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Epidemiology

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B.S. in Public Health
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An abstract of
A thesis submitted to the Faculty of the
Rollins School of Public Health of Emory University
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Abstract

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Introduction

Diet is a known modifiable factor that can either have negative or positive implications for the incidence of colorectal cancer, a cancer that has affects roughly 1 in 23 men and 1 in 26 women[28]. There are many aspects of a diet that can lead to outcomes of interest, in this case, metabolomic markers that are linked to colorectal cancer risk.

Methods

Fiber is what we investigated within a study population of 15 people, both males (n=8) and females (n=7), as well as Black (n=7), White (n=7), and Hispanic (n=1) individuals using a subset of data from an ongoing randomized controlled clinical trial. By comparing variables of interest at each participants' baseline and 6-month visits.

This randomized clinical feeding trial is being conducted to gain an understanding of the implications a specific diet has on weight-related variables and gastrointestinal (GI) tract transit times, among other things. Data was collected from each participant at baseline and 6 months, standardized as needed, and compared using both a Wilcoxon Signed Rank Test and a Spearman's Correlation Tests.

Results

Overall, results did not provide sufficient evidence to support the hypothesis that an increased fiber intake (g/ 1,000 kilocalories) is associated with a decrease in weight, BMI, waist circumference, and total percent body fat and a decrease in GI tract transit time variables. When stratifying on sex, we saw that the associations among men were more robust for changes in weight, waist circumference and total percent body fat (p-values < 0.05), than those observed within females.

Discussion

What this study provides is a basis of knowledge that can be expanded upon. Other macronutrients can be compared to these outcome variables over a longer period. A larger sample size will also prove to be beneficial in producing more robust findings. There is a need for more research surrounding these topics. A broader scope of knowledge can be achieved and eventually be shared within the cancer research community, potentially leading to a decreased burden of colorectal cancer in the United States.

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My father passed away from colon cancer when I was young, therefore, this research holds a special place in my heart. I dedicate this work to him and all of those who have lost a family member to cancer.

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Chapter 1: Literature Review

How Diet Affects Health

Understanding the implications of chronic disease occurrence in individuals is not a straightforward path. There are many factors that, when paired with underlying aspects such as genetic makeup, can place an individual at an increased risk for chronic diseases such as obesity, type 2 diabetes, and cancer. Diet is a known risk factor for the aforementioned diseases; however, it is something humans require for survival, thus creating a complex framework. One million people in the United States die from diet-related diseases every year[1]; thus, diet needs to be looked upon as a preventative measure for chronic diseases such as obesity and cancer. Eating patterns and behaviors that lead to chronic disease occurrence are influenced at multiple levels, starting as broad as governmental policy-making, to individual decision-making, family impact, and biological composition[2]. Previous studies have provided some evidence for the beneficial effects of a high fiber diet on colorectal cancer prevention, but few have focused on exploring the mechanisms whereby fiber reduces risk. What this study intends to explore is the implementation of a diet that is high in fiber and how that simultaneously leads to the presence of metabolomic markers known to potentiate weight loss and attenuate the risk of colorectal cancer, a cancer that is heavily linked to diet and energy balance.

High Fiber Diet and Health

Many diets exist with the goal of reducing body weight and limiting one's risk of chronic diseases such as obesity and cancer; however, research points us to investigate the benefits of a diet that contains high levels of fiber. Fiber falls under the category of carbohydrates and can be found at higher levels in fruits and vegetables, whole grains, and legumes. Primary purposes of including fiber in one's diet are to improve bowel health and to reach a weight that is healthy for the individual[3], both of which are intended outcomes of this study. Past studies have also shown that fiber contributes to reduced risk for coronary heart disease, stroke, diabetes, and many more diseases. Unfortunately, according to an article from Nutrition Reviews, it is estimated that the majority of people living in the United States consume less than half of what is recommended for daily fiber intake[4]. This study seeks to examine the benefits a high fiber diet can provide to a participant who is classified as high risk for diet-related diseases, specifically colon cancer and obesity.

High Fiber Diet and Obesity

Approximately 1 in 3 adults living in the United States are obese[5]. Obesity is a complex disease that if left unmanaged, can lead to a surfeit of other health issues and diseases. As a major cause of millions of premature deaths worldwide every year, obesity continues to climb in prevalence, with the World Health Organization projecting that 20% of the adult population around the world will be obese by 2030[6]. In some areas of the world, including the United States, where the prevalence of obesity is very high, diets have become more westernized, meaning characterized by larger portion sizes, heavily processed products, products containing abnormally high levels of sugar, etc., therefore often lacking in other important dietary components such as fiber. For example, a study conducted in 2009, including approximately 5,000 men and women from the Netherlands Cohort Study evaluated the association of fiber intake measured by food frequency questionnaire with BMI. Among men, each additional gram of fiber consumed was associated with a marginally significant 3% (95% CI: -1-8%) lower risk of being obese (BMI $\geq$30) compared to normal BMI (BMI <25). No significant differences were seen in the female population, which was attributed to reporting bias. A related study including 3083 adults in

Belgium which collected dietary intake data via two non-consecutive dietary recalls did not report a significant association between dietary fiber intake and BMI but did find a significant inverse relationship for waist circumference. Essentially, it was reported that as fiber intake increased, waist circumference was lower, with a reported beta value of -0.118 (95% CI -0.181- -0.055; $p<0.0001$) amongst men who had a mean total fiber intake of 17.8 grams per day and amongst women who had a mean total fiber intake of 15 grams per day[8]. There are limitations to these studies, and others that were looked at, due to the response rates and self-reporting of variables such as anthropometric variables and food intake, but they do align with the general hypothesis.

High Fiber Diet and Cancer

In 2021, the number of new cases in the United States reached almost 2 million people, out of which a little over 600,000 people have died[9]. Cancer is a disease that has modifiable and non-modifiable risk factors, meaning that certain risk factors can be changed, such as diet and physical activity levels, or certain risk factors are unable to be changed throughout one's life, such as the genome someone was born with. Diet is an exposure that can either attenuate the risk of developing cancer, or strengthen the risk, depending on what the diet is comprised of. Previous research suggests a high fiber diet may have beneficial effects related to cancer prevention, specifically colon cancer prevention. A prospective nested case control study was conducted that involved a much larger study sample of adults in the United Kingdom (579 cases of colorectal cancer patients and 1,996 controls), which sought to utilize dietary data collected by food records in order to understand how the differences in fiber intake affected colon cancer risk[11]. An inverse relationship was observed; those in highest quintile of energy-adjusted fiber intake had a lower risk of colon cancer (OR:0.66; 95% CI:0.45-0.96) compared to those in the lowest quintile of intake.

Diet and the Gut Microbiome/Metabolic Health

What was once thought of as simply an aspect of the digestive system, the human microbiome is an extraordinarily complex system that has implications for many other mechanisms throughout the body. Much progress has been accomplished over the past several years regarding microbiome research and discovery; not only how it affects not only our digestive system, but mood and mental state, immune system, and disease pathogenesis[12]. As defined by the National Human Genome Research Institute, a microbiome is a community of microorganisms that exists in a particular environment[13], in this case, we will be discussing the human gut microbiome. Microbiomes are located all over the human body, with microbes existing in the mouth, skin, and reproductive organs, and the gut microbiome can be found in the small and large intestines. The gut microbiome, comparable to an organ, is uniquely determined and designed by one's own DNA as early as infancy, when the baby passes through the mother's birth canal[14]. The structure of the human gut microbiome changes throughout one's lifespan. Much can be attributed to the alteration the gut microbiome undergoes throughout someone's life, but one of the most pertinent influences is diet. On average, a human being's GI tract, which contains the gut microbiome, is responsible for passing about 60 tons of food throughout the human lifetime, making diet a key player in the health and well-being of the gut microbiome[15]. To understand the microbiome and its effectiveness, there are existing gut metabolomic markers that are collectible and measurable, which in turn allows us to understand how a specific diet, such as a high fiber diet, contributes to the gut microbiome and may lead to weight loss, obesity prevention, and colon cancer prevention.

Gut Metabolome Markers

To observe the effects of a high-fiber diet over the course of 6 months, this study will measure metabolic markers including short chain fatty acids and bile acids and will track the gut transit time. Short chain fatty acids (SCFAs) are molecules that are found in the gut microbiome and are generated with the digestion of fibrous foods[16]. The three most commonly studied SCFAs are acetate, propionate, and butyrate, and they provide a plethora of benefits ranging from anti-obesity to anticancer outcomes once they are absorbed into the bloodstream[17,18]. Depending on the amount of fiber that is consumed, the SCFAs that are generated are used as an energy source in other surrounding organs such as the colon and muscles[18]. Fiber-rich diets increase SCFAs, thought to be highly beneficial for colon cancer prevention, as they are responsible for histone deacetylases and apoptosis promotion in the colon[19]. The same beneficial relationship cannot be said with confidence regarding obesity. An article discussing the relationship between SCFAs and weight loss explains that there is no clear causal pathway between the two. There is conflicting evidence whether SCFAs promote weight loss by encouraging the release of hormones that decrease appetite, and in turn, caloric intake, or if SCFAs lead to an excess caloric intake[20]. Bile acids are another biomarker used to assess the effects of a high-fiber diet on obesity and colon cancer prevention. Bile acids are the products of cholesterol metabolism and are cytotoxic when they collect in high concentrations[21]. There are two kinds of bile acids that exist in the body. Primary bile acids are those that are formed in the hepatocyte – a liver cell -, whereas secondary bile acids are those that are formed from primary bile acids via colonic bacteria[22,23]. In high concentrations, bile acids have the capability to send signals to other molecules in the body and alter their functions, which can lead to cancer[24]. When a diet containing high levels of fiber is consistently consumed, the concentration of secondary bile acids decreases and is excreted through stool, because they attach themselves to fiber molecules[25,26]. This is important because bile acids may promote carcinogenesis when their concentration becomes too high for the body to manage. The last marker that will be analyzed to help understand the effects of a high fiber diet is the gut transit time or the time it takes for food to transport from the mouth upon food intake, to the end of the intestine when it needs to be excreted[26]. Gut transit times may vary from person to person based on diet and differ between males and females with average transit times being 30-40 hours among men, but natal sex women sometimes reach 100-hour transit times[27]. It is important that gut transit time be included as a covariate in studies pertaining to the microbiome, diet, and diet-related diseases. A technology known as the SmartPill can be used as a tracking device to map transit time through regions of the GI tract. It can be easily swallowed, and it then excreted via stool during a normal trip to the bathroom. A fibrous diet helps to increase the weight of stool, which can minimize the amount of carcinogenic exposure within the GI tract and can decrease transit time[6].

Chapter 2: Materials and Methods

Introduction

Colorectal cancer (CRC) is among the 5 most common cancers in the United States. It has a lifetime risk of about 1 in 23 for men and 1 in 26 for women and is expected to be the cause of over 50,000 deaths in the year 2023[28]. Occasionally referred to as simply colon cancer, CRC is a form of cancer that is found in either the colon, rectum, or both. Polyps, or abnormal cellular growths[29], which can be removed, are precursors to CRC. While diet overall has been studied in relation to CRC risk, a diet that requires further investigation is one that is rich in fiber. Fiber is a type of carbohydrate that is found in foods like dry beans (cooked), fruits, vegetables, and various nuts and seeds. It is a beneficial aspect to a diet, as it aids in the regulation of sugars found in the body, thus helping to manage hunger and blood sugar levels[30]; however, its linkage to CRC risk remains ambiguous. In part, this may be due to measurement error in diet assessment which may attenuate associations.

We can study the mechanistic operations of fiber as it is digested and processed in the body, and how that can alter other bodily functions and mechanisms that are known linkages to CRC protection; the prime example being observed here is gastrointestinal (GI) transit time. Defined as the time it takes for food to be transported from the mouth following the intake of food to the end of the intestine prior to excretion[27], the speed of a GI transit time can provide information pertaining to CRC risk, and whether fiber intake modifies this association. The average transit times for men and woman are about 30-40 hours and about 100 hours[19], respectively, with a time exceeding that potentially posing an increased risk to CRC. The importance of speeding up the GI transit time can be attributed to the benefits of short-chain fatty acids (SCFA) and the risks of bile acids. SCFAs are a known benefit and can lower the risk of CRC via histone deacetylases and apoptosis promotion in the colon[17,31]. Bile acids are known to be quite harmful[24]; however, are shown to pose less of a threat to the body when there is an increase of fiber intake, as bile acids can latch on to fiber molecules that exist within stool, which is then excreted[32]. Literature suggests that the increase of fiber has the ability to increase the speed of the GI transit time, which will limit the tract's exposure to harmful bile acids. The role of fiber in determining the speed of the GI transit time is what was examined in this study.

With the purpose of understanding the relationship between the implementation of a fiber-rich diet and the increased protection of CRC (using the proxy of an increased gut transit time), a 6-month long blinded clinical feeding trial is being conducted. Selected preliminary analyses were conducted for this thesis including 15 participants, a subset of the total planned enrollment in the ongoing trial. Measures of obesity (i.e., a reduction in weight, BMI, and waist circumference) will also be observed, as obesity and CRC are known comorbidities[33].

Methods

Participants

The subjects in this study were 15 adults aged 40-75 years old. There were 8 men and 7 women. To be eligible for the trial, participants needed to have a BMI between 25-40 kg/m^2 with an interest in losing weight and have had a colonoscopy sometime within the 3 years prior to enrollment that found adenoma(s) in between the size range of 0.5 and 1 cm. Other eligibility requirements for participants included English speaking, ambulatory, and able to provide consent.

Recruitment

Prior to enrollment, every participant provided informed consent and went through a screening process in order to determine if they met the aforementioned inclusion criteria. Food allergies and aversions were also recorded along with traveling plans for the year. Other exclusion factors include the participant having a serious medical condition, history of Colorectal Cancer, bowel resection, polyposis syndrome or any inflammatory bowel issue/disease. Participants could not have smoked regularly in the year leading up to potential enrollment, or have complicated dietary restrictions, unusual exercise behavior, regular usage of medications that are known to affect anything related to GI tract, or be currently pregnant, breastfeeding, or planning to become pregnant. Viable candidates were given an ID number in REDCap; the data collection tool that was utilized throughout the duration of this study to collect information regarding all in-person visits (i.e., baseline and 6-month visits).The current analyses include only participants who completed both their baseline and 6-month appointment as of 04-15-2023.

Entrees

For the first three months of the study, participants receive two entrees per day, prepared by trained kitchen staff at Emory University Hospital Georgia CTSA Clinical Research Center (EUH GCRC). Participants picked up their allotted number of entrees weekly, some fresh and some frozen. For the second three months, the same process is carried out, however, participants receive one entree per day rather than two.

All entrees are pre-portioned by the study bio-nutrition staff with suggestions made for appropriate side dishes consistent with promoting healthy weight loss.

Legume Fiber Diet

Participants assigned to this diet will be consuming about 250g of legumes a day (30g), with a goal of consuming 40 or more grams of fiber a day.

Healthy American Diet

Participants assigned to this diet (the control diet) receive similar types of entrees to the legume diet, however, legumes are replaced with lean meat and chicken.

Randomization

At the closure of the enrolled participant's baseline appointment, they are randomized to their assigned diet, whether it be the control diet (Healthy American Diet) or the intervention diet (Legume Fiber Diet). This is a blinded randomization process done using R Software to generate random numbers that will be used to assign diet plans.

Measurement of Variables

Variables intended for analysis included sex, age, race, height (m), weight (kgs), BMI (kg/m^2), systolic and diastolic blood pressure (mm Hg), waist circumference (cm), calories consumed (kcal), whole gut (minutes), gastric emptying time (minutes), colonic transit time (minutes), bowel transit time (minutes), total body fat (%), and dietary intake of macronutrients including fiber intake (both in g and g/1,000 kcals), protein intake (g/1,000 kcals), trans-fat intake (g/1,000 kcals).

Anthropometric Variables

The anthropometry variables (height, weight, BMI, systolic blood pressure, waist circumference) were collected following NHANES guidelines with participants while in light clothing and without shoes. These variables were recorded in REDCap for each participants' baseline and 6-month appointment. These variables were collected by trained research staff at the EUH GCRC. Variables sex, age, and race was self-reported.

Nutritional Variables

24-hour diet recalls were collected over the telephone / Zoom by trained interviewers utilizing the web-based ASA24 tool[34] at baseline and 6-months for each participant to collect data on calories consumed, fiber intake, protein intake, and trans-fat intake. Two of these phone conversations were conducted around the time of each participants' baseline and 6-month appointments. The ASA24 software automatically codes and organizes the data with final values available for download. Participants' two days of data per timepoint were averaged for each time.

Whole Gut Transit Time Variables

We measured gut transit time by utilizing the SmartPill; a device to collect data on a person's GI tract, including pressure, pH, and temperature. Prior to ingesting the SmartPill, the participant must have consumed a small nutritional snack bar, along with 50mL of water. With the assistance of water, the participant swallows the SmartPill, which then connects to a corresponding receiver worn by the participant and displays software that will show progress through the intestine. Following the participant taking the SmartPill, they are given instructions to avoid heavy exercise, alcohol consumption, smoking, and any medications that could alter the SmartPill results. They are asked to record any GI-related symptoms and side effects and to return the SmartPill receiver after 5 days. The SmartPill does not get returned, as it gets excreted from the body as any normal bowel movement. The data collected from this test is used to calculate whole gut transit time (in minutes), along with gastric emptying time, colonic transit time, and bowel transit time (all reported in minutes).

Total Percent Body Fat Variable

This variable was collected via a dual-energy X-ray absorptiometry (DEXA), a medical imaging test used to estimate body bone density, body fat, and muscle composition. For this study, the variable of interest was the participant's total % body fat at baseline and 6-months. Following the body scan, results were recorded and stored for each participant.

Analysis

Data were analyzed using SAS. Cutoff points for categorical variables BMI, systolic and diastolic blood pressure, and waist circumference were determined based on existing literature. Descriptive statistics (i.e., frequencies, mean, and standard deviation) were obtained by running frequency and mean procedures. Fiber intake, protein intake, and trans-fat intake were all converted from their original raw values (reported in grams) to grams per 1,000 calories. This allowed the variables to be easily comparable across varying caloric intakes. Participant data pertaining to total calories consumed was excluded if the individual participant reported consuming less than 800 calories in one day (seen from ASA24 diet recall data). The univariate procedure was run to obtain Wilcoxon Signed Rank values and the correlation procedure was run to obtain a Spearman's correlation coefficient for each variable of interest.

Results

As seen in Table 1, there were more men in this study (n=8) than women (n=7) and a majority of the study population was over the age of 50 (n=13). There was an even number of Black/African Americans and White/Caucasians (n=7 for both populations); and one subject who identified as Hispanic. Most of the study population were classified as obese at the beginning and end of the study period (n=11 and n=9, respectively). For both systolic and diastolic blood pressure, the majority fell into the "normal" category at both baseline and 6 months (systolic: n=7 and n=7, respectively; diastolic: n=11 and n=12, respectively). Looking at waist circumference, most of the study population were in the "extra-large" category at baseline and 6 months (male: n=8 and n=7, respectively; female: n=6 and n=5, respectively.

Table 1. Characteristics of Participants at Baseline and 6 Months

Variables	Baseline n = 15 n (%)	6 Months n = 15 n (%)
Sex		
Male	8 (53.3%)	
Female	7 (46.67%)	
Age		
<50	2 (13.3%)	
>50	13 (86.7%)	
Race		
Black/African American	7 (46.7%)	
White/Caucasian	7 (46.7%)	
Hispanic	1 (6.7%)	
BMI (kg/m^2)		
Healthy (18.5-24.9)	1 (6.7%)	3 (20.0%)
Overweight (25.0-29.9)	3 (20.0%)	3 (20.0%)
Obese (>=30.0)	11 (73.3%)	9 (20.0%)
Systolic Blood Pressure (mm Hg)		
Normal (<120 mmHg)	7 (46.7%)	7 (46.7%)
At Risk (120-139 mm Hg)	5 (33.3%)	6 (40.0%)
High Blood Pressure (≥140 mm Hg)	3 (20.0%)	2 (13.3%)
Diastolic Blood Pressure (mm Hg)		

Normal (<80 mm Hg)	11 (73.3%)	12 (80.0%)
At Risk (80-89 mm Hg)	4 (26.7%)	3 20.0%)
High Blood Pressure (≥ 90 mm Hg)	0 (0.0%)	0 (0.0%)
Waist Circumference (cm)		
Male		
Normal (<94)	0 (0.0%)	0 (0.0%)
Large (94-102 cm)	0 (0.0%)	1 (12.5%)
Extra Large (>102 cm)	8 (100.0%)	7 (87.5%)
Female		
Normal (<80)	0 (0.0%)	1 (14.3%)
Large (80-88 cm)	1 (14.3%)	1 (14.3%)
Extra Large (>88 cm)	6 (85.7%)	5 (71.4%)

Table 2 provides means and standard deviations for each variable of interest from baseline and 6 months, overall and stratified by sex for the treatment groups (both Legume and Healthy American Diets combined). To visualize the trends of these variables for males, females, and overall, the differences of the reported means were also included, and these were calculated by subtracting the baseline value from the 6-month value. Variables expected to have decreased (i.e., portray a positive value) include weight, BMI, systolic and diastolic blood pressure, waist circumference, total body fat, and all transit times. Looking at these differences in the unstratified ("All") category, expectations were met for weight, BMI, both blood pressures, waist circumference, total body fat, and gastric emptying time. Results very when we stratify on sex. calories consumed and trans-fat intake, fiber intake (g) show an increase in men and a decrease in women, however, protein intake, colonic transit time, and bowel transit time shows the opposite. Variables we expected to increase (i.e., portray a negative value), include fiber intake (g) and fiber intake (g/1,000 kcals), which can be seen for the "All" category and the males in fiber intake (g). No increase is seen for any of the categories in the fiber (g/1,000 kcals) variable or for females in the fiber intake (g) variable.

Table 2. Averages at Baseline and 6 Months with Differences Stratified by Sex			
	Baseline n = 15	6 Months n = 15	
Variables	Mean (±SD)	Mean (±SD)	Difference of Means*
Total Calories Consumed (kcal)			
All	1418.7 (±400.2)	1391.7 (±442.1)	27.0
Male	1366.5 (±436.0)	1662.0 (±399.5)	-295.5
Female	1296.6 (±422.6)	1073.9 (±249.4)	222.7
Fiber Intake (g/1000 kcals)			
All	14.5 (±7.0)	14.2 (±5.7)	0.3
Male	14.9 (±7.1)	14.3 (±5.6)	0.6
Female	14.1 (±6.8)	14.0 (±5.8)	0.1
Fiber Intake (g)			
All	18.8 (±7.1)	21.1 (±11.4)	-2.3
Male	19.5 (±5.4)	26.5 (±12.2)	-7
Female	18.0 (±8.5)	14.5 (±6.0)	3.5
Protein Intake (g/1000 kcals)			
All	49.0 (±9.5)	49.4 (±15.5)	-0.4
Male	50.5 (±9.0)	46.3 (±9.6)	4.2
Female	47.3 (±9.7)	52.8 (±19.6)	-5.5
Trans-Fat Intake (g/1000 kcals)			
All	34.3 (±9.5)	36.0 (±7.5)	-1.7
Male	32.6 (±9.9)	37.0 (±8.2)	-4.4
Female	36.2 (±8.6)	34.9 (±6.4)	1.3
Weight (kg)			
All	97.9 (±22.8)	93.6 (±23.3)	4.3
Male	116.1 (±12.3)	111.4 (±13.9)	4.7
Female	77.1 (±13.5	73.3 (±15.0)	3.8
BMI (kg/m^2)			
All	32.6 (±4.4)	31.4 (±5.4)	1.2
Male	35.1 (±3.1)	34.1 (±4.3)	1
Female	29.7 (±4.6)	28.2 (±3.4)	1.5
Systolic Blood Pressure (mm Hg)			
All	125.1 (±15.7)	123.0 (±15.0)	2.1
Male	129.6 (±13.7)	128.8 (±13.5)	0.8
Female	120 (±18.4)	116.9 (±15.6)	3.1
Diastolic Blood Pressure (mm Hg)			
All	71.4 (±9.7)	71.0 (±9.50)	0.4
Male	76.1 (±9.3)	74.6 (±8)	1.5
Female	66 (±8.6)	66.7 (±10.7)	-0.7
Waist Circumference (cm)			
All	109.3 (±14.2)	104.3 (±13.8)	5
Male	120.6 (±8.5)	114.5 (±7)	6.1

Female	96.5 (±7.5)	92.8 (±11.4)	3.7
Total Body Fat (%)			
All	42.2 (±4.2)	39.1 (±4.8)	3.1
Male	40.3 (±4.6)	37.3 (±4.2)	3
Female	44.5 (±3.0)	41.1 (±5.3)	3.4
Whole Gut Transit Time (minutes)			
All	2470.8 (±1503.2)	2778.7 (±1554.7)	-307.9
Male	2441.3 (±1537.2)	2058.3 (±955.4)	383
Female	2505.3 (±1742.6)	3602.1 (±1869.8)	-1096.8
Gastric Emptying Time (minutes)			
All	215.4 (±201.4)	205.9 (±135.8)	9.5
Male	159.0 (±75.0)	156.0 (±54.5)	3
Female	281.2 (±270.7)	263.0 (±173.3)	18.2
Colonic Transit Time (minutes)			
All	1771.6 (±1351.0)	2197.5 (±1609.2)	-425.9
Male	2202.6 (±1400.2)	1478.5 (±1043.7)	-816.7
Female	1268.8 (±1094.6)	3019.3 (±1743.0)	-1750.5
Bowel Transit Time (minutes)			
All	1705.9 (±1230.1)	2429.8 (±1629.7)	-723.9
Male	1909.3 (±1243.4)	1695.4 (±1064.0)	213.9
Female	1468.7 (±1170.5)	3269.1 (±1754.0)	-1800.4
*Calculated by: Baseline - 6 Months; a negative value indicates an increase; a positive value indicates a decrease			

In Table 3, we present overall and sex-stratified results for differences between variables at baseline and 6 months with Wilcoxon Signed Rank Tests of whether their median values differ (p-value alpha set at 0.05). We can see large differences in weight (17 kg; 0.02), BMI (9 kg/m^2; 0.25), waist circumference (17 cm; 0.02), and total body fat (15%; 0.04), with differences in weight, waist circumference, and total body fat being a statistically significant difference (all p-values are <0.05). There were no significant differences observed among women. In Table 2, we saw the differences of each variable from baseline to 6-months, and in Table 3, we can see how strong these differences are. Overall, the highest, as well as the most statistically significant, reported measurements correspond to variables weight (51.10 kg; 0.002), BMI (38.00 kg/m^2; 0.03), waist circumference (51.00 cm; 0.002), and total body fat (51.00%; 0.002).

Table 3. Comparison of Variables of Interest from Baseline and 6-Months Stratified by Sex

Variables	Mean of Differences (±SD)[1]	Wilcoxon Signed Rank (p-value)*
Weight (kg)		
All	4.31 (±3.92)	51.1 (0.002)
Male	4.71 (±3.99)	17 (0.02)
Female	3.84 (±4.10)	11 (0.08)
BMI (kg/m^2)		
All	1.23 (±1.69)	38 (0.03)
Male	0.99 (±1.80)	9 (0.25)
Female	1.49 (±1.65)	11 (0.08)
Systolic Blood Pressure (mm Hg)		
All	1.93 (±9.32)	17 (0.35)
Male	0.87 (±11.37)	3.5 (0.67)
Female	3.14 (±6.99)	6.5 (0.33)
Diastolic Blood Pressure (mm Hg)		
All	0.47 (±8.04)	3 (0.87)
Male	1.50 (±8.35)	3.5 (0.66)
Female	-0.71 (±8.16)	-2 (0.78)
Waist Circumference (cm)		
All	4.99 (±5.40)	51 (0.002)
Male	6.13 (±6.27)	17 (0.02)
Female	3.69 (±4.31)	10 (0.11)
Total Body Fat (%)		
All	3.19 (±3.02)	51 (0.002)
Male	3.05 (±2.99)	15 (0.04)
Female	3.34 (±3.29)	12 (0.05)
Total Calories Consumed (kcal)[2]		
All	-10.96 (±456.91)	-2.5 (0.90)
Male	-295.74 (±232.68)	-16 (0.02)
Female	368.39 (±405.97)	7.5 (0.16)
Fiber Intake (g)		
All	-2.33 (±11.29)	5 (0.80)
Male	-7.01 (±13.80)	-7 (0.38)
Female	3.02 (±3.56)	10 (0.11)
Fiber Intake (g/1000 kcals)		
All	0.33 (±5.66)	7 (0.72)
Male	0.58 (±7.68)	2 (0.84)
Female	0.05 (±2.40)	1 (0.94)
Protein Intake (g/1000 kcals)		
All	-0.35 (±14.11)	-1 (0.98)
Male	4.22 (±12.24)	6 (0.46)
Female	-5.57 (±15.17)	-7 (0.30)

Trans-Fat Intake (g/1000 kcals)		
All	-1.75 (±10.70)	-5 (0.80)
Male	-4.40 (±9.65)	-6 (0.46)
Female	1.27 (±11.76)	3 (0.67)
Whole Gut Transit Time (minutes)[3]		
All	-217.92 (±1656.79)	-6.5 (0.68)
Male	349.14 (±1597.74)	3 (0.69)
Female	-879.50 (±1596.61)	-4.5 (0.44)
Gastric Emptying Time (minutes)[3]		
All	0.15 (±263.38)	-11.5 (0.45)
Male	0.86 (±56.80)	1 (0.94)
Female	-0.67 (±403.26)	-4.5 (0.44)
Colonic Transit Time (minutes)[3]		
All	-422.08 (±1922.29)	-11.5 (0.45)
Male	516.00 (±1640.32)	4 (0.58)
Female	-1516.50 (±1723.47)	-7.5 (0.16)
Bowel Transit Time (minutes)[3]		
All	-729.69 (±1798.88)	-19.5 (0.19)
Male	-24.29 (±1587.79)	-1 (0.94)
Female	-1552.67 (±1797.68)	-7.5 (0.16)

*alpha set at 0.05
[1] Calculated by: Baseline - 6 Months
[2] Missing one participant's value
[3] Missing two participants' values

As shown in Table 4, we can see the Spearman Correlations of the relationship between Fiber Intake (g/1000 calories, total g) and each outcome variable of interest, with a specific interest in the transit time variables (whole gut transit time, gastric emptying time, colonic transit time, and bowel transit time), reported in minutes. The correlation coefficient values reflect the exposure change from baseline to 6 months and the outcome change from baseline to 6 months. For weight and fiber (g/1,000 kcals), there is a correlation of 0.45 (0.26) for males and a marginally significant correlation of 0.71 (0.07) for females. In contrast, for BMI, there is a correlation of 0.31 (0.46) and 0.50 (0.25) with fiber (g/1,000 kcals). For both weight and BMI, women have the stronger correlation. Both systolic blood pressure and whole gut transit time show inverse relationships with fiber intake for males and females. For systolic blood pressure, there is a significant correlation of -0.76 (0.03) for females for energy-adjusted fiber intake. For whole gut transit time, Spearman correlations with energy-adjusted fiber intake for males was -0.50 (0.25) and -0.77 (0.07) for females. Another relationship that is shown to be statistically significant includes the relationship between fiber intake and waist circumference within males, with a reported value of 0.83 (0.01). Looking at these results overall, weight in relation to energy-adjusted fiber intake shows a significant positive correlation of 0.53 (0.04), as does waist circumference, with a value of 0.72 (0.003).

Table 4. Spearman's Correlation Between Fiber Intake and Outcome Variables of Interest between Baseline and 6 months; Stratified by Sex

	Spearman's Correlation Coefficient (p-value)*	
Variables	Fiber Intake (g/1000 calories)	Fiber Intake (g)
Weight (kg)		
All	0.53 (0.04)	0.23 (0.42)
Male	0.45 (0.26)	0.14 (0.74)
Female	0.71 (0.07)	0.43 (0.33)
BMI (kg/m^2)		
All	0.33 (0.23)	0.33 (0.23)
Male	0.31 (0.46)	0.07 (0.87)
Female	0.50 (0.25)	0.71 (0.07)
Systolic Blood Pressure (mm Hg)		
All	-0.71 (0.003)	-0.64 (0.01)
Male	-0.76 (0.03)	-0.90 (0.002)
Female	-0.39 (0.38)	-0.54 (0.22)
Diastolic Blood Pressure (mm Hg)		
All	-0.37 (0.17)	-0.64 (0.01)
Male	-0.66 (0.08)	-0.83 (0.01)
Female	0.16 (0.73)	-0.22 (0.64)
Waist Circumference (cm)		
All	0.72 (0.003)	0.17 (0.55)
Male	0.83 (0.01)	0.62 (0.10)
Female	0.00 (1.00)	-0.46 (0.29)
Total Body Fat (%)		
All	0.45 (0.09)	0.09 (0.76)
Male	0.40 (0.32)	0.21 (0.61)
Female	0.50 (0.25)	-0.11 (0.82)

Whole Gut Transit Time (minutes)[1]		
All	-0.48 (0.10)	-0.07 (0.82)
Male	-0.50 (0.25)	-0.32 (0.48)
Female	-0.77 (0.07)	0.03 (0.96)
Gastric Emptying Time (minutes)[1]		
All	0.28 (0.36)	-0.51 (0.07)
Male	0.39 (0.38)	0.07 (0.88)
Female	-0.09 (0.87)	-0.09 (0.004)
Colonic Transit Time (minutes)[1]		
All	-0.28 (0.35)	-0.01 (0.99)
Male	-0.50 (0.25)	-0.32 (0.48)
Female	0.03 (0.96)	0.77 (0.07)
Bowel Transit Time (minutes)[1]		
All	-0.13 (0.68)	0.25 (0.42)
Male	-0.29 (0.53)	-0.07 (0.88)
Female	0.03 (0.96)	0.77 (0.07)

*alpha set at 0.05

[1]Missing two participants' values

Discussion

The overall purpose of this study was to investigate the association between the 6-month long implementation of a high-fiber diet and the presence of metabolomic markers that are known protectants of colon cancer. We did observe longitudinal differences between many of the outcome variables of interest, as well as strong correlations for dietary fiber intake with outcomes; however, likely due to the small sample size in these preliminary analyses, the majority of the findings do not reach significance.

Both males and females lost weight during follow-up, regardless of the diet they were randomly assigned to, with significant weight loss observed among males. Males also showed decreases in waist circumference and total body fat at 6 months, with values of 6.13 cm (±6.27) and 3.05% (±2.99), respectively. When we look at waist circumference in relation to fiber intake (g/1,000 kcals) a positive correlation is seen, however, this contrasts a study that was previously mentioned, which stated that an increase of fiber correlated with a decrease in waist circumference in men[8]. These variables are closely related to each other and are all affected by diet. A high fiber diet is known to improve these variables (i.e., decrease them), due to its tendency to improve satiety when consuming meals and contain fewer calories even when consuming a higher volume of food[3]. This suggests that consuming a healthy and well-managed diet has the capability of producing desirable results regarding the reduction of weight, BMI, waist circumference, and total body fat percentage.

There was evidence of a decreased GI transit time in response to an increase in fiber in this study. As seen in Table 4, an inverse relationship can be seen in both men and women, meaning that as both groups had an increase in fiber intake per 1,000 kilocalories, a decrease in whole gut transit time (in minutes) was observed (-0.50, p-value: 0.25; -0.77, p-value: 0.07, respectively). This was an expected outcome, due to literature providing evidence of fiber's tendency to shorten transit time in the GI tract[35]. Decreasing transit time within the GI tract is a well-studied factor in reducing colon cancer risk because it can lessen the exposure of carcinogens and other harmful stool components (i.e., bile acids) to the GI region, thus decreasing the risk of GI related cancers such as colorectal cancer[36].

The whole gut transit time variable provided the expected outcome; however, it only provides the overall picture of what occurs within the GI tract during food digestion. The other variables that were included, gastric emptying time, colonic transit time, and bowel transit time, represent other aspects of what occurs within the GI tract. These three additional transit time variables produced varying findings and were not consistent across males and females. Some literature reflects this, as one study concluded that while incorporating more fiber into a daily diet can decrease the components of gastrointestinal transit, addressing the type and form of the fiber can alter the effects[35]. Another study implies a similar conclusion, stating that no difference was found in final transit times after the implementation of 4–6-week high fiber diet (22g fiber/day), however, the participants in this study had undergone total colectomies and ileorectal anastamosis[37]. Perhaps fiber does not have a significant impact on minimizing the overall Gastrointestinal transit time, however, stratifying on more specific mechanisms (i.e., the colonic transit time or large intestine transit time) could show differences.

Strengths of this study include a lack of misclassification bias within the data collection of the anthropometric variables, as strict NHANES guidelines were followed by trained medical staff at the EUHGCRC. This study being a randomized clinical feeding trial is a robust strength. While it is a more costly type of study design, it is the gold standard of study designs and is known to produce the strongest findings when compared to other study designs such as a cohort or case-control study. When looking at the GI tract transit times, the whole gut transit time variable was initially going to be the only examined variable, however it would not have provided enough results to give us a better understanding of how fiber effects this mechanism, therefore, we included other mechanisms that make up the overall GI transit path. Another strength of this study was the aid provided to the study participants to increase diet adherence and overall morale throughout their time on the study. For the duration of the 6-month period, each participant received a weekly call, either from the registered dietician on the study or from a trained research assistant to essentially gather information pertaining to changes in weight, emotions, food intake, etc. These conversations served as an integral part of maintaining relationships with the participants, thus boosting motivation and lessening burn out.

This study has some limitations as well. The sample size of this study was very small, resulting in very few significant findings. Due to the small sample size, results could also be easily skewed if even one value was deemed an outlier. Self-reporting/recall bias is a potential concern, as many of the variables analyzed were self-reported by the participants, including fiber intake, protein intake, trans-fat, and calories consumed. All these variables sourced from the data gathered from the diet recalls conducted through ASA24, and there could be potential for under reporting, due to participants wanting to appear “healthy” for the study.

This is an ongoing study that has a goal of a population size of 60 participants, therefore, much is still to be learned about the associations that were investigated in this study. Future analyses will compare intervention-specific change in outcomes among participants in the two treatment groups.

Chapter 3: Public Health Implications

While this study did not produce robust findings, it provided promising results for favorable effects of a healthy diet on some health outcomes related to obesity and colorectal cancer. The sample included an even distribution of sex and race, important for drawing generalizable results. As previously mentioned, this is an ongoing study and has an expected sample size of about 60 participants with additional power for evaluating associations.

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