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ثبكة المعلومات الجامعية







#### MEASURMENT OF MYCOBACTERIAL ANTIGEN A 60 SPECFIC IMMUNOGLOBULINS IgG, IgA AND IgM IN THE SERA OF PULMONARY TUBERCULOUS PATIENTS

Thesis Submitted in Partial Fulfillment for Master Degree In Clinical Pathology

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#### List of Abbreviation

WHO: World health organization.

HIV: Human immune deficiency virus.

B.C: Before century.

A.D: After delivery.

IWGMT: International working group of mycobacterial Taxonomy.

M.T.B: Mycobacterial Tuberculosis

MOTT: Mycobacterium officer than tuberculosis

C M I: Cell mediated immunity.

DTH: Delayed type hypersensitivity.

IFNy: Interferone  $-\gamma$ .

NK: Natural killer.

ADCC: Antibody - dependant, cell mediated cytotoxicty.

TNF: Tumor necrosis factor.

CTLS: Cytotoxic T- lymphocytes.

OT: Old tuberculin.

PPD: Purified protein derivative.

ELISA: Enzyme linked immunosorbant assay.

ADA: Adenosine deaminase.

BCG: Bacille calmette - Guerin

A.M.: Alveolar macrophage.

AIDS: Acquired immune deficiency syndrome.

RIf: Rifampin.

INH: Isoniazide.

**D.O.T.:** Directly observed therapy

SM: Streptormycine

EMB: Ethambitol.

PZA: Pyrazinamide.

PAs: Para amiono – salicylic.

NALC: N acetye -L - cysteine.

Z.N: Ziehl - Neelsen.

A F B: Acid fast bacilli.

IUAT: International union against tuberculosis.

LJ: Lowenstein - Jensen.

ATS: American thoracic society.

DNA: Deoxyribonucleic acid.

GIC: Gas liquid chromatography.

HPLC: High performance liquid chromatography.

rRNA: Ribosomal ribonucleic acid.

PCR: Polymerase chine reaction.

SACT: Solid phase antibody competition test.

TMA: Thermo stable macromolecular antigen.

A 60: Antigen 60.

TCH: thiophen -2 – carboxylic acid hydrazid.

LAM: lipoarabino mannan.

IgA: immunoglobulin A.

IgG: immunoglobulin.G.

IgM: immunoglobulin M.

CMN group: Corynebacterium - Mycobacterium and No-cardia.

D. W.: Distilled water

O.D. I.: Optical density index

A. L.: Antibody index.

T. M. B.: Tetramethyl benzidinc.

FC: Fraction crystalline

TH1:T- helper 1.

TH2:T- helper 2.

1L2:Inter leukine 2.

CD: Cluster of differentiation.

T. lymphocytes.: Thymus dependant lymphocytes.

B. lymphocytes: Bone marrow lymphocytes.

ADCC: Antibody dependent cellular cytotoxicity.

C5a: complement 5a.

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# INTRODUCTION AND AIM OF THE WORK

#### INTRODUCTION

Tuberculosis is still a major health problem in most developing countries and its incidence is rising in many industrial countries. The diagnosis of tuberculosis depends primarily on identification of mycobacteria and on clinicoradiological evidence of the disease (Heijjaj et al., 1999). The rising incidence of tuberculosis worldwide means an increasing burden on diagnostic facilities, which should be objective, reproducible, so serological tests compared to other diagnostic methods are faster and do not necessarily require samples that contain the tuberculibacilli (Pereira et al., 2000). Serological tests available for diagnosis of tuberculosis can provide the valuable information about host immune response to the mycobacterial infection (Sieminska et al., 1999).

A renewed attempt to develop of a serodiagnostic test was initiated when antigen A 60 became available which is a major component of mycobacterial cytoplasm preferred from the cytoplasm of mycobacterium bovis (Cocito, et al., 1991). For early diagnosis of tuberculosis, especially in the patients without adequate specimens for examination or sputum examination is negative, the enyme-linked immunosorbent assay (ELISA) is a simple, rapid and inexpensive method among many diagnostic tools (Lilf, et al., 1998).

The Aim of this work is to evaluate the role of A 60 antigen in the serological diagnosis of tuberculosis using the ELISA technique for the detection of A60 specific IgA, IgG and IgM antibodies in the sera of patients with pulmonary tuberculosis.

## REVIEW OF LITERATURES

#### Mycobacterium Tuberculosis

#### Historical background:

Tuberculosis is a disease of great importance, as it remains in the words of World Health Organization (W.H.O.) reports, "the most important specific communicable disease in the world". (Davidson, 1985).

According to estimates of W.H.O. in 1990 there was approximately 8 million active cases, of which 7.6 million new cases were in developing countries and 400000 new cases were in industrialized countries (Kochi, 1991). In 1990, death due to tuberculosis occurred in an estimated 2.9 million people world wide and 40.000 occurred in developing countries (Murray, 1994).

Thus, tuberculosis is still a major cause of disease and death and its elimination will be extremely difficult as long as poverty, over population, and human immunodefficency virus (HIV) infection characterize large portions of the earth. It is already deemed the number one preventable cause of death in developing countries (Murray, 1990).

The tubercle bacillus was discovered in 1882 when Robert Koch, succeeded in isolating the tubercle bacillus from pathological material.

Tuberculosis appears to be as old as humanity itself. Skeletal remains of prehistoric humans dating back to 8000 B.C. found in Germany, show clear

evidence of the disease. Egyptian skeletal dating from 2500 to 1000 B.C. have revealed evidence of Pott's disease of the spine.

Perhaps the best proof of tuberculosis has come from an Inca mummy of an 8 years old boy who lived about 700 A.D. The radiographic picture of the lumbar spine showed evidence of pott's disease, and the smears of the lesion revealed acid fast bacilli, most likely Mycobacterium bovis (Marrio, et. al.1998).

Tubercle bacilli can remain in viable form for many years in the tissues of healthy persons. When they produce disease, it runs a chronic and protracted course that gives time for transmission to susceptible hosts.

The infection can produce disease in a human being after decades of dormancy. Thus, the infection becomes endemic when a large proportion of the population is infected. It can produce an epidemic when introduced into a population of which only a small portion is immunologically protected by already having been infected (David. 1999).

#### The Mycobacterium

#### **Definition:**

Mycobacterium is the only genus in the family Mycobacteriaceae, straight or slightly curved rods, but coccobacillary, filamentous and branched forms also occur. Cells are gram positive—acid fast, non motile and non-sporing. Some