Tuberculosis is an ancient disease & it remains the leading cause of death of human being. It is estimated that yearly about 9 million people in the world are attacked with TB, with 1.7 million deaths (Singh, 2008). It is mainly caused by *Mycobacterium tuberculosis* which destroys the lungs & is highly infectious. It is caused by inhaling cough droplets & dust particles containing tubercle bacilli which produce small inflammatory lesions in the lung- known as tubercle and so disease is known as tuberculosis. After droplet infection by tubercle bacilli from the lung they enter into the blood circulation and via blood the macrophages are activated, which form a granuloma around the site of primary infection. In most people the primary lesions is self healing but some bacilli still remain alive inside the macrophages, which may reactivate disease, after latent period which may be few months or the few year. The disease can also spread internally via blood circulation, infecting glands, bones, brain (tubercular meningitis), skin & internal organs; in this form it is called miliary tuberculosis. The disease also produces cavities when affects bone commonly known as bone TB.

Mainly in human four very closely related species are responsible for tuberculosis, *Mycobacterium tuberculosis*, (human tubercle bacilli), *Mycobacterium bovis* (bovine tubercle bacilli), *Mycobacterium microti* (vole tubercle bacilli), & *Mycobacterium africanum* (intermediate form). In HIV patient, atypical mycobacteria also cause tuberculosis like disease i.e. (*M. avium, M. intracellur, M.kanssii*).

**MORPHOLOGY:**

With Ziehl Neelsen Stain, *Mycobacterium tuberculosis* look slender, straight or slightly curved rod with beaded or barred appearance. They are acid fast due to presence of mycolic acid in cell wall. They are weakly or variable Gram staining. They are non-motile, non-sporing and non-capsulated bacilli.
CLASSIFICATION

Mycobacteria causing human disease may be classified as follows:

Mycobacteria

1. Cultivable mycobacteria
   (a) Typical tubercle bacilli.
   - Human type *M. tuberculosis*.
   - Bovine type *M. bovis*.
   - Vole type *M. microti*.
   - Human type *M. africanum*.

(b) Atypical mycobacteria (Runyon’s classification)

   Group- 1 Photochromogen
   - *M. kansasii*
   - *M. marinum*
   - *M. simiae*
   - *M. asiaticum*

   Group- 2 Scotochromogen
   - *M. scrofulaceum*
   - *M. szulgai*
   - *M. xenopi*
   - *M. gordonae*
   - *M. celatum*

   Group- 3 Non Photochromogen
   - *M. avium-intracelluar complex*
   - *M. paratuberculosis*
   - *M. terrae-triviale*
   - *M. shimoidae*

   Group- 4 Rapid growers
   - *M. fortuitum/chelonea complex*
   - *M. thermoresistible*
(c) Saprophytic mycobacteria.

- *M. smegmatis*
- *M. phlei*
- *M. stercoris*
- *M. thermo*

2. Non-cultivable mycobacteria

- *M. leprae* (cause leprosy in human)

**PATHOGENESIS:**

**ENCOUNTER AND ENTRY:**

The infection is commonly acquired by inhalation of aerosols or dust particles containing tubercle bacilli and sometimes by way of ingestion and inoculation into skin (lupus vulgaris)[Fig. 1]. The inoculation size of tubercle bacilli required to cause infections is usually high.

![Dissemination of Tuberculosis](image)

**Figure: 1.1 Entry and encounter of TB bacilli**
SPREAD, MULTIPLICATION, AND DAMAGE:

Damage is caused by uncontrolled, progressive, chronic inflammation, and by organism living within macrophages.

Figure 1.2 Spread, multiplication and damage by TB Bacilli

Types of Tuberculosis

Tuberculosis is divided into three clinically important categories:

I. Primary TB
II. Secondary Reactivated TB
III. Disseminated TB

I. **Primary Tuberculosis**

Primary tuberculosis refers to the infection process which eventually eliminates the pathogen or results in a stalemate between the *Mycobacteria* and the immune system. With most TB infections, the immune system is able to contain, although not eliminate, the *Mycobacteria* within the tubercle, preventing the spread of bacteria and progression of the disease. *M. tuberculosis* can remain in this impasse of dormant infection for many years.
II. Secondary or Reactivated Tuberculosis

The infection can become reactivated if the *Mycobacteria* are able to rupture the tubercle and spread through the lungs. This reactivation typically happens to those with a weakened or suppressed immune system, such as those with HIV infection, may fail to contain the primary infection, and organisms may invade the blood stream.

III. Disseminated Tuberculosis

The spread of the disease within the body may result if infected macrophages moving through the blood and lymph transport the bacteria to other sites. Blood borne organisms can then localize and cause disease in almost any individual organ of the body or they can disseminate and cause a potentially fatal infection known as miliary tuberculosis. Miliary TB may infect any number of organs, including the lungs, liver, and spleen. It is a complication of 1–3% of all TB. Once infected, symptoms of disseminated TB correspond to the locations infected. The antiquated term "consumption" arose from the myriad of symptoms associated with disseminated tuberculosis, when those infected seemed to slowly waste away. (http://tami-port.suite101.com)
**TUBERCULOSIS DISEASES**

**(A) Pulmonary tuberculosis**
- Fibrocaseous TB
- Cavity TB
- Tuberculous bronchopneumonia
- Caseous pneumonia
- Hematogenous or miliary TB in lungs
- Apical lesions or perifocal lesion
- Generalized spread of tuberculosis
- Healing by fibrosis-calcification
- Pleural effusion and Empyema

**(B) Extra Pulmonary Tuberculosis**

Most of the important organs that are affected include bones, kidney, brain, lymph node, female pelvic organs (ovaries, tubes, endometrium), testis and intestine.

**HIV –TB CO-INFECTION**

In developing country tuberculosis is the most common life threatening opportunistic infection in patient with HIV. About 25 to 65 % patients with HIV/AIDS have tuberculosis of any organ (Narain et al. 2002; Gothi et al. 2004; Sharma et al. 2004). Of the 9.4 million people who became ill with TB in 2009, an estimated 1.0-1.2 million were HIV positive (11-13%) (Harries et al. 2004; Corbett et al. 2003).

HIV reactivates latent TB by weakening the natural defenses of infected persons. Of the 5.1 million HIV infected people in India, about half of them are co infected with *M.tuberculosis*, approximately 2,00,000 of these co-infected person will develop active TB each year in association with HIV infection. (Khatri et al. 2002)

Thus tuberculosis is a leading cause of morbidity and mortality in patient with HIV/AIDS. (Human immunodeficiency virus) (Harries et al.2004; Raviglione et al. 1992). The course of disease is determined by some factors like malnutrition, drug abuse, poverty, unemployment, homelessness.
EPIDEMIOLOGY

The recent survey of WHO Says 98% of cases in developing countries with an increase of ~3% annually, 10% in African countries. 80% of cases seen in 27 countries; about half in 5 countries: India, China, Indonesia, Nigeria and Bangladesh. The average prevalence of all forms of tuberculosis in India is estimated to be 5.05 per thousands, prevalence of smear positive cases 2.7 per thousand and average annual incidence of smear positive cases at 84 per 1, 00,000 annually (Chakraborty 2004). In India every day more than 5,000 develop TB disease and more than 1,000 people die, because of tuberculosis. (i.e. 1 death in every one and half minutes) (WHO. 2008). In Gujarat state all districts have examined more than 140 TB suspects/lakh pop in 1st quarter 2009. (Gujarat State report. 2010)

WHO estimates that in 2008, 440000 MDR TB cases emerged and 150000 deaths were caused by MDR TB (WHO. 2010). The proportion of TB cases that have MDR TB is highest in eastern Europe and central Asia, while around half of the world’s cases of MDR TB occur in China and India (WHO. 2011). As of August 2010, 59 countries had reported at least one case of XDR TB. The main goal of DOTS components of the Global plan is to reduce the global burden of TB mortality by 50 % by 2015, compared with 1990 (WHO. 2011-15).
Figure: 1.4 Global epidemiology of Tuberculosis

Figure: 1.5 Proportion of MDR TB among new TB cases, 2009

Source: WHO Geneva; WHO Report 2004: Global Tuberculosis Control, Surveillance, Planning and Financing
**For programming monitoring purpose estimated cases in East & South zones have been kept at the national level of 75

**Figure: 1.6 Epidemiology of Tuberculosis in India**

**Figure: 1.7 Epidemiology of Tuberculosis in Gujarat and Surat**
**Laboratory diagnosis:**

It is divided mainly in three ways

(1) Direct evidence
(2) Indirect evidence
(3) Newer technique

(1) **Direct evidence:**

Mycobacterial Species either by directly observes in staining method or by culture method.

(1) Microscopy: It is done by various ways.
   - ZNCF technique
   - Floreance technique
   - Cold staining technique
   - Silver Impregnation method

(2) Culture technique:

Conventional mycobacterial cultures are done on Lowenstein Jensen medium or the Middle brook medium. It takes 4-8 week for the growth of mycobacteria & another 4-8 weeks for the drug sensitivity.

**Newer Rapid Culture Techniques**

(a) Bactec
(b) Mycobacteria Growth Indicator Tube (MGIT) System
(c) Luciferase Reporter Mycobacteriophage test (LRM TEST)
(d) Molecular amplification Technique For Diagnosis-PCR

(2) **Indirect evidence:**

- Skin test (tuberculin test)
- Immunological Methods
  (1) Antibody based tests

(a) RIA (Radio Immuno assays)
(b) SAFA (Modified Soluble Ag Fluorescent Ab)

(c) ELISA (Enzyme Linked Immuno-Sorbant Assays)

(2) Antigen based Tests

★ ELISA
★ RIA
★ Haemagglutination test Latex Agglutination test
★ Circulating Immune Antibodies complexes
★ Monoclonal Antibodies by SACT.

▼ ADA Enzyme
▼ Gas Liquid chromatography
▼ Histopathology
▼ High Performance liquid Chromatography

(3) Newer Technique

▼ PCR (Line Probe Assay)
▼ Fast Plaque Technique
▼ Colorimetric methods: MTT and REMA methods
▼ The Nitrate Reductase Assay
▼ The Microscopic Observation Broth – Drug susceptibility Assay (MODS)
▼ The Thin - layer agar method (Palomino et al.,2007)

PREVANTION, CONTROL AND TREATMENT

There are various ways to prevent tuberculosis. A method for immunization against tuberculosis with living attenuated bovine tubercle bacilli i.e. BCG Vaccine has been known. The use of BCG stimulates partial immunity against tuberculosis but the effectiveness of BCG in preventing tuberculosis in adult is limited.

Tuberculosis can be treated effectively by a combination of anti-tubercular drugs. Patient with tuberculosis, who fail to complete ‘Standard’ course of anti tubercular therapy are at increased risk for treatment failure and they may play a role in both the emergence of drug resistant strains of *Mycobacterium tuberculosis* and further spread of tuberculosis in the society.
The choice of combinations of drugs & regimens have been based on the properties of different drugs particularly on their bactericidal activity, intracellular penetrability, resistance pattern of Mycobacteria in vivo activities, Pharmacological behavior & toxicity.

Different anti-tubercular drugs posses the different properties Rifampicin (RIF) & Isoniazid (INH) are most powerful bactericidal drugs active against all populations of tubercle bacilli. Pyrazinamide is active against tubercle bacilli inside macrophages, where as Streptomycin is active against rapidly multiplying extracellular tubercle bacilli. Ethionamide & Thioacetazone are bacteriostatic drugs used in association with more powerful bactericidal drugs to prevent the emergency of drug resistant bacilli. First line drugs for initial treatment are Isoniazid, Rifampicin, Streptomycin, Pyrazinamide and Ethambutol which are routinely used once active infection is suspected (Gandhi et al. 2006).

Drugs of second choice are PAS (Para amino Salicylic acid), Ethionamide, Kanamycin Cycloserine, Capreomycin and Ofloxacin which usually reserved for use as backup, because of their serious side effects and/or lack of efficiency when resistance to first line drugs is found (Gandhi et al.2006).

Majority of these drugs are bacteriostatic than bactericidal and therefore when treatment is stopped the organisms become resistant.

To resolve the problem of TB the National Tuberculosis Programme (NTP) in India was implemented in 1962 by establishing District TB Centers (DTCs), TB Clinics and TB hospitals. To improve & strengthen tuberculosis control activities, the Government of India launched the Revise National Tuberculosis Control Programme (RNTCP) in 1997 and covers almost the whole country with excellent results by the end of 2005. Directly Observed Treatment, Short Course Chemotherapy (DOTS) means that the patient swallows short course anti tubercular drugs in the presence of health worker or other trained individual (Module for Laboratory Technicians, Central TB division. 2005)

There are two phases in the treatment of tuberculosis,

- The intensive phase which is of 3 months,
- The continuation phases for 4 and 5 months.
According to the WHO, under this program second line anti tubercular drugs are used to control the prevalence of MDR-TB & thus DOTS-PLUS should be implemented in selected areas with moderate to high levels of MDR-TB.

**WHAT IS MDR-TB & XDR TB ?**

Drug resistant TB is widespread and found in all countries surveyed. It emerges as a result of treatment mismanagement and is passed from person to person in the same way as drug sensitive TB.

Multi Drug Resistant Tuberculosis (MDR-TB) is defined as disease caused by *M.tuberculosis* resistant to at least Rifampicin and Isoniazid, the two most important first line anti-tubercular drugs. (Skenders et al.,2005) Multi-drug resistance (MDR) has become a major concern to control TB particularly in the developing countries (Cohn *et al.*, 1997). The development of mutations in different genes of mycobacterium leads to drug resistance and subsequent MDR-TB (Petrini and Hoffner, 1999).

Management of MDR-TB entails intense chemotherapy for up to 2 years which is very damaging to a patient’s health due to high levels of drug toxicity (WHO Report, 2008). Because of treatment failure for MDR-TB, extensively drug resistant tuberculosis (XDR TB) emerged.

Extensively drug resistant (XDR TB) is a form of TB caused by bacteria resistant to all the most effective drugs (i.e. MDR-TB plus resistance to any fluoroquinolones and any of the second line anti-TB injectable drugs viz. Amikacin/ Kanamycin /Capreomycin).

There is emergence of multi-drug resistant (MDR) and extensively-drug resistant (XDR) strains of *M. tuberculosis* all over the world including India (Singh. 2007). The emergence of resistance to antimicrobials, though is a natural biological occurrence, has become an important public health issue in many developing countries as the treatment of TB requires the use of more expensive drugs for a longer treatment period. There is, therefore, an urgent need for new, inexpensive TB drugs which are more effective and with fewer side effects.
The use of herbs and other alternative therapies for the treatment of tuberculosis is on the increase. Natural products continue to play a most significant role in the drug discovery and development process (Newman and Cragg, 2007), and plants are recognized as a useful source of highly active antimycobacterial metabolites (Gibbons, 2005; Pauli et al., 2005).

Medicinal plants offer a great hope to fulfill these needs and have been used for curing diseases for many centuries. These have been used extensively as pure compounds or as a crude material. Only a few plant species have been thoroughly investigated for their medicinal properties (Heinrich et al. 2001). India is one of the few countries in the world which has unique wealth of medicinal plants and vast traditional knowledge of use of herbal medicine for cure of various diseases (Gupta et al. 2004; Sharma. 1998). So far, few plants have been tested against mycobacteria and a few plants which showed anti-tubercular activity were Salvia hypargeia, Euclea natalensis, etc. (Gautam et al. 2007; Lall et al. 2001; Raut et al. 2006). The increasing incidence of MDR and XDR-TB worldwide highlight the urgent need to search for newer anti-tuberculosis compounds/ drugs. Therefore, the present study was carried out to check the antimycobacterial activity of aqueous extracts of two medicinal plants i.e. Garlic (Allium sativum) and Turmeric (Curcuma longa) against MDR isolates of M. tuberculosis and reference susceptible strain M. tuberculosis H37Rv.