

CHAPTER-I

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**INTRODUCTION**

## **“Molecular Biochemical Studies On Human Serum Afflicted With Lung Related Health Problems”**

The World Health Organisation (WHO) defines health as a state of complete physical, mental and social well being and not merely the absence of disease and infinity. From a bio-chemical point of view health may be considered as a situation in which all of the many thousands of intra and extra-cellular reactions occur in the body without any deleterious or adverse effect. All diseases are manifestations of abnormalities of molecules Chemical reactions or processes. The causative factors are several, namely, physical factors, chemical agents, biological agents, lack of oxygen, genetic disorders, nutritional imbalances and endocrine imbalances. Enzymes play control roles in health and disease. A significantly large number of people suffer from lung problems such as Asthma, Tuberculosis, Bronchitis, Emphysema etc. In these disease states, severe lung tissue injury or uncontrolled cell growth enzymes are released in to the blood measurement of these intracellular enzymes in blood serum therefore, provides physicians with invaluable diagnostic and piagnostic information. Defects in enzyme function frequently cause disease. <sup>(1)</sup>

Among the biological agents which cause pathological states in human subjects, the bacteria, viral and mycoplasma are notorious, as they are air-borne. The prime entry point for these disease causing organism is through the lungs where they cause a number of lung-related diseases debilitate the entire system it was thought worth while to study the lung related health problems in the context of clinical biochemistry and medical biochemistry.

The various types of lung diseases, which will be tentative subject of present study, are: -

- 1) Asthma.
- 2) Tuberculosis.
- 3) Emphysema.
- 4) Bronchiectesis.

Normal life process requires that an adequate supply of oxygen be delivered to the tissues and that the waste products of cell metabolism be removed. These functions are carried out by a cooperative effort of the respiratory and circulatory systems. The respiratory system oxygenates the blood and removes carbon dioxide. The circulatory system transports these gases in the blood stream. (2)

In what follows now in a systematic description of lung biogenesis, etiology of lung related diseases and possible causes with a physiological and clinical treatise.

**LUNG:** - The lung appears first as an epithelial bud at the caudal end of the laryngotracheal groove on the 26<sup>th</sup> day after ovulation, and is derived from endoderm to form the epithelium of the airways and of the acini. As it elongates, it becomes invested in mesenchyme derived from mesoderm. The mesenchyme itself develops into the connective tissue, cartilage, smooth muscle and vessels of the lung. The developing lung bud after dividing into two halves elongate, growing caudally on either side of the oesophagus. By about 33 days the trachea gets separated from the foregut and tracheal pouches representing the five lobes are clearly seen. Full adult complement of segments developed by 41 days and the completion of the bronchial tree as far as the terminal bronchioles occur by 16 weeks. From 16 to 26 weeks the lung's appearance can be described as canalicular. Lung reaches a stage of maturity at about 26 weeks, when it is capable of supporting life. At the time of birth there is an adult, complement of about 24 million terminal bronchioles. Some 127 million alveoli are present at about one year and in most cases, reaching this number by the age of two. By the age of 8 years the number may grow upto 280 million.

On an estimate, the numbers in adult may vary between 200 and 600 million. The weight of the normal adult lung averages 350 to 425 gms. Fully developed lungs are two, spongy organs surrounded by a thin, moist membrane called the pleura. Each lung is composed of smooth, shiny lobes, there being 'three' lobes on right one, while the left one has 'two'. Approximately 90% of the lung is filled with air and only 10% is solid tissue,

the air is carried from the trachea in to the lung through flexible airways called bronchi.

The bronchioles lead to the grape-like clusters of microscopic sacs called alveoli. There are over 300 million of these tiny sacs, which are composed of a thin membrane through which oxygen and carbon dioxide pass to and from capillaries. Capillaries carry blood through out the body. Normal person breathes per day nearly 25,000 times and inhales 10,000 liters of air. Red blood cells contain factors that fight pollutants and white blood cells are the critical fighters in the body. The lungs are the only organs to receive all the blood from the heart with each heartbeat. The functional unit of the lung is called acinus. It is formed by the cluster of respiratory bronchioles, alveolar ducts and alveoli derived from a single terminal bronchiole. The alveolar walls are perforated by numerous pores of kohn, which allows the passage of bacteria and exudates between adjacent alveoli. Adjacent to the alveolar cell membrane is the glycoprotein containing cell coat upon which is a thin film of phospholipids, phosphatidyl choline (lecithin), the pulmonary surfactant. Lecithin is critical to the alveolar wall as it serves to lower the surface tension of the alveolar lining and maintain the stability of the alveoli. Surfactant is synthesized in type II epithelial cells and stored in the osmophilic lamellated bodies of such cells. Inadequate surfactant activity is believed to play a role in the respiratory distress syndromes in infants and adults. The structure of bronchioles differ in several respects from that of the alveolus on one hand and larger bronchi on the other. The bronchiolar surface is covered with cilia, which are surrounded by a water protein layer rich in 'lysozyme' and 'immunoglobulins'. Unlike the alveoli, the surface layer contains no surfactant and no mucus cells. In the walls of the bronchioles, nonciliated granulated clara cells secrete the mucus, a poor living protein. In some inflammatory conditions of bronchioles, there is goblet cell metepiasia of the bronchiolar lining with an increase in mucus production and a diminution in the number of clara cells.

Each day the respiratory airways and alveoli are exposed to over 10,000 liters of air containing hazardous dusts, chemicals and microorganisms.

Inhaled particles are deposited largely according to their size. Thus particles over 10 $\mu$ m are deposited largely in the turbulent air flow of the nose and upper airways. Particles of 3 to 10 $\mu$ m lodge in the trachea and bronchi by impactions, and smaller particles, about the size of most bacteria 1.0 to 5.0 $\mu$ m are deposited in the terminal airways and alveoli. In effective clearance of particles from these three sites is believed to be crucial to the pathogenesis of pulmonary infections and of the slowly developing pneumoconiosis. <sup>(3)</sup>

**LUNG DISEASES:** - Pulmonary diseases are divided into two categories-

- 1) **OBSTRUCTIVE DISEASE** (or) airway disease characterized by an increase in resistance to air flow owing to partial or complete obstruction at any level, from the trachea and larger bronchi to the terminal and respiratory bronchioles and,
- 2) **RESTRICTIVE DISEASE** characterized by reduced expansion of lung parenchyma, with a decrease total lung capacity. The major obstructive disorders are 'Emphysema', 'Chronic Bronchitis' 'Bronchiectasis' and 'Asthma'. In patients with this 'pulmonary functional tests' show increased pulmonary resistance and limitation of maximal expiratory air flow rates during forced expiration. Expiratory obstruction may results either from anatomic airway narrowing, such as it is classically observed in Asthma or from loss of elastic recoil of the lung, which characteristically occurs in emphysema.

**ASTHMA:** - It is a Greek word meaning breathless (or) to breath with open mouth. Asthma is a disease characterize by increased responsiveness of the tracheobronchial tree to various stimuli, potentiating paroxymal construction bronchial airways. Asthma is a common disorder occurring in upto 5% of the adult population and perhaps upto 10% of children. It is increasing in prevalence and severity and has a rising mortality over 4000 per annum.

Asthma is widely distributed but very variable in its prevalence. Asthma is more common in the countries with markedly under developed ways of life. The incidence of diagnosed asthma as recorded in the USA is highest in

the 1<sup>st</sup> year of life. In childhood the incidence is higher in boys than in girls but reverse in the age group 15- 50yrs and reverse again in the older age group when the incidence among men increases once more. The prevalence of bronchial responsiveness has been shown to follow a similar pattern, with boys having greater responsiveness than girls, adult women having higher levels than men and older men having similar levels of hyper responsiveness as older women. There is some broad correlation between the prevalence asthma allergen such as pollen or house-dust mite, reaginmediated, atopic and intrinsic to which was added a third, mixed pattern in which both intrinsic and extrinsic factors are operative.

i) **Atopic or allergic asthma**: - This is the most common type of asthma. It is triggered by environmental antigens such as dust, pollens, animal dander and foods, but potentially any antigen is implicated. A positive family history of atopy is common and asthmatic attacks are often preceded by allergic rhinitis, urticaria and eczema. Serum IgE levels are usually elevated. A skin test with the offending antigen results in an immediate wheal and flare reaction, a classic example of type-I IgE-mediated hypersensitivity reaction. IgE-mediated hypersensitivity elicits an acute immediate response and a late phase reaction. In case of air born antigens, the reaction occurs first on sensitized mast cells on the mucosal surface, the resultant mediator release opens the mucosal intercellular tight junctions and enhances penetration of antigen to the more numerous sub mucosal mast cells. Mast cells play a key role in the effector limb of asthma. These highly granular tissue cells have long been implicated in the pathogenesis of allergic conditions, particularly asthma. Direct stimulation of sub epithelial vagal receptors provokes reflex bronchoconstriction, which occurs within minutes after stimulation and is called the acute or immediate response. The mediators of IgE-triggered reactions include both primary and secondary mediators. The primary mediators include (i) histamine which causes bronchoconstriction by direct and cholinergic reflex actions, increased venular permeability and increased bronchial secretions and (ii) eosinophilic (ECF) and neutrophilic chemotactic factors (NCF), which selectively attract eosinophils and

neutrophils. The secondary mediators include (i) Leukotriene C<sub>4</sub>, D<sub>4</sub>, and E<sub>4</sub>, extremely potent mediators that cause prolonged bronchoconstriction as well as increased vascular permeability and increased mucus secretion. (ii) Prostaglandins D<sub>2</sub> (PGD<sub>2</sub>), elicits bronchoconstriction and vasodilation and (iii) Platelet activating factor (PAF) causes aggregation of platelets and release of histamine and serotonin from their granules. The acute reaction is associated with bronchoconstriction edema, mucus secretion, flushing and in certain instances hypotension. It is followed by late phase reaction occurring several hours later and lasting for two days. The late phase reaction is caused by the recruitment of a battery of leukocytes, basophils neutrophils and eosinophils by the chemotactic factors derived from mast cells. These leukocytes release a second wave of mediators that stimulate the late reaction. Histamine releasing factor (HRF) produced by various cell types, induce release of histamine from basophils, causing bronchoconstriction and edema. In addition neutrophils cause further inflammatory injury and the major basic protein of eosinophils causes epithelial damage. The presence of both immediate and delayed reactions in IgE mediated events, help to explain the prolonged manifestations of asthma and similar allergies. At the chemical level, the role of acid hydrolases serum to be very important.

(ii) **Food, Drink, Drug-induced asthma**: - It occurs in less than 10% of patients. The foods most frequently suspected are milk, eggs, fish, cereals, nuts and chocolates, meats containing antibiotics fed to animals or tenderized with enzymes are recognize as occasional causes. Entry of antigen in to the blood stream provokes an IgE mediated reaction. Food may also provoke asthma via mechanisms that may not be related to IgE mediated allergy. Preservatives such as benzoates, sodium nitrite and sodium metabisulphate, synthetics antioxidants, dyes such as tetrazine and flavourings may be found in many foods and may provoke asthma. Redwines contain a number of congeners that give them their distinctive flavours also may provoke attacks of asthma. By a direct effect on mast cells causing liberation of mediators. Alcohol itself is a mild bronchodilators. It has been postulated that asthma associated with

chewing betel-nut is chemically mediated by cholinergic stimulation. Changes in dietary salt intake could alter the activity of the bronchial smooth muscle cellular sodium pump, which may be relevant to bronchial reactivity. Dietary polyunsaturated fats, notably  $\Omega$ -6-fatty acids such as linoleic acid may influence the development of allergic sensitization by increasing the formation of PGE<sub>2</sub>, promoting Th<sup>2</sup> lymphocyte responses and IgE generations. Drugs are an important but occasional cause of asthmatic attacks. Aspirin sensitive asthma is a somewhat fascinating type occurring in patients with recurrent rhinitis and nasal polyps. These individuals are exquisitely sensitive to very small doses of aspirin and they experience not only asthmatic attacks but urticaria. Aspirin triggers asthma in these patients by inhibiting the cyclooxygenase pathway of arachidonic acid metabolism without affecting the lipoxygenase route, thus tripping the balance toward elaboration of the bronchoconstrictor leukotrienes.

(iii) **Occupational asthma**: - A most common type of work-related respiratory disease in industrialized countries, is stimulated by fumes, organic and chemical dusts, gases and other chemicals, very minute quantities of chemical are required to induce the attack, which usually occurs after repeated exposure. Asthma related to chemical and vegetable agents occur more commonly in cigarette smokers, it may be due to the effect of smoke on bronchial epithelium, improving access of the sensitizer to sub mucosal lymphocytes and mast cells.

(iv) **Allergic bronchopulmonary aspergillosis**: - Skin sensitivity to *A. fumigatus* is commonly present in atopic individuals and occurs also in a proportion of 'non-atopic' patients with chronic asthma. The frequency with which this occurs varies between 16 to 38 percent. These patients develop both an immediate type-I IgE induced reaction and a 4 to 6 hour type-III IgE-mediated response. Aspergillus induced IgE-mediated mast cells degranulation causes bronchoconstriction and increased vascular permeability. The latter allows anti-Aspergillus antibodies to enter bronchi, combine with antigen, form immune complexes and trigger inflammation and pulmonary damage. Blood

eosinophilia levels rise to  $1-3 \times 10^9/L$ . Attacks are recurrent and tend to occur at first predominantly in the winter months.

Asthma can also be precipitated non-specifically by cold, exercise and emotional stress. Indeed an important feature of patients with asthma of all types immunologic or otherwise, is the hyperreactivity of the airways to non-specific irritants and bronchoconstrictor agents. Patients with asthma even mild asthma, have a variety of inflammatory cells such as mast cells, neutrophils, macrophages in their bronchial walls and mediators released from such cells example leukotrienes may be involved in the state of hyper irritability. Eosinophils cells are attracted by chemotactic factor released by mast cells. They themselves can release mediators, including leukotrienes and platelet activating factor. Eosinophils also produce the basic major protein (MBP) of their granules and MBP has been shown to be toxic to respiratory epithelium and to accumulate in the lungs and sputum of patients with asthma.

**MORPHOLOGY:** - The morphologic changes in asthma patients lungs are over distended because of over inflation and there may be small areas of atelectasis. The most striking macroscopic finding in occlusion of bronchi and bronchioles by thick, tenacious mucous plugs. The mucous glands become enlarged. The submucosa is oedematous, infiltrated with eosinophils and lymphocytes and contains dilated capillaries with swollen endothelial cells. Mast cells may be found in the submucosa, especially in association with small vessels. Eosinophils are a conspicuous feature of the inflammatory exudates mucous plugs may occur throughout airways of all sizes, sometimes reaching the smallest respiratory bronchioles by aspiration.

**CLINICAL FEATURES:** - Asthma may be present at any age including the extremes of life, but the majority of cases are seen before the age of 25 years. The most common symptom is breathlessness usually associated with chest tightness, wheezing cough are often present and nocturnal awakening tends to be a common feature. In an acute attack, the respiratory rate is rapid and tachycardia is common. Sputum is light pale green or white streaks found in the

mucus along with eosinophils, Curchmann's spiral and Charcot-Leyden crystals are also seen in the sputum. There is in the region of a tenfold increase in the number of mast cells spontaneously produced and induced sputum from asthmatic patients. The peak expiratory flow (PEF), maximal mid-expiratory flow (MMEF) and forced expiratory volume in the first second (FEV<sub>1</sub>) are all decreased in asthma. In severe asthmatic attack the PEF and FEV<sub>1</sub> are generally less than 50% of predicted values with a MMEF of 10 to 30% predicted values. Life threatening features include exhaustion, cyanosis, bradycardia, hypotension, confusion and coma.

The aim of management are to recognize asthma, abolish symptoms restore normal or best possible long term airway function reduce the risk of several attacks, enable normal growth to occur in children, minimize absence from school or work. All patients should be encouraged not to smoke and also to avoid passive smoking, as far as possible, an even temp and humidity in all rooms. The control of exercise induced asthma requires therapeutic intervention rather than avoidance of exercise. All B-adrenoreceptor antagonists are absolutely contra indicated in all patients with asthma. Aspirin also induces worsening of asthma and sometimes is accompanied by urticaria.<sup>(4)</sup>

**TUBERCULOSIS:** - Tuberculosis caused by mycobacterium remains to be the largest single cause of death from any infectious disease worldwide. One third of the world's population is infected with the tubercle bacillus. The WHO and United Nations joint program on HIV and AIDS estimated that 35 million adults and 1.4 million children were infected with HIV, world wide by end of 2000. An estimated 15 million adults AIDS deaths and 3.8 million pediatric deaths have occurred world wide since the beginning of the pandemic.

The Indian estimates of HIV infection prevalence are between 2.8 and 4.2 million. The WHO Tuberculosis Program estimates that 95% (8 million) of world's tuberculosis morbidity and 9.8% (2.9 million)-tuberculosis mortality is contributed by developing countries. There are more than 12 million dually infected persons globally and India accounts for more than 1

million of them. More than 1000 people die of TB and over 20,000 people contract the disease every day in India.

According to published research India alone accounts for nearly 1/3 rd of the global TB burden. It also uncovers that in India 3 lakh children are forced to leave school every year because their parents have TB and 1 lakh women lose their families because of the social stigma attached to the disease. <sup>(5)</sup>

**I) PULMONARY TUBERCULOSIS** : - Tuberculosis of the lungs is divided into primary infection and reinfection.

(i) **Primary Pulmonary Tuberculosis**: - The first infection with the tubercle bacillus is known as primary tuberculosis and usually includes involvement of the draining lymph nodes in addition to the initial lesion. All lobar segments are at equal risk of being seeded by inhaled infected droplets and in 25% of cases there may be more than one primary focus. Within days the infection spreads to regional lymph node with enlargement of hilar, mediastinal or subcarinal nodes. A left-sided pulmonary focus may lead to bilateral adenopathy, whereas right-sided lesions only result in right-sided adenopathy. The combination of the primary focus and the draining lymph nodes is known as the primary complex, which consists of (i) a parenchymal subpleural lesion, either first above or just below the interlobes fissure between the upper and lower lobes and (ii) enlarged caseous lymph nodes draining the parenchymal focus.

(ii) **Reinfection stage**: - Most causes of secondary pulmonary tuberculosis represent reactivation of an old possible subclinical infection. There are of two possible subclinical infections. These are two possible ways by which tuberculosis in adults may develop, (a) endogenous reactivation from the focus of primary infection and (b) introduction of fresh bacilli from exogenous. According to Aschoff, the reinfection in young adults is due to an exogenous infection of a massive dose of mycobacterium tuberculosis hominis in persons with primary infection. In

reinfection in the adults, the lesion in lung is usually near the apex of the right lung. People without primary infection in childhood have poor resistance and suffer from the unabated assault of massive dose of a virulent strain of mycotuberculosis. This is specially noticed in Nepalis and other hill tribes, when they come to the plain and suffer from the infection, which is often serious and becomes fatal. They cannot offer any resistance to the infection and quickly succumb to a spreading exudative type of lesion. The right lung more frequently affected than the left lung and the lesion starts initially or around the small bronchiole. The inflammation causes obstruction of the lumen leading to collapse of the alveoli associated with exudation and formation of bronchopneumonic patches with caseation.

**PATHOLOGY:** - Deposition of tubercle bacilli in the alveoli of the lung is followed by vasodilation and an influx of poly morphonuclear leucocytes and macrophages to the area. After several weeks the poly morph numbers diminish and macrophages predominate. The macrophages develop pale foamy cytoplasm rich in lipid and crowd together as epithelioid cells to form the tubercle or unit lesion of tuberculosis. Some mononuclear cells fuse to form the multinucleated or langhan's giant cell. Lymphocytes surround the outer margin of the tubercle and in the centre of the lesion a zone of caseous necrosis may appear that may subsequently calcify. Primary complex probably occurs via the lymphatics in the majority of infected subjects, resulting in the seeding of tubercle bacilli to other parts of the lung as well as other organs. Within an hour of the bacilli reacting the lung they reach the lymph glands and often the blood stream.

Reactivated pulmonary tuberculosis is most often seen in the upper lung zones and is limited in extent to the posterior segment. The high ventilation perfusion ratios, with alveolar  $P_{O_2}$  elevated relative to other zones is believed to predispose to reactivation at these sites. Proliferation of tubercle bacilli in the caseous centers is followed by

softening and liquefaction of the caseous material which may discharge into a bronchus with resultant cavity formation. Whereas  $10^4$  bacilli per gram are found in caseous tissue, up to  $10^9$  organisms may be harboured within a single cavity.

(iii) **Chronic Fibrocaseous Tuberculosis**: - This is the commonest type of lesion that is frequently encountered in tuberculosis. It is found in patients near about the age of 20 or above and the period of gap that remains between the primary and the secondary infections. In this condition the patient is allergic after a primary infection of tuberculosis in childhood, as a result of mycotuberculosis infection. Near about the age of adult period, a reinfection takes place with a massive dose of mycotuberculosis or in rare cases, this may be reactivation of a latent focus (endogenous). This results in breaking down of the tissue due to allergic producing necrosis with the formation of a cavity. This is due to heightened antigen and antibody reaction of the sensitized allergic cells resulting in necrosis. The necrosed tissue is discharged into bronchus and coughed out along with the sputum. The cavity is lined by a yellow-gray caseous material and is more or less walled off by fibrous tissue, thrombosed arteries may traverse these cavities to produce apparent fibrous bridging bands, when such cavitation occurs in the apices, the pathways for further dissemination of the tuberculous infection are prepared. The infective material spreads to the lymph nodes via the lymphatics and other areas of the lung or other organs. Cavitory fibrocaseous tuberculosis may affect one, many or all lobes of both lungs in the form of isolated minute tubercles, confluent caseous foci, or large areas of caseation necrosis.

(iv) **Miliary Tuberculosis**: - Lympho hematogenous dissemination may give rise to miliary tuberculosis, confined only to the lungs or involving other organs also. The distribution of miliary lesions

depends on the pathways of dissemination. Tuberculosis infection may drain via the lymphatics through the major lymphatic ducts in to the right side of the heart and thence spread into a diffuse blood borne pattern throughout the lungs alone. Because most of the bacilli are filtered out by the alveolar capillary bed, the infective material may not reach the arterial systemic circulation. Some bacilli pass through the capillaries to enter the systemic circulation and produce distant organ seedings. In other circumstances a tuberculous focus may erode directly into a pulmonary artery and thence be spread only in the pattern of supply of this single vessel to produce a localized miliary dissemination within the alveolar parenchyma. On the other hand, extension into a pulmonary vein is likely to be followed by disseminated miliary tuberculosis throughout the body or isolated organ tuberculosis.

In all instances, the miliary type of lesion takes place and the lung or other organs are studded with small millet seed like multiple focal lesion varying from one to several millimeters in diameter and are distinct, yellow white, firm areas of consolidation that usually do not have grossly visible central caseation necrosis. In the chronic conditions, the lesions undergo fibrotic changes. In the lungs the intervening alveoli may be either empty or contain caterrhal cells. In the case of miliary tuberculosis, the dose of bacillaria is massive, overwhelming and continually repeated. It is more frequent occurrence in the middle aged and elderly.

- (v) **Tuberculosis Bronchopneumonia:** - In the highly susceptible, highly sensitized individual, the tuberculous infection may spread rapidly throughout large areas of lung parenchyma to produce a diffuse bronchopneumonia, or lobar exudative consolidation, at one time descriptively referred to as 'galloping consumption'. Numerous bacilli are usually present in such exudates. <sup>(4)</sup>

**CLINICAL FEATURES:** - Tuberculosis affects any part of the body, patients of pulmonary tuberculosis catch infection often through cough, chest pain, fever and blood in the sputum are the main symptoms of tuberculosis. In a few cases, the child may be unwell with loss of appetite, fretfulness and failure to gain weight cough is not usual but may occur mimic the paroxysms of whooping cough when lymph nodes or tuberculosis granulation tissue impinge on the bronchial wall. Wheeze may be result of the same process sputum production is rare in children. Auscultation of the chest is usually unrewarding but occasionally crepitations may be heard over an extensive primary focus.

In case of miliary tuberculosis in children, and acute or subacute febrile illness onset but often especially in adults, the onset is insidious with gradual development of vague ill health, malaise, anorexia, weight loss and fever, cough breathlessness, haemoptysis and night sweats are less common. Headache as a feature suggests associated tuberculosis meningitis. Sputum may be mucoid, purulent or blood stained. Hepatomegaly, nuchal rigidity, lymphadenopathy and splenomegaly may be found in a proportion of cases choroidal tubercles are found in over 90% of children but less common in adults. Anemia is usual in cryptic miliary tuberculosis and ESR is often elevated. A variety of blood dyscrasias, including leucopenia, pancytopenia, aplastic anaemia, leukaemoid reactions, leucoerythroblastic anemia and polycythaemia, have also been seen. The liver function tests commonly disturbed, with elevation of transaminases and alkaline phosphatase. Hyponatraemia and hypokalaemia are also commonly seen. Finally amenorrhoea may be a presenting symptom, usually in severe tuberculosis. It is common for a patient with a history of recurrent colds for a number of months, exacerbations of cough. Patients may occasionally present with apparent acute pneumonia.

**BRONCHIECTASIS:** - The word being derived from Greek roots, bronchion meaning windpipe and ektasis a stretching out. Bronchiectasis is a permanent dilation and destruction of bronchi or bronchioles, this irreversible dilatation may be varicosa or sacular as opposed to cylindrical changes which often

temporarily follow pneumonia. There is extensive destruction and fibrosis of the intervening lung parenchyma. The pleura is usually fibrotic and thick with dense adhesions to the chest wall. The walls of the distended bronchi are inflamed and fibrosed. The mucosa may be ulcerated and lumen is filled with foul smelling pus. It results from bronchial obstruction with atelectasis following any number of infections such as bacterial pneumonia, measles, pertussis, tuberculosis and other granulomatous infections and some viral pneumonias and aspiration. <sup>(6)</sup>

**Aetiology & Pathogenesis:** - Most bronchiectasis is acquired during childhood. If the infection is the primary cause, the chronic inflammation of the bronchial wall causes destruction of the musculoelastic tissue resulting in dilation of bronchi whose walls are damaged. Dilatation causes accumulation of secretion and added infection with further injury to the bronchial wall. There is also a physiological block due to paralysis of cilia or metaplasia of columnar epithelium into a squamous type. All these interfere with the normal mechanism of removal of secretion. The frequency of a previous pulmonary or bronchial infection such as fibrosing pneumonia, asthma, TB or measles. Whooping cough, influenza with respiratory involvement support the theory of primary infection. Spirochaete and fusiform bacilli, often found in the dilated bronchi are secondary invaders than primary aetiological agents. Much of the alveolar tissue adjacent to the dilated bronchi is compressed, non functional and may be involved in the inflammatory process.

**CLINICAL FEATURES:** - Symptoms of bronchiectasis include chronic cough production of copious amounts of purulent sputum, hemoptysis and occasionally malodorous, sinusitis and antecedent serious chest infection occurring in healthy youngsters, age 7 or less. It is common in underdeveloped areas where it leads to respiratory failure, inanition and death in the fourth or fifth decade. Under these circumstances emphysema, corpulmonate, clubbing, pulmonary osteoarthritis amyloidosis and metastatic brain abscesses are common. Alternatively, with upper lobe involvement usually following

tuberculosis, recurrent massive hemoptysis may occur with little evidence of infection so called, dry bronchiectasis.

Immunodeficiency states that may lead to bronchiectasis include congenital or acquired 'panhypogammaglobulinemia', common variable immunodeficiency selective IgA, IgM and IgG subclass deficiencies and acquired immunodeficiency from cytotoxic therapy. AIDS, lymphoma, multiple myeloma, leukemia and chronic renal and hepatic diseases. Most of the patients with bronchiectasis have 'panhypogammaglobulinemia', presumably reflecting an immune system response to chronic airway infection.

During development of bronchiectasis in adult life, in a few patients it has been reported that some of the 'proteases' released from phagocytes during pyogenic infection may be more liable to cause bronchial wall damage if unopposed by anti proteases, of which 'α1-antitrypsin' is one. There have been occasional reports of bronchiectasis developing as a consequence of the inhalation or aspiration of toxic or irritant substances, either in liquid form or when contained in smoke or fumes. When the disease is sufficiently developed the characteristic finding is that of persistent early and midinspiratory crackles, which are localized to one or more areas. These crackles are frequently described as 'coarse' and are not shifted by coughing, unlike other lower pitched interrupted noises produced by loose secretion in large airways. <sup>(3)</sup>

**MICROBIOLOGY:** - Patients with bronchiectasis whose lungs are colonized by *PS aeruginosa* produce various substances including 1-hydroxy phenazine and also produce another pigment, pyocyanin, this is toxic to ciliated epithelium this property also shared by both *H.influenzae* and *streptopneumoniae* being highly chemotactic stimulate the migration of neutrophil leucocytes from capillaries through the bronchial wall to its lumen. These neutrophils are likely to result in the release of proteinases with consequent local pulmonary damage, these factors, that inhibit respiratory ciliary activity.

Anaerobic organisms are probably responsible for the distressing symptom of foul-smelling purulent sputum production was common. Any effort

to grow anaerobes from sputum is wasted because of their invariable habit of massive colonization of the mouth, although transtracheal aspiration has shown that they occasionally reside in the lower respiratory tract in bronchiectasis. They may be important not only as pathogens in their own right but also as  $\beta$ -lactamase producers, contributing to the failure of  $\beta$ -lactam antibiotics to control other more common pathogens. Amyloidosis is now a very rare complication of bronchiectasis-fibrils of amyloid. A protein may be deposited in any tissue in the body. If this increases sufficiently in quantity, it may disrupt normal cellular structures, producing functional disturbances. Proteinuria occurs as a result of renal glomerular disruption.

Hepatosplenomegaly and cardiac involvement may also occur. There have been occasional cases of lung cancer reportedly developing in an area of scarred bronchiectasis lung. <sup>(5)</sup>

**EMPHYSEMA (OR) COPD:** - Emphysema is defined as a condition of the lung characterized by abnormal, permanent enlargement of the airspaces, distal to the terminal bronchioles, accompanied by destruction of their walls and without obvious fibrosis. Chronic Obstructive Lung Disease (COLD) also known as Chronic Obstructive Pulmonary Disease (COPD) includes several related irreversible conditions that limit the ability to exhale. The two major diseases in this category are emphysema and chronic bronchitis. Emphysema and chronic bronchitis occur together so frequently that they are usually considered as a single entity, designated chronic obstructive pulmonary disease. Thus chronic bronchitis is defined in clinical terms, whereas emphysema is defined pathologically. Chronic bronchitis has been classified into three forms. (1) simple bronchitis defined as hypersecretion of mucus. (2) chronic or recurrent mucopurulent bronchitis in the presence of persistent or intermittent mucopurulent sputum and (3) chronic obstructive bronchitis when chronic sputum production is associated with airflow obstruction. The use of the term 'chronic obstructive bronchitis' arose from the 'British hypothesis' that persistent recurrent infection and thus chronic sputum production resulted in damage to the airways and hence airway obstruction.

The chief clinical manifestation of any type of chronic pulmonary disease are dyspnea and cyanosis. 'Dyspnea' is a sensation of shortness of breath. 'Cyanosis' is a blue tinge of the skin and mucous membrane that results from an excessive amount of reduced hemoglobin in the blood. Reduced hemoglobin is dark purple, red in contrast to normally oxygenated blood, which is bright red. The three main anatomic derangements in chronic obstructive pulmonary diseases are inflammation and narrowing of the terminal bronchioles, (2) dilation and coalescence of pulmonary air spaces and (3) loss of lung elasticity. These derangements in turn cause severe disturbances in pulmonary functions.

The presence of Emphysema implies that there has been enlargement of airspaces resulting from destruction of alveolar walls. Three major types of emphysema are recognized, according to the distribution of enlarged air spaces within the acinar unit.

- (1) 'Centriacinar Emphysema', in which enlarged airspaces are initially clustered around the terminal bronchiole.
- (2) 'Panacinar Emphysema', where the enlarged airspaces are distributed throughout the acinar unit.
- (3) 'Periacinar Emphysema', where the enlarged airspaces are long at edge of the acinar unit.

'Periacinar Emphysema' occurs less commonly than centriacinar or panacinar emphysema. When it occurs extensively in a sub pleura position and may be associated with pneumothorax. Centriacinar emphysema is more common in the upper zones of the upper and lower lobes. Where as panacinar emphysema may be found any where in the lungs but is more prominent at the bases and may be associated with  $\alpha_1$ -AT deficiency. Both types of emphysema can occur alone or in combination. Smokers can develop both centriacinar and panacinar emphysema and that those patients with centriacinar emphysema had more abnormalities in their small airways, with more muscle and smaller luminal diameters, than those with predominantly panacinar emphysema. Moreover, the patients with centriacinar emphysema had greater AHR than those with panacinar emphysema, the AHR in the former correlated with the

numbers of lymphocytes in the airways. The early stages of the disease, where a relationship has been shown between a decrease in FEV<sub>1</sub> and loss of alveolar attachments in COPD patients the best relationship with percentage predicted FEV<sub>1</sub> was the inter alveolar cell attachment distance. <sup>(3)</sup>

**PATHOGENESIS:** - The majority of the work on the pathogenesis of COPD relates to the development of emphysema and derives from the obstructive of an association between a deficiency of  $\alpha_1$ -AT and the development of early-onset emphysema, above observations from the basis of the 'protease-antiprotease' theory of the pathogenesis of emphysema. The hypothesis states that in healthy lungs the release of 'proteolytic enzymes' from inflammatory cells does not cause lung damage because of inactivation of these proteolytic enzymes by an excess of inhibitors. However, in conditions of excessive enzymes load-or where there is an absolute or functional deficiency of anti protease an imbalance develops between proteinases, and antiproteinases and in favour of proteinases, leading to uncontrolled enzyme activity and degradation of lung connective tissue in alveolar walls, leading to emphysema, the elastase was the most important proteolytic enzymes in the pathogenesis of emphysema.

The fact is that  $\alpha_1$ -AT deficiency develops emphysema in early life, particularly if they smoke, provides a simple concept for the development of emphysema resulting from a disturbance in the balance between elastase release and its activation by  $\alpha_1$ -AT. An increase in the proteinase burden, due to either the presence of increased numbers of inflammatory leucocytes in the airspaces or the release of excess protease from the leucocytes and a functional deficiency of protease inhibitors and an abnormality in the repair mechanisms for lung connective tissue. Most of the lung  $\alpha_1$ -AT is derived from the plasma, although monocytes and macrophages can also contribute the protein. Deficiency of  $\alpha_1$ -AT in the serum also results in a deficiency in the lung living fluid since  $\alpha_1$ -AT enters the lung largely by diffusion from the plasma. It appears that the development of emphysema and hence the decrease in life expectancy of  $\alpha_1$ -AT deficient subjects, occurs particularly in the presence of the additional risk factor of smoking. The

mechanisms for the development of pulmonary emphysema can be described under three major headings (1) increased protease burden. (2) decreased antiprotease function and (3) decreased synthesis of elastin. Enzymes other than neutrophils elastase have been identified in the lungs, including two present in 'neutrophils cathepsin G' and 'protease 3 cathepsin G' is a relatively weak elastolytic enzyme compared with neutrophil elastase, cathepsin G has a major role in elastin degradation alone and hence the development of emphysema. 'Protease 3' is another human neutrophil serine protease that can induce experimental emphysema.

Patients with COPD have been divided on clinical grounds and blood gas abnormalities into two extreme presentations. Type A patients or 'pink puffer' have severe dyspnea, a normal or low PaCO<sub>2</sub>, only a mild decrease in PaO<sub>2</sub> at rest, and a low DLCO. These patients are hypoxaemic only at a late stage in the disease and therefore do not develop pulmonary hypertension. Cor pulmonale and consequently fluid retention and secondary polycythaemia. In contrast, 'blue bloaters' or type B patients present with cough and sputum production and are likely to develop hypoxaemia and hypercapnia earlier in the course of their disease and hence cor pulmonale, fluid retention and polycythaemia. The 'pink puffers' were thought to have predominantly emphysema and 'blue bloaters' were the bronchitic type.

**MORPHOLOGY:** - Lungs fixed in a state of inflation, the abnormal fenestration in the walls of the alveoli the complete destruction of septa walls and the distribution of damage within the pulmonary lobule. These septal changes are found in the walls of respiratory bronchioles, alveolar ducts and alveolar spaces, in stage adjacent alveoli fuse to produce even larger abnormal airspaces and possibly blebs or bullae. Often the respiratory bronchioles and vasculature of the lung are deformed and compressed by the emphysematous distortion of the air spaces. When panacinar emphysema developed produces voluminous lungs, overlapping the heart and hiding it, when the anterior chest wall is removed. The macroscopic feature of centriacinar emphysema are less impressive. The lungs may not appear particularly pale or voluminous unless

the disease is well advanced. Generally, the upper two thirds of the lungs are more severely affected large apical blebs or bullae are more characteristic of irregular emphysema secondary to scarring.

**CLINICAL FEATURES:** - The characteristic symptoms of COPD are breathlessness on exertion. Sometimes accompanied by wheeze and cough. Production 60ml of mucoid sputum often occurring predominantly in the morning for many years. Severe breathlessness is often affected by changes in temperature and occupational exposure to dust and fumes. Some patients have severe orthopnoea. A productive cough occurs in up to 50% of cigarette smokers. In the presence of severe airway obstructions with the generation of high intra thoracic pressures, paroxysms of cough may produce syncope and cough fractures of the ribs. Patients with COPD often complain of chest tightness during exacerbation of breathlessness. Particularly during exercise and this is sometimes difficult to distinguish from ischaemic cardiac pain. Hemoptysis in association with purulent sputum may be due to inflammation or infection. Weight loss and anorexia are features of severe COPD. In general symptomatic COPD develops after 20 pack-years of smoking occupational exposure to dust has an additive effect on the decline in lung function as has been shown in coalmines, where both smoking and years of dust exposure contribute to the decline in FEV<sub>1</sub>. Variable degree of tachypnoea may be present in patients with severe COPD.

In patients with severe emphysema, cough is often slight, over distention is severe, diffusing capacity is low and blood gas values are relatively normal. Such patients may over ventilate and remain well oxygenated and therefore are euphorically if some what ingloriously designated as 'pink puffer'. On the other hand, patients with chronic bronchitis more often have a history recurrent infection, abundant purulent sputum, hypercapnic and severe hypoxemia, prompting the equally inglorious designation of 'blue bloaters'. A hazard in severe emphysema, in addition to the respiratory difficulties, the development of cor pulmonale and eventual congestive heart failure. Death in

most of these patients is due to (1) right sided failure (2) respiratory acidosis and coma, and (3) massive collapse of the lungs secondary to pneumothorax.

### **OTHER TYPES OF EMPHYSEMA: -**

(i) Compensatory Emphysema. (ii) Senile Emphysema. (iii) Obstructive over inflation. (iv) Bullous Emphysema. (v) Interstitial Emphysema.

**Compensatory Emphysema:** - This term is sometimes used to designate dilation of alveoli in response to loss of lung substances elsewhere. It is best exemplified by the hyper expansion of the residual lung parenchyma that follows surgical removal of a diseased lung or lobe. In most instances this constitutes compensatory hyper inflation as there is no accompanying destruction of septal walls.

- (i) **Senile Emphysema:** - Senile emphysema refers to the over distended sometimes voluminous lungs found in the aged. These changes result from age related alteration of the internal geometry of the lung larger alveolar ducts and smaller alveoli. There is no significant loss of elastic tissue and because there is no destruction of lung substance and the respiratory deficit is usually minimal, a better designation for such aging lungs would be senile hyper inflation.
- (ii) **Obstructive Over Inflation:** - Obstructive over inflation refers to the condition in which the lung expands because air is trapped within it. A common cause is sub total obstruction by a tumor or foreign object. A classical example is congenital lobar over inflation in infants. This is a congenital anomaly, probably due to hypoplasia of bronchial congenital cardiac and lung abnormalities. Over inflation in obstructive lesions occurs either ball-valve action of the obstructive agent so that air enters on inspiration but cannot leave on expiration or the bronchus may be totally obstructed, but ventilation through 'collaterals' may bring in air from behind the obstruction. These collaterals are represented by the pores of Kohn and other direct

accessory bronchiolo alveolar connections. Obstructive over inflation can be a life-threatening emergency because the affected portion extends sufficiently to compress the remaining normal lung.

- (iii) Bullous Emphysema: - Bullous emphysema refers merely to any form of emphysema that produces large subpleural blebs or bullae. They represent localized accentuations of one of the four forms of emphysema are most often subpleural and occurs near the apex, sometimes in relation to old tuberculous scarring. On occasion rupture of the bullac may give rise to pneumothorax.
- (iv) Interstitial Emphysema: - The entrance of air in to the connective tissue stroma of the lung mediastinum or subcutaneous tissue is designated interstitial emphysema. In most instances, alveolar tears in pulmonary emphysema provide the avenue of entrance of air in to the stroma of the lung but rarely a wound of the chest that allows air to be sucked in or a fractured rib that punctures the lung substances may underline this disorder. Alveolar tears usually occur when there is a combination of coughing plus some bronchiolar obstruction, producing sharply increased pressures within the alveolar sacs children with whooping cough and bronchitis, patients with obstruction to the airways and individuals who suddenly inhale irritant gases provide classic examples.

Progressive accumulation of air may dissect through the fibrous connective tissue of the alveolar walls and into and along the fibrous septa of the lung to reach the mediastinum and thence possibly the subcutaneous tissues. If the collection of air small, it usually has no clinical importance. However, extensive insufflation of the lung may encroach upon the small blood vessels to create serious impairment of blood flow through the lungs. When interstitial air treks into the subcutaneous tissue, the patients may literally blowup into an alarming, although usually harmless. 'Walt Disney Balloon' appearance with marked swelling of the head and neck and crackling

crepitation all over the chest. In most instances such air is resorbed promptly as soon as the point of entrance is sealed. <sup>(4)</sup>

### CAUSES OF OBSTRUCTIVE LUNG DISEASES

- (i) **SMOKING**: - Cigarette smoke is the cause of over 80% of all cases of chronic obstructive lung disease. It contains irritants that inflame the air passages, setting off a cascade of biochemical events that damage cells in the lung, increasing the risks both for Chronic Obstructive Lung Disease (COLD) and lung cancer. It is now thought that emphysema develops when inflammation caused by smoke incites the body's immune system, particularly large white blood cells called macrophages, to over produce an infection-fighting enzyme known as macrophage elastase. This overactive enzyme attempts to reverse the inflammation by attacking healthy cells in bronchioles as if they were foreign proteins like bacteria or viruses. Blocking the enzyme might help to prevent or halt the progression of emphysema. Although smokes appear to have sufficient and even high amounts of the protective alpha-1-antitrypsin (AAT) protein that ordinarily neutralizes these proteases, smoke generates oxygen-free radical particles that deactivate the AAT and render it ineffective.

Another important enzyme produced in the liver is microsomal 'epoxide hydrolase', which is responsible for the breakdown of harmful oxidants found in the cigarette smoke. Two variants of the gene regulating the enzyme cause it to act either rapidly or slowly. A 1997 study showed that, compared to healthy people, those with COLD are four to five times more likely to have the genetic variant that shows the action of this enzyme, possibly making such people more vulnerable to lung damage. Cigarette smoke causes chronic bronchitis through inflammation and damage to the airways. It also damages the cilia, hair-like waving projections that

move bacteria and foreign particles out of the lungs increasing the risk for infections that can lead to chronic bronchitis.

- (ii) **GENETIC FACTORS:** - Genetic factors that cause lungs to be hyper-reactive to stimulants and allergens may also increase the risk for COLD. An inherited condition that causes a deficiency in AAT can trigger early-onset emphysema, even in non smokers known as alpha-I-antitrypsin deficiency (AIAD) related emphysema, it accounts for only about 3% of all emphysema cases. The AAT protein is produced in the liver and neutralizer the effects of the protease enzymes, most importantly neutrophils elastase, which attacks the cell linings in the lungs, without adequate amounts of AAT, the enzymes destructive action is even more pronounced, causing early progressive damage to the lungs. In such cases, both the walls of the alveoli and the airways leading to them are damaged. Because smoke deactivates may residual amounts of AAT that these patients are able to produce, patients with AIAD who smoke have no chance at all for escaping emphysema. Researches recently identified a group of patients who might have an inherited form of COLD that is unrelated to AIAD. In such patients, a genetic susceptibility may increase the effects of smoking so that severe COLD develops at an earlier age than usual. People exposed for a long time to toxic fumes, industrial smoke, dusts from mines and other air pollutants are also at increased risk for COLD. About 2 million people have emphysema, of those who have this condition, 55% are men and 45% are women, although the prevalence in women increased by 24% between 1982 and 1993. Thinness is associated with emphysema; obesity is linked to chronic bronchitis. <sup>(9)</sup>

A great deal of whole has already been done with regard to clinical pathology and investigation and literature abounds in such studies. Acid phosphatase levels in a serum and cells are

has been reported in which a deficiency of lysosomal acid phosphatase is observed. In contrast, in the lysosomal storage disease mucopolipidosis II (I-cell disease), although the patients fibroblast are deficient in several other lysosomal hydrolases, the level of acid-phosphate is reportedly normal. <sup>(Ref) 95</sup> Von Figura et al found that this Acid phosphatase is infact present in the lysosomal fractions of mucopolipidosis II fibroblasts as well as in normal fibro blasts.

However the proposed work that focuses on the role of lysosomal acid hydrolases in the sera from patients afflicted with lung related problems has hitherto not been undertaken. This puts the proposed work into a rather peculiar situation and definite advantage for pursuing this exciting research area in cell Biology, Developmental Biology, Clinical Biochemistry and medical Biochemistry and molecular Biochemistry.

## OBJECTIVES

The present investigation ~~what follows in next pages~~ is aimed to bridge gap in the knowledge of the lung-related diseases by taking for parameter that might be useful for clinical evaluation of above diseases.

As the blood serums inevitably contains the cellular as well as soluble components or products of a metabolic, physiologic, immunologic reactions investigation were carried out on the human sera derived from patients affected with Asthma, Tuberculosis, Bronchiectasis and Emphysema as well as Normal subjects.

Determination of the following parameters constituent of blood will serve to imminent on the etiology of these diseases are help in diagnostic evaluation.

The parameters studied are:-

1. To determine the value of Hemoglobin in normal and lung patients.
2. To determine the Red Blood cells and white Blood cells count in the blood of control subjects and pulmonary patients.
3. To estimate the amount of serum glucose and protein in normal and clinical patients.
4. To investigate the enzymes like Serum Glutemate Oxalo Amino Transferase, Serum Glutemate, Alanine Amino Transferase, Acid Phoshatase and Lactate Dehydrogenase activity is detected in the serum of normal subject and lung diseased patients.