GROWTH DEFICIENCY IN CYSTIC FIBROSIS IS OBSERVABLE AT BIRTH AND PREDICTIVE OF EARLY PULMONARY FUNCTION

by

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Growth Deficiency in Cystic Fibrosis is Observable at Birth and Predictive of Early Pulmonary Function

Abstract

by

REBECCA JOAN NELSON

Previous studies have identified a positive correlation between weight and pulmonary function of patients with cystic fibrosis (CF). As decreased weight in patients with CF has been observed at birth, this study aimed to identify the predictive quality of birth weight on later complications of CF. Results showed that males with CF (n=40) were on average 10.25 oz lighter at birth (p<0.01) and females with CF (n=39) were on average 9.04 oz lighter at birth (p=0.01) than national averages, matched for gender. Birth weight was correlated with FEV₁% at age 6 (p=0.04) but not at ages 10, 15 and 20. Birth weight was not predictive of percent ideal body weight, the age of *Pseudomonas* colonization, or the incidence of cystic fibrosis related diabetes. These findings suggest that birth weight is predictive of early FEV₁%; however, in older patients, other variables contribute to the majority of CF related complications.
Cystic fibrosis (CF) is an autosomal recessive genetic disorder that affects approximately one in every 3,500 live births. The disease leads to the accumulation of a sticky mucus throughout the respiratory tract and pancreas, ultimately leading to respiratory disease and digestive complications.

A major medical complication for patients with CF is their inability to gain weight. Currently, there is an effort to improve the nutritional health of these patients at younger ages, as better nutritional status has been shown to decrease other medical complications and increase pulmonary function (Forrester, Knox, Smyth, & Fogarty, 2012; Konstan et al., 2003; Stephenson et al., 2013; Steinkamp & Wiedemann, 2002; Yen, Quinton, & Borowitz, 2012; Zemel, Jawad, FitzSimmons, & Stallings, 2000). Many studies have investigated the positive correlation between weight and pulmonary function of young patients with CF (Konstan et al., 2003; Steinkamp & Wiedemann, 2002; Yen et al., 2012; Zemel, 2000). While there are no definitive data regarding the ideal weight and age at which weight becomes predictive of pulmonary function, there is a general consensus that weight is positively correlated with pulmonary function. Consequently, one clinical goal for patients with CF is to increase the patient’s weight, starting as early as possible (Konstan et al., 2003; Steinkamp & Wiedemann, 2002; Yen et al., 2012; Zemel et al., 2000). Although most of the research on body weight and pulmonary function in CF has focused on children and adults, decreased body weight is observed in patients with CF as early as birth. Festini et al. (2005) conducted an 11-year cohort study in Italy, which concluded that infants affected with CF were on average 246.2 g (8.7 oz) lighter than unaffected infants. With no consensus about
what age is predictive of future medical complications, in conjunction with the underweight phenotype that is observable from birth, it is logical to examine the possibility that birth weight is associated with later medical complications seen in older children, adolescents, and adults with CF.
CHAPTER 2: QUESTION AND AIMS

Question

Is there a correlation between birth weight and the severity of medical complications associated with cystic fibrosis?

Purpose

The purpose of this research was to examine whether birth weight is predictive of the development of disease phenotypes seen in patients with cystic fibrosis.

Aims

The aims of this study were to:

1. Determine the mean of birth weights of patients with CF treated at the Cystic Fibrosis Center at Rainbow Babies and Children’s Hospital in Cleveland, OH, and compare these birth weights to national averages matched for gender.
   - It was hypothesized that patients with CF will have a significantly lower birth weight than their unaffected, matched control population.

2. Determine if there is a correlation between birth weight and variability of clinical phenotype of patients with CF.
   - It was hypothesized that individuals with higher birth weights will have higher forced expiratory volume for 1-second (FEV$_1$%) scores, higher body mass index (BMI) and fewer days of IV antibiotics at ages 6, 10, 15, 20, and 25 years. It was also hypothesized that individuals with higher birth weights
will be older at the time of their primary *pseudomonas* colonization, and have a lower incidence of cystic fibrosis related diabetes (CFRD).
CHAPTER 3: BACKGROUND

Cystic fibrosis is an autosomal recessive genetic disease that affects approximately one in every 3,500 Caucasian births and an estimated 30,000 people in the United States. When the phenotypic findings of CF were first attributed to a singular disease process in 1938, the median age of survival was six months (Orenstein, 2001). However, over the past century, advances in medical treatment have increased the average lifespan of a patient with CF to 37.8 years (CFF patient registry, 2013). Even with these treatment advances, such as new medication regimes, nutritional guidelines, and medical devices, CF remains the most common life-shortening inherited disease in the Caucasian population.

Genetics of Cystic Fibrosis

Cystic fibrosis is caused by mutations in the cystic fibrosis transmembrane conductance regulator gene (CFTR). This gene is located on chromosome 7q31.2, and is composed of 27 exons spanning approximately 250 kb. Since CFTR’s discovery in 1989, over 1,940 disease-causing mutations have been identified (Fanen, Wohlhunter-Haddad, Hinzpeter, 2014). The most common mutation is a deletion of a phenylalanine at position 508 (ΔF508). Recent reports state that 86.7% of patients with cystic fibrosis in the United States had one or two ΔF508 mutations, and 47% are homozygous for that mutation (CCF Patient Registry, 2013). Still, 8.2% of patients in the United States have at least one unidentifiable mutation (CCF Patient Registry, 2012).
The CFTR gene encodes a chloride channel of the same name, the cystic fibrosis transmembrane conductance regulator. While present in multiple cell types, this protein is highly expressed in the apical membrane of epithelial cells. This channel allows for the transport of chloride ions across the cell membrane. CFTR chloride channels work in tandem with sodium channels in order to maintain the hydration of the epithelial cell secretions. Proper hydration allows the mucus secreted by these cells to move freely. Mutations in the CFTR gene cause a loss of function of the CFTR protein. The exact consequence of losing the CFTR protein is tissue specific. For example, the CFTR channels in the epithelial cells that line the bronchial airways are meant to transport chloride out of the cell. This causes sodium and water to leave the cell as well. The secretion of water hydrates the mucus that lines the apical membrane of the cell. This creates fluid mucus, which is able to flow freely over the epithelial cells. In individuals with cystic fibrosis, the abnormal chloride transport causes abnormal sodium transport and water osmosis. This leads to an electrolyte imbalance across the epithelial cell membrane, which results in decreased hydration of the cell secretions. In the bronchial airways of an individual with cystic fibrosis, the chloride cannot be transported out of the cell. Consequently, sodium and water are also retained in the cell and the mucus on the apical membrane becomes viscous. This thick and sticky mucus accumulates in the lungs, which makes it difficult to breathe and provides an ideal environment for bacteria to multiply (Gibson, Burns, & Ramsey, 2003; Nussbaum et al., 2007).
Not all CFTR mutations affect gene function in the same way. There are currently five classes of mutations:

I. A nonsense mutation prematurely halts synthesis of the CFTR RNA. The gene transcription is flagged for degradation.

II. The mutation causes a problem with protein folding in the endoplasmic reticulum. The abnormal protein is degraded.

III. The CFTR protein is folded and transported to the cell membrane but the channel does not open.

IV. The protein is in the cell membrane and the channel opens, but the channel cannot transport chloride as well as it should.

V. Mutations in the regulatory regions of the gene cause a decrease in amount of CFTR made in the cell.

Figure 1: Different classes of CFTR mutations. Common mutations in these classes include: Class I) R1282X II) ΔF508, N1303K III) G551D IV) R117H, R347P V) 3349 +10KB C->G (Rogan et al. 2011). Image taken from (Gelfond & Borowitz, 2012).
These mutation classifications provide some genotypic-phenotypic correlations. Patients homozygous for class I, II, or III mutations typically present with classic CF phenotypes. Patients that are homozygous for class IV or V mutations often have more mild disease phenotypes: pancreatic sufficiency, less severe pulmonary disease, and increased average lifespan when compared to those with classic CF. Patients who have one class I, II, or III mutation and one class IV or V typically show a phenotype closer to those with class IV or V mutations (Rogan, Stoltz, & Hornick, 2011).

In addition to different phenotypic manifestations, genotype specific treatment options have recently become available. Kalydeco (generic: Ivacaftor) is a drug currently used to treat individuals who are heterozygous or homozygous for the G551D CFTR mutation, a Class III mutation. The drug is known as a CFTR potentiator, meaning that it increases the ability of activated CFTR channels to open and remain open, thus allowing for adequate chloride transport (Ramsey et al., 2011; Van Goor, Yu, Burton & Hoffman, 2013).

Phenotypic Manifestations

Cystic fibrosis is a multisystem disease affecting the respiratory, digestive, and reproductive systems as well as the sweat glands. In the respiratory system, the accumulation of mucus obstructs the patient’s airways and creates an ideal environment for bacterial infection. Both obstruction and infection reduce the lung’s ability to respire at full capacity and ultimately cause pulmonary disease, the first major medical complication in cystic fibrosis (Orenstein, 2001). While CF creates a number of medical problems, the most
common cause of death is due to respiratory failure, which contributes to approximately 90% of all cystic fibrosis related mortalities (Courtney et al., 2007; Orenstein, 2001).

The pancreas is the main digestive organ affected by CF. The mucus that accumulates in the pancreas inhibits the secretion of the pancreatic enzymes responsible for breaking down fats and proteins in the duodenum. Approximately 80% to 90% of patients with CF have pancreatic insufficiency, meaning that these digestive enzymes are not being secreted from the pancreas, resulting in a decrease in absorption of fats and proteins. Along with pancreatic insufficiency, approximately 20% of patients with CF between the ages of 10 and 30 develop cystic fibrosis related diabetes (CFRD). CFRD occurs when the islets of Langerhans are damaged or destroyed, resulting in insulin deficiency. Patients with CFRD have symptoms similar to diabetes unrelated to CF such as polyuria, polydipsia, lethargy, and unexplained weight loss. These digestive complications related to CF contribute to overall growth failure in many patients, including shorter stature and failure to gain and maintain weight (Orenstein, 2001).

**Advances in Treatment of Cystic fibrosis**

Matthews et al. (1964) outlined the four therapies for patients with cystic fibrosis: 1) therapy of the obstructive pulmonary lesion, 2) therapy of the pulmonary infections, 3) therapy of the digestive defect, and 4) therapy of the sweat glands. Present treatment has evolved substantially since 1964, yet the core principles of the therapies remain (Davis, 2006).

*Therapy of the obstructive pulmonary lesion*
This treatment aims to clear the mucus that accumulates in the lungs of patients with CF. In the past, this was achieved by medications and physical therapy. Medication was used to reduce the viscosity of the mucus and open the airways. Physical therapy included deep breathing and coughing exercises such as postural drainage. Postural drainage involves a medical professional or parent rapidly clapping a patient’s back, shoulders, or chest with a cupped hand to loosen mucus in the lungs (Matthews et al., 1964). Today’s therapies for airway clearance are similar but have become more specific to cystic fibrosis and utilize more sophisticated technology. For example, Pulmozyme® is an aerosolized medication, which makes the mucus in the lung more fluid. The medication, composed of recombinant DNA, degrades extracellular DNA that can cause the mucus to become viscous (Fuchs, et al., 1994). Postural drainage is still used today; however other methods of airway clearance have proven to be more efficient and can be performed independently. For example, oscillating positive expiratory pressure devices, such as the Flutter™, are handheld apparati that create vibrations in the lungs when a patient blows into them (Konstan, Stern, & Doershuk, 1994). High-frequency chest wall oscillation, called the Vest or Oscillator, is a vest-like apparatus that rapidly inflates and deflates to create vibrations in the chest wall and subsequently dislodges mucus from the airways (Warwick & Hansen, 1991).

*Therapy of the pulmonary infections*

Prevention and treatment of frequent airway infection is accomplished through the use of antibiotics. Matthews et al. (1964) outlined the importance of frequent monitoring for infection and antibiotic treatment. Initially, patients would have routine sputum cultures
tested for bacterial organisms; if organisms were identified, patients would receive systemic or aerosolized antibiotics such as penicillin. Today, patients still undergo frequent sputum cultures, but they are treated with antibiotics specific to CF. Tobramycin (TOBI®) is a widely used antibiotic to treat endobronchial *Pseudomonas aeruginosa* infections. It can be taken orally or aerosolized (Hodson, Roberts, Butland, Smith, & Batten, 1987; Ramsey et al. 1993). Patients with severe pulmonary exacerbation may require IV antibiotics, which may require hospitalization but can also be administered at home (Harrison & Rucker, 1974).

*Therapy of the digestive defect*

In the past, diet and pancreatic enzyme replacement were suggested to treat patients with pancreatic insufficiency. It was recommended that infants follow a low-fat, high protein diet, and older children were to follow a high protein diet with moderate restriction of fatty foods. In addition, patients were prescribed pills or powders of pancreatic enzymes to replace those not produced by the patient (Matthews et al., 1964). In the late 1980s and early 1990s both diet and pancreatic enzyme recommendations went through significant changes. It was recommended that patients follow a high fat, high calorie diet (Ramsey, Farrell, & Pencharz, 1992), and gastronomy tube feeding was implemented for patients who were malnourished (Steinkamp, von der Hardt, 1994). Finally, enteric-coated enzymes were introduced, which allowed for the oral medication capsule to withstand the highly acidic nature of the stomach (Carroccio et al., 1988). Current nutritional recommendations are discussed below.

*Therapy to treat inflammation*
Davis (2006) suggested that the suppression of inflammation be included in the “Pillars of Therapy.” Ibuprofen has been shown to slow the rate at which FEV\textsubscript{1}% and percent of ideal body weight declines (%IBW) (Konstan, Byard, Hoppel, & Davis, 1995). The long-term use of ibuprofen has also shown to be beneficial in a CFF registry-wide study (Konstan, Schulchter, Xue, & Davis, 2007).

**Growth Deficiency of Patients with CF**

*Growth and Survival*

While the pulmonary and pancreatic complications that arise in patients with CF have been extensively studied, the growth deficiency that many patients encounter has not been well characterized. In fact, until the late 1980s, growth was viewed as a nonentity when addressing a patient’s medical problems. This was revised when a pivotal study examined the differences in survival and growth of patients with CF followed at clinics in Boston and Toronto (Corey, McLaughlin, Williams, & Levinson, 1988). While both clinics treated similar patient populations in similar climates, patients at the Toronto clinic had much better survival and growth parameters compared to the Boston clinic patients.

Between 1972 and 1981, the mean age of survival was 30 years at the Toronto clinic (n=534, 57% male), and 21 years at the Boston clinic (n=499, 58% male). The female patients’ height for age percentile was on average 44 in Toronto and 33 in Boston; the males’ average was 42 in Toronto compared to 33 in Boston. Weight for age had similar differences; females and males in Toronto were on average at the 31\textsuperscript{st} and 43\textsuperscript{rd} percentiles respectively, while in
Boston they were on average at the 29th and 35th percentiles, respectively (Corey, McLaughlin, Williams, & Levinson, 1988).

Interestingly, there was no difference found in pulmonary function, measured in the two populations, suggesting that the differences in survival were not related to lung function, but rather to growth. The authors speculated that the divergence in growth was a result of the variation in nutritional guidelines between the two clinics. In Toronto, physicians had been recommending a high fat, high calorie diet with 20-30 pancreatic enzyme capsules at every meal. In Boston, patients were told to follow a low fat, high calorie diet, with much lower pancreatic enzyme supplementation. The investigators indicated that little research had been done on the long-term effects of nutritional intervention and their impact on survival, and future studies could illuminate this possible correlation (Corey et al., 1988).

This was an incredibly valuable study as it was the first to establish a link between growth and survival. However, one of the most remarkable factors surrounding this research is that the authors indicated that there was no difference between the pulmonary functions of the patients at the two centers. Pulmonary function is typically used as an indicator for survival. Yet, in the Corey et al study (1988), Toronto patients lived on average nine years longer than the patients treated in Boston, even though there was no significant difference in pulmonary function between the two centers (Corey et al., 1988). While this finding does not make sense in light of more recent research that has identified a correlation between growth, pulmonary function, and survival (Liou et al., 2001), it may be explained by the changing treatment for CF. Perhaps more recent respiratory medications
and therapies have resulted in there being a correlation between weight and pulmonary function; patients who weigh more respond better to the medication and consequently have better pulmonary function. This increase in average pulmonary function since the Boston Toronto study (Wang, O’Leary, FitzSimmons, & Khoury 2002) allows for a wider distribution of FEV$_1$% scores, thus creating the opportunity to observe correlations between a medical parameter and pulmonary function. The fact that there was no significant difference between the pulmonary function of the populations in Boston and Toronto limits the generalization of that study to current medical practice. However, this study should be viewed as evidence that even without the recent advances in treatment, both height and weight are correlated with the survival of the patient.

Another limitation of the Boston/Toronto study is that the research methodology was purely observational rather than an experimental nutritional study. The authors did not manipulate any conditions at either center. Therefore, the conclusion that the difference in survival rates was a reflection of each center’s recommended diet cannot be fully supported. Even though there was a difference in suggested diets at each center, the patients may or may not have followed these recommendations. This study did not monitor the diets of these patients and thus there was no way to measure diet adherence other than anthropometric measurements. While this is an influential study on nutrition and survival in CF, it must be acknowledged that there is a distinct lack of evidence that the patients at these two centers were actively following two different diets.

*Nutrition and Pulmonary Function*
As stated above, research by Corey et al. (1988) led to a multitude of studies devoted to finding a correlation between nutritional status and clinical outcomes. Some research has examined the short-term consequences of nutritional status on pulmonary function by measuring nutrition through weight and height analysis of the individual. Pulmonary function is typically measured through forced expiratory volume in one second (FEV$_1$%), or the volume of air the tested individual can exhale in one second. Besides its ability to quantify pulmonary function, FEV$_1$% is a predictor of mortality in adults with CF and is used to monitor the health status of patients with CF (Courtney et al., 2007; Johnson, Butler, Konstan, Morgan, & Wohl, 2003; Schluchter, Konstan, & Davis, 2002; Schluchter, Konstan, Drumm, Yankaskas, & Knowles, 2006). By comparing these two variables, researchers have been able to identify what was not present in the Corey et al. (1988) study, the correlation between growth and pulmonary function at different ages.

Weight-for-age was positively correlated with 5-year survivorship in a study of patients with cystic fibrosis who were 5½ years or older (n=5,820). This study found that the more a patient weighed at the beginning of the study, the higher the probability that they would still be alive five years later (Liou et al., 2001). Such conclusions strengthen the evidence that nutrition is an important factor in the health and survival of a person with cystic fibrosis.

Konstan et al. (2003) also identified the importance of improving weight in young patients with CF. This study measured weight-for-age, height-for-age, percent ideal body weight, and signs of pulmonary distress, in patients at ages three and six (n=931). The growth indexes at age three were not correlated with signs and symptoms of pulmonary
disease at age three; however, the growth indexes at age three were positively correlated with pulmonary function at age six. In addition, pulmonary function was highest for the patients who had a weight-for-age that remained above the 10th percentile from age three to six years, and lowest for those patients who remained below the 10th percentile throughout the study. The authors concluded that the relationship between growth parameters and pulmonary function are not fixed, and that nutritional intervention has the potential to substantially influence pulmonary function (Konstan et al., 2003).

The influence of nutrition on pulmonary function was an incidental finding in a paper by Wang et al. (2002). This retrospective study of patients, ages six to 10 years, was conducted between 1982 and 1990, using data from the Cystic Fibrosis Foundation National Patient Registry (n=3625). The authors examined whether early diagnosis impacted pulmonary prognosis in children. They determined that in patients born after 1987, early diagnosis was important for pulmonary function outcome. Children who had an early asymptomatic diagnosis (made from family history or newborn screening) had a better FEV1% at ages six and 10 when compared to early symptomatic diagnosis, later asymptomatic diagnosis, and later symptomatic diagnosis. In addition, individuals with an early asymptomatic diagnosis prior to 1987 were more likely to be above the 5th percentile for weight between ages six and 10. These correlations were not seen before 1987, which is most likely reflective of the implementation of new early interventions and nutritional treatments that were implemented around 1987, such as new nutritional guidelines, gastronomy tube feedings, and CF specific antibiotics (Hodson et al., 1987; Ramsey et al., 1992; Steinkamp & von der Hardt, 1994).
**Nutrition and Pulmonary Function in Adults**

Nutrition has been shown to be associated with pulmonary function in adults with CF as well as children. Forrester et al. (2012) conducted a cross sectional study of adult patients (18+ years) with CF from the Cystic Fibrosis Registry in the United Kingdom (n=2096). The study found that lean muscle mass (measured by serum creatinine levels) and body mass index (BMI) were both positively and independently correlated with FEV₁% and forced vital capacity (FVC). They concluded that being overweight, as defined by a BMI greater than 25 kg/m², was in fact linked to better lung function.

A similar study conducted by Stephenson et al., (2013) examined the relationship between FEV₁% and BMI in the adult population of CF patients in Canada (n=909). In this study, the authors placed each participant into one of three categories determined by his or her BMI: underweight, adequate weight, and overweight/obese. Comparisons were then made between and within these categories. Similar to the findings in past research, this study concluded that FEV₁% was positively associated with BMI. However, within groups, it was noted that the effects of BMI on pulmonary function were only significant in the underweight and adequate weight populations. A 10% increase in BMI led to a 4% increase in FEV₁% in underweight individuals, a 5% increase in FEV₁% in individuals with adequate weight, and a 2% increase in FEV₁% for overweight and obese individuals (Stephenson et al. 2013).

**Long Term Association Between Nutritional Status and Pulmonary Function**
The majority of studies have examined the short-term benefits of nutritional intervention; however, a recent study conducted by Yen, Quinton, and Borowitz (2012) focused on the long-term outcomes of such an intervention. The study used data from the Cystic Fibrosis Foundation Registry, specifically looking at patients born between 1989 and 1992 (n=3142). The authors compared the early nutritional status of these patients and the clinical outcomes and survival at 18 years or older. At age four, weight and height were found to be predictive of future growth throughout childhood. Weight-for-age percentile (WAP) was positively correlated with lung function (measured by FEV₁%). Patients with a WAP ≥50% at age four (n=677) spent fewer days in the hospital, were less likely to have impaired glucose intolerance, and significantly less likely to have CFRD at age 18 when compared to those individuals with a WAP ≤50%. In addition, WAP at age four was also positively correlated with survival at age 18. By 18 years of age, 91% of the total study population was still alive. Interestingly, 82.6 % of the patients whose WAP at age four were <10% (n=310) were still alive, while 96.6% of the patients whose WAP at age four were ≥50 (n=677) were still living (Yen et al., 2012). This study shows a clear correlation between growth and clinical outcome/survival in CF patients.

As outlined above, numerous studies have examined the effect of a patient’s weight on his or her pulmonary function. These articles have concluded that weight, or weight gain, is positively correlated with pulmonary function. However, there is no clear evidence that greater weight causes better pulmonary function. Indeed it is possible that individuals who have better pulmonary function are able to gain more weight than those with lower
pulmonary function. Walker and Gozal (1998) studied patients with CF who required gastrostomy tube (GT) feedings (n=21). Baseline weight for age and FEV$_1$% were recorded before the GT placement. After two years of GT feeds, the patients were classified as either having a positive change in their weight for age (responders), or a negative change in their weight for age (nonresponders). The patients who had a baseline FEV$_1$% >40 were more likely to be responders. Conversely, patients who had a baseline FEV$_1$% <40% were more likely to be nonresponders. There was no significant difference between the baseline weights or average age of the responders and nonresponders. The authors suggested that patients with higher FEV$_1$% are able to respond to nutritional intervention, whereas patients with lower FEV$_1$% may have experienced such deterioration in their lung function that nutritional intervention was ineffective.

Walker and Gozal (1998) concluded that pulmonary function is predictive of weight gain, however, this conclusion may not be generalizable to all patients with CF, as the study population had low baseline FEV$_1$%. The nonresponders had severe lung disease with an average FEV$_1$% of 23.2 (±3.4%). The responders had moderate lung disease with an average FEV$_1$% of 61.3 (±6.6%). This study, in fact, provides evidence that when patients have moderate or severe lung disease, FEV$_1$% is predictive of the ability to gain weight.

It is likely that weight and pulmonary function are predictors of better health. Change in one affects the change in the other. It is currently unclear if one measure causes a greater effect than the other.

_Nutritional Recommendations_
As previously mentioned, all of these studies have focused on how a patient’s weight is related to his or her pulmonary function. From these studies, suggested nutritional guidelines for patients with CF include increasing the caloric intake to as much as twice the recommended amount for individuals without CF. If a patient is unwilling to gain weight, which can occur as a result of decreased appetite, illness, or peer pressure, it is suggested that they be referred to a nutritionist and/or have a behavioral intervention in order to demonstrate the importance of maintaining this high calorie diet in the patient. In addition, CF centers are instructed to monitor their patients’ growth indices every three months (Borowitz, Baker & Stallings, 2002). Patients 20 years or younger should maintain a BMI above the 50th percentile as dictated by their age and sex-matched cohort. The ideal BMI for male patients 21 years and older is 23 and for females 21 years and older, a BMI of 22 is suggested. These weight recommendations are derived from population studies that identified the best weight for lung function (Stallings, Stark, Robinson, Feranchak, & Quinton, 2008).

Low Birth Weight in Patients with CF

While low weight-for-age has been shown as a cause for concern in children and adults with CF, these are not the only age groups who are considered to be smaller than their unaffected peers. Patients with CF may have lower birth weights when compared to healthy infants (Haeusler, Frisch, Wardhör, & Götz, 1994). A retrospective cohort study in Tuscany, Italy, compared the birth weights and gestational ages of babies affected by cystic fibrosis (n=70) to unaffected babies (n=290,059). Both populations encompassed the vast
majority of babies born in this area, between 1991 and 2002. The mean birth weight of babies with CF was 246.2 g (8.7 oz) lighter than the mean birth weight of unaffected babies. In addition, 14.4% of these newborns with CF were delivered preterm (36 6/7 weeks or less); only 5.52% of newborns without CF were preterm. There was no significant difference in the birth weights of these preterm affected and unaffected neonates; however, there was a significant difference in the birth weights of the affected and unaffected neonates born at term. The authors proposed that the difference in weight between affected and unaffected babies was generated during the final weeks of gestation (Festini et al., 2005). This study suggests that low weight in patients with CF is observable as early as birth.

Although considerable research has been devoted to the correlation between childhood weight percentile and later pulmonary function and/or health of the individual, no attention has been paid to the correlation between birth weight and later medical complications. While many studies have shown that weight and weight gain are influential in predicting or improving the pulmonary prognosis of patients with CF, there is no consensus regarding at what age weight becomes predictive. Since individuals with CF fall into the lower weight percentiles as early as birth, it is only logical to study the effects of birth weight on a patient’s medical phenotypes.

From the previous work that has been done on the correlation between nutritional status and pulmonary function, it was hypothesized that this current study would find that birth weight is predictive of clinical manifestations of cystic fibrosis. A higher birth weight was hypothesized to be associated with better pulmonary function (FEV₁%), higher BMI and
fewer medical complications: later colonization of *Pseudomonas aeruginosa*, lower incidence of CFRD, and fewer nights receiving IV antibiotics.
**SIGNIFICANCE FOR GENETIC COUNSELING**

If data suggest that birth weight is predictive of the clinical manifestations of cystic fibrosis, there will be cause to reevaluate the treatment of these patients. Birth weight may become the earliest known prognostic indicator, from which a patient’s treatment plan can be determined. Clinicians may want to instigate more aggressive treatments earlier on for patients with lower birth weight, as it has been found that more aggressive treatments for patients with mild symptoms improves FEV₁% (Johnson et al., 2003). They may also want to increase the surveillance for CF related medical issues, like CFRD, in patients with lower birth weights if this correlation is supported by the conclusions of this study.

This study could also highlight the need for further research to identify the cause of the low birth weight seen in these babies. If a cause can be identified it is possible that a prenatal treatment could also be found and implemented. If birth weight were correlated with the severity of cystic fibrosis, a prenatal treatment to increase birth weight would be extremely beneficial. As previously mentioned, there are currently multiple drugs that aim to correct the specific genetic mutation or correct the defect caused by the mutation. Ivacaftor, which was discussed earlier, was found to increase weight of patients 6 years and older by an average of 3.1 kg (Ramsey et al., 2011). Ataluren (PTC124), a drug currently in phase 3 of clinical trials, targets Class I mutations and allows the ribosomes to read through nonsense mutations. During a clinical trial for Ataluren, children showed more long lasting effects of the medication than the adults. These effects include: increased expression of active CFTR protein in the apical membrane on cells in the nasal passage, and increased chloride transport (Serment-Gaudelus et al., 2010). Perhaps similar prenatal intervention
could increase birth weight and effectively decrease the clinical severity of the patient’s CF.

If this is the case, wide based carrier screening for cystic fibrosis or prenatal screening for the disease may become standard in order to identify the pregnant women who should be offered this prenatal treatment.

Regardless of the study’s outcome, the information collected will still have importance to genetic counseling. If birth weight is not correlated with clinical manifestation of the disease, then weight gain early in life, as previously recognized, is correlated with a better prognosis (Wang et al., 2002; Konstan et al., 2003; Yen et al., 2013). This finding will reinforce the need for parents, clinicians, and patients to improve their child’s, patient’s, or own weight in order to improve prognosis.
CHAPTER 4: PROJECT DESIGN AND METHODS

Study Design

The purpose of this descriptive retrospective cohort study was to identify the relationship between birth weight and the variability of disease phenotypes of cystic fibrosis. This study included patients with cystic fibrosis cared for at the Cystic Fibrosis Center (CFC) at Rainbow Babies and Children’s Hospital (RBCH) in Cleveland, Ohio. The majority of the medical information for these patients was already recorded in the CFC’s electronic database, PortCF, which collates the patients’ measurements, treatments, and test results during every inpatient and outpatient visit to the CFC. A complete list of the information from PortCF that was used in this study can be found in Appendix A. In addition, a four-question survey was placed in every patient’s chart to obtain necessary information not collected in PortCF at the beginning of this study. The questionnaire (Appendix B), used during a patient’s visit to CFC, prompted the medical professionals working with the patient to ask the patient or his/her parent or guardian about the patient’s birth weight, birth length, and gestational age. Answers to these questions were written on the questionnaire and recorded in PortCF.

Participants

All patients cared for at the CFC were asked to answer the questionnaire. The follow exclusion criteria were used for Aim 1: Determine the range of birth weights of patients cared for at the CFC.

1. Birth weight was not available
2. The patient was pancreatic sufficient
3. The patient was born prior to 37 weeks gestation
4. The patient was a twin

Patients were excluded for prematurity and twin pregnancy to control for additional causes of low birth weight (Alexander, Kogan, Martin, Papiernik, 1998; Olsen, Groveman, Lawson, Clark, Zemel, 2010). Patients were excluded due to pancreatic sufficiency, as these patients tend to have less acute clinical manifestations of cystic fibrosis. Pancreatic sufficiency is more likely to be present in the patients with class IV or V mutations (McKone, Emerson, Edwards, Aitken, 2003). Individuals with these mutations can have some working CFTR protein. Excluding individuals who were pancreatic sufficient made it more likely that participants in the study population did not have functioning CFTR protein.

The following exclusion criteria were used for Aim 2: Determine if there is a correlation between birth weight and variability of clinical phenotype of patients with CF.

1. Birth weight was not available
2. The patient was pancreatic sufficient
3. The patient was not born between 1975 and 2005

Patients who received lung transplants were included in the study up until the age of their transplant. Patients were excluded for pancreatic sufficiency for the same reasons listed above. Patients born before 1975 were excluded to control for the introduction of new treatments for CF. Patients born before 1975 were not exposed to treatments such as aerosolized TOBI® and enteric-coated pancreatic enzymes until they were teenagers or
adults (Carroccio et al., 1988; Ramsey et al., 1993). Therefore, these patients’ early health measurements, such as FEV₁% and age at *Pseudomonas* colonization, are expected to be worse than patients who were taking these medications.

Patients born after 2005 were excluded because they would not have had enough time at the CFC to generate enough data required for analysis.

**Data Collection**

Data for birth weight, birth length, and gestational age were not collected by the CFC prior to the start of this study. To collect this information, a brief questionnaire was placed in each patient’s chart (Appendix B). The questionnaire had three main questions: birth weight, birth length, and gestational age. Since some parents might have only remembered if their child was full term or premature, gestational age was split into two sections: one section that asked if the baby was born at term, and one that inquired about the gestational age. The questionnaire prompted the health professional to ask the patient and/or their parent or guardian all four questions and to fill out any information they received. For identification purposes, there was a section in the top right corner of the questionnaire for the patient’s hospital admission sticker.

Once completed, the questionnaires were collected by the CFC’s Cystic Fibrosis Computer Center and the data were entered into PortCF.
Aim 1: Determine the range of birth weights of the patients with CF treated at the CFC at RBCH in Cleveland, OH, and compare these birth weights to national averages matched for gender.

In order to assess the range of birth weights of this population, birth weights were collected thorough the questionnaire. Control birth weights were obtained through the national birth weight averages published by the Center for Disease Control and Prevention (CDC, 2001). The mean birth weight of newborns with cystic fibrosis was compared to the weight listed as the 50th percentile on the CDC growth chart, matched for gender.

Additional analyses were done to explore factors that influence birth weight, including sex, year of birth, race, genotype, gestational age at birth, pancreatic status, and presence of meconium ileus at birth.

Aim 2: Determine if there is a correlation between birth weight and variability of clinical phenotype of patients with CF.

There were four categories of information that were collected from the PortCF including demographic information, pulmonary measures, gastrointestinal complications, and nutritional status (Appendix A).

To characterize pulmonary function, FEV₁% was examined at five time periods in the patient’s life. For this analysis, a patient’s best measurement during each time period was used for analysis. In order to capture as many patients as possible, each time period included three years: ages 5-7, 9-11, 14-16, 19-21, and 24-26. For simplicity’s sake, these time points will be referred to by the median age: age 6, 10, 15, 20, and 25, respectively.
To characterize nutritional status percent ideal body weight (%IBW) was examined at three time periods during childhood (Ages 6, 10, and 15) and BMI was examined at two
time periods during adulthood (Ages 20 and 25). Similarly to FEV₁%, the patient’s best measurement during each time period was used for analysis. Each time period for
nutritional status was three years long, during the same ages that FEV₁% was examined. Presence or absence of CFRD was also used to characterize nutritional status.

To characterize the pulmonary exacerbation and infection, the number of nights spent at the hospital and at home on IV antibiotics during ages 6, 10, 15, 20, and 25 was collected. These numbers were combined in order to generate a total number of nights spent on IV antibiotics during the year. The age of Pseudomonas colonization was also used to characterize pulmonary exacerbation and infection. At the CFC, patients are classified as colonized after three consecutive clinic visits with cultures positive for Pseudomonas.

Statistical Analysis

Demographic information was summarized using means, standard deviations, and frequencies. One-sample t-tests were used to compare the mean birth weight of individuals with CF and the national average birth weight published by the CDC, matched for gender. Independent sample t-tests were used to compare the birth weight of different demographics, genotypes, and phenotypes, controlled for gender.

Linear regressions were used to compare birth weight with FEV₁%, and with %IBW/BMI at ages 6, 10, 15, 20, and 25.
Data analysis was performed using IBM SPSS Statistics 21 for Mac version. Graphs were created using Microsoft Excel 2008 for Mac version 12.3.6.

**Data Safety and Monitoring**

This study was part of a larger study protocol conducted at the Cystic Fibrosis Center at Rainbow Babies and Children’s Hospital, which was approved by the Institutional Review Board (IRB) at University Hospitals Case Medical Center.
CHAPTER 5: RESULTS

Participants

There are a total of 352 patients cared for at the CFC. During the data collection period between May 10, 2013, and February 21, 2014, 238 patients were seen in the CFC.

Among these patients, 128 surveys were received. In Aim 1, 79 patients were studied (Table 1) and 95 patients were included in Aim 2 (Table 2).

Table 1: Participants excluded from Aim 1

<table>
<thead>
<tr>
<th>Exclusion criteria</th>
<th>Number of participants</th>
</tr>
</thead>
<tbody>
<tr>
<td>Did not provide birth weight</td>
<td>17</td>
</tr>
<tr>
<td>Reported a birth weight ±2 SD from his or her gender’s average birth weight</td>
<td>5</td>
</tr>
<tr>
<td>Pancreatic sufficient</td>
<td>8</td>
</tr>
<tr>
<td>Premature (born &lt;37 weeks gestation)</td>
<td>16</td>
</tr>
<tr>
<td>Pancreatic sufficient and premature</td>
<td>3</td>
</tr>
<tr>
<td>Total number of participants in Aim 1</td>
<td>79</td>
</tr>
</tbody>
</table>

Table 2: Participants excluded from Aim 2

<table>
<thead>
<tr>
<th>Exclusion criteria</th>
<th>Number of participants</th>
</tr>
</thead>
<tbody>
<tr>
<td>Did not provide birth weight</td>
<td>17</td>
</tr>
<tr>
<td>Reported a birth weight 2 SD away from his or her gender’s mean birth weight</td>
<td>5</td>
</tr>
<tr>
<td>Pancreatic sufficient</td>
<td>11</td>
</tr>
<tr>
<td>Born before 1975</td>
<td>4</td>
</tr>
<tr>
<td>Total number of participants in Aim 2</td>
<td>91</td>
</tr>
</tbody>
</table>
Aim 1

Aim 1 was to determine the range of birth weights of the patients with CF and compare these birth weights to national averages, matched for gender. Preliminary statistical analyses were preformed to verify the exclusion criteria: birth weight is correlated to gestational age, pancreatic function, and twin pregnancy. These analyses supported the exclusion criteria (Appendix C).

CF Birth Weight Vs. National Averages

As CF can cause growth deficiency in children and adults, it was hypothesized that patients with CF would have a significantly lower birth weight than the national birth weight averages, matched for gender. One sample t-tests were conducted with data from the CDC serving as the national average for infant birth weight. Figure 2 shows the average birth weight for males with CF at the CFC ($M= 3239.80$ g, $SD=367.83$ g, $n=40$) was significantly lower than the national average of 3530.20 g (7 lbs 12 oz), $t$ (39) = -5.00 $p<0.01$. The birth weight for females with CF at the CFC ($M=3142.94$ g (6 lbs 15 oz), $SD=422.75$ g, $n=39$) was also significantly lower than the national average of 3399.19 g (7 lbs 7.8 oz), $t$(39)=-3.79 $p=0.01$. These results suggest that growth deficiency in cystic fibrosis is observable at birth.
Figure 2: The average birth weight of males (n=40) and females (n=39) with CF compared with national averages.

**Gender**

An independent samples t-test conducted between males and females with CF indicated that there was no significant difference between the birth weight of the two sexes $t(77) = 1.09, p=0.28$ (Figure 3). Consequently, the genders were not separated in all calculations for Aim 1, except for the birth weight of patients with cystic fibrosis compared to national averages.
Figure 3: The average birth weight of males and females with CF, $p=0.28$

**Current Age**

Analysis was done to examine any differences between current age of the patient and birth weight. To test for differences between birth weight and year of birth, a One-Way Analyses of Variance (ANOVA), was used. Participants were grouped by current age: ≤4 years, 5 to 9 years, 10 to 14 years, 15 to 19 years, 20 years, and ≥25 years. It was hypothesized that there would be no difference in average birth weight among the age groups. The analysis showed that there was no effect of year of birth on birth weight, $F(5, 78)=0.414$, $p=0.42$ (Figure 4). These results indicate that there has been no significant change in average birth weight for infants with CF for at least the past 25 years.
Figure 4: The average birth weight of patients with CF grouped by age on December 31, 2013 by ANOVA analysis, $p=0.42$

**Genotype**

In order to test for differences between birth weight and genotype, an independent samples t-test was conducted with birth weight as the dependent variable and genotype (categorized as ΔF508 homozygotes and ΔF508 heterozygotes) as the independent variable. It was hypothesized that ΔF508 heterozygotes would have higher birth weights, as these individuals may have some functioning CFTR protein. Results indicated that there were no statistically significant difference between birth weight of participants who were ΔF508
homozygous (n=48) and those who were ΔF508 heterozygous (n=31), t(77) = -1.18, p=0.24 (Figure 5). Due to the small sample size of participants without at least one ΔF508 mutation, these individuals could not be included in the calculations. These results do not support the hypothesis; ΔF508 heterozygotes do not have significantly higher birth weights than ΔF508 homozygotes.

Figure 5: The average birth weight of patients who are ΔF508 homozygous and patients who are ΔF508 heterozygous, p=0.24
Meconium Ileus

An independent samples t-test was also used to test for a difference in birth weight between those born with (n=28) and without a meconium ileus (n=51). It was hypothesized that there would be no significant difference between the average birth weights of the two groups, and the data supported this hypothesis, $t(77)=-0.396$, $p=0.70$ (Figure 6). The presence of meconium ileus at birth does not influence birth weight.

Figure 6: The average birth weight of patients born with MI and without MI, $p=0.70$
Aim 2

The second aim of this study was to determine if there is a correlation between birth weight and variability of clinical phenotype of patients with CF. A variety of clinical measures were originally chosen to quantify the participants’ clinical phenotype. Pulmonary function was measured by best FEV₁% during ages 6, 10, 15, 20, and 25. Nutritional status was measured by best %IBW at ages 6, 10, and 15, and best BMI for ages 20 and 25. Severity of and susceptibility to pulmonary infection were measured by determining the number of days spent receiving IV antibiotics during ages 6, 10, 15, 20, and 25, as well as the age that Pseudomonas colonization was identified in each patient. Finally, digestive complications of CF were explored by comparing the birth weights of patients with CFRD to patients without CFRD.

These initial variables required modification once data collection was completed. Due to a small sample size (n=5), calculations at age 25 could not be included in these analyses. Additionally, analysis for the number of days spent receiving IV antibiotics was not able to be calculated as these data were not recorded in PortCF until the year 2000.

Pulmonary Function

Preliminary analyses were performed to determine if any additional variables were correlated with pulmonary function. Gender, year of birth, gestational age, and presence of MI at birth were all tested using independent samples t-tests and ANOVA. No correlations were identified; therefore all patients who met inclusion criteria were grouped together for these analyses.
Forced expiratory volume in one-second (FEV₁) measures the amount of air that a person can forcibly exhale in one second. FEV₁% is the percent-predicted FEV₁ based on normal controls matched for age, sex, height, and weight. Therefore, FEV₁% is used to compare the lung function of the patient to normal controls.

Linear regressions were preformed to evaluate the correlation between birth weight and pulmonary function. It was hypothesized that birth weight would be predictive of FEV₁% at ages 6, 10, 15, and 20. For all ages, best FEV₁% during that age was the dependant variable and birth weight was the independent variable.

Birth weight significantly predicted FEV₁% at age 6, \( b=0.01, t(54)=2.104, p=0.04 \). This indicates that for every 100 grams that birth weight increased, FEV₁% increased by 1%. Birth weight also explained a significant proportion of variance in FEV₁% at age 6, \( R^2=0.08, F(1, 54)=4.43, p=0.04 \) (Figure 7). This signifies that 8% of the variance observed in FEV₁% can be attributed to birth weight. These results suggest that birth weight is predictive of pulmonary function at age 6.
Figure 7: The relationship between birth weight and FEV$_1$% at age 6 (n=55), $R^2=0.08$, $p=0.04$

Birth weight trended towards significantly predicting FEV$_1$% at age 10, $b=0.01$, $t(58)=1.98$, $p=0.05$. There was a trend towards birth weight predicting the variability of FEV$_1$% at age 10, $R^2=0.07$, $F(1, 58)=3.94$, $p=0.05$ (Figure 8). These results suggest that birth weight may be predictive of pulmonary function at age 10. However, this cannot be determined with the population studied.
Figure 8: The relationship between birth weight and FEV$_1$% at age 10 (n=59), $R^2=0.07$, $p=0.05$

There was no correlation between FEV$_1$% and birth weight at age 15, $b=-0.001$, $t(44) = -0.013$, $p=0.93$ (Figure 9), or at age 20, $b=0.006$, $t(16) = 0.417$, $p=0.683$ (Figure 10). These results suggest that at ages 15 and 20, additional variables that were not measured in this study are more strongly correlated with FEV$_1$% than birth weight.
Figure 9: The relationship between birth weight and FEV₁% age 15 (n=45), p=0.93
Figure 10: The relationship between birth weight and FEV$_1$% at age 20 (n=17) $p=0.68$

Additional analyses were used to characterize the relationship between birth weight and FEV$_1$%. It was hypothesized that individuals who had a birth weight above the mean birth weight for the CF population would have a higher mean FEV$_1$% than the individuals who had a birth weight lower than the mean birth weight of the CF population, matched for gender.

The patients were separated into two groups: those born below the average birth weight for patients with CF, matched for gender (below average) and those born above the average birth weight for patients with CF, matched for gender (above average). The average birth weights of each group were determined through descriptive statistics. Independent samples t-tests were used to find the differences between the two groups. The results of
this analysis were similar those of the linear regressions; at age 6 patients, patients with an above average birth weight had significantly higher average $FEV_1\%$ than patients with a below average birth weight. There was no significant difference between the average $FEV_1\%$ of the two groups at ages 10, 15, and 20 (Table 3).

Table 3: The differences between $FEV_1\%$ of patients above and below the mean birth weight for CF patients, matched for gender

<table>
<thead>
<tr>
<th>Age</th>
<th>Mean $FEV_1%$ of patients $\geq$50th percentile of birth weight (SD)</th>
<th>Mean $FEV_1%$ of patients &lt;50th percentile of birth weight (SD)</th>
<th>Difference</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>6</td>
<td>105.31% (17.04) n=29</td>
<td>96.13% (14.92) n=26</td>
<td>9.18%</td>
<td>0.04</td>
</tr>
<tr>
<td>10</td>
<td>105.06% (14.20) n=30</td>
<td>98.47% (13.65) n=29</td>
<td>6.59%</td>
<td>0.08</td>
</tr>
<tr>
<td>15</td>
<td>95.69% (15.22) n=25</td>
<td>94.23% (15.28) n=19</td>
<td>1.47%</td>
<td>0.75</td>
</tr>
<tr>
<td>20</td>
<td>77.62% (19.93) n=10</td>
<td>69.58% (11.72) n=6</td>
<td>8.04%</td>
<td>0.39</td>
</tr>
</tbody>
</table>
Figure 11: The average FEV₁% separated by birth weight group

**Percent Ideal Body Weight (%IBW)**

Preliminary analyses were performed to determine if any additional variables were correlated with %IBW. Gender, year of birth, gestational age, and presence of MI at birth were all tested using independent samples t-tests and ANOVA. No correlations were identified for most of the above variables. However, an independent samples t-test identified a difference in %IBW at age 6 of patients born with MI (\(M=70.00\%, SD=19.80\%, n=27\)) and those born without MI (\(M=56.13\%, SD=23.98\%, n=30\)). At age 6, patients born with MI had a mean %IBW 13.86% higher than patients born without MI, \(t(55)=2.37, p=0.02\). There was no significant difference between the %IBW of patients born with MI and without MI at ages 10 and 15 (Figure 12).
Figure 12: Average percent ideal body weight for patients born with and without MI

Due to the difference in %IBW at age 6, patients born with and without MI were separated during the linear regression for all ages. Linear regressions were used to test the hypothesized that patients with higher birth weights would have higher %IBWs at ages 6, 10, and 15 (Figure 13). These analyses were preformed to better understand the correlation between birth weight and later growth.
<table>
<thead>
<tr>
<th>Age</th>
<th>Born With Meconium Ileus</th>
<th>Born Without Meconium Ileus</th>
</tr>
</thead>
<tbody>
<tr>
<td>6</td>
<td><img src="image1" alt="Graph" /> R² = 0.004</td>
<td><img src="image2" alt="Graph" /> R² = 0.229</td>
</tr>
<tr>
<td>10</td>
<td><img src="image3" alt="Graph" /> R² = 0.039</td>
<td><img src="image4" alt="Graph" /> R² = 0.106</td>
</tr>
<tr>
<td>15</td>
<td><img src="image5" alt="Graph" /> R² = 0.019</td>
<td><img src="image6" alt="Graph" /> R² = 0.106</td>
</tr>
</tbody>
</table>

Figure 13: Linear regressions of percent ideal body weight (%IBW) predicted by birth weight, separated by MI status. In patients born with MI, there was no significant correlation between birth weight and %IBW at ages 6 (n=28, p=0.74), 10 (n=24, p=0.37), and 15 (n=14, p=0.640). For patients born without MI, there was a significant correlation at age 6 (n=30, p<0.01), and a trend towards significance at ages 10 (n=35, p=0.06) and 15 (n=31, p=0.08).
At age 6, 22.9% of the variability seen in %IBW can be attributed to birth weight. These analyses support the hypothesis that birth weight is predictive of %IBW but only in patients born without MI. There is no significant correlation between birth weight and %IBW in patients born with MI.

**Pseudomonas Colonization**

There were 42 patients who had a reported *Pseudomonas* colonization. An independent samples t-test determined that there was no significant difference in the age of colonization for males and females, $t(40)=0.130, p=0.94$. Therefore, males and females were combined for this analysis. The average age of colonization was 8.36 years ($SD=4.88$). An independent samples t-test was used to test the correlation between birth weight and early *Pseudomonas* colonization (colonized younger than 10-years-old). Two groups were compared: patients 10 years and older who were not colonized and patients who were colonized before age 9 (Figure 14). While it was hypothesized that *Pseudomonas* colonization would be negatively correlated with birth weight, results showed no significant difference between the average birth weights of these two groups, $t(46) = -0.09, p=0.93$. 
Figure 14: Average birth weight of who had an identified *Pseudomonas* colonization before age 9 and patients over age 10 without an identified *Pseudomonas* colonization, $p=0.93$

A Kaplan-Meier survival curve was used to visualize the time of colonization based on birth weight (Figure 15). Patients were divided into birth weight groups: birth weight above (n=49) and below (n=41) the average for the CF population, matched for gender. There were 22 patients above and 20 patients below the average birth weight who had a reported colonization. It was hypothesized that patients in the below average group would have an earlier age at colonization; however, a Tarone-Ware test confirmed that there was no significant difference in the age of colonization between the birth weight groups.
These data support the conclusion that birth weight is not correlated with age of *Pseudomonas* colonization.

Figure 15: The Kaplan-Meier survival curve shows the age of *Pseudomonas* colonization by birth weight group. Patients who had not had a reported colonization (censored) are marked on the curve by a cross at his or her age on December 31, 2013, *p*=0.68

**Cystic Fibrosis Related Diabetes (CFRD)**

There were 16 patients who had developed CFRD. It was hypothesized that patients with a lower birth weight would be more likely to develop CFRD. To test this hypothesis, patients who had developed CFRD were compared to the adult patients who did not have CFRD. The average age of CFRD diagnosis was 17.25 years (*SD*=5.54, *n*=16). The average
current age for patients with CFRD was 19.69 ($SD=6.28$, $n=16$) and 24.89 years ($SD=8.15$, $n=16$) for those without CFRD. An independent sample t-test indicated that there was no significant difference between the birth weight of patients with CFRD, $t(30)=-1.58$, $p=0.13$ (Figure 16). This suggests that birth weight does not contribute to the development of CFRD. However, this analysis is limited. There were a small number of individuals with CFRD, and the 18 patients placed in the no CFRD group may develop CFRD at a later date.

![Figure 16: The average birth weight of patients with CFRD ($n=15$) and the Adult patients without CFRD ($n=16$), $p=0.13$](image)

Through observation, it was noted that of the patients with CFRD, it seemed that birth weight was positively correlated with age of CFRD diagnosis. A linear regression was used to test this observation and indicated that birth weight explains a significant proportion of variance in age of CFRD diagnosis, $R^2=0.26$, $F(1, 15)=4.97$, $p=0.04$ (Figure 17). This signifies that 26% of the variability in age of CFRD diagnosis can be attributed to birth
weight. While this data is striking, it is important to note that one patient with very low birth weight may be skewing this data. This patient was born at 35 weeks gestation, had a birth weight of 1417.48 grams, and was diagnosed with CFRD at 10 years of age. Even though CFRD was not more prevalent in the premature population, it is possible that this one patient is affecting the overall trend due to low birth weight and early age of diagnosis.

Figure 17: The relationship between birth weight and age at CFRD diagnosis, $R^2=0.26$, $p=0.04$. 
CHAPTER 6: DISCUSSION

The correlation between weight and pulmonary function in patients with cystic fibrosis has been extensively characterized. Data has shown in both short-term and long-term studies of children and adults with CF, that weight at a certain age is positively correlated to FEV\(_1\)% at later ages (Forrester, Knox, Smyth, & Fogarty, 2012; Konstan et al., 2003; Stephenson et al., 2013; Steinkamp & Wiedemann, 2002; Yen, Quinton, & Borowitz, 2012; Zemel, Jawad, FitzSimmons, & Stallings, 2000). As patients with CF are typically underweight, there is a large effort to increase weight in order to increase pulmonary function. While the weight of children and adults with CF has been extensively studied, there has been little research done on the birth weight of patients with CF. This study aimed to characterize the birth weight of patients with cystic fibrosis and examine the correlation between birth weight and later medical phenotypes of CF. Identifying very young patients who are at higher risk to develop more severe phenotypic manifestations of CF such as lower FEV\(_1\)%, lower weight and height, earlier age of *Pseudomonas* colonization, and CFRD, would be beneficial in order to institute more aggressive treatment at younger ages.

**Aim 1**

*Birth Weight Of Patients with CF Compared to the National Average*

It was found that males and females with CF have significantly lower birth weights (10.25 oz and 9.04 oz respectively) than the national average birth weights, matched for gender. These differences in birth weight are similar to those found in the study of neonatal characteristics of patients with CF in Tuscany, Italy (Festini et al, 2005). Festini et al. found
that the males with CF were on average 261.2 g (9.21 oz) lighter than males without CF and females were on average 262.2 g (9.25 oz) lighter than females without CF, which suggests a prenatal origin of low weight in patients with CF.

The growth deficiency in children and adults with CF has previously been attributed to three main factors: lower caloric intake, higher energy expenditure, and maldigestion. Lower caloric intake, caused by decreased appetite or unwillingness to eat, can be linked to a number of factors, including chronic illness, gastrointestinal reflux, and negative body image (Haller et al., 2014). Higher energy expenditure has been attributed to chronic pulmonary infections, which leads to a higher resting metabolic rate. Finally, maldigestion has been explained by pancreatic insufficiency, which causes fat malabsorption, and uncontrolled CFRD, which can cause glycosuria (Haller et al., 2014; Pencharz & Durie, 2000). Low caloric intake can be ruled out as causes of overall prenatal growth deficiency since fetal nutrition is maintained by maternal nutrition and placental functioning. Therefore, low birth weight may be caused by higher fetal energy expenditure, maldigestion, or other factors that have yet to be explored.

Rogan et al. (2010) identified a possible explanation for lower birth weight with research on newborn pigs and humans with CF. These researches noted that neonatal insulin-like growth factor 1 (IGF1) levels in newborn pigs (n=9) and newborn babies (n=23) with CF were significantly lower than the control populations (n=11 and n=41, respectively). The authors concluded that lower IGF1 levels at birth might be related to lower weight at birth in both newborn pigs and humans with CF. IGF1 levels have been previously identified as positively correlated to birth weight in newborns without CF (n=53) (Giudice et al., 1995).
This lends evidence to the conclusion that low levels of IGF1 may be a factor in lower birth weight of patients with CF. Additional research on this topic is needed to further delineate the cause of low birth weight in cystic fibrosis.

**Contributing Factors to Birth Weight**

Multiple factors were tested in this study to identify variables that contributed to birth weight. As expected, premature patients were on average 803.70 grams (28.35 oz) lighter birth weights than full term patients. This is consistent with prenatal growth curves prediction that babies gain around 100-200 grams per week between 30-40 weeks gestation (Olsen et al., 2010).

Patients who were pancreatic sufficient had birth weights 322.88 grams (11.39 oz) heavier than patients who were pancreatic insufficient. This could indicate that the presence of fetal pancreatic enzymes contributes to fetal growth. A research study involving fetal rabbits concluded that the ingestion of amniotic fluid contributes to 10-14% of fetal growth (Mulvihill, Stone, Debas, & Fonkalsrud, 1985). If this is true, pancreatic enzymes might play a role in the break down and absorption of nutritional molecules from amniotic fluid. In this case, growth deficiency in pancreatic insufficient patients might stem from malabsorption of amniotic fluid. On the other hand, the milder CF mutations that are typically identified in patients with pancreatic sufficiency may lead to less severe fetal growth restriction as patients with pancreatic sufficiency are more likely to have some working CFTR protein (McKone et al., 2003). After all, mice with CF have been observed to have lower birth weight compared to wild type mice (Hodges & Darrah, personal...
communications, May 6, 2014), even though CF in mice does not involve pancreatic
dysfunction (Colledge et al., 1995). It is possible that a lack of CFTR protein causes higher
fetal energy expenditure, and therefore reduced fetal growth, while patients with some
working CFTR would have lower fetal energy expenditure and higher rates of growth by
comparison.

There was no significant difference between the birth weights of patients born
with and without meconium ileus. These results mirror those found by Munck et al. (2006)
and Festini et al. (2005). As the meconium ileus is found in the ileum, or the final section of
the small intestine, it is unlikely that fetal MI would affect absorption of nutrients.

Aim 2

This study aimed to identify the effect of birth weight on pulmonary function,
nutritional status, susceptibility to *Pseudomonas* colonization, and the development of
CFRD. It was hypothesized that the birth weight would be correlated with higher FEV₁ %,
higher %IBW/BMI, older age of *Pseudomonas* colonization, and lower risk of developing
CFRD.

*Predictor of FEV₁ % at Young Ages*

This study found that while birth weight is predictive for FEV₁ % at age 6,
statistical significance is not found at ages 10, 15, or 20. This indicates that as patients age,
factors other than birth weight likely contribute to a patient’s FEV₁ %. Declines in FEV₁ %
have been associated with body mass index (BMI), *Pseudomonas* colonization, pancreatic
status, and CFRD (Kerem et al., 2014). As this study did not identify any association between birth weight and later growth indices (%IBW), *Pseudomonas* colonization, or CFRD, it makes sense that birth weight would lose some its predictive quality once these environmental factors and disease phenotypes become more prevalent and influential on the patient’s FEV<sub>1</sub>%. For example, the average age of *Pseudomonas* colonization in this study was 8.36 years. Since birth weight was not predictive of age of *Pseudomonas* colonization, the onset of *Pseudomonas* would influence FEV<sub>1</sub>% independently from birth weight.

The lack of correlation between birth weight and FEV<sub>1</sub>% at ages 15 and 20 suggest that birth weight does not predict later pulmonary function. This reinforces the idea that weight and weight gain in childhood is important for later FEV<sub>1</sub>% (Zemel et al., 2000). Since birth weight does not seem to influence pulmonary function in older children and young adults, nutritional support remains to be a very important part of increasing FEV<sub>1</sub>%.

*Predictor of %IBW*

Previous research has identified a positive correlation between birth weight and weight as an adult (Parsons, Power, Logan, & Summerbell, 1999). It has been hypothesized that prenatal development serves as a “critical period” for determining how the individual will metabolically process food and store fat as a child and adult (Wells, 2007). A similar association was found in the CF population born without MI. The striking difference in the correlation of %IBW and birth weight between patients born with and without MI may be explained by early intervention. Newborn screening for CF was not implemented in Ohio until 2006. Therefore, while patients born before 2006 with MI were likely diagnosed
shortly after birth, it’s possible that patients born before 2006 without MI were not diagnosed with CF until later childhood. This would create a gap in opportunity for early nutritional and pulmonary intervention, which has been shown to improve long-term growth (Farrell et al, 2001). In addition, patients with MI may require early and aggressive nutritional intervention after surgical correction of the MI, including the use of a g-tube. Under this theory, patients born without MI may not receive early intervention and consequently birth weight becomes predictive of long-term growth. Patients with MI do not have correlations between birth weight and %IBW because early intervention decreases birth weight’s influence on long-term growth. Therefore, these findings may not suggest that the presence or absence of MI influences the correlation between birth weight and %IBW, but rather early intervention eliminates the correlation.

**Risk Factor for CFRD**

The thrifty gene hypothesis postulates that pre and perinatal growth deficiency effects the development of the pancreas. The impairment of the pancreas subsequently predisposes these individuals to the development of type II diabetes later on in life (Hales & Barker, 1992). It is difficult to apply this hypothesis to the CF population as these patients are already at a high risk for pancreatic dysfunction. In this study there was no significant difference in the birth weight of individuals with CFRD and adults without CFRD. This suggests that birth weight does not predispose certain individuals to CFRD. However, of the individuals in the studied population who developed CFRD, there was a significant positive
correlation between birth weight and age of CFRD. Therefore, if a patient will develop CFRD, birth weight explains to approximately 26% of the variability in age of onset.

**Study Limitations**

There were multiple limitations to this study. Primarily, all analyses used self-reported birth weights. The self-reporting nature of the study may have introduced false information regarding birth weight into the data. For some of the participants, recalling birth weight required knowing about their own birth weight or recalling their child’s birth multiple decades in the past. This likely decreased the validity of the reported birth weights.

It is possible that the only patients who responded to the questionnaire were those who remembered their own/their child’s birth weight. This would explain the young study population. It is likely that the younger population of patients attend their appointments with one or both of their parents. A parent may be able to recall birth weight much better than a child; therefore the nature of the questions may have created a biased sample towards the younger participant population. This greatly limited the study, as analyses on later phenotypic and anthropometric measures in the teens and adult years had much fewer respondents than the measures of the younger children.

As alluded to above, the analyses were based off a small sample size with low response rate. Some comparison studies, such as pancreatic status and CFRD, were performed with less than 20 people in a group. Therefore, the statistical significance of these analyses may not be valid. Further analyses on larger populations are required in order to determine the generalizability of these results.
Finally, studies on *Pseudomonas* colonization and CFRD were preformed using the patient’s current clinical data. It is possible that some of the unaffected patients will develop these conditions in the future. Therefore the conclusions of these analyses may not be accurate.

**Directions for Future Research**

This study identified multiple trends towards significance: FEV$_1$% at age 10 and %IBW at ages 6 and 10. Since these calculations did not reach statistical significance, additional research is needed. Perhaps a larger study that includes multiple CF centers would generate a large enough patient population for statistical significance to be identified. In addition, a larger patient population may include older participants, therefore expanding the analyses into the second decade of life.

Since growth deficiency is observable at birth, and can influence early pulmonary function, future research could be done prenatal interventions to increase birth weight. For example, research could be conducted on the feasibility and efficacy of correcting CFTR in utero with Kalydeco™. In addition, further research could examine the impact of initiating aggressive treatments in earlier in childhood for patients with lower birth weights in order to increase FEV$_1$% at age 6.

Given that the patients in this study had lower birth weights then the general population, birth weight may be a useful in predicting the likelihood that a baby flagged by newborn screening will have CF, even before genetic testing and sweat tests are preformed.
This could potentially decrease the false positive rate with the newborn screen and reduce parental stress.

Finally, as the cause of low birth weight is still unknown, additional research could aim to identify the reason that babies with CF are smaller than unaffected babies. It is possible that learning about prenatal growth deficiency could lead to more information about postnatal growth deficiency.
CHAPTER 7: CONCLUSION

This study found that patients with CF have significantly lower birth weights than unaffected babies. This suggests that the CFTR mutation influences prenatal growth. The eight full term, pancreatic sufficient patients had significantly higher birth weights than the full term, pancreatic insufficient patients. This may indicate that pancreatic function contributes to fetal growth or that the milder phenotypes leading to pancreatic sufficiency do not affect birth weight as noted in the more severe phenotypes that result in pancreatic insufficiency. While other variables that may contribute to birth weight, such as sex, birth year, presence of meconium ileus, and genotype were analyzed, no differences reached statistical significance.

There was little correlation found between birth weight and later medical complications of CF; however, there were some correlations and trending correlations noted in the first decade of life. Study data showed that birth weight was correlated with \( \text{FEV}_1\% \) at age 6 and trended towards correlation at age 10, but the correlation was not present at ages 15 and 20. This suggests that birth weight may influence and predict early pulmonary function; however, as a child ages, additional variables that were not measured in the study may exert a stronger influence on pulmonary function.

The correlation between birth weight and \%IBW trended towards significance at ages 6 and 10, but there was no correlation at age 15. Similar to \( \text{FEV}_1\% \), this may suggest that birth weight has some early influence on growth.
No correlations were identified between birth weight with *Pseudomonas* colonization or the incidence of CFRD. This suggests that individuals with lower birth weight are not more susceptible to *Pseudomonas* colonization or to developing CFRD.

To the author’s knowledge, this is the first study in the United States to examine birth weight in patients with CF, and the first study in the world to examine the correlation between birth weight and later medical complications in CF. This study confirmed that low birth weight in patients with CF is also observable in the United States, suggesting that growth deficiency in CF begins prenatally. In addition, this study identified that up to 8% of the variation in FEV₁% at age 6 can be attributed to birth weight. Birth weight is not currently thought of as a variable for pulmonary function; therefore, this finding signifies that there is at least one factor for pulmonary function set in stone at birth. However, as patients get older, there is no correlation between birth weight and pulmonary function. Consequently, long-term health in patients with CF does not seem to be correlated with birth weight.
Appendix A

The following information will be obtained from PortCF. For the measures that cover multiple ages, the patient will be included in the study up to their current age. If the patient was currently one of the ages where the data is measured, data from this age will only be included in the study if the patient has had four or more appointments where these measures were taken during the year.

Demographics
• Year of Birth
• Gender
• Race
• Genotype: ΔF508 Homozygous, ΔF508 Heterozygous, Other.
• Is the patient a twin?

Pulmonary Status
• The best FEV₁% recorded when the patient was between ages
  o 5 and 7
  o 9 and 11
  o 14 and 16
  o 19 and 21
  o 24 and 26
• The patient’s age when Pseudomonas colonization was identified.
  o Pseudomonas colonization will be identified when the patient has had three positive Pseudomonas cultures in a row.

Gastrointestinal Status
• Is the patient pancreatic sufficient or insufficient?
• Was the patient born with Meconium Ileus?
• Does the patients have CFRD?

Nutritional Status
• The patient’s highest Percent Ideal Body Weight (%IBW) between the ages
  o 5 and 7
  o 9 and 11
  o 14 and 16
• The patient’s highest Body mss index (BMI) between the ages
  o 19 and 21
  o 24 and 26
1. What was the patient’s birth weight?
   _____ lbs _____ oz

2. What was the patient’s birth length?
   _______ inches

3. Was the patient born at full term (after 36 to 42 weeks of pregnancy)? Mark one
   _____ Yes
   _____ No

*If known, how many weeks pregnant was the patient’s mother when she gave birth to the patient?
   _________ weeks
Appendix C: Verification of Aim 1 exclusion criteria

Gestational Age

An independent samples t-test was used to test the hypothesis that patients born prematurely would have lower birth weights than patients born full term. This analysis found that patients born prematurely had lower birth weights than patients born full term. Patients with CF born premature (<37 weeks gestation) were significantly lighter ($M=2388.28$ g, $SD=572.90$) than patients born full term ($M=3191.98$ g, $SD=396.35$), $t(93)=6.82$, $p<.01$ (Figure 18). The significant differences between patients born premature and full term supports the exclusion of all patients born premature the aim 1 analyses.

Figure 18: Average birth weight of patients with CF born prematurely and patients born full term
**Pancreatic sufficient**

It was predicted that patients who were pancreatic sufficient would have higher birth weights than patients who were pancreatic insufficient. An independent samples t-test found statistical significance between the birth weights of full term patients with pancreatic sufficiency ($M=3514.87$ g, $SD=381.87$ g, $n=8$) and full term patients with pancreatic insufficiency ($M=3191.99$ g, $SD=396.35$ g, $n=79$), $t(85)=2.202$, $p=.03$ (Figure 19). This analysis supported the exclusion of pancreatic sufficient patients from the analyses in aim 1; pancreatic sufficient patients have significantly higher birth weights than pancreatic insufficient patients.

![Figure 19: Average birth weight of patients who are pancreatic sufficient and patients who are pancreatic insufficient](image-url)
Twin Pregnancy

There were two sets of twins in the study population. Both sets were born premature (34 weeks and 36 weeks). Therefore, the twins were excluded with the participants born premature.
REFERENCES


48. Stephenson AL, Mannik LA, Walsh S, Brotherwood M, Robert R, Darling PB,


