SRM MEDICAL COLLEGE HOSPITAL AND RESEARCH CENTRE

KATTANKULATHUR

HANDBOOK ON TUBERCULOSIS

DEPARTMENT OF MICROBIOLOGY

HOSPITAL INFECTION CONTROL COMMITTEE

Prepared By
Dr. Shweta Sagar Naik
Dr. Manjula S.R.

Authorized By
Dr K.V. Leela,
HOD (Microbiology)

Approved By
Dr A. Sundaram, Dean
Dr Nalini Jayanthi, HOD (Respiratory medicine)

INDEX

Scope:

The aim of this handbook is to provide the basic information for TB diagnosis and highlight the options available for TB laboratory diagnosis,

Introduction:

Tuberculosis (TB) is an infectious disease caused by Mycobacterium tuberculosis. Tuberculosis commonly affects the lungs, but can also affect other parts of the body. About a quarter of the world's population is infected with M. tuberculosis. TB can affect anyone anywhere, but most people who develop the disease are adults, there are more cases among men than women, and 30 high TB burden countries account for almost 90% of those who fall sick with TB each year.

Sign and symptoms:

Tuberculosis most commonly affects the lungs. Common symptoms of active lung TB are:

- A persistent cough of more than two weeks that brings up phlegm and blood at times
- Breathlessness, which is usually mild to begin with and gradually gets worse
- Lack of appetite and weight loss
- A high temperature of 38ºC (100.4ºF) or above
- Extreme tiredness or fatigue
- Night sweats

Less commonly TB infection can occur in other organs of the body, as :Lymph nodes, bones and joints, digestive system, nervous system, bladder and reproductive system. This is known as extra pulmonary TB. People who have latent TB infection do not feel sick, do not have any symptoms, and cannot spread TB to others

DIAGNOSTICS:

Diagnostics are needed to detect exposure, diagnose disease and drug resistance, and monitor improvement under treatment in both primary healthcare and laboratory settings. High accuracy, short time to obtain a result, and simple use at an affordable price can be significant challenges for current TB diagnostic tools.

Physical examination:

Physical examination findings associated with TB depend on the organs involved. Patients with pulmonary TB have abnormal breath sounds, especially over the upper lobes or involved areas. Rales or bronchial breath signs may be noted, indicating lung consolidation

Radiology :Chest X ray/CT scan

Chest X-ray (CXR) has historically been one of the primary tools for detecting tuberculosis (TB), especially pulmonary TB. CXR has high sensitivity for pulmonary TB and thus is a valuable tool to identify TB as a differential diagnosis for patients, especially when the X-ray is read to identify any abnormality that is consistent with TB. However, CXR has poor specificity; as many CXR abnormalities that are consistent with pulmonary TB are seen also in several other lung pathologies. Moreover, there is significant intra- and

inter-observer variation in the reading of CXRs. Relying only on CXR for TB diagnosis leads to over diagnosis, as well as under diagnosis. Rigorous efforts should always be made to base a TB diagnosis on bacteriological confirmation (sputum-smear microscopy, culture or a molecular test)

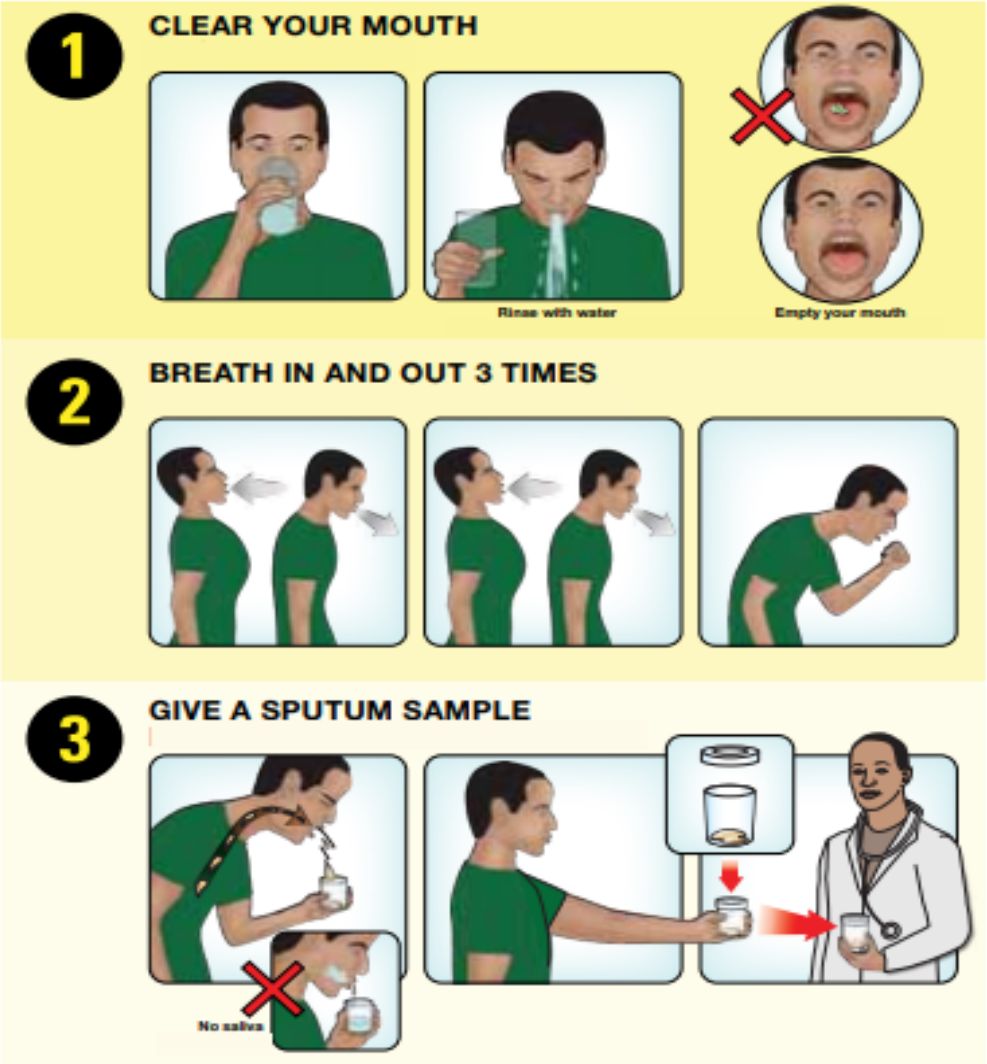
LABORATORY DIAGNOSIS:

A high-quality laboratory system that uses modern diagnostics is a prerequisite for the early, rapid and accurate detection of TB and drug resistance.

SPECIMEN COLLECTION:

Sputum Collection

Specimens should be collected in a well ventilated area, away from the general sample collection area/ washrooms.



SPECIMEN	METHOD OF COLLECTION	TIMINGS	CONTAINER
PULMONARY			
Sputum	Deep cough, thick, mucoid, white-yellow	Two samples, early morning and spot OR two spot one hour apart	Sterile container
Bronchial alveolar lavage (BAL)	Aseptic collection, minimum volume 20-50 ml	Any time	Sterile container
Bronchial secretions	Aseptic collection, minimum volume 2-5 ml	Any time	Sterile container
Gastric lavage	Aspiration of the gastric content	Early morning	Sterile container
EXTRAPULMONARY			
Body fluids • Spinal • Pleural • Pericardial • Synovial • Ascitic	Aseptic collection by aspiration techniques or surgical procedures.	Any time	Sterile container
Tissue	Aseptic collection by surgical procedures.	Any time	Sterile container with normal saline (0.9%) Never collect in formalin.
Urine	Adequate cleansing of external genitalia prior to collection	Three early morning samples, 500 ml	Sterile container
Bone marrow	Aseptic method of aspiration	Any time	Heparin vacutainer
Swabs	Sub optimal specimens and not recommended		
Blood	Not recommended as low diagnostic yield and high possibility of contamination		

Specimens should be transported to the laboratory as soon as possible after collection with Specimen Requisition Form. If delay is unavoidable, the specimens should be refrigerated 2-8^{0}C to inhibit the growth of other unwanted micro-organisms .Sputum samples can be transported from PHCs in icepacks.

ACID-FAST BACILLI MICROSCOPY (AFB) PREPARATION AND STAINING

The purpose of AFB microscopy is to detect acid-fast bacilli (AFB) by microscopic examination of clinical specimens and cultures. Both living and dead (viable and non-viable) bacilli will stain and be counted. A semi-quantitative grading system is used to report the number of AFB observed in stained smears.

These smears are stained with Ziehl-Neelsen stain or fluorescent stains, either auramine O or auramine/rhodamine. The Ziehl-Neelsen stains acid-fast organisms pink and the background debris stains blue. A positive smear is approximately 10^4 bacilli per ml or greater. With fluorescent stain, organisms fluoresce bright yellow/orange red against a black background. The most important advantage of the fluorescence technique is that slides can be examined at a lower magnification, thereby minimizing the

time required and increasing the sensitivity by 10%. Sputum smear microscopy is currently most widely used first test for diagnosis: it is inexpensive and widely available but not very accurate – and children and HIV+ people often cannot produce a sputum sample.

MOLECULAR TESTS: Molecular tests provide an accurate result for diagnosis and drug resistance in hours has been a great step forward, but it requires infrastructure and is often not affordable in many low-resource settings.

Real-time polymerase chain reaction (PCR) assays: The biosafety precautions required for these tests are similar to those for smear microscopy and allows the use of the assay outside of conventional laboratories. Training requirements are minimal.

Platform	Xpert MTB/RIF® (Ultra)	Truenat™
Manufacturer	Cepheid, US	Molbio, India
Principle	Cartridgebased)	Chip-based
Steps	Single step	3 steps: 1. Sample preparation, 2.Mtb assay, 3.If positive: Rif assay
Time taken	Xpert: 110 min Xpert ultra: 80 min	Sample preparation: Rif assay: 1 hr
Specimens	Sputum, on processed sputum sediment and on selected extrapulmonary specimens (Pus, CSF) Low sensitivity in other fluids	Sputum, BAL, extrapulmonary fluids and tissues
Sensitivity	Xpert:131 cfu/ml Xpert Ultra:13.6 cfu/ml Ultra is less specific, more sensitive	100 cfu/ml
Electric supply	Continuous	Battery operated

Other methods of detection:

METHOD	PRINCIPLE	TAT	ADVANTAGE
CULTURE	Growth of bacteria can be observed by culturing on special solid or liquid culture media. Gold standard.		
Solid media	LJ media slants	8 weeks	Economical, discrete colonies seen, less contamination
Liquid media	MGIT 320: Fluorogenic	6weeks	Shorter TAT, more yield

Drug susceptibility (can be done on solid or liquid media)	• **First-line drugs:** streptomycin, isoniazid, rifampicin, ethambutoland pyrazinamide (SIREP). • **Second line drugs :**fluoroquinolones (gatifloxacin, levofloxacin, moxifloxacin, ofloxacin) ,injectable drugs (amikacin, capreomycin, kanamycin), Cycloserine, Ethionamide, PAS, clofazimine • **Third line drugs:** doubtful or unproven efficacy: rifabutin, Macrolides (clarithromycin), thioacetazone (T);thioridazine;Amoxixillin-clavulanate, Imipenem and meropenem. • **Newer TB drugs: Linezolid**, delamanid, bedaquiline : XDR TB cases.		
Urinary LAM	Lipoarabinomannan antigen detection by lateral flow	25 minutes	Less sensitive, Rapid
MOLECULAR TESTS	Detect the DNA		
Line probe assays (LPAs)	reverse hybridization detects mutations associated with drug resistance	5 hours	First and second line drug resistance detected in hours, less sensitive , Require PCR infrastructure/reference lab
Loop-mediated isothermal amplification (LAMP)	Target DNA is amplified at a fixed temperature	<1 hr	Resistance cannot be detected, test can be done at lower labs
Pyrosequencing:	Targeted sequencing by synthesis	<6 hrs	Gives presence of known drug resistance mutations, highly sensitive
Next-generation WGS	Whole Genome Sequencing (WGS)	Days	Shows the exact genotype including complete drug resistance profile

What is latent TB? What are the symptoms of latent TB?

Latent TB is the presence of M. tuberculosis in the body without any signs and symptoms, or radiographic or bacteriology evidence of TB. TB treatment is not recommended for latent TB. Only certain "target" groups including immunocompromised people who are at risk from progressing from latent to active TB should receive treatment.

Tests for latent TB: IGRAs and the TST are designed to detect latent TB infection. They are 'indirect tests' - they do not detect the actual TB bacilli but instead an immune response that suggests past or present exposure to TB bacilli. They are therefore expected to have poor specificity for active TB in high-burden settings.

	Mantoux skin test/TST	**Interferon gamma release assay (IGRA)**
Principle	Purified protein derivative injected intradermally shows induration which indicates immune response.	Indirectly measures the body's immune response to antigens derived from TB bacteria
Patient visit	Single	Two: 2nd after 48 hrs
Cost	Expensive, requires Laboratory infrastructure	Inexpensive, simple

BCG vaccination	No effect	False positive in younger patients

SPECIAL CONSIDERATIONS:

TB and PLHIV:

Worldwide, TB is one of the leading causes of death among people living with HIV. The Government of India is committed to ending TB by 2025, five years ahead of the global End TB target with National TB Elimination Programme (NTEP). This aims to provide universal access to TB care to achieve TB-free India with zero deaths and poverty due to TB. National AIDS Control Programme and NTEP have developed a "National framework of joint TB/HIV Collaborative Activities". As per this, all patients in TB clinics should be screened for HIV and rapid molecular test CBNAAT will be offered to all presumptive TB cases among PLHIV for early diagnosis of TB.

TB and COVID: The COVID-19 pandemic and TB – impact and implications

The COVID-19 pandemic threatens to reverse recent progress in reducing the global burden of TB disease due to health services disruption and economic impact of the pandemic, which will worsen at least two of the key determinants of TB incidence: GDP per capita and undernutrition. Negative impacts on essential TB services include the reallocation of human, financial and other resources from TB to the COVID-19 response.

TB and Pediatric:

Conventional TB diagnostics for children involved appropriate use of clinical details, chest radiology and tuberculin skin test, with much less focus on microbiology, due to poor yield (AFB smear) or access issues (MTb cultures). Universal DST strategy has led to change in the diagnostic pathways to include NAAT for every patient where a biological specimen can be procured. Routine chest imaging is done as initial screening test as testing of respiratory specimens from radiologically positive cases improves the yield of NAAT. The management of TB in children now has undergone a sea change with drug sensitivity directed therapy becoming the corner pillar. NTEP approved rapid NAAT has become the core investigative modality and erstwhile clinic-radiological approach of diagnosis is used only when NAAT fails in a clinically probable case.

TB in HCW:

Health care workers (HCWs) are at an increased risk of acquiring tuberculosis (TB) as well as DR-TB compared to the general population, especially in low-resource, high-TB-burden settings, where HCWs are in more frequent and prolonged contact with people in an infectious stage of active TB.

All HCWs should be screened for TB upon hire (i.e., preplacement) as well as annually. TB screening is a process that includes:

- Baseline TB risk assessment and TB symptom evaluation,
- Chest x-ray
- CBC/ESR
- Additional evaluation for TB disease as needed.

All health care personnel should receive TB education annually. TB education should include information on TB risk factors, the signs and symptoms of TB disease, and TB infection control policies and procedures.

TB infection control:

The organization of TB infection control at the level of health care setting can be considered as being in three parts: administrative controls, environmental controls, and respiratory protection controls.

Administrative measures should include:

- Promptly identifying people with TB symptoms (triage), separate infectious patients, control the spread of pathogens (cough etiquette and respiratory hygiene) and minimize time spent in health care facilities;
- Provide a package of prevention and care interventions for health workers, including HIV prevention, antiretroviral therapy, and isoniazid prevention therapy (IPT) for HIV positive health workers;
- Reduction of diagnostic delays, the use of rapid diagnostic tests, the reduction of turnaround time for sputum testing and culture, and the prompt initiation of treatment.

Environmental controls are of two types:

- Primary environmental controls consist of controlling the source of infection by using local exhaust ventilation. Examples of this are hoods, tents or booths. It also involves diluting and removing contaminated air by using general ventilation.
- Secondary environmental controls consist of controlling the airflow to prevent contaminated air in areas adjacent to the airborne source. Environmental controls also include the use of ultraviolet germicidal irradiation (UVGI) fixtures, when inadequate ventilation cannot be achieved.

Outpatient Settings

- Screen for respiratory symptoms as early as possible upon patient's arrival at the health care facility
- Provide patient education on cough hygiene and sputum disposal
- Segregate and Fast-track patients with respiratory symptoms

Inpatient settings

- Minimize hospitalization of TB patients
- Establish separate rooms, wards, or areas within wards for patients with infectious respiratory diseases
- Educate inpatients on cough hygiene and provide adequate sputum disposal
- Establish safe radiology procedures for patients with infectious respiratory disease, including smear-positive TB cases or TB suspects
- Use of protective equipment in situations that pose a high risk of exposure to TB disease.

References:

1. www.tbcindia.nic.in
2. www.cdc.gov
3. www.who.int
4. www.stoptb.org