Assessing quality-assured diagnoses made by TB laboratories

Quality-assured TB laboratory

Objectives: at the end of the assessment reviewers should comment on –

- the laboratory network and its structure;
- whether WHO's recommended diagnostic technologies are used, and whether there is a quality-assurance programme;
- laboratory performance analysis;
- whether there are procedures to ensure that staff take part in continuing education activities;
- the procurement of laboratory equipment and supplies;
- the safety measures and practices;
- data recording and reporting;
- the strengths and weaknesses of the laboratory network, and any improvements needed.

Background:

Sputum specimens should be obtained for microscopic examination from all patients suspected of having pulmonary TB. In most settings, the diagnosis of TB is confirmed by the presence of acid-fast bacilli (AFB) by sputum-smear examination. Sputum-smear microscopy is the most widely used method for detecting TB, but it has a number of drawbacks, including low sensitivity (especially for people who are HIV-positive and for children) and an inability to determine whether the organism is resistant to anti-TB medicines. The conventional diagnosis of drug-resistant TB relies on bacterial culture and drug-susceptibility testing (DST), slow and cumbersome processes. While awaiting the results of culture and DST, patients may be inappropriately treated, resistant strains may continue to spread, and drug resistance may become amplified.

HIV infection is responsible for the large increase in the proportion of patients with smear-negative pulmonary TB and extrapulmonary TB. Thus, in settings where the prevalence of HIV is high and MDR-TB is significantly present, it is necessary for diagnostic algorithms to include conventional and rapid diagnostic technologies (such as molecular line probe assays and the Xpert MTB/RIF test [Cepheid, Sunnyvale, CA]).

When feasible, laboratory services for microscopy, culture and rapid DST should be introduced in a phased manner at the appropriate referral levels of the health system. In settings where comprehensive DST is not feasible, the diagnosis of drug-

resistant TB requires surveillance of new patients, retreatment patients and other high-risk groups to collect population-representative data on drug resistance. Depending on a country's capacities and strategies for introducing the use of new and rapid diagnostic technologies, sputum samples may be referred to a higher-level laboratory where those technologies are used (that is, molecular testing such as the line probe assay or liquid culture); in most cases these will be the regional reference laboratory and the central reference laboratory.

Location: microscopy centre or intermediate-level laboratory facility

Staff to be interviewed: laboratory manager or director, laboratory technicians

Assessment

a. What are the laboratory's general operations like? What is the workload? What tasks are performed at the laboratory, and how many staff are there?

    i. What is the laboratory's catchment area and the population served?

    ii. How many technicians work in the laboratory, and what qualifications do they have? What is their workload? What proportion of time does each technician devote to processing AFB smears?

    iii. How many microscopy laboratories are there per population of the catchment area? How many sputum specimens are processed daily, weekly and monthly?

    iv. How many smears are performed during each reporting period?

    v. On average how many smear-microscopy procedures are performed by each laboratory technician each day?

    vi. Have the technicians received special training on sputum collection and smear microscopy? If yes, when was the training and who provided it?

    vii. (Observe general biosafety conditions in the laboratory.) Is the airflow appropriately directed? Are the smears prepared and stained, and reagents stored, in an appropriate area?

b. Are national laboratory guidelines and standard operating procedures for sputum processing available and used? (If yes, ask to see a copy.) Were the national guidelines prepared by the national reference laboratory?

c. Are laboratory technicians or staff responsible for collecting sputum from patients? If yes,

    i. (Review the procedures for collecting specimens.) Are specimens collected using the "spot, early morning, spot" approach or the "spot, early morning" approach? (Please describe the procedure or attach the standard operating procedures to the report.)

ii. Where is sputum collected? Observe whether sputum is collected in well ventilated areas or outside the building.

iii. Are sputum-collection containers adequate? How are they labelled?

iv. Who is responsible for instructing people suspected to have TB on how to produce sputum? (Observe how sputum samples are collected from patients or ask staff about the procedures for collecting samples.) Are proper instructions provided to all suspected TB cases?

d. Does the laboratory receive sputum specimens from an outpatient clinic or facility? If yes,

i. How is the specimen delivered to the laboratory? How is the specimen stored before being sent to the laboratory? How long, on average, is the specimen stored before being sent to the laboratory? How long does it take for the specimen to reach the laboratory?

ii. How long does it usually take for results to be reported once a specimen has been received?

iii. How are results reported to the clinic or the health-care facility?

e. What type of equipment is used?

i. What type of microscope is used: light or fluorescent? If a light microscope is used, is it of good quality? How old is it? Is it monocular or binocular? What is the light source? (Ask the technician to show you an AFB-positive slide, and observe it under the microscope to assess the quality of the microscope.)

ii. Are microscopy slides reused? (Observe how the slides are labelled and stored.) How long are slides kept? What types of slides (smear-negative or smear-positive) are kept for quality control, and for how long are they kept?

f. Are the reagents of good quality? Is the supply adequate? (Review the quality and supply of the reagents.) Are the reagents prepared centrally or locally?(Ask staff to describe the process of preparing the reagents.)

g. Is the laboratory register complete?

i. Have all smear examinations performed in the laboratory been recorded in the laboratory register?

ii. Have the following fields been included in the register and have they been completed: date the specimen was received, name and address of the patient, name of the referring facility, reason for sputum-smear microscopy, result, basic management unit and TB laboratory register number (for newly diagnosed and registered

patients)?

iii. Does each patient have at least two (or three) results from the sputum-smear examination?

iv. If light microscopy is used, are the results quantified properly? They should be reported as: Negative if 0 AFB/100 fields; 1–9 if 1–9 AFB/100 fields; 1+ if 10–99 AFB/100 fields; 2+ if 1–10 AFB/ field; 3+ if >10 AFB/ field in 20 fields.

v. If fluorescence microscopy is used are the results reported properly? They should be reported as shown in the table below.

If a 200x objective is used	**If a 400x objective is used**	**Results to be reported**
No AFB in 1 length	No AFB in 1 length	No AFB observed
5–9 AFB in 1 length	3–24 AFB in 1 length	Scanty
50–499 AFB in 1 length	25–249 AFB in 1 length	1+
25–250 AFB per field	12–125 AFB per field	2+
250 AFB per field	125 AFB per field	3+

AFB, acid-fast bacilli.

vi. What is the workload like? (To measure changes in the workload, count the number of smears tested during the past month and compare the total with the number of smears tested during the same month in the previous year. If there is a significant difference, ask the laboratory technician about the possible reasons for the difference.)

vii. Does the laboratory participate in a quality assurance programme?(If so, describe the programme.) Is feedback on the results of quality assurance testing provided to technicians in a timely manner? (Review any recent results. Determine whether additional training or supervision is offered to technicians who have less-than-adequate results.)

For laboratories performing solid or liquid culture, rapid diagnostic tests (such as, the Xpert MTB/RIF or line probe assay) or culture and DST –

[Note: If the reviewer visits a laboratory that performs culture and DST, the following general questions may be asked in preparation for a more thorough review by an expert at a later time. Are any other tests besides smear microscopy or culture and DST being implemented. If yes, which ones?]

h. What is the general organization of the laboratory? What is the link with the national reference laboratory or the supranational reference laboratory?

i. How many technicians have been designated to perform culture tests? How many perform DST? Have the technicians received special training on culture and DST? If yes, when was the training provided and who provided it?

j. Are the environmental conditions in the laboratory adequate? (Review the laboratory's infrastructure and environmental conditions, including water sources, electricity supply, general maintenance, safety and the management of waste.)

k. What is the quantity and quality of equipment, supplies and reagents?

l. Are there contracts and logbooks to record when equipment is serviced? (If yes, review them.)

m. How many laboratories perform TB culture? At which level of the health-care services are the culture laboratories? What is the number of culture laboratories per population?

n. How many laboratories perform DST? At which level of the health-care services is DST performed? What is the number of laboratories performing DST per population?

o. Are standard operating procedures for culture and DST available? If yes, were they developed by the national reference laboratory?

p. How many cultures were performed during the past quarter (or other relevant period of time)? How many cultures were positive? What proportion of TB cases is confirmed by culture?

q. How many DSTs were performed during the past quarter (or other relevant period of time)? How many DSTs were conclusive?

r. Are machines available for Xpert MTB/RIF testing? If yes, at which level of the health-care system are they? Are the indications for using the Xpert MTB/RIF test well defined? Is there a clear algorithm or standard operating procedure for staff to follow when using the Xpert MTB/RIF test?

s. How many Xpert MTB/RIF tests were performed during the past quarter (or other relevant period of time)? How many Xpert MTB/RIF tests were positive for TB and how many were positive for rifampicin resistance?

t. What infection-control procedures are in place?

   i. How is infection control handled during the transportation and management of specimens?

   ii. How well are biosafety cabinets and culture rooms maintained?

   iii. Is ventilation appropriate, with negative pressure where necessary? Are high-efficiency particulate air (HEPA) filters used and maintained?

   iv. Are respirators and masks stocked for personal protection ?

u. Does the national reference laboratory provide external quality assurance? (Describe these procedures.)

Indicators for: **Quality-assured diagnoses by TB laboratories**		
Indicator	***Calculation***	***Source of information***
Number of TB microscopy laboratories to the population	Numerator: number of TB microscopy laboratories Denominator: population	Routine reports on the national TB programme's resources, official demographic data
Workload of each microscopist at the TB laboratory	Number of slides examined each day by each laboratory technician (the average number can be calculated for the past week, past month or past quarter, and compared with the average calculated for the same period of the previous year)	TB microscopy laboratory register
Proportion of TB microscopy laboratories participating in an external quality assurance programme	Numerator: number of TB microscopy laboratories participating in an external quality assurance programme Denominator: total number of TB microscopy laboratories	Routine reports on the management of the TB laboratory network
	Numerator: TB microscopy	Routine reports on

Proportion of TB microscopy laboratories with acceptable proficiencies as assessed by external quality assurance	laboratories with acceptable proficiencies during evaluation by external quality assurance (for example, no more than 2 major errors by a laboratory each year) Denominator: number of TB microscopy laboratories participating in an external quality assurance programme	external quality assurance evaluations
Proportion of microscopy slides that are smear-positive	Numerator: number of smear-positive slides identified during a specified period Denominator: total number of slides tested for AFB during the same period	TB microscopy laboratory register
Proportion of positive cultures	Numerator: number of positive cultures identified during a specified period Denominator: total number of cultures performed during the same period	Culture laboratory register
Proportion of contaminated cultures	Numerator: number of contaminated cultures identified during a specified period Denominator: total number of cultures performed during the same period	Culture laboratory register
Proportion of Xpert MTB/RIF tests[a] positive for *Mycobacterium tuberculosis*	Numerator: number of Xpert MTB/RIF tests positive for *M. tuberculosis* during a specified period Denominator: total number of	Xpert machine register

	Xpert MTB/RIF tests performed during the same period	
Proportion of Xpert MTB/RIF tests showing resistance to rifampicin	Numerator: number of Xpert MTB/RIF tests positive for *M. tuberculosis* and showing resistance to rifampicin during a specified period Denominator: total number of Xpert MTB/RIF tests positive for *M. tuberculosis* during the same period	Xpert machine register
Proportion of DSTs showing resistance to rifampicin (to be reported separately for new cases and previously diagnosed cases)	Numerator: number of DSTs showing resistance to at least rifampicin during a specified period Denominator: total number of DSTs in which susceptibility to rifampicin was tested during the same period	DST register

AFB, acid-fast bacilli; DST, drug-susceptibility testing.
[a] Xpert MTB/RIF test manufactured by Cepheid, Sunnyvale, CA.