controls. For more information, refer to the CDC's "Guidelines for Preventing the Transmission of *Mycobacterium tuberculosis* in Health-care Settings, 2005" (*MMWR* 2005;54[No. RR-17]) at http://www.cdc.gov/mmwr/pdf/rr/rr5417.pdf .

Detailed information about diagnostic testing available at the Alaska State Public Health Laboratory (ASPHL) is available in the “Laboratory Services Manual”. It is available at: http://dhss.alaska.gov/dph/Labs/Documents/LaboratoryTests.pdf

Highlights include:

- “Tuberculosis/Mycobacterium Detailed Collection Instructions” (for the provider and laboratorian) and “Instructions for Collecting Sputum Samples” (for the patient).
- A linked table of contents so you can click on the test of interest in the table of contents and it will take you directly to that page.
- General information about who can request testing, and how to obtain supplies, request forms, and shipping boxes.
- Tests are listed alphabetically and include specific information about the sample required, storage and transport recommended, and when you can expect results back.

The ASPHL website also contains contact information, explanations of the services offered, and the forms needed to request testing and supplies. It is available at: http://dhss.alaska.gov/dph/Labs/Documents/LaboratoryTests.pdf

Table 5: **SPECIMEN COLLECTION METHODS AND TYPES FOR PULMONARY TUBERCULOSIS**

Pulmonary Tuberculosis Collection Method	Specimen Type
Spontaneous sputum collection occurs when the patient can cough up sputum without extra assistance.	▪ 5–10 ml of sputum from deep in the lung (Submit in sterile 50 mL conical tube with 50 mg of sodium carbonate preservative)
Induced sputum collection should be considered if a patient needs assistance in bringing up sputum.*	▪ 5–10 ml of sputum from deep in the lung (Submit in sterile 50 mL conical tube with 50 mg of sodium carbonate preservative)
Gastric aspirates can be submitted for the diagnosis of pulmonary tuberculosis (TB) in young children who cannot produce sputum.	▪ 5-10 ml of gastric contents (Adjust to neutral pH with 100 mg of sodium carbonate immediately following collection)
Bronchoscopy can be used in the following situations: ▪ If a patient cannot produce sputum by the above three methods[6] or ▪ If a patient has a substantial risk of drug-resistant TB and has initial routine studies that are negative[7] or ▪ In a patient in whom there is suspicion of endobronchial TB[8] or ▪ If a variety of clinical specimens for the diagnosis of pulmonary TB or other possible diseases need to be obtained	▪ Bronchial washings ▪ Bronchoalveolar lavage ▪ Transbronchial biopsy
* It is important to specify if the sputum is induced or not, because induced sputum is more watery and appears to be just saliva. Some laboratories may throw out induced sputum and report it as an inadequate specimen.	

Refer to Table 6 for collection methods and specimen types for extrapulmonary TB.

Table 6: **SPECIMEN COLLECTION METHODS AND TYPES FOR EXTRAPULMONARY TUBERCULOSIS**

Extrapulmonary Tuberculosis		
Collection Method	**Specimen Type**	
Extrapulmonary specimen collection from tissue and other body fluids can be submitted for the diagnosis of extrapulmonary tuberculosis.	**Examples of tissues (biopsy)*** ▪ Lymph node ▪ Pleural ▪ Bone/joint ▪ Kidney ▪ Peritoneal ▪ Pericardial	**Examples of fluids** ▪ Pleural ▪ Cerebrospinal ▪ Blood ▪ Urine ▪ Synovial ▪ Peritoneal ▪ Pericardial
*** Do not place specimens in formalin.**		

How to Perform Spontaneous Sputum Collection at a Healthcare Facility

1. Collect the specimen in a specialized room or booth designed for cough-inducing procedures.
2. Instruct the patient on how to collect the sputum sample.
   a. Put a mark at the 5 ml level on the sputum tube (if not already marked) to show the patient the minimum amount of sputum needed. (Most laboratories consider 5 to 10 ml an adequate amount.)
   b. Review with the patient how to collect sputum.
3. Make sure the specimen container and laboratory requisition are filled out completely before shipping.
   a. On the specimen container, record the patient name and the date and time of collection.
   b. Complete Test Request Form. These are located on the ASPHL website: http://dhss.alaska.gov/dph/Labs/Pages/publications/default.aspx
4. Make sure the specimen and laboratory requisition are packaged into appropriate shipping containers, per laboratory instructions.

Refer to the “Specimen Collection and Shipment Supplies” topic in the Supplies, Materials, and Services section, and see the “Specimen Shipment topic,” which follows.

5. If possible, send the specimen on the day it is collected. If this is not possible, refrigerate the specimen until it is sent on the next day.
6. Unless shipping by express delivery, such as Gold Streak, do not keep specimens to send all three on the same day.
7. Use the most rapid transport to the laboratory: You, courier, overnight carrier, or WE mail.

Make every effort to submit specimens to the laboratory within 24 hours of collection. Normal flora can overgrow any mycobacteria in the specimen and make it unusable. If specimens cannot be submitted within 24 hours, keep in mind that ASPHL will not run a specimen over 10 days old. Know how long it takes the specimen to get to the laboratory from the time it leaves your hands, and submit specimens accordingly.

How to Direct a Patient to Perform Spontaneous Sputum Collection at Home

If a patient will be collecting sputum specimens at home, provide the following guidance.

1. Put a mark at the 5 ml level on the sputum tubes (if not already marked) to show the patient the minimum amount of sputum needed (5 to 10 ml is an adequate amount).
2. Review with the patient how to collect sputum.
3. Make arrangements for a healthcare worker to pick up the specimen or for the patient, a family member, or a friend to drop off the specimen.

Induced Sputum Collection at a Healthcare Facility

If the patient cannot produce sputum spontaneously, then make arrangements for induced sputum to be collected at a facility. Facilities where sputum can be collected include the respiratory therapy department of a local hospital or TB clinic. Facilities should have appropriate respiratory protection, environmental controls, and policies and procedures.

How to Collect Gastric Aspirates

The following are basic guidelines for collecting gastric aspirates:

- Collect the specimen after the patient has fasted for 8 to 10 hours and, preferably, while the patient is still in bed.
- Put sample into 50 mL conical tube with 100 mg of sodium carbonate preservative.
- Collect a specimen daily for three days.

For additional information on how to collect a gastric aspirate and prepare the specimen for transport, see the guide and Francis J. Curry International Tuberculosis Center's online video *Pediatric TB: A Guide to the Gastric Aspirate (GA) Procedure* at http://www.currytbcenter.ucsf.edu/topics-interest/pediatric-tb .

Bronchoscopy or Collection of Extrapulmonary Specimens

Physicians who plan to collect (extrapulmonary) specimens should send part of the specimen (not in formalin) to the microbiology laboratory to be forwarded to the ASPHL for acid-fast bacilli (AFB) smear and culture, in addition to any other tests or pathology examinations the physician plans to obtain. A post-bronchoscopy sputum specimen should be sent for AFB smear and culture.

- **Bronchoscopy:** Refer the patient to a local specialist.
- **Extrapulmonary specimens:** These specimens will be collected by the physician performing the diagnostic work-up.

Specimen Shipment

There are three main categories of transportation methods: medical couriers, ground transportation, and air transportation. Category B Infectious Substances (raw diagnostic specimens, such as sputum, blood, or tissue) can be mailed through the US Postal Service (USPS), air shipped by private carrier (e.g., Federal Express, Airborne Express, Gold Streak, etc.), or transported by a medical courier. Specimens collected for AFB smear and culture are Category B Infectious Substances (raw diagnostic specimens, such as sputum, blood, or tissue) and should be labeled and shipped as such.

Shipment of dangerous goods by USPS is regulated by the US Department of Transportation. Specific shipping instructions from the Centers for Disease Control and Prevention (CDC) can be found in the publication by the US Department of Health and Human Services (DHHS) *Public Health Mycobacteriology: A Guide for the Level III Laboratory.* Packaging and shipment of specimens by USPS should meet the following regulations:

- Public Health Service/CDC: 42 CFR, Part 72—Interstate Shipment of Etiologic Agents at http://www.cdc.gov/od/ohs/biosfty/shipregs.htm
- USPS: 39 CFR and USPS Domestic Mail Manual C023.1.1, International Mail Manual 135, and USPS Publication 52
- US Department of Transportation: 49 CFR, Parts 171–180 (August 14, 2002) at http://www.access.gpo.gov/nara/cfr/waisidx_04/49cfrv2_04.html
- The Department of Labor, Occupational Safety and Health Administration (OSHA): 29 CFR 1910.1030[9]

For shipments by private carriers, follow International Air Transportation Association (IATA) instructions. *Mycobacterium tuberculosis* pure cultures are defined as infectious substances/etiologic agents when shipped by private carrier and must be shipped in packaging approved by the United Nations (UN), according to IATA Packing Instruction 602. Diagnostic specimens are defined as human or animal specimens, including excreta, secreta, blood and its components, tissue, tissue fluids, and cultures of nontuberculous mycobacteria being transported for diagnostic or investigational purposes. Diagnostic specimens must be packaged according to IATA Packing Instruction 650.[10]

Specimens must be shipped according to current federal, state and local laws. Upon request, Alaska State Public Health Laboratory provides ambient temperature shipping boxes that meet current shipping regulations.

Refer to the shipping regulations that are listed under “Resources and References” at the end of this section. Personnel who handle, package, and ship infectious materials must be trained in these procedures.

For more information, contact ASPHL Lab at 907-334-2100. To request lab supplies, visit the website: http://dhss.alaska.gov/dph/Labs/Pages/publications/default.aspx

Use the *Anchorage Public Health Laboratory Request Form* to request tuberculosis laboratory services from the Alaska State Public Health Laboratory in Anchorage. It is available in the Forms section of the manual **18.1**.

To obtain specimen collection and transport supplies, see the topic on "Specimen Collection and Shipment Supplies" in the Supplies, Materials, and Services section **16.1**.

Resources and References

Resources for Laboratory Services

Detailed descriptions of recommended laboratory tests; recommendations for their correct use; and methods for collecting, handling, and transporting specimens have been published.

For more information on laboratory testing for tuberculosis (TB), see the following:

- ATS, CDC, IDSA. "Controlling Tuberculosis in the United States: Recommendations from the American Thoracic Society, CDC, and the Infectious Diseases Society of America" (*MMWR* 2005;54[No. RR-12]). Available at: http://www.cdc.gov/mmwr/PDF/rr/rr5412.pdf .
- ATS, CDC, IDSA. Diagnosis of Tuberculosis in Adults and Children. *Clinical Infectious Diseases 2017*; 64(2):1-33. Available at: https://www.cdc.gov/tb/publications/guidelines/pdf/cid_ciw694_full.pdf
- National Committee for Clinical Laboratory Standards. *Susceptibility Testing of Mycobacteria, Nocardiae, and Other Aerobic Actinomycetes; Approved Standard* [Document no. M24-A] (Wayne, PA; 2003).

Resources for Specimen Collection and Shipment

- CDC. "Guidelines for Preventing the Transmission of *Mycobacterium tuberculosis* in Health-care Settings, 2005" (*MMWR* 2005;54[No. RR-17]). Available at: http://www.cdc.gov/mmwr/pdf/rr/rr5417.pdf .
- CDC. *Public Health Mycobacteriology: A Guide for the Level III Laboratory* (Atlanta, GA; 1985).
- Francis J. Curry International Tuberculosis Center. *Pediatric TB: A Guide to the Gastric Aspirate (GA) Procedure* (Francis J. Curry National Tuberculosis Center Web site). Available at: http://www.nationaltbcenter.edu/products/product_details.cfm?productID=ONL-06 .
- International Air Transport Association (IATA). IATA Web site. Available at: http://www.iata.org/index.htm .
- National Jewish Medical and Research Center. *How to Mail Specimens and Cultures to the National Jewish Mycobacteriology Laboratory* (Denver, CO: March 2005).
- National Jewish Medical and Research Center. *Instructions (for Patients) for Collecting and Mailing Sputum Specimens* (Denver, CO: March 2005).
- National Tuberculosis Controllers Association—National Tuberculosis Nurse Consultant Coalition. *Tuberculosis Nursing: A Comprehensive Guide to Patient Care* (Atlanta, GA; 1997):39–42.

- US Department of Transportation. Hazardous Materials: Revision to standards for infectious substances. Part III 49 CFR Part 171. Federal Register (August 14, 2002).
- USPS. *Mailing Standards of the United States Postal Service: Domestic Mail Manual* (USPS Web site). *Available* at: http://pe.usps.com/ .

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[2] ATS, CDC, IDSA. Controlling tuberculosis in the United States: recommendations from the American Thoracic Society, CDC, and the Infectious Diseases Society of America. *MMWR* 2005;54(No. RR-12):19.

3 APHL, The Future of Tuberculosis Laboratory Services: A Framework for Integration/Collaboration/Leadership. 2004. Available at: https://stacks.cdc.gov/view/cdc/11399

[4] CDC. Diagnostic microbiology. In: Chapter 5: Treatment of LTBI. *Core Curriculum on Tuberculosis (2013)* [Division of Tuberculosis Elimination Web site]. Updated 2013. Available at: https://www.cdc.gov/tb/education/corecurr/pdf/chapter5.pdf . Accessed January 18, 2017.

[5] ATS, CDC, IDSA. Diagnosis of Tuberculosis in Adults and Children. *Clinical Infectious Diseases 2017*; 64(2):1-33.

[6] Iseman, MD. *A Clinician's Guide to Tuberculosis, 2000.* 1st ed. Philadelphia, PA: Williams & Wilkins; 2000:135–136.

[7] Iseman, MD. *A Clinician's Guide to Tuberculosis, 2000.* 1st ed. Philadelphia, PA: Williams & Wilkins; 2000:135–136.

[8] Iseman, MD. *A Clinician's Guide to Tuberculosis, 2000.* 1st ed. Philadelphia, PA: Williams & Wilkins; 2000:135–136.

[9] National Jewish Medical and Research Center. *How to Mail Specimens and Cultures to the National Jewish Mycobacteriology Laboratory*. Denver, CO; March 2005:2.

[10] National Jewish Medical and Research Center. *How to Mail Specimens and Cultures to the National Jewish Mycobacteriology Laboratory*. Denver, CO; March 2005:5–7.