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Non-Alcoholic Fatty Liver Disease and Metabolic Syndrome: The Role of Gender Differences and Menopausal Status of Women

Kimberly Kushner
Walden University

Walden University

College of Health Sciences and Public Policy

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Kimberly Kushner

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Review Committee
Dr. W. Sumner Davis, Committee Chairperson, Public Health Faculty
Dr. Harrison Ndetan, Committee Member, Public Health Faculty
Dr. Simone Salandy, University Reviewer, Public Health Faculty

Chief Academic Officer and Provost
Sue Subocz, Ph.D.

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Abstract

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Kimberly Kushner

MS, Colorado State University-Pueblo, 2006

BS, Colorado State University-Pueblo, 2001

Dissertation Submitted in Partial Fulfillment

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Abstract

There are few studies that investigate the association between non-alcoholic fatty liver disease (NAFLD) and metabolic syndrome (MetS) predominantly in menopausal women. This quantitative study investigated the association between MetS and NAFLD while controlling for age, gender, ethnicity, and menopausal status of women. The theoretical framework that helps establish and guide the research was the diffusion of innovation (DOI). A multivariable logistic regression was conducted using secondary data from the National Health and Nutrition Examination Survey. There is a statistically significant association between MetS, and NAFLD after controlling for age (OR = 2.96, 95% CI = 2.319–3.778, $p < 0.001$). However, ethnicity was not statistically significant. After further controlling for gender, the relationship remained statistically significant and exhibited a decrease in risk for females compared to males (OR = -.734, 95%CI = .387–.596, $p < 0.001$). Lastly, focusing on women only, and further controlling for menopausal status, the association between MetS and NAFLD was still statistically significant (OR = 2.227, $p < 0.001$). Mexican Americans, other Hispanics, and Non-Hispanic Black women were at higher risk (95% CI 1.102–7.039, 1.321–5.351, and 1.255–3.013, respectively). Implications for positive social change include adopting proactive and preventative strategies that can improve the quality of life for vulnerable patients and reduce morbidity and mortality associated with MetS and NAFLD.

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Dedication

This dedication is for my parents, who are deseased; they always believed in me and that gave me the drive to continue my education. This is also for my three daughters, my husband, my brother and sister in law, close friends, and the good Lord. I was able to complete my PhD because of the support of my family and friends with the love and support I received. My husband and my girls have also made me work harder for this degree, as they were so encouraging and supportive. I am very sad my parents will not be able to see me graduate with my PhD; it has always been a dream of mine. However, I am still thankful to have my brother to experience this with me, since my parents are no longer with us. I hope to be an inspiration to not only my daughters but also other women in science.

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Table of Contents

List of Tables

Chapter 1: Introduction to the Study

Well before COVID-19 came into the picture, obesity was a major public health concern. It has been well understood that obesity can increase an individual's risk of disease and early death including an increased risk factor to the current pandemic. The prevalence of obesity has increased significantly since 1975, not just in the United States but worldwide (American Heart Association, 2021). It is estimated that obesity has tripled since that time and that in 2016, more than 1.9 billion adults were overweight. Over 650 million of those individuals were put into the obese category. To add to these devastating numbers are the approximately 39 million children under the age of 5 that are overweight or obese that was reported in 2020 (American Diabetes Association, 2021).

In addition, to the increasing rate of obesity, an increase of non-alcoholic fatty liver disease (NAFLD) and metabolic syndrome (MetS) have also gone up. Traditionally, the fatty liver disease had been seen in people who consumed a large amount of alcohol (30 g/day and 20 g/day for women) respectively (Yan-Lan, 2018). However, in recent studies, excessive abdominal fat has been correlated to NAFLD. Furthermore, MetS has been recognized as a contributing factor of hepatic steatosis (Godoy-Matos et al., 2020).

Studies suggest that NAFLD can follow a two-step model that includes a "first hit" and then followed by a "second hit." Hepatic steatosis falls under the umbrella of NAFLD and is called the "first hit"; if fat keeps accumulating it can lead to a "second hit" that is seen in more advanced liver damage. The "second hit" is seen when inflammatory cytokines can lead to fibrosis (Yan-Lan, 2018). Once the liver has accumulated excessive fat in the liver, inflammation will lead to cell death or ballooning, which results in nonalcoholic steatohepatitis (NASH). Over time, NASH will damage the liver by inducing chronic inflammation and hepatocellular ballooning resulting in fibrosis, cirrhosis, liver cancer, and eventually liver failure (Johns Hopkins Medicine, 2021).

However, recently accepted theories have been adopted and purposed to be termed the “multiple-hit model” due to modern technology and new research. The new model is widely used to include the complexity of the disease and how it can be associated with MetS (Yan-Lan, 2018). Studies suggest that NAFLD is a multifactorial disease that may be identified with population screenings to determine those individuals who are more at risk (Yan-Lan, 2018).

NAFLD has been linked not just to obesity but also shares common features of MetS, which can lead to the progression of NASH. Currently there are no pharmaceutical drugs per se that can treat NASH, so lifestyle changes are critical to prevent irreversible damage (BMJ Best Practice, 2021).

Blood tests such as the alanine aminotransferase (ALT) and the aspartate aminotransferase (AST) are effective screening tools along with imaging. An increase in the ALT and AST ratio along with an increased number of MetS factors put the patient at high risk. Transient elastography (TE) is non-invasive imaging that is used for fibrosis evaluation, as a liver biopsy has many downfalls (Godoy-Matos et al., 2020). NAFLD can be reversed if caught early enough since NASH is a chronic progressive disease. Early diagnosis of NAFLD before it progresses to NASH is essential to a positive prognosis (Yan-Lan, 2018).

The liver is the largest internal organ and performs many life-sustaining events per day. Due to the vital function, the liver is important to effectively measure potential hepatic injury correctly. A very effective blood test to assess possible liver damage is the ALT and AST. ALT and AST are very important hepatic enzymes that may indicate hepatic injury and malfunction (Moriles & Azer, 2021). ALT is typically found in the cytosol of liver cells and is detectable in blood at low levels. Damage to the plasma membrane of the hepatocytes due to loss of integrity or necrosis will release ALT into the circulation at a high concentration. The ALT values are subjective, in that they can allow clinicians to identify hepatocellular injury, however, not the cause (Moriles & Azer, 2021).

For example, NAFLD and other diseases, and even medications, can affect the liver, so more diagnostic tests would need to be conducted to narrow down the cause of cellular injury (Moriles & Azer, 2021). Like ALT, AST is normally found in the blood in low concentrations and can indicate liver damage if levels are increased. Typically depending on the lab normal values of ALT are 7-55 units per liter (U/L) and the AST is 8-48 U/L (Moriles & Azer, 2021). Biomarkers like the AST and ALT can be conducted to establish if more invasive testing such as a biopsy is necessary (Lopez-Amador, 2017).

Research indicates that there is evidence that there is a high prevalence of MetS found in those who have NAFLD (Golabi, et al., 2018). There are five conditions identified under MetS, and if an individual is diagnosed with three out of the five, they have MetS. The five conditions of MetS are increased waist circumference (WC), hypertension, high triglycerides, increased fasting glucose, and low high density lipoprotein (HDL) levels (Golabi, et al., 2018). Inadvertently, due to the increase in obesity, the prevalence of MetS will also increase with an increase of NAFLD as well. Overweight and obese individuals are most at risk of developing MetS and NAFLD (Godoy-Matos et al., 2020). Nonetheless, overweight or obesity may not be the only risk factor; weight distribution can have a large impact when it comes to metabolic disease. An individual can look thin and fit on the outside and be metabolically sick, whereas an obese individual may be metabolically sound (Godoy-Matos et al., 2020).

Pathophysiological Driver of Metabolic Disease

The pathophysiological driver in MetS can be linked to abdominal obesity and is widely used to estimate MetS. WC can measure abdominal fat distribution, which has been more reliable in detecting MetS than body mass index (BMI) alone (Godoy-Matos et al., 2020). Furthermore, a study conducted in 2017 followed a cohort for 4.4 years and found that visceral fat was associated (longitudinally) with the incidence of NAFLD (Godoy-Matos et al., 2020).

Abdominal obesity is an accumulation of adipose connective tissue. Adipose connective tissue is made up of specialized cells called adipocytes. These fat cells play an important role in insulation, protection, cushioning, and storage of energy. Many factors influence the distribution of adipose tissue that can be a risk factor for disease (Mittal, 2019). In addition, MetS, NAFLD, and increased adiposity can play a major role in the prediction of these chronic diseases by the areas that it is found in. For example, adipose tissue that is found underneath the skin is referred to as subcutaneous adipose tissue

(SAT) and is not associated with chronic disease (Mittal, 2019). However, visceral adipose tissue (VAT) lines internal organs and drains directly into the hepatic portal circulation. SAT distribution tends to be higher in premenopausal women, whereas VAT tends to be higher in postmenopausal women (Mittal, 2019). There are health risks associated with excess SAT and VAT distributions; however, VAT has a higher morbidity rate. A close relationship exists between VAT and metabolic disease, hepatic steatosis, hypertension, and overall mortality (Mittal, 2019).

Menopausal Women Risk Factors

For menopausal women, these diseases can appear before the patient understands what is going on. Menopause is a normal part of a woman’s life and is also referred to as the change of life. Typically, menopause occurs at around 52 years of age on average. A female is menopausal when her menstrual cycle stops permanently (Office on Women's Health, 2019). However, this does not happen overnight, and a female could be in perimenopause for several years. While all these changes are occurring, one may not see the changes or potential risk factors until it is too late. After menopause, there is an increase in very dangerous chronic diseases such as osteoporosis, heart disease, and stroke (Office on Women's Health, 2019).

Estrogen has long been known to not only protect bones but all internal organs, especially the heart, as it is cardioprotective. The loss of estrogen can increase cholesterol and high blood pressure, which can lead to cardiovascular disease (Cleveland Clinic , 2021). In addition, for reasons unknown women accumulate fat around the abdominal region at an increased rate during perimenopause and post menopause. When a female is in her younger years, fat tends to accumulate in more of a pear-shaped pattern; however, after the loss of estrogen, it changes to an apple pattern. This type of distribution of fat increases the risk of MetS and NAFLD. As estrogen levels start to drop, women can gain excess fat that accumulates in the abdomen. Fat distribution in this manner may suggest a fatty liver. Hormone replacement therapy (HRT) may not be an option for many women but has been found to reduce the risk of cardiovascular disease (Better Health, 2018).

Positive Social Change

Even though this research has a target population of menopausal women, I believe this research can help young women who may have to have a hysterectomy for medical reasons. When a young woman has a hysterectomy, she will experience many of the same signs and symptoms due to the loss of estrogen. There are many medical conditions that may require a hysterectomy, including heavy menstrual cycles that may be caused by endometriosis, untreated pelvic inflammatory disease, polycystic ovary syndrome (PCOS) adenomyosis, prolapsed uterus, and cancer (National Health Service, 2019). A hysterectomy may be the only treatment option if cancer has been detected and the possibility of an early menopause must be acknowledged (NHS, 2019). My hope is that this information can not only help menopausal women, but also help those women who could potentially face early menopause.

Chapter 1 will provide an overview and growing concerns associated with obesity. In this chapter, a full description of chronic diseases linked to obesity will be discussed. The primary focus will be on menopausal women, which will solidify the significance of the study. The literature review Chapter 2 will outline the gap in research regarding menopausal women and obesity. Chapter 1 will also include some key terms such as definitions, limitations, delimitations, and social change. An overview of the problem statement, the purpose of the study, and the research questions will also be provided in Chapter 1.

Background of Study

There have been previous studies that focus on the connection between MetS and NAFLD in both animals and humans. However, in the United States, data suggests that the most neglected individuals include children and menopausal women. Recent studies have suggested that researchers need to understand the natural history of NAFLD and identify factors associated with the progression of the disease (Lindenmeyer & McCullough, 2018). NAFLD is becoming an epidemic and will continue to grow as obesity increases. The evolving understanding of the association between NAFLD and other chronic diseases is crucial. To further understand the hypothesis that NAFLD can be the result of MetS, theoretically, the treatment for MetS could also be the treatment for NAFLD (Lindenmeyer & McCullough, 2018).

Problem Statement

Obesity has become one of the most significant public health risks in the United States. Typically, hightly caloric foods and a sedentary lifestyle can be the cause of this global public health issue (Tiwari & Balasundaram, 2022). In addition, overweight and obese patients add to a growing burden of obesity-related morbidity and mortality, adding to an already overburdened healthcare system (Centers for Disease Control and

Prevention, 2020). Many health concerns may accompany obesity, which can contribute to severe disease and disability (CDC, 2020).

Despite the plethora of information and studies conducted in this field, there are still many gaps that remain. For example, there are very few studies that investigate the association between gender differences, adolescents, and menopausal women in the relationship between obesity and common chronic diseases, such as NAFLD and MetS (Tanaka et al., 2019).

Several growing diseases that have life-threatening consequences include NAFLD and MetS. These chronic and potentially progressive diseases have been linked to obesity, and even more specifically excess abdominal obesity (Tanaka et al., 2019). With the lack of research, irreversible damage can occur within this population, as diagnostic guidelines have not been well studied (Tanaka et al., 2019).

Purpose of Study

The purpose of this study was to investigate the gender differences and the impact of menopausal status of women in the relationship between MetS and NAFLD. In conducting a literature review, it is evident that there are gaps in research about NAFLD and menopausal women, as well as well-defined gender differences. According to El Khoudary et al. (2020), menopause on average happens in women who are 52 years or older. There have been many studies that focus on NAFLD and MetS; however, studies focused on menopausal women are lacking (Lonardo & Suzuki, 2020). There are many changes that happen to a female after menopause that may put them at risk of adverse health outcomes (El Khoudary et al., 2020). Furthermore, with the lack of research menopausal women may not have resources needed to adopt a healthy life.

Early diagnosis of this condition has not been well established, and often patients get the diagnosis when the advanced liver disease has occurred. In this study, elevated liver enzymes AST, ALT, and AST/ALT ratio were analyzed to determine early signs of NAFLD along with special imaging (Tanaka et al., 2019).

In addition, MetS has been associated with NAFLD; therefore, MetS was also analyzed in this study. MetS is defined by the National Institutes of Health as having three or more of the following conditions: large WC, high triglycerides, reduced HDL cholesterol, elevated blood pressure, high microalbumin, and elevated fasting blood glucose level or glucose tolerance (Engin, 2017). A gap in this type of research is identified as women who have experienced their last menstrual period (Tanaka et al., 2019).

Research Questions

RQ1: Is there an association between MetS and NAFLD, controlling for age and ethnicity?

$H_1$1: There is an association between MetS and NAFLD, controlling for age and ethnicity.

$H_0$1: There is no association between MetS and NAFLD, controlling for age and ethnicity.

RQ2: Is there gender difference in the association between MetS and NAFLD, controlling for age and ethnicity?

$H_1$2: There is a gender difference in the association between MetS and NAFLD, controlling for age and ethnicity.

$H_0$2: There is no gender difference in the association between MetS and NAFLD, controlling for age and ethnicity.

RQ3: Does the menopausal status of women affect the relationship between MetS and NAFLD, while controlling for ethnicity?

$H_1$3: Menopausal status of women does affect the relationship between MetS and NAFLD, while controlling for ethnicity.

$H_0$3: Menopausal status of women does not affect the relationship between MetS and NAFLD, while controlling for ethnicity.

Theoretical and Conceptual Framework

Diffusion of innovation (DOI) is a theory that was introduced in 1962 by E. M. Rogers (LaMorte, 2019). The framework can be used to help explain how an idea can gain acceptance and spread over time. For the diffusion of a new process to be successful, the individuals must be able to adopt a new idea or behavior. Changing one's behavior can be a process that will take time, as some people are more receptive to change than others (LaMorte, 2019). The adoption of successful behavior changes can be accelerated by using the DOI. The goal is to have an intervention that will be promoted to people in a specific population or social system (LaMorte, 2019).

Implementing DOI can be successful in helping disseminate obesity prevention. This theory has been used in many different fields to study how people translate new ideas (Shin-Yu & Jiang, 2017). Using this method can be useful in understanding the depth of knowledge an individual may have on treatments of disease. Furthermore, DOI can instill knowledge of disease and how it relates to the real world. Modern technology may be an effective way to incorporate DOI and encourage positive intervention strategies (Shin-Yu & Jiang, 2017).

There are two main parts of the DOI model, which include innovation and diffusion. The innovation portion is new ideas or thoughts, which are useful for the individuals (Shin-Yu & Jiang, 2017). Innovations can have an impact on the methods that are involved in the modification of attitudes and behaviors that may contribute to unhealthy eating. Diffusion is an innovation that is completed within a specific period. Social networking would be an example, as individuals can use social networks to reach certain goals (Shin-Yu & Jiang, 2017). Overall, the DOI theory can encourage social networking as a communication channel to implement an innovation. Educational strategies can be used as well as innovative strategies for obesity prevention (Shin-Yu & Jiang, 2017).

Using social media may encourage research to extend beyond behavior. To see a positive social change the implementation of group activities may provide some beneficial results. For example, joining a group of golfers that walk long distances, gardening, hiking, etc. In turn, people may see physical activity, which is important in weight control, as a fun and exciting activity instead of a non-enjoyable task (Puterbaugh, 2016).

Studies suggest that when people are trying to lose weight, they do better if they are in the company of others who have the same goal. In addition, incorporating enjoyable physical activity can significantly positively affect health. However, if it is not an enjoyable activity, they must do it alone, the outcome may be negative (Puterbaugh, 2016). Social networking may be a way for obese patients to communicate with each other and to find an activity that may be in line with their interest.

Overall, the way to make public health better is to encourage, educate, and support the community. Chronic diseases such as NAFLD and MetS are a result of excessive weight in most cases. If there are tools that can be used, such as social media, to educate and empower people about these chronic diseases, it may have positive outcomes (Puterbaugh, 2016).

Nature of the Study

To answer the RQs, secondary data analysis (SDA) was used with a quantitative research method. SDA will involve investigations on data that has already been collected from a previous study (Ruggiano, 2019). Secondary data tend to have large sample sizes and may be collected over a specific period, giving the researcher the advantage to detect change over time (Ruggiano, 2019).

The variables that will be used and analyzed are MetS (WC, reduced HDL, increased blood pressure, elevated blood sugar, and high triglycerides), all these variables can be found in the codebooks. Furthermore, the data to determine NAFLD will be the WC, TE, elevated ALT, and AST levels. Gender, ethnicity, and age are also available in the data sets.

The variables that are included in the codebook were recoded to reflect MetS, NAFLD, and gender as dichotomous, age as ordinal, and ethnicity as nominal. Regular descriptive statistics were analyzed along with a multiple binary logistic regression model. Quantitative research focused on data that are in numerical form.

Definitions of Terms

Adipose connective tissue: Adipose tissue is a loose connective tissue that serves as a cushion and insulation throughout the body (Mittal, 2019).

Alanine transaminase (ALT): ALT is also an enzyme that provides energy to the hepatocytes. ALT is also found circulating in the blood, however, when the liver is damaged the ALT will be released into the blood at high levels (Mayo Clinic, 2021). The upper limit of normal for ALT is 55 IU/L (Mayo Clinic, 2021).

Aspartate aminotransferase (AST): AST can also be called serum glutamic-oxaloacetic transaminase (SGOT). AST is a very important enzyme that helps metabolize amino acids. AST can be found in the blood at low levels; however, if elevated it may indicate liver damage (Mayo Clinic, 2021). AST can be found in other organs as well, such as the heart. The upper limit of normal for AST is 48 IU/L (Mayo Clinic, 2021).

Blood pressure (BP): According to the American Heart Association (AHA), blood pressure that consistently measures 130-139 systolic and 80-89mm Hg diastolic has Stage 1 hypertension (AHA, 2020).

Body mass index (BMI): BMI is a screening tool that is used to determine body fatness (CDC, 2020). BMI is calculated by an individual's weight in kilograms divided by the square of height in meters. The values are BMI less than 18.5 is underweight, BMI 18.5-24.9 normal/healthy weight, BMI 25.0-29.9 is overweight, and a BMI of 30.0 or higher is considered obese (CDC, 2020).

Fasting glucose: After a patient has fasted overnight, a blood test will be drawn to measure blood glucose levels in the blood. A value of 100-125 mg/dl is an indication of prediabetes, and 126 ml/dl or above indicates diabetes (CDC, 2021).

Hepatic steatosis: Modulated by visceral fat accumulation, hepatic steatosis usually occurs in the setting of MetS (Lindenmeyer & McCullough, 2018).

High-density lipoprotein (HDL): HDL is a protein carrier of lipid-based molecules and carries it to the liver. High levels of HDL indicate a decreased risk of heart disease and stroke. The normal adult range for HDL is 45-70 mg/dl for men and 50-90 mg/dl for women (University of Rochester Medical Center, 2021).

Menopause: Menopause is a natural and normal process of aging for women. Menopause is when a female has not had a menstrual cycle in 12 months. This transition can happen usually in her 40s and 50s; however, the average age is 51 (National Institute on Aging, 2021).

Metabolic syndrome (MetS): Metabolic syndrome is characterized by five disorders: abdominal obesity (waist circumference), hypertension, high triglycerides, high fasting glucose, and decreased HDL levels.

Nonalcoholic fatty liver disease (NAFLD): NAFLD is the buildup of excessive fat in the liver with the absence of alcohol. NAFLD is evident by a histological or radiological examination (Lindenmeyer & McCullough, 2018).

Nonalcoholic steatohepatitis (NASH): NASH is chronic inflammation that builds up and causes liver cell damage that can result in fibrosis. Furthermore, there is liver cell injury which presents as hepatocyte ballooning (Lindenmeyer & McCullough, 2018).

Subcutaneous adipose tissue (SAT): SAT is adipose tissue beneath the skin (Mittal, 2019).

Transient elastography (TE): TE is imaging that is non-invasive for the evaluation of liver fibrosis.

Triglycerides: Triglycerides are measured in the blood in milligrams per deciliter (mg/dl). If an individual has triglyceride levels above 150 mg/dl, they have an increased risk for chronic heart disease (Medline Plus, 2021).

Visceral adipose tissue (VAT): VAT is adipose tissue lining internal organs (Mittal, 2019).

Waist circumference (WC): According to the Centers for Disease and Prevention (CDC), WC can be another way to estimate severe disease. If a male has a WC of more than 40 inches and a non-pregnant female has a WC of more than 35 inches, he/she has an increased risk of disease (CDC, 2020).

Assumptions

Because secondary data were used in the study, the assumption is the that the data was collected correctly. To analyze laboratory data quality assurance is valuable, so all can benefit from high-quality laboratory clinical data (Hajia, 2019).

The data that were used in this research were obtained from the National Health and Nutrition Examination Survey (NHANES 2017-2018). The laboratory team included three American Society of Clinical Pathologists, medical laboratory technicians, and a certified phlebotomist (NHANES, 2018). On the NHANES website, there is a full discloser of the policies and procedures that are used to collect the laboratory results.

Included in the policies and procedures are the laboratory work stations, equipment used, how blood and other values were collected, training, etc.

Scope and Delimitations

According to the North American Menopause Society (NAMS), the average female will go through menopause at approximately 52 years of age (The North American Menopause Society, 2021). Therefore, women who are younger than 52 years of age were excluded from this study when analyzing menopausal status.

Individuals that were selected to answer the research questions had an elevated BMI and had three of the five conditions required for a diagnosis of MetS. In addition, these individuals had to be 52 years or older, elevated liver enzymes, WC of over 35", and a TE for evaluation of fibrosis in the liver.

Limitations

How well results represent true findings in research when comparing an outcome between the test subjects and individuals not included in the study its validity. Validity applies to many different types of studies including associations, interventions, diagnosis, etc. The extent to which the observed results do represent the truth in the population of interest is considered internal validity (Delahanty et al., 2016).

Limitations that were anticipated early on was the strength of the study. Thankfully, that was alleviated by constructing a complex data plan. This type of data plan will ensure the true population is represented. The data contains a large sample size of individuals, and it is imperative that the information that was provided was accurate. A lifestyle change may translate into something different for each person (Delahanty et al., 2016). When obesity is the main focus of a modifiable risk factor for MetS, the causes may be difficult to determine. For example, obesity can co-occur with many other risk factors, so reducing BMI may not reduce the risk factors associated with obesity (Delahanty et al., 2016). Some individuals have a high BMI and remain metabolically healthy, which may contradict that obesity is a risk factor for metabolic disease (Franks & Atabaki-Pasdar, 2016).

Contradictions in a study may lead to potential confounders, which may result from a bias that is introduced when the accuracy of participants' answers may lead to false or biased information (Franks & Atabaki-Pasdar, 2016). Confounders can imply that there is a non-causal relationship between abdominal obesity and metabolic disease. There may be a weakness in questionnaires that assess lifestyle or diet, which can lead to bias in that some participants may be educated about diet and not answer the questions accurately (Franks & Atabaki-Pasdar, 2016).

Internal validity can show that this is a true representation and not due to methodological errors (Patino & Ferreira, 2018). The internal validity of this data suggests that the data has been collected in a manner that indicates it is accurate (Patino & Ferreira, 2018). The focus was to demonstrate if there is an association between MetS and NAFLD. Once internal validity had been accomplished, I was able to proceed with the external validity of the study (Patino & Ferreira, 2018).

The external validity allowed me to determine that the results may be generalizable to other populations. In this case, I evaluated if the results from the study appllied to all genders respectively (Patino & Ferreira, 2018).

Significance

In 1980, pathologist Jurgen Ludwig suggested that NASH was observed in patients without a drinking addiction. The histological findings showed that there were similar disease outcomes that were mainly found in alcoholic steatohepatitis (Tanaka et al., 2019). These findings suggested that there were fatty changes that lead to hepatocyte necrosis, ballooned hepatocytes, lobular inflammation, and perisinusoidal fibrosis (Tanaka et al., 2019). Typically, patients that were diagnosed with this condition had diabetes, hypertension, and obesity to mention a few. In many cases, NAFLD can progress to liver cirrhosis and eventually hepatocellular carcinoma (Tanaka et al., 2019).

NAFLD is highly associated with visceral obesity and can be the result of MetS, which includes insulin resistance (IR) and diabetes. There is still some debate on IR and a high sugar diet contributing to NAFLD, as a prospective study was conducted (Tanaka et al., 2019). Therefore, if abdominal obesity can be predictive of NAFLD, a more robust intervention strategy can be designed and implemented (Tanaka et al., 2019). Some studies suggest that groups that share the same goals may help encourage physical activity and healthy eating. Often, when an individual has support from others who are in the same situation, they can support and encourage positive social change. For example, joining a group that has a community garden would offer an enjoyable physical activity that an individual would stick to and want to participate in (Puterbaugh, 2016). Many of these groups exist on social media and can be a very powerful tool in incorporating physical activity into the lives of these individuals.

Overall, the significance of menopausal women who have increased abdominal obesity are at risk of NAFLD and may produce a risk for liver dysfunction that may lead to liver carcinoma. In contrast, adults will face the same challenges and may require liver transplants later on in life. Checking liver enzymes AST and ALT can be good indicators of overall liver function (Tanaka et al., 2019). These important laboratory tests can show any potential damage to the liver that may be leading to NAFLD. AST and ALT are abnormal liver function tests that help diagnose NAFLD. Elevated levels of these laboratory findings along with fibrosis markers and imaging exams lead to an accurate diagnosis (Tanaka et al., 2019). Furthermore, IR can be the main contributor to NAFLD as well as prediabetes (Tanaka et al., 2019).

Prediabetes is a condition where an individual has high blood glucose levels and is associated with MetS; however, they are not high enough to be diagnosed with Type-II Diabetes. The CDC estimates that there are approximately 88 million people who have prediabetes (CDC, 2020). Further, the CDC suggests that people with prediabetes can alter lifestyle choices to prevent Type-II diabetes. Detecting patients who have elevated liver function tests and prediabetes may be able to make lifestyle changes that could positively impact their health in the future. NAFLD and prediabetes need to be diagnosed early with those patients with central obesity to implement a healthier lifestyle.

Summary

In Chapter 1, I provided an introduction and background regarding abdominal obesity and the link to NAFLD. The focus of this study was to see if there is an association between NAFLD and menopause, as many times weight increase can occur.

To study NAFLD, it is important to measure the events that take place that leads to liver disease. Obesity and metabolic disease have been associated with NAFLD and have been shown to increase in women after menopause.

Obesity and all the complications associated with obesity, such as NAFLD, have been recognized as a public health concern. Furthermore, women who gain weight, especially in the abdomen, after menopause are at a high risk of chronic disease. Certain tests can be conducted to predict many of these health concerns promptly, so lifestyle changes can be implemented. Blood tests such as the AST and ALT are good indicators of potential liver damage. Regarding metabolic health, other non-invasive exams may also provide helpful information in determining the risk of NAFLD. These non-invasive exams include WC, blood pressure (BP), LDL and HDL levels, and fasting blood glucose levels. In contrast, special ultrasound technology, as seen in TE, can also guide practitioners in the degree of fibrosis in the liver. In Chapter 2, I will provide current literature on obesity and MetS as it relates to NAFLD.

Chapter 2: Literature Review

In Chapter 2, I will provide an assessment of the literature gathered regarding NAFLD and menopausal women. Research does not address the association between NAFLD and menopause. The research has shown little information in the assessment of risk factors of disease after menopause.

Ross et al. (2020) argued that BMI is not sufficient to assess the cardiometabolic risk associated with central obesity. Researchers in this study tryed to identify the gaps in the knowledge of WC decrease the possibility of chronic disease. The authors suggested that WC can refine adverse health risks than BMI alone. Some gaps remain in the refinement of WC threshold values in regards to age, sex, and ethnicity.

Lonardo & Suzuki (2020) researched sexual dimorphism in human diseases mainly focusing on NAFLD. The researcher aimed at discussing the sex differences in NAFLD to provide information that is needed for clinical treatment. The author suggested a potential research gap in menopausal women and adolescents.

Choi et al. (2019) studied BMI and WC to measure obesity and abdominal obesity. The researchers were focusing on the effects of abdominal obesity and how it puts an individual at risk for major adverse cardiac events (MACE). The study concluded that those with abdominal obesity were more at risk for MACE than those who were generally obese.

Chen et al. (2019) looked at obesity and abdominal obesity among individuals in China, abdominal obesity was measured using BMI and WC. The scientist concluded that abdominal obesity has increased significantly especially in males, which puts them at greater risk of chronic disease.

Tantanavipas et al. (2019) did a study on polycystic ovary syndrome (PCOS) and NAFLD due to the similar clinical presentation both diseases show. The clinical presentations include obesity, IR and MetS. In this study, researchers found that the WC was a predictive factor that can determine NAFLD for women with PCOS and those women that were healthy. Future research may need to determine steatosis staging. There were some disagreements concerning the steatosis stage, as the United States has limitations when fat accumulation is greater than 20%.

Yang et al. (2016) wanted to determine the association between NAFLD and MetS. Researchers determined that NAFLD is an emerging chronic disease that may lead to hepatocellular carcinoma and cirrhosis. One of the main contributing factors indicated that this chronic liver disease is closely associated with obesity and cardiovascular disease. A statistical linear relationship was observed between NAFLD and waist circumference. One of the limitations of this study was the authors could not establish a temporal association between NAFLD and MetS due to the cross-sectional design.

Young et al. (2020) researchers looked at lean NAFLD (normal BMI) vs. obese patients and found more severe and abnormal lab results in those who are obese. Even though lean patients had NAFLD they had less severe biomarkers for inflammation, less hepatocyte ballooning, and fewer metabolic abnormalities. One limitation is that the data overrepresented studies from Asia.

Yue et al. (2019) were interested in cholecystectomy vs. central obesity or IR as it relates to the risk of NAFLD. In the researchers' findings, cholecystectomy and NAFLD do share some of the same etiologies, such as abdominal obesity and insulin resistance. Limitations that the authors pointed out were the use of ultrasound to detect NAFLD and the lack of information that pertains to residual confounding variables.

Lin et al. (2021) wanted to look at the association among MetS and obesity-related indices with NAFLD. The authors noted that there was a difference in these morbidities between men and women. However, the population they studied lived in southern Taiwan and was not generalized to other populations.

In research conducted by Popa et al. (2016), 2,681 subjects aged 20-79 were classified into cardiometabolic phenotypes of the Romanian population-based on BMI. Participants had to have the presence of abdominal obesity and MetS. There is a high prevalence of abdominal obesity and MetS in the Romanian population and an association with kidney function and cardiometabolic factors (Popa, 2016).

Sigit et al. (2020) conducted a cross-sectional study of middle-aged adults with a sample size of 10,575 men and women (Indonesian and Dutch) diagnosed with MetS with specific BMI and WC measures. Data obtained from this Indonesian National health Surveillance and the Netherlands Epidemiology of Obesity Study Prevalence of MetS was 28% and 46% in Indonesian men and women, and 36% and 24% in Dutch men and women. The most prominent components were abdominal obesity in Dutchmen and more Indonesian women than men have MetS.

In a meta-analysis conducted by, Ping et al. (2018), anthropometric indices was conducted to measure SAT and VAT. The authors found that WC could be a good indicator for estimating VAT and SAT. There were 29 publications included in this meta-analysis, and the results stated that the values of SAT and VAT differed in many areas. For example, they did find statistical significance in patients with advanced age, ethnicities, and body shapes (Ping, et al., 2018).

Studies suggest that hormonal changes acquired after menopause may contribute to excess abdominal fat. Excess abdominal fat can lead to NAFLD. NAFLD can contribute to a set of comorbidities for those women who have gone through menopause. Venetsanaki & Polyzos (2019) found that women who were deficient in estrogen tend to have more severe cases of NASH. In addition, there have been limited studies done to show if HRT had any effect on NAFLD. NAFLD increased the risk of Type 2 diabetes and cardiovascular disease. The focus of the study conducted by Venetsanaki et al. was to determine possible therapeutic perspectives for women who get NAFLD after menopause (Venetsanaki & Polyzos, 2019).

Setroane et al. (2020) studied the prevalence of NAFLD and MetS in premenopausal and postmenopausal women in Ho Municipality. In this cross-sectional study, researchers took 88 premenopausal and 97 postmenopausal women who were patients at Ho Teaching hospital and Ho Municipal Hospital from November 2018 to January 2020. Blood samples, measurements, and other examination procedures were conducted to determine the health of the women and to establish a baseline of values.

Setroane et al. (2020) found that MetS and NAFLD were more prevalent in postmenopausal women than in premenopausal women.

In a study conducted by Da-Zhi et al. (2018) in Eastern China, researchers examined the potential effects that MetS and NAFLD had on menopausal women regarding osteoporosis. Osteoporosis is a very common skeletal disease that decreases bone density leaving the patient more vulnerable to bone fractures. Regardless of gender, NAFLD has been linked to an increased risk factor for the development of osteoporosis. In addition, it was also observed that individuals that had MetS also had an increased risk of osteoporosis. The one determinate factor that stood out was women with both conditions had a higher prevalence of osteoporosis after menopause. Ideally, that makes sense because estrogen is known to be bone protective, so to add insult to injury NAFLD and MetS come into the picture. The study concluded that women who have both NAFLD and MetS should be closely monitored for osteoporosis, as both are independent risk factors (Da-Zhi et al., 2018).

Chen et al. (2019) performed a comparative cross-sectional study in an urban population in China that had liver and metabolic disease. A new term that has been adopted by many is metabolic associated fatty liver disease (MAFLD), which is liver disease associated with metabolic dysfunction. Males and females were enrolled in the study with a close eye on menopausal women. In the population studied, the prevalence of MAFLD increased for women in the 18-49 age group. However, females over 50 had a significant increase in MAFLD, unlike their men counterparts. In men, the older they got, the least amount of MAFLD was seen, while in the younger age groups there was a high prevalence of MAFLD. This study also found that women who do not produce estrogen any longer were at an increased risk of developing MNAFLD (Yu-ling, et al., 2021).

Literature Search Criteria

Several databases were used in the literature review search criteria. I used the research questions and anatomy and physiology terms as a guide to peer reviewed articles and websites. Many of the sources I acquired came for the Walden Library and Google Scholar. The key words and phrases that were used include *waist circumference, abdominal obesity, metabolic syndrome, AST levels, ALT levels, blood pressure, body mass index, menopause, menopause and obesity, laboratory blood values, normal range of fasting glucose levels, age of menopause, hypoglycemia, insulin resistance, metabolically sick individuals, liver ultrasound exams, NASH, NASH values, NAFLD and central obesity, adiposity, NAFLD risk factors, causes of NAFLD,* and *women and NAFLD.*

The main databases used in the search of sources included PubMed, MEDLINE, NCBI, NHANES, CDC, Cochrane library, EMBASE, Federal and local health agencies, and CINAHL. The literature used provided evidence-based studies that focused on the prevalence of MetS as it relates to NAFLD. There are several studies that show the increased risk factor for MetS and NAFLD when a high WC was observed. Furthermore, some of the studies suggest that blood liver enzyme levels can be a very resourceful tool in determining the potential degree of damage to the liver.

I provided an overview of NAFLD, MetS, and the criteria that must be met to be diagnosed with these diseases. In addition, I provided an overview of specific blood tests that can give practitioners an indication of disease. The laboratory tests that will be discussed include AST, ALT, fasting blood glucose, lipid panel. The AST and ALT will be used to determine liver disease and the fasting blood glucose and lipid panel will aid in the diagnosis of MetS. In addition, measurements of the WC and blood pressure (BP) will also be analyzed. Ultrasound technology values will be in place of a liver biopsy, as data have not been provided for biopsies in the data obtained.

NAFLD

NAFLD has become a common complication of an array of conditions in where fat accumulates in the liver. It has been estimated that one in three adults and one in 10 children in the United States have this disorder. Typically, a fatty liver was seen in people who consumed excessive amounts of alcohol. However, over time there have been high numbers of this same condition seen in individuals that did not have a history of alcohol use including children (Jakhete et al., 2020).

Some individuals can go on to develop a more severe condition called NASH. NASH is a chronic condition in where the excess fat accumulation and inflammation leads to scarring and cirrhosis of the liver. When the hepatocytes are replaced with scar tissue due to cirrhosis, the liver cannot work properly, therefore requiring a liver transplant (Jakhete et al., 2020).

Cause of NAFLD

Obesity is one of the most common risk factors associated with NAFLD. Abdominal adiposity is a more specific risk factor, due to the distribution of fat. In addition to obesity, another condition associated with NAFLD is a condition known as

MetS (Jakhete et al., 2020). MetS is part of metabolic disease that includes IR, overweight or obesity, increased levels of LDL, decreased levels of HDL, and high BP. A patient must be diagnosed with three of the five conditions above to have MetS (Jakhete et al., 2020).

NAFLD Diagnosis

Medical professionals have many tests at their disposal to diagnose NAFLD. Furthermore, due to the advancement in medical technology, NASH can also be diagnosed, as it can also present with NAFLD. A patient history is a list of medical conditions that would make the patient more likely to develop NAFLD. Included in the history the practitioner is concerned when there is a history of being overweight or obese and MetS (National Institute of Diabetes and Digestive and Kidney Diseases, 2021). A physical exam will also be performed to check for a potential enlarged liver and other signs of MetS such as darkened skin over the knuckles, elbows, and knees (NIDDKD, 2021).

Laboratory tests are also used in the diagnosis of NAFLD. Two of the important laboratory tests that are performed include elevated enzymes of the ALT and AST. The upper limit of normal for ALT is 55 IU/L and the upper limit of normal for AST is 48 IU/L (Mayo Clinic, 2021). In addition, imaging test are also typically ordered to determine the amount of fat in the liver. Imaging testing is also very important because it can determine if the patient has NAFLD or NASH (NIDDKD, 2021).

TE is the imaging that is used to measure the amount of thickening or scarring that can occur in the liver. The TE that was used in this study was done by the FibroScan.

The FibroScan uses kiloPascal's (kPa) to measure the thickness of the liver and ranges from 2 to 75. An individual that falls into the 2 to 7 kPa range is within normal range, with the average 5.3 kPa. There are 4 stages of scaring that can occur with includes F0 = No scarring, F1 = mild fibrosis, F2 = moderate fibrosis, F3 = severe fibrosis, and F4 = cirrhosis or advanced fibrosis (MedlinePlus, 2021).

Metabolic Syndrome

MetS has become a common syndrome that is seen in many patients due to the increase of obesity. It has been suggested that MetS may far surpass smoking for the leading risk factor for cardiovascular disease and other chronic illnesses.

IR tends to increase the likelihood of developing MetS. When a patient has IR, the body is not able to use insulin correctly and this can lead to high blood glucose levels. IR is closely linked to obesity, among other risk factors such as genetics (National Heart, Lung, and Blood Institute, 2020).

MetS Diagnosis

There are five conditions that make up MetS. A patient is diagnosed with MetS if they have three of the five conditions, which include high blood glucose, low levels of HDL, high levels of triglycerides, large WC, and high BP (AHA, 2021).

Blood Glucose

Glucose is a source of energy that can come from the food you eat, or the body can produce in times of need. Insulin is the hormone that is produced in the beta cells in the pancreas that carries the glucose into the cells, so it can be used to produce energy. A peptide hormone, insulin, is the regulator for carbohydrate, lipid, and protein metabolism.

Insulin will maintain glucose homeostasis by facilitating cellular glucose uptake. For the purpose of this study, I will focus on IR that is a result of type 2 diabetes mellitus (T2DM) and is also linked to MetS (ADA, 2021).

When the body is exposed to sugar and high carbohydrate diets day in and day out, insulin can become less effective. For example, if the cells are already full of so much sugar, because they can only take so much, then the excess will be sent to the liver (ADA, 2021). The liver can also only hold onto so much glucose before it is turned into triglycerides. After time, the liver is so consumed with fat that it will eventually spill over into the abdominal cavity. All these events contribute to MetS and therefore can be linked to not only T2DM but NAFLD as well (ADA, 2021). According to the American Diabetes Association, a fasting plasma glucose (FPG) should be less than 100 mg/dl to be in normal range. Before the blood is drawn the patient is instructed to fast for 8 hours and may only have water to drink. A diagnosis of diabetes is concluded when a FPG is greater than or equal to 126 mg/dl (ADA, 2021).

Triglycerides

Typically, when the word triglyceride is mentioned good cholesterol, bad cholesterol, and heart disease follow. The most common type of fat found in the body is triglycerides and can be measured in the blood. There are two values that are very important to health and that is High-density lipoprotein cholesterol (HDL) and low-density lipoprotein (LDL). In fact, these are not cholesterol themselves but a lipoprotein that carries the cholesterol (AHA, 2021).

Even though these molecules are not cholesterol molecules they are still referred to as the good and bad cholesterol. LDL is considered the "bad" cholesterol because it carries cholesterol into the arteries. In contrast, the HDL is called the "good" cholesterol because it carries the cholesterol into the liver and out of the arteries. If a patient has elevated triglycerides (150 mg/dL) combined with low HDL and high LDL levels, it is a risk factor for chronic disease (AHA, 2021). A risk factor for chronic disease including MetS is an HDL level that is less than 40 mg/dL for males and less than 50 mg/dL in females (AHA, 2021).

Waist Circumference

Abnormal WC is an important risk factor that can determine the distribution of adipose connective tissue. The WC can be obtained in a clinical setting and is an anthropometric measurement of obesity. Typically, individuals who carry more fat in the abdominal region and carry an "apple shape" distribution of fat have an increased risk for MetS and NAFLD. In many cases obesity, even more specific abdominal obesity, usually coexists with MetS and NAFLD (Jiang et al., 2018).

An increased WC has been an essential component of determining the risk of chronic disease. It has been observed in many studies that even individuals with a normal BMI are at risk of disease if they have abnormal abdominal fat. Risk for chronic disease increases with a WC more than 40 inches in males and 35inches in females (Jiang, et al., 2018).

High Blood Pressure

Hypertension, or high BP is another risk factor that falls under MetS. Many individuals that have MetS have hypertension, due to the high sugar content in the blood. BP is taken by using a sphygmomanometer which compresses the brachial artery, therefore, leading to a measurement. BP is reported in two numbers systolic or top number and diastolic or bottom number (AHA, 2016). The significance of the systolic is the pressure exerted against the artery wall with each heartbeat, and diastolic is the reading between heart beats when the heart is at rest (AHA, 2016)

According to the American Heart Association, hypertension is a value greater than a systolic value of 130 mmHg and a value greater than 85 mmHg for diastolic blood pressure. The AMA has listed a systolic between 130-139 mm/Hg or a diastolic 80-89 mm/Hg hypertension stage 1. Stage 2 hypertension is systolic 140 mm/Hg or diastolic 90 or higher, and values above these is a hypertensive crisis and an emergency (AHA, 2016).

The number of readings before a diagnosis of hypertension has been up for debate. However, the AHA recommends at least three readings on different days to establish a true reading. Recently, it has been suggested that home monitoring also be implemented before a treatment plan due to recent studies (Bello et al., 2018). A study that was recently released from the Journal of the AHA, found that using readings from the evening, at least three different days indicated a reliable blood pressure reading (Bello et al., 2018). In this study, however, the patients were instructed to take the readings at home, subsequently the data that will be used in this research study was conducted in the laboratory.

Link Between NAFLD and MetS

I presented NAFLD and MetS individually, now I will explain the link between the two potentially fatal conditions. The defining criteria for MetS was listed above, however, NAFLD is not one of those criteria or conditions. NAFLD can be a common liver manifestation because of MetS (Lin et al., 2021). There are countless studies that have successfully shown the association between MetS and NAFLD, it has been suggested to change the name to metabolic dysfunction-associated fatty liver disease (MAFLD). However, there is further research needed to figure out how the name change impacts clinical hepatic outcomes (Lin et al., 2021).

Typically, the fat accumulation that occurs in NAFLD is due to metabolic dysregulation and is usually implicated in patients who are overweight or obese. After many years of eating over processed and high carbohydrate foods there can be increased inflammation and oxidative stress that can lead to mitochondrial dysfunction (Lin et al., 2021).

According to the American Liver Foundation, approximately 25 percent of adults in the United States have NAFLD, and an estimated 20 percent of those individuals have NASH (American Liver Foundation, 2021). When analyzing the prevalence of MetS due to the increased cases of obesity it is no surprise that the cases increased in men and women. However, there was a significant increase seen in women between 2011-2016 from 31.7% to 36.6%, adults ages 20-39 from 16.2% to 21.3%) and Hispanics from 32.9% to 40.4% (Shmerling, 2020).

Menopause

The cessation of menstrual cycles (amenorrhea) for a full year indicates a female has transitioned from a reproductive to a nonreproductive chapter in her life also referred to as menopause (El Khoudary et al., 2020). During this normal process of ageing there are significant changes in hormones, metabolism, physiological changes, and mental challenges to mention a few. The average age for a woman to go through menopause is approximately 52 years of age and is premature if menopause happens before 40 years of age. There are many changes that can be detrimental to health during this phase of life, and in many cases many females will spend 40-45% of their life in a postmenopausal phase (El Khoudary et al., 2020).

The ovaries make many different forms of estrogen, which includes estrone (E1), 17α- and 17β-estradiol (E2), and estriol (E3). The most common and robust form of estrogen for a female is 17β-estradiol (Marongiu, 2019). As the ovaries transition in the decline of producing and secreting hormones, there is also a decrease in the amount of estrogen receptors (ER). The decrease in ER can induce irregular menstrual cycles and inconsistencies in estrogen levels that can lead to unpleasant symptoms (Marongiu, 2019). Menopause doesn't happen overnight so a woman can be in perimenopause for up to 10 years, which can start the biological changes that may lead to unfavorable outcomes in menopause (Marongiu, 2019).

Some of the hormonal changes a woman can experience during this transition is decrease levels of estradiol and follicle-stimulating hormone (FSH). Decline of these hormones, especially estradiol, present many negative symptoms such as mood changes, vasomotor disturbances, sleep deprivation, weight gain, metabolic disturbances etc. (El Khoudary et al., 2020).

It has been well documented that these hormonal changes can contribute to the risk of chronic progressive diseases. Studies have shown that the hormone changes may lead to metabolic alterations, increase in body composition, development of inflammation, visceral adiposity, etc. (El Khoudary et al., 2020). Due to these changes and the complications implemented there needs to be better screening and intervention strategies at this stage of life.

Menopause and the Link to NAFLD, and MetS

Metabolic changes do occur after menopause due to the lack of estrogen after menopause. According to John Hopkins there are a group of estrogens that regulate normal sexual and reproductive development made by the ovaries, small amounts may also come from the adrenal glands and adipose cells (JHM, 2022). However, that is far from all that estrogen is important for in the female body (JHM, 2022). Estrogen effects so much more than just the reproductive organs, which is not stressed enough in many studies (JHM, 2022). Estrogens are very important for providing protection and efficiency in the urinary tract, cardiovascular system (heart and blood vessels), breasts, skin, mucous membranes, brain, pelvic muscles etc. Estrogen also has a huge influence on major organ systems such as the musculoskeletal, cardiovascular, and the nervous system (JHM, 2022).

A recent study released by the Translational Genomics Research Institute (TGen), suggested that women who are post-menopausal is at a higher risk of NAFLD, that can potentially lead to NASH. This puts an added burden on the healthcare system that already has an estimated $292 billion annual cost. It is estimated that more than 100 million U.S. citizens will develop NAFLD in the next decade. The estimates provided does not include long term disabilities, loss of work and productivity, and potential liver transplant (The Translational Genomics Research Institute, 2020). NASH is the leading cause of liver transplants among women indicating many women have NASH instead of NAFLD and these numbers are expected to rise. It is expected that the cases of NAFLD will increase to 27 million cases by 2030 (TGRI, 2020). This recent study stressed that there is a critical gap in research that is focused on post-menopausal women and the risk of NAFLD.

Individuals, who have NAFLD, have the potential to progress into NASH, which can cause cirrhosis, and then eventually lead to hepatocellular carcinoma (Robeva et al., 2021). Research that was conducted in animals showed that postmenopausal females had more prevalence of NAFLD, and that cirrhosis and hepatocellular carcinoma was more progressive in this stage of life (Robeva et al., 2021).

The loss of estrogen after menopause is called the hypoestrogenic period, which can cause a metabolic imbalance that can enable the onset of MetS. The onset of MetS can lead to NAFLD causing dyslipidemia and IR which in turn increases adipose tissue to accumulate in the liver. The devastating effects of NAFLD can ultimately lead to NASH, cirrhosis, and carcinoma (Robeva et al., 2021).

Lack of estrogen may encourage visceral adiposity, due to alteration of metabolism, which can lead to metabolic disturbances and NAFLD. One of the main reasons NAFLD is prevalent in post-menopausal women is because a specific estrogen called estradiol, which is produced by the ovaries, before menopause is responsible for glucose and lipid metabolism. Furthermore, stimulation of glycogen synthesis instead of lipogenesis or gluconeogenesis is also conducted in the presence of estradiol (Robeva et al., 2021). Lipogenesis and gluconeogenesis can be a main contributor to the buildup of fat in the liver, as it serves as a storage container (Robeva et al., 2021).

In addition to protecting organs and metabolism, estradiol has anti-inflammatory effects which can decrease atherosclerosis accumulating in the vessels. Many studies have shown that consistent high levels of inflammation can lead to many chronic diseases such as cardiovascular and liver disease. Too much inflammation over time can cause buildup of plaques in the arteries, and excess fatty acids in the liver (Robeva et al., 2021).

Furthermore, in addition to the hypoestrogenic phase of menopause, there are other hormonal factors that may be involved in the progression of MetS and NAFLD. A condition called hyperandrogenemia was seen in females that suffers from PCOS also had an increased risk of NAFLD. Nonetheless, women that had NAFLD and were postmenopausal, did have an increase of testosterone in comparison to those females that were postmenopausal and no diagnosis of NAFLD (Robeva et al., 2021). The association of testosterone and its role in facilitating NAFLD has not been well studied, as it is difficult to extrapolate pre- and post-menopausal testosterone levels. With that said this research study is extremely important as it addresses MetS and NAFLD plus the associated complications in the context of sex differences (Robeva et al., 2021).

Theoretical Framework

The need to incorporate research into clinical care is built on evidence-based practices (EBP). The strong influence of EBP is the evidence presented is superior as well as the best clinical experience with both family and patient values. Using this strategy is very important because it involves and allows patients to contribute in his/her care (Stewart et al., 2019). In addition, the new research that comes out, which can be often, can be implemented as well.

Throughout time, there have been certain frameworks that support the use of EBP's (Stewart et al., 2019). The classical theory that has influenced incorporating science into the clinical settings is diffusions of innovations (DOI). Five decades ago, the DOI theory was developed by Everett Rogers, which explains factors that may influence the adoption of innovations. Individuals were put into categories in reference to how quickly the new ideas were incorporated (Stewart et al., 2019).

The idea of using DOI is to incorporate, and idea and it will eventually diffuse through a targeted population. In the case of this study the target population would be menopausal women, or better yet peri-menopausal women (LaMorte, 2019). The goal is to have selected women adopt a new idea or behavior to prevent common disease seen in the population in question. The hope is that menopausal women will adopt specific screenings and lifestyle changes as a part of a social system. However, the adoption of a new idea may not be as receptive to some individuals (LaMorte, 2019). For example, it is important to eat healthy and exercise at any age, however, when women lose estrogen, it is essential. In addition, there are many annual tests and screenings that should be done regardless. Now, the question is, will the targeted population have specific characteristics that would prevent change? Turns out that there are some people in general who are more open to change than others (LaMorte, 2019).

It has been suggested that to fill in the gaps of DOI may be to implement the social networks theory (SNT). Combining SNT with DOI can alleviate some potential biases found in the DOI framework when introducing interventions (Iqbal & Zahidie, 2021). In many cases, there is a pro-innovation and a potential individual blame bias that can happen in using DOI. DOI does not consider some of the influences of culture and society regarding individual change. In addition, SNT may help fill in the gaps by explaining behavior change that is based on the influence of social networks (Iqbal & Zahidie, 2021). Both DOI and SNT seem to complement each other in that DOI focuses on individuals and SNT will take into account the influence of social networks associations when it comes to behavior change (Iqbal & Zahidie, 2021). Furthermore, DOI and SNT would be a valuable tool when implementing an intervention that may improve the quality of life for menopausal women (Iqbal & Zahidie, 2021).

In studies, that implemented DOI adoption of a new behavior did not happen in a social system. There are some individuals who are more open to behavioral change. When trying to implement or promote an innovation, there are typically different strategies that will need to be addressed. With that in mind there are five adopter categories that the targeted population may fall into which includes innovators, early adopters, early majority, late majority, and laggards (LaMorte, 2019). An important disclaimer is that this is just generalization and not all individuals will fall into these categories the way it is depicted here. For example, not all laggards have a low social status, and a low education level. In contrast, not all innovators have the highest social status and financial resources (International Design Foundation, 2021).

Innovators

This group of individuals are first in line, they tend to want to try the innovation first. People in this group tend to be interested in new ideas and embrace them. Typically, people in this group have a high social status, and have financial options that can secure them if the adoption is a failure (IDF, 2021).

Early Adopters

Enjoying leadership roles early adopters are known to welcome change, as they are already aware of the need to change. Like the innovators, the early adopters usually have a high social status and financial resources. Advanced educational degrees are common in the early adopters group. However, they do tend to be a little more inconspicuous when it comes to adopting an innovation (IDF, 2021).

Early Majority

Individuals who fall into the early majority group typically want to see evidence of the innovation before adopting it themselves. Individuals in the early majority group, tend to wait a significant amount of time to adopt an innovation. Common among this group is an above average social status, and typically associate with early adopters usually not holding a leadership position (IDF, 2021).

Late Majority

Very skeptical of change, the late majority individuals, will only try the innovation when others have successfully implemented the innovation by the vast majority of people. Typically, success stories of other individuals that have success will appeal to this population. In addition, they usually have low financial resources and a below average social status (IDF, 2021).

Laggards

Very conservative and very skeptical, this population is the most difficult to bring on board. Many times, laggards are driven by tradition and must be provided statistics and pressure from other groups. An aversion to an innovation, laggards mostly have contact with family and close friends, which constitutes as a low social status. Laggers also tend to have the lowest financial resources than the other groups (IDF, 2021).

Summary

It has been well documented that weight gain can lead to many chronic debilitating diseases. In addition, in this chapter scholarly literature supports the need for continued research for menopausal women. After menopause women are more at risk for metabolic and liver disease because of added weight (Office on Women's Health, 2019). Not every female will experience this, however, if there were support groups, screenings, and treatments available it may alleviate problems to help an already burdened healthcare system. There are specific physical and laboratory exams that can be done to prevent, diagnose and treat early. If women were educated on what happens to her body after menopause it might be enough of an incentive to participate in early screenings (Office on Women's Health, 2019). The important laboratory exams could include liver enzyme levels, fasting glucose, and triglycerides. Other non-invasive test that can detect chronic disease is BP, BMI, possible imaging, and WC to determine fat distribution (ADA, 2021).

The distribution of adipose tissue is very important because fat distribution can increase the risk of MetS and NAFLD. The increased cases of obesity is a pandemic that causes many deaths and disabilities, however, there are not many useful strategies to implement that works (ADA, 2021). Therefore, if there is a specific group that is plagued by the risk of obesity early detection and lifestyle changes could be beneficial. Furthermore, sex-specific strategies should be implemented especially for menopausal women, the target group, who have lost the ability to produce estrogen (NAMS, 2021).

Estrogen protects females from many potentially fatal diseases, so when the ovaries fail it opens pandoras box. This group of females are underrepresented and are a vulnerable group, as heart disease and NAFLD was thought to be more prevalent in men. In many cases the loss of estrogen may cause metabolic alterations, which may contribute to excess weight gain, that can lead to MetS, NAFLD, and possible progress to NASH (NAMS, 2021).

Subsequently, a lifestyle change would need to be implemented, therefore the theoretical method DOI was used. DOI can diffuse an innovation through a population to induce behavior change (Iqbal & Zahidie, 2021). EBP’s can also be beneficial as it introduces evidence-based research that may be influential to patients who are not too sure about change. Due to some gaps in DOI, SNT can ease some of those gaps. DOI tends to focus on the individual whereas SNT can assess the connection between social networking and behavior change (Iqbal & Zahidie, 2021).

In chapter 3, I will discuss the methodologies that was used to determine the presence of MetS and NAFLD in menopausal women. Included in the methods will be information on the data that was collected, collection methods for samples, and the identification of the target population.

Chapter 3: Research Method

In this chapter, I will discuss the research design. I addressed many relevant articles that show evidence of an increased risk factor of MetS and NAFLD after menopause. Other relevant material in this chapter will include the methodology used, population of interest, procedures used to collect the data, and the instruments used to collect and analyze the specimens.

Research Design and Rationale

I used a secondary research method with existing data that was publicly available. I downloaded the codebook of secondary data from NHANES from 2017-2018. NHANES supplies an array of cross-sectional examination surveys; these can be obtained by mobile examination units or clinics (NHANES, 2018). The use of a cross-sectional quantitative approach was based on the measurements of data collected from laboratory and observational methods.

The secondary data I used in the study include biochemical tests, physical measurements, and clinical information. Data collected pertain to chronic diseases such as obesity, serum cholesterol levels, and hypertension to mention a few. To limit validity issues, laboratory tests and medical examinations have specific protocols that are standard to insure comparability across sites. According to NHANES, since 1999, data are collected continuously every year from civilians in the United States population (Healthy People 2020, 2021).

A cross-sectional design can provide a snapshot of the frequency of a health-related issue at a given point in time. This type of design can help to determine and assess the burden of disease on a particular population. The defined population that will be used in this study are women who are 52 years or older, as they are indicators of women who have gone through menopause, in most cases. In addition, a cross-sectional study design will be valuable in the frequency of MetS and NAFLD within the population of menopausal women. A cross-sectional design allowed me to analyze the data including the conditions of metabolic syndrome and laboratory values assessed in NAFLD (NHANES, 2020).

Research Variables

The DV for Research Question 1 (RQ1) is MetS (dichotomous), the IV for RQ1 is age (ordinal) and ethnicity (nominal) and NAFLD (dichotomous). The DV for RQ 2 is MetS (dichotomous) the IVfor RQ 2 is gender (dichotomous), also adding in covariates (CoV2) as age (ordinal) and ethnicity (nominal) and NAFLD (dichotomous). Lastly, the DV for RQ3 is MetS, the IV is menopausal status of women (nominal) and CoV3 will be ethnicity (nominal). and NAFLD (dichotomous). All the codebook data were taken from NHANES 2017-18, and the statistical analysis that was used is the multiple binary logistic regression (NHANES, 2020).

Study participants in the data set obtained had blood drawn by a certified phlebotomist, and the amount of blood varied by age. The blood vials were stored according to NHANES policies and transported to various laboratories across the United States. Examinations occurred in mobile examination centers (MEC) in a controlled environment (NHANES, 2020). A controlled environment was used to allow the physical measurements to be done under standardized conditions. There is a correlation analysis that will include data such as age and gender (NHANES, 2020).

Methodology

Population

The participants in the NHANES 2017-2018 included 16,211 individuals from 30 different survey locations. In that population of people, 9,254 completed and interview and 8,704 of those individuals were examined. Included in the data were men, women, and children laboratory and examination results (NHANES, 2020). The codebook indicated I would be able to analyze the results to determine if the patient met three of the five conditions for a diagnosis of MetS. Furthermore, I was able to determine abnormal liver enzymes and TE scores regarding NAFLD (NHANES, 2020). My target population was women over 52 years of age.

Sampling and Sampling Procedure

A complex data plan was prepared to ensure proper inferences about the population. Data were taken from NHANES, a nationally represented data set, that may have underrepresented or overrepresented certain groups. An adjustment needed to be made in order to get accurate estimates of the population. To do this in SPSS, a CSS plan was created, and a logistic regression was conducted under complex samples rather than a normal logistic regression. I used age (52 years or older) and gender to target the population of interest (Elfil & Negida, 2017). Gender was divided into males and females and then recoded to reflect variables from 0 years to 150 years (NHANES, 2020).

When analyzing gender, there are 4,629 males and 4,625 females with a total of 9,254 participants. For the purpose of this study, I only used the 4,625 female participants; any females younger than 52 years of age and males were excluded when analyzing menopausal women. However, when I analyzed gender differences, men were included in the analysis. To protect confidentiality of the participating individuals, some of the variables were recoded. In addition, race-ethnicity variables are included and can be found in the demographic codebook (NHANES, 2020).

Data Collection Strategies and Research Instrumentation

A multistage probability design was used to obtain data from a population of individuals living in the United States according to the NHANES website. The staff that was involved in the data collection had special training to recognize and respect different cultures. Examination data were obtained in a MEC (CDC, 2017).

The MEC provides a controlled environment so the physical measurements can be done under standardized conditions. Making the MEC convenient, it was open to participants 5 days a week, and randomized participants into morning and evening sessions (CDC, 2017). Each examination that was conducted at the MEC had computerized data collection with a built-in data entry quality control. Quality control was of upmost importance, and many quality assurance analyses were completed for each examination. Participants had the opportunity to ask questions and refuse participation in the study at any time (CDC, 2017).

Blood Collection

According to the information on the NHANES website, biological specimens were also collected at MEC, including the collection, processing, storing and shipment of those specimens. The biological specimens that were collected for testing include blood, urine, and vaginal/penile swabs. In the analysis of this research study, the biological specimen used is blood samples. Blood was collected by a trained and certified phlebotomist, and then stored and sent to the appropriate lab (CDC, 2017). The laboratory team that handles the specimens include an American Society of Clinical Pathologists medical technologist, laboratory technicians, and certified phlebotomist. There is a chief medical technologist that will have the responsibility of overseeing all of the activities of the laboratory team. The chief medical technologist will also be involved in quality control, calibration of equipment, and maintenance (NHANES, 2020).

Blood work can play a very significant role in the diagnosis of disease. The data that I obtained from the laboratory data included triglycerides, fasting glucose, HDL, AST, and ALT. In order to assess the risk of MetS, there are five conditions that fall under the metabolic umbrella (AHA, 2021). A patient must be diagnosed with three of the five conditions set by experts in the field. Of the five conditions, three of the tests are blood levels of triglycerides, FPG, and HDL levels. In contrast, there are blood tests that are very important in the diagnosis of NAFLD as well. The AST and ALT are very useful in the diagnosis of potential liver disease along with a TE (NHANES, 2020).

The values that I used in the statistical analysis for blood values that fall under MetS was triglycerides (above 150 mg/dl), FPG (greater than 100 mg/dl), and HDL levels (lower than 40 mg/dl) respectively. The blood collection data used in abnormal liver enzymes include the AST (48 IU/L or higher) and the ALT (55 IU/L or higher), as determined by blood test results from the data (NHANES, 2020). In addition, to the laboratory data imaging such as the TE should also be used to assess if there is any significant damage to the liver (NHANES, 2020).

Instrumentation Used for Blood Specimens

Participants were fasting when blood collection was scheduled as stated in the procedure manual of the collection of blood specimens. The blood is collected by a certified phlebotomist that performs the venipuncture. A NaF evacuated tube system is used to collect the blood and then centrifuged to separate the plasma from the erythrocytes. The participants status and name were recorded immediately and then stored for analysis (CDC, 2017). After collection the vials of blood, were stored at -70 C until a licensed medical technologist can analyze the specimen. On the day of testing, the specimens will be thawed, mixed, and then refrozen to -70 C. Each manifest form will include patient sample ID, test name, date of collection, shipment ID, shipment date, lab name, lab ID and the survey year. The results of the tests are exported from the NHANES database and uploaded to Westat through a secure transfer done weekly (CDC, 2017).

The instrument that is used to analyze the blood specimen is the Roche/Hitachi Cobas C Chemistry Analyzer-C311(fasting blood glucose) and the Roche Cobas 6000 c501 model for the (AST, ALT, HDL, and triglycerides). Quality control procedures are used consistently to ensure the accuracy of the output of test results. There are at least 20 runs performed to establish a target control values and limits. Controls are tested with every run and then reviewed daily by a lab supervisor (CDC, 2017).

Transient Elastography

The TE is a specialized ultrasound used to measure liver stiffness and was conducted at the MEC. The TE is housed in Trailer three of the MEC. The room of the trailer has all of the appropriate equipment to conduct the examination. Before the procedure is done, the technologist provided a fasting questionnaire to the participant. Fasting was a requirement for the correct interpretation of the results. Participants are advised they may consume diet soda, black coffee, tea, and artificial sweeteners (CDC, 2018). The extent of fibrosis are measured in kilopascals (kPa). The amount of scarring, if present, will be measured by the Fibroscan, and a fibrosis result will be provided. The results are reported with a fibrosis score such as a fibrosis score of F1-F1 mild scarring, F2 moderate scarring, F3 severe liver scarring, and F4 advanced scarring leading to cirrhosis. Each of the fibrosis scores fall into a specific range indicating the level of fibrosis measured in the liver (Memorial Sloan Kettering Cancer Center, 2018).

Instrumentation Used Transient Elastography

The noninvasive exam enlists the FibroScan (model 502 V2 Touch) that has a handheld transducer (CDC, 2018). The transducer taps in rapid successions on the participant’s abdomen to determine if there is liver stiffness and to what extent or stage of damage has occurred. The FDA has approved the FibroScan Model 502 Touch, that is used in the examination (CDC, 2018).

Anthropometric Measurements

The measurements of the human body calculated into dimensions of bone, muscle, and adipose tissue is called anthropometry. The calculations stem from the participants weight, stature, recumbent length, skinfold thickness, circumferences (head, waist, limb), and limb length and breadths, such as the shoulder and wrist (NHANES, 2017).

The WC is a very important way to measure the amount of abdominal adiposity, as it is a risk factor for disease. The more abdominal adiposity an individual has, the more likely they may be at risk for metabolic and liver disease. The values that I used for this analysis is a WC of 35” or greater indicates increased abdominal adiposity. This value pertains to women, as that is the target population (Jiang, et al., 2018).

Instrumentation Used Anthropometric Measurements

NHANES supplies data that are used to evaluate many health disparities such as heart disease based on body composition. The goal for NHANES is to use high-quality body measurements using standardized procedures with calibrated equipment. All of the anthropometry measurements will take place in Trailer 1 of the MEC. All participants are eligible for the anthropometry examination and completed depending on the age of the participant (NHANES, 2017).

There are two people who are in the room with the participant during examination: a health technologist and a recorder. In addition, the extra person can alert the examiner if there is an issue (NHANES, 2017). For example, in WC measurements, the recorder will ensure the tape measure is parallel to the floor and snug against the skin.

A study data base called Integrated Survey Information System (ISIS) will be used to store the results. To ensure quality control ISIS will alert the technologist if recorded values fall outside of the preprogrammed edit range (NHANES, 2017).

The WC is of interest for this study. The WC is the measurement of the circumference of the abdominal adiposity using a retractable steel measuring tape. Participants will be provided standard MEC shirt and pant gown during the examination and be asked to change from street close to those provided (NHANES, 2017). The participants were instructed to stand straight with their arm placed on the opposite shoulder. The measurement is taken on the right side; the hip area is palpated to locate the right ilium of the pelvis. A measuring tape is extended around the waist and recorded using measurements to the nearest 0.1 cm (NHANES, 2017).

Quality control is of utmost importance, as NHANES is a widely used data sources. In gathering anthropometrics, most often errors are seen in body positioning, locating body landmarks correctly, reading results, and recording results into ISIS. To reduce recording errors, pre-programmed edit ranges are used, and ISIS will catch those readings and send an “out of range” message. In addition, a consulting anthropometry expert will visit the MEC twice a year to verify the protocols are implemented correctly (NHANES, 2017).

Blood Pressure

Accurate and reliable BP readings has been linked to better health outcomes. For example, if an individual is aware of their BP, then lifestyle changes or medical intervention may be indicated. BP is measured, and there are two values that are presented: the systolic and diastolic values. The instrument used is the auscultatory/manual technology or the oscillometer automated device (NHANES, 2019). Furthermore, systolic and diastolic BP are taken from the mean arterial pressure while using a device-specific algorithm, and oscillometric pulse is detected by the BP cuff. The BP cuff will increase the pressure up to 180 mm of Mercury (mmHg), and then the deflation of the cuff proceeds (NHANES, 2019).

When analyzing MetS one of the five conditions included high BP, so these readings are very important. Many Americans are plagued with high BP, but what is the true definition of high blood pressure or hypertension. A systolic pressure reading over 130 mm/Hg and a diastolic reading of 85 mm Hg is considered hypertension (AHA, 2020). There are various stages of hypertension, however, my focus is on hypertension in general. If an individual has stage 1 hypertension there is time for intervention in modification of diet and exercise in many cases. There was a total of 4 readings taken from each individual, however, I will only use three per recommendations from the AHA. The AHA recommends that three separate high readings should be obtained before the diagnosis of hypertension (AHA, 2016).

Instrumentation used Blood Pressure

The instrument that was used in the study is the Omron IntelliSense Blood Pressure Monitor model HEM-907XL (CDC, 2019). This model comes with a battery pack and four various cuff sizes. The Omron HEM-907XL has been approved by the FDA and is used as a clinical blood pressure monitor instrument. This blood pressure cuff has also passed the AAMI validation and has been used in many epidemiological studies.

In obtaining the BP reading, there are collection procedures that will indicate an accurate result. For example, the position that the individual was in, such as standing, sitting, legs crossed supported arm etc. (CDC, 2019). The cuff size was also of great importance, as an accurate reading could not be reached if the cuff was too small or too large. To eliminate possible errors all the participants were given gowns, and the cuff was placed on a bare middle-upper arm (CDC, 2019).

Collecting the BP was done by a health technician (HT) and the mobile examination center (MEC). Training used for HT is provided by the NCHS personnel and Westat (CDC, 2019). The training includes the skills and standards that are used to obtain accurate BP results. The HT is instructed to acquire three BP readings that are one minute apart ensuring no talking, using cell phone, or any other potentially distracting event (CDC, 2019).

Reliability and Validity of Instrumentation

There are two very important fundamental features that must be conducted to ensure accurate and dependable research. The instruments used in research need to provide valid and reliable results (Mohajan, 2017). How well the instruments used in the study performs is the validity of that instrument. However, when data is obtained, the amount of trust in the results depends on the reliability. An instrument that provides measurements must be able to control for random errors (Mohajan, 2017). Validity provides the truth in the findings, and the reliability is the stability of the findings.

When using secondary data in research it is essential to assess the validity and reliability of the methods that were used to collect the data. Reliability is used in research to measure consistency and trustworthiness without bias. Quantitative research must have consistency and repeatable results that can be repeated. In addition, validity should provide measurements for what it's designed to do. For example, instrumentation used to collect data must accurately measure values that are being studied (Mohajan, 2017). In contrast, the validity of quantitative research involves the instruments' ability to measure what it is intended to measure. Validity can provide credibility or internal validity and transferability or external validity. Internal validity will provide information on how the groups involved in the study were selected to ensure legitimacy. If the results are transferable to other groups of participants, then the results show external validity (Mohajan, 2017).

Quality control (QC) procedures are incorporated to ensure that NHANES is collecting and documenting data accurately and reliably. These procedures are in place to reduce any examiner variability, reduce errors, and ensure data quality (NHANES, 2020). The QC program has very strict procedures that takes place in the MEC's where the examinations take place. There are major elements that are involved in the QC program and that includes training, equipment monitoring and maintenance, and site visits by the National Center for Health Statistics (NCHS) and Westat. Westat is a company that many government agencies contract to provide data collection services (NHANES, 2020).

Quality Control Data Collection: Blood Specimens

Blood collection also followed strict standards of quality control. A trained phlebotomist interviewed the participant ensuring there were not issues that would exclude them from the venipuncture. Some of the biochemical blood draws required the patient be in a fasted state, so a questionnaire was given to confirm compliance (CDC, 2017). Participants who were 12 years and older received a hard copy verification from was given that included questions such as, “Did you eat or drink anything, other than plain water, after 11:30 p.m. last night?” If the participant marked “Yes” it was highlighted in pink indicating, they have not met the required fast time of nine hours (CDC, 2017).

Each participant has a bracelet that contains his/her ID number in a barcode and the phlebotomist used a scanner to log it into the computer. The participant will wear this bracelet the entire visit. The supplies that are used by the phlebotomist are sterile and only used once and the work area is clean and uncluttered (CDC, 2017). All instruments used in the laboratory must have documented quality control and entered in the Laboratory QC module. All of the lot numbers and expiration dates for the vacutainer tubes used in the blood collection were entered into the QC modules. The centrifuges that were used at the MEC must operate at two to eight 0C, and the blood is spun down in the refrigerated centrifuge (CDC, 2017). Specimens must be frozen, or it will affect the integrity and quality of the specimen, so dry ice and frozen gel packs are on hand in case of an outage. All of the freezers and refrigerators have a temperature guard system installed to monitor their temperature at all times. In addition, the MEC lab does have a backup generator to preserve the integrity of the specimens (CDC, 2017).

Transportation of the specimens can be done using the International Air Transport Association (IATA), which regulates and publishes dangerous goods. The specimens are packed in dry ice (this is classified as dangerous goods) and used air transport via Federal

Express (CDC, 2017). The laboratory team consists of three American Society of Clinical Pathologists- certified medical technologists and a certified phlebotomist. The chief medical technologist is responsible for the operations of the MEC lab including quality control, equipment calibration and maintenance (CDC, 2017).

Quality Control Data Collection: Transient Elastography

Eligibility criteria must be met before the liver ultrasound TE (LUTE) before the exam was conducted. Participants of the study may be excluded if he/she cannot lie down on the exam table, current pregnancy, implanted medical device, or have an injury or lesion on the right side of the abdomen (NHANES, 2018). In addition, the participant was given a fasting questionnaire to ensure correct interpretation of the results. The question will specify the last time the participant ate or drank anything, with the understanding that they are allowed to consume diet soda, black coffee, or tea with artificial sweeteners (NHANES, 2018).

One critical part of QC for the TE is routine cleaning to ensure optimal operation. The wands and machine must be disinfected according to a detailed scheduled outline. The wands do have mechanical parts and must be serviced and calibrated by a company called Echosens (NHANES, 2018). Quality assurance is performed on the Fibroscan by Shear Wave Liver Fibrosis Phantoms. The phantoms are used as a tool to detect any potential inconsistencies between the three different Fibroscan machines. The phantoms will measure the wand and the ultrasound wave speed for one hundred measurements (NHANES, 2018).

Quality Control Data Collection: Anthropometric

Regarding the collection of the anthropometry data WC validity and reliability were limited best as possible. There was an anthropometry expert that would visit the MEC twice per year. During visits the expert will meet with the technologist performing the exam to review any issues that was encountered and reported to Westat (NHANES, 2017). There will be a summary of any issues forwarded to the appropriate NCHS project officer. In addition to observations of measurements one of the essential functions of a body measurement consultant is to serve as the "gold standard" anthropometry examiner. For example, the consultant will perform a repeat anthropometry examination on all the participants following the examination performed by a technologist. This gold standard replicated results is a very important aspect of quality control for anthropometry measurements (NHANES, 2017).

Quality Control Data Collection: Blood Pressure

Likewise, strict quality control methods are in place for BP collection as well. Each time the technician logs in to the application he/she has a prompt to do a QC check. The QC checks are done daily and will not show up on the screen until another check is required (usually the next day). The NCHS personnel and Westat will visit the MEC in regular intervals (NHANES, 2019). To ensure quality of data collection the staff will generate reports from the ISIS intraweb. The amount of BP examinations and the times collected will be sorted by age, and the technologist that collected the BP. In addition, a Westat specialist in coordination with a NCHS project officer will introduce retraining sessions frequently (NHANES, 2019).

Data Analysis Plan

The program I have chosen to analyze data in this research study is the Statistical Package for the Social Sciences (SPSS v. 27). I did receive IRB approval before analyzing the data. SPSS is always updating and improving the software package to keep up continued demand of statistical output. In Chapter 4, my research findings are provided with the help of SPSS. SPSS has the ability to collect data from different types of files and help researchers perform complex data analysis. For example, SPSS can read and analyze files from different statistical packages, spreadsheets, and databases (Statistics Solutions, 2022) Charts, tables, descriptive statistics etc., are very precise and easy to manipulate. Many helpful features come with SPSS such as techniques that account for missing values, and “split file” for comparative studies. SPSS can provide many inferential and multivariate statistical procedures as well (Statistics Solutions, 2022).

I have various data sets from the codebook I downloaded that were used in the statistical analysis. My target population was women over 52 years of age, using a categorical scale. In my analysis all of the variables were recoded to reflect male=1 and female =2, women under 52 years =0 and women over 52 years =1. Metabolic syndrome (MetS) was also recoded to reflect a diagnosis of MetS by having three out of five health conditions. In order to have a diagnosis of MetS a patient must have three of these five conditions which include hypertension, high levels of triglycerides, elevated fasting glucose, low HDL levels, and an increased waist circumference. The new recoded variable for MetS done in SPSS is Metabolic Syndrome and the values are No MetS=0 and Yes MetS =1.

Furthermore, Non-Alcoholic Fatty Liver Disease (NAFLD) was recoded by using the data that was collected from the liver ultrasound that measured liver stiffness. The patient was given a fibrosis score (F0, F1, F2 and so on) depending on the level of thickness which could indicate liver damage the higher the score. The values were recoded to reflect liver stiffness by 0=F0, 1=F1, 2=F2, 3=F3, and 4= F4.

A descriptive analysis was conducted first by looking at charts and graphs to characterize the demographics of the participants. The codebook contains males and females under 52 years of age and over age 52. To select for females only to answer RQ 3, I recoded this variable by using a mathematical equation (gender=2) & (age= recode =1) in "Data select cases" in SPSS. After recoding those variables, I went back and ran a descriptive statistic, and the output will reflect females and age was 52 years of age or older. MetS is the DV and, my IV's include NAFLD, age, ethnicity, gender and menopausal status.

I used categorical variables in my analysis. Categorical variables are further divided into nominal, dichotomous, or ordinal variables (Statistics Solutions, 2022). I used many different variables to collect and analyze risk factors of MetS, demographic data, and NAFLD data. Age is a scale variable and measured age in years. Age and gender were used in determining the relationship between MetS and NAFLD. Dichotomous variables in the codebook, include MetS, gender, ALT analysis, and AST analysis. The ordinal variables are NAFLD and race using non-Hispanic white females as a baseline.

After conducting the descriptive statistics, the output information provided revealed the variable dispersion. I did include the national population estimates to calculate different demographic indicators. Due to the use of data from NHANES weighted frequencies, percentages, and standard errors were done to produce an unbiased statistical analysis. The importance of using weighted data is to ensure the data that is in the codebook reflects the target population. In addition, the standard error (SE) allowed me to assume that the data I have is a sample from a larger population (CDC, 2020). The significance of running a SE is the ability for this statistical test to quantify the variation in sample means. Along with the SE the standard deviation will provide the spread of individual data values around the mean (Agency for Healthcare Research and Quality, 2016).

I did complete a multivariable regression analysis to ascertain the strength of the relationship between the DV MetS (dichotomous) and the IV's NAFLD(dichotomous), and age (ordinal), ethnicity (nominal), gender (dichotomous), and menopausal status (nominal) as my IV's (Hasan, 2020). The multivariable model that was used is the logistic regression for binary outcomes, to establish whether the IV's are associated with the binary outcome (Grant, Hickey, & Head, 2019). The rationale of using logistic regression is that it seems to be a biologically reasonable model to describe the relationship between my DV's, IV's and covariates. To answer my research questions, the multivariable logistic regression was a beneficial model to describe the relationship between MetS and the set of predictors. I have identified helpful statistical analysis including, applying multistage probability survey design, odds ratio, and a 95% confidence interval (CI) (Grant, Hickey, & Head, 2019). A p-value of 0.05 will be used to establish statistical significance in my statistical-analysis (Hasan, 2020).

Research Questions and Hypotheses

RQ1: Is there an association between MetS and NAFLD, controlling for age and ethnicity?

$H_1$1: There is an association between MetS and NAFLD, controlling for age and ethnicity.

$H_0$1: There is no association between MetS and NAFLD, controlling for age and ethnicity.

RQ2: Is there gender difference in the association between MetS and NAFLD, controlling for age and ethnicity?

$H_1$2: There is a gender difference in the association between MetS and NAFLD, controlling for age and ethnicity.

$H_0$2: There is no gender difference in the association between MetS and NAFLD, controlling for age and ethnicity.

RQ3: Does the menopausal status of women affect the relationship between MetS and NAFLD, while controlling for ethnicity?

$H_1$3: Menopausal status of women does affect the relationship between MetS and NAFLD, while controlling for ethnicity.

$H_0$3: Menopausal status of women does not affect the relationship between MetS and NAFLD, while controlling for ethnicity.

Ethical Procedures

I did obtain approval from Walden's Institution Review Board (IRB) before conducting and manipulating the data. The data was obtained from NHANES for the purpose to conduct meaningful research on the prevalence of major diseases and risk factors. All the health measurements used in this analysis was conducted in a specially designed mobile center. Advanced computers and technology were used in the collection and storage of participant information according to the CDC and NHANES website. The data collected is made available to researchers, universities, health care providers and educators.

All the data that is collected from participants is kept in strict confidence. NHANES has an informed consent process, and the participants are assured that the health information that is collected from them is used for stated purpose (CDC, 2018). The data collected is not to be used or released for any other purpose without the consent of the participant in accordance with section 308(d) of the Public Health Service Act (42 U.S.C. 242m). The privacy of the participants is in federal compliance with the Information Protection and Statistical Efficiency Act of 2002 (CIPSEA, Title 5 of Public Law 107-347). To be in compliance of CIPSEA, each employee has taken an oath to protect the participants privacy and is punishable with a jail term of up to 5 years, and a fine up to $250,000 dollars or both. An employee must not willfully disclose any identifiable information about a participant under any circumstance (CDC, 2018).

Summary

Chapter 3 included the type of statistical analysis that will be used to analyze the data, overview of the target population, sample procedures, data collection and instrumentation used, quality control methods and ethical issues. Quantitative methodology was used MetS as the dependent variable and NAFLD as the independent variable accounting for age and ethnicity. The target population that was used were women 52 years of age or older, as it typically indicates menopause has occured. Chapter 4 will include a more in-depth explanation of the data and any barriers or confounders found from the statistical analysis. Visual SPSS output is available to support the data collection and recoding of variables.

Chapter 4: Results

Introduction

In this chapter, I used quantitative analysis to assess if there is an association between MetS and NAFLD status. I obtained approval from the Walden Institutional Review Board (IRB), approval number 07-12-22-0117464, to proceed with the statistical analysis. The purpose of this study was to explore the gaps in reasearch in regards to MetS and NAFLD. One of the main target populations in this study are menopausal women, which is reflected as women over 52 years of age. To determine is an individual had MetS, a series of tests had to be completed and conducted by professionals in the medical field. There were five conditions that a patient had to be diagnosed with in order to have metabolic disease.

Likewise, to determine if an individual had NAFLD, a TE was perfomed in a MEC approved facility. Even though a liver biopsy is the “gold standard” to detect liver fibrosis, TE is suggested for a non-invasive method to monitor liver fibrosis. It is unrealistic, due to the amount of individuals that have diabetes or MetS, to perfom a liver biopsy. Using the TE can lead to better health outcomes, as it can monitor patients for an extended period of time determined by the healthcare professional.

The participants for this study were obtained from NHANES. Due to the nature of a nationally used data set, care in the statistical analysis was implemented. The samples are not random; instead, they are a complex, mulitstage probability sampling design. This design is used to represent civilian and non-institutionalized population in the United States. This method of collecting data can lead to oversampling of certain population subgroups. In this case, special statistical software in SPSS was done to analyze complex survey design (CDC, 2018).

According to the CDC (2018), NHANES creates sample weights that are used in a complex survey designs. The sample weights include survey nonresponse, oversampling, and post-stratification to ensure the estimates are representative of the U.S. civilian noninstitutionalized population. In addition, NHANES, will provide post-stratified weights to match the poulation controls for each domain, according to the Estimation Procedures Document on the NHANES website. In this analysis, MEC exam weight (wtmec2yr) was used in designing a complex data plan and can be found under demographic files. The two-year cycle obtained for this analysis is a nationally representative sample, as indicated on the NHANES website (CDC, 2018).

RQ1: Is there an association between MetS and NAFLD, controlling for age and ethnicity?

$H_1$1: There is an association between MetS and NAFLD, controlling for age and ethnicity.

$H_0$1: There is no association between MetS and NAFLD, controlling for age and ethnicity.

RQ2: Is there gender difference in the association between MetS and NAFLD, controlling for age and ethnicity?

$H_1$2: There is a gender difference in the association between MetS and NAFLD, controlling for age and ethnicity.

$H_0$2: There is no gender difference in the association between MetS and NAFLD, controlling for age and ethnicity.

RQ3: Does the menopausal status of women affect the relationship between MetS and NAFLD, while controlling for ethnicity?

$H_1$3: Menopausal status of women does affect the relationship between MetS and NAFLD, while controlling for ethnicity.

$H_0$3: Menopausal status of women does not affect the relationship between MetS and NAFLD, while controlling for ethnicity.

Data Analysis

In this study, the correct analysis of the data is critical in order to find the answers to the research questions. The variables analyzed include DV: MetS and the IV: NAFLD

(Liver Stiffness F0, F1, F2, F3 and F4), Gender (Male=1, Female=2) and Race (1=Mexican American, 2=Other Hispanic, 4= Non-Hispanic Black, 6= non-Hispanic, 7=Other Race-Including Multiracial, 8= Non-Hispanic White (Reference Group).

I was able to use SPSS to design a complex data design to ensure the integrity of the analysis. The specific design includes strata (includes levels of stratification), clusters (grouping), and weights (Zou et al.,). To use this feature, there are a few steps to complete before running the statistical analysis. The first step in creating an analysis plan was to obtain the weighted 2yr MEC data from NHANES. In SPSS, under analyze, complex samples was selected, saved, and labeled as such. This saved file can allow identification of the stratification, sample weights, and clustering variables. The output Strata defines a distinct subpopulation or strata, cluster defines the observational units, or clusters, and the sample weights are calculated automatically (IBM, 2021). To accomplish this, I included these variables from the codebook, WTMEC2YR (Full sample 2 year MEC Exam Weight), SDMVPSU (Masked Variance pseudo-PSU, and SDMVSTRA (masked variance pseudo-stratum; see Table 1).

Included in the 2017-2018 NHANES demographic files are 15 masked variance (MVUs) strata and 30 masked primary sampling units (PSUs). Each stratum has two PSUs, and the MVUs are a collection of secondary sampling units (NHANES, 2020). The MVUs are used to produce variance estimates. These estimates are based on the actual survey dample strata and PSUs. To protect the privacy of the participants and to reduce discolure risks, MVUs are used (NHANES, 2020). The two-year sample weights used in this anaylsis (WTMEC2YR) was recommended by NHANES and are provided in the codebook. NHANES offers further details and turorials on sample weight and other analytic issues in the NHANES Analytic Guidelines (NHANES, 2020).

Table 1

Complex Data Design Summary

			Stage 1
Design variables	Stratification	1	Masked variance pseudo-stratum
	Cluster	1	Masked variance pseudo-PSU
Analysis information	Estimator Assumption		Sampling with replacement

Note. From “National Health and Nutrition Examination Survey “. NHANES, 2017-2018 Demographics Variable List.

Exploratory Study Results

Individuals diagnosed with MetS and NAFLD were the primary focus of this study. The main target population is women over 52 years of age in the hopes menopause has taken place. As previously stated, there are five conditions that must be met before a diagnosis of MetS and an increase in liver stiffness for a diagnosis of NAFLD.

A complex data plan was prepared to ensure proper inferences about the population. Data were taken from NHANES, a nationally represented data set, that may have underrepresented or overrepresented certain groups. An adjustment needed to be made in order to get accurate estimates of the population. To do this in SPSS, a CSS plan was created, and a logistic regression was conducted under Complex Samples rather than a normal logistic regression (see Tables 2 and 3). There were a total of 201,427,118 participants, of the unweighted cases there were 5,950, and 3,304 were invalid, leaving a total of 9,254 participants for this study.

Table 2

Sample Design Information

		N
Unweighted cases	Valid	5950
	Invalid	3304
	Total	9254
Population size		201427118.343
Stage 1	Strata	15
	Units	30
Sampling Design Degrees of Freedom		15

Note. From “National Health and Nutrition Examination Survey “. NHANES, 2017-2018 Demographics Variable List.

Table 3

Demographics Variable List

Categorical Variable Information

		Weighted count	Weighted percent
Metabolic syndrome	No metabolic syndrome	150070123.353	74.5%
	Yes metabolic syndrome[b]	51356994.989	25.5%
Race recode	Non-Hispanic White	65931203.154	32.7%
	Mexican American	29392713.618	14.6%
	Other Hispanic	17478414.106	8.7%
	Non-Hispanic Black	49029107.484	24.3%
	Non-Hispanic Asian	28382467.517	14.1%
	Other Race - Including Multi-Racial	11213212.464	5.6%
Population Size		201427118.343	100.0%

Note. From "National Health and Nutrition Examination Survey ". NHANES, 2017-2018 Demographics Variable List.

To answer RQ1: Is there an association between MetS and NAFLD, controlling for age and ethnicity?

A logistic regression analysis was conducted using a complex sample to investigate [*if NAFLD, Age, Gender, and Ethnicity had an association to MetS*]. The outcome of interest was [*MetS]*. The possible predictor variables were [*Age, and Ethnicity]*. Based on the results of the multivariable logistic regression there is a statistically significant association between MetS, and NAFLD after controlling for age (OR=2.96, 95% CI=2.319-3.778, p<0.001). However, ethnicity was not statistically significant (p=0.237). The Nagelkerke indicates that the predictors explain about 11.5% of the variance, in the response variable [Nagelkerke R squared = .115] (see Table 4).

Table 4

Pseudo R Squares

Cox and Snell	.078
Nagelkerke	.115
McFadden	.071

Note. Dependent variable: Metabolic syndrome (reference category = Yes Metabolic syndrome).
Model: (Intercept), Race recode, Liver stiffness, Age recode
From "National Health and Nutrition Examination Survey ". NHANES, 2017-2018 Demographics Variable List.

The model indicated that the independent variables [*Age, Gender, and Non-Alcoholic Fatty Liver Disease*] were significantly significant ($p < .05$) controlling for age, and gender. However, the independent variable [*Ethnicity*] was found not to be significant ($p > .05$). The predictor variable [*Non-Alcoholic Fatty Liver Disease*] in the analysis did contribute to the model. The Wald = [40.875], p=. 000 (See Table 5).

Table 5

Tests of Model Effects

Source	df1	df2	Wald F	Sig.
(Corrected model)	7.000	9.000	15.461	.000
(Intercept)	1.000	15.000	537.740	.000
Race recode	5.000	11.000	1.611	.237
Liver stiffness	1.000	15.000	40.875	.000
Age recode	1.000	15.000	89.793	.000

Note. Dependent Variable: MetS (reference category = Yes metabolic syndrome) Liver stiffness=NAFLD
Model: (Intercept), Race recode, Liver stiffness, Age recode
From "National Health and Nutrition Examination Survey ". NHANES, 2017-2018 Demographics Variable List.

The estimate of the odds ratio favored a [positive] relationship of approximately 45% chance of every unit increase in liver stiffness (NAFLD) the more likely an individual will have metabolic syndrome. Controlling for age proved to be a significant predictor with an OR (2.96) indicating those individuals 52 or older are 3 times more likely to have MetS. Controlling for ethnicity, the model indicated it was not statistically significant; however, it is important to note that Mexican Americans are 1.8 times more likely to have MetS than the reference group Non-Hispanic White (See Table 6).

Table 6
Parameter Estimates

Metabolic Syndrome	Parameter	B	95% confidence Interval Lower	Upper	Exp (B)	95% Confidence Interval for Exp (B) Lower	Upper
	(Intercept)	-2.327	-2.601	-2.052	.098	.074	.128
	Race recode = 1	.593	.194	.992	1.809	1.214	2.697
	Race recode = 2	.227	-.262	.716	1.255	.770	2.046
	Race recode = 4	.111	-.153	.375	1.117	.858	1.455
	Race recode = 6	.112	-.290	.514	1.119	.748	1.673
	Race recode = 7	-.070	-.396	.256	.932	.673	1.292
	Race recode = 8	.000[a]	.	.	1.000	.	.
	Age recode	1.085	.841	1.329	2.960	2.319	3.778
	Liver stiffness	.441	.294	.588	1.554	1.342	1.800

Note. Dependent Variable: Metabolic syndrome (reference category = No metabolic syndrome
Race Recode= 1
Model: (Intercept), Race recode, Age recode, Liver stiffness
a. Set to 0 because this parameter is redundant
From "National Health and Nutrition Examination Survey ". NHANES, 2017-2018 Demographics Variable List.
Race Recode: (1=Mexican American, 2=Other Hispanic, 4= Non-Hispanic Black, 6= non-Hispanic, 7=Other Race-Including Multiracial, 8= Non-Hispanic White (Reference Group).

RQ 2: Is there gender difference in the association between MetS and NAFLD, controlling for age and ethnicity?

A complex data logistic regression was conducted to examine [*if there are gender differences in the association between MetS and NAFLD*]. Further controlling for gender, the relationship remains statistically significant and exhibited a decrease in risk for females vs. males (OR=-.734, 95%CI .387-.596, $p<0.001$). The outcome of interest was [*MetS*] another possible predictor variable is [*Gender Differences] (*See Table 7*).*

Table 7

Categorical Variable Information

		Weighted Count	Weighted Percent
Metabolic syndrome[a]	No metabolic syndrome	150070123.3	74.5%
	Yes metabolic syndrome[b]	51356994.98	25.5%
Gender	Male	98012625.77	48.7%
	Female	103414492.5	51.3%
Population size		201427118.3	100.0%

Note. a. Dependent variable and b. reference category

From "National Health and Nutrition Examination Survey ". NHANES, 2017-2018 Demographics Variable List.

The value of the Nagelkerke is .075 [Nagelkerke Pseudo R squared = .075]. Controlling for age and ethnicity, the predictor variable [*Gender Differences*] conducted in the regression did contribute to the model (see Table 8).

Table 8

Pseudo R Squares	
Cox and Snell	.051
Nagelkerke	.075
McFadden	.046

Note. Dependent Variable: Metabolic syndrome (reference category = Yes metabolic syndrome)

Model: (Intercept), Race recode, Gender new, Liver stiffness.

From "National Health and Nutrition Examination Survey ". NHANES, 2017-2018 Demographics Variable List.

The Wald= [52.328], (see Table 9), the unstandardized B= [-.734], SE= [.101], indicating for every unit increase in [*Gender Differences*], MetS will decrease by .73 units (see Table 10).

Table 9

Tests of Model Effects

Source	df	Wald Chi-Square	Sig.
(Corrected model)	3.000	131.437	.000
(Intercept)	1.000	74.004	.000
Race recode	1.000	9.410	.002
Gender new	1.000	52.328	.000
Liver stiffness	1.000	56.553	.000

Note. Dependent Variable: MetS (reference category = Yes metabolic syndrome)
Model: (Intercept), Race recode, Gender new, Liver stiffness
From “National Health and Nutrition Examination Survey “. NHANES, 2017-2018

Demographics Variable List.

Table 10

Parameter Estimates

				95% Confidence Interval			95% Confidence Interval for Exp (B)	
Metabolic syndrome	Parameter	B	Std. Error	Lower	Upper	Exp (B)	Lower	Upper
No metabolic syndrome	Intercept	1.770	.182	1.382	2.159	5.873	3.983	8.658
	Race recode	0.61	.020	.019	.103	1.063	1.019	1.108
	Gender new = .00	-.734	.101	-.950	-.518	.480	.387	.596
	Gender new = 1.00	.000[a]	.	.	.	1.000	.	.
	Liver stiffness	-.482	.064	-.618	-.345	.618	.539	.708

Note. Dependent variable: Metabolic syndrome (reference category = Yes metabolic syndrome)
Model: (Intercept), race recode, gender new, and liver stiffness
From “National Health and Nutrition Examination Survey “. NHANES, 2017-2018

RQ3: Does the menopausal status of women affect the relationship between MetS and NAFLD, while controlling for ethnicity?

A logistic regression was conducted to investigate if the [*Menopausal status of women would affect the relationship between MetS and NAFLD, while controlling for Ethnicity*]. The outcome of interest was [*MetS*] a possible predictor variable is [*Menopausal status]* which includes women over 52 years of age. Focusing on women only, and further controlling for menopausal status, the association between MetS and NAFLD was still statistically significant (OR=2.227, p<0.001). The Nagelkerke had a value of .050 [Nagelkerke Pseudo R squared = .050] (see Table 11).

Table 11

Pseudo R Squares

Cox and Snell	.035
Nagelkerke	.050
McFadden	.030

Note. From “National Health and Nutrition Examination Survey “. NHANES, 2017-2018 Demographics Variable List. Dependent Variable: Metabolic Syndrome = Yes (metabolic syndrome). Model: (Intercept), Race Recode, Gender New, Liver Stiffness.

Controlling for Ethnicity, the predictor variable [*Menopausal status*] conducted in the regression did contribute to the model. The odds ratio indicates that Mexican Americans, Other Hispanic, and Non-Hispanic Black women are statistically significant. However, the same is not true with Non-Hispanic Asians and Other race, as the confidence interval includes 1 for both (see Table 12). While controlling for ethnicity the odds ratio for developing [*MetS*] increased approximately 20% for every 1 unit increase of [*NAFLD/Liver Stiffness*] (see Table 13).

Table 12

Odds Ratio 1[a]

	Metabolic syndrome	Odds Ratio	95% Confidence Interval Lower	Upper
Mexican American vs. Non-Hispanic White	Yes, Metabolic syndrome	2.785	1.102	7.039
Other Hispanic vs. Non-Hispanic White	Yes, Metabolic syndrome	2.658	1.321	5.351
Non-Hispanic Black vs. Non-Hispanic White	Yes, Metabolic syndrome	1.945	1.255	3.013
Non-Hispanic Asian vs. Non-Hispanic White	Yes, Metabolic syndrome	1.635	.821	3.257
Other Race Including Multi-Racial vs Non-Hispanic White	Yes, Metabolic syndrome	1.976	.672	5.811

Note. Dependent Variable: Metabolic syndrome (reference category= No metabolic syndrome)

Model: (Intercept), Race recode, Liver stiffness

Table 13

Odds Ratios 2a

Units of Change		Metabolic Syndrome	Odds Ratio	Lower	Upper
Non-alcoholic fatty liver disease	1.00	Yes metabolic syndrome	1.227	.937	1.609

Summary

The purpose of this study was to examine the association between MetS and NAFLD while controlling for age, gender, and ethnicity. A complex data plan was prepared to ensure proper inferences about the population. Data was taken from NHANES, a nationally represented data set, that may have underrepresented or overrepresented certain groups. An adjustment will need to be made to get accurate estimates of the population. To do this in SPSS a CSS plan was created, and a logistic regression was conducted under complex samples rather than a normal logistic regression.

Based on the results of the multivariable logistic regression there is a statistically significant association between MetS, and NAFLD after controlling for age (OR=2.96, 95% CI=2.319-3.778, $p<0.001$). However, ethnicity was not statistically significant ($p=0.237$). In addition, even though ethnicity was found to not be significant, Mexican Americans were more likely to have MetS. The odds ratio indicated that for every unit increase in liver stiffness the more likely the individual will have MetS. Age was also an important indicator predicting that individuals over 52 are 3 times more likely to have MetS.

After further controlling for gender, the relationship remains statistically significant and exhibited a decrease in risk for females vs. males (OR=-.734, 95%CI .387-.596, $p<0.001$). The purpose was to examine if there are gender differences in the association between MetS and NAFLD while controlling for gender. The results in this analysis indicated that females have a decrease in MetS compared to males.

Lastly, focusing on women only, and further controlling for menopausal status, the association between MetS and NAFLD was still statistically significant (OR=2.227, $p<0.001$). The results were unexpected regarding ethnicity. Turns out Mexican Americans, Other Hispanic, and Non-Hispanic Black women over 52 years old were more likely to be diagnosed with MetS (95% CI 1.102-7.039, 1.321-5.351, & 1.255-3.013). The odds ratio showed a 20% chance if a woman is of these ethnicities. Age did show some significance; however, ethnicity did seem to play an important role in the development of the disease almost more than age. Furthermore, when looking at gender differences ethnicity did not have significance until males were taken out of the equations.

Chapter 5: Discussion, Conclusions, and Recommendations

Discussion

Menopause can bring forward life changes that a women many not understand or be completely unaware of. Some of the physiological and biological changes can increase a number of chronic diseases due to hormonal changes. As discussed in this paper, an increase of MetS and NAFLD has been deamed an epidemic. However, there are gaps in the research when it comes to health outcomes for women who have went through menopause. It is important to understand the prevelance of these associated chronic diseases to implement health care practices that can implement disease education and interventions.

There needs to be more investigations into the health of menopausal women to decrease health disparities and to lessen the public health burden. Furthermore, there are some younger women who may require a full hysterectomy, which can induce menopause, and she may face the same challenges. If programs could be developed to educate and screen women after menopause there would be more awareness and support for this group of individuals.

Therfore, in this reasarch the target population was menopausal women. I included gender differences and race to account for confounders. I also created a complex data plan to capture the true population. I based menopausal status on women who are 52 years or older, as that is the average age of menopause, according to the Menopause Guidebook (NAMS, 2021). Due to the fact that metabolic and liver disease increase with age, I believe estrogen may play an important role in the development of chronic disease. One of the classical implications of MetS and NAFLD is the accumulation of visceral fat in the abdominal region.

Interpretation of the Results

The results from this study are consistent with research that has been done previously. There is a high percentage of individuals in general that have MetS and NAFLD with increased age. After preparing a complex data plan design to capture the true population, a logistic regression was conducted. The complex data plan was used to get a more accurate esimate of the population. When the CSS plan was designed in SPSS, I was able to run the logistic regression.

RQ1: Is there an association between MetS and NAFLD, controlling for age and ethnicity?

$H_1$1: There is an association between MetS and NAFLD, controlling for age and ethnicity.

$H_0$1: There is no association between MetS and NAFLD, controlling for age and ethnicity.

To determine if there is an association between MetS and NAFLD, controlling for age and eithnicity provided valuable results. Based on the results of the multivariable logistic regression there is a statistically significant association between MetS, and NAFLD after controlling for age (OR=2.96, 95% CI=2.319-3.778, $p<0.001$). However, ethnicity was not statistically significant ($p=0.237$). The predictors of interest age, gender, and ethnicity and was correctly specified and satisfied 11.5% of the population

(Nagelkerke R squared = .115). All of the predictor variables were found to be significant, with a p value less than .05, with the exception of ethnicity. Ethnicity was found to be insignificant as a predictor of MetS; however, I think it is important to note that it was significant in Mexican Americans.

The other predictor variables favored a positive relationship, according to the Wald test 40.875 with a p value of 0.001. This is a very important finding, as it is indicating that there is a 45% chance of every unit increase in liver stiffness the more likely the individual will have MetS. In controlling for age, the OR was 2.96, leading to an likelyhood of having MetS 3 times more likely after the age of 52. Controlling for race indicated that for other Hispanic, Non-Hispanic Black, Non-Hispanic Asian, other Race including Multi-Racial, and Non-Hispanic White were not a predictor. However, Mexican Americans were 1.8 times more likely to have MetS than the reference group.

RQ2: Is there gender difference in the association between MetS and NAFLD, controlling for age and ethnicity?

$H_1$2: There is a gender difference in the association between MetS and NAFLD, controlling for age and ethnicity.

$H_0$2: There is no gender difference in the association between MetS and NAFLD, controlling for age and ethnicity.

A complex data logistic regression was done to determine if there was gender differences in the association between MetS and NAFLD. After further controlling for gender, the relationship still remains statistically significant (OR=-.734, 95%CI .387-.596, p<0.001). Gender differences did contribute to the model as indicated with a

Nagelkerke Pseudo R Squared of .075. Analyzing the unstandardized B (-.734), SE (.101), and Wald (52.328) indicated that for each unit increase in gender differences, MetS will decrease by .73 units.

RQ3: Does the menopausal status of women affect the relationship between MetS and NAFLD, while controlling for ethnicity?

> $H_1$3: Menopausal status of women does affect the relationship between MetS and NAFLD, while controlling for ethnicity.
>
> $H_0$3: Menopausal status of women does not affect the relationship between MetS and NAFLD, while controlling for ethnicity.

Focusing on women only, and further controlling for menopausal status, the assiciation between MetS and NAFLD was still statistically significant (OR=2.227, p<0.001). A logistic regression using a complex data plan model indicated that the model was appropriately specified with a Nagelkerke Pseudo R square of .050. While controlling for ethnicity, menopausal status assured me that it was indeed a predictor variable, and did contribute to the model. However, according to the OR Mexican Americans, other Hispanics, and Non-Hispanic Black women were statistically significant (95% CI 1.102-7.039, 1.321-5.351, & 1.255-3.013). When analyzing Non-Hispanic Asians and other race, ethnicity was insignificant. When controlling for ethnicity, the odds of developing MetS increased approximately 20% for every 1 unit increase of NAFLD.

Interpretation in the Context of DOI

Presented early on in the dissertation, a rationale for conducting the research was guided by a strong theoretical framework. The theoretical framework helped to establish the structure that guided the research conducted in this dissertation. After completing the literature review, I was able to identify gaps in the research and decide the variable and the relationship I wanted to further understand. The interpetation of the findings clearly demonstrated that the relationship between MetS, NAFLD, and menopause was formed from investigating prevous research (Statistics Solutions, 2022).

The DOI theoretical framework explained the variables in the study. The purpose of using DOI is to introduce new evidence-based material that can improve health care and the health outcomes of patients. This innovation would be communicated through certain channels over time, as many people are not open to drastic changes all at once (Dearing & Cox, 2018). In this case, the dependent variable (MetS) can diffuse in society and hopefully morph into a wave of knowledge and acceptance. Many times, to address certain barriers in healthcare, it may require doctors, nurses, and other stakeholders to coordinate delivery of this method. In order to sustain a new innovation, it is important for the stakeholders to support improved methodologies based on current research. New ideas can take time and multiple levels of adoption including delivery and support of the innovation to reduce barriers (Dearing & Cox, 2018). If patients are aware of the negative health outcomes at a particular point in their life, perhaps they can adopt a healthier lifestyle.

To be successful in implementation, additional steps may need to be taken early in the process to increase the chances it will be perceived positively. The goal is to be able to implement practices that lead to positive health outcomes and incorporate research to practice adaptations. Research has indicated that after menopause, metabolic changes may induce excess abdominal adiposity and other chronic conditions (Dearing & Cox, 2018). If women are aware of possible negative health outcomes, they may work on researched base medical practices that may improve the quality of life.

Limitations of the Study

The findings in this research study expanded the knowledge on menopausal women and chronic disease such as MetS and NAFLD. However, there are limitations that were observed in the current study. Even though TE is a reliable test to detect NAFLD, it is not the gold standard. The gold standard in diagnosing NAFLD is a liver biopsy. A liver biopsy is a risky invasive procedure with high costs in medical bills. The patients in this study only had the TE performed and not a liver biopsy, so there may have been some misdiagnosis. Furthermore, there could have been some patients that did in fact have NAFLD and the TE did not detect it, like a liver biopsy would have. Furthermore, many of these patients may not have been correctly diagnosed with MetS, because they did not have all five conditions that are required for the diagnosis.

Women was the main population of interest in this dissertation and represented by being 52 years or older. In the data set used, there was not specific data available for just women who went through menopause. With data limited to an average age, this study may have missed those individuals who went through menopause before age of 52. In this case, the association of MetS and NAFLD may have left out many women who did not fall into this age group.

Recommendations and Implications for Positive Social Change

The main study age group was limited to 52 years or older. It is important to get a data set that contains all women who have gone through menopause regardless of age. Thus, further research should capture women who has gone through menopause. Moving forward, I believe that NHANES should create a specific question on menopausal status. In addition, based on the analysis findings in Chapter 4 and 5, it is important to invest in certain ethnicities. Even though ethnicity was not significant overall, it was significant primarily for Mexican Americans.

Moving forward, this study has the potential to induce social change and create awareness of these chronic diseases. MetS and NAFLD can compromise the quality of life for a patient and for the most part is preventable. Adopting proactive and preventative strategies can improve the quality of life for vulnerable patients. Research into better screening strategies to help eliminate the morbidity and mortality associated with MetS and NAFLD is essential. One important strategy could be to enhance healthcare workers' education, in line with patient education to control and prevent these diseases. Shaping public health policies to incorporate research-based evidence in practice will allow possible screenings and prevention. More research in this area is essential and can bring positive social change and improve the quality of life for many individuals.

References

Agency for Healthcare Research and Quality. (2016, November 18). *HCUP*. Agency for Healthcare Research and Quality: https://www.hcup-us.ahrq.gov/tech_assist/standarderrors/508/508course_2016.jsp

American College of Gastroenterology. (2020). *Non-alchoholic fatty liver diesease (NAFLD)*. American College of Gastroenterology: https://gi.org/topics/fatty-liver-disease-nafld/

American Diabetes Association. (2021). *Diabetes*. American Diabetes Association: https://www.diabetes.org/a1c/diagnosis

American Heart Association. (2016, October 31). *How high blood pressure is diagnosed*. American Heart Association: https://www.ahajournals.org/doi/full/10.1161/JAHA.118.008658

American Heart Association. (2020). *Understanding blood pressure readings*. American Heart Association: https://www.heart.org/en/health-topics/high-blood-pressure/understanding-blood-pressure-readings

American Heart Association. (2021, March 25). *About metabolic syndrome*. American Heart Association: https://www.heart.org/en/health-topics/metabolic-syndrome/about-metabolic-syndrome

American Liver Foundation. (2021). *NASH definition & prevalence*. American Liver Foundation: https://liverfoundation.org/for-patients/about-the-liver/diseases-of-the-liver/nonalcoholic-steatohepatitis-information-center/nash-definition-prevalence/

Bello, N., Schwartz, J., Kronish, I., Oparil, S., Anstey, E., Wei , Y., Cheung, Y., Munter, P., & Shimbo, D. (2018). Number of measurements needed to obtain a reliable estimate of home blood pressure: results fthe improving the detection of hypertension study. *Journal of the American Heart Association*, *7(20),* Article e008658. https://doi.org/10.1161/JAHA.118.008658, Vol 7 No. 20.

Better Health. (2018, December 2). *Menopause and weight gain*. Better Health: https://www.betterhealth.vic.gov.au/health/conditionsandtreatments/menopause-and-weight-gain

BMJ Best Practice. (2021, October 23). *Hepatic steatosis*. BMJ Best Practice: https://bestpractice.bmj.com/topics/en-us/796

Centers for Disease Control and Prevention. (2017, January). *MEC laboratory procedures Manual*. Centers for Disease and Prevention: https://wwwn.cdc.gov/nchs/data/nhanes/2017-2018/manuals/2017_MEC_Laboratory_Procedures_Manual.pdf

Centers for Disease Control and Prevention. (2018, November 6). *How the national health and nutrition examination survey keeps your information confidential*. Centers for Disease Control and Prevention: https://wwwn.cdc.gov/nchs/nhanes/continuousnhanes/documents.aspx?BeginYear=2017

Centers for Disease Control and Prevention. (2018, February). *Liver ultrasound transient elastography procedures manual*. Centers for Disease Control and Prevention: https://wwwn.cdc.gov/nchs/data/nhanes/2017-2018/manuals/2018_Liver_Ultrasound_Elastography_Procedures_Manual.pdf

Centers for Disease Control and Prevention. (2018, January 1). *Module 2: study design*. Centers for Disease Control and Prevention: https://wwwn.cdc.gov/nchs/nhanes/tutorials/module2.aspx

Centers for Disease Control and Prevention. (2018, January 1). *Module 3: weighing data NHANES*. Centers for Disease Control and Prevention: https://wwwn.cdc.gov/nchs/nhanes/tutorials/module3.aspx

Centers for Disease Control and Prevention. (2019, June). *NHANES blood pressure procedures manual*. Centers for Disease Control and Prevention: https://wwwn.cdc.gov/nchs/data/nhanes/2019-2020/manuals/2019-Blood-Pressure-Procedures-Manual-508.pdf

Centers for Disease Control and Prevention. (2020, January 1). *Section 7: measures of spread*. Centers for Disease Control and Prevention: https://www.cdc.gov/csels/dsepd/ss1978/lesson2/section7.html

Centers for Disease Control and Prevention. (2020, September 17). *Waist circumference*. Centers for Disease Control and Prevention: https://www.cdc.gov/healthyweight/assessing/index.html

Centers for Disease Control and Prevention. (2021, August 10). *Diabetes tests*. Centers for Disease Control and Prevention: https://www.cdc.gov/diabetes/basics/getting-tested.html

Chen, Y., Peng, Q., Yang, Y., Zheng, S., Wang, Y., & Lu, W. (2019). The prevalence and increasing trends of overweight, general obesity, and abdominal obesity among Chinese adults: a repeated cross-sectional study. *BMC Public Health, 19(1),* 1293. https://doi.org/10.1186/s12889-019-7633-0

Choi, D., Choi, S., Son, J., Woo Oh, S., & Park, S. (2019). Impact of discrepancies in general and abdominal obesity on major adverse cardiac events. *Journal of American Heart Association, 8(18),* Article e013471. https://doi.org/10.1161/jaha.119.013471.

Cleveland Clinic . (2021, October 5). *Postmenopause*. Cleveland Clinic : https://my.clevelandclinic.org/health/diseases/21837-postmenopause

Da-Zhi, C., Xu, Q., Wu, X., Chao, C., Zhang, L., Shi, K., Shi, H., & Li, L. (2018). The combined effect of nonalcoholic fatty liver disease and metabolic syndrome on osteoporosis in postmenopausal females in eastern china. *International Journal of Endocrinology, 2018*, Article e2314769, 8. https://doi.org/10.1155/2018/2314769.

Dearing, J., & Cox, J. (2018). Diffusion of innovations theory, principles, and practice. *Health Affairs, 37(2).* https://doi.org/10.1377/hlthaff.2017.1104.

Delahanty, L., Riggs, M., Klioze, S., Chew, R., England, R., & Digenio, A. (2016). Maximizing retention in long-term clinical trials of a weight loss agent: use of a dietitian support team. *Obesity Science & Practice*, *2(3),* 256-265. https://doi.org/10.1002%2Fosp4.57.

El Khoudary, S. A., Aggarwal, B., Beckle, T., Hodis, H., Johnson, A., Langer, R., Limacher,M., Manson, J., Stefanick, M., & Allison, M. (2020). Menopause transition and cardiovascular disease risk: implications for timing of early prevention: A scientific statement from the American Heart Association. *AHA Journals: Circulation*, *142(25),* Article e506-e532. https://doi.org/10.1161/CIR.0000000000000912.

Elfil, M., & Negida, A. (2017). Sampling methods in clinical research; an educational review. *Emerg (Tehran)*, *5(1),* Article e52. PMC5325924

Engin, A. (2017). The definition and prevalence of obesity and metabolic syndrome. *Advances in Experimental Medicine and Biology, 960*, 1-17. https://doi.org/10.1007/978-3-319-48382-5_1

Franks, P., & Atabaki-Pasdar, N. (2017). Causal interence in obesity research. *Journal of Internal Medicine*, *281(3)*, 222-232. https://doi.org/10.1111/joim.12577

Godoy-Matos, A., Junior, W., & Valerio, C. (2020). NAFLD as a continuum: from obesity to metabolic syndrome and diabetes. *Diabetology & Metabolic Syndrome*, *60(2020),* 12. https://doi.org/10.1186/s13098-020-00570-y.

Golabi, P., Otgonsuren, M., Avila, L., Sayiner, M., Rafiq, N., & Younossi, Z. (2018). Components of metabolic syndrome increase the risk of mortality in nonalcoholic fatty liver disease (NAFLD). *Medicine*, *97(13),* Article e0214. https://doi.org/10.1097/MD.0000000000010214

Grant, S., Hickey, G., & Head, S. (2019). Statistical primer: multivariable regression considerations and pitfalls. *European Journal of Cardio-Thoracic Surgery*, 55(2), 179-185. https://doi.org/10.1093/ejcts/ezy403.

Hajia, M. (2019). Secondary use of laboratory data: potentialities and limitations. *Iranian Journal of Pathology*, *14(3)*, 188-192. https://doi.org/10.30699/IJP.2019.95692.1942

Hasan, N. (2020, July 8). *Logistic regression using SPSS*. University of Miami: Statistical Supporting Unit. https://sites.education.miami.edu/statsu/wp-content/uploads/sites/4/2020/07/Logistic-Regression-Webinar.pdf

Healthy People 2020. (2021, December 28). *National health and nutrition examination survey*. Healthy People 2020: https://www.healthypeople.gov/2020/data-source/national-health-and-nutrition-examination-survey

Hirsova, P., Bohm, F., Dohnalkova, E., Nozickova, B., Heikenwalder, M., Gores, G., & Weber, A. (2020). Hepatocyte apoptosis is tumor promoting in murine nonalcoholic steatohepatitis. *Cell Death & Disease, 80 (2020*), 11. https://doi.org/10.1038/s41419-020-2283-9.

IBM. (2021, January 1). *IBM SPSS Complex Samples 28.* IBM: https://www.ibm.com/docs/en/SSLVMB_28.0.0/pdf/IBM_SPSS_Complex_Samples.pdf

Interaction Design Foundation. (2021). *Adopter categories for new products*. Interaction Design Foundation: https://www.interaction-design.org/literature/topics/adopter-categories-for-new-products

Iqbal, M., & Zahidie, A. (2021). Diffusion of innovations: a guiding framework for public health. *Scandinavian Journal of Public Health*, *50(5).* https://doi.org/10.1177%2F14034948211014104.

Jakhete, N., Jesudian, A., & Kwo, P. (2020, January 1). *Non-alcoholic fatty liver disease (NAFLD)*. American College of Gastroenerology: https://gi.org/topics/fatty-liver-disease-nafld/

Jiang, B., Gu, T., Zhou, K., Zheng, Y., Guo, Y., & Lu, Y. (2018). Fatty liver as a potential surrogate for waist circumference in the diagnosis of metabolic syndrome: A population-based study among Chinese adults. *International Journal of Endocrinology*, 2018, Article e7903982. https://doi.org/10.1155/2018/7903982.

John Hopkins Medicine. (2022). *Estrogen's effects on the female body*. John Hopkins Medicine: https://www.hopkinsmedicine.org/health/conditions-and-diseases/estrogens-effects-on-the-female-body

Johns Hopkins Medicine. (2021, October 1). *Nonalchoholic fatty liver disease*. Johns Hopkins Medicine: https://www.hopkinsmedicine.org/health/conditions-and-diseases/nonalcoholic-fatty-liver-disease

Kim, H., Kim, J., Shin, G., Han, J., & Kim, J. (2019). Association between abdominal obesity and cardiovascular risk factors in adults with normal body mass index: based on the sixth Korea national health and nutrition examination survey. *Journal Obesity Metabolic Syndrome, 28(4),* 262-272. https://doi.org/10.7570/jomes.2019.28.4.262

LaMorte, W. (2019, September 9). *Diffusion of innovation theory*. Boston University School of Public Health: https://sphweb.bumc.bu.edu/otlt/mph-modules/sb/behavioralchangetheories/behavioralchangetheories4.html

Lim, S., Kim, J., & Targher, G. (2021). Links between metabolic syndrome and metabolic dysfunction-associated fatty liver disease. *Trends in Endocrinology & Metabolism*, *32(7)*, 500-514. https://doi.org/10.1016/j.tem.2021.04.008.

Lin, I., Lee, M., Wang, C., Wu,W., & Chen, S. (2021). Gender differences in the relationships among metabolic syndrome and various obesity-related indices with nonalcoholic fatty liver disease in a Taiwanese population. *International Journal of Environmental Research and Public Health*, *18(3),* 857. https://doi.org/10.3390/ijerph18030857

Lindenmeyer, C., & McCullough, A. (2018). The natural history of nonalcoholic fatty liver disease - an evolving view. *Clinical Liver Disease*, *22(1),* 11-21. https://doi.org/10.1016/j.cld.2017.08.003

Lonardo, A., & Suzuki, A. (2020). Sexual dimorphism of NAFLD in adults. focus on clinical aspects and implications for practice and translational research. *Journal of Clinical Medicine, 9(5),* 1278. https://doi.org/10.3390/jcm9051278

Lopez-Amador, N., Nolasco-Hipolito, C., Rojas, M., & Carvajal, O. (2017). Liver enzymes in patients diagnosed with non-alcoholic fatty liver disease (NAFLD) in Veracruz: a comparative analysis with the literature. *Clinical Investigation*, *7(1).* https://doi.org/10.4172/Clinical-Investigation.1000107

Marongiu, R. (2019). Accelerated ovarian failure as a unique model to study peri-menopause influence on Alzheimer's disease. *Aging Neuroscience, 11(2019),* 242. https://doi.org/10.3389/fnagi.2019.00242.

Mayo Clinic. (2021, August 18). *Liver function tests*. Mayo Clinic: https://www.mayoclinic.org/tests-procedures/liver-function-tests/about/pac-20394595

Medline Plus. (2021, September 27). *Triglycerides*. Medline Plus: https://medlineplus.gov/triglycerides.html

MedlinePlus. (2021, September 9). *Elastography*. MedlinePlus: https://medlineplus.gov/lab-tests/elastography/

Memorial Sloan Kettering Cancer Center. (2018, February 27). *Understanding your fibroscan results*. Memorial Sloan Kettering Cancer Center: https://www.mskcc.org/cancer-care/patient-education/understanding-your-fibroscan-results

Mittal, B. (2019). Subcutaneous adipose tissue. *Indian Journal of Medical Research*, *149(5*), 571-573. https://doi.org/10.4103/ijmr.IJMR_1910_18

Mohajan, H. (2017, October 1). *Two criteria for good measurements in research: validity and reliability*. Munich Personal RePEc Archive. *17(3*), 58-82. https://mpra.ub.uni-muenchen.de/83458/1/MPRA_paper_83458.pdf

Moriles, K., & Azer, S. (2021, May 9). *Alanine amino transferase*. Treasure Island: StatPearls Publishing. https://pubmed.ncbi.nlm.nih.gov/32644704/

National Health and Nutrition Examination Survey. (2017, January). *Anthropometry procedures manual*. Centers for Disease Control and Prevention: https://wwwn.cdc.gov/nchs/data/nhanes/2017-2018/manuals/2017_Anthropometry_Procedures_Manual.pdf

National Health and Nutrition Examination Survey. (2018, February). *Liver ultrasound transient elastography procedures manual*. National Health and Nutrition Examination Survey: https://wwwn.cdc.gov/nchs/data/nhanes/2017-2018/manuals/2018_Liver_Ultrasound_Elastography_Procedures_Manual.pdf

National Health and Nutrition Examination Survey. (2018). *NHANES 2017-2018 procedure manuals*. Centers for Disease Control and Prevention: https://wwwn.cdc.gov/nchs/data/nhanes/2017-2018/manuals/2017_MEC_Laboratory_Procedures_Manual.pdf

National Health and Nutrition Examination Survey. (2019, June). *Blood pressure procedures manual*. Centers for Disease Control and Prevention: https://wwwn.cdc.gov/nchs/data/nhanes/2019-2020/manuals/2019-Blood-Pressure-Procedures-Manual-508.pdf

National Health and Nutrition Examination Survey. (2020). *NHANES 2017-2018*. Centers for Disease Control and Prevention: https://wwwn.cdc.gov/nchs/nhanes/search/datapage.aspx?Component=Laboratory&CycleBeginYear=2017

National Health Service. (2019, February 1). *Why it's necessary for a hysterectomy*. National Health Service: https://www.nhs.uk/conditions/hysterectomy/why-its-done/

National Heart, Lung, and Blood Institute. (2020, May 28). *Metabolic syndrome*. National Heart, Lung, and Blood Institute: https://www.nhlbi.nih.gov/health-topics/metabolic-syndrome

National Institute of Diabetes and Digestive and Kidney Diseases. (2021, April). *Diagnosis of NAFLD & NASH*. National Institute of Diabetes and Digestive and Kidney Diseases: https://www.niddk.nih.gov/health-information/liver-disease/nafld-nash/diagnosis

National Institute on Aging. (2021, September 20). *What is menopause?* National Institute on Aging: https://www.nia.nih.gov/health/what-menopause

Office on Women's Health. (2019, March 18). *Menopause basics*. Office on Women's Health: https://www.womenshealth.gov/menopause/menopause-basics

Patino, C., & Ferreira, J. (2018). Internal and external validity: can you apply research study results to your patients? *Jornal Brasileiro de pneumologia*, *44(3)*,183. https://doi.org/10.1590/s1806-37562018000000164

Ping, Z., Pei, X., Xia, P., Chen, Y., Guo, R., Hu, C., Imam, M., Chen, Y., Sun, P., & Liu, L. (2018). Anthropometric indices as surrogates for estimating abdominal visceral and subcutaneous adipose tissue: A meta-analysis with 16,129 participants. *Diabetes Research and Clinical Practice*, *2018(143),* 310-319. https://doi.org/10.1016/j.diabres.2018.08.005

Popa, S., Mota, M., Popa, A., Mota, E., Serafinceanu, C., Guja, C., Catrinoiu, D., Hancu, N., Lichiardopol, R., Bala, C Popa, A., Roman, G., Radulian, G., Timar, R., & Mihai, B. (2016). Prevalence of overweight/obesity, abdominal obesity and metabolic syndrome and atypical cardiometabolic phenotypes in the adult Romanian population: PREDATORR study. *Journal Endocrinology Investigates*, *39(9)*,1045-53. https://doi.org/10.1007/s40618-016-0470-4

Puterbaugh, J. (2016). Reducing societal obesity: establishing a separate exercise model through studies of group behavior. *Journal of Obesity*, *2016(2016),* Article e6734043. https://doi.org/10.1155/2016/6734043

Robeva, R., Mladenovic, D., Veskovic, M., Hrncic, D., Bjekic-Macut, J., Stanojlivic, O., Livadas, S., Yildiz, B., & Macut, D. (2021). The interplay between metabolic dysregulations and non-alcoholic fatty liver disease in women after menopause. *Elsevier*, *151*, 22-30. https://doi.org/10.1016/j.maturitas.2021.06.012.

Ross, R., Neeland, I., Yamashita, S., Shai, I., Seidell, J., Magni, P., Santos, R., Arsenault, B., Cuevas, A., Hu, A., Griffin, B., Zambon, A., Barter, P., Fruchart, J., Eckel, R., Matsuzawa, Y., & Despres, J. (2020). Waist circumference as a vital sign in clinical practice: a consensus statement from the IAS and ICCR working group on visceral obesity. *Nature Reviews Endocrinology*, *16(2020),* 177-189. https://doi.org/10.1038/s41574-019-0310-7

Ruggiano, N., & Perry, T. (2017). Conducting secondary analysis of qualitative data: Should we, can we, and how? *Sage Journals*, *18(1).* https://doi.org/10.1177/1473325017700701.

Setroame, A., Affrim, P., Yawson, A., Kwadzokpui, P., Adrah, F., Bless, H., Mohammed, L., Bawah, A., & Alidu, H. (2020). Prevalence of metabolic syndrome and nonalcoholic fatty liver disease among premenopausal and postmenopausal women in Ho Municipality: a cross-sectional study. *BioMed Research International, 2020*, Article 2168381, 9. https://doi.org/10.1155/2020/2168381.

Shin-Yu, A., & Jiang, Y. (2017). Integration of diffusion of innovation theory into diabetes care. *Journal of Diabetes Investigation, 8(3),* 259-260. https://doi.org/10.1111/jdi.12568

Shmerling, R. (2020, October 2). *Metabolic syndrome is on the rise: What it is and why it matters*. Harvard Health Publishing Harvard Medical School: https://www.health.harvard.edu/blog/metabolic-syndrome-is-on-the-rise-what-it-is-and-why-it-matters-2020071720621

Sigit, F., Tahapary, D., Trompet, S., Sartono, E., Van Dijk, K., Rosendaal, F., & De Mutsert, R. (2020). The prevalence of metabolic syndrome and its association with body fat distribution in middle-aged individuals from Indonesia and the Netherlands: a cross-sectional analysis of two population-based studies. *Diabetology & Metabolic Syndrome, 12(2020)*, 2. https://doi.org/10.1186%2Fs13098-019-0503-1.

Statistics Solutions. (2022). *SPSS statistics help*. Complete Dissertation by Statistics Solutions: https://www.statisticssolutions.com/academic-research-consulting/dissertation-consulting-services/spss-statistics-help/

Stewart, R., Beidas, R., & Mandell, D. (2019). Stop calling them laggards: strategies for encouraging nonadopters to incorporate evidence-based practices. *The American Psychiatric Association, 70(10),* 958-960. https://doi.org/10.1176/appi.ps.201900031.

Tanaka, N., Kimura, T., Fujimori, N., Nagaya, T., Komatsu, M., & Tanaka, E. (2019). Current status, problems, and perspectives of non-alcoholic fatty liver disease research. *World Journal of Gastroenterology*, *25(2*), 163-177. https://doi.org/10.3748/wjg.v25.i2.163.

Tantanavipas, S., Vallibhakara, O., Sobhonslidsuk, A., Phongkitkarun, S., Valibhakara, S., Promson, K., & Sophonsritsuk, A. (2019). Abdominal obesity as a predictive factor of nonalcoholic fatty liver disease assessed by ultrasonography and transient elastography in polycystic ovary syndrome and healthy women. *BioMed Research International*, *2019(9047324)*, 9. https://doi.org/10.1155/2019/9047324

The North American Menopause Society. (2021). *Menopause guidebook*. The North American Menopause Society: https://www.menopause.org

The Translational Genomics Research Institute. (2020, August 17). *Postmenopausal women at risk for nonalcoholic fatty liver disease, review suggests*. Science Daily: https://www.sciencedaily.com/releases/2020/08/200817191747.htm

Tiwari, A., & Balasundaram, P. (2022). Public health considerations regarding obesity. *Public Health Considerations Regarding Obesity*. In StatPearls. StatPearls Publishing. https://www.ncbi.nlm.nih.gov/books/NBK572122/). Treasure Island: StatPearls Publishing.

University of Rochester Medical Center. (2021). *Lipid panel with total cholesterol: HDL ratio*. University of Rochester Medical Center: https://www.urmc.rochester.edu/encyclopedia/content.aspx?ContentTypeID=167&ContentID=lipid_panel_hdl_ratio

Venetsanaki, V., & Polyzos, S. (2019). Menopause and non-alcoholic fatty liver disease: A review focusing on therapeutic perspectives. *Current Vascular Pharmacology, 17(6),* 546-555. https://doi.org/10.2174/1570161116666180711121949.

Yang, K., Hung, H., Lu, C., Chang, H., Lee, L., & Huang, K.-C. (2016). Association of non-alcoholic fatty liver disease with metabolic syndrome independently of central obesity and insulin resistance. *Scientific Reports 6*, Article e27034. https://doi.org/10.1038/srep27034

Yan-Lan, F. E. (2018). Pathogenesis of non-alcoholic fatty liver disease in children and adolescence: From “two hit theory” to “multiple hit model”. *World Journal of Gastroenterology, 24(27),* 2974-2983. https://doi.org/10.3748/wjg.v24.i27.2974

Young, S., Tariq, R., Provenza, J., Satapathy, S., Faisal, K., Choudhry, A., Friedman, S., & Singal, A. (2020). Prevalence and profile of nonalcoholic fatty liver disease in lean adults: systematic review and meta-analysis. *Hepatology Communication, 4(7),* 953-972. https://doi.org/10.1002/hep4.1519.

Yue, W., Sun, X., & Du, T. (2019). Cholecystectomy versus central obesity or insulin resistance in relation to the risk of nonalcoholic fatty liver disease: the third US National Health and Nutrition Examination Survey. *PubMed, 19(1),95.* https://doi.org/10.1186/s12902-019-0423-y

Yu-ling, C., Li, H., Li, S., Xu, Z., Tian, S., Wu, J., Liang, X., Li, X., Liu, Z., Xiao, J., Wei, J., Ma, C., Wu, K., Ran, L., & Kong, L. (2021). Prevalence of and risk factors for metabolic associated fatty liver disease in an urban population in China: a cross-sectional comparative study. *BMC Gastroenterology*, *21(212)*. https://doi.org/10.1186/s12876-021-01782-w.

Zou, D., Lloyd, J., & Baumbusch, J. (2020). Using SPSS to analyze complex survey data: A primer. *Journal of Modern Applied Statistical Methods*, *18*(1), 2 – 22. https://doi.org/10.22237/jmasm/1556670300.