Tuberculosis (*Mycobacterium tuberculosis*)

The Oregon State Public Health Laboratory (OSPHL) will test appropriately submitted specimens from patients suspected of having tuberculosis. The OSPHL will test specimens submitted to help monitor therapy of previously diagnosed TB patients or to aid in contact investigations.
The OSPHL accepts primary specimens from County Health Programs and their affiliates. Primary specimens from Private Care facilities will be accepted at the request of a State or County TB Public Health program. In addition to *M. tuberculosis* isolates submitted to this laboratory, in compliance with OAR 333-018-0018, the OSPHL will accept unidentified AFB smear positive isolates to rule out *M. tuberculosis*. Susceptibility testing will be performed on all confirmed *M. tuberculosis* isolates. Private Laboratories may submit NALC-NaOH processed sputum pellets for nucleic acid amplification testing (NAAT). Private sector submitters will be charged a fee for this service. Submitters should contact the OSPHL 503-693-4100 for additional billing information.

Primary Specimens (Local Health Departments)

An AFB smear and culture is routinely performed on all submitted primary specimens. Additionally, a NAAT will be performed on at least one respiratory sample from a previously undiagnosed patient. The NAAT performed by the OSPHL is the Cepheid GeneXpert® MTB/RIF assay. Cepheid GeneXpert® testing is FDA cleared for AFB smear positive and smear negative respiratory samples from patients not on anti-tuberculosis therapy (TB-Rx) and who have a presentation consistent with active pulmonary tuberculosis. Patients with less than 72 hours of TB-Rx or who have not had TB-Rx in the previous 12 months may be tested.

Testing is always performed in conjunction with AFB smear and culture. If NAAT is not requested, this should be indicated on the Test Request Form. Repeat testing of additional samples from a specific patient may occur in the following instances:

- The smear is negative and the GeneXpert detects MTB.
- The smear is positive and the GeneXpert does not detect MTB.
- The initial smear is negative and the GeneXpert does not detect MTB, but a later specimen on the same patient is smear positive.

Submitting Samples

Three to five AFB negative specimens, collected on consecutive days, may be required to assure a negative diagnosis. However, one AFB positive specimen may be sufficient to confirm a suspect diagnosis. Do not pool specimens.

Collection Instructions

All required collection and shipping materials are available from the OSPHL.

1. **Expectorated sputum**
   a. The best specimen is the first deep sputum obtained in the morning after clearing the saliva from the mouth.
   b. Collect a series of three to five single, early morning samples in the collection tubes provided. Use a separate tube for each sample. **A volume of 5 to 10 mL is Optimal. However, smaller volumes will be accepted. Results from samples of less than 1.0 mL will be reported with a qualifier.**

c. Securely cap tubes to avoid leakage.
d. Decontaminate outside of specimen tube with appropriate disinfectant.
e. Label tube with two patient identifiers and collection date.
f. Complete and submit a Test Request Form for each specimen.
g. Refrigerate specimens pending shipment to OSPHL.

2. Bronchial wash/induced sputa

There is no minimum volume for this specimen type. Follow same guidelines as for expectorated sputum.

3. Gastric lavage

a. Specimens which cannot be processed within four hours of collection must be neutralized. For each 35 – 50 mL of specimen, use 1.5 mL of sterile 40% anhydrous disodium phosphate (Na_2HPO_4) or two pH 7.4 pHydrion buffer capsules or tablets.
b. Secure lid against leakage and label specimen tube with two patient identifiers, and collection date.
c. Complete and submit a Test Request Form for each specimen.
d. Store refrigerated pending shipment.

4. Urine

A three-to-five day series of single, early morning, mid-stream urines should be submitted. Do not submit pooled specimens.

a. Aseptically transfer up to 35 mL of urine to provided tube.
b. Secure lid against leakage and label collection tube with two patient identifiers and collection date.
c. Complete and submit a Test Request Form for each specimen.
d. Store refrigerated pending shipment.

5. Body fluids (CSF, thoracentesis, pleural fluid, synovial fluid, etc.)

a. Aseptically transfer specimen to plastic specimen tube. There is no minimum volume.
b. Secure lid and label collection tube with two patient identifiers, and collection date.
c. Complete and submit a Test Request Form for each specimen.
d. Store refrigerated pending shipment.

6. Tissue

a. Aseptically transfer specimen, along with a minimal amount of sterile saline as needed to prevent desiccation, to sterile plastic tube.
b. Secure lid against leakage and label tube with two patient identifiers, specimen source, and date of collection.
c. Complete and submit a Test Request Form for each specimen.
d. Store refrigerated pending shipment.
e. Transport to OSPHL on wet ice or ice packs as soon as possible. Call the OSPHL for additional packaging and shipping instructions as needed.

7. Wounds and lesions

a. Aseptically obtain purulent material with a sterile swab or by aspiration or washing.
b. Aseptically transfer specimen to a sterile tube. Add a small amount of 7H9, 7H11 or sterile isotonic saline to prevent specimen dehydration.
c. Secure lid against leakage and label specimen tube with two patient identifiers, specimen source and collection date.
d. Complete and submit a Test Request Form for each specimen.
e. Store refrigerated pending shipment.

Precautions

1. All specimens should be kept refrigerated pending shipment.
2. Do not pool specimens. Transport each specimen as soon as possible after collection.
3. Whenever possible, collect and ship specimens early in the week.
4. Ship samples according to current federal regulations. Refer to the Shipping and Transport section of this Guide to Services.

Note: Improperly packaged or leaky specimens present a hazard and may be rejected.
Note: Isolates of *M. tuberculosis* are classified as Category A for shipping.

Private Care Laboratories

Secondary Specimens: AFB Isolates submitted for identification/ confirmation of organism ID and/or TB susceptibility testing

1. Submit AFB culture isolates on Lowenstein-Jensen, 7H10, or other comparable solid AFB media. Failure to submit "pure" cultures may delay or adversely affect the identification process.
2. AFB "Positive" liquid media tubes from automated TB systems, such as MGIT or BTA, will be accepted for testing.
   Note: these liquid media culture results can be adversely affected by inadequate organism load (potential false negatives) or blood in the media (potential false positives). Cultures with mixed AFB organisms or cultures with bacterial contamination may yield misleading results
3. Evaluate DNA probe results from liquid culture with caution. Results should be correlated with specimen, smear/culture morphology, and available patient data, before releasing patient results to charting system.
4. Submitters should retain subcultures of all submissions as a backup precaution.

Processed concentrates of primary sputum pellets for NAAT testing

1. Submitters must process samples using CDC NALC-NaOH decontamination methodology
2. A minimum of 0.5 mL of the processed/concentrated pellet (stored and shipped at 2-8ºC) should be submitted to the OSPHL for receipt within 2 days of the processing date. Friday shipments must be received by noon.
3. Ship according to Category B regulations.
4. Private care facilities will be assessed a fee for this test. Please contact the OSPHL for billing information or if additional information or guidance is required, (503) 693-4100.

AFB Culture Test Results Reporting

First Preliminary Report (AFB smear report)

1. Acid Fast Stains are reported within 24 hours of receipt.

2. Initial positive results for new patients are called or faxed on day of test.
3. Faxed reports for all submissions are generated and sent within one business day.
4. Submitters should check Copia then contact the OSPHL if a report has not been received within 5 days of specimen receipt.

Second Preliminary Report (GeneXpert/NAAT, tested primary samples)

1. Positive GeneXpert results are called or faxed on day of test. GeneXpert testing is performed five days a week or as needed.
2. GeneXpert reports are faxed within one business day.
3. Submitters should check Copia then contact the OSPHL if a report is not received within 5 days of testing.

Note: Cultures are monitored for up to 8 weeks before being reported as culture negative for AFB.

Additional Preliminary Report (Culture Results)

1. Additional preliminary reports are generated for any culture growing acid fast bacilli. This report will include organism identification as either *M. tuberculosis* complex or *M. avium* complex by the Hologic DNA-Probe.
2. No additional identification tests are performed on AFB organisms testing negative for either *M. tuberculosis* complex or *M. avium* complex. If further identification is needed, contact the OSPHL. The OSPHL may be able to forward organism to a reference laboratory for further identification.
3. Private care laboratories may submit isolates for DNA-probe ID as described in the isolate submission section.
4. All results of *M. tuberculosis* complex from new patients will be called to submitter on day of test.

Final Culture Report

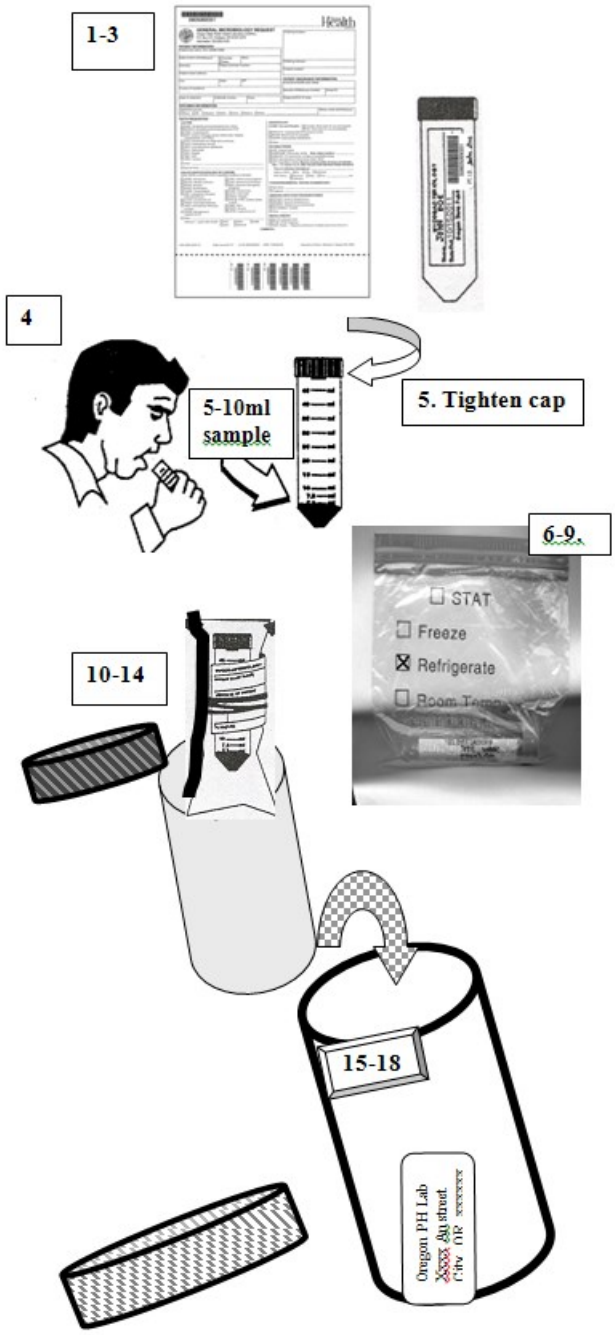
1. Cultures with no growth at eight weeks are reported as “AFB (acid fast bacilli) NOT ISOLATED”.
2. Final reports for positive cultures will include colony count information if available.

Antibiotic Susceptibility Testing

1. Antibiotic susceptibility testing is performed on initial TB isolates from new patient cases.
2. Susceptibility testing is repeated if a patient’s additional samples are TB culture positive 3 months or more after the original susceptibility testing was performed.
3. Additional susceptibility tests will be done on request. Antibiotic susceptibility testing of *M. tuberculosis* isolates is available to all submitters.
4. Preliminary Susceptibility results of MGIT I.R.E. and final MGIT PZA testing are typically available within two to three weeks following culture identification of *M. tuberculosis*.
5. Final Indirect Susceptibility results are usually available four to five weeks following organism identification.
6. “Resistant” susceptibility results are reported as soon as possible after completion of test. Results may be accessed via Copia. All hard copy results are faxed within 24 hours of test completion.

Specimen Packaging and Shipping

1. OSPHL provides complete kits for packaging and shipping of tuberculosis test specimens.
2. Label specimen with two patient identifiers and collection date.
3. Complete all sections of the Test Request Form.
4. Attach one of the OSPHL bar-coded Test Request Form labels to the specimen.
5. Transfer specimen to collection tube.
6. Secure the lid with adhesive tape to prevent leakage during transport.
7. Wrap the tube with the pad of absorbent material.
8. Place the tube sideways into the plastic bag.
9. Expel the air from the bag.
10. Close bag by zipping the top seal.
11. Put the Test Request Form into outer bag pocket.
12. Wrap bag around tube.
13. Place the sealed bag and slip into the inner metal tube which must bear a biohazard label.
14. Screw the lid onto the container.
15. Make sure lid is secured.
16. Place the metal tube into the outer shipping container.
17. Make sure the lid is secured.
18. Refrigerate specimen pending shipment to OSPHL.
19. Send specimens to the OSPHL as soon as possible after collection. Specimens should be received within 5 days of collection.



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