1. INTRODUCTION

1.1 Epidemiological Study

Globally, about 300 million people are suffering from bronchial asthma. The evidence for a world-wide increase in asthma cases is strong and consistent. Now, asthma is considered as a serious health and economic hazard. Asthma costs the United States $56 billion each year. In 2008, asthma caused 10.5 million missed days of school and 14.2 million missed days of work. In 2010, 8.7 million adults had asthma. That is equal to 1 in 12 adults. 7 million children had asthma. That is equal to 1 in 11 children. In United States, in the year 2009, asthma caused 4,79,300 hospitalizations, 1.9 million emergency department visits and 8.9 million doctor visits. A pattern of geographical distribution began to emerge from international studies such as the European Community Respiratory Health Survey (ECRHS 1996) and the International study on Asthma and Allergies in Childhood (ISAAC 1998), that provided comparable data on the prevalence of asthma in different parts of the world; the highest prevalence of asthma was observed in Westernized countries, intermediate values in other Western countries and the lowest prevalence occurred in most developing countries like India, Pakistan, China, and in eastern Europe.

The hypothesis made to explain the epidemic trend, fall into two main groups: One that points to increasing exposure to aggressive factors, and the other that implicates decreasing exposure to protective factors. The important aggressive factors are airborne indoor or outdoor pollutants (Behrend 1995) high salt intake (Burney 1987) indoor allergens (Woolcock 1995) drugs (Wjst 1997) and vaccines (Rook GAW1998). The important protective factors are antioxidants (Soutar 1997), microbial burden (Strachan 1989 and Martinez 1999) and physical exercise (Platts - Mills 1998).

Hence Asthma is itself defined as a ‘Multifactorial disease’ with a large series of causative, inducing, triggering and aggravating factors, interacting with the expression of genetic background at a given age.

ISAAC Phase 1 studies demonstrated a significant correlation between the prevalence of asthma in a given country and that of allergic rhino conjunctivitis and atopic eczema (1998). In the British cohort studies, the occurrence of hay fever was inversely related to the overall number of siblings. It is also consistently noted that farmers rarely suffer from atopic diseases, including allergic asthma. Children of full
time farmers were even more protected from atopy than those of part-time farmers (Braun – Fahrlander 1999).

Epidemiological survey of allergic diseases were done in different countries to know the cause. The following were the prevalence of allergic diseases Asthma, Atopy, and Airway hyper responsiveness in several countries.

**Table 1.1**

**Prevalence of asthma in children in studies using airway hyper responsiveness as a test**

<table>
<thead>
<tr>
<th>Name</th>
<th>Study year</th>
<th>Number of children studied</th>
<th>Age</th>
<th>Current asthma</th>
<th>Diagnosed asthma</th>
<th>AHR</th>
<th>Atopy (SPT)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Australia</td>
<td>82</td>
<td>1487</td>
<td>8 to 10</td>
<td>5.4</td>
<td>11.10</td>
<td>10.1</td>
<td>29.3</td>
</tr>
<tr>
<td></td>
<td>86</td>
<td>1217</td>
<td>8 to 11</td>
<td>6.7</td>
<td>17.3</td>
<td>10.0</td>
<td>31.9</td>
</tr>
<tr>
<td></td>
<td>91</td>
<td>1575</td>
<td>8 to 11</td>
<td>9.9</td>
<td>30.8</td>
<td>16.0</td>
<td>37.9</td>
</tr>
<tr>
<td>England</td>
<td>80</td>
<td>1613</td>
<td>8</td>
<td>8.0</td>
<td>16.0</td>
<td>?1.1</td>
<td></td>
</tr>
<tr>
<td>Denmark</td>
<td>87</td>
<td>527</td>
<td>7 to 16</td>
<td>5.3</td>
<td></td>
<td>16.0</td>
<td>31</td>
</tr>
<tr>
<td>Indonesia</td>
<td>81</td>
<td>406</td>
<td>7 to 15</td>
<td>1.2</td>
<td>2.3</td>
<td>2.2</td>
<td>24.1</td>
</tr>
<tr>
<td>China</td>
<td>88</td>
<td>3067</td>
<td>11 to 17</td>
<td>1.9</td>
<td>2.4</td>
<td>4.1</td>
<td>?30</td>
</tr>
<tr>
<td>Papua New Guinea</td>
<td>85</td>
<td>257</td>
<td>6 to 20</td>
<td>0</td>
<td>0</td>
<td>1.0</td>
<td>17</td>
</tr>
</tbody>
</table>

Legend

AHR - Airway hyper responsiveness  
SPT - Skin Prick Test


To estimate changes in prevalence of respiratory symptoms and the reported diagnosis of asthma, eczema and hay fever, Ninan and Russell studied two groups of 8 - 13 year old primary school children in Aberdeen, Scotland in 1964 (n=2743) and 1989 (n=3942). The results showed that prevalence of wheeze and episodes of shortness of breath were higher in boys than in girls and significantly increased between 1964 and 1989 (wheeze 10.4% Vs 19.8%; shortness of breath: 5.4% Vs 10.0%).

Rim pela et al studied the change in prevalence of asthma and allergic rhinitis among 12-18 year old Finnish adolescents from 1977 (n=4335) to 1991 (n=3059) in order to determine if any increase correlated with either a particular sub-population or
changes in genetic susceptibility. Results showed that the prevalence of asthma and allergic rhinitis almost tripled between 1977 and 1991 (asthma 1.0% Vs 2.8%; allergic rhinitis 5.0% Vs 14.9%) and was similar in all groups. The author concluded that this represented a real increase in prevalence which could possibly be explained by changes in indoor air pollution over the study period. Between 1991 and 1993 Peat et al compared the prevalence and severity of asthma and allergic sensitization in 6394 children (8 - 11 years) living in seven climatic regions of New South Wales, Australia. The results showed all regions to have high prevalence of wheeze. The authors concluded that there may be some relation to current asthma prevalence and different levels of allergic sensitization.

The above studies show that expression of atopic diseases arises from a combination of genetic, environmental and immunological influences. Much epidemiological evidence suggests that environmental changes may be implicated in the recent ‘allergy epidemic’. Candidate factor include: reduced exposure to microbial antigens, exposure to specific allergens, pro-inflammatory ‘Western’ diets and organic and inorganic environmental pollutants in early life (Holt 1997).

Various hypotheses and factors have been put forward previously like:

- Genetic factors
- Non-Genetic factors
  i) Allergen hypothesis, ii) Diet hypothesis, iii) Hygiene hypothesis, iv) Antenatal and postnatal infection, v) Intestinal microbial flora (microbial load), which together constitute non-genetic factors.

### 1.1.1 Genetic Factors

Robert Cooke and Albert Vander Veer established that 48.4% of their atopic probands had a family history of atopy, compared with 14.5% in the control population. Further studies had indicated that environmental factors also play a large role. Hence, it is very difficult to identify which genes contribute to a complex multifactorial disease such as asthma and atopy.
1.1.1 Candidate – Gene approach

A good example of the use of the candidate – gene approach in the identification of a region on chromosome 5, region 5q 31-33, by Marsh et al (1982) that appears to be linked to the development of asthma using a candidate – gene approach. In Amish cohort study, this region was investigated because it includes cluster of cytokine genes, among them the genes that encode 1l-3,1l-4,5,9 and 13 as well as the gene that encodes granulocyte – macrophage colony – stimulating factor.1L-4 is thought to be a good candidate gene, since it induces the Ig class switch to IgE (Marsh et al. 1994)

1.1.2 Genome Scans in Asthma and Atopy

The first genome - wide scan to be completed for asthma and atopy – related phenotypes used 269 micro satellite makers across the whole genome for four quantitative traits (log IgE, skin test index, log eosinophil count, log slope for BHR and 1 quantitative trait atopy). This study identified b region of potential linkage, on chromosomes 4q35, bp21.3-23, 7p35, 11q13, 13q14.3-32.2 and 16p24.1. Another study group in U.S.A. identified 6 novel regions on 5p15, 17p11.1, 11p15, 19q13, 2q22-33 and 21q21 showing linkage to the asthma phenotype. They also detected linkage to the previously reported as showing to the asthma phenotypes. 5q22-33, 6p21.3-23, 12q14-24.2, 13q14.143-32.2 and 8p23.3

1.1.2 Non Genetic Factors
1.1.2.1 Pollution hypothesis

The ‘pollution hypothesis’ explains why atopic diseases are more frequent in urban and industrialized area and ‘not’ why they are less frequent in large and disadvantaged families or in heavily polluted cities of eastern Europe or the Republic of China.

1.1.2.2 Diet hypothesis

In Diet hypothesis, there is strong epidemiological evidence that increased dietary n-3 PUFA (Poly Unsaturated Fatty Acids) intake is associated with lower prevalence of asthma and respiratory disease (Hodge 1996, Peat 1992). Prevalence of asthma and atopic diseases are more in those who have decreased consumption of anti-
oxidants (e.g. Vitamin C). This nutrient hypothesis, however do not explain the sib ship size and birth order effect.

One of the most significant dietary changes with increasing urbanization is declining consumption of anti-inflammatory (and anti-th1) n-3 poly unsaturated fatty acids (n-3 PUFA) and increasing intake of pro inflammatory (pro-Th2) saturated fat and synthetic and n-6 PUFA. There is strong epidemiological evidence that increased dietary n-3 PUFA intake is associated with lower prevalence of asthma and respiratory disease.

The action of n-3PUFA is:

a) Inhibits lymphoproliferation
b) Inhibits pro inflammatory cytokines.
c) Inhibits MHC class-1 expression antigens and PGE2 which normally enhances IgE production.
d) Inhibits migration of inflammatory cells into local tissues by down regulating adhesive molecule expression.
e) n-3 PUFA are important for successful pregnancy and foetal development.
f) n-3 PUFA are concentrated in the foetal circulation (at 10-15 times maternal levels)

Declining exposure to eosinophil and food-borne microbes, may therefore underline the allergy and asthma (Sepp 1997). But, there is some consistency among all these hygiene related exposures that it is early life exposure that confers protection (de Meer, 2004).

1.1.3 Hygiene

Infections in the early life give protection or causes asthma. There is now more or less consensus that the maturation of the immune system after birth is largely dependent on microbial stimulation. As the placenta is embryonic in origin, it contains genetic material from both parents and may be treated as antigenically foreign by the maternal immune system. There is evidence that Th1 driven all mediated immunity which is detrimental to the feto-placental unit is down regulated during pregnancy. This inhibition of cell mediated Th-1 responses appear to be mediated by hormonally driven shift towards Th-2 responses(1L-4,1L-5,1L-10,and 1L-13) at the feto-maternal interface
which form hormonal immunity. This adaptive ‘Th-2 shift’ in pregnancy was first proposed by Wegmann and colleagues in 1993. This ‘Th-2 shift’ is called as immune deviation.

**Figure 1.1**

**Pre-natal and post-natal events**

Infants, who subsequently develop atopy, show failure of immune deviation and subsequent consolidation of their neonatal patterns of Th2 polarized allergen specific immunity. In addition to post-natal factors, it is possible that antenatal factors interfere with Th1 maturation. After birth the new born infant repeatedly encounters high level of inhalant allergens from the environment and these exposures shape the nature of developing the memory cell populations. In 1978, Stannegard reported a primary defect of lower number of lymphocytes in children with atopic dermatitis.

It is well established that infection and other microbial pressure may drive the immune system towards the Th1 like response. An infection at the time of sensitization facilitates skewing towards Th-1 type response even in higher responder rate with a genetic propensity for IgE antibody formation.

It was recently reported that the treatment of children with broad spectrum antibodies during the first 2 years of life was associated with an increased risk for allergy during adolescence and early adulthood, independent of why the antibiotics were given. This would indicate that the effects could have been the consequence of the impact on the intestinal flora. The total mucosal surface area of the adult human intestinal tract is up to 3.00m², making it the largest body area interacting with environment. It is colonized with over 10^{14} microorganisms; weight over 1kg and corresponding to more than 10 times the total number of cells in the body. Thus the gut
flora is quantitatively the most important source if microbial stimulation. Rook and Stanford recently suggested two major syndromes that could be the result of inadequate microbial stimulation early in life. One is called as ‘input deprivation syndrome’, where there is inadequate priming of T helper cells, leading to an incorrect cytokine balance and the second they coined as ‘uneducated T-cell regulation syndrome’, where there is a failure to fine tune the T cell repertoire in relation to epitopes that are cross-reactive between self and micro-organisms. The gut immune system has the capacity to distinguish between potentially harmful antigens, for example microbes, and harmless antigens, for example, foods. Lactobacilli and eubacteria were more common and the counts were higher in Estonia than in Swedish infants, whereas the reverse was true for clostridiums. Bifid bacteria were more prevalent in non-allergic children, while the counts of coliform bacteria were higher in the atopic children. The findings do however indicate that it may be at least as important to look at changes in the internal environment as to look in the external environment when trying to explain regional differences in the prevalence of allergy. There is preliminary evidence that lower CD14 levels at 16 weeks of gestation are associated with atopic eczema at 1 year of age.

1.1.4 Other antenatal exposures which may relate to the development of asthma

There is growing evidence of relationship between in utero tobacco smoke exposure and the subsequent development of asthma in childhood. In the East Boston neighbourhood, health study, Hanrahan et al studied 80 infants and found an association between maternal smoking during pregnancy and decline in expiatory flow at functional residual capacity, approximately 4 weeks after birth. It is becoming clear that events in very early life when the immune system is more relevant to environmental influences hold the keys to both disease etiology and to potential intervention strategies. Hence general improvements in public health and decreased early childhood exposure to bacteria in western culture may unmask Th-2 propensity in these predisposed individuals.
Table 1.2

Summary of genome – wide scans for asthma and atopy – related phenotypes

<table>
<thead>
<tr>
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<tbody>
<tr>
<td>2pter</td>
<td></td>
<td></td>
<td></td>
<td>BHR/asthma</td>
<td>IgE/RAST</td>
</tr>
<tr>
<td>2q22-33</td>
<td>Asthma-Hispanic</td>
<td>IgE (Der p 1)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3p24.2-22</td>
<td></td>
<td></td>
<td></td>
<td>BHR/asthma</td>
<td></td>
</tr>
<tr>
<td>4q35</td>
<td>Asthma-African-American</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5p15</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5q23-33</td>
<td>Asthma-Caucasian</td>
<td>IgE (Der p 1)</td>
<td></td>
<td>Asthma, eosinop</td>
<td>IgE/RAST</td>
</tr>
<tr>
<td>6p21.3-23</td>
<td>Asthma-Caucasian</td>
<td>IgE (Der p 1)</td>
<td>IgE/Eos</td>
<td></td>
<td></td>
</tr>
<tr>
<td>7p35</td>
<td></td>
<td></td>
<td></td>
<td>IgE/BHR</td>
<td></td>
</tr>
<tr>
<td>8p23.3</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>9 (D9S925 and DS1784)</td>
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<td></td>
<td></td>
<td>Asthma/BHR</td>
<td>Asthma, IgE/RA</td>
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<tr>
<td>11p15</td>
<td>Asthma-Caucasian</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>11q13</td>
<td></td>
<td></td>
<td></td>
<td>IgE/skin test</td>
<td></td>
</tr>
<tr>
<td>12q13</td>
<td>Asthma-Caucasian</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>12q14-24.2</td>
<td>Asthma-Caucasian</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td></td>
<td>Asthma-Hispanic</td>
<td></td>
<td></td>
<td>Asthma</td>
<td></td>
</tr>
<tr>
<td>13q14.3-32.2</td>
<td>Asthma-Caucasian</td>
<td>IgE (Der p 1)</td>
<td>Atopy</td>
<td>BHR</td>
<td></td>
</tr>
<tr>
<td>14q11.2-13</td>
<td>Asthma-Caucasian</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>16q24.1</td>
<td></td>
<td></td>
<td></td>
<td>IgE/BHR</td>
<td></td>
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<tr>
<td>17p11.1-11.2</td>
<td>Asthma- African Americans</td>
<td></td>
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<td>19q13</td>
<td>Asthma-Caucasian</td>
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<td>Asthma</td>
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<td>Asthma-Hispanic</td>
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<td>Asthma/BHR</td>
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<td>Linkage criteria</td>
<td>P &lt; 0.01</td>
<td>P &lt; 0.001</td>
<td>P &lt; 0.001</td>
<td>P &lt; 0.01</td>
<td>P &lt; 0.01</td>
</tr>
</tbody>
</table>

Legend

*Der p1, Principal house dust mite allergens,* 
*BHR, Bronchial Hype responsiveness, RAST,* 
*Radio Allergosorbent Test for allergen specific IgE,* 
*IgE, Total Serum IgE.*
Despite intensive efforts of the last decade, no susceptibility genes for asthma and atopy have been identified with any certainty. Evidence has been presented that several regions of the human genome-chromosomes 5q 31-33, 6p 21.3, 7q35, 11q13, 12q14.3-24.1, 13q14.2, 14q11.2-13, 16p12-contain genes implicated in the expression of asthma and allergy. Understanding of the genetic basis of asthma and atopy will promote the identification of important pathogenic disease mechanisms, the development of novel therapeutic interventions, and the development of methods for early diagnosis and disease prevention.

Although, in the early period of asthma epidemic, more cases are being reported in different countries, later when detailed survey was taken in different population, the spatial distribution of asthma showed uniformity with in many developed countries. Although same levels of outdoor concentration of ozone (O₃) and nitrogen dioxide (NO₂) showed urban–rural geographical differences, there was surprisingly little evidence of major urban–rural gradients in asthma and morbidity.

The occurrence rate was more or less same in different area. The study conducted by Gaur (2006), to find out current prevalence of bronchial asthma and allergic rhinitis belonging to different occupational strata of urban and rural population of Delhi with 5900 subjects - comprising of urban 4348 people and rural 1552 people using questionnaire, clinical evaluation by physician found out no significance difference in asthma prevalence between urban and rural population. In New Zealand, a cohort study conducted by Willkie (1995) using ISAAC questionnaire to know the prevalence asthma symptoms in children living in Christ - Church and Hornby, an industrial suburb, and the authors found no evidence of an increase in asthma symptoms in children living in the industrialized suburbs of Hornby. In a detailed study of 1576 life-long non-smokers in Beijing, China (XuX 1993), living in different area like industrial, residential and suburban, it was observed that the prevalence of ‘Wheeze’ did not differ greatly between the three geographical area. The study conducted in mid-Western and Eastern cities of USA in the late 1970’s and 1980’s showed no significant variation in the occurrence of ‘wheeze’ with the levels of any air pollutants (Dockery, 1989, Ware, J.H., 1986).
In the study conducted by Alameldin (2012) with 1048 students (482 boys and 566 girls) in Assist District, Egypt, over all prevalence of bronchial asthma was 6.2% and no significant difference was found between urban and rural areas (P = 0.075).

### 1.2 Asthma

#### 1.2.1 Definition

Asthma is defined as a chronic inflammation of the airways in which inflammation of the central event which leads to the development of bronchial hyperreactivity and asthmatic symptoms – International consensus Report on Diagnosis and Treatment of asthma (1992).

Immunologically, it can be defined as dysfunction of immune system.

Asthma is now defined as a chronic inflammatory disorder of the airways, associated with widespread but variable airflow obstruction that is often reversible spontaneously or with treatment. Increased airways responsiveness and airway inflammation characterized by Th2 lymphocyte infiltration with eosinophils and neutrophils are characteristic of the disorder.

Asthma is a complex disorder involving a combination of genetic and environmental interactions that lead to airway inflammation characterized by T-helper 2 cell polarization and airway wall remodelling accompanied by extensive epithelial dysfunction.

#### 1.2.2 Causes of asthma

In certain patients, allergens like airborne (dust mite, dander, hairs of cat, dog, goat etc.,) or blood - borne (viral antigen, food etc.,) trigger on asthmatic attack. They are called as extrinsic causes. In other causes, an asthmatic attack can be induced by exercise or cold, apparently independent of allergen stimulation (intrinsic asthma).

The asthmatic response can be divided into two phases: a) Early phase and b) Late phase

The early response occurs within minutes of allergen exposure and primarily involves histamine, leukotrienes (LTC4) and prostaglandin (PGD2). The effects of these mediators lead to Bronchoconstriction, vasodilation and some build-up of mucus.
The late response occurs hours later and involves additional mediators, including IL-4, IL-5, IL-6, TNF-α, eosinophil chemotactic factor (ECF), and platelet-activating factor (PAF).

**Figure 1.2**

**Early phase and Late phase**

These mediators induce expression of adhesion molecular on endothelial cells, including eosinophils and neutrophils into the bronchial tissue.

These events lead to occlusion of the bronchial lumen with mucus, proteins, and cellular debris; sloughing of the epithelium; thickening of the basement membrane fluid build-up in the intestinal tissue and alveolar sac causing oedema; and hypertrophy of bronchial smooth muscle. A mucus plug often in asthma forms and adheres to the...
bronchial wall. The mucus plug contains clusters of detached epithelial cell fragments, eosinophils, some neutrophils, and spinals of bronchial tissue known as Curschmann’s spinals.

1.2.3 Various Types of Asthma

1.2.3.1 Extrinsic or Allergic asthma

1.2.3.2 Intrinsic asthma

1.2.3.3 Cough variant asthma

1.2.3.4 Exercise Induced asthma

1.2.3.5 Occupational asthma

1.2.3.6 Aspirin Induced asthma

1.2.3.7 Nocturnal asthma

1.2.3.8 Health Conditions that mimic or co-exist with asthma

1.2.3.1 Extrinsic or Allergic asthma

Allergies and asthma often go hand in hand. Allergic rhinitis (also called hay fever) is inflammation of the inside lining of the nose and is the single most common chronic allergic disease. In those with allergic rhinitis, increased sensitivity (allergy) to a substance causes your body’s immune cells to release histamines in response to contact with the allergens. Histamines, along with other chemicals, lead to allergy symptoms. The most common allergens enter the body through the airway.

With allergic rhinitis, you may feel a constant runny nose, ongoing sneezing, swollen nasal passages, excess mucus, weepy eyes and a scratchy throat. A cough may result from the constant post nasal drip. Many times asthma symptoms are triggered by allergic rhinitis.

1.2.3.2 Intrinsic asthma

Cold, dry air induced asthma, exercise induced asthma are intrinsic in nature.

1.2.3.3 Cough variant asthma

In cough variant asthma severe coughing is the predominant symptoms. There can be other causes of cough such as post nasal drip, chronic rhinitis, sinusitis, or gastroesophageal reflux disease (GERD or heart burn). Sinusitis causes postnasal drip

A Comparison study of Serum Histamine binding capacity in normal and allergic patients
leading to cough. Asthma associated with sinusitis will cause severe cough, because of sinusitis with asthma is common.

1.2.3.4 Exercise Induced asthma

Of many types of asthma, exercise induced asthma occurs in few people. Although people with asthma experience some degree of breathing difficulty with exercise, many people including football and hockey players experience some degree of asthmatic symptoms with exercise. asthma experience some degree of symptoms with exercise. With exercise induced asthma, airway narrowing peaks 5 to 20 minutes after exercise begins making it difficult to breathe.

1.2.3.5 Occupational asthma

Some people handling with cleaning materials in houses or work places also will develop occupational asthma. With this type of asthma patient get difficulty in breathing and asthma symptoms just on the day they are on job. Because of the irritant substances that are handled by these workers, they may develop running nose, congestion, eye irritation. Sometimes people with this type of asthma suffer with or have a cough instead of the typical asthma wheezing. Workers working in cement factories, coir industries, hairdressers, wood workers and farmers are some of the who develop occupational asthma.

1.2.3.6 Aspirin Induced asthma

Aspirin Induced asthma affects 5-8% of asthmatics. It may cause life-threatening bronchospasm as well as dermal, naso-ocular and gastro-intestinal symptoms. Patients with this susceptibility have elevated levels of urinary leukotrienes at baseline and even higher urinary leukotriene levels following aspirin challenge. The physiological effects of aspirin challenge in aspirin-sensitive patients, pre-treated with zileuton is almost completely blocked, as such patients failed to develop any clinically significant adverse effects and urinary LTE₄ levels were reduced by 68%. The bronchospasm in aspirin-sensitive asthma is mediated by leukotrienes and that leukotriene modifiers are the treatment of choice for these patients.
1.2.3.7 Nocturnal asthma

Night time asthma also called nocturnal asthma is a common type of the disease. If one is having asthma, having symptoms are much higher during sleep because asthma is powerfully influenced by the sleep wake cycle (circadian rhythms) Asthma symptoms if wheezing, cough and trouble breathing are common and dangerous particularly at night time.

It is thought that this may be because of increased exposure to allergens (asthma triggers), cooling of the airways, reclining positions or even hormone secretions that follow a circadian pattern. Diseases like GERD (Gastro Oesophageal Reflux Disease) and sinusitis may cause asthma at night time due to acid escaping from the stomach reaching the trachea in GERD and the viscid mucus secretions reaching the trachea, irritates trachea causing cough and asthma at night time.

1.2.3.8 Health Conditions that mimic or co-exist with asthma

Children with asthma, uncontrolled by conventional treatment need careful re-evaluation to establish the cause of symptoms. Persistent provoking factors may play a part. Alternative diagnosis must be considered. Diseases that mimic or co-exist with asthma are:

- Gastro oesophageal reflex disease GERD
- Cystic fibrosis
- Bronchiectasis
- Tracheomalasia
- Immune deficiency
- Right sided aortic arch
- Vascular Ring

They can be diagnosed based upon history, clinical factures and investigations such as MRI (Magnetic Resonance Image), C.T. scan (Computerized Tomography scan) Endoscopic test to rule out reflux oesophagitis, x ray chest, blood analysis, etc.,

1.2.4 Social and Economic Impact of Asthma

According to 2002 statistics from the centre for disease control, 20 million people suffer from asthma in the United States and 12 million per year experience an
asthma attack. About 5000 people die each year from asthma. In years, the prevalence of asthma in the western world has doubled. Approximately 3 million Americans are allergic to peanuts and tree nuts (almonds, cashews and walnuts). At least 50% of serious reactions are caused by accidental exposures to peanuts, tree nuts or their products. There is a staggering cost – estimated to be almost 56 billion in terms of lost work time and for caregivers.

1.3 Pathophysiology of Asthma

1.3.1 Expression of atopic disease

Over the last 10-20 years, the atopy and asthma increase progressively. It has made the need to identify the factors responsible, and to develop preventive strategy urgent. Recent studies suggest that the development of immune responses to specific environmental allergens begins in vitro. Here the maternal immune system mediates interactions between the feto-placental unit and environment. This interaction may influence differential patterns of T-cell memory response to allergens in atopic (TH2) and non-atopic (TH1) individuals.

Indeed, there is convincing evidence that the immune system is primed to respond to environmental antigens before birth. IgE specific for certain allergens can be detected at birth. TH1 immaturity which predates allergic disease expression is now considered to be an important factor in disease pathogenesis. In neo-natal period, in both allergic and non-allergic individuals, there will be more TH2 cells. Normal infants show rapid down regulations of this TH2 cell response in the first year of life, and progressive development of reciprocal TH1 like pattern of immunity. This is called ‘immune deviation’, but, in allergic children, there is failure of this immune deviation that leads to TH2 polarized allergen specific immunity.

Delayed maturation of APCs has been proposed as a mechanism of the perinatal TH1- immaturity associated with atopic disease. The APCs produce IL-12, a major factor in promoting TH1 differentiation in response to allergens. While mature IL-12 promotes TH1 differentiation, defective IL-12 signaling results in a TH2 differentiation.

Other factors that could influence immune deviation include antigen exposure, maternal atopic status, the nature of antigen presentation etc. Genetically predisposed
individuals - those with Th1 maturation - maybe more susceptible to environmental influences which favor Th2 selection.

Hence, expression of atopic disease arises from a combination of genetic, developmental and immunological influences.

1.3.2 Mechanism of degranulation

IgE – mediated degranulation begins when an allergen cross – links IgE that is bound to the Fc receptor on the surface of a mast cell or basophil. Binding of IgE to Fc&RI apparently has no effect on a target cell. It is only after allergen cross – linkage is indicated by the finding that monovalent allergens which cannot cross-link the fixed IgE, also do not initiate degranulation.

**Figure 1.3**

*General mechanism underlying immediate type I hypersensitivity reaction*

1.3.3 Intracellular events that trigger mast cell degranulation

The intracellular signalling events that result is mast-cell degranulation are multifaceted, involving co-operation among various protein and protein and lipid kinases and phosphatases and rearrangement of cytoskeleton.
Cross-linkage of Fc & R1 receptors in mast cells, activates phosphorylation. Within 15 seconds after cross-linkage of Fc & R1, methylation of various membrane phospholipids is observed, resulting in an increase in membrane fluidity and the formation of Ca\(^{2+}\) channels. An increase of Ca\(^{2+}\) reaches a peak within 2 minutes of Fc&RI cross-linkage. The increase in Ca\(^{2+}\) promotes the assembly of microtubules and the contraction of microfilaments, both of which are necessary for the movement of granules to the plasma membrane. This granule fusion with the membrane of the cell leads to exocytosis of the primary mediators, associated with mast-cell degranulation.

In addition the Ca\(^{2+}\) increase causes hydroxylation of membrane phospholipids, leading to the formation of arachidonic acid which is converted into two classes of potent lipid mediators: Prostaglandins and leukotrienes.

**Figure 1.4**

*Kinetics of major biochemical events that follow cross-linkage of bound IgE on cultured human basophils*

Along with Phospholipid methylation and Ca\(^{2+}\) increase, there is a transient increase in the activity of membrane-bound adenylate cyclase, with a rapid peak of its reaction product, cyclic adenosine monophosphate (cAMP), reached about 1 minute after cross-linkage of Fc\(\varepsilon\)R. the effects of cAMP are exerted through the activation of cAMP- development protein kinases, which phosphorylate proteins on the granule
membrane, thereby increasing the permeability of the granules to water and Ca$^{2+}$. The increase in cAMP is transient and is followed by a drop in cAMP appears to be necessary for degranulation to proceed. When cAMP levels are increased by certain drugs, the degranulation process is blocked.

1.3.4 The cascade of reactions

The cascade of reactions that take place after the entry of allergens into the body till the development of classic asthma are given below:

Susceptible person exposed to any allergen $\rightarrow$ Activation of lymphocyte, which transform into effector cells like Th2 and plasma cells $\rightarrow$ Th2 lymphocytes and mast cells, secrete interleukin 4 and 5, which promotes Th2 differentiation and switch to IgE synthesis $\rightarrow$ These IgE antibodies get attached to the receptors of the cell membrane of mast cells $\rightarrow$ Now the mast cells are said to be sensitized to that particular allergen $\rightarrow$ When the same allergen settle over the IgE antibodies cross linking it causes transduction of signal in the mast cell $\rightarrow$ Release of chemical mediator like histamine which binds with H1 receptors of target cells $\rightarrow$ causing contraction of bronchial smooth muscles, increased capillary and venule permeability, increased mucus secretion by goblet cells of the alveoli $\rightarrow$ causes asthma symptoms like wheezing, itching, sneezing, coughing etc.
1.4 Various tests available to diagnose Asthma

The following tests are usually done to diagnose bronchial asthma and allergic rhinitis and also to rule out other common conditions which may also cause chronic cough and sometimes ‘wheeze’ like pulmonary tuberculosis, bronchiectasis, chronic obstructive pulmonary disease, eosinophilia, etc.

1.4.1 Blood Test

By doing routine blood analysis, like total count, differential count and haemoglobin, we can diagnose whether the patient is having anaemia which may be the cause for dyspnoea on exertion in which may mimic wheezing in bronchial asthma. Raised eosinophil count may indicate that the person is having either allergic diseases or worm infestations like hook worm (Necator americanus) or round worms (Ascaris lumbricoides) or both.

Mantoux test, Erythrocyte Sedimentation rate test and X–ray chest will be useful in diagnosing ‘pulmonary tuberculosis which may also cause wheezing and ‘chronic cough’.
Immunoglobulin ‘E’ level may be increased in systemic lupus erythematosis, psoriasis, etc., apart from bronchial asthma and allergic rhinitis. But in systemic lupus erythematosis (SLE) there will be butterfly shaped eruption in the face, on both sides of nose and in psoriasis, there will be pinkish violet patches of scaly lesions all over the body.

The methods and procedures of various tests are discussed below.

1.4.1.1 Total count, Differential count

A Levy counting chamber with Improved Neubauer ruling is used for counting white blood cells. The most commonly used diluting fluid for WBC is dilute athec acid. The cells in the four large corner squares are counted using the low power (10x) objective with 10x eye piece. Calculation WBC/µl = (N*20*10)/4=50*N

The differential white blood cell count is done with a well stained peripheral blood-smear. The number of each type of white blood cell is then expressed as the percentage of the total number of cells counted. Usually hundred cells are counted in a representative area of the smear.

1.4.1.2 Erythrocyte sedimentation rate

The Erythrocyte sedimentation rate can be done either by Wintrobe ESR method or Westergren ESR method. 1.6ml of blood and 0.4ml of saline sample is used. The readings are taken at 30minutes and 60minutes. The reading from the pipette in mm which the top of the RBC column has fallen is the ESR reading. The ESR readings are influenced by the physiological factors like plasma factor, the number of cells, age, sex and pregnancy. The following laboratory factors may also influence the ESR reading 1) temperature, 2) time, 3) anti coagulants, 4) length of ESR tubes, 5) inclination of the tube, 6) movement or agitation of the tube. The ESR will be raised in patients having tuberculosis and rheumatic fever.

1.4.1.3 Absolute Eosinophil count

Occasionally, it is desired to directly quantitate the number of eosinophils present in the blood, even though it is also possible to get an approximate figure by multiplying the total WBC count by the percentage of eosinophils in the differential
WBC count. This is especially true in patients with filariasis, tropical eosinophilia and eosinophilic lung.

The normal eosinophil count – will be 40 to 440 cells/cu.mm. The eosinophil count will be more in allergic rhinitis and bronchial asthma. The eosinophil count will also be increased in worm infestation.

1.4.1.4 Haemoglobin estimation
1.4.1.4.1 Clinical significance

Haemoglobin (Hb) has the major function of supplying Oxygen (O₂) to the tissue cells. Hb estimation is one of the commonest screening tests for the diagnosis of anaemia. Decreased levels of haemoglobin concentrations are observed in all varieties of anaemia, resulting from haemorrhage or from deficiency of iron, Vitamin B₁₂ or Folic acid. Increased levels of haemoglobin concentration is observed in polycythaemia Vera, congenital cyanotic heart disease and in haemo concentration due to various clinical causes like heat stroke and dehydration.

The international committee for Standardization in Haematology (ICSH) recommends the Cyanmethemoglobin (CMG) method as a standard method for the estimation of Haemoglobin.

1.4.1.4.2 Estimation of haemoglobin

The haemoglobin is estimated based on the oxidation of haemoglobin and its derivatives (except sulph-haemoglobin) to methaemoglobin in the presence of alkaline potassium ferricyanide. Methaemoglobin reacts with potassium ferricyanide to form cyanmethemoglobin, which has maximum absorption at 540 nm. The colour intensity measured at 540 nm is proportional to the total haemoglobin concentration.

The normal expected value of haemoglobin for male and female were given in appendix

1.4.2 Mantoux test

This test is useful to know whether person has contracted tuberculosis infection or not.
1.4.2.1 Materials
   5 Tu and 10 Tu solution; soap and water, cotton, Insulin Syringe.

1.4.2.2 Mantoux test and its significance
   A standard dose of 5 tuberculin units (0.1 ml solution) is taken in a new 1 cc insulin syringe. The left forearm is cleansed with soap solution. After the area becomes dry, 0.1 ml of tuberculin solution is injected intradermally. Readings are taken 48 hours to 72 hours later.

   The reaction is read by measuring the diameter of induration (palpable, raised, hardened area) across the forearm in millimetres. If there is no induration, the result should be recorded as ‘0 mm’. In children below 10 years, a reading of ‘5 mm’ and below and in adults a reading of ‘10 mm’ and below will be insignificant.

1.4.3 Total Immunoglobulin ‘E’ estimation
1.4.3.1 Principle of Assay
   The MAGIWELL IgE (MERCK) quantitative was a solid phase enzyme linked immunosorbent assay ((ELISA). The wells were coated with anti-IgE antibodies. The samples, standards and controls were included in the wells with enzyme conjugate which was another antibody directed towards a different region of IgE molecules and chemically conjugated with horse radish peroxidase. Unbound enzyme conjugate was washed off and the amount of bound peroxidase was proportional to the concentration of the IgE present in the samples, Standards and controls upon addition of the substrate and chromogen, the intensity of colour developed was proportional to the concentration on IgE in the serum.

   The expected normal values in various age groups are given in the appendix.

1.4.4 Estimation of Specific Immunoglobulin E concentration in serum against food allergens by Euroline Test kit
1.4.4.1 Principle
   The Euroline test kit provides a semi-quantitative in vitro assay for human IgE antibodies to food allergens in serum or plasma. The test kit contains test strips coated
with parallel lines of 21 different allergen extracts. The test strips are first moistened and then incubated with first reaction step with patient serum. If samples are positive, specific antibodies of class IgE will bind to the allergens. To detect the bound antibodies, a second incubation is carried out using an enzyme-labelled monoclonal antihuman IgE (enzyme conjugate) catalysing a colour reaction.

The test strip includes the following allergens.

**Figure 1.6**

**Test strip**

![Test strip image](image)

### 1.4.4.2 Interpretation of the results

In order to evaluate the signals, the band positions and intensity of staining were taken into consideration. By comparing the incubated test strips with the pointed evaluation strip, the allergens against which IgE antibodies were raise can be identified. The signals can be divided into 4 classes which correspond to the bands.
Table 1.3
Classification of signals which correspond to the bands

<table>
<thead>
<tr>
<th>Class</th>
<th>Results</th>
<th>Characteristics</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>Negative No clinical significance</td>
<td>No band</td>
</tr>
<tr>
<td>+</td>
<td>Low allergens in specific IgE concentration, partial clinical significance</td>
<td>Weak band signal</td>
</tr>
<tr>
<td>++</td>
<td>Moderate Allergens in Specific IgE concentration, often with clinical symptoms</td>
<td>Clear band signal</td>
</tr>
<tr>
<td>+++</td>
<td>High allergens in specific IgE concentration, clinical symptoms in most cases</td>
<td>Intense band signal</td>
</tr>
</tbody>
</table>

In the digital evaluation system ‘EURO Line Scan’ the intensity of the bands is calculated in EAST classes of 0 – 6. EAST is the observation for Enzyme – Allergo – Sorbent Test and with respect to the concentration grades identical to the well-known RAST system – (Radio – Allergo – Sorbent Test) used in allergy diagnostics.

The number denotes the classes of concentration and the classes are divided into the following concentrations:

Table 1.4
EAST (Enzyme – Allergo – Sorbent Test) classes of 0-6

<table>
<thead>
<tr>
<th>Class</th>
<th>Concentration (Ku/6)</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>&lt; 0.35 Ku/L</td>
<td>No specific antibodies detected</td>
</tr>
<tr>
<td>1</td>
<td>0.35 Ku/L – 0.7 Ku/L</td>
<td>Very low antibody titre frequently no clinical symptoms where sensitization is present</td>
</tr>
<tr>
<td>2</td>
<td>0.7 Ku/L – 3.5 Ku/L</td>
<td>Low antibody titre, existing sensitization, frequently with clinical symptoms in the upper range of class</td>
</tr>
<tr>
<td>3</td>
<td>3.5 Ku/L – 17.5 Ku/L</td>
<td>Significant antibody titre, clinical symptoms usually present</td>
</tr>
<tr>
<td>4</td>
<td>17.5 Ku/L – 50 Ku/L</td>
<td>High antibody titre, almost always with clinical systems</td>
</tr>
<tr>
<td>5</td>
<td>50 Ku/L – 100 Ku/L</td>
<td>Very high antibody titre</td>
</tr>
<tr>
<td>6</td>
<td>100 Ku/L</td>
<td>Very high antibody titre</td>
</tr>
</tbody>
</table>
1.4.4.3 Clinical Significance

The most frequently occurring allergy is a type 1 hyper sensitivity reaction, in which specific IgE antibodies are formed. The allergic symptoms occur shortly after contact with the allergen. This type of allergy is called an immediate type reaction.

The allergies can also be caused by ingested food, apart from airborne allergens. The most common foods causing allergic reactions are peanuts, soy, wheat, shellfish, fish, milk, eggs and tree nuts.

A food allergy is an IgE mediated reaction which leads to symptoms within hours of having ingested the food. IgE antibodies directed against inhalation allergens may cause food allergies by cross – reacting with components of plant derived food. For instance, patients suffering from birch pollen allergy may develop allergies to apples, carrots, celery, hazelnuts, potatoes or kiwis. Table shows some of the examples of cross-reactivity between airborne allergens and food allergens.

<table>
<thead>
<tr>
<th>Pollinosis</th>
<th>Associated food allergy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grass</td>
<td>Tomato, Potato, carrot, celery, garlic, onion, wheat, rice, green pea, peanut, apple, peach, orange, watermelon, melon, kiwi</td>
</tr>
<tr>
<td>Birch</td>
<td>Hazelnut, walnut, apple, peas, carrot, celery, potato, orange, kiwi</td>
</tr>
<tr>
<td>Magwort</td>
<td>Celery, carrot, spices, green bean, mustard, hazelnut</td>
</tr>
<tr>
<td>Ragweed</td>
<td>Watermelon, melon, cucumber, banana</td>
</tr>
<tr>
<td>English plantain</td>
<td>Melon</td>
</tr>
<tr>
<td>Latex</td>
<td>Avocado, potato, banana, tomato, chestnut, kiwi</td>
</tr>
</tbody>
</table>

The assay of specific IgE antibodies to food allergens is useful to treat bronchial asthma and allergic rhinitis patients by asking them to avoid allergic food materials during treatment period.

Few test results are given below to show the various food materials which are commonly causing allergy to patients.
1.4.5 Spirometric study in bronchial asthma patients

The important function of respiratory system is to separate and extract oxygen from the ambient air, to deliver it to the tissues of the body and to remove carbon-dioxide (CO₂) produced during the process of metabolism by the tissues and expels it into the atmosphere.

Asthma, a chronic recurrent inflammatory disease of the airways, is usually episodic in nature and reversible, either spontaneously or with treatment in early stages. However, in chronic conditions, airflow limitation occurs as a result of varying degrees of airway hyper responsiveness, airway oedema and bronchial constrictions. Hence the spirometric value reveal the working condition of lungs. Any pathology of alteration in the structure of lung tissue affect spirometric values.

The two lungs contain approximately 300 million alveoli, giving a surface area of more than 70 square meters between the trachea and the alveolar sac. According to E.R. Weibel (1963) a Swiss anatomist, the air passage divides 23 times. The whole trachea- bronchial tree can be divided in to two major zones, depending on their functions namely
• Conducting zone - this extends from the nose and mouth up to the terminal bronchial and the total capacity of the zone is about 150 ml. This is called the ‘dead space’.

• Respiratory zone - this is made up of respiratory bronchioles, alveolar ducts and the alveoli. Its volume is approximately 4 litres. The air in the alveoli is separated from the blood in the pulmonary capillaries by the respiratory membrane, consisting of alveolar wall and capillary wall.

In normal children, the development of lung is similar to increase in stature. In children with asthma, the general development is often retarded and this is an indicator of existing disease condition like asthma. If treatment is effected early, the development of the child can often be kept above 25th or even 50th centiles. So early diagnosis of asthma is important. For early diagnosis, reliable tests are essential.

In adults, after biological development has finished by the mid 20’s in healthy subjects, there is a gradual increase in Residual Volume which results in tidal volume moving towards Total Lung Capacity (TLC) and consequently a gradual reduction in Vital Capacity (VC). Due to the natural process of tissue loss due to ageing, respiratory muscle strength decrees and compliance of the chest wall is reduced. There is a decreases in the static elastic recoil of the lung which increases the work of breathing

**Figure 1.8**

**Evolution of Lung volumes with ageing**

![Graph showing lung volumes with age](image)

TLC – Total Lung Capacity;  
VC – Vital Capacity;  
RV – Residual Volume; (Adapted from Crapo et al., 1992)

Age (years)

To measure the effect of disease on pulmonary function, spirometry is indicated. In case of bronchial asthma, the ‘hall mark’ for diagnosis is establishing reversible or variable airway obstruction. In bronchial asthma, there is wide spread narrowing of the
bronchial airways which changes in severity over short period of time, either spontaneously or under treatment.

However, irreversible air flow obstruction develops in some patients. More over accelerated loss of pulmonary function over time has been reported in certain prospective studies in asthma patients. (Lange et al., 1998, Peat et al., 1987, Covar et al., 2004). Many asthmatic patients spirometric value show an accelerated and progressive loss of lung function over time. In one study by Lange et al., (1998), it was proved that there was greater decline in forced expiratory volume in 1 second (FEV₁) in asthmatic patients in comparison with healthy subjects. The accelerated decline in lung function does not occur in all patients. So many risk factors such as young age, height of the patient, duration of disease, smoking, gender, total immunoglobulin E level have been identified which affect FEV₁, FEV₁% spirometric values of asthmatic patients of different age groups.

1.4.5.1 Assessment of Forced Vital Capacity

The simple FVC (Forced Vital Capacity) manoeuvre is performed using Helios 702 electronic spirometer. It is a pre calibrated and computerised spirometer connected to a laptop computer and to Xerox copying instrument. As per the guidelines of American Thoracic society (ATS) statement on standardization of spirometry – 1994 update, the spirometry is recorded in sitting position. The weight and height of the patient are taken. Patient’s age, sex, whether the person is a smoker or ex-smoker are noted down. Then the test is done as follows:

To start with, the patient is instructed to breathe in fully. The patient is advised to bite the mouth piece lightly and lips are closed around the mouth piece so that air from the lungs will be expelled only through the mouth piece to the spirometer. Patient is instructed to blast air out as fast and long as he can until the lungs are completely empty. The exhalation is continued for at least 6 seconds. Immediately after full exhalation, the patient is instructed to breathe in again as forcibly and fully as possible

When airway obstruction is demonstrated, in order to know whether it is partly or completely reversible, bronchodilator test is done. This is performed by repeating the expiratory manoeuvre after administration of fast acting bronchodilator – 200 mg of
metered dose of salbutamol inhaler. A 15% improvement in all measurements is accepted as significant reversibility which is normally observed in bronchial asthma.

The following spirometer parameters are recorded for analysis:

1. Forced Vital capacity (FVC)
2. Forced Expiratory Volume (FVC)
3. FEV$_1$ in percentage.

Figure 1.9
Photo of the Spirometry procedure

The forced expiratory volume in 1 second increases up to the age of 10 years. It increases steadily in childhood. At the age of 16, the maximum capacity is reached. This maximum capacity will be there up to the age of 35 when it begins to decline (Rijcken, 1991). The major determinants of Forced Expiratory Volume (FEV) are age, male gender (Covan et al., 2004) duration of the disease (Lee et al., 2007), more prominent eosinophilic airway inflammation, asthma exacerbations (Bai et al., 2007) and smoking (Lee et al., 2007).

A study has been conducted to determine the effect of height of patients, ageing, duration of suffering, smoking and total immunoglobulin E level upon the spirometric values FEV$_1$ and FEV$_1$% of asthmatic patients of different age groups. Eighty three patients of different age groups, of both sexes, coming from the urban and the rural areas and suffering from bronchial asthma of variable duration of years were selected.
The patients were divided into six groups. All patients were thoroughly examined and necessary entries were made in the case sheets. The total immunoglobulin E level was estimated by ELISA method. It was used as an aid to diagnose atopy in children and adults.

The spirometric values FEV\textsubscript{1} and FEV\textsubscript{1}\% were reduced before the administration of bronchodilator drug in all patients and increased after the administration of the same. These findings were thus in line with the perception that asthma is characterized by the presence of reversible air flow obstruction. The marked decline in FEV\textsubscript{1} and FEV\textsubscript{1}\% values in patients suffering from asthma for a long duration maybe due to at least three mutually independent mechanisms by which one can reach a low level of FEV\textsubscript{1} in later adult life. They are reduced growth, premature and accelerated decline in lung function.

### Table 1.6

**Forced Expiratory Volume in 1 second and FEV\textsubscript{1} in percentage (FEV\textsubscript{1}\% ) pre and post bronchodilator means values compared with predicted normal values**

<table>
<thead>
<tr>
<th>S. No</th>
<th>Age groups</th>
<th>No. of patients</th>
<th>Mean±SEM Pre-Broncho</th>
<th>Mean±SEM Post-Broncho</th>
<th>Predicted Normal value, FEV\textsubscript{1}(L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>10 years and below</td>
<td>10</td>
<td>1.1±0.1</td>
<td>1.2±0.1</td>
<td>73.9±4.4 78.9±4.7 1.5</td>
</tr>
<tr>
<td>2</td>
<td>11 to 20 years</td>
<td>15</td>
<td>1.9±0.12</td>
<td>2±0.11</td>
<td>85.7±3.3 93±3.5 2.2</td>
</tr>
<tr>
<td>3</td>
<td>21 to 30 years</td>
<td>13</td>
<td>2±0.20</td>
<td>2.3±0.18</td>
<td>73.5±5.8 82.6±3.4 2.8</td>
</tr>
<tr>
<td>4</td>
<td>31 to 40 years</td>
<td>16</td>
<td>1.7±0.15</td>
<td>1.8±0.14</td>
<td>67.8±6.3 72.9±6.1 2.6</td>
</tr>
<tr>
<td>5</td>
<td>41 to 50 years</td>
<td>13</td>
<td>1.5±0.15</td>
<td>1.6±0.16</td>
<td>63.5±5.2 67.6±5.2 2.3</td>
</tr>
<tr>
<td>6</td>
<td>51 to 60 years</td>
<td>15</td>
<td>1.4±0.15</td>
<td>1.5±0.15</td>
<td>60.7±5.35 65.9±5.22 2.2</td>
</tr>
</tbody>
</table>

**p<0.005, * p<0.05, SEM - Standard Error of Mean, FEV1 (L) - Liters of Air**

In the present study, there was a definite accelerated decline in FEV1 and FEV1\% values of lung function tests. Thompson and Spears (2005) have reported that smoking and asthma are associated with poor symptom control and impaired therapeutic responses to anti-asthma drug. But Wrik et al., (1992\_3), in their 10 year follow up of 180
asthmatic patients, reported that there was no relation between the rate of decline in lung function and the number of cigarettes smoked. But, compared with the asthmatic non-smokers, smokers with asthma have worse symptom control (Althuis et al, 1999) and increased mortality rate (Marquette et al., 1992).

**Table 1.7**

Relationship of asthma in adults to decline in FEV₁

<table>
<thead>
<tr>
<th>Author</th>
<th>Subject</th>
<th>Follow - up</th>
<th>FEV₁ decline</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peat et al., (1987)</td>
<td>92 asthmatics could not assess smoking indefinitely</td>
<td>18 Yrs.</td>
<td>Asthma 15 ml &gt; Non – asthma</td>
</tr>
<tr>
<td>Buist and Vollmer (1987)</td>
<td>35 adult asthmatics 2 cohorts</td>
<td>9 -11 Yrs.</td>
<td>Employee's asthma &gt; decline then non - asthma only among non - smokers screening asthma &gt; decline than non-asthmatics only among smokers</td>
</tr>
<tr>
<td>Lange et al., (1998)</td>
<td>1095 asthmatics</td>
<td>15 Yrs.</td>
<td>Asthma 14 ml/Yr &gt; non – smokers</td>
</tr>
<tr>
<td>Fletcher et al., (1976)</td>
<td>17 male smokers with asthma</td>
<td>8 Yrs.</td>
<td>Asthma 22 ml/Yr &gt; non - smokers</td>
</tr>
<tr>
<td>Schechter et al., (1984)</td>
<td>Adult smokers; no adjustment for age, gender of smoking</td>
<td>6 Yrs.</td>
<td>Asthma 18 ml &gt; non – smokers</td>
</tr>
</tbody>
</table>

In the study, there was marked decline in FEV₁ and FEV₁% values in patients above 50 years of age. But Peat et al., (1987) did not find any influence of age on the functional decline over several years in asthma, whereas (Ulrik et al., 1992 b) reported steeper decline in FEV₁ values with ageing.

Therefore, it was clearly evident from the study that asthma patients showed marked decline in all lung functions when compared to the predicted values of that age group, and further the spirometric values of FEV₁ and FEV₁ % were influenced by the height of the patients, ageing, duration of suffering above 30 years of age, and smoking, but the gender and sum total immunoglobulin E level had no influence upon spirometric values of FEV₁ and FEV₁%
From the above study, it is evident that spirometric study is an important investigation to diagnose Bronchial asthma, but similar readings may be obtained in patients with chronic bronchitis with emphysema (COPD) without allergic component. Hence spirometric study may be one of the important diagnostic tool but it is not the diagnostic test for diagnosing bronchial asthma with certainty.

**Figure 1.10**

**Spirometry. Normal Study**
In the study conducted at Asthma Research Centre, Pollachi with 196 atopic patients to know the prevalence of food allergens, results showed the main food allergens in various age groups were potato, crab and peanut. Allergy to cow’s milk, egg and milk powder were milk, egg, peanut, chicken, soya and fish. These results suggested that the food allergens vary from place to place and the sound knowledge of food allergens prevalent in that area are essential to diagnose and treat the food allergy and atopic diseases.

1.4.6 X-ray chest

Radiological analysis of the chest will be useful to find out certain causes of wheezing like foreign body in the trachea or any tuberculosis infection or other conditions like Pneumothorax or hydrothorax or massive pleural effusion etc., which may cause difficulty breathing.
1.4.7 Blood pressure

The arterial blood pressure (B.P.) is the pressure column of blood in the arterial system. Clinically, blood pressure is expressed as systolic blood pressure (SBP)/Diastolic blood pressure DBP and the normal average 120/70 mm Hg. The blood pressure is affected by emotions, heredity, meals, sleep etc. Increased blood pressure causes congestive heart failure. The heart failure also will cause dyspnoea or exertion and it is called cardiac asthma. In order to differentiate cardiac asthma from respiratory asthma it is essential to record blood pressure of all wheezing patients.

1.4.8 Electrocardiogram (ECG)

Body is a volume conductor (i.e.) body fluid are good conductor of electricity because it contains large quantities of electrolytes; Therefore electrical changes occurring in the heart with each heart beat are conducted all over the body and can be picked up from the body surface. The record of these electrical fluctuations during cardiac cycle is called electro cardiogram. The following types of abnormalities can be diagnosed using E.C.G.

- Heart block
- New rhythm centre.
- Myocardial infarction.

Effects due to change in the ionic composition of blood as seen in decrease in Na ion concentration in ECF is associated with low voltage ECG, in hyper kalemia – increase in K+ in ECF produces QRS complex prolonged and abnormal in appearance; T wave is tall and peaked due to altered polarisation of myocardial cells; in hypo kalemia – decrease K+ ion in ECF, produces PR interval prolongation and ST segmental depression. So ECG is very useful in differentiating dyspnoea due to myocardial infarction ir congestive cardiac failure or bronchial asthma.
1.5 Clinical features

Wheezy episode may occur at any age in any season with any trigger factors. It may be a single episode or recurrent episodes, preceded by stuffy nose with uneasiness marked by running nose leading to irritation of the child. Frequent rubbing of the nose or noisy breathing indicates partial or complete nasal obstruction. Sneezing, conjunctival congestion and watering from eye may be present, if the onset is allergic in nature.

1. Wheeze: It may vary from person to person, season to season, from time to time, depending on the site and type underlying lesion and other trigger factors. Children with intense wheeze may present with air hunger, inability to drink, to suck and swallow and audible wheeze during expiration, in addition to all components of dyspnoea. Clinically there will be subcostal, lower intercostal, upper intercostal, and suprasternal retraction, alae nasi flaring, retrosternal retraction.

2. Increased respiratory rate and heart rate. In wheezing, essential underlying pathology is the defect in ventilation, whereas in pneumonia the defect is in perfusion and diffusion.

3. A child with wheezing, presenting with drowsiness and low blood pressure is an indicator of poor prognosis – It indicates anaemia leading to respiratory failure and at times circulatory failure.

4. Hyperthermia is not a common feature in children with wheezing. However the presence of hyperthermia may indicate infection. Prompt measures to bring down the temperature along with effective antimicrobial therapy, O₂ supplementation and bronchodilator should be instituted.

5. Electrolyte imbalance and associated

6. Congestive cardiac failure should be borne in mind while managing these children.
Wheezing episode – course and outcome

Uneasiness or itching in the nose or throat } → Sneezing → Running nose

Nasal stuffiness
Nasal block } → difficulty breathing → Noisy breathing
Throat infection → cough → Bronchial congestion
Chest tightness → difficulty expiration → No chest finding
Chest heaviness → expiratory difficulty → Auscultatory wheeze
Work of breathing → Audible wheeze

(sub costal, lower intercostal
recession, Suprasternal, → Restlessness → Airway hunger
upper intercostal recession,
ster mastoid prominence,
Alae nasi flare, retrosternal retraction) → Hypoxia

Course and outcome of wheezing tendency
1. In younger children, less than 3 years
   a) 90% outgrow wheezing in immediate future.
   b) 10% continue to wheeze.
   c) Overall 40% to 50% become asthmatic later.
2. In older children more than 3 years
   a) 80% outgrow wheezing in immediate future.
   b) 20% continue to wheeze.
   c) Majority become asthmatic later.
1.5.1 Early identification of potential wheezers

1. Excessive sweating, more in the scalp region.
2. Cold extremities.
3. Noisy breathing.

1.5.2 Associated problems observed in wheezy children.

1. Convulsions.
2. Urinary tract infection (UTI) in wheezy children 15% to 16% of patients are with U.T.I. These children may present with intractable wheeze or frequent wheezy episodes. Treatment of urinary tract infection along with bronchodilators in these wheezy children has resulted in relief of wheezing for a prolonged period.
3. Infestations: Ascarial infestation, hook worm infestation – in 60% of cases in asthma clinic, Institute of Child Health, Egmore, Chennai and in 15% with giardial infection.

1.5.3 Complications of wheezing

1. Status asthmaticus.
2. Acid base imbalance
3. Dehydration, exhaustion
4. Transient congestive cardiac failure
5. Segmental atelectasis
6. Anoxic convulsion
7. Hypoxaemia
8. Pneumothorax.
10. Acute respiratory failure

1.6 Various treatments available for Bronchial Asthma and Allergic Rhinitis

Asthma is characterised by episodic, reversible bronchospasm, wheezing, cough and dyspnoea, associated with endobronchial inflammation and airway hyper reactivity.
Abnormalities of airway physiology and immuno pathology in asthma have been thought of as reversible, either spontaneous or as a result of treatment. This assumption has recently undergone re-evaluation. It is found that air flow obstruction is not completely reversible despite clinical remission and that asthmatics have a greater rate of decline in lung function than non-asthmatics.

A number of histopathological abnormalities have been described in asthma like thickening of the sub epithelial collagen layer lamina reticularis, and deposition of submucosal scar type collagen. The functional consequences of these abnormalities and their role in the natural history of asthma remain unknown, although fixed airflow and persisting BHR maybe attributable to fibrotic and other structural changes of the airway wall. In normal airway there are several readily distinguishable layers by light microscopy. They are

a) The surface epithelium
b) Basement membrane,
c) Lamina propria
d) Smooth muscle
e) Submucosa
f) Cartilage
g) Adventitia.

The major components, collagen and elastin are the primary determinants of the physical properties if the bronchioles. Larger airways are additionally dependant on smooth muscle, vasendaity and oedema in same circumstances for rigid support.

Lung collagens are continuously produced and degraded throughout life, although synthesis rate decrease with age. The balance between synthesis and degradation of extra cellular matrix is essential to maintain tissue integrity following injury is reflected by cytokines derived from cells present in chronically inflamed tissues. Biopsies taken from asthmatic airways have demonstrated altered levels of matrix metalloproteins (TIMPs) compared with normal controls as well as increased levels of TIMPs in broncho alveolar lavage fluid. These imbalances could lead to further tissue damage, accumulation of matrix components and fibrosis observed in patients with chronic asthma.
Airway inflammation is a fundamental component of asthma and is characterised by epithelial destruction, muscular hypertrophy, apparent thickening of the basement membrane and an inflammatory infiltrate mainly consisting of eosinophils, macrophages, activated T-lymphocytes and mast cells. The continuous process of injury and healing is associated with specific changes associated with airway fibrosis and remodelling. The following growth factors are known regulators of fibroblasts proliferation and collagen synthesis:

a) Platelets Derived Growth Factor(PDGF)
b) Transforming Growth Factor-β(TGF-β),
c) Fibroblast Growth Factor(FGF),
d) Interlukins-1(IL-1)
e) Tissue Necrosis Factor-α(TNF-α)

1.6.1 Current Treatment Strategies

Asthma is a chronic inflammatory disorder. Hence the major goal of treatment is to reverse or present airway inflammation thus improving symptoms and presenting decline in lung function. However, despite adequate therapy that controls symptoms it is clear that there can be ongoing airway inflammation and progressive decline in lung function. There is also evidence that structural changes can develop early in the disease, so it is likely early intervention will be needed to prevent this occurring.

1.6.2 Preventive Measures – By avoiding allergen exposure

The role of allergens exposure and sensitization in relation to asthma prevalence and severity have been demonstrated from varied studies in different countries like UK, USA, Australia etc.

Allergen exposure causes sensitizing in susceptible individuals. When exposed to same allergens after sensitization, it causes allergic diseases by release of histamine and other chemical mediators from mast cells. The main and common indoor allergens are home dust mites, cats, dogs, and cockroaches. Peat et al conducted a series of epidemiological studies in Australia and provided strong evidence of the role of exposure to mite allergens in child hood asthma. In region with high mite allergen exposure, more children were skin test positive to mites. Those who were sensitized to
house dust mite were at significant risk of having current asthma and the magnitude of risk increased with increasing exposure. The risk of mite-sensitized children have asthma apparently doubled with every doubling of Der p1 level. A pattern emerges in which sensitized patients have more severe disease, if their exposure to offending allergens is high, than when it is low. The effectiveness of allergen reduction with treatment of asthma was first suggested by studies in which patients were removed from their homes into a low allergen environment. Later measures aimed at a reduction in allergen levels were attempted in patient’s homes.

1.6.3 Allergen avoidance in homes

a) For controlling home dust mites and mite allergens

- Bed and Bedding – the most effective and probably most important avoidance measures is to cover mattress, pillows and duvets with covers that are impermeable to mite allergens.

- Washing – at 55 Degree Celsius- kills mites in the bedding. If it is cold cycle of laundry adding a concentration of 0.3% benzyl benzoate or dilute solutions of essential oils provide alternative method for mite control.

- Carpets, upholstered furniture – exposure to strong sunlight for at least 3 hours kills mites, steam cleaning may also used to kill mites. The following compounds are used a) Acaricides - chemicals that kill mites are called acaricides - (e.g.) benzyl benzoate b) liquid nitrogen-freezing with liquid nitrogen can kill mites. c) Tannic acid - the protein denaturing properties of tannic acid are well recognized and is used for the reduction of indoor allergen levels in the home – dust. d) Vacuum cleaning - intensive vacuum cleaning may remove large amounts of dust from carpets, reducing the size of the allergen reservoir. e) Humidity control - High levels of humidity in the micro habitats are essential for mite population growth and reducing humidity may be an effective control method f) Air filtration and ionizers.

b) Pet allergen avoidance: Complete avoidance of pet allergens is all but impossible as sensitized patients can be exposed to pet allergens not only in homes with pets, but also in homes without pets and in public buildings and public transport.
1.6.4 Measures for reducing cat & dog allergens exposure

a) Removal of cat/dog from the home.
b) If the pet cannot be removed,
   • Keep the pet out of main living areas and bedroom.
   • Have the pet washed twice a week.
   • Replace carpets with linoleum or wood flooring.
   • Fit allergen impermeable bedding covers.
   • Use a vacuum cleaner with an integrated HEPA filter and double thickness bags.

1.6.5 Treatment of bronchial asthma

1) Corticosteroid therapy.
2) Xanthine group of drugs.
3) Other new therapies.
   a) At sensitization level.
   b) Inhibition of Th2 activation during secondary antigen presentation.
   c) Counteracting the effects of Th2 cell activation during secondary antigen presentation.
   d) Th2 cytokine antagonism.
      i) Anti IL-4
      ii) Anti IL-5
   e) Non – Th2 – based anti-inflammatory
      i) Anti chemokines/cell adhesive blockers/anti – TNF-α
      ii) Anti T-Cell approach
      iii) Anti IgE
      iv) Phosphodiesterase inhibitors
      v) Airway remodelling
4) Long acting β2-agonists
5) Anticholinergics
6) Leukotriene modifiers in the treatment of asthma

The goals of first line asthma therapy have been defined by the National Asthma Education and Prevention Programme (NAEPP) and include
a) asthma control with near normal airway function,

b) absence of asthma symptoms

c) maintenance of activity without limitation,

d) prevention of exacerbation and an acceptable tolerability profile.

1.6.5.1 Steroid therapy

With the recognition that asthma is a chronic inflammatory disease of the airways, there is now widespread consensus for the earlier introduction of inhaled corticosteroid (ICS) whole population data suggests that early use of ICS protects against exacerbation leading to hospitalisation. Guidelines from North America recommend incremental steps based on symptoms and baseline lung function prior to the introduction of treatment. The common steroids used as inhaler are beclomethasone, budesonide and fluticasone. Oral steroids are betamethasone, dexamethasone, methylprednisolone, prednisolone, and triamcinalone.

1.6.5.2 Xanthine Group of drugs

Theophylline, etophylline, doxophylline etc, are some of the Xanthine group of drugs. They bear structural and pharmacological similarity to caffeine.

1.6.5.2.1 Medical Uses

The main actions of theophylline, belonging to Xanthine family are

a) Relaxing bronchial smooth muscle

b) Increasing heart muscle contractivity and efficiency, as a positive isotropic

c) Increasing renal blood flow

d) Anti-inflammatory effects

e) Central nervous system stimulatory effect mainly on the medullary respiratory centre.

1.6.5.2.2 Mechanism of action

1. Competitive non-selective phosphodiesterase inhibitor, which raises intracellular cAMP, activates PKA, inhibits TNF-alpha and inhibits leukotriene synthesis and reduces inflammation.
2. Nonselective adenosine receptor antagonist, antagonising A1, A2 and A3 receptors almost equally, which explains many of its cardiac effects.

3. Theophylline has been shown to inhibit TGF-beta-mediated conversion of pulmonary fibroblasts into myo fibroblasts in COPD and asthma via cAMP-PKA pathway and suppresses mRNA, which codes for the protein collagen.

Theophylline, important and frequently used Xanthine derivative drug is metabolised extensively in the liver (upto 70%). It undergoes N-demethylation via cytochrome P450 1A2. Smokers and people with hepatic impairment metabolize it differently. Theophylline is excreted unchanged in the urine (upto 10%) clearance of the drug is increased in these conditions:

Children 1 to 12, teenagers 12 to 16, adult smokers, elderly smokers, cystic fibrosis, hyperthyroidism. Clearance of the drug is decreased in elderly, acute congestive heart failure, cirrhosis, febrile viral illness.

1.6.5.3 Other new therapies

The pathogenesis of asthma can be regarded as a two-step phenomenon: The first step consists of sensitization to an aero-allergen which involves the development of antigen specific TH2 cells. The second step consists of targeting the TH2 driven allergic inflammation to the lower airways. This inflammatory process is orchestrated and regulated by a complete network of controlling mutually interacting cytokines and growth factors, secreted not only from a range of inflammatory cells, but also from structural tissue components, including epithelial cells, fibroblasts, and smooth muscle cells. This later phenomenon causes remodelling of the airway walls, resulting in a number of structural alterations. Novel treatment strategies can interfere at various levels with in this sequence of events

1.6.5.3.1 At sensitization level

Several approaches can be considered to interfere at this level –

1. To induce tolerance by oral administration of high amounts of antigen (mg. Dose), for example after birth by incorporating them in transgene plants or fruits.
2. Another possibility is to facilitate the switch from TH2 to Th1 cell development by giving exogenous IL-12 cytokine and inducing its endogens productions. The effectiveness of this approach has clearly been demonstrated in mice. However, the applicability by this approach in man is faced with few problems which have to be solved.

**Figure 1.12**

*Treatment of Bronchial Asthma (At sensitization level)*

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1.6.5.3.2 **Inflammation of the lower airways**

Current concepts on the pathogenesis of asthma postulate that once sensitized, secondary antigen exposure induces in predisposed individuals a Th2 – driver inflammation in the lower airways. Therefore, limiting TH2 all involvement constitutes a logical therapeutic target. This can be achieved by

1. Inhibition of Th2 – activation during secondary antigen presentation.
2. Counteracting the effect of Th2 cell activation during secondary antigen presentation. It can be anticipated that Th2 activation during secondary antigen
presentation can be counteracted by inducing the endogenous production or by exogenously administering antagonising cytokines such as IL – 12, IFN – γ, or IL – 10. Various experiments with IL – 12 and IFN – γ cytokines proved to be disappointing. The inducers like attenuated mycobacteria or the administration of digodeoxy nucleotides(ODN) which cause endogenous production of these cytokines, when given intratracheally, the protective effects like inhibition of antigen induced eosinophil influx and bronchial hyper responsiveness, last for 6 weeks, opening the prospect of a novel type of immuno therapy.

1.6.5.3.3 Counteracting the effects of Th2 cell activation during secondary antigen presentation

The activation of Th2 can be prevented either by giving exogenous antagonizing cytokines or by promoting endogenous production of cytokines IL-12, IFN-γ or IL-10. It has been proved that IL-12 given during secondary allergen exposure reduces IL-5 production. But the role of IL-12 as a therapeutic option for allergic disorders is not clear.

IL-10, a pleiotropic immunomodulatory cytokines down regulates both Th1 and Th2 driven inflammatory processes. IL-10 has been shown to reduce collagen type-1 synthesis. It also reduces vascular smooth muscle proliferation. Inducing the endogenous production of beneficial cytokines like IL-10, IFN-γ, IL-12 may be another line of treatment. Oligodeoxynucleotides(ODN) a powerful immunomodulator, contracts Th2 stimulation by acting upon monocytes and macrophages and causing release of IL-10, IL-12, IL-18, TNF-α etc., it also downregulates MHC class antigen expression by mononuclear cells.

1.6.5.4 Th2 cytokine antagonism

A third approach to limit Th2 cell development is by antagonising the cytokines produced by Th2 cells. The main cytokines involved in this experiment 1L – 4 and 1L – 5 with human data are now emerging.
1.6.5.4.1 Anti IL -4

Th2 cell development may be limited by antagonizing the cytokines produced by it. The important cytokines that are produced by Th2 cells are IL-4 and IL-5. IL 4 is mainly involved with pathogenesis of allergic reactions. It is demonstrated that inhalation of IL-4 induces airway eosinophilia and in allergic people bronchial hyperresponsiveness. Hence, by controlling or preventing the action of IL-4 can have good effect in asthma treatment. This can be achieved by giving neutralizing anti IL-4 antibodies, thus blocking the interaction with the receptor.

1.6.5.4.2 Anti IL -5

IL- 5 antigenism can be achieved at various levels

- by interfering with IL-5 gene transcription and translation
- by interfering with IL- 5 production
- by interfering with IL- 5 induced signal transduction mechanism
- blocking the interaction between IL- 5 and its receptor on eosinophils
- using monoclonal antibodies against IL- 5 such as SB240563 and SCH55700. These antibodies reduces eosinophil counts but did not influence the allergen induced early and late asthmatic response.

1.6.5.5 Non – Th2 – based anti-inflammatory

Treatments like anti chemokines/cell adhesion blockers/anti-TNF-α new approaches to treat asthma they are discussed below anti T-Cell approaches, anti IgE approach, phosphodiesterase inhibitors.

1.6.5.5.1 Antichemokines/cell adhesive blockers/anti – TNF-α

Expression of adhesion molecules on vascular endothelium and chemokine activity are necessary to recruit inflammatory cells with airway mucosa. Blocking of the CCR3 receptors found to be effective in controlling allergic manifestation in invivo.
animal models. Alternatively, the adhesion molecules (VCAM-1) on endothelium may be blocked. TNF-α regulates expression of adhesion molecules VCAM-1. Increased levels of TNF-α is noted in asthmatic airways. In rheumatoid arthritis humanized TNF-α antibodies, have been used with noticeable improvements, but its effects in asthma has yet to be assessed.

1.6.5.5.2 Anti T-Cell approach

Till date our experience with T- cell suppression is limited. The T- cell activity may be suppressed by administration of anti-CD4 antibodies; but whether it will be safe on long term treatment has to be considered. Another novel but potentially more effective method is to inhibit cytokine production with P38 MAPkinase activity. This kinase family regulates the transcription of several proinflammatory cytokines and chemokines in a number of cells. P38 MAPkinase inhibitors are more effective at reducing cytokine production from Th2 cells than from Th1 cells.

1.6.5.5.3 Anti IgE

Elevated levels of immunoglobulin E level in circulating blood is an important feature of allergic diseases like bronchial asthma, allergic rhinitis. Anti IgE antibodies such as E25 or CGP56901 have been developed. Administration of E25 repeatedly blocks both antigen induced early and late asthmatic response. It also causes increase in sputum eosinophil counts. Anti IgE, it is proved that also reduced the preallergen level of eosinophilia. IgE antibodies E25 was given systematically and repeatedly to patients having moderate to severe allergic asthma. It is noticed that the highest dose improved symptoms significantly and also requirement of steroids has markedly come down.

1.6.5.5.4 Phosphodiesterase inhibitors

The activities of inflammatory cells like eosinophils etc., are suppressed by increased intracellular cAMP level. It causes relaxation of airway smooth muscle cells and inhibits their proliferation. cAMP is degraded by phosphodiesterase enzymes. It is
found out that inhibition of high affinity binding sites of HPDE4 is related to side effects whereas the anti-inflammatory effect is mediated by LPDE4. Hence, any novel compounds that lose affinity for HPDE4 but not for LPDE4 could proved an important step forward.

1.6.5.5 Airway Remodelling

At last a venue for novel therapeutic approach is airway remodelling. It was recently reported that more selective inhibition of matrix metalloproteinases (MMP2 and MMP9) prevents allergen induced eosinophil influx and bronchial hyper responsiveness in a murine model, opening the prospect for a novel form of antiasthma treatment.

1.6.5.6 Long acting β-2 agonists drugs

β-2 agonists relax airway smooth muscle through stimulation of β-2 adrenergic receptors which activate adenylylate cyclase, leading to an increase in intracellular cAMP and a decrease in intracellular calcium (ca++) concentration. Two types of β2- agonists

a) Short acting: feneterol, salbutamol, terbutaline – have a rapid onset of action and relieve symptoms for 3 to 6 hours. Short acting β2 agonists shall be used in patients with normal lung function.

b) Long acting β2 – agonists:Bambuterol, Formoterol, Salmeterol. They can be used to improve symptom control, particularly at night or with exercise induced symptoms and are indicated for use on a long time basis. Salmeterol and formoterol are administered as inhalers.

1.6.5.7 Anti-cholinergic – drugs

Ipratropium, bromide, oxitropiumbromide. Anticholinergics relax airway smooth muscle by blocking vagal reflex bronchoconstriction, and inhibits early phase response. They have a slow onset but prolonged duration of action.
1.6.5.8 Anti-inflammatory agents

1.6.5.8.1 Steroids

1.6.5.8.1.1 Inhaled steroids

Beclomethasone, Budesonide, Flutasone, they are the mainstay of prophylactic therapy in adults and children using a medium or large volume spacer in conjunction with an MDI will enhance lung deposition and reduce oropharyngeal deposition.

1.6.5.8.1.2 Oral Steroids

Betamethasone, dexamethasone, methylprednisolone, prednisolone, tiramcilone, used as short course rescue therapy in the control of asthma exclamations, it is associated with side effects. Main side effect is suppression of the hypothalamic pituitary adrenal axis. Dosage: Prednisolone 1mg/kg body weight.

1.6.5.8.2 Non- Steroidal anti-inflammatory agents

Drugs sodium cromoclycate, nedocromil sodium, are for prophylactic use. They prevent the release of bronchospastic mediators from mast cells and pate phase response and prevent exercise induced asthmatic attacks. Administering is by inhalation.

1.6.5.9 Xanthine derivatives

Include aminophylline, etophyline and theophylline. Long term treatment with sustained release theophylline is effective in controlling symptoms and improving lung function. Sustained release preparations are useful in controlling nocturnal symptoms.

1.6.5.10 Sympathomimetic

Include adrenaline, ephedrine, isoprenaline, orciprenaline. Produce bronchodilation by stimulating B2 adrenegic receptors in bronchial smooth muscle.
1.6.5.11 Leukotriene receptor antagonists

A new class of drugs known as leukotriene modifiers include both cytokine antagonists (such as montelukast, zafirlukast, pranlukast) as well as 5-lipoxygenase inhibitors (zileuton) also represent the first asthma therapies that have evolved from our understanding of the pathophysiology of the disease.

Asthma pathogenesis is characterised by many features which can be attributed to the actions of leukotrienes – bronchoconstriction, hyperresponsiveness, increased microvascular permeability with tissue oedema, hyper-secretion of mucus and eosinophil recruitment. Thus the discovery of medications that selectively inhibit the formation or action of leukotrienes represents a novel rational way to control asthma.

Figure 1.13
Leukotriene biosynthesis, their effects and points of Therapeutic interruption

Leukotrienes are synthesised from arachidonic acid via the action of 5-lipoxygenase activating protein (FLAP) and they play important roles in mediating airway inflammation. Leukotriene modifiers include both 5-LO inhibitors and cysteinyll leukotriene antagonists.
Leukotriene modifiers were tested and found to be effective in a) laboratory induced asthma such as exercise induced asthma, cold induced asthma, aspirin induced asthma and allergen induced asthma, b) asthmatic bronchoconstriction and c) chronic persistent asthma.

1.6.5.11.1 Leukotriene modifiers are safe

The presently available asthma treatments may be complicated by several side effects. For instance, beta agonists may cause tachycardia, palpitations and headaches. Theophylline has a very narrow toxic therapeutic window, interacts with many medications and may cause tremors, nausea and several other ill effects. Systemic corticosteroids have a number of adverse side effects like hyperglycaemia, growth retardation, hypertension, insomnia and oedema, cataracts, thrush adrenal suppression and bone loss. By contrast, leukotriene modifiers continue to have an excellent safety profile and offer the opportunity to minimize dosage and potential risks of many of the above medications.

1.6.6 About Serum Histamine Binding Capacity

At present about 300 million people around the globe and about 15-20 million people in India are suffering from the disease. Although the people in a particular area either urban or rural or industrial area are exposed to the same air pollutants, only 10 to 15% of the population develop allergic reactions (Lancet 1998) and remaining 85 to 90% of the people are not affected. It was first found out by Parrot et al (1953) that the serum of normal individuals possessed a certain amount of histamine binding capacity which were either absent or low in allergic persons.

Parrot et al (1958) tested and reported that normal persons have 25% histaminopexic power, but the allergic individuals do not have or less power of capacity.

It is reported by Karl-Landsteiner in 1920-1930s that when a hapten is conjugated with a carrier like albumin or globulin and injected subcutaneously, the hapten-carrier complex elicits 3 types of antibodies.
a) antibody to hapten which constitutes a major portion
b) antibody to carrier
c) antibody to hapten-carrier conjugate.

Here histamine is a hapten and globulin acts as a carrier. The carrier is a large immunogenic protein. Hapten is a small molecule with 5000 daltons in molecular weight. Although they are allergens, they are not immunogens i.e. they are incapable of inducing a specific immune response.

When the same hapten is coupled with a large immunogenic protein called a carrier, it yields an immunogenic hapten-carrier conjugate. Animals, immunised with such a conjugate produce antibodies, specific for three types of antigenic determinant

1) the hapten determinant.
2) unaltered epitopes on the carrier protein.
3) epitopes formed by regions of both the hapten and the carrier molecule in combination.

By itself, a hapten cannot function as an immunogenic epitope. But when multiple molecules of a single hapten are coupled to a carrier protein, (or even to a non-immunogenic homopolymer), the hapten becomes accessible to the immune system and can function as an immunogen.

Table 1.8

<table>
<thead>
<tr>
<th>Injection with</th>
<th>Antibodies formed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hapten (Histamine)</td>
<td>None</td>
</tr>
<tr>
<td>Protein carrier (Globulin)</td>
<td>Anti-Globulin (minor)</td>
</tr>
<tr>
<td>Hapten-carrier Conjugate (Histamine+Globulin)</td>
<td>Anti-Histamine (major)</td>
</tr>
<tr>
<td></td>
<td>Anti-Globulin + Hapten (minor)</td>
</tr>
</tbody>
</table>

“A hapten-carrier conjugate is the immunogen in this illustration and the hapten is an antigen that is not by itself immunogenic. The immunogen contains multiple copies of the hapten - a small non-immunogenic organic compound, in this case, Histamine linked to a large protein carrier such as Globulin. Immunisation with Histamine alone elicits no anti-histamine antibodies, but immunisation with Histamine+Globulin elicits three types of antibodies. Of these, anti-histamine antibody is predominant, indicating that in this case the hapten is the immunodominant epitope or antigenic determinant, as it often is in such conjugates”.

A Comparison study of Serum Histamine binding capacity in normal and allergic patients
Karl Landsteiner, had done pioneering works in 1920s and 1930s about hapten molecules.

From the above epidemiological and various other studies, it is evident that allergens, like air pollutants, antenatal and postnatal infections, infestations, diet, chemicals etc. are only precipitating or triggering or exacerbating the existing asthmatic conditions and are not causative factors. Because, if the occurrence rate of bronchial asthma is 10 to 15% and it has more or less equal in people living in industrial area or urban or rural area, why the remaining 85% to 90% of the people are not affected, although they are also exposed to the same allergens? Hence the real ‘causative factor’ must be within the allergic person only.

Investigations or Parrot et al 1953 showed that the serum of normal subjects possess 20% to 30% more histamine – binding capacity as compared to allergic patients in whom it was 0 to 5% only.