AFB microscopic examination of tuberculosis patients (50 years old or above) admitted in National Institute of Diseases of the Chest and Hospital (NIDCH)

A dissertation is submitted for the partial fulfillment of the course of Pharmaceutical Research (PHRM 404) of the Department of Pharmacy,

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Submitted by:

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Declaration by the Research candidate

I, Tasnuva Tamanna, hereby declare that the dissertation entitled "**AFB microscopic examination of tuberculosis patients (50 years old or above) admitted in National Institute of Diseases of the Chest and Hospital (NIDCH)",** submitted by me to the Department of Pharmacy, East West University, in partial fulfilment of the requirements for the award of the degree of Bachelor of Pharmacy (B.PHARM) is a complete record of original research work carried out by me during the period 2011-2012 under the supervision and guidance of **Dr. Sufia Islam,** Associate Professor, Department of Pharmacy, East West University and it has not formed the basis for the award of any other Degree/Diploma/Fellowship or other similar title to any candidate of any University.

Place: Dhaka

Date: 10.07.2012

Signature of the candidate

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Thesis Certificate

This is to certify that the thesis entitled "**AFB microscopic examination of tuberculosis patients (50 years old or above) admitted in National Institute of Diseases of the Chest and Hospital (NIDCH)"**, submitted to the Department of Pharmacy, East West University, in partial fulfilment of the requirements for the award of the degree of Bachelor of Pharmacy (B.PHARM) is a complete record of original research work carried out by Tasnuva Tamanna (ID. 2008-3-70-030) during the period 2011-2012 of her research in the Department of Pharmacy at East West University, under my supervision and guidance and the thesis has not formed the basis for the award of any other Degree/Diploma/Fellowship or other similar title to any candidate of any University.

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Thank you

Abstract

Objective:
The purpose of the study is to focus on the AFB microscopic examination outcome of the patients (50 years old or above) to monitor whether they are responding to the anti-TB drug after one month of treatment or not.

Methods and materials:
In this study, AFB microscopic examination of the TB patients of 50 years old or above has been given the superior attention. Study patients have been selected randomly from the admitted patients in NIDCH. The reports of first and second (after one month of duration) AFB microscopic examination were compared to determine the level improvement. In total 20 patients were enrolled among them 7 were accessible for the follow up test. In order to compile the socio-demographic information a semi-structured questionnaire was prepared and filled up after taking the verbal consent from each patient. The haematological report of each patient at the beginning of disease identification was also assembled along with diagnosis report of AFB microscopic examination.

Result: The overview of the study shows that among 7 patients 2 patients (28.5 %) were properly responding to the anti-TB treatment. These two patients showed all negative result in the second AFB microscopic examination report. Rest of the 5 patients (71.43 %) was slowly improving with the anti-TB drugs rather being all negative in the result of second test. Another important feature is that, although follow up diagnosis is an inseparable element of the management of TB disease but only 7 patients (35%) out of total 20 patients available to perform the follow up AFB microscopic examination after one month of the detection of disease and initiation of treatment. These five patients had smear positive result in any of the specimens of the second test. Another important finding of this preliminary study includes 15% patients having family history of tuberculosis. Both the patients with smoking habit and male gender were identical that is 85%.

Discussion: Responding toward anti-TB medication was not rapid for every patients only 28.5% patients showed proper response to the treatment. Only 35 % were available for performing follow up test indicating the negligence of patients about the proper management of disease and the possibility of relapse.

Conclusion: To finish off the overview of the study, it is evident that follow up tests are required to come to a concrete conclusion about the effectiveness of treatment. Even though, it shows that patients were responding towards the treatment but more effective management is required. Further follow up test of these patients will help to come in an unambiguous conclusion.

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Chapter 1

Introduction

1.1 Tuberculosis

Tuberculosis is one of the most important health problems in developing countries like ours'. Tuberculosis mainly occurs due to bacilli belonging to the *Mycobacterium tuberculosis* complex. In the medical dictionary, tuberculosis is defined as "Tuberculosis (TB) is a potentially fatal contagious disease that can affect almost any part of the body but is mainly an infection of the lungs. It is caused by a bacterial micro-organism, the tubercle bacillus or *Mycobacterium tuberculosis*." According to WHO, "Tuberculosis, or TB, is an infectious bacterial disease caused by *Mycobacterium tuberculosis*, which most commonly affects the lungs. It is transmitted from person to person via droplets from the throat and lungs of people with the active respiratory disease."

Human being is the only carrier or reservoir of bacilli. *M. tuberculosis* multiplies slowly in comparison with other bacteria; therefore, TB has slower progress or development than most other bacterial infection. *Mycobacterium tuberculosis* is responsible for over 90% of all cases of tuberculosis (Peclzar M. J., 2009).

According to WHO "Over 9 million people fell ill and 1.7 million people died from TB in 2009, including nearly 600,000 women. The vast majority of deaths occur in developing countries like ours'.

1.2 Classification and cellular characteristics of mycobacterium tuberculosis complex (MTBC)

The *Mycobacterium* genus was described by Lehman and Neuman in 1896. It was identified based on some characteristics namely shape of the colonies, growth rate, and biochemical reactivity. Till now, seventy one species have been identified within this genus. These species are subdivided in two main groups depending on their growth rates.

1. The rapidly growing *Mycobacterium* species: A few number of rapidly growing *Mycobacterium* species are pathogenic for humans or animals and majority of them are non-pathogenic. Examples of them include *Mycobacterium abscessus*, *M. fortuitum*, *M. porcinum*.
2. The slowly growing *Mycobacterium* species: Majority of them are pathogenic for humans and/or animals such as the species of the MTB complex [MTBC], *M. leprae*, *M. ulcerans*, *M. avium*).

MTB complex is composed of seven different species which are-

- *Mycobacterium tuberculosis* (Koch, 1882)
- *M. bovis* (Karlsen and Lessel, 1970),
- *M. africanum*
- *M. microti* (Reed, 1957),
- *M. canettii* (still not officially recognized on the list of Bacterial Names with Standing in Nomenclature)
- *M. caprae*
- *M. pinnipedii*

Each member of MTBC is associated with a specific primary host but the responsible species for tuberculosis disease in human are *Mycobacterium tuberculosis* (MTB), *M. africanum*, and *M. canettii*. Among them *Mycobacterium tuberculosis* is the main species causing TB disease in human.

The cellular characteristics of the members of mycobacterium tuberculosis complex (MTBC) mentioned hereunder-

- Shape: Rod-shaped bacteria (0.2–0.6 μm wide, 1–10 μm long).
- Motility: Non-motile.
- Encapsulation: Non-encapsulated.

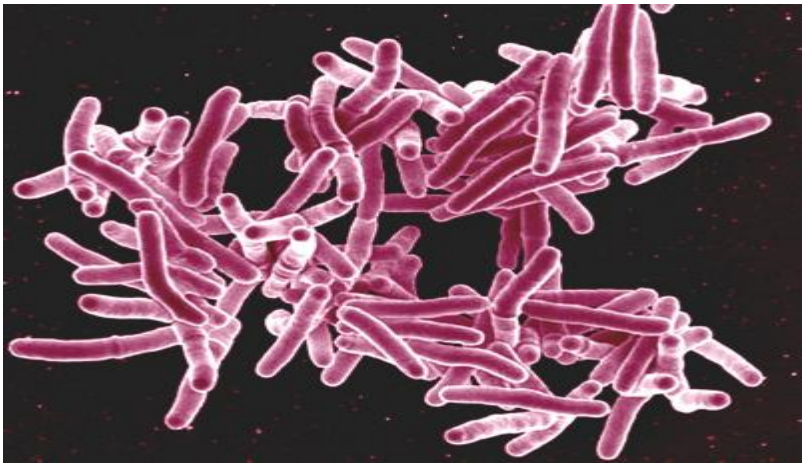

Figure 1.1: *M. tuberculosis.*

- Type: Gram positive.
- Types according to their need of oxygen to survive: Aerobes (growing most successfully in tissues with a high oxygen content such as lungs), or facultative anaerobes.
- Cellular target of infection: Usually infects mononuclear phagocytes (e.g., macrophages).

- ❖ Ability of synthesizing essential material: As deduced from its genome, *Mycobacterium tuberculosis* has the potential to manufacture all of the machinery necessary to synthesize its essential vitamins, amino acids, and enzyme co-factors.
- ❖ Cell wall: *Mycobacterium tuberculosis* has an unusual cell wall. There is an additional layer beyond the peptidoglycan layer. This additional layer is rich in unusual lipids, glycolipids, and polysaccharides.
- ❖ Identification: These bacteria can be detected with the help of optical microscopy after Ziehl–Neelsen (ZN) acid-fast stain of sputum from a person with active TB. In microscopic field, the bacilli appear as thin red rods against all other materials in the sputum pick up the blue counter stain (Godreuil S. *et al.* 2007).
- ❖ Growth rate: They have slow growth rate.
- ❖ Required temperature for multiplication: Temperature of 37 °C provides an ideal environment for the bacilli to replicate.
- ❖ Response to ultraviolet ray: These bacilli are rapidly destroyed in the ambient environment by ultraviolet rays (sunlight) (Khaled N.A. & Enarson D. A. 2003).

1.3 Transmission and multiplication of *Mycobacterium tuberculosis*:

The success of the pulmonary TB depend on four successive stages which are-

a) Bacilli phagocytosis
b) Intracellular multiplication
c) The stationary stage
d) The pulmonary form of TB

Brief description of each stage is given below-

a) **Bacilli phagocytosis:** Normally, the mature macrophages phagocytise the bacilli that reach to the pulmonary alveolus. This step is the first stage of infection which takes place in the first week following particle inhalation. Two main factors namely the bacillus virulence and the bactericidal activity of the macrophage largely govern this stage. In general, the bacteria are destroyed by the alveolar macrophages and the infection is blocked at this stage. Otherwise, they begin an intracellular cycle of multiplication.
b) **Intracellular multiplication:** The duration of second stage ranges between the 7th and the 21st day. It is also called the symbiotic stage. In this stage, the bacteria whose escape themselves from being destroyed by the alveolar macrophages will multiply. After cellular lyses, they are released and can infect other circulating macrophages thus continue their multiplication.

c) **Stationary stage:** Due to the induction cell-mediated immunity in host, bacterial growth becomes steady. This is the third stage of the infection and also known as primary infection. Because of delayed-type hypersensitivity, the macrophages in which bacilli multiply are destroyed and bacterial toxins and cellular products are released. This phenomenon leads to the formation necrosis where pseudo-equilibrium is developed between inactivated and mature macrophages. At this stage, either the number of infected cells decreases due to the phagocytises of released bacilli by the mature macrophages or increases if the bacilli multiply in the inactivated macrophages. In this stage, the bacilli may become dormant referred to as a "latent infection" or the latent organisms can eventually begin to develop the disease condition known as "TB reactivation."

d) **Pulmonary form of TB (PTB):** The infection reaches to the final stage when the equilibrium between the inactivated and mature macrophages is broken down. In this last stage, formation of a cavity is detected by pulmonary radiography. The liquefied material is present in this cavity which provides an excellent growth media for the bacteria. This condition also causes the destruction of macrophages. At this stage of the disease, the person can transmit the infection to others by releasing the bacilli into the air. If the person remains untreated, the disease will turn to a chronic TB leading to even death (Godreuil S. *et al*., 2007).

1.4 Difference between TB infection and TB disease

1.4.1 Tuberculosis infection: TB disease is transmitted form one person to anther through droplet infection. When a patient with TB disease cough, sneeze they contaminate the area with the causative agent of TB that is *Mycobacterium tuberculosis*. These bacilli stay suspended in the air as droplet. Healthy people become infected with disease by inhalation of these droplets. It does not mean that a person inhaling the air containing the *Mycobacterium tuberculosis* has been developed the disease. Around 90% of the infected people do not develop TB disease because their immunity spares them from the development of TB disease. In case of tuberculosis infection, the tubercle bacilli remain present in the body but the intact immune system is keeping them under control. In TB infected person, the bacilli are engulfed by the pulmonary macrophages and pulmonary macrophages act as scavengers as well. The TB infection turns to TB disease when the immune system is compromised because then the bacilli multiply and spread to other sites in the body. Around 10% of the infected people progress to TB diseases in their lifetime. Even if, the people who are only infected with TB rather progressing to the TB disease have following criteria

- ❖ Do not show any symptoms.
- ❖ Do not even feel sick.
- ❖ Show positive skin test (Mantoux test).
- ❖ Cannot spread the TB to others (Guidelines for Health Care Providers, 2002–2005).

This is also known as the dormant / latent tuberculosis infection (LTBI)

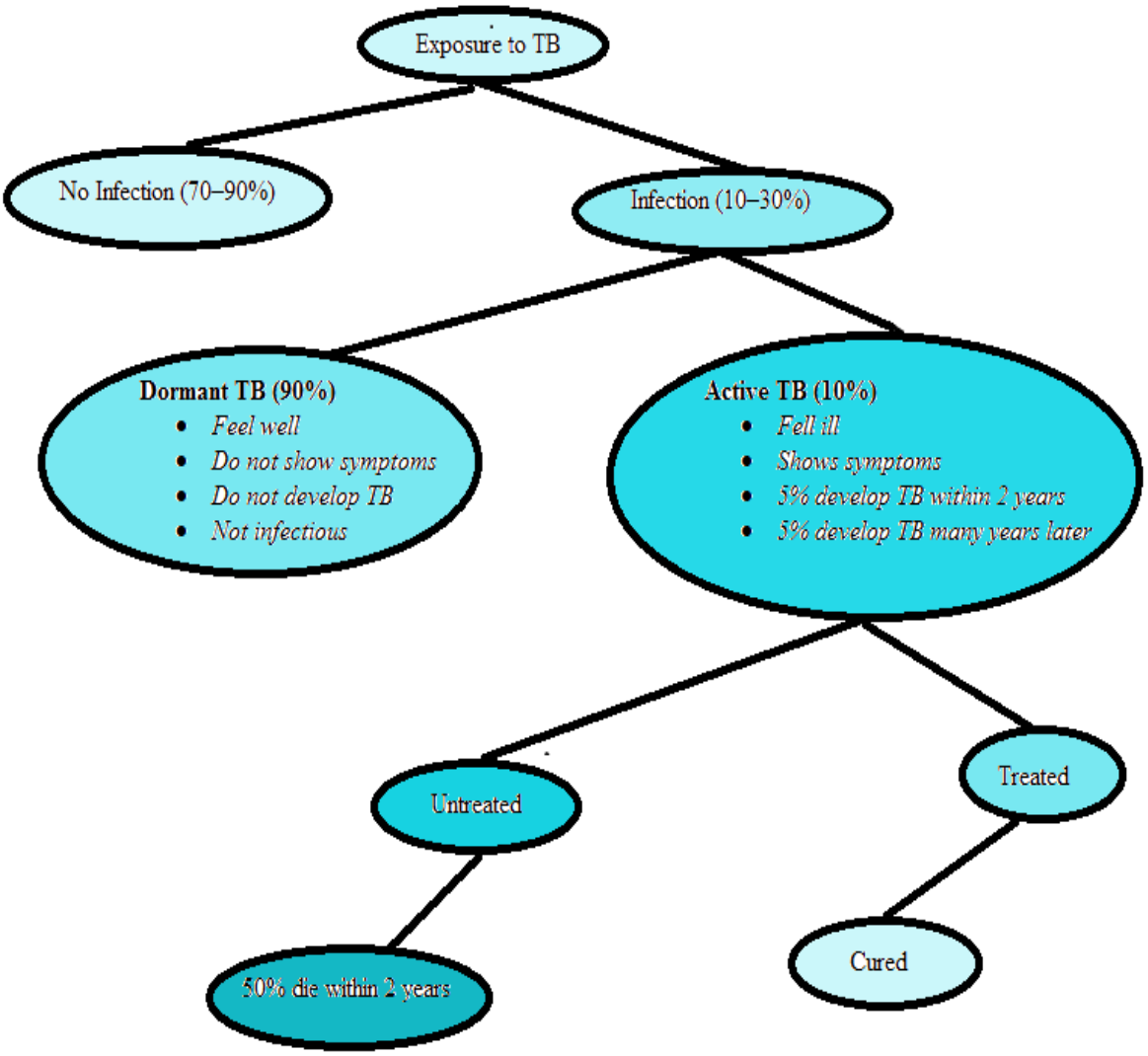

Figure 1.2: Flow chart showing the difference between TB disease and TB infection.

1.4.2 Tuberculosis disease:

When the TB bacilli multiply in the lungs and other organs during the life time of the infected person, they produce signs and symptoms of TB disease. Tuberculosis disease mainly

develops due to the failure of immune system to keep the tubercle bacilli under control. As a result, the bacilli do not only reside within the macrophage rather begin to multiply rapidly. Normally, about 10% infected individual turns to TB patient. The remaining 90% infected individual will live free of disease during the rest of their lives. About 5% of the infected individuals develop TB diseases within two years and the remaining develops TB disease in the old age which is known as reactivation of the disease.

Therefore, TB disease can be distinguished from the TB infection as such the infection with presence of sign and symptoms of TB (WHO, 2008).

1.4.3 Risk Factors for active TB disease in persons with LTBI: The persons with latent tuberculosis infection (LTBI) are at increased risk to progress to have the active disease condition. Such conversion of tuberculosis infection to TB disease is accelerated if any of the following are being observed:

- ❖ Human immunodeficiency virus (HIV) infection
- ❖ Use of injectable drugs
- ❖ Diabetes mellitus (especially insulin-dependent)
- ❖ Silicosis
- ❖ End-stage renal disease
- ❖ Chronic immunosuppression such as-
  - Organ transplant
  - Prolonged corticosteroid treatment
- ❖ Hematologic or reticuloendotheial diseases such as
  - Leukemia
  - Hodgkins' disease
- ❖ Malnutrition and clinical situations associated with rapid weight loss such as-
  - Cancers of the head and neck
  - Intestinal bypass or gastrectomy
  - Chronic mal-absorption
  - Low body weight (more than ten percent below ideal body weight)
- ❖ Radiographic findings consistent with old or healed lesions in the lung (Jensen P. A. *et al*, 2005).

1.5 Classification of tuberculosis:

Tuberculosis can be classified depending on various aspects. Classification of tuberculosis based on different aspects is being described below.

1.5.1. Classification according to whether it is active or dormant like

a. Inactive TB and
b. Active TB

a. Inactive TB

Inactive TB infects the lungs but it is not growing. Normally, the person with inactive TB infection has no symptoms and the chest x-ray may be normal. The only manifestation of inactive tuberculosis is the reaction to the tuberculin skin test (TST) or interferon-gamma release assay (IGRA) (Iseman D. & Daley D, 2012).

b. Active TB

Active TB is the form of tuberculosis disease where the causative bacteria are rapidly multiplying and invading different organs of the body. A person with active TB disease is able to transmit infectious particles to others by airborne conduction when he or she has coughed. Actually, when the TB starts to grow in the lung of a person, it turns to be active. Normally, multi-drug treatment is employed to treat active TB disease (Milstien D.J., 1996).

1.5.2. Classification according to pathogenesis of the disease-

a. Class 0
b. Class 1
c. Class 2
d. Class 3
e. Class 4
f. Class 5

It is also known as WHO (World Health Organization) classification of TB. Description of each class are shown below-

a. Class 0

When the individuals have had no exposure to tuberculosis and have a negative tuberculin test is considered to be belonging to “Class 0”. This test determines if a person has already been infected with tuberculosis mycobacterium by measuring how sensitive a person’s immune system is to proteins, called tuberculins, from the tuberculosis mycobacterium cultures (Lingohr-Smith M, 2011). That means no history of TB exposure and no evidence of *M. tuberculosis* infection or disease is found in case of “Class 0” individuals (CDC, 2011).

b. Class 1

When the individuals have been exposed to tuberculosis, but did not have a positive tuberculin skin test are placed in “Class 1”. That means in such case there has been exposure but no evidence of infection (CDC, 2011). Treatment for latent tuberculosis infection should be initiated if the person is immune compromised and especially if they are HIV positive. Other recommended alternative treatments include a two month daily regimen of rifampin and pyrazinamide or rifampin alone for four months (Lingohr-Smith M, 2011).

c. Class 2

When the individuals have a positive tuberculin skin test, but no evidence of active tuberculosis can be considered of belonging “Class 2”. Some persons in class 2 may be treated for latent tuberculosis infection (Lingohr-Smith M, 2011). That means the feature of this class includes positive reaction to TST (tuberculin skin test), negative bacteriological studies (smear and cultures), and no bacteriological or radiographic evidence of active TB disease (CDC, 2011).

d. Class 3

When the individuals have active tuberculosis and exhibit symptoms of the disease are placed in “Class 3” category. Patients with active tuberculosis may have abnormalities in the upper lung lobes that can be detected by a chest radiograph. Patients with past active tuberculosis may have nodules and fibrotic scars in the upper lung lobes. Individuals in class 3 will remain in class 3 until treatment of tuberculosis is completed (Lingohr-Smith M, 2011). That means the feature of this class includes positive culture for *M. tuberculosis* or positive reaction to TST and clinical, bacteriological, or radiographic evidence of current active TB (CDC, 2011)

e. Class 4

When the individuals do not have active tuberculosis disease, but have had a previous episode of tuberculosis and have a positive reaction to the tuberculin skin test are placed in “Class 4” (Lingohr-Smith M, 2011). That means the feature of this class includes past medical history of TB disease, abnormal but stable radiographic findings, positive reaction to the TST, negative bacteriologic studies (smear and cultures) and no clinical or radiographic evidence of current active TB disease (CDC, 2011).

f. Class 5

Individuals are placed in “Class 5” when suspected of having tuberculosis with a pending diagnosis. After a tuberculin skin test, mycobacterium culture and chest x-ray of the person should be classified in one of the other preceding classes (Lingohr-Smith M, 2011)). That

means the feature of this class includes signs and symptoms of active TB disease but incomplete medical evaluation (CDC, 2011).

1.5.3. Classification according to the severity of tuberculosis disease

a. Primary tuberculosis
b. Secondary tuberculosis
c. Disseminated tuberculosis and
d. Miliary tuberculosis

a. Primary Tuberculosis

It can be defined as primary tuberculosis when tuberculosis affects a person who had never been exposed to the causative agents prior. In case of primary tuberculosis, the source of bacterium is external. The symptoms of such form of TB disease have much more similarities with those of pneumonia. In primary tuberculosis, the lymph nodes get affected causing them to be swelled. Lesions are also developed which are removed during treatment. But, the exclusion of the lesion does not specify that bacterial are removed permanently. This is because; as the bacteria may have turned into a latent phase and if it remains untreated then recurrence of TB will happen at favourable condition (Mir A., 2012).

b. Secondary Tuberculosis

Another name of secondary tuberculosis is post-primary tuberculosis. This type of tuberculosis occurs in a person who had been infected with TB prior. The difference between primary tuberculosis and secondary tuberculosis is that, in case of primary TB, the bacterium goes into an inactive phase or dormant phase whereas in case of secondary tuberculosis, the bacterium regains its active mode and patients show the symptoms. Due to the highest oxygen pressure in lungs, secondary tuberculosis is mostly localised there. Secondary tuberculosis is more infectious than primary tuberculosis. Again, secondary TB also boosts up the possibility of the spreading of the infection to other organs such as kidneys, heart and brain (Mir A., 2012).

c. Disseminated Tuberculosis

When the tuberculosis has infected the entire body system, it is termed as disseminated tuberculosis. It is a very rare type of the disease. Primarily infection of disseminated tuberculosis occurs to bones of spines, hips, joints and knees, the genital tract of women, the

urinary tract and even the central nervous system. It can even affect the cerebrospinal fluids, the gastrointestinal tract, the adrenal gland, skin of the neck and even the heart.

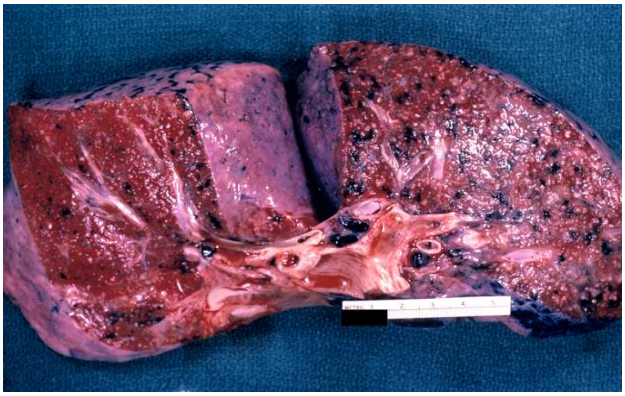

Figure 1.3: Dissection of lung from this patient with disseminated tuberculosis.

This is a gross photograph of a cut section of lung from this patient with disseminated tuberculosis. The numerous small white nodules scattered throughout this lung tissue represent individual tuberculosis granulomas. In addition, note the dark areas throughout the lung which represent deposits of anthracotic pigment (Mir A., 2012).

d. Miliary Tuberculosis

The most severe type of tuberculosis infection is known as miliary tuberculosis in which total of the blood stream gets infected with the bacterium. Appearance of numerous tiny lesions can be seen in such type of infection

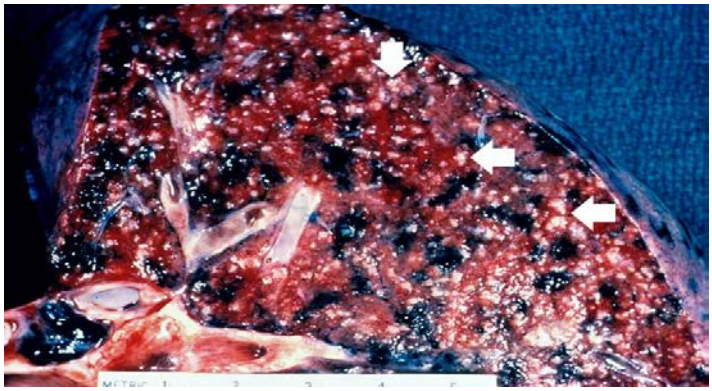

Figure 1.4: Lesions appeared in miliary tuberculosis.

This is a closer view of the same section of lung containing multiple white granulomas which are now more easily identified (arrows). These lesions are referred to as miliary tuberculosis. Dark areas of anthracosis are also prominent in this lung.

Anaemia results when the infection reaches to the bone marrow. The infection in the blood leads to uncontrolled multiplication of white blood cells which ultimately resulting to

leukaemia-like conditions. In case of miliary tuberculosis, the host immune response becomes insufficient (Bhowmik D. *et al,* 2009).

1.5.4 Classification according to the anatomical site involved-

a. Pulmonary tuberculosis (PTB)
b. Extra-pulmonary (EP) form of tuberculosis

a. Pulmonary tuberculosis

PTB has certain signs which are quite specific which include prolonged cough (more than two weeks), sputum production and chest pain. There are some other symptoms associated with PTB which are less common namely weight loss, anorexia, fatigue, moderate fever, night sweats. But the most prevalent sign of pulmonary tuberculosis is presence of blood in sputum (haemoptysis). There is a variability of all the signs of PTB and these develop in a chronic, insidious manner.

The advanced forms of PTB and its' complication are common outside the developed countries which are as follow-

- ❖ Respiratory insufficiency results from the extension of the lesions.
- ❖ Massive haemoptysis resulting from large cavities with hyper-vascularisation and erosion of vessels.
- ❖ Accumulation of pus in the pleural space.
- ❖ Pnemothorax due to the rupture of a cavity in the pleural space.

Pulmonary tuberculosis can be again divided into the following classes-

- Primary Tuberculosis Pneumonia
- Tuberculosis Pleurisy
- Cavitary Tuberculosis
- Laryngeal Tuberculosis

Each is being described hereunder-

Primary tuberculosis pneumonia

It is an uncommon type of TB that presents as pneumonia and is very infectious. Patients have a high fever and productive cough. It occurs most often in extremely young children and the elderly. It is also seen in patients with suppression of immunity, such as HIV-infected and AIDS patients, and in patients on long term corticosteroid therapy (Bhowmik D. *et al*, 2009).

Tuberculosis Pleurisy

One of the common complications of primary tuberculosis or in conjunction with pulmonary infiltrate typical of post primary tuberculosis is pleural effusion or tuberculosis pleurisy.

The inner surface of the chest wall and the surface of the lungs are coated by the parietal and visceral pleura having a potential space of 10-24 µm between the two pleural surfaces. Normally, the space between two pleural spaces is filled up with approximately 1 ml of fluid. The presence of this fluid maintains the balance between hydrostatic and oncotic forces in the visceral and parietal pleural vessels. It also assists in extensive lymphatic drainage. Pleural effusion is resulted from the disturbance of this fluid leading to the accumulation of large amounts of fluid in the pleural space under pathologic conditions.

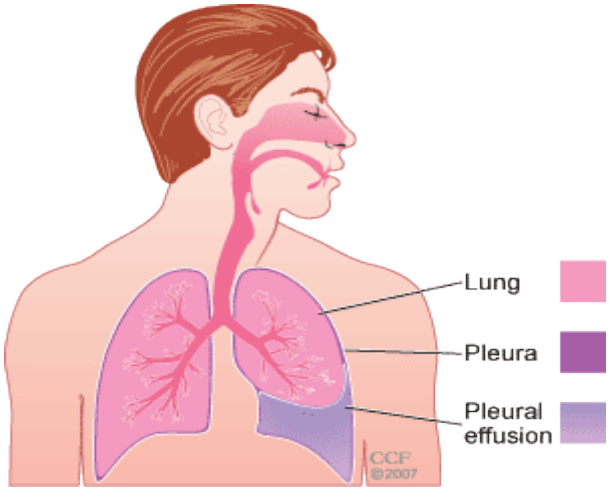

Figure 1.5: Pleural effusion.

The diagnosis of tuberculosis pleural effusion (TPE) is difficult as lymphocytic exudates observed in TB pleural effusion also can take place in other disease such as malignancy, collagen vascular disease and lymphoma (Soe Z. *et al*, 2010).

Cavitary TB

Cavitary TB involves the upper lobes of the lung. The bacteria cause progressive lung destruction by forming cavities, or enlarged air spaces. This type of TB occurs in reactivation disease. The upper lobes of the lung are affected because they are highly oxygenated (an environment in which *M. tuberculosis* thrives). Cavitary TB can, rarely, occur soon after primary infection. Symptoms include productive cough, night sweats, fever, weight loss, and weakness. There may be haemoptysis (coughing up blood). Patients with cavitary TB are highly contagious (Bhowmik D. *et al*, 2009).

Laryngeal TB

TB can infect the larynx, or the vocal chord area. It is extremely infectious (Bhowmik D. *et al*, 2009).

Site and symptoms of different types of are shown in the following figure

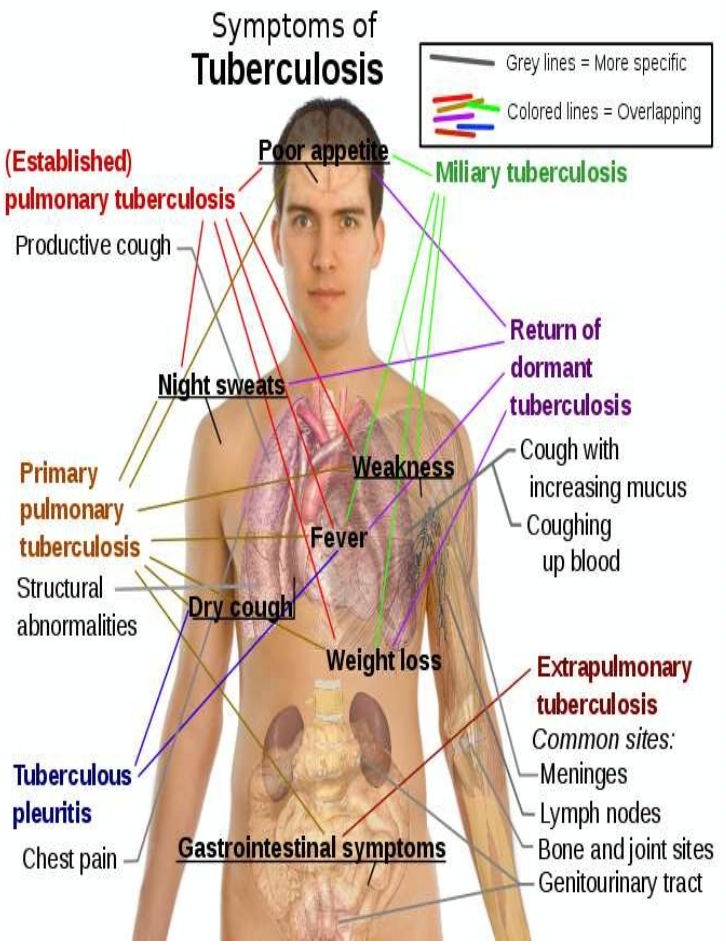

Figure 1.6: Site and symptoms of different types of tuberculosis.

b. Extra-pulmonary (EPTB) forms of tuberculosis

Mycobacterium tuberculosis can spread to the entire organism starting from an initial pulmonary localization during silent phase. Therefore, active TB can progress in many other parts of the human body like in lymph nodes, meninges, vertebrae, joints, genital organs, and even kidney. Such spread out of tuberculosis throughout the body is termed as extra-pulmonary tuberculosis.

The common clinical characterization of EP form of TB include-

- ❖Insidious evolution
- ❖Cold lesions often accompanied by deterioration of physical condition
- ❖Lack of response to symptomatic or non- symptomatic infectious treatment
- ❖They are often isolated but may be associated with a pulmonary localization.

According to the organ where the tuberculosis has been spread out, extra-pulmonary tuberculosis can be subdivided into the following categories

- Lymph node TB
- TB of bone and joints
- Ascetic abdominal TB
- Genito-urinary TB
- TB pericardial effusion
- TB meningitis

Lymph node TB

Lymph node TB is a frequent pathology in central Asia and certain areas of Africa such as in Senegal, Djibouti. This form of tuberculosis is very much common with the children and HIV patients.

These are characterized with non-inflammatory adenopathies, cold and painless, single or multiple, usually bivalent, evolving chronic mode towards softening and fistulisation. Cervical localization is the most common among all others.

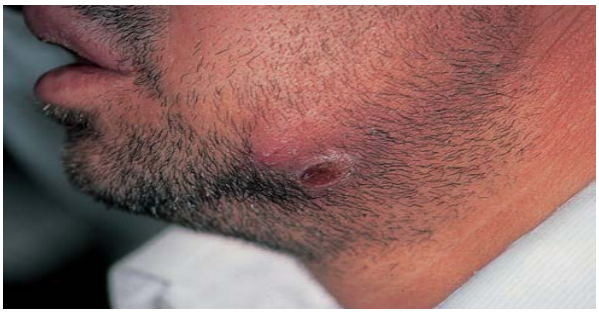

Figure 1.7: Involvement and fistula formation in lymph node tuberculosis.

Lymph node TB is non-infectious, generally not leading the patients' life in danger except when it is accompanied with second condition like HIV, cancer.

Adenopathies normally disappear within less than three month with the proper initiation of treatment. Paradoxical reaction can be observed at the beginning of the treatment that is appearance of abscesses, fistulas or other lymph nodes but alteration of the treatment should not be taken (F. Varaine *et al*, 2010).

TB of bones and joints

Due to better vascularisation and oxygenation of osteo-articular structures during growth, these forms of tuberculosis are mostly common in children.

The joints most often infected by such form of TB are the hips, knees, elbows, and wrist. Half of the patients having arthritis have pulmonary tuberculosis at the same time. Like arthritis, osteitis is also distinguished from common bacterial infection by contrast of symptoms.

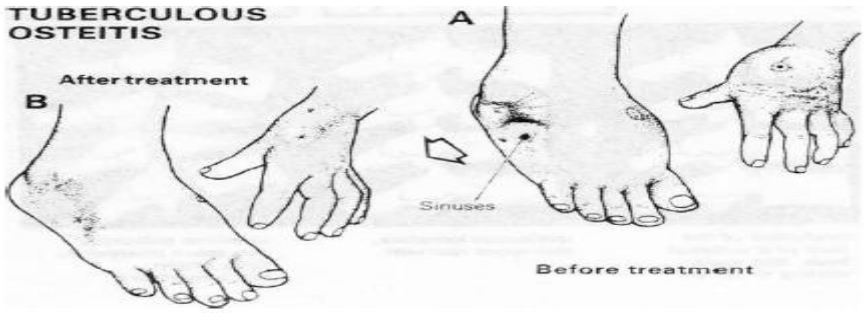

Figure 1.8: Osteitis tuberculosis

In the above figure, A represents the condition before treatment and B represents the condition after treatment in case of bone and joint tuberculosis.

Spondylodiscitis or Pott's disease (infection) can happen at any age where the effect mainly occurs in vertebrae and disks leading to the destruction and deformation of the spine. It is a severe form of tuberculosis which must be taken with priority for the treatment. This condition may require surgical consultation (F. Varaine *et al*, 2010).

Ascitic abdominal TB

Ascites mean accumulation of fluid in abdomen. TB ascites can be identified as peritorial localization of the infection. The common diagnostic problem of this rare form of TB disease in tropical region arises due to the frequency of all types of chronic ascites. An ascites puncture is characterized with diagnostic augment of a translucent yellow-coloured liquid, rich in lymphocytes, an exudative nature. Exudative ascites may occur because of carcinoma or bacterial-infection of transudate (F. Varaine *et al*, 2010).

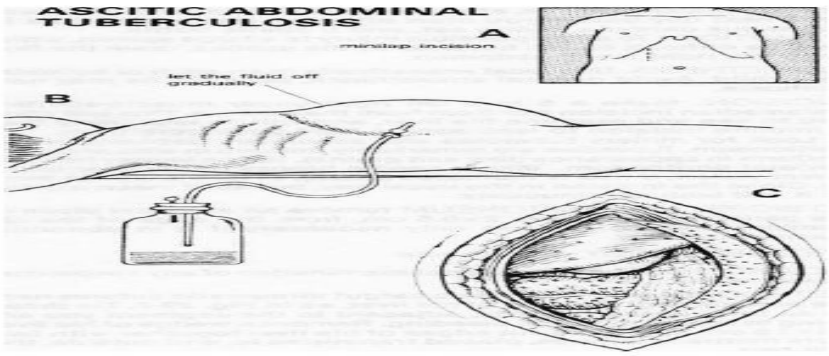

Figure 1.9: Ascetic abdominal tuberculosis

In this diagram A, B, and C represent ascetic abdominal tuberculosis, draw off of fluid, and miliary tubercles of the parietal peritoneum, liver and gut respectively.

Genitourinary TB

Such type of tuberculosis can remain asymptomatic for a long period of time until urinary signs extend to the genital tract. More specifically, genitourinary TB can be defined as the localization of the infection to the renal system. The presence of micro or macroscopic haematuria and a “sterile” pyuria by microscopy are considered in case of the diagnosis of genitourinary TB.

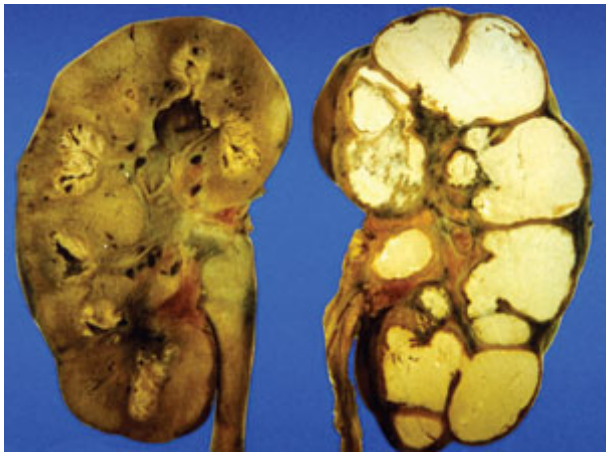

Figure 1.10: A post-mortem specimen demonstrating cessation in the renal cortices of a patient affected by tuberculosis.

In women, haematogenous path can be another way of genital tract contamination. Some non-specific symptom of genital localization can be abdominal pain, leucorrhoea, vaginal bleeding which are variable. It can be extended to peritoneum which can be a basis of ascites. In men, genital localization is secondary to renal localization and it is accompanied often with cold epididymitis, which is responsible of causing scrotal pain (F. Varaine *et al*, 2010).

TB pericardial effusion

TB pericardial effusion is one type of tuberculosis infection of the pericardial membrane (pericardium) that envelops the heart. Infection of the pericardium causes build-up of fluid (effusion) around the heart. This accumulation of fluid constrains the pumping action (tamponade) that is life threatening. Sometimes thickening of the pericardium occurs without an effusion due to tuberculosis infection. Such type of thickening can also lead to constrain the pumping action (constrictive pericarditis).

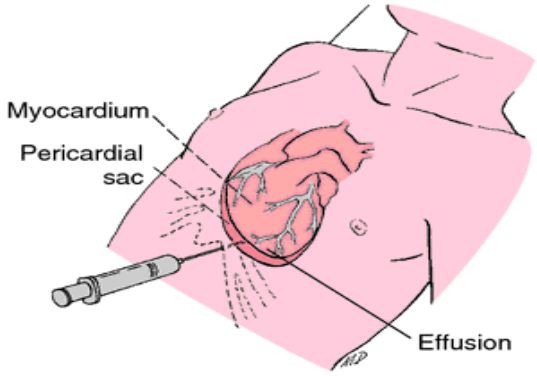

Figure 1.11: Pericardial effusion.

Chest pain, shortness of breath, oedema of the lower limbs, and sometimes ascites are the clinical manifestation of such form of tuberculosis. Clinical examination can also be associated with pericardial friction rub, raised jugular pressure, and tachycardia, enlarged heart (Mayosi B. M., 2009).

TB meningitis

TB meningitis can be defined as infection of the meninges by *Mycobacterium tuberculosis* which is the most common form of CNS tuberculosis. Meningitis is membranes which envelops the central nervous system. Cranial nerve roots may be affected in case of inflammation in the brain stem subarachnoid area (Thwaites G. *et al*, 1999).

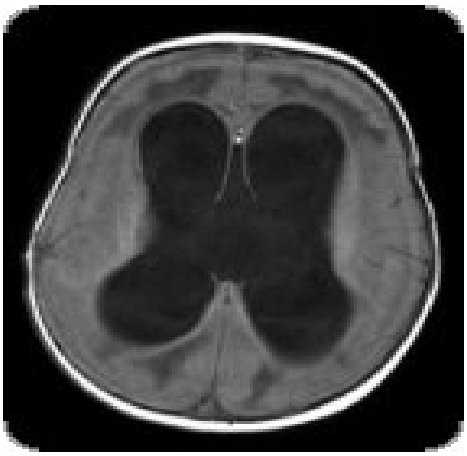

Figure 1.12: TB meningitis.

Children are highly prone to TB meningitis during the first year following primary infection. It is characterized with headache, irritability, fever and alteration of physical condition. In most the cases, vomiting, stiff neck, hypotonia in infant, photophobia can appear which are mainly know as meningeal syndromes (Bhowmik D. *et al*, 2009).

1.6 Symptoms of tuberculosis:

As mentioned before, people with tuberculosis infection do not have any infection unless it progresses to active diseases. Sign and symptoms of tuberculosis vary according to the anatomical site of infection. The symptoms of both pulmonary TB and extra- pulmonary TB are mentioned below.

1.6.1 Symptoms of pulmonary TB:

A patient with pulmonary TB may have one or more of the following symptoms:

- Respiratory symptoms which include-
  - Persistent cough with sputum (thick, cloudy mucus from the lungs) for more than 2 weeks.
  - Haemoptysis (coughing up blood from the respiratory tract).
  - Shortness of breath
  - Chest pain (WHO, 2008)
- General symptoms:
  - Loss of weight
  - Fever
  - Night sweats
  - Tiredness/ fatigue
  - Anorexia (loss of appetite)
  - General malaise (general feeling of being unwell) (Manual of prevention and control procedure, 2010)

A person with persistent cough for three weeks or more can be suspected to have pulmonary tuberculosis.

1.6.2 Symptoms of extra- pulmonary TB

Although the signs and symptoms of extra-pulmonary TB depend on the site involved, but most common symptoms are as follows-

- TB lymph adenitis: Swelling of lymph nodes
- Pleural effusion:
  - Fever
  - Chest pain
  - Shortness of breath
- TB arthritis: Pain and swelling of joints.
- TB of the spine: Radiological findings with or without loss of function.

- ❖ Meningitis:
    - Headache
    - Fever
    - Stiffness of neck
    - And subsequent mental confusion (American thoracic society, 2000).

1.7 Diagnosis of tuberculosis:

Diagnosis of tuberculosis patients varies according to the anatomical site involved. But preliminarily some diagnosis tests are performed for all suspected patients such as sputum-smear microscopy, culture of TB bacilli, tuberculin skin test (not in Bangladesh), and X- ray.

Diagnosis tests of pulmonary tuberculosis involves-

- ❖ AFB microscopic examination (sputum- smear microscopy)
- ❖ Radiographic examination (X-ray)
- ❖ Tuberculin Skin Test
- ❖ Culture of TB bacilli

The diagnosis of extra-pulmonary TB should always be made by a graduate physician or specialist and often requires special examinations such as X-ray examinations, biopsies, FNAC, etc.

Description of each type of diagnosis pattern are illustrated below-

1.7.1 AFB microscopic examination (sputum smear microscopy): In Bangladesh, where most of people live under the poverty line and where the prevalence of tuberculosis is high, AFB microscopic examination (sputum smear microscopy) is likely to be the cost-effective tool for diagnosing patients with tuberculosis infection. AFB microscopy is an easy, economical, suitable and appropriate method which is relatively simple to perform and to read.

Microscopic examination of sputum of the TB suspected persons is done by the Ziehl-Neelsen method. Over 65% of pulmonary TB patients are smear-positive and will be detected by this method. The rest of the pulmonary tuberculosis patients are smear- negative as the number of bacilli in their sputum is too low to be identified through this method.

The Ziehl-Neelsen (ZN) staining technique is a method of choice for AFB microscopic examination as this method provides consistently good results without requiring special equipment. The advantages Ziehl-Neelsen (ZN) staining technique include-

- Better standardisation and quality assurance

- Outweigh the disadvantages of long term storage.

By this method it is easy to detect the micro-organism whereas other cold staining procedures such as the Kinyoun and Tan Thiam Hok methods have difficulty in detecting acid-fast bacilli (AFB) and the staining also fades rapidly.

Ziehl-Neelsen reagents includes-

1. **Ziehl's carbol fuchsin:** To prepare the Ziehl's carbol fuchsin working solution, 10 ml of Solution A is mixed with 90 ml of Solution B**.**

Solution A (fuchsin alcoholic stock solution) contains basic fuchsin and alcohol (95%). Solution B (aqueous phenol solution) contains phenol crystals and distilled water.

2. **Decolourising agent solutions:** It is an acid-alcohol solution that contains alcohol (95%) and 35% concentrated hydrochloric acid.

When alcohol is unavailable 25% aqueous sulfuric acid solutionis also used as decolourising agent solution.

3. **Methylene blue counterstaining solution 0.3%**: It contains methylene blue chloride and distilled water.

1.7.1.1. AFB microscopic (sputum smear microscopy) technique: The entire technique of AFB microscopy (sputum smear microscopy) can be described as follows-

Smear preparation: Smear is prepared according to the following steps.

Smearing: First of all, the container should be opened carefully in order to avoid aerosol production. After that a wood or bamboo stick is broken down to be used as applicator in order to select yellow, purulent particle of sputum with jagged end of the broken wood or bamboo stick applicator. The sputum is then smeared uniformly over the middle area of the slide with a continuous rotational movement. The slides are then placed on dryer with smeared surface upwards. Within the drier the smear is air dried for about 30 minutes.

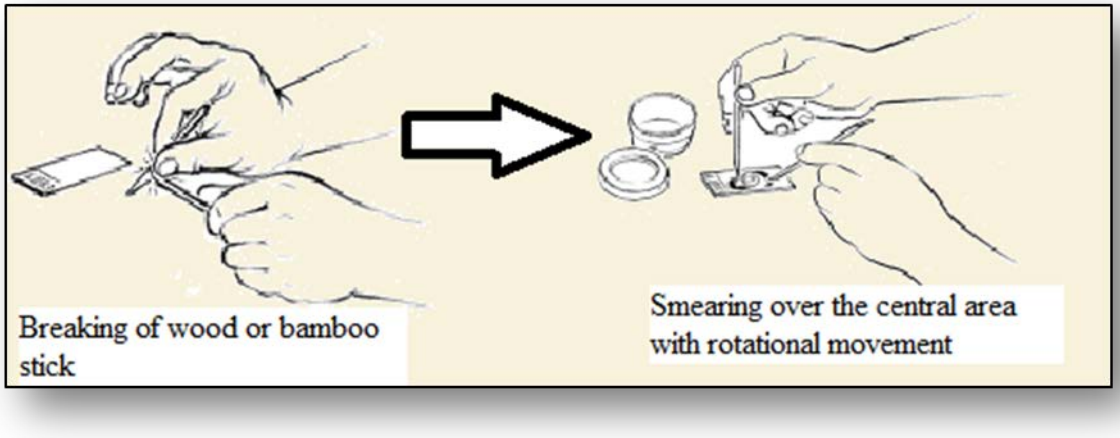

Figure 1.13: Smearing of the sputum.

The recommended applicators are only used once.

Fixing: Dried smears can be fixed by holding the slide with forceps and passing the smear side up over the flame five times for about four seconds. Over heating should be avoided very carefully.

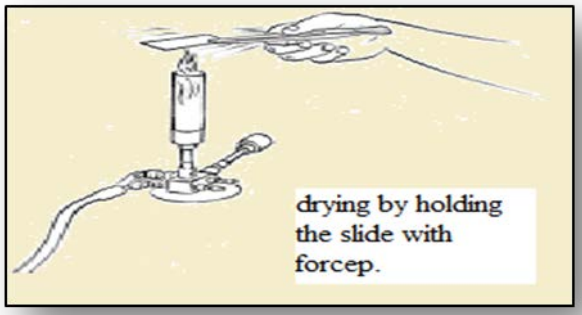

Figure1.14: Fixing.

Staining: The next step after fixing is staining where the fixed slides are placed on the staining rack with the smeared side upward. There should be 1 cm gap between the slides and those must not be touched to each other. The smear dried slides are then individually covered with 0.3% Ziehl's carbol fuchsin (filtered) working solution. After that a strip of absorbent paper like a filter paper or even a newspaper is placed for holding the staining solution. It will also avoid the deposition of fuchsin crystals on the smear. Then, the slides should be heated from the bottom with the help of a bunsen burner or an alcohol lamp or an alcohol soaked cotton swab until vapour starts to rise. Staining solution must not to be permitted to boil and drying of staining should also be prohibited. The next step is to rinse the slides smoothly with water in order to remove excess carbol fuchsin and excess rinsing water should also be drained off from slides. At the end of this stage, sputum smear will appear red in colour. The slides still need to be covered up with hot, steaming carbol fuchsin over five minutes.

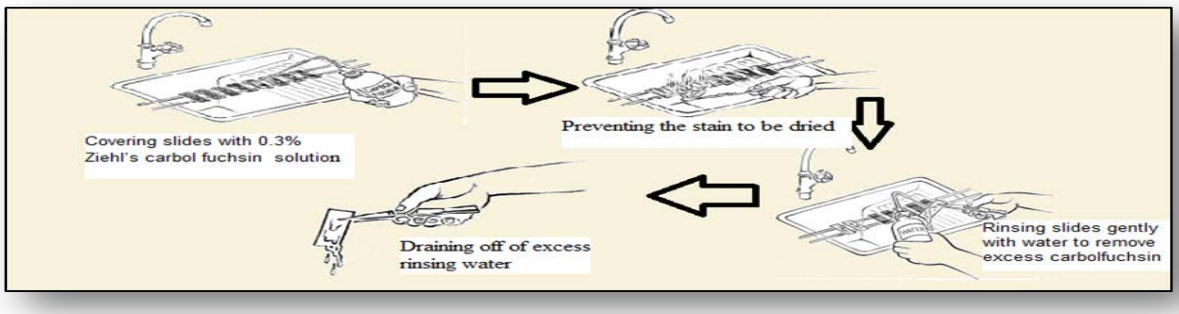

Figure 1.15: Staining.

Decolourising: In this step, slides are covered with 25% sulfuric acid or acid-alcohol solution followed by standing for about 3 minutes. After that, the red colour should have almost entirely disappeared. When it is required the sequence will be repeated until the red colour disappears completely. There should be enough care for avoiding over decolourisation. The sulfuric acid or acid alcohol and the excess stain must be washed away gently by using water. Finally, excess rinsing water must be drained off from slides.

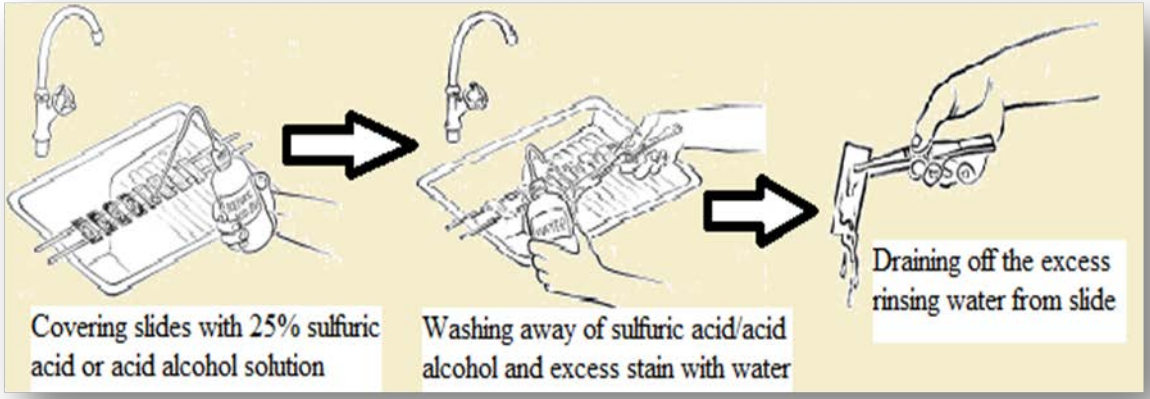

Figure 1.16: Decolourising.

Counterstaining: Final step of smear preparation starts with covering slides with 0.3% methylene blue counterstaining solution followed by standing for one minute. At the end, slides are rinsed individually with water which are then eventually allowed to be air dried.

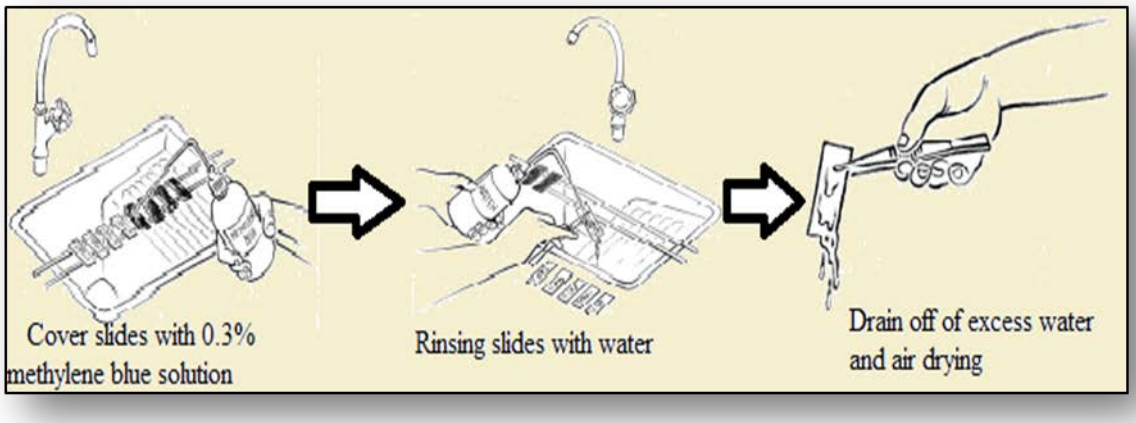

Figure 1.17: Counterstaining.

Usually the Ziehl-Neelsen staining technique requires five minutes for staining, three minutes for decolourising and only one minute counterstaining.

B. Microscopic examination: Acid-fast bacilli are appeared as bright red or pink against the blue background. Acid-fast bacilli differ greatly in shape, from short, coccoid to elongated filaments. These bacilli may appear as uniformly or unevenly stained. The acid-fast bacilli can even appear as granular (Akther M. *et al*, 2000).

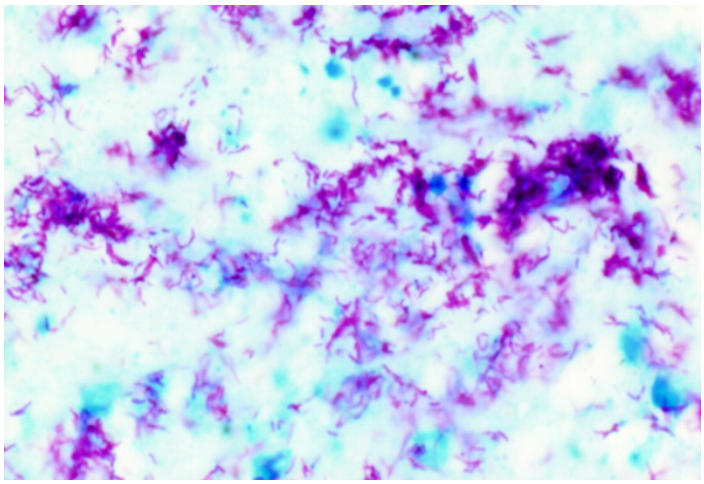

Figure 1.18: MTB in acid fast stain of sputum.

The result of AFB microscopic examination is interpreted based on the amount of microbes present in the specimen. The grading standard is given as follow-

Table: 1.1: Grading of the AFB microscopic examination

Examination	**Grading**
More than 10 AFB per oil immersion field	3+
1–10 AFB per oil immersion field	2+
10–99 AFB 100 oil immersion fields	1+
1–9 AFB per 100 oil immersion fields	Scanty
No AFB in 100 oil immersion fields	Negative

(RNTCP glance, 2012)

1.7.1.2. Sample and consequences of AFB microscopic examination:

Generally, a person with cough for more than 3 weeks duration, even in absence of other symptoms, is advised for AFB microscopic examination (sputum smear microscopy). In sputum smear microscopic examination, three sputum samples namely the SPOT SPUTUM, EARLY MORNING SPUTUM, and SPOT SPUTUM are collected within two days.

Three consequences may take place which are described hereunder-

- When all the three samples are smear positive or two are smear positive and one smear negative, the patient is considered to have tuberculosis and are given treatment with CAT I but if the patient have previous history of treatment more than one month then the treatment is started with CAT II.
- In case of result where one of three samples is smear positive and other two are smear negative, three samples are collected again for the microscopy examination. If the results remain same then chest X-Ray (PA view) is recommended. When the X-Ray report represents a positive result the physician starts the treatment either with CAT I or CAT II.
- In case of all smear negative samples, physician prescribes the patients antibiotic for 1-2 weeks. After taking the antibiotics if symptoms persist, three sputum samples are collected again. In case of negative result, a chest X-Ray should be taken. If the X-Ray report represents positive results then it is defined as smear negative pulmonary tuberculosis and if the repost represents negative result then the patient is not considered to be a tuberculosis patient (WHO, 2008).

The possible outcomes of AFB microscopy and corresponding treatments have been illustrated by a flow chart according to the National Guideline and Operational Manual for Tuberculosis Control (WHO, 2008).

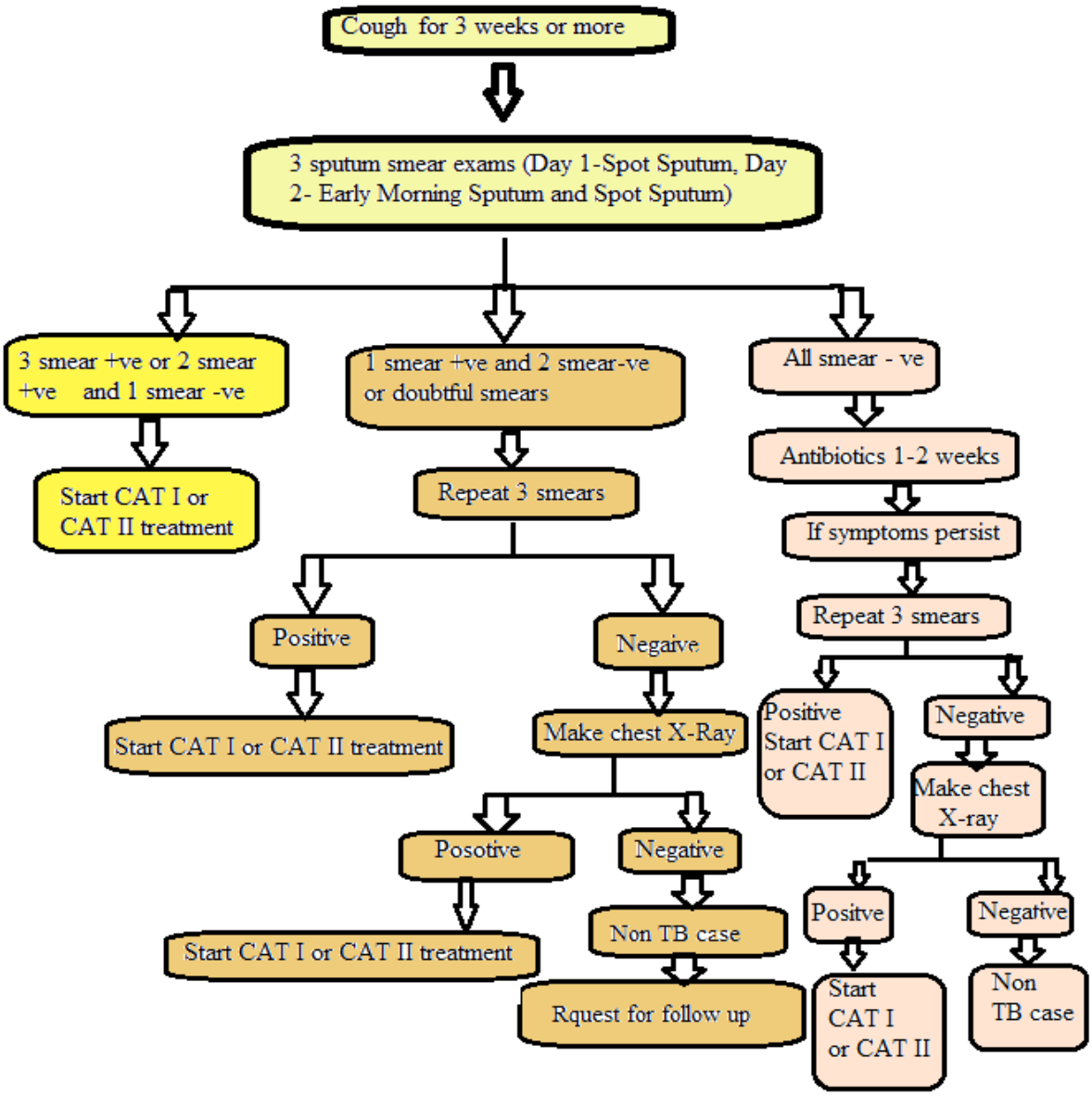

Figure 1.19: Sample and consequences of AFB microscopic examination (sputum smear microscopy).

1.7.2. Radiographic examination:

Although, the chest X-ray is a supportive diagnosis method of tuberculosis disease but it has considerable limitations. As many other conditions can show chest X-ray abnormalities which are more or less similar to that of TB, radiographic examination cannot be a confirmative technique to identify this disease. Instead of this, a chest x-ray(PA view) is

useful to enhance the confirmation of TB disease in a person having only one of three sputum smears that are positive or persistent TB symptoms with negative sputum smears. The term posterior/anterior (PA) means to the direction of the X-ray beam which moves across the patient from posterior (back) to anterior (front). To reduce magnification and improve sharpness the PA view is usually taken at a distance of six feet.

If X-ray abnormalities which are indicative of tuberculosis are found in a non-TB related medical observation then sputum specimens should be collected and recommended for AFB smear and culture examination to ensure suspicion.

Normally, the abnormalities are most common in upper lobes or apical segments of lower lobes.

The following presentations in X-ray are suggestive to active pulmonary tuberculosis-

- Infiltrates
- Nodules (are round shadows with clearly defined borders varying in size from a micro nodule having diameter less than 3 mm to a nodule having diameter more than 3 mm and less than 1 cm)
- Consolidation
- Cavities (A cavity is an area with a fairly thick wall more than 1 mm in which an area of bronchial drainage may be evident. Cavities sometimes contain liquid at the base)
- Fibrosis (John D. W., 2012)

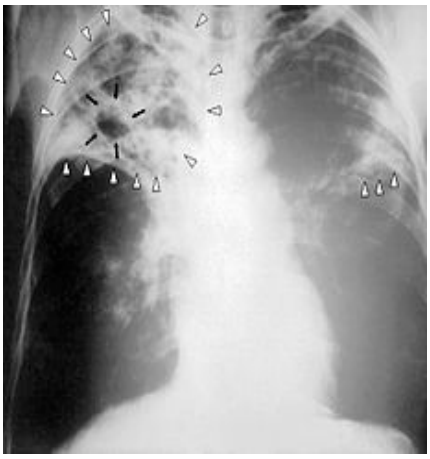

Figure 1.20: Chest X-ray of a person with advanced tuberculosis.

In the above figure, white arrow-heads are indicating ling infection and the black arrows are indicating the formation of a cavity.

The following presentations in X-ray are suggestive to previous or presumed inactive PTB include:

- Apical fibrosis
- Upper lobe fibronodular abnormality
- Pleural (fibro) calcification
- Upper lung zone bronchiectasis
- Thoracoplasty or partial pneumonectomy
- Healed primary lesion (Ghon focus / complex) (Manual for prevention and control of tuberculosis, 2010).

1.7.3. Tuberculin skin test (Mantaux Test): This test is mainly used as a supporting tool to the TB diagnosis in young children. It is mainly effective in children who do not take BCG vaccine. But in case of patients with BCG vaccine, this test will always provide a positive result. Therefore, in the context of Bangladesh, tuberculin skin test is not considered to be an effective way to identify tuberculosis either in adult or geriatric patients. In Bangladesh where there is population with high TB prevalence, the tuberculin skin test has minute value in the diagnosis of TB disease in adults. *M. tuberculosis* infection cannot be differentiated from TB disease by a positive Tuberculin Skin Test report. A false-positive test result can be obtained due to previous exposure to environmental mycobacterium. On the contrary, a false negative tuberculin skin test result can be obtained in TB patients due to severe malnutrition, miliary TB, HIV infection and other immuno-compromised condition.

1.7.3.1. Tuberculin skin test (TST) procedure: The TST is performed using the Mantoux method. In Mantoux method, small amount of tuberculin units (TU) of protein purified derivative (PPD) is intra-dermally injected into volar aspect (palm side) of the forearm. Multiple puncture skin testing devices should be avoided. The test is read within 48 to 72 hours by a trained health care worker, who looks for a reaction (induration) on the arm.

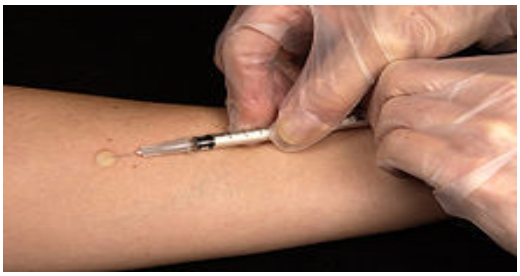

Figure 1.21: Tuberculin skin test.

The situations in which tuberculin skin test can be considered include-

- ❖ Recent contacts of persons who have, or suspected to have clinically active TB.
- ❖ In case of casual contacts, for example, visitors at home, at work or at clubs, tuberculin skin test could be performed only when the source case is regarded as

highly infectious such as laryngeal disease, strongly smear-positive pulmonary disease or endobronchial TB.

- Groups at high risk of recent infection with *M. tuberculosis* such as medically underserved populations, and personnel and long term residents in some hospitals, nursing homes, mental institutions and correctional facilities.
- Health care workers at risk of infection with *M. tuberculosis*.
- Recent immigrants especially those who have from countries with high rates of TB infection the first time
- Persons with HIV infection.
- Persons with abnormal chest X-rays suggestive of previous TB.
- Persons with other medical conditions that increase the risk of TB (Munsiff S. *et al*, 2005).

As stated before, it is not that much effective in case of Bangladesh as our country has high prevalence of tuberculosis; therefore; the physicians rely on other diagnosis procedures to identify the tuberculosis patients.

1.7.4. Culture of TB bacilli: Culture If resources permit and adequate, quality-assured laboratory facilities are available, culture should be included in the algorithm for evaluating patients with negative sputum smears. However, it takes about six weeks to provide a definite result, and is not accessible to most patients. Therefore, it is unsuitable as routine procedure. The probability of finding acid-fast bacilli in sputum smears by microscopy is directly related to the concentrations of bacilli in the sputum. Sputum microscopy is likely to be positive when there are at least 10 000 organisms per ml of sputum. At concentrations below 1000 organisms per ml of the sputum, the chance of observing acid-fast bacilli in a smear is less than 10%. In contrast, a properly performed culture can detect organism even concentrations below 100 organisms per ml.

Culture of TB bacilli is the most rigorous method of diagnosing tuberculosis. The specificity and sensitivity of this test is much higher than smear microscopy as each live bacillus forms colonies on culture. The equipment and running costs for performing culture are much higher than those for microscopy; culture also necessitates a high level of training of laboratory technicians (American thorasic society, 2000).

1.7.5 FNAC: These are special tests performed to confirm extra pulmonary TB to be referred to concerned specialists (WHO, 2008). Fine-needle aspiration cytology (FNAC) is a simple

and cost-effective tool having high diagnostic accuracy for tuberculous lesions. FNAC is used for the diagnosis of tuberculous lesions presenting as

- Superficial lumps and bumps
- Ulcers
- Sinuses
- And for deep-seated, space-occupying lesions under imaging guidance.

As in histology, FNA cytodiagnosis of mycobacterial diseases depends on demonstration of epithelioid granuloma and/or necrotic material in the smear (Das D. K., 2000).

1.7.5 Novel detection method of tuberculosis disease: One of the major research areas for tuberculosis (TB) focuses both on the diagnostics and biomarkers that can provide prognostic data about the disease course and response to treatment. Young children are at high risk of progressing to TB after exposure, and may suffer from disseminated forms of the disease. A novel B-cell assay has been tested and evaluated called the antibodies in lymphocyte supernatant, or ALS, which has performed very well in diagnosing TB disease both in Asia and Africa (manuscript in preparation).

The principle of ALS assay is similar to that of the enzyme-linked immunosorbent spot assay, measuring antibody-secreting cells in cultures of peripheral blood mononuclear cells (PBMCs). The ALS assay detects antibody secretion from in vivo activated plasma B cells that migrate throughout the peripheral circulation in response to TB antigens that are present during active disease but not latent TB infection. In the study performed in icddrb, the steps of ALS methodology for children included

Step 1: Phlebotomy of 3.5 ml of blood in order to isolate 5 million PBMCs.

Step 2: Then these cells are incubated in tissue culture plates without stimulation for 48–72 h.

Step 3: After that, the supernatant is collected, placed into BCG-coated microtitre plates.

Step 4: Finally, the IgG responses to BCG are measured by ELISA.

A positive titer is defined as ≥0.350 optical density units, calculated from data on healthy children.

In its present format, the method requires skilled personnel and equipment for PBMC separation from blood samples, sterile cell cultures and ELISA (Thomas T. *et al*, 2011).

1.8 Treatment of tuberculosis

Although treatment of tuberculosis varies according to age, body weight but more of less five anti-TB drugs are mainly prescribed to give proper treatment to a TB patients which are-

- ❖ Isiniazid
- ❖ Rifampicin
- ❖ Streptomycin
- ❖ Pyrazinamide
- ❖ Ethambutol

Isoniazid and refampicin mainly act as bactericides whereas streptomycin and pyrazinamide provide complementary bactericidal action. Ethambutol is a bacteriostatic drug. The action of ethambutol is associated with bactericidal action in order to avoid the emergence of resistance.

Abbreviations of these drugs are used internationally when these are being prescribed. The abbreviation of each drug is given below:

Table 1.2: Name of the drug used and their abbreviation

Name of the drug	Abbreviation
Isiniazid	H
Rifampicin	R
Streptomycin	S
Pyrazinamide	Z
Ethambutol	E

1.8.1 Treatment phases: Effective treatment of tuberculosis can be divided into two phases likewise –

(a) Initial or intensive phase

(b) Continuation phase

Short description of each phase are mentioned below-

(a) The initial or intensive phase: In initial phase, drugs are administered daily for

- ❖ Two months in new cases.
- ❖ Three months in re-treatment cases.

The major purpose of this phase is to

- ❖ rapidly diminish and eliminate the multiplication of bacilli
- ❖ restrict the development of acquired resistance to the prescribed drugs.

During the intensive phase, the tubercle bacilli are killed rapidly and the infectious patients quickly (within approximately two weeks) become non-infectious as well.

(b) The continuation phase: During this phase, drug is administered daily.

- ❖ to complete the rest of the treatment duration according to category
- ❖ and to eliminate the remaining bacterial population.

According to WHO, the standard treatment regimen for diagnosis category is as follows-

Table 1.3: The standard treatment regimen for diagnosis category

TB diagnosis category	Patient category	Treatment regimen	
		Intensive phase (daily)	Continuation phase (daily)
CAT I	• New smear-positive patients • New smear- negative PTB • Extra- pulmonary TB • Concomitant/associated HIV/AIDS • Sputum smear-positive PTB with history of treatment of more than one month	2(HRZE)	4 (HR)
CAT II	• Relapse • Treatment failure after Cat. I • Treatment after default • Others	2(HRZE)S / 1(HRZE)	5 (HR)E

Here the numbers indicate the length of treatment in month and two (or three or four) drugs within brackets indicates that fixed dose combinations are being used.

As fixed dose combination (FDC) incorporates 2 to 4 different drugs within same tablets, it provides great advantages to-

- ❖ Improve adherence
- ❖ Avoid patients taking only a part of prescribed drugs.

1.8.2 Dosages of FDC tablets: FDC tablets are composed as follows:

Table 1.4: Combination of FDC tablet

FDC	Combination
4-FDC	rifampicin 150 mg + isoniazid 75 mg + pyrazinamide 400 mg + ethambutol 275 mg
2-FDC	rifampicin 150 mg + isoniazid 75 mg

The dosages of FDC tablets vary according to the pre-treatment weight of the patients in both CAT I and CAT II treatment. Dosages according to pre-treatment weight which have to be prescribed are shown in following two tables.

Table 1.5: The dosages of FDC tablets according to the pre-treatment weight of the patients in CAT I treatment.

	Category I	
	Intensive Phase	Continuation Phase
Pre-treatment weight (kg)	Daily (First two months)	Daily (Next 4 months)
	Number of 4FDC tablets	Number of 2 FDC tablets
30 – 37	2	2
38 – 54	3	3
55 – 70	4	4
> 70	5	5

Table 1.6: The dosages of FDC tablets according to the pre-treatment weight of the patients in b CAT II treatment

Category II				
	Intensive phase		Continuation phase	
Pre-treatment weight (kg)	Daily (first 3 months)	Daily (first 2 months)	Daily (next 5 months)	
	Number of 4FDC Tablets	Injection Streptomycin in	Number of 2FDC tablets	Ethambutol 400mg (Number of tablets)
30 – 37	2	500mg	2	2
38 – 54	3	750mg	3	3
55 – 70	4	≤750 mg	4	3

In case of adult 1mg of streptomycin can be prescribed.

A very important feature of the tuberculosis control is the adherence to treatment. As a proportion of patients stop treatment before completion, for various reasons therefore strict adherence to treatment should be ensured. It is needed in order to cure the patients and prevent the development of drug-resistant TB. To obtain this goal, Directly Observed Treatment (DOT) is a very vital component in the internationally recommended policy package for TB control. It is knows as DOT strategy.

DOT ensures that the right anti-TB drugs, in the right doses, at the right intervals and for the right period are being administered by the TB patients. According to the DOT strategy, all patients, irrespective to the treatment category, should receive all doses of the anti-TB drugs (WHO, 2008).

1.9 Incidence of tuberculosis in global aspects:

Tuberculosis (TB) is a major public health problem in Bangladesh since long. Estimates suggest that daily about 880 new TB cases and 176 TB deaths occur in the country (WHO, 2008). For centuries all over the world tuberculosis has been a major killer. Still now, it ranks

as one of the greatest health problems. Tuberculosis (TB) is second only to HIV/AIDS as the greatest killer worldwide due to a single infectious agent. According to WHO, in 2010, 8.8 million people had been affected with TB and 1.4 million died from this infectious disease. Over 95% of TB deaths occur in low-income and middle-income countries (WHO, 2012). Tuberculosis is more likely to affect people in poor countries those may be malnourished and have poor access to healthcare.

The following 22 countries accounts for 80% of the TB cases in the world-

1. Afghanistan
2. Bangladesh
3. Brazil
4. Cambodia
5. China
6. Democratic Republic of Congo
7. Ethiopia
8. India
9. Indonesia
10. Kenya

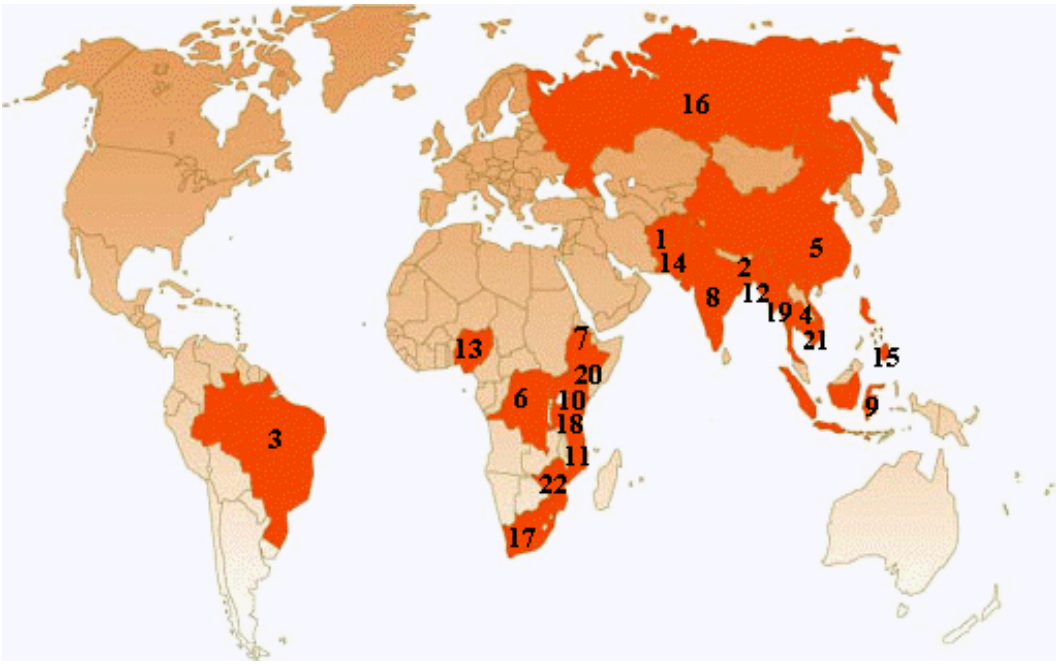

Figure 1.22: Twenty two countries those account for 80% of the TB globally.

11. Mozambique
12. Myanmar

13. Nigeria
14. Pakistan
15. Philippines
16. Russia
17. South Africa
18. Tanzania
19. Thailand
20. Uganda
21. Viet Nam
22. Zimbabwe (WHO, 2005).

The incidence of tuberculosis disease is increasing alarmingly. According to WHO, statistics shows that occurrence of TB is remarkable enough to be aware. WHO reported that TB cases are being increased with time. In 1980 the rate of TB cases was 1000000 where as in 2005 it turned 5000000. That means, in 25 years the incidence of TB increased five times (Vidula M., 2009).

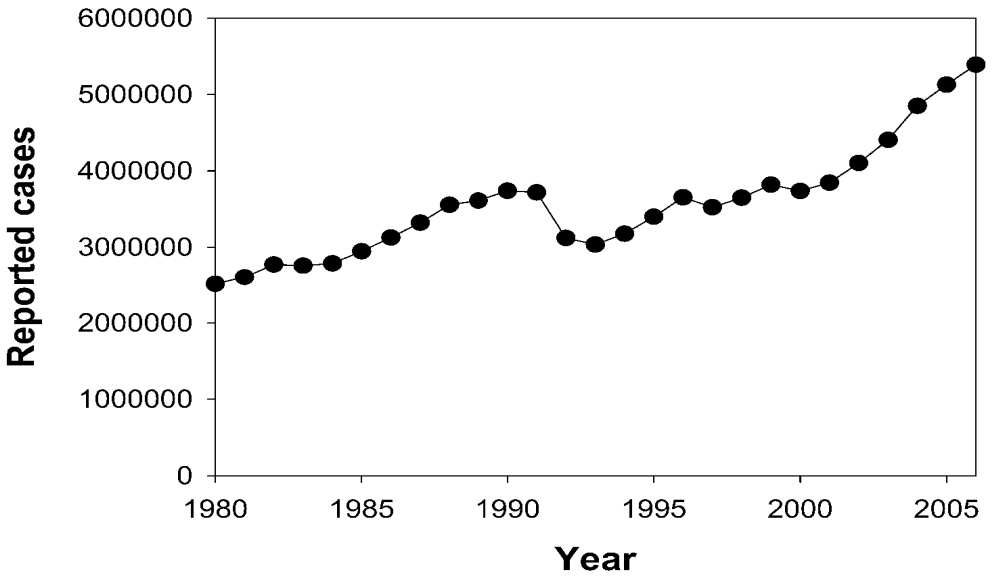

Figure 1.23: Incidence of TB.

TB alone took more than 200 million lives in the last century. The disease now spreads at the rate of one person per second or claiming lives of 2,000 people per day in the South East Asian region. Official estimates say that TB infects one person every two minutes and kills one person every 10 minutes in Bangladesh. Over 300,000 people are being infected with this preventable disease every year, the rate being 111 per 100,000. The rate of infection was less

than 40 per 100,000 ten years back (Banglapedia). Bangladesh ranks sixth among the 22 high burden countries of TB (Chowdhury D. F. A.,2012).

Estimation is done by WHO that provides the following statistic about tuberculosis over the world.

Table 1.7: Estimated WHO regional statistic for 2010

Region	Incidence	Prevalence	Deaths	Population
Africa	2,300,000	2,800,000	250,000	836,970,000
Americas	270,000	330,000	20,000	933,447,000
Eastern Mediterranean	650,000	1,000,000	95,000	596,747,000
Europe	420,000	560,000	61,000	896,480,000
South-East Asia	3,500,000	5,000,000	500,000	1,807,594,000
Western Pacific	1,700,000	2,500,000	130,000	1,798,335,000
Global Total	8,840,000	12,190,000	1,056,000	6,869,573,000

1.10 Significance of the study:

As tuberculosis is a great concern all over the world, therefore, control of this disease is essential. Bangladesh is more vulnerable to TB than the developed countries. Data shows that incidence of all TB cases is 227/100K/YR and incidence of new sputum smear positive (SS+ve) TB cases is 102/100K/YR. 70,000 deaths occurs per year due to TB in our country (Zaman K *et al*, 2006). The statistical data demonstrates that people of our country are at high risk of getting contaminated with the causative agent TB. Not only for detection rather for monitoring the effectiveness of medication as well as level of improvement follow up AFB microscopy tests should be performed to monitor the level of improvement. As it is a contagious disease, proper management should be taken to cure the infected person along

with to prevent the transmission from him/her to others. In order to achieve proper management, the patients have to be monitored thought out the course of the treatment period. Follow up AFB microscopy test will confirm the condition of the patients that is whether they are improving with medication or turning to MDR (Multi Drug Resistance) tuberculosis. In this context, this study “AFB microscopy examination of tuberculosis patients (50 years old or above) admitted in National Institute of Diseases of the Chest and Hospital (NIDCH)” is worthy enough to perform.

Chapter 2

Methods and Materials

2.1 Methodology:

To carry out the study, permission from the hospital director has been collected. The fundamental study has been started by taking data from the hospital management authority and form patients as well. To collect the diagnosis reports such as AFB microscopy report and haematological test report of the sample patients, the hospital authority has been considered as the source of information.

The major focus of this study has been given to outcome of AFB microscopy diagnosis test of patients with advanced age. The aim has been to observe the AFB test report after one month of first AFB report. By comparing these two outcomes, the level of progression in case of each patient and the awareness among the patients about the management of tuberculosis along with follow up can be estimated.

The follow up test has also been done in the National Institute of Diseases of the Chest and Hospital (NIDCH) with the aid of nurses. A form had to be filled up with the registration number, patients' name, age, and other necessary information to do the AFB microscopy test. Then sputum sample were collected by the nurses. Three specimens for each patient have been collected namely Spot specimen, Early morning specimen, and Spot specimen. The report of AFB microscopy test ensures whether the patient is smear positive tuberculosis patient or not. The form of AFB microscopy test is being attached here.

Some additional information like weight, gender, accommodation, smoking habit, educational level and many others have also been accumulated from the hospital but these information have also been cross-checked by asking the patients individually.

To have an overall idea about the awareness and living condition responsible for their disease condition, a number of additional information have also been brought together by preparing a questionnaire and asking them to the sample patients. That questionnaire includes the vaccination information, other family member having tuberculosis, literacy state occupation and so on.

2.2 Study Place:

The study has been conducted at National Institute of Diseases of the Chest and Hospital (NIDCH). National Institute of Diseases of the Chest and Hospital (NIDCH) is a state supported research institute and hospital in Bangladesh.

2.3 Inclusion criterion:

In this research project only pulmonary tuberculosis patients were enrolled. And the focus of the project has been given to-

- AFB microscopy examination and the follow up out come

In addition to this, some other information have also been collected likewise-

- Haematological report
- Socio-demographic data.

The patients who has been enrolled in this study were-

- Pulmonary tuberculosis patients
- Patients of 50 years old or above

2.4 Exclusion criteria:

From this study following patients have been excluded

- Patients with extra pulmonary tuberculosis
- MDR patients
- Patients with age of below 50 year.

2.5 Study type:

The study type of this project can be defined as preliminary prospective study on the tuberculosis patients of 50 or above 50 years old.

2.6 Study period:

This prospective study has been conducted about one year of time.

2.7 Sample size:

Total sample volume was taken on 20 patients among them 17 were male and 3 were female.

2.8 Sampling technique: The patients were taken in a random manner.

2.9 Study population:

The study population has been belonging from the tertiary hospital that is National Institute of Diseases of the Chest and Hospital (NIDCH). Almost all of them were from deprived societal areas. The study population were of age of 50 years old or above. Most of them were from rural or semi-urban area.

2.10 Data analysis:

Data of the entire study have been analysed by using Microsoft Excel. To give the visual representation, bar diagram, column and pie chart have been used.

Chapter 3

Result

3.1Some basic information of the patients:

3.1.1. Age of patients: Distribution of tuberculosis patients according to age are shown in the following table.

Table 3.1: Distribution of tuberculosis patients according to age

Age Range	Number of Patients
50-59 years	11
60-69 years	05
70-79 years	03
80-90 years	01

After the completion of the study on the occurrence of tuberculosis at different age range of patients (with age 50yrs or above), a basic overview described by Table 3.1 suggests that there are 11 patients have tuberculosis within 50-59 years; 5 patients within 60-69 years; 3 patients within 70-79 years and 1 patient within 80-90 years. This phenomenon was observed due to the presence of higher number of patient in the hospital within the age range of 50-59 years that was 11 patients in the study. As mean age of our country people is 59 years, so most of the patients in the hospital are found within 50-59 years and these people are exposed to polluted environment on their working place. Above 60 years, generally people do not go for work may be the reason of finding this type of analysis.

3.1.2 **Weight of patients:** Distribution of the patients of 50 years old or above according to their body weight is being shown with pie chart.

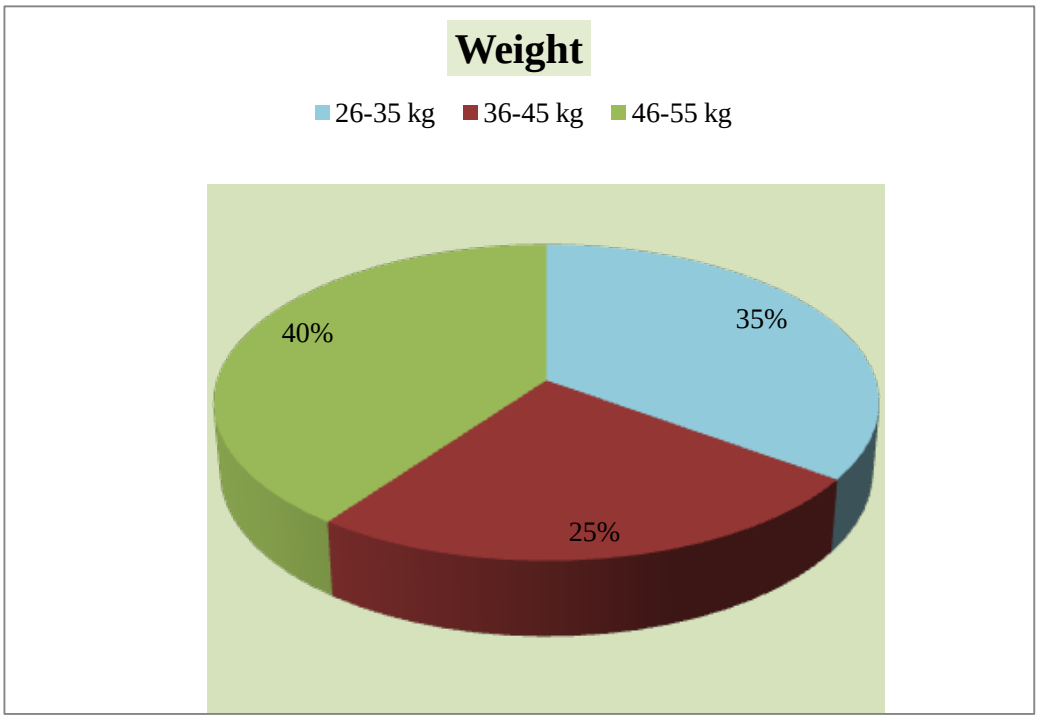

Figure 3.1: Distribution of the patients of 50 years old or above according to their body weight.

In the study, tuberculosis occurred in 7 patients whose average weight was within 26-35 kg; 5 patients within 36-45 kg and 8 patients within 46-55 kg. Due, tuberculosis normally weight loss occurs but in this study a reverse view has been found. It may be happened due to small sample volume or in advanced age normally people get bulkier. So the result of study, observed 8 patients their average weight is 46-55 kg done on the following pie chart.

3.1.3 Gender of patients: Distribution of the patients according to gender their gender is being described with help of following tabulation.

Table 3.2: Distribution of the patients according to gender

Gender	Number of patients
Male	17
Female	3

From 20 patients, the study illustrates that 17 patients were male and 3 patients were female. The data shown in the tabulation represents higher number of male patient and reasoning behind this is quite simple. As most of the male people are gone outside for work on their working place, they are easily exposed to the contaminated air which is associated with *Mycobacterium tuberculosis*. On the other hand, male have smoking tendency that increases the incidence of tuberculosis than that of female. Such case is rarely found on female.

3.1.4 Vaccination: Distribution of the patients according to immunization with BCG vaccine is being demonstrated in the following table.

Table 3.3: Distribution of the patients according to immunization with BCG vaccination

Features	Number of patients
Immunized with BCG	1
Non- immunized with BCG	0
Don't know their immunization status	19

BCG (Bacilli Calmette Guerin) vaccine was taken by patient found in 1 patient but here 19 patients did not know whether or not they were immunized with such type of vaccine. Most of the people in Bangladesh suffering from tuberculosis are not conscious enough about different types of diseases as well as vaccine or sera also. For this reason even though they

were immunized did not know about this, such result shown by a bar diagram. This data shows that how much careless the people are about their health concern.

3.2 Some additional information about patients:

3.2.1 Smoking tendency: The smoking habit and the initiative after getting tuberculosis with smoking habit of the patients are illustrated by the following table.

Table 3.4: Smoking history of the patients

Feature	Number of patients
Patients having smoking habit before developing TB disease	17
Patients who quit smoking habit after developing TB disease	17

Smoking may be a risk factor that worsens the condition in case of tuberculosis. The table gave an overview that from the 20 patients, 17 patients had smoking habit before infected by TB whereas only 3 patients (female) did not smoke before getting infected by TB. The table shows that 17 patients who quit smoke after having TB and as 3 patients did not have smoking habit all of the patients quitted smoking after infected by TB. Smoking does not influence the growth of or pathogenesis of tuberculosis but it degenerates the respiratory system that aggravate condition accelerating the development of TB disease

3.2.2. Risk factors: Other risk factors patients having along with tuberculosis are shown below with a bar diagram.

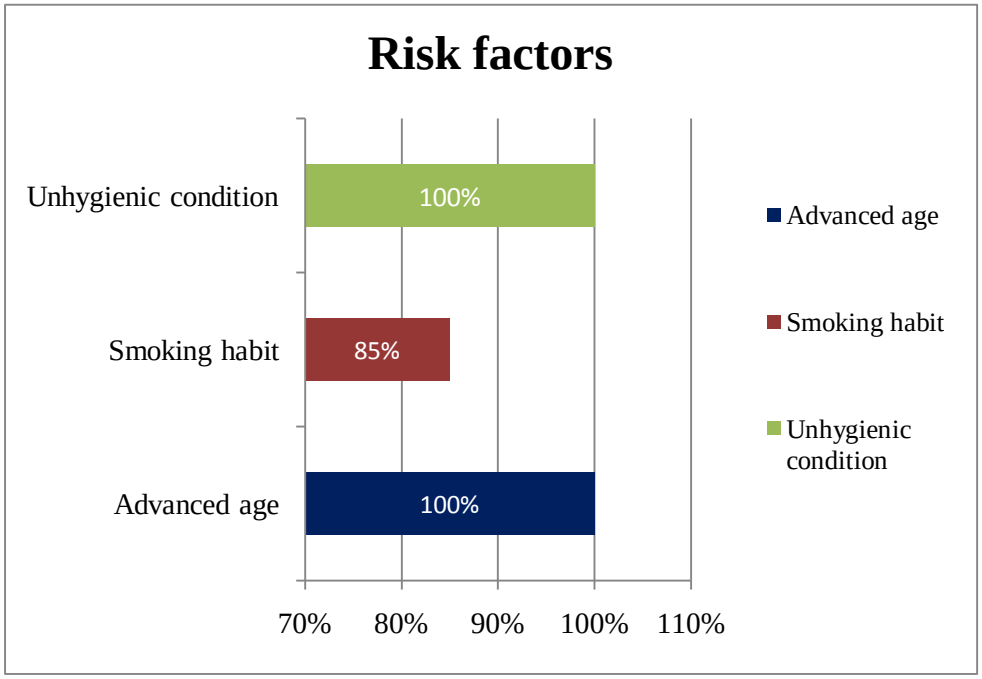

Figure 3.2: Distribution of patients according to other risk factors.

In case of considering other risk factors associated with the tuberculosis patient, all of the patients of the tested sample were living in an unhygienic condition. The advanced age is another important risk factor among all patients as well. All of the patients (100%) were 50 years old or above. Because of being very elderly some of them could not maintain personal hygienic properly. Besides, 17 patients (85%) had the smoking habit that was described earlier and and 20 patients (100%) were with unhygienic practice.

3.2.3. Education level: Distribution of the patients according to their educational level is shown by a column diagram hereunder.

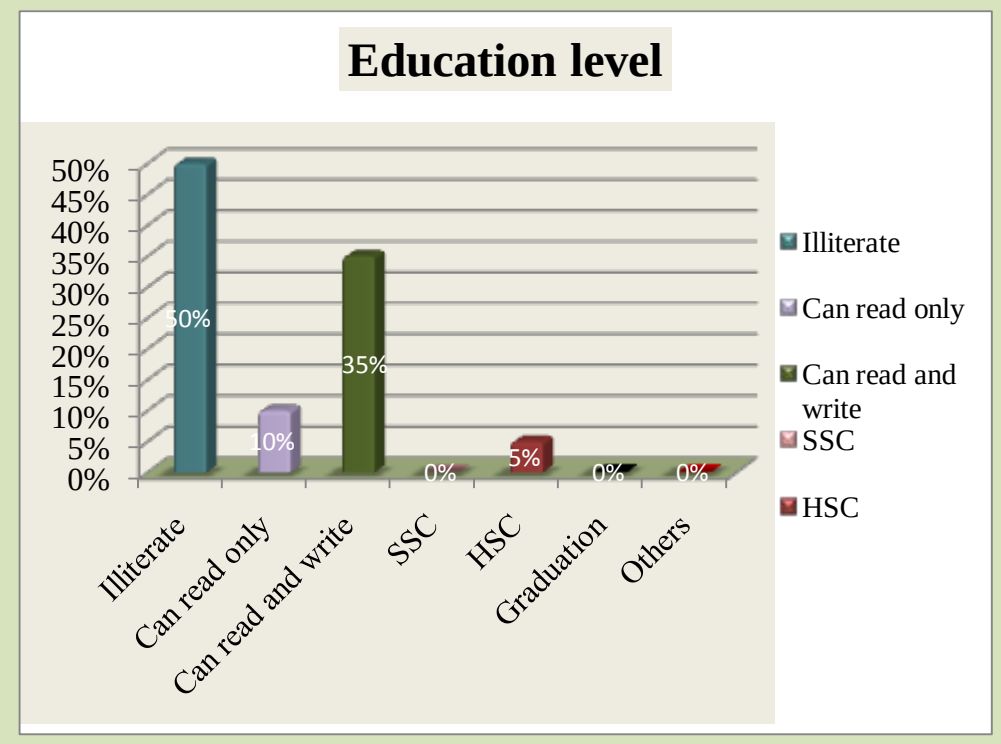

Figure 3.3: Distribution of the patients according to their educational level

From the 20 patients, 10 patients were illiterate (50%), 2 patients (10%) can read only, 7 patients (35%) can read and write and from which 1 patient (5%) pass HSC examination. In the educated patient no one (0%) have graduation or other degrees. So from this data most of the patient are illiterate and did not know enough about dangerous view of the tuberculosis. On other hand, as they due to having no or very small knowledge of tuberculosis they do not take enough prevention technique and spread it to others because of carelessness. Due to the lack of consciousness in the illiterate people tuberculosis became more prominent on them.

3.2.4 Working disability and family history of TB: Distribution of the patients according to the working disability and other members having (and/or had) TB disease is illustrated in following table.

Table 3.5: Distribution of patients according to working disability and family history of TB

Feature	Number of patients
Patients with working disability	20
Patients without working Disability	00
Patients having other family members with TB	03
Patients not having other family members with TB	17

From the analysis, all of the 20 patients have working disability after the infection with tuberculosis and no patient are found without working disability. Different types of working disability are occurred in case of patients who are affected by tuberculosis most vulnerable is respiratory disability. In this study another reason of working disability was there advanced age.

From the 20 test samples, there were 3 patients having family member affected with tuberculosis and 17 patients have no family member who were affected by tuberculosis. Generally tuberculosis is not a genetic disorder. There is no evidence of people suffered by tuberculosis if it is occurred in any family member. Three among 20 patients showed to have family history of tuberculosis perhaps as it is a contagious disease and transmitted from one to another.

3.2.5 Living condition of patients: Distribution of the patients according to living condition is shown below with the help of a column diagram.

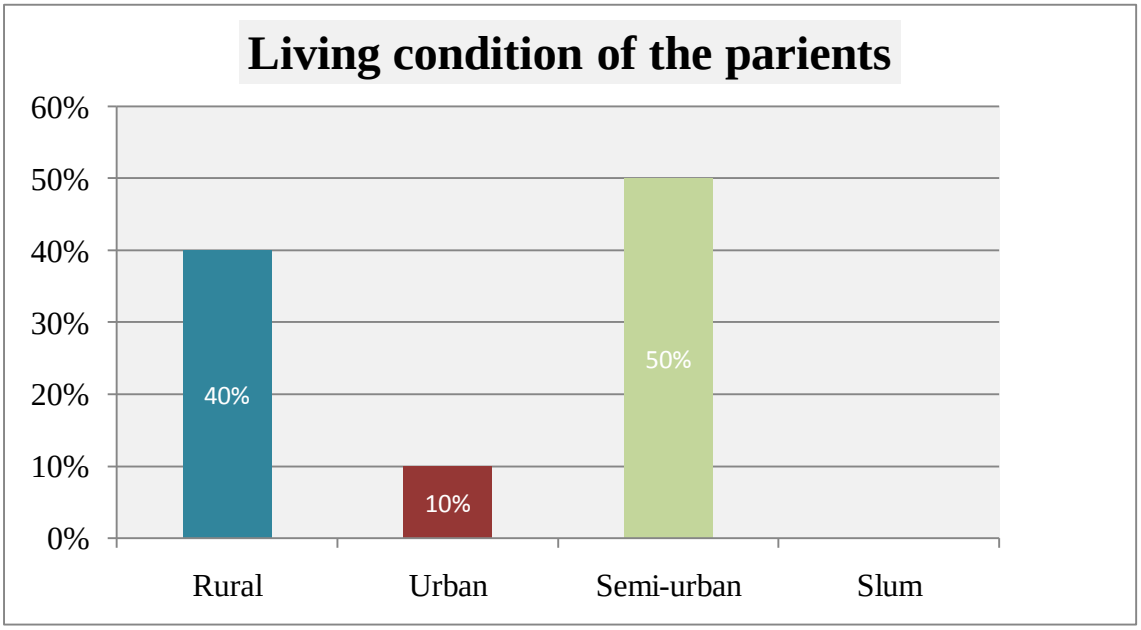

Figure 3.4: Distribution of the patients according to living condition

There were 8 patients (40%) who were living in rural; 2 patients (10%) in urban; 10 patients (50%) in semi-urban areas from the 20 patients. No patient (0%) was from the slum area. Mainly patient from the rural and semi-urban area are not well concerned about the disease as well as these areas are mainly exposed to the polluted environment which is a great source of microbial contamination. Due to the higher number of patient from semi-urban area are found in the hospital the column diagram gave such report.

3.2.6 Living status: Distribution of the patients according to the living status is being shown in the following table.

Table 3.6: Distribution of the patients according to the living status

Living status	Number of patients
Rich	0
Upper middle	1
Lower middle	8
Poor	11

In the study, from the 20 patients 1 patient was from upper middle, 8 from lower middle and 11 patients were from the poor family. There was not a single patient from the rich family. As the 85% people of Bangladesh are poor, most of them do not know what tuberculosis is, what are its symptoms and what is the corrective and preventive measure that should be taken. They are not healthier due to needy condition of their lives.

3.3 Diagnosis outcomes of the patients

3.3.1 AFB microscopic examination

3.3.1.1. **Result of spot sample (1st specimen):** The result of the first specimen of all patients are being stated in the following table.

Table 3.7: Result of 1st specimen of all the patients

Result of 1st specimen	Number of patients
Negative	3
Scanty	3
1+	3
2+	6
3+	5

In the 1st specimen of AFB microscopy test analysis, 3 patients showed negative; 3 patients showed scanty, 3 patients showed +1; 6 patients showed 2+ and 5 patients showed 3+ results which were described by the following pie chart. Although 3 shows negative in the result of first specimen in the AFB microscopic examination but they two of them have been shown to be positive in the 2nd or third 3rd specimen. As the total result is considered by taking all the result of three specimens so that among these three patients two are smear positive pulmonary tuberculosis patients.

3.3.1.2 Result of early morning sample (2nd specimen): The result of the second specimen in AFB microscopy test of all patients is being illustrated with bar diagram below.

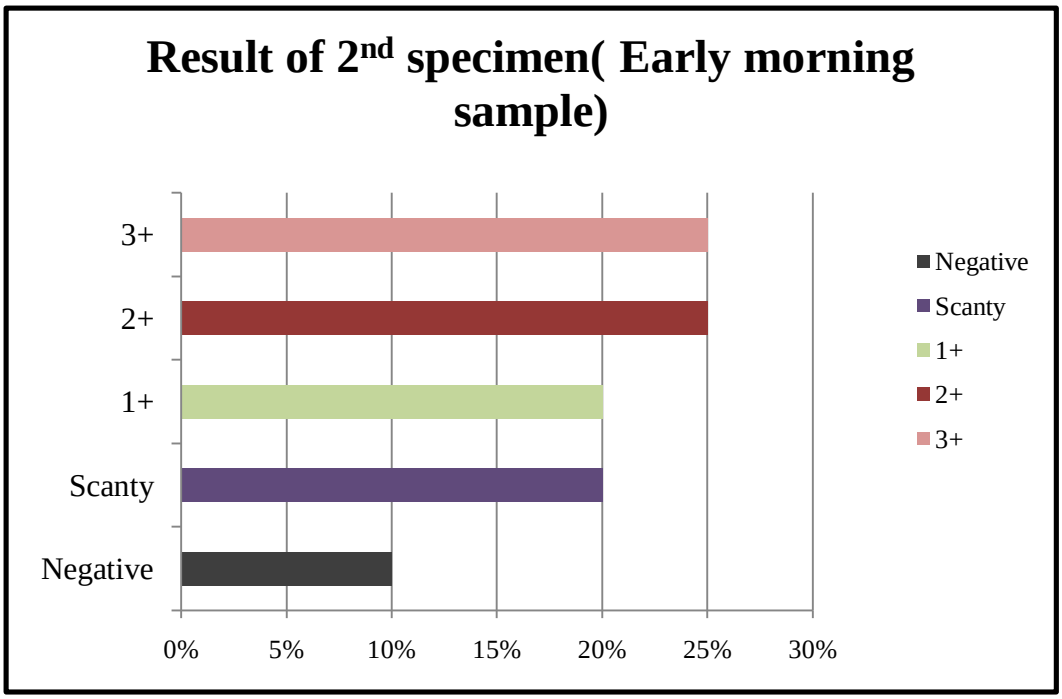

Figure 3.5: Result of early morning sample (2nd specimen)

In the 2nd specimen (early morning sample) of AFB microscopic test analysis, 2 patients(10%) showed negative; 4 patients (20%) showed scanty, 4 patients (20%) showed +1; 5 patients (25%) showed 2+ and 5 patients (25%) showed 3+ results that was described by the following bar diagram.

3.3.1.3. Result of spot sample (3rd specimen): The result of the second specimen in AFB microscopic examinaion of all patients is being illustrated with column diagram below.

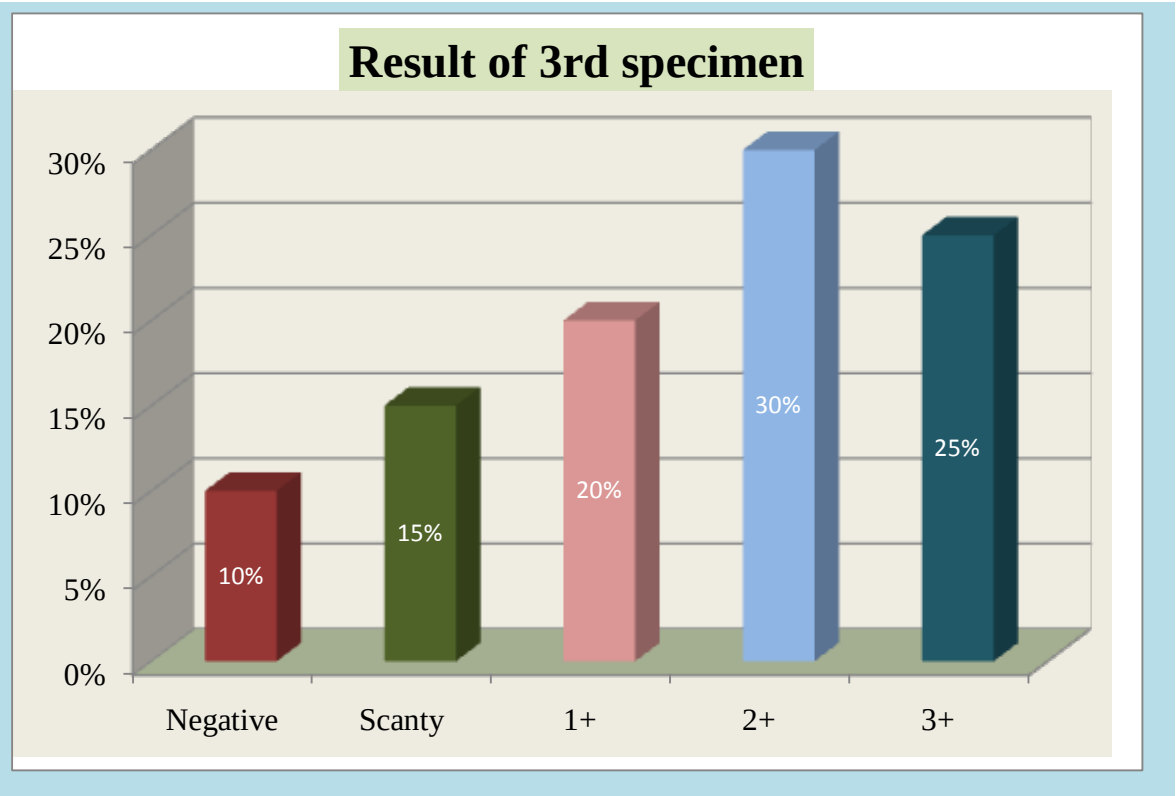

Figure 3.6: Result of early morning sample (3rd specimen).

In the 3rd specimen of AFB microscopy test analysis, 2 patients(10%) showed negative; 3 patients(15%) showed scanty, 4 patients(20%) showed +1; 6 patients(30%) showed 2+ and 5 patients (25%)showed 3+ results that was described by the following bar diagram.

So from the investigation, there was one patient who had smear negative tuberculosis as he showed negative result in all specimens and other 19 patients showed smear positive tuberculosis as they showed positivity in any of the following specimens.

3.3.2. Haematological/CBC test analysis:

3.3.2.1: Haematological and ESR report: The haematological with ESR report of patients are shown in the following table.

Table 3.8: Haematological with ESR report of all patients

Test Name	**Number o the patients**			**Unit**	**Ref. Value**
	Within Normal Range	**Above Normal Range**	**Below Normal Range**		
Hb	3	0	17	gm/dl	Male : 12.5-17.5 Female : 11.5- 16.5
Total count of WBC	10	10	0	/cmm	3,900-10,000
ESR	2	18	0	mm/1st hr	Male: 12 mm or less Female: 15mm or less
Platelet count (PC)	4	16	0	/cmm	140,000- 390,000
PCV	2	0	18	%	Male: 40.0-52.0 Female : 35.0-47.0

Table: 3.8 represents a gross report on the level of haemoglobin (Hb) level in the blood. 3 patients had the haemoglobin level within the normal range that is 12.5-17.5 gm/dl for male and 11.5-16.5 gm/dl for female; 17 patients had the haemoglobin level below the normal range. There was no patient having the haemoglobin content above the normal range that means haemoglobinamea. So in most cases haemoglobin level was below the normal range that means tuberculosis may lead to anaemia.

From the study of the CBC (Complete Blood Count) test in between 20 patients, it was possible to measure the total count of WBC (White Blood Cell). Ten patients had the total WBC count within the normal range that is 3900-10000 /cmm and 10 patients had the total WBC count above the normal range. There was no patient with the total WBC content below the normal range. So the normal range of total count of WBC and above the normal range is similar in the test patients. That means there is no effect of TB disease on the total count of WBC.

Another crucial feature of CBC (Complete Blood Count) test is ESR (Erythrocyte Sedimentation Rate) level count. From the 20 patients of test sample, 2 patients had the ESR level within the normal range that is 12 mm or less/ 1^{st} hour for male and 15 mm or less/ 1^{st} hour for female; 18 patients had the ESR level above the normal range. There was no patient having the ESR level content below the normal range. So in case of ESR level report, the most tubercular patients have the ESR level above the normal range which can be an indication of the tuberculosis disease.

In the CBC (Complete Blood Count) test of the platelet count from the 20 patients, 4 patients have the total platelet count within the normal range that is 140,000-390,000 /cmm and 16 patients have the total platelet count above the normal range. There was no patient having the total platelet count content below the normal range. So, it is clear that tuberculosis has impact on the total platelet count of the patient.

Finally, the most common test of CBC (Complete Blood Count) is checking the PCV (Packed Cell Volume) level. In the analysis from the 20 patients, 2 patients were with the PCV level within the normal range that is 40.0-52.0% for male and 35.0-47.0% for female; 18 patients were with the PCV level below the normal range. There was no patient having the PCV level content above the normal range. The PCV level was found below the normal range associated with the tested tubercular patients.

3.3.2.2. Differential WBC (white blood cell) count of all patients: The report of the differential WBC (white blood cell) count of all patients are illustrated with the help of tabulation as bellow-

Table 3.9: Differential WBC (white blood cell) count of all patients

Test Name	Number of patients		Unit	Ref. Value
	Within Normal Range	Out of Normal Range		
Neutrophil	11	9	%	40 – 75
Lymphocytes	7	13	%	20 – 47
Monocytes	16	4	%	02 – 10
Eosinophils	13	7	%	00 – 01
Basophils	18	2	%	00 – 01

Neutrophil level test is under the test of differential count of WBC (White Blood Cell) test. From the viewpoint of the study among the 20 patients, 11 patients had the neutrophil level within the normal range that is 40-75% and 9 patients were out of the normal range. In this case, the patients had normal range of neutrophil level. So there may be no relation of neutrophil level with the tubercular patients.

In case of lymphocyte level table 3.9 represents that among the 20 patients, 7 patients had the lymphocyte level within the normal range that is 20-47% and 13 patients were out of the normal range. So in the most of the tubercular patients, the lymphocyte level was out of the normal range.

Monocyte level test is also under the test of differential count of WBC (White Blood Cell) test. From the viewpoint of the study among the 20 patients, 16 patients had the monocytes level within the normal range that is 2-10% and 4 patients were out of the normal range. In this case the patients had normal range of monocyte level. So there may be no relation of monocyte level with the tubercular patients.

In case of eosinophil level, data showed that among the 20 patients, 13 patients had the eosinophil level within the normal range that is 0-1% and 7 patients were out of the normal range. In this case, the patients had normal range of eosinophil level. So there may be no relation of eosinophil level with the tubercular patients.

Finally, data of the basophil level test which is also under the test of differential count of WBC (White Blood Cell) test represents that among the 20 patients, 18 patients had the basophil level within the normal range that is 0-1% and 2 patients were out of the normal range. In this case, the patients had normal range of basophil level. So there may be no relation of basophil level with the tubercular patients.

3.4.Result of follow up patients:

Patient number: 01

A male patient with age 74 years had diagnosed for tuberculosis on 21st of April, 2012. The report of AFB microscopic examination showed smear positive pulmonary tuberculosis. All of the specimens resulted to be "3+". After one month of treatment with anti-TB, the AFB microscopy test had been done again on 21st of May, 2012 to observe whether he was improving or not. The follow up result showed 1+ for all specimens. The result of both initial and follow up AFB microscopic examination are given below.

Table 3.10: Outcome of the first AFB microscopic examination of patient 01

Date	Specimen	**Result**				
		Negative	Scanty	1+	2+	3+
21.04.2012	1.					√
	2.					√
	3.					√

Table 3.11: Outcome of the follow up AFB microscopic examination of the patient 01 after one month

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
21.05.2012	1.			√		
	2.			√		
	3.			√		

From the follow result, it can be concluded that this patient is improving slowly.

Patient number: 02

This patient was a 52 years old male who had diagnosed for tuberculosis on 8th of April, 2012. From the report of AFB microscopic examination, it was evident that he had smear positive pulmonary tuberculosis. The AFB microscopy result of the first specimen was "3+" and both the second and third specimen showed to be "1+". For the follow up study, the AFB microscopy test had been taken again on 2nd of May, 2012 to observe whether he was improving or not. The follow up result showed all negative for all specimens. The result of both initial and follow up AFB microscopic examination are given below.

Table 3.12: Outcome of the first AFB microscopic examination of patient 02

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
08.04.2012	1.					√
	2.			√		
	3.			√		

Table 3.13: Outcome of the follow up AFB microscopic examination of patient 02 after almost one month

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
02.05.2012	1.	√				
	2.	√				
	3.	√				

This patient was improved properly with anti-TB treatment. So, after completing the full course of anti-TB therapy he will get rid of disease properly.

Patient number 03:

A male patient with age 55 years had diagnosed for tuberculosis on 11th of April, 2012. The report of AFB microscopy was smear positive pulmonary tuberculosis. All of the specimens resulted to be "2+". After almost one month of treatment with anti-TB, the AFB microscopic examination had been performed again on 19th of May, 2012 to observe the level of improvement. The follow up result showed negative for 1st specimen, "2+" for the second specimen and 1+ for the third specimen. The result of both initial and follow up AFB microscopic examination are given below.

Table 3.14: Outcome of the first AFB microscopic examination of patient 03

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
11.04.2012	1.				√	
	2.				√	
	3.				√	

Table 3.15: Outcome of the follow up AFB microscopic examination of patient 03 after almost one month

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
19.05.2012	1.	√				
	2.				√	
	3.			√		

The follow up result of this patient is a bit hazy, but it can be assumed that he is responding to the anti-TB treatment very slowly. To come to any clear statement about the level of improvement further follow up tests are required.

Patient number: 04

This patient was a 60 years old male who had diagnosed for tuberculosis on 18th of April, 2012. The AFB microscopy result of the first specimen was scanty (S-4) for first specimen and "1+" for both the second and third specimens. For the follow up study, the AFB microscopic examination had been taken again on 16th of May, 2012 to observe whether he was improving or not. The follow up result showed negative for 1st specimen and scanty for the 2nd and 3rd specimens. The result of both initial and follow up AFB microscopic examination are given below.

Table 3.16: Outcome of the first AFB microscopic examination of patient 04

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
18.04.2012	1.		√			
	2.			√		
	3.			√		

Table 3.17: Outcome of the follow up AFB microscopic examination of patient 04 after almost one month

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
16.05.2012	1.	√				
	2.		√			
	3.		√			

The follow up result showed that he was improving with the an-TB treatment.

Patient number: 05

A male patient with age 50 years had diagnosed for tuberculosis on 24th of April, 2012. The report of first specimen was "1+" and both second and third specimens reported "3+". After almost one month of treatment with anti-TB, the AFB microscopic examination had been performed again on 20th of May, 2012 to observe the level of improvement. The follow up result showed scanty for the first specimen and negative for both second and third specimens. The result of both initial and follow up AFB microscopic examination are given below.

Table 3.18: Outcome of the first AFB microscopic examination of patient 05

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
24.04.2012	1.			√		
	2.					√
	3.					√

Table 3.19: Outcome of the follow up AFB microscopic examination of patient 05 after almost one month

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
20.05.2012	1.		√			
	2.	√				
	3.	√				

.

The follow up result showed that he was improving with anti- TB treatment.

Patient number: 06

This patient was a 50 years old male who had diagnosed for tuberculosis on 18th of April, 2012. The AFB microscopic examination showed "3+" for all specimens. For the follow up study, the AFB microscopy test had been taken after one month of treatment on 19th of May, 2012 to observe whether he was improving or not. The follow up result showed negative for all specimens. The report of both initial and follow up AFB microscopic examination are given below.

Table 3.20: Outcome of the first AFB microscopic examination of patient 06

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
18.04.2012	1.					√
	2.					√
	3.					√

Table 3.21: Outcome of the follow up AFB microscopic examination of patient 06 after one month

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
19.05.2012	1.	√				
	2.	√				
	3.	√				

This patient showed excellent improvement with one month of treatment.

Patient number: 07

A female patient with age of 50 years had diagnosed for tuberculosis on 8th of April, 2012. The report of AFB microscopy was smear positive pulmonary tuberculosis. The first specimen resulted to be "2+" and the second and third specimen showed scanty in the report of AFB microscopic examination. After almost one month of treatment with anti-TB, the AFB microscopy test had been performed again on 5th of May, 2012 to observe the level of improvement. The follow up result showed scanty for first specimen and negative for second and third specimens. The report of both initial and follow up AFB microscopic examination are given below.

Table 3.22: Outcome of the first AFB microscopic examination of patient 07

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
8.04.2012	1.				√	
	2.		√			
	3.		√			

Table 3.23: Outcome of the follow up AFB microscopic examination of patient after 07 almost one month

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
05.05.2012	1.		√			
	2.	√				
	3.	√				

The follow up report showed that this patient is improving gradually with the anti-TB treatment.

Chapter 4

Discussion

&

Conclusion

4.1 Discussion:

Even though, with proper treatment patients should show smear negative result within 2 weeks (NHS Oxfordshire, 2002) but this preliminary study shows that among the seven patients two responded completely towards the treatment but the rest of the patient were improving in a slower manner. It shows that those five patients (71.43 %) that is majority who performed the second diagnosis were not responding properly towards the anti- TB course. It can be said that, as the patients were 50 years old or above, therefore, they become weak naturally and their immune system could not fight with the *Mycobacterium tuberculosis* properly and the response towards treatment was slow as well.

An additional finding of this preliminary study is that, the second follow up diagnosis test could be performed with 7 patients (35%) and the rest of the 13 patients (65%) did not agree to come for the 2nd follow up in the hospital after being discharged. Hence, this phenomenon indicates that awareness about the disease was also negligible among the TB patients even after getting the anti-TB treatment therapy.

Another significant result represents that among the 20 patients 3 (15 %) had other family members who infected and developed tuberculosis prior and 17 patients (85%) showed no family history of tuberculosis. As a result, there has been a doubt whether TB transmits genetically or not. The natural history of the disease after primary infection has been subject to debate (Lillebaek T., 2001).

One of the most vital findings of the study includes, 85% patients had shown to have smoking habit. This data represented that there is an association between tobacco use and tuberculosis disease. In 2004, WHO Tobacco Free Initiative (TFI), the WHO Stop TB! Department (STB) and the International Union against Tuberculosis and Lung Diseases (The Union) responded to the figure out the evidence of association between tobacco use and tuberculosis disease. All relevant studies up to July 2005 were considered and 50 studies were reviewed for the strength of evidence. The conclusion of this review stated as “This review indicates that passive or active exposure to tobacco smoke is significantly associated with tuberculous infection and tuberculosis disease. Active smoking is significantly associated with recurrent tuberculosis and tuberculosis mortality. These effects appear to be

independent of the effects of alcohol use, socioeconomic status and a large number of other potential confounders" (WHO, 2007).

An added outcome of the study shows that among 20 patients 17 (85%) were male. This data represents that male are more vulnerable to TB disease than female. According to a study, TB prevalence in male is higher in Sahariya tribe (Sharma P. R., *et al* 2010).

The limitation of the study was that I could not make the follow up result of all patients I have enrolled. The reason behind this drawback of the study was funding. As most of the patients lived outside the Dhaka, therefore, they were not comfortable to come for follow up check up as it was quite non-feasible. If, the fund behind this research program could be enough to bear their transportation cost and accommodation cost then more follow up result could be gathered to come to a more precise conclusion. In fact, the best way is to monitor the patient throughout the study period.

4.2 Conclusion:

From this study it can be concluded that, smear positive tuberculosis can be get well very quickly if they take the treatment properly. In managing patients with tuberculosis, the importance of follow-up visits should therefore be emphasized because a reliable and readily available tool can be used to predict response to treatment. It should be remembered that he or she have to complete the full course of treatment. During the treatment procedure, the patient should be concerned not to miss even a single dose. In case of advance age, the dormant phase of infection turns to active form so that proper treatment has to be taken and maintained though out the complete course.

Annexure

1.Case Report Form (CRF) for patients

Topic: AFB microscopy examination of tuberculosis patients (50 years old or above) admitted in National Institute of Diseases of the Chest and Hospital (NIDCH).

Prepared by-

Tasnuva Tamanna

ID# 2008-3-70-030

Patients' Identification:

1. Patient's name: ……
2. Age: …….
3. Registration number: ……..…

Socio-demographic history:

1. Gender:
    a. Male b. Female
2. Education level:
    - Illiterate
    - Can read only
    - Can read and wrote
    - SSC
    - HSC
    - Graduate or higher
    - others
3. Body weight: …….
4. Place of residence:
    a. Rural
    b. Urban
    c. Semi-Urban
    d. Slum
5. Living status:

a. Rich
b. Upper middle
c. Lower middle
d. Poor

Patients' awareness about the disease:

1. Do you know it is contagious?
   a. Yes b. No
2. Is there anyone in your family ever infected with tuberculosis?
   a. Yes b. No
3. Did you take vaccine (BCG)?
   a. Yes b. No c. Do not know

Patients' occupation

1. What is your occupation? …….
2. Do you have any working disabilities?
   a. Yes b. No
   If yes, then what?.....

Patients' smoking history:

1. Did you have smoking habit before diagnosing for tuberculosis identification?
   a. Yes b. No
2. Did you quit smoking after being confirmed to have TB?
   a. Yes b. No

Risk factors:

- Malnutrition
- Unhygienic
- Smoking habit
- Advanced age

Diagnosis:

AFB microscopy:

Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
	1.					
	2.					
	3					
Follow up						
Date	Specimen	Result				
		Negative	Scanty	1+	2+	3+
	1.					
	2.					
	3.					

Hematological Report

Hb: _____

Total count of WBC: _____

ESR: _____

Platelet count (PC): _____

PCV: _____

Differential count of WBC:

Neutrophils:

Lymphocytes:

Monocytes:

Eosinophils:

Basophils:

Immature Cells: _____

2.

TB 05

NATIONAL TUBERCULOSIS CONTROL PROGRAMME (NTP)

Directorate General of Health Services, Bangladesh

Request Form for AFB Microscopy Examination

(The completed form with results should be sent promptly by the Laboratory to the referring facility)

Name of Referring Facility[1]/Providers: _____ Date: _____

Name of Patient: _____ Age: _____ Sex: ☐ M ☐ F

Occupation _____ Name of Father / Husband _____

Full Address of Patient: _____

_____ Telephone no. (if any): _____

OPD Reg. No. (if any); (For suspects only): _____

Reason for examination: ☐ Diagnosis ☐ Follow-up If follow-up, No. of month of Treatment: _____

Disease Classification: ☐ Pulmonary ☐ Extra-pulmonary (EP) If EP, Site: _____

Nature of Specimen: ☐ Sputum ☐ Urine ☐ Pus ☐ Other, specify: _____

Specimen identification no: _____ Patient TB Registration No: _____
(For follow-up patients)

Signature of person requesting examination: _____

Name & designation of person requesting examination: ----------

1. Including all public and private health facility/providers

---

RESULTS (To be completed in the Laboratory)

Lab Registration No: _____

Visual appearance of the specimen (if it is sputum): ☐ Muco-purulent ☐ Blood-stained ☐ Saliva

Microscopy results

Date of Collection*	Specimen	Result				
		Negative	Scanty (1-9)	1+	2+	3+
	1					
	2					
	3					

Sputum collected by:

Signature: _____

Name: _____

Examined by:

Signature of Medical Tech (Lab) _____

Name: _____

Date: _____

Name of Lab/Organization: _____

* To be completed by the person collecting the sputum

ÁvZwjwLZ m¤§wZcÎ

M‡elK t Zvmbyfv Zvgvbœv

M‡elYvi D‡Ïk¨t

TB GKwU wbivgq A‡hvM¨ dzmdz‡mi †ivM hv cÖKvk cvq `xN©‡gqv`x ‡k-®§vhy³ Kvwk Ges k¦vmKó e„w×i gva¨‡g| GwU Dbœqbkxj †`kmg~‡n cÖvßeq¯‹‡`i Amy¯'Zv Ges g„Zz¨i Ab¨Zg cÖavbKviY| mvaviYZ: a~gcvb, cwi‡ekRwbZ Zvgv‡Ki †auvqv, Avf¨šixb evqy`~lY Ges †ckvMZ Kvi‡Y †auvqv ev ‡auvqvi KYv/KwYKvi ms¯ú‡k© Avmv n‡"QTBnIqvi KviY|

AskMÖnYKvixi wbKU †_‡K cÖZ¨vkv t

Avcwb hw` GB M‡elYvq AskMÖn‡Yi Rb¨ m¤§wZ †`b, Zvn‡j Avgiv Avcbv‡K wKQ ycÖk œewR‡Ám Ki‡ev|

†MvcbxqZv, bvgnxbZv Ges wbðqZv t

Avgiv Avcbv‡K Avk¦¯— KiwQ †h, Avcbvi wbKU †_‡K msM„nxZ Z_¨ Ges Avcbvi ¯^v¯'¨MZ Ae¯'v m¤ú‡K© KvD‡K Rvb‡Z †`qv n‡ebv| GB M‡elYvq †Kvb KvMRc‡Î Avcbvi bvg _vK‡e bv| Avcbvi bvg A_ev e¨w³MZ Z_¨(hv Øviv Avcbv‡K Lyu‡R †ei Kiv hv‡e)†Kv_vI cÖKvk Kiv n‡e bv ev KvD‡K †`qv n‡e bv|

fwel¨‡Z e¨envi †hvM¨ Z_¨ t

GB M‡elYvq msM„nxZ Z_¨ fwel¨‡Z M‡elYvi Kv‡R e¨envi Kiv n‡Z cv‡i|

AskMÖnY bv Kivi, cÖZ¨vnvi Kivi AwaKvi t

GB M‡elYvq Avcbvi AskMÖnY †¯^"Qvq| Zvi gv‡b n‡"Q, Avcwb GB M‡elYvq Avcbvi AskMÖnY eR©b ev cwinvi Ki‡Z cv‡ib| †h ‡Kvb mgq Avcwb PvB‡j Avcbvi AskMÖnY cÖZ¨vnvi Ki‡Z cv‡ib, hw`I Avcwb c~‡e© e‡jwQ‡jb †hGBM‡elYvq AskMÖnY Ki‡eb| †h ‡Kvb ai‡bi A¯^w¯—Ki cÖ‡kœi DËi Avcwb Gwo‡q †h‡Z cv‡ib|

Avcbvi wK †Kvb cÖkœ Av‡Q? n¨uv/bv

Avcwb wK GB M‡elYvq AskMÖn‡Yi Rb¨ ivRx Av‡Qb? n¨uv/bv

GLb Avgiv Avcbv‡K GB M‡elYvq AskMÖn‡Yi Rb¨ Avgš¿Y Rvbvw"Q| Avcwb hw` ivRx _v‡Kb,Zvn‡j AbyMÖnc~e©K AskMÖn‡Yi m¤§wZi wb‡`©kbv ¯^i*f*c bx‡P wba©vwiZ ¯'v‡b Avcbvi bvg A_ev ¯^v¶i w`b|

_____ _____

AskMÖnYKvixi ¯^v¶i A_ev bvg ZvwiL

_____ _____

M‡el‡Ki¯^v¶i ZvwiL

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