INTRODUCTION

Cystic fibrosis (also known as CF, mucovoidosis, or mucoviscidosis) is the most common lethal autosomal recessive childhood disorder in Caucasians, occurring in 1/2,500 births (Wilcken et al., 1995 and Massie et al., 2005). In its most common form, CF manifests as progressive lung dysfunction, pancreatic insufficiency, and intestinal disease. Although technically a rare disease, cystic fibrosis is ranked as one of the most widespread life-shortening genetic diseases. It is most common among nations in the Western world; one in twenty-two people of Mediterranean descent is a carrier of one gene for CF, making it the most common genetic disease in these populations. An exception is Finland, where only one in 80 people carry a CF mutation (Hytonen et al., 2001). In the United States, 1 in 4,000 children are born with CF. In 1997, about 1 in 3,300 Caucasian children in the United States was born with cystic fibrosis. In contrast, only 1 in 15,000 African American children suffered from cystic fibrosis, and in Asian Americans the rate was even lower at 1 in 32,000. In Turkish CF patients, 36 mutations accounted for 75% of all CF chromosomes, and 41 CF chromosomes remained unidentified (Kilinc et al., 2002).

The CFTR gene

The gene which harbors mutations responsible for disease was identified in 1989 (Riordan et al., 1989 and Rommens et al., 1989), after which the protein it encodes was determined to function as a chloride channel that indirectly controls sodium transport. Since then, genetic testing has expanded our understanding of the spectrum of disease that CF represents. The large gene named Cystic Fibrosis Transmembrane Conductance Regulator (the CFTR gene), consists of 27 coding exons. Molecular Location on chromosome 7q31.3 (Kerem et al., 1989): base pairs 116,907,252 to 117,095,950, the CFTR gene span 250,000 bases encoding 1,480 amino acids (Zielenski et al., 1991). The CFTR protein has multiple membrane-spanning regions, two nucleotide-binding domains (NBD), and an R domain which contains sites to which phosphate groups can be attached. The encoded gene product, the CFTR glycoprotein, is a cyclic adenosine monophosphate (cAMP)-regulated low voltage chloride channel without preference for direction of ion secretion. All the 1480 amino acids occupies two membrane-spanning domains (MSD) with six Trans membrane (TM) segments each, two nucleotide-binding domains (NBD) and a
regulatory domain (R). CFTR belongs to the adenosine triphosphate (ATP)-binding cassette family, and is mainly expressed in the apical membrane of epithelial cells in exocrine glands (Hanrahan et al., 2004, Amaral, 2005, Ackerman and Clapham, 1997) (Figure 1.1a, 1.1b and 1.2).

Figure 1.1 a: Location of CFTR Gene in chromosome No. 7

Figure 1.1 b: CFTR gene

Figure 1.2 (A & B): Ackerman & Clapham (1997)
CFTR Gene & Gene product

- ABC35
- ABCC7
- cAMP-dependent chloride channel
- CF
- cystic fibrosis transmembrane conductance regulator, ATP-binding cassette (sub-family C, member 7)
- MRP7

Protein Structure & Function:

The CFTR gene encodes a 170 kDa membrane-based protein with an active transport function that regulates chloride transport. As a consequence of chloride transport, the CFTR protein regulates multiple ion channels and cellular processes, most notably the epithelial sodium (Na⁺) channel (also known as ENaC). CFTR is a type of protein classified as an ABC (ATP-binding cassette) transporter or traffic ATPase. These proteins transport molecules such as sugars, peptides, inorganic phosphate, chloride, and metal cations across the cellular membrane. CFTR transports chloride ions (Cl⁻) ions across the membranes of cells in the lungs, liver, pancreas, digestive tract, reproductive tract, and skin. The structure of the complete CFTR protein has not yet been experimentally determined. This is because membrane proteins, such as CFTR, with substantial hydrophobic ("water-fearing") regions are extremely difficult to crystallize, and X-ray crystallography can only be carried out on protein crystals. By comparing the CFTR protein sequence with that of other known ABC transporters, models depicting the structure of CFTR have been proposed. CFTR is made up of five domains: two membrane-spanning domains (MSD1 and MSD2) that form the chloride ion channel, two nucleotide-binding domains (NBD1 and NBD2) that bind and hydrolyze ATP (adenosine triphosphate), and a regulatory (R) domain. ΔF508, the most common CF-causing mutation, occurs in the DNA sequence that codes for the first nucleotide-binding domain (NBD1) (Figure 1.3).
While most ABC transporters consist of four domains (two membrane-spanning and two nucleotide-binding domains), CFTR is the only one known to possess a regulatory domain. Modification of the regulatory domain, either through the addition or removal of chemical phosphate groups, has been shown to regulate the movement of chloride ions across the membrane.

Although there are no structures of the entire CFTR protein in the Protein Data Bank (PDB), an international archive of molecular structure data, a structure for a similar ABC transporter is available from the PDB. Structure of a CFTR-related ABC protein (Msba) caseydk1 in *E. coli* has been studied. The PDB ID for this protein is IJSQ. PDB also contains a structure based on the theoretical model of the first nucleotide-binding domain (NBD1) (Figure 1.4). PDB ID for this entry is 1NBD (Figure above). In addition, PDB contains synthetic peptide structures (25 to 26 amino acids long) of NBD1’s alpha helical region containing the ΔF508 mutation. PDB IDs for structures of these peptides are 1CKW, 1CKX, 1CKY, and 1CKZ.
Cystic Fibrosis in India

Many medical professionals consider Cystic Fibrosis a disease affecting only the Caucasian population. As such, it was considered extremely rare in the Indian subcontinent until a decade ago. However, recent reports suggest that Indian children do indeed have CF; however, CF in India is still being misdiagnosed and misrepresented.

Due to lack of studies, the precise incidence of CF in the Indian population is unknown. The estimated prevalence in migrant populations in the UK and US vary from 1 in 10,000 to 1 in 40,000. There are no large community based studies that give a clear idea about the disease burden in India, and with the lack of this critical data, CF does not get appropriate attention from policy makers. Even if the prevalence of CF in India is 1 in 10,000 births, there may be 3000 children born with cystic fibrosis annually in different parts of India. Therefore, India would hold the largest population of CF patients in the world today (Kabra et al., 1999, 2000).
As a result of the widespread belief that CF does not occur in Indians, the disease is rarely suspected and even if it is suspected the diagnosis is rarely confirmed due to the poor availability of facilities for diagnosis. The median age of diagnosis among Indian Americans is 12 months compared with 6 months among Caucasian American children and reflects a low index of suspicion for the disease even among Indians in western countries (Powers et al., 1996). Recent reports suggest that genetic and clinical profile of Indian children with CF may be different (Kabra et al., 1996 and Spencer et al., 1994). These reports suggest that the diagnosis of CF is delayed in Indian children which may result in severe malnutrition by the time the condition is eventually diagnosed of the bad prognostic indicators for survival (Kabra et al., 1999).

**Mutations in CFTR gene:** Protein is synthesized by a process of transcription of a single stranded DNA to mRNA, which in turn is translated to the final protein. Every amino acid is determined by a codon of 3 sequential nucleotides. Stop codons will terminate the protein synthesis. On the DNA level, there are three mechanisms for mutations; deletions, insertions or substitutions of one or more base pairs, resulting in missense (incorporation of an incorrect amino acid), nonsense (point mutations that convert a codon to a stop codon), frame shift (substitutions or deletions that often cause disruption of the reading frame) and splice site mutations (often at junction of exon-introns). There are today over 1930 known CFTR mutations listed in the CFTR mutation database (www.genet.sickkids.on.ca). The severity of disease in cystic fibrosis varies greatly, generally based on the specific types of CFTR mutations that are present (Tsui et al., 1997). These mutations have been divided into five different classes, based on the fundamental defects that they cause in the CFTR protein (Figure 1.4). Class I and II mutations (Table 1.1) result in no CFTR protein at the cell surface and are present in patients with more severe disease (Tsui et al., 1997). In contrast, Class III, IV, and V mutations (Table 1.1) have diminished activity and can result in milder disease (www.genet.sickkids.on.ca/cftr).
Table 1.1: Classification of CFTR mutations

<table>
<thead>
<tr>
<th>Class</th>
<th>Description</th>
<th>Example</th>
</tr>
</thead>
<tbody>
<tr>
<td>Class I</td>
<td>Defective protein synthesis</td>
<td>No CFTR</td>
</tr>
<tr>
<td>Class II</td>
<td>Defective processing</td>
<td>CFTR degraded</td>
</tr>
<tr>
<td>Class III</td>
<td>Defective regulation</td>
<td>Impaired response to ATP</td>
</tr>
<tr>
<td>Class IV</td>
<td>Transmembrane mutations</td>
<td>Diminished ion flow</td>
</tr>
<tr>
<td>Class V</td>
<td>Intronic splice sites/CFTR pro mutations</td>
<td>Decreased abundance</td>
</tr>
</tbody>
</table>

**Common Disease causing Mutations:**

About 70% of mutations observed in CF patients result from deletion of three base pairs in CFTR's nucleotide sequence. This deletion causes loss of the amino acid phenylalanine located at position 508, which resides in the first nucleotide-binding domain (Figure 1.6), in the protein; therefore, this mutation is referred to as ΔF508 or F508 (Figure 1.5).

![CFTR Sequence](image)

*Figure 1.5: The ΔF508 deletion is the most common cause of cystic fibrosis. The isoleucine (Ile) at amino acid position 507 remains unchanged because both ATC and ATT code for isoleucine.*

Patients with two of these mutations suffer from classic CF symptoms: bronchiectasis, pancreatic insufficiency, male infertility, and hepatic cirrhosis (Figure 1.6). Since CF is an autosomal recessive disorder, disease phenotypes are only observed in individuals with two inherited mutations in the CFTR gene. It is the effect of these two CFTR mutations on the function of the CFTR protein that, ultimately, determines the clinical phenotype seen in patients. CF, however, is a complex disorder and other factors such as modifying genes, environment, and treatment affect disease progression and severity.
Figure 1.6: CFTR gene, protein, and mutations. The CFTR gene spans 250 kilobases and is encoded by 24 exons that transcribe a 1,480-amino-acid protein. The CFTR protein has 12 membrane-spanning regions bisected by a regulatory region (R) and a nucleotide-binding domain (NBD-1). This binding catalyzes hydrolysis of ATP in order to open the chloride channel; it is counter-regulated by another nucleotide-binding region (NBD-2) at the C-terminus that closes the channel. The location and frequency of the five most common mutations are indicated; the frequency is based on the screening of 43,849 CF chromosomes (Mishra et al., 2005 and Scriver et al., 2001) (www.genet.sickkids.on.ca/cftr/resource/Table1.html).

With normal CFTR, once the protein is synthesized, it is transported to the endoplasmic reticulum (ER) and Golgi apparatus for additional processing before being integrated into the cell membrane. When a CFTR protein with the ΔF508 mutation reaches the ER, the quality-control mechanism of this cellular component recognizes that the protein is folded incorrectly and marks the defective protein for degradation. As a result, ΔF508 never reaches the cell membrane (Figure 1.7). People who are homozygous for ΔF508 mutation tend to have the most severe symptoms of cystic fibrosis due to critical loss of chloride ion transport. This upsets the sodium and chloride ion balance needed to maintain the normal, thin mucus layer that is easily removed by cilia lining the lungs and other organs. The sodium and chloride ion imbalance creates a thick, sticky mucus layer that cannot be removed by cilia and traps bacteria, resulting in chronic infections. While the mechanism that leads to lung damage is not fully understood, lung disease is the leading cause of morbidity and mortality among CF patients.
In general, when mutations in CFTR result in a non-functional protein, ENaC (epithelial sodium (Na\(^+\)) channel) activity increases, and sodium transport across the membrane is augmented. In the lungs and intestine, these results in the accelerated uptake of water from the lumen, leaving dehydrated mucous layers. Conversely, in the sweat gland, defective chloride transport impairs sodium uptake in the sweat duct, resulting in elevated NaCl levels in sweat (Figure 1.8). Accordingly, sweat chloride has allowed effective non-invasive diagnosis for decades.

Despite the large number of CFTR mutations that have been identified, a small number of patients have clinical evidence of CF, including a positive sweat-chloride test, but no identifiable CFTR gene defect. For example, in one study of non-classical (mild) CF, 40% of patients did not have any detectable mutations, despite exhaustive analysis (Groman et al., 2002 and 2005). It is not exactly clear what the underlying defect is in these cases; however, factors other than CFTR mutations appear to lead to phenotypes indistinguishable from CF in some patients. It is clear, however, that even with exhaustive searches CF mutations may miss identifying the underlying cause.
Figure 1.8: Cystic-fibrosis disease mechanisms. In the sweat glands, a defective CFTR channel results in excess sodium and chloride concentration in secretions. This abnormality is the biochemical basis for sweat testing. In the lung, impaired chloride transport causes increased sodium transport into the cell. Water follows sodium and the mucus layer to become dehydrated and more viscous, trapping pathogenic bacteria and leading to chronic respiratory illness. In the pancreas, it is proposed that CFTR impairment causes clogging of the acini and inappropriate activation of enzymes leading to pancreatic insufficiency and malabsorption. CFTR protein is red, other channels and transporters are grey (Colombo et al., 2007, Cohn et al., 2005 and Mishra et al., 2005).
Pathophysiology of Cystic-fibrosis

Clinical Aspects: Cystic fibrosis affects the entire body

- Lungs and sinuses
- GI tract, liver and pancreas
- Endocrine system
- Reproductive system

Cystic fibrosis affects epithelial cells in organs where the CFTR protein is found, including lungs, pancreas, intestine, vas deferens, liver, and sweat glands. It is the distribution of the CFTR in these organs that explains much of the multiorgan nature of CF. Defects in CFTR function within these organs results in lung disease, pancreatic insufficiency, multifocal biliary cirrhosis, male infertility, and increased sweat ion loss.

**Lungs:** The most serious complications of CF occur in the lungs, due to abnormal epithelial-cell transport of Cl\(^-\), resulting in altered surface fluid (Chmiel et al., 2003) (Figure 1.9). The airway surface fluid is decreased due to an increased uptake of sodium (and water), resulting in a dehydrated mucous. These changes impede the necessary ciliary clearance of microorganisms and debris in the lungs, promoting obstruction of the airways and infections. These recurrent infections lead to airway impairment and can cause permanent damage to the lungs (Chmiel et al., 2003). CF patients become infected with specific bacteria, such as *Staphylococcus aureus* or *Haemophilus influenza*, early in life. As disease progresses, *Pseudomonas aeruginosa* and *Burkholderia* spp may infiltrate the lung (Parameswaran et al., 2007). Despite current therapies, lung disease in CF patients still worsens over time; milder forms of CF are associated with a later onset of lung disease, which progresses as a slower place (Yiallouros et al., 2007). Lung involvement is responsible for greater than 90% of the mortality in CF patients.
Figure 1.9: Malfunctioning of lungs and pancreas

Sinus illness

*Aspergillus fumigatus* - A common fungus which can lead to worsening lung disease in people with CF. Lung disease results from clogging the airways due to mucosa build-up and resulting inflammation (Figure 1.10). Inflammation and infection cause injury and structural changes to the lungs, leading to a variety of symptoms. In the early stages, incessant coughing, copious phlegm production, and decreased ability to exercise are common. Many of these symptoms occur when bacteria that normally inhabit the thick mucus grow out of control and cause
pneumonia. In later stages of CF, changes in the architecture of the lung further exacerbate chronic difficulties in breathing. Other symptoms include coughing up blood (hemoptysis), changes in the major airways in the lungs (bronchiectasis), high blood pressure in the lung (pulmonary hypertension), heart failure, difficulties getting enough oxygen to the body (hypoxia), and respiratory failure requiring support with breathing masks such as bilevel positive airway pressure machines or ventilators (Rowe et al., 2001). In addition to typical bacterial infections, people with CF more commonly develop other types of lung disease. Among these is allergic bronchopulmonary aspergillosis, in which the body's response to the common fungus *Aspergillus fumigatus* causes worsening of breathing problems. Another is infection with *Mycobacterium avium complex* (MAC), a group of bacteria related to tuberculosis, which can cause further lung damage and does not respond to common antibiotics. Mucus in the paranasal sinuses is equally thick and may also cause blockage of the sinus passages, leading to infection. This may cause facial pain, fever, nasal drainage, and headaches. Individuals with CF may develop overgrowth of the nasal tissue (nasal polyps) due to inflammation from chronic sinus infections. These polyps can block the nasal passages and increase breathing difficulties (Ramsey et al., 1992 and Maldonado et al., 2004).

**Gastrointestinal, liver and pancreatic disease**

**Pancreas:** The exocrine function of the pancreas is responsible for the secretion of enzymes essential for the breakdown of food. In the pancreas, defective CFTR protein causes reduced $\text{HCO}_3^-$ secretion, leading to congestion of acini and inappropriate activation of pancreatic proteases. This process effectively impairs secretion of the pancreatic enzymes necessary for digestion. Approximately 85% of patients with CF have exocrine pancreas insufficiency, which manifests as poor nutrition and increased fat in stool. This results in weight loss, abdominal pain, and flatulence (Baker et al., 2005). Replacement of pancreatic enzymes and careful diet planning can overcome many of these problems.

The thick mucus seen in the lungs has a counterpart in thickened secretions from the pancreas, an organ responsible for providing digestive juices which help break down food. These secretions block the movement of the digestive enzymes into the duodenum and result in irreversible damage to the pancreas, often with painful
inflammation (pancreatitis) (Cohn et al., 1998). The lack of digestive enzymes leads to difficulty absorbing nutrients with their subsequent excretion in the feces, a disorder known as malabsorption (Figures 1.7 and 1.8). Malabsorption leads to malnutrition and poor growth and development because of calorie loss. Individuals with CF also have difficulties absorbing the fat-soluble vitamins A, D, E, and K. In addition to the pancreas problems, people with cystic fibrosis experience more heartburn, intestinal blockage by intussusceptions, and constipation (Malfroot and Dab, 1991). Older individuals with CF may also develop distal intestinal obstruction syndrome when thickened feces cause intestinal blockage (Khoshoo and Udall, 1994).

Prior to prenatal and newborn screening, cystic fibrosis was often diagnosed when a newborn infant failed to pass feces (meconium). Meconium may completely block the intestines and cause serious illness. This condition, called meconium ileus, occurs in 10% of newborns with CF (Eggermont and Boeck, 1991). In addition, protrusion of internal rectal membranes (rectal prolapse) is more common in CF because of increased fecal volume, malnutrition, and increased intraabdominal pressure due to coughing (Kulczycki and Shwachman, 1958).

**Endocrine disease and growth**

The pancreas contains the islets of Langerhans, which are responsible for making insulin, a hormone that helps regulate blood glucose. Damage of the pancreas can lead to loss of the islet cells, leading to a type of diabetes that is unique to those with the disease (Moran et al., 1994). This Cystic Fibrosis Related Diabetes (CFRD) shares characteristics that can be found in Type 1 and Type 2 diabetics and is one of the principal non-pulmonary complications of CF (Alves et al., 2007). Vitamin D is involved in calcium and phosphorus regulation. Poor uptake of vitamin D from the diet because of malabsorption can lead to the bone disease osteoporosis in which weakened bones are more susceptible to fractures (Haworth et al., 1999). In addition, people with CF often develop clubbing of their fingers and toes due to the effects of chronic illness and low oxygen in their tissues.

Poor growth is a hallmark of CF. Children with CF typically do not gain weight or height at the same rate as their peers, and occasionally are not diagnosed
until investigation is initiated for poor growth. The causes of growth failure are multifactorial and include chronic lung infection, poor absorption of nutrients through the gastrointestinal tract, and increased metabolic demand due to chronic illness.

**Liver disease.** While pulmonary and pancreatic disease occurs in 90% of CF patients, liver manifestations occur in no more than one-third of patients. In the hepatic biliary system, CFTR is expressed in cholangiocytes and gall-bladder epithelial cells but not hepatocytes (Cohn *et al.*, 1993). The main role of CFTR within these cells is to regulate the fluid and electrolyte content of bile; its absence or dysfunction results in impaired secretory function, secondary to increased bile viscosity and bile-duct occlusion (Colombo *et al.*, 2007). This stasis results in damage to the hepatocytes and increases pro-inflammatory cytokines, growth factors, and lipid peroxidation. These derangements prompt liver stellate cells to synthesize collagen, leading to fibrosis (Colombo *et al.*, 2007). Liver disease is the most common non-pulmonary cause of death resulting in approximately 2.5% of all CF mortality (CFF, 2003). Thickened secretions also may cause liver problems in patients with CF. Bile secreted by the liver to aid in digestion may block the bile ducts, leading to liver damage. Over time, this can lead to cirrhosis, in which the liver fails to rid the blood of toxins and does not make important proteins such as those responsible for blood clotting (Williams *et al.*, 1992 and Colombo *et al.*, 2006).

**Infertility.** Most males with CF are infertile as a result of azoospermia (complete lack of sperm) secondary to the congenital bilateral absence of the vas deferens (CBAVD) (Popli *et al.*, 2007). In patients with mild disease, infertility may be the first indication that they may have CF. Due to advances in reproductive medicine; spermatozoa can be retrieved in order to overcome the infertility. While 1% to 2% of CBAVD occurs in infertile males without CF, as many as 80% of men with CBAVD have CFTR gene mutations (Khaitov *et al.*, 2005). Infertility affects both men and women. At least 97 percent of men with cystic fibrosis are infertile but are not sterile and can have children with assisted reproductive techniques (McCallum *et al.*, 2000). These men make normal sperm but are missing the tube (vas deferens), which connects the testes to the ejaculatory ducts of the penis (Dodge, 1995). Many men found to have congenital absence of the vas deferens during evaluation for infertility
have a mild, previously undiagnosed form of CF (Augarten, 1994). Some women have fertility difficulties due to thickened cervical mucus or malnutrition. In severe cases, malnutrition disrupts ovulation and causes amenorrhea (Gilljam et al., 2000).

Other organ systems. There are a number of associated morbidities in patients with CF. These manifestations affect the intestine and upper airway, and include sinusitis, nasal polyps, distal ileum obstruction, and meconium ileus. Up to one-fifth of newborns with CF present with meconium ileus, the retention of the meconium after birth. The identification of meconium ileus is nearly pathognomonic of CF, since it occurs so infrequently in patients without CF. Small-bowel obstruction can also occur in older children and adults in patients with severe disease, sometimes requiring surgical intervention to alleviate the obstruction (Chuan, 2001). Most CF patients develop sinus disease (90% to 100%), while 10% to 32% develop abnormal lesions of the nasal mucosa (nasal polyps) (Ramsey et al., 1992). In undiagnosed patients with mild CF, recurring sinus inflammation and/or nasal polyps may prompt the screening for CF. Changes in the CFTR gene also have been associated with chronic inflammation of the tissues that line the sinuses (rhino sinusitis). This condition causes sinus pain and pressure, headache, fever, and nasal congestion or drainage. Other respiratory problems, including several conditions that partially block the airways and interfere with breathing, are also associated with CFTR mutations. These conditions include bronchiectasis, which damages the passages leading from the windpipe to the lungs (the bronchi), and allergic bronchopulmonary aspergillosis, which results from hypersensitivity to a certain type of fungal infection. Additional genetic and environmental factors likely play a part in determining the risk of these complex conditions.

Sign and symptoms: Hallmarks of CF

The hallmarks of cystic fibrosis are salty tasting skin, normal appetite but poor growth and poor weight gain, excess mucus production, frequent chest infections and coughing/shortness of breath. Males can be infertile due to congenital absence of the vas deferens. Often, symptoms of CF appear in infancy and childhood. Meconium ileus is a typical finding in newborn babies with CF. As the child grows older, he or she will have to exercise to release mucus stuck to the alveoli. Cilial epithelial cells in the patient have a mutated protein that instead of creating the right resin that is used
to prevent the alveoli from collapsing, it makes a thicker resin, mucus. This makes the oxygen extraction in the alveoli harder as the molecules must travel through the mucus leading to breathlessness. Since the mucus stays there most of the time bacteria will thrive in it, causing multiple, chest infections whose symptoms are -

- difficulty breathing;
- constant cough which expels thick mucus;
- excessive appetite, with weight loss;
- bowel disturbances;
- skin which tastes salty;
- repeated or prolonged bouts of pneumonia;
- failure to thrive.

**Inheritance of CFTR Gene:**

![Autosomal Recessive Inheritance Diagram](image)

**Figure 1.11: Autosomal Recessive inheritance**
People who have inherited only one copy of the mutated gene have no symptoms of the disease. Two mutated genes must be present for CF to appear. This means that if both parents are CF carriers (they both have one copy each of the mutated gene), their children would only show CF symptoms if they had inherited both faulty copies of the CFTR gene, one from each parent (Figure 1.11). When the CFTR gene is mutated, it either produces a CFTR protein that does not work or as in a large number of cases, there is no CFTR protein produced at all. When there is no CFTR protein presents this is because part of the DNA code in the CFTR gene is missing, making the CFTR protein shorter than normal. The cell’s quality control system destroys the CFTR protein, as it is too short. CF affects different people in different ways and to varying degrees. Although in all cases, they have the same basic problem, an abnormality in the glands that produce or secrete sweat and mucus. Sweat is needed to cool the body; mucus is needed to lubricate the respiratory, digestive, and reproductive systems and preventing them from drying out and from becoming infected.

Objective:

The present work was carried out with the following objectives.

1. Identification of most common \( \Delta F508 \) mutation in cystic fibrosis patients using allele refractory mutation system.

2. Identification and characterization of unknown mutation in CFTR gene from cystic fibrosis patients and their family members.

3. To estimate 5T allele and its frequencies.

4. To establish correlation between genotype and phenotype.