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The Relationship between Selected Nutrients and Depression among Smokers

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I. Introduction

In 2006, 20.2% of Americans were classified as cigarette smokers, with a median smoking prevalence of 22.2% for men and 18.5% for women. Every year, more than 438,000 Americans die from smoking cigarettes even though smoking remains the most preventable cause of death in the United States (MMWR, 2007). According to the Surgeon General's Report regarding the health consequences of cigarette smoking, smoking causes a variety of diseases and harms every organ of the body. Despite this, the Surgeon General states that quitting smoking has immediate benefits and can ultimately reduce the risk of developing these diseases (U.S. Department of Health and Human Services, 2004). Because of these findings, research regarding the co-occurring problems that smokers face, such as depression and nutritional deficits, is a necessity.

It is estimated that millions of Americans suffer from depression, leading to both physical and psychological ailments (NASD, 2002). While the cause of depression remains unclear, it has been proposed that many factors influence the occurrence of depressive symptoms (Brown, Madden, Palenchar, & Cooper-Patrick, 2000). In recent studies, a correlation has been found between smoking status and depressive symptoms with an increased prevalence of depressive symptoms among smokers, especially among women (Brown, Madden, Palenchar, & Cooper-Patrick, 2000; Son, Markovitz, Winders, & Smith, 1997). In addition to this, it has also been shown that depressive symptoms have a negative impact on the smoker's ability to quit smoking (Brown, Madden, Palenchar, & Cooper-Patrick, 2000), thus leading to further health problems. This has important implications in clinical practice because it indicates the need to further investigate the relationship between smoking and other factors that may further intensify the health risks that smokers face.

Of the factors influencing depression, diminished serotonergic (5-HTergic) function is a widely accepted theory that is attributed to the onset and course of depression. The 5-HTergic system is complex and thus has a variety of clinical implications. While most of the cell bodies of 5-HTergic neurons are located in the raphe nuclei in the brain stem, the axons of these neurons innervate almost the entire brain. While the exact function of 5-HT has been difficult to define, it is

known that 5-HT is generally an inhibitory neurotransmitter involved with mood, anxiety, and sleep induction. Although strong connections between 5-HT and depression have been revealed, not all patients who have diminished 5-HTergic function show depressive symptoms and not all people with depressive symptoms have diminished 5-HTerigic function. Because of this, diminished 5HT is described as being a vulnerability factor in developing depression (Jans, Riedel, Markus, Blokland, 2007).

A variety of factors have been attributed to altered 5-HT function (Jans, Riedel, Markus, Blokland, 2007). Studies in the past have indicated that low levels of tryptophan, an essential amino acid required to produce both vitamin B3 (niacin) and 5-HT via an enzyme called tryptophan hydroxylase, have been found in people who exhibit depressive symptoms (Coppen, Eccleston, Craft & Bye, 1973; Cowen, Parry-Billings & Newsholme, 1989). Vitamin B6 (pyridoxine) is required for the conversion of tryptophan to niacin and serotonin. When vitamin B6 is deficient, the body cannot complete this conversion and the body then excretes compounds that are not fully metabolized. In addition to this, it is important to note that tryptophan converts to niacin before serotonin. If a person has a niacin deficiency, the body will use the tryptophan to make niacin to address the deficiency and not convert to serotonin, thus leading to a serotonin deficit (Kline, 2006). Smoking is thought to decrease these conversions. Because of this, taking niacin and pyridoxine intake into consideration is important when examining tryptophan intake in relation to depressive symptoms and smoking.

In addition to tryptophan, omega-3 polyunsaturated fatty acids (omega-3 PUFAs) have been proposed to influence the occurrence of depressive symptoms. Omega-3 PUFAs, which are essential fatty acids, are highly concentrated throughout the brain and central nervous system and affect neurotransmitter synthesis. Studies have indicated an inverse relationship between docosahexaenoic acid (DHA), an omega-3 PUFA, and depression, thus implicating that a long-term low dietary intake of DHA is associated with an increased incidence of depression in adults (Mamalakis et al, 2006). The INTERMAP study, which examined dietary intake in male and female smokers versus that of ex-smokers and never smokers, indicated that smokers in general have less

healthful diets than nonsmokers, with a lower intake of a variety of nutrients. Among these nutrients are omega-3 PUFAs, fiber, vitamin C, vitamin E, beta carotene, iron, and calcium (Dyer, Elliott, Stamler, Chan, Ureshima & Zhou, 2003).

It is evident that previous studies have shown that depression and nutritional alterations are common problems among smokers. Research has been focused on the relationships between depression and smoking, nutrition and smoking, and nutrition and depression, but little research has been designed to examine whether a relationship exists among these three variables. This secondary analysis aims to study the relationship between depressive symptoms among smokers and the intake of tryptophan, controlling for niacin and pyridoxine intake, and omega-3 PUFAs. Another primary aim of this study is to look at the influence nicotine dependence has on stress, depression, and the selected nutrients. A secondary aim of this study is to explore the relationship between sociodemographics, smoking level, body mass index (BMI) and depression.

II. Review of Literature

Literature searches were conducted on Medline and PubMed to identify studies relevant to this hypothesized relationship among smoking, depression and selected nutrients. In this review of literature, relevant studies are grouped into five categories: smoking and depressive symptoms; depressive symptoms and stress; omega-3 PUFAs and depressive symptoms; tryptophan, depressive symptoms, and smoking; and smoking and omega-3 fatty acids.

Smoking and Depressive Symptoms

Because of the high correlation between smoking and depression, various studies have been designed to further examine this relationship. In a study by Son, Markovitz, Winders, and Smith (1997), data from the Coronary Artery Risk Development in Young Adults (CARDIA) Study was used to examine whether depressive symptoms were independently associated with smoking and nicotine dependence among cigarette smokers. The sample consisted of 3,933 participants ages 23-35. Black men, black women, white men, and white women were represented in this study. The Center

for Epidemiologic Studies Depression (CES-D) Scale (Radloff, 1977) was used to measure the severity of depressive symptoms, while nicotine dependence and cigarette smoking status was determined through a questionnaire administered by an interviewer. A serum cotinine level of greater than or equal to 19 ng/ml was used to confirm smoking status. Nicotine dependence was measured by the amount of time prior to the participant smoking his or her first cigarette of the day with dependence defined as smoking a cigarette within 30 minutes of arising. Results indicated that depressive symptoms were more prevalent among smokers; however, this relationship was not statistically significant among white men. When nicotine dependence was taken into consideration, the dependent smokers in all the groups showed higher CES-D scores.

The results of this study validate the correlation between smoking and depression with a higher occurrence of depressive symptoms among those who are nicotine dependent. This study does not address the possibility that people take up smoking because they are depressed. In addition, the researchers used only one question to define nicotine dependence. Despite these limitations, although this study is cross-sectional in nature, its large sample size and statistically significant findings make it both reliable and valid. It is a well designed study that validates that smoking is related to depressive symptoms.

Like the previous study discussed, Brown, Madden, Palenchar, and Cooper-Patrick (2000) also aimed to further examine the relationship between depressive symptoms and smoking. In their study, 526 people aged 18-64 were recruited in the waiting room of various urban primary care offices. Although this sample is not as large as the sample in the CARDIA study, it includes both older and younger populations. Like the CARDIA study, the CES-D scale was used to measure depressive symptoms. Smoking status was measured using questions from a modified version of the Fagerstrom Test for Nicotine Dependence. The number of cigarettes smoked per day and the time it takes for the patient to smoke their first cigarette of the day were used to measure nicotine dependence. Similar to the CARDIA study, this study found that the severity of depressive symptoms increased with nicotine dependence.

While the Brown et al study further validates the fact that smoking is correlated with depressive symptoms, the participants did not represent the general population. The participants were primarily white females less than 50 years of age, who were educated beyond high school and had steady employment. In addition, validation of smoking status was based on self-report measures. This study failed to take into consideration other factors that could influence depressive symptoms. This is especially important because the participants in this study were seeking medical attention. The medical conditions of the participants could have had a great deal of influence on the severity of the depressive symptoms reported. Despite these limitations, the major findings of this study are congruent with the CARDIA study. This further emphasizes the correlation between smoking and depressive symptoms.

Another recent study by Kenney, Holahan, North, and Holahan (2006), sought to examine this relationship, but did not take nicotine dependence into consideration. This study, which utilized the data from the National Survey of Midlife Development in the United States, examined 2,087 American workers. The researchers measured depression using their own questions, which resembled a modified Beck Depression Inventory (Kenney et al, 2006). They measured smoking status by classifying the participant as a current smoker, a current nonsmoker, a past smoker, or a past nonsmoker. The results of this study validate the researchers' initial hypothesis that the level of depressive symptoms predicted smoking status. Unlike the CARDIA study, smoking status was determined through self report measures. In addition to this, the cross-sectional nature of this study is a limitation. However, the large sample size and statistical significance of the results increase the reliability and validity of the findings.

While the three studies discussed all have limitations, it is evident from their findings that there is a relationship between smoking and depressive symptoms. The Kenney et al study did not need to take nicotine dependence into account in order to achieve a statistically significant correlation between smoking and depressive symptoms. This contradicts the findings of the CARDIA study, in which nicotine dependence needed to be considered in order for the results to be statistically significant. Further studies that take these limitations into consideration are needed. A

study in which racial and age groups are equally represented and does not completely rely on self-report measures for smoking status would further enhance the understanding of the relationship between smoking and depressive symptoms.

Stress and Smoking

A recent study by Croghan, Bronars, Patton, Schroeder, Nirelli, & Thomas, et al (2006) sought to examine the relationship between body image, smoking, stress, and self-esteem. While it is unknown if negative mood precedes smoking or visa versa, studies have shown that smokers use smoking as a form of self-medication. Negative mood, also known as negative affect, includes symptoms such as stress, anger, sadness, distress, and low self-esteem. Croghan et al sought to identify these factors among young adults and determine if they are related to smoking status. Researchers in this study used a convenience sample of 1456 students between the ages of 18 and 24 at three Midwestern universities. Of the 1456 participants, 483 were smokers and 973 were nonsmokers. Participants were invited to take a self-administered survey that included items from the Body-Areas Satisfaction Scale (BASS), Perceived Stress Scale (PSS), Rosenberg Self-Esteem Scale (SE), and Positive and Negative Affect Schedule (PANAS), as well as questions regarding tobacco use and lifestyle choices (Croghan et al, 2006).

The results from this study indicate that smokers had higher levels of stress and lower self-esteem. In addition, multiple logistic regression analyses indicated a strong association between stress and smoking status, but not smoking status and negative affect. Researchers concluded that this may be due to the fact that negative affect may be a mediator of stress and smoking. It is also important to consider that these findings are limited due to self-report and researchers only focused on the young adult population, most of whom were white. Despite these limitations, the large sample size and statistically significant findings validate the notion that smoking, negative affect, and stress are related and warrant further study (Croghan et al, 2006).

Another study by File, Dinnis, Heard, and Irvine (2002) sought to examine sex differences in mood between light smokers and nonsmokers. The participants in the study included 36 students in

Great Britain. The subjects included 9 female smokers, 9 female nonsmokers, 8 male smokers, and 10 male nonsmokers. The smokers smoked 5-12 cigarettes per day for at least one year. Baseline anxiety and depression were measured using the Hospital Anxiety and Depression Scale. Mood and aggression baseline data were also collected. Testing was scheduled for each smoker at a time when they usually smoked and testing began 5 minutes after smoking. Volunteers were then shown 20 images, each for 3 seconds, and then were asked to recall as many as possible 20 minutes after viewing the images. Subjects were then asked to complete a digit-symbol substitution test while counting gaps in music that played in the background in order to measure psychomotor speed and attention. Other cognitive tests that were administered included matching tests and an Auditory Serial Addition Test (File et al, 2002).

Results indicated that smokers, who smoked 5 minutes before testing, were more likely to feel dizzy and had greater symptoms of sweating and dry mouth during the protocol. In addition smokers were significantly more discontented, troubled, tense, and slow. There was also significant data that supported the hypothesis that smokers were more furious, impatient, unfriendly, annoyed, and disgusted after completing the tests. The cognitive tests also caused smokers increased stress and decreased attention when compared with nonsmokers and there were no significant differences between genders. Despite these findings, the smokers performed better on the memory tests. Researchers concluded that these results are most likely a result of both genetic factors and the direct effects of nicotine, mediated by the 5-HT system (File et al, 2002).

Limitations of this study include the small sample size and the fact that the study design did not allow the researchers to determine whether the mood differences observed between the smokers and nonsmokers were due to the long-term effects of nicotine. In addition, abstinent state was not observed in the smoking group. Despite these limitations, this study provides strong evidence that supports the association between smoking and stress and suggests the need for further research on this topic (File et al, 2002).

Omega-3 PUFAs and Depressive Symptoms

Various studies have been designed to determine if a relationship exists between the intake of polyunsaturated fatty acids and depressive symptoms. A recent study by Mamalakis, Kalogeropoulos, Andrikopoulos, Hatzis, Kromhout, Moschandreas, et al. (2006) found an inverse relationship between depression and long chain n-3 PUFAs in a sample of 130 healthy adults from Crete. This is the first study designed to examine this relationship in a non-homogenous sample using these methods. Researchers used a Greek translation of the Zung Self-rating Depression Scale (ZSDS) to measure the level of depression (Fontoulakis et al, 2001). Adipose tissue samples were collected by aspirating subcutaneous buttock tissue. Adipose tissue fatty acid composition is a biomarker that indicates dietary fat intake over a one to three year period. The study's results suggest that the inverse relationship between depression and n-3 long chain fatty acids indicates a lower dietary intake of these fatty acids over time. Mamalakis, et al hypothesize that this relationship is due to the fact that fatty acids inhibit pro-inflammatory cytokine synthesis, which is associated with depression, by suppressing inflammation and immune reactivity markers.

Despite these findings, the Mamalakis, et al study does have limitations. Because this study is a cross-sectional design, the findings cannot be used to imply that there is a cause-effect relationship between depression and fatty acid intake. In addition to this, the study was composed of mostly non-depressed subjects. Mamalakis, et al do state that studies such as this have not yet been conducted in the depressed population. These limitations provide future aspects to consider for further research on this topic.

Another study, by Kiecolt-Glaser, Belury, Porter, Beversdorf, Lemeshow, and Glaser (2007), also aimed to investigate the relationship between depressive symptoms and fatty acid intake. However, this particular study aimed to examine the relationship between PUFA levels and depressive symptoms in relation to the cytokine synthesis involved in inflammation. The sample in this study consisted of 43 older adults with a median age of 66.7 years. Depressive symptoms were measured using the CES-D Scale and immunological assays and fatty acid analyses were used to measure cytokine and fatty acid levels. Despite the small sample size and cross-sectional nature of this study, results indicate that a higher ratio of omega-6:omega-3 fatty acids was correlated with

higher levels of depressive symptoms, both of which increase proinflammatory cytokine levels. This is because a higher omega-6:omega-3 ratio suggests that there is a lower level of omega-3 intake. The Keicolt-Glaser, et al study used serum PUFA levels instead of the tissue aspiration method used in the Mamalakis, et al study. Despite this, both studies indicate that there is an inverse relationship between the dietary consumption of PUFAs and depressive symptoms in adults.

Although the two previously discussed studies have shown results implying this relationship, a study by Hakkarainen, Partonen, Haukka, Virtamo, Albanes, and Lönnqvist (2004) found no relationship between mood level and dietary intake of omega-3 fatty acids. In this study, the sample consisted of 29,133 men ages 50-69 in Finland. Data were used from the previously conducted ATBC Cancer Prevention Study, which was a randomized, double-blind, placebo-controlled study. Although this study was well designed, the measurements used in the Hakkarainen, et al study significantly weaken the results. Depressive symptoms were measured by asking the participants if they felt symptoms of depressed mood or anxiety and were recorded three times per year. History of major depression and suicide, as it appeared on a death certificate, were also taken into consideration. Omega-3 fatty acids were measured by a food frequency questionnaire that asked the participants to recall their dietary intake over a year. There was no scale used to measure depressive symptoms and no patient history was taken into consideration besides history of treatment for depressive symptoms and the presence of suicide on a death certificate. Although the randomized, double-blind, placebo-controlled design of this study is stronger than the Mamalakis, et al and the Keicolt-Glaser, et al study, the measures were weaker overall. Because of this, it is inconclusive whether or not there is a relationship between omega-3 fatty acids and depressive symptoms. More research needs to be completed in this area before a true relationship can be confirmed.

Tryptophan, Depressive Symptoms, and Smoking

Similarly, there has been a hypothesized inverse correlation between tryptophan levels and depression, with smoking exacerbating this relationship. To first address the link between tryptophan

and depression, tryptophan is an essential amino acid that plays a vital role in brain processes. Both vitamin B3 (niacin) and the neurotransmitter serotonin are synthesized in the brain from tryptophan, with the help of vitamin B6 (pyridoxine). Tryptophan, being an essential amino acid, must be supplied to the body through dietary intake. Because of this, according to the Booij, et al (2005) study, acute tryptophan depletion (ATD) is a method that displays the effects of decreased levels of serotonin in the brain in 50-60% of patients who are depressed and taking serotonergic antidepressants to treat their depression. This is because depleting tryptophan levels ultimately depletes serotonin and diminished serotonin levels are highly associated with depression. Because of this relationship, ATD in experimental conditions causes these patients to display symptoms of relapse in their depressive symptoms even while on medication.

The added element of smoking in this relationship was recently publicized. A recent study by Spring, et al (2007), reported that smoking magnified this ATD response in those with a history of major depression. Researchers recruited a total of 73 subjects with and without histories of smoking and major depression. A 21-item Hamilton Depression scale was used to evaluate level of depression and personal and familial history of depression were obtained (Hamilton, 1967). Smoking was defined as smoking more than ten cigarettes per day for at least one year. In order to have a history of major depression, personal and familial major depressive disorder was necessary. All subjects followed a low tryptophan diet and smokers were encouraged to smoke regularly in order to prevent nicotine withdrawal during the acute study. One group was given a placebo, while the other group was given an ATD drink to decrease tryptophan levels in the body. Five hours after being given the drinks, the subjects were exposed to negative guided imagery and mood was assessed. Results demonstrated that smoking heightened depressive symptoms in response to acute reduced tryptophan in subjects with a history of major depressive disorder (Spring, et al, 2007).

A double-blind study by Pergadia, Spring, Konopka, Twardowska, Shirazi, & Crayton (2004) reported similar findings. Seven smokers with and without a history of depression participated, were recruited via advertisements, and smoked an average of 18.9 cigarettes per day. The subjects were

asked to consume a tryptophan-depleting drink or a similar tasting placebo on two days separated by a one week interval. Following consumption, they were asked to sit in a waiting area and were allowed to read neutral material and smoke as they pleased. Five hours after consumption, participants listened to negative material via headphones. They were asked to respond to a Modified Hamilton Rating Scale for Depression (Hamilton, 1960) at three, five, and seven hours after consumption of the drinks. To be sure that the participants were smoking as usual, exhaled carbon monoxide levels were taken every two hours. Brain scans were also completed during the testing period. Despite the extremely small sample size, the Pergadia, et al study concluded that tryptophan depletion caused depressed mood in smokers with and without a history of major depression. Although these findings differ slightly from the Spring et al study in which depressed mood was only seen in smokers with a history of depression, they verify the relationship between tryptophan, depression, and smoking.

Smoking and Omega-3 Fatty Acids

Various studies have aimed to look at smoking and nutrition. In recent analyses, smokers have been determined to have less than adequate dietary intakes of a variety of nutrients. A recent study by Dyer, Elliott, Stamler, Chan, Ureshima & Zhou (2003), examined the diet of smokers in relation to nonsmokers. This study examined the dietary intake in 4680 participants in four countries using data from the INTERMAP study, which is an epidemiologic study that examined the relationship between nutrients and other dietary factors. Using 24-hour recalls, this study indicated that smokers in general have less healthful diets than nonsmokers, with a lower intake of a variety of nutrients, such as omega-3 PUFAs, fiber, vitamin C, vitamin E, beta carotene, iron, and calcium. This study reports that these findings are consistent with many previous studies that have aimed to examine the relationship between select nutrients and smoking status.

A study by Marangoni, Colombo, DeAngelis, Cambaro, Agostoni, Giovannini, and Galli (2004) aimed to look at the metabolic aspect of this problem in a cellular model. Researchers used healthy human mammary gland cells and observed their characteristics under normal conditions.

Cigarette smoke was introduced to these cells through aspiration and a pump. Interpretation of this data indicates that cigarette smoke negatively affects omega-3 fatty acid synthesis.

Another study by Pawlosky, Hibbeln, & Salem (2007), characterized a broader aspect of the relationship between omega-3 PUFAs and smoking. Because smoking increases circulating free radicals and polyunsaturated fatty acids (PUFAs) have been determined to be highly susceptible to free radical oxidation, it has been hypothesized that chronic smoking may decrease the amount of PUFAs in plasma and red blood cells. The study by Pawlosky et al (2007) examined the effects of smoking on omega-3 fatty acid metabolism in 5 female nonsmokers, 5 female smokers, 5 male nonsmokers, and 5 male smokers. For 3 weeks, subjects ate a beef-based diet provided by the research kitchen. During the final week of the study, after an overnight fast, subjects were administered 1 g of α-Linolenic acid, the most abundant omega-3 fatty acid in the North American diet, in low fat yogurt prior to their morning meal. With the exception of the 8 hour sample, 40 mL of blood was taken at 24, 48, 72, 96, and 160 hours under fasting conditions. Plasma fatty acids were analyzed using gas chromatography and GC-mass spectrometry and concentration-time curves were used to assess kinetic rate parameters. Results from the study showed that smoking accelerated the metabolism of omega-3 fatty acids. These findings suggest that smokers would therefore need to consume more omega-3 fatty acids than nonsmokers.

Although the sample size of the Pawlosky et al (2007) study was relatively small, it further examined the relationship suggested by Marangoni et al (2004). Because of the results implied by these two studies, it may be suggested that two factors play into the lower omega-3 fatty acid levels in smokers. The first of these factors is that smokers tend to consume less omega-3 fatty acids than nonsmokers. The second of these factors is that cigarette smoke alters PUFA synthesis because of the free radicals that result from smoking, thus affecting PUFA metabolism on the cellular level. Because of these findings, studying the general relationship between smoking and omega-3 fatty acid intake is essential in order to determine the implications of this relationship. The added impact of the inverse relationship between depression and long chain n-3 PUFAs may also affect these variables.

Conclusion

The studies presented in this review of literature imply that various relationships exist between smoking, depression, stress and the selected nutrients. Research has demonstrated an increased incidence of depressive symptoms and stress among smokers across both genders and all races. In general, studies have indicated that smokers have less than adequate intake of a variety of nutrients, especially omega-3 fatty acids and tryptophan, which have been shown to have an inverse relationship with depressive symptoms. Because there is no research regarding the interaction of smoking, depression, stress, and these selected nutrients, this secondary analysis aims to study the interactions between these four variables.

III. Methods:

This secondary analysis aims to analyze four main research questions: (1) What is the relationship between depressive symptoms among smokers and the intake of tryptophan, controlling for niacin and pyridoxine intake? (2) What is the relationship between depressive symptoms among smokers and the intake of omega-3 PUFAs? (3) Of tryptophan intake, omega-3 PUFA intake, and smoking level, what is the best predictor of depression? (4) Is there a significant difference in tryptophan intake, omega-3 intake, depressive symptoms, and perceived stress between nicotine dependent smokers and non-nicotine dependent smokers? A secondary aim of this study is to explore the relationship between sociodemographics, smoking level, body mass index (BMI) and depression.

It is hypothesized that smokers with increased depressive symptoms will have lower than the recommended levels of these nutrients in their diets and that nicotine dependent smokers will have greater depressive symptoms, higher perceived stress, and lower than the recommended dietary intake of tryptophan and omega-3 fatty acids.

Theoretical Framework

This study uses the biobehavioral approach to examine the relationship between nutrition and depression among smokers. This is a multidisciplinary approach that seeks to examine the biological and behavioral determinants of health status as well as the effects of biological and psychosocial influences on behavior (Kozlowski, Henningfield, & Brigham, 2001).

Design

This study uses a descriptive, correlational design in which a correlation matrix will be used to determine any relationships that exist among smoking, depression, tryptophan, omega-3 PUFAs, and antioxidants.

Participants

The primary study aimed to examine the effects of ethnicity and menthol/non-menthol cigarette preference on nicotine dependence, smoking topography, and cigarette smoke constituent exposure. The inclusion criteria included 136 African American and Caucasian male or female smokers 18-50 years of age who identified themselves as a person who has smoked 10-40 cigarettes/day for one year. Women were to be in the mid to late follicular phase of the menstrual cycle, confirmed by progesterone levels. Exclusion criteria included other forms of tobacco use, taking regular prescription medications, history of liver, endocrine or pulmonary disease, drug or alcohol abuse, and pregnant or lactating women. Health status was determined upon admission via a detailed history and physical examination. Recruitment of the sample in the primary study involved placing advertisements in neighborhood newspapers and informational flyers in work settings, community centers and clinics. Subjects were paid $250 to participate in the primary study.

Sample Size Justification

For the correlational analysis involving Pearson's r, a sample size of 85 will have an 80% power to detect a medium effect at α=0.05. For the multiple regression analysis with three predictors (smoking, tryptophan and omega-3 PUFAs), a sample size of 76 will have an 80% power

to detect a medium effect at α=0.05. The probable sample size of 136 subjects more than satisfies this requirement (Cohen, 1992).

Procedure

In the primary study, adult cigarette smokers who met the inclusion criteria participated in a 36-hour protocol at the Ohio State University Clinical Research Center (CRC). During this time, measures of nicotine dependence, nicotine and carbon monoxide increases pre to post-cigarette, cotinine concentrations, sensory perceptions post-cigarette, and smoking topography were obtained while the participants smoked their usual brand of cigarettes ad lib. Twenty-four hour urine samples were also obtained to analyze cigarette smoke constituents of menthol and its metabolite, tobacco specific nitrosamines, and polycyclic aromatic hydrocarbons.

Instruments

For this secondary analysis, four measures were used to examine the research questions. The plasma nicotine and cotinine levels were assayed by high performance liquid chromatography-tandem mass spectrometry (LC-MS/MS) at Mayo Laboratories. The LC-MS/MS method has a coefficient of variation for both intra-assays and inter-assays of nicotine and cotinine ranging from 4.9% to 11% with excellent correlations (r>0.98) to comparison methods (Moyer et al., 2002). This will be used to quantify level of cigarette smoking.

The Vio Food Frequency Questionnaire (VioFFQ) was used to asses the subject's nutritional intake. Each subject completed the VioFFQ in order to obtain information regarding the types and quantities of foods consumed over the past 90 days. The subject answered questions based on food grouping, frequency of intake, and portion size. The software used the information obtained from this questionnaire to estimate each subject's daily nutrient intake over the past three months (Viocare Technologies, Inc). In this secondary analysis, tryptophan, pyridoxine, niacin, omega-3, and antioxidant intake were analyzed. The VioFFQ is based on information obtained from The Women's Health Initiative Food Frequency (Patterson et al, 1999).

The Center for Epidemiologic Studies Depression Scale (CES-D) was used to assess the subject's depressive symptoms. The (CES-D) is a 20 item scale used to assess depressive symptomatology in the general population. Subjects rated the severity of their depressive symptoms during the past week on a 4 point likert scale. This scale ranges from 0 to 3, in which 0 means the subject experiences the symptom rarely and 3 indicates the subject experiences the symptom most of the time. The subject is given a score ranging from 0 to 60, which is the total of the 20 items with a score greater than or equal to 16 indicative of depressive symptomatology. The CES-D Scale is found to have very high internal consistency and test- retest repeatability (Radolff, 1977).

The Perceived Stress Scale-10 (PSS) was used to measure the degree to which the subjects considered situations stressful over the previous month. This is a 10-question tool scored on a 5-point likert scale and is the only established index of general stress appraisal. It was developed by Cohen, Kamarck, & Mermelstein (1983) and is found to have high internal reliability (alpha=0.78). Higher scores on the PSS are associated with increased perceived stress and scores are also associated with depressive and physical symptomology (r=0.52-0.76) (Cohen et al, 1983).

Time to first cigarette of the day (TTF) will be used to measure nicotine dependence. A study by Heatherton, Kozlowski, Frecker, Rickert, and Robinson (1989) performed regression analyses using TTF and average daily consumption of cigarettes (CPD) to predict carbon monoxide, nicotine, and cotinine levels. In three independent samples, subjects who had their first cigarette earlier after awakening had higher levels of carbon monoxide, salivary and plasma cotinine, and nicotine than subjects who postponed having their first cigarette. Both TTF and CPD were concluded to be indicative of heaviness of smoking and are the most practical, non-invasive, and powerful predictors of nicotine dependence (Heatherton et al, 1989). For the purposes of this secondary analysis, subjects will be divided into two groups with smoking within 30 minutes of waking classifying subjects as nicotine dependent. This time frame is supported by the Fagerstrom Tolerance Questionnaire, which measures physiological tolerance to nicotine (Fagerstrom, 1978).

Human Subjects

IRB approval was obtained by the investigators of the primary study.

Data Analysis

Descriptive statistics, including mean, standard deviation, and ranges, were used for smoking level (cotinine ng/mL), depression (CES-D), perceived stress, time to first cigarette, and nutrient intake. For research questions 1 and 2, Pearson's correlation (Pearson's r) and Kendall's Tau B were used to describe the linear relationship between smoking, depression, and the selected nutrients. For research question 3, multiple regressions will be used to analyze data in order to determine which variable best predicts depression because it allows simultaneous testing and modeling of the multiple independent variables present in the study. T-Tests were used to examine differences in smoking patterns among races and BMI differences among genders. Mann-Whitney tests were used to determine the difference in tryptophan intake, omega-3 intake, depressive symptoms, and perceived stress between nicotine dependent smokers and non-nicotine dependent smokers.

IV. Results

Subject Characteristics:

The primary study examined 136 African American and Caucasian male or female bioconfirmed smokers 18-50 years of age who identified themselves as a person who has smoked 10-40 cigarettes/day for at least one year. There were 108 complete data sets used in this secondary analysis. In this sample, about half of the participants were female and 45% African American. The mean age was 28.3 years. Participants smoked an average of 15.7 cigarettes per day and had an average baseline cotinine level of 218.7 ng/mL. Time to first cigarette ranged from 1.0 minutes to 99.0 minutes, with an average of 21.8 minutes. The participants had an average body mass index (BMI) of 27.8 and average educational level was 12.9 years. The Vio Food Frequency Questionnaire indicated an average omega-3 intake of 2.32 grams/day and an average tryptophan intake of 1.29 grams/day among the subjects. CES-D scores ranged from 3 to 43, with an average

score of 14 points. Perceived Stress scores ranged from 12 to 33 with an average score of 21.2 (See Table 1). It was noted that omega-3 intake, tryptophan intake, pyridoxine intake, and niacin intake were positively skewed. Correcting for skewness by taking the square root or log of the variables did not significantly change the correlation and regression analyses. Results reported are with untransformed data.

Table 1: Demographic and Clinical Characteristics of Sample (N=108)

VARIABLE	*SAMPLE CHARACTERISTIC Mean (SD)*	RANGE
Age (yrs)	28.3 (8.3)	18-50
Gender (%Female) (% Male)	46.3 53.7	----
Race (% Caucasian) (% African American)	55.6 44.4	----
Average education (yrs)	12.9 (1.7)	9.0-17.0
Average Body Mass Index (BMI)	27.8 (7.7)	18.1-57.5
Cigarettes per day	15.7 (6.2)	4.0-40.0
Average baseline Cotinine (ng/mL)	218.68 (128.94)	19.0-695.0
Time to first cigarette (TTF in minutes)	22.78 (21.28)	1.0-99.0
Average CES-D score	14.02 (7.52)	3.0-43.0
Perceived stress score (PSS)	21.22 (3.95)	12.0-33.0
Average omega-3 intake (g/day)	2.32 (1.66)	0.25-9.90
Average tryptophan intake (g/day)	1.29 (.98)	0.17-5.43
Average pyridoxine intake (mg/day)	2.71 (2.15)	0.37-12.87
Average niacin intake (mg/day)	32.31 (25.52)	5.62-138.59

Depressive Symptoms and Tryptophan

Pearson's correlation was used to determine if a relationship exists between depressive symptoms and tryptophan intake among the 108 participants (Aim 1). Because pyridoxine is needed for the production of tryptophan and increased niacin intake allows for more production of serotonin, both pyridoxine and niacin intake were taken into consideration when running correlation statistics. Controlling for pyridoxine and niacin intake, Pearson's correlation between tryptophan and CES-D scores is -.109 (See Table 2). The relationship is inverse, indicating increased depressive symptoms with low intake of tryptophan; however, statistics indicate no significant relationship (p=.265) between CES-D scores and intake of tryptophan among the 108 subjects.

Table 2: Correlation between CES-D score and tryptophan intake, controlling for pyridoxine and niacin intake (N=108)

Control Variables			TRYPTOPHAN	CESDTOTAL
Pyridoxine & Niacin	TRYPTOPHAN	Correlation	1.000	-.109
		Significance (2-tailed)	.	.265
		df	0	104
	CES-D TOTAL	Correlation	-.109	1.000
		Significance (2-tailed)	.265	.
		df	104	0

Depressive Symptoms and Omega-3 PUFAs

It was hypothesized that subjects would display increased depressive symptoms with decreased intake of Omega-3 PUFAs (Aim 2). Pearson's correlation was used to assess this relationship. Pearson's correlation for the CES-D scores and omega-3 PUFA intake is -.015 (See Table 3). Again, the inverse relationship indicates increased CES-D scores with lower intake of omega-3 PUFAs; however, the relationship is not statistically significant (p=.881).

Table 3: Correlation between CES-D score and Omega-3 PUFA intake (N=108)

		OMEGA-3	CES- D TOTAL
OMEGA-3	Pearson Correlation	1.000	-.015
	Sig. (2-tailed)		.881
	N	108.000	108
CES-D TOTAL	Pearson Correlation	-.015	1.000
	Sig. (2-tailed)	.881	
	N	108	108.000

Smoking and Depressive Symptoms

Pearson's correlation was again used to examine the relationship between depressive symptoms and level of smoking, which was indicated by cotinine level. Literature supports the hypothesis that an increased amount of depressive symptoms would be associated with higher baseline cotinine values. Among the sample of 108 subjects, the relationship between baseline cotinine values and CES-D scores has a Pearson's correlation of -.015 (See Table 4). With a significance of .895, there is no significant relationship between these two variables among the 108 subjects.

Table 4: Correlation between baseline cotinine and CES-D score (N=108)

		CES-D TOTAL	BASELINE COTININE
CES-D TOTAL	Pearson Correlation	1.000	-.015
	Sig. (2-tailed)		.889
	N	108.000	108
BASELINE COTININE	Pearson Correlation	-.015	1.000
	Sig. (2-tailed)	.898	
	N	108	108.000

Smoking and Nutrition

Pearson's correlation was used to examine the relationship between the subjects' nutrition profiles and smoking status as indicated by baseline cotinine level. Results indicate that none of the selected nutritional indicators (omega-3, tryptophan, niacin, vitamin B6, fat, and calories consumed) were significantly correlated with smoking status (see Table 5).

Table 5: Correlation between smoking status and selected nutrients (N-108)

Correlations

		Baseline Cotinine	Omega-3	Tryptophan	Niacin	Vitamin B6
Baseline Cotinine	Pearson Correlation	1.000	.114	.042	.003	-.023
	Sig. (2-tailed)		.242	.665	.976	.816
	N	108.000	108	108	108	108
Omega-3	Pearson Correlation	.114	1.000	.852**	.784**	.759**
	Sig. (2-tailed)	.242		.000	.000	.000
	N	108	108.000	108	108	108
Tryptophan	Pearson Correlation	.042	.852**	1.000	.937**	.882**
	Sig. (2-tailed)	.665	.000		.000	.000
	N	108	108	108.000	108	108
Niacin	Pearson Correlation	.003	.784**	.937**	1.000	.957**
	Sig. (2-tailed)	.976	.000	.000		.000
	N	108	108	108	108.000	108
Vitamin B6	Pearson Correlation	-.023	.759**	.882**	.957**	1.000
	Sig. (2-tailed)	.816	.000	.000	.000	
	N	108	108	108	108	108.000

**. Correlation is significant at the 0.01 level (2-tailed).

Predictors of Depression

Multiple regression was used to analyze the multiple hypothesized, independent predictor variables of depressive symptoms (Aim 3). Again, pyridoxine and niacin intake were taken into consideration when describing tryptophan intake. Of the three hypothesized predictors of depression, omega-3 intake, tryptophan intake, and baseline cotinine level, there was no significant data supporting that these variables could adequately predict increased depressive symptoms in the sample. Levels of significance are displayed in Table 6.

Table 6: Potential predictors of depressive symptoms (N=108)

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95% Confidence Interval for B	
		B	Std. Error	Beta			Lower Bound	Upper Bound
1	(Constant)	13.973	1.762		7.928	.000	10.477	17.468
	PYRIDOXINE	-.256	1.212	-.073	-.211	.833	-2.659	2.147
	NIACIN	.118	.137	.400	.861	.391	-.154	.389
	OMEGA-3	-.195	.872	-.043	-.224	.824	-1.924	1.534
	TRYPTOPHAN	-2.066	2.601	-.270	-.794	.429	-7.225	3.093
	BASELINE COTININE	2.910E-5	.006	.000	.005	.996	-.012	.012

a. Dependent Variable: CESDTOTAL

Stress and Depression

According to Cohen (1983), higher scores on the PSS are associated with increased perceived stress and scores are also associated with depressive symptomatology. Pearson's correlations were used to assess the relationship between the CES-D score and the PSS score among the 108 participants. Results indicate that CES-D score and PSS score are highly correlated ($p<.001$). Results are displayed in Table 7.

Table 7: Correlation between CES-D score and PSS score (N=108)

Correlations

		CESDTOTAL	PSS
CES-D TOTAL	Pearson Correlation	1	.519**
	Sig. (2-tailed)		.000
	N	108	107
PSS	Pearson Correlation	.519**	1
	Sig. (2-tailed)	.000	
	N	107	107

Stress, Nicotine Dependence, Depression and Nutrition

Another aim of this study was to determine if there is a significant difference in tryptophan intake, omega-3 intake, depressive symptoms, and perceived stress between nicotine dependent smokers and non-nicotine dependent smokers (Aim 4). Nicotine dependence was defined by smoking the first cigarette of they day within 30 minutes of arising. Because 87 of the 108 participants smoked their first cigarette of the day within 30 minutes of arising, Mann-Whitney tests were used to determine if there was a significant difference in nutrient intake, stress and depression between the group that smoked within 30 minutes of arising and the group that did not. Results indicate that the nicotine dependent smokers consumed lower levels of tryptophan (p=.018) and higher PSS scores (p=.058). Mann-Whitney tests are displayed in Table 8.

Table 8: Differences in tryptophan intake, omega-3 intake, CES-D scores, and PSS scores between nicotine dependent smokers (TTF<30 min) and non-nicotine dependent smokers (TTF>30 min) (N=107 for PSS and 108 for tryptophan, omega-3, and CES-D total)

Ranks

	TTF	N	Mean Rank	Sum of Ranks
PSS	1.00 (Before 30 min)	86	51.20	4403.00
	2.00 (After 30 min)	21	65.48	1375.00
	Total	107		
TRYPTOPHAN	1.00 (Before 30 min)	87	50.99	4436.00
	2.00 (After 30 min)	21	69.05	1450.00
	Total	108		
OMEGA-3	1.00 (Before 30 min)	87	53.37	4643.00
	2.00 (After 30 min)	21	59.19	1243.00
	Total	108		
CES-D TOTAL	1.00 (Before 30 min)	87	52.78	4591.50
	2.00 (After 30 min)	21	61.64	1294.50
	Total	108		
Baseline Cotinine	1.00 (Before 30 min)	87	58.67	5104.50
	2.00 (After 30 min)	21	37.21	781.50
	Total	108		

Test Statistics[a]

	PSS	TRYPTOPHAN	OMEGA3	CESDTOTAL	CotBase
Mann-Whitney U	662.000	608.000	815.000	763.500	550.500
Wilcoxon W	4403.000	4436.000	4643.000	4591.500	781.500
Z	-1.898	-2.371	-.765	-1.166	-2.818
Asymp. Sig. (2-tailed)	.058	.018	.444	.244	.005

a. Grouping Variable: TTF

Sociodemographics, smoking level, BMI, depressive symptoms, and nutrition

A secondary aim of this study was to explore the relationship between sociodemographics, smoking level, body mass index, depressive symptoms, and nutritional intake.

Sociodemographics and Depressive Symptoms

Pearsons correlation was used to examine the relationship between sociodemographics and depression rating on the CES-D questionnaire. The variables selected included age at which the subject began smoking, years of education, gender, race, and age at the time of the study. Of these variables, level of education was the only variable that was significantly correlated with CES-D score (p=.076). The negative, low correlation implies an inverse relationship between education level and CES-D scores, indicating that subjects with lower levels of education exhibited higher scores on the CES-D, implying increased depressive symptoms.

In addition, age was positively correlated with race (p=.001), indicating that the older participants were generally African American. Years of education was also significantly correlated with race (p=.002). This inverse correlation implies that the Caucasian participants generally had higher levels of education (See Table 9).

Table 9: Correlation between sociodemographics variables and CES-D score (N=108)

Correlations

		GENDER	CES-D TOTAL	AGE	RACE	EDUCATION
GENDER	Pearson Correlation	1	.140	-.095	-.046	.017
	Sig. (2-tailed)		.149	.327	.639	.862
	N	108	108	108	108	108
CES-D TOTAL	Pearson Correlation	.140	1	-.039	.148	-.171
	Sig. (2-tailed)	.149		.688	.127	.076
	N	108	108	108	108	108
AGE	Pearson Correlation	-.095	-.039	1	.365**	.047
	Sig. (2-tailed)	.327	.688		.000	.632
	N	108	108	108	108	108
RACE	Pearson Correlation	-.046	.148	.365**	1	-.290**
	Sig. (2-tailed)	.639	.127	.000		.002
	N	108	108	108	108	108

EDUCATION	Pearson Correlation	.017	-.171	.047	-.290**	1
	Sig. (2-tailed)	.862	.076	.632	.002	
	N	108	108	108	108	108

**. Correlation is significant at the 0.01 level (2-tailed).

BMI and Gender

Another area of interest was the relationship between BMI and gender. Using independent sample t-tests, there is a significant difference in body mass index between genders (p=.026), implying that the female participants had higher body mass indexes than male participants (See Table 10).

Table 10: Differences in BMI among genders (N=108)

Group Statistics

	Gender	N	Mean	Std. Deviation	Std. Error Mean
BMI	1 (Male)	58	26.04823	5.911377	.776202
	2 (Female)	50	29.39400	9.331304	1.319646

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
									95% Confidence Interval of the Difference	
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	Lower	Upper
BMI	Equal variances assumed	10.455	.002	-2.256	106	.026	-3.345775	1.482833	-6.285635	-.405914
	Equal variances not assumed			-2.185	80.488	.032	-3.345775	1.530998	-6.392274	-.299276

Age and Smoking Status

Another area of interest involved the relationship between subject's ages, baseline cotinine, age at which the subjects began smoking, and number of cigarettes smoked per day. Pearson's correlation was used to examine the relationship between these four variables. Analyses indicate that baseline cotinine was positively correlated with age (p=.002), implying that cotinine values generally increased with age. Because cotinine baseline is indicative of level of smoking, the significant, moderate positive relationship between cotinine and number of cigarettes per day was expected (p=.001). In addition, age was also significantly correlated with number of cigarettes per day. This reiterates the notion that, in general, number of cigarettes smoked per day increased with age (p=.001). The age at which the subject began smoking was inversely correlated with number of cigarettes smoked per day (p=.025), indicating that the sooner the subjects began smoking, the more cigarettes they currently smoke per day (see Table 8).

Table 8: Correlation between subject ages, baseline cotinine, age at which the subjects began smoking, and number of cigarettes smoked per day (N=108)

Correlations

		COTININE BASELINE	AGE	AGE BEGAN	CIGS PER DAY
COTININE BASELINE	Pearson Correlation	1.000	.293**	-.079	.403**
	Sig. (2-tailed)		.002	.418	.000
	N	108.000	108	108	108
AGE	Pearson Correlation	.293**	1.000	.112	.196*
	Sig. (2-tailed)	.002		.250	.042
	N	108	108.000	108	108
AGE BEGAN	Pearson Correlation	-.079	.112	1.000	-.216*
	Sig. (2-tailed)	.418	.250		.025
	N	108	108	108.000	108
SMOKE PER DAY	Pearson Correlation	.403**	.196*	-.216*	1.000
	Sig. (2-tailed)	.000	.042	.025	
	N	108	108	108	108.000

Correlations

		COTININE BASELINE	AGE	AGE BEGAN	CIGS PER DAY
COTININE BASELINE	Pearson Correlation	1.000	.293**	-.079	.403**
	Sig. (2-tailed)		.002	.418	.000
	N	108.000	108	108	108
AGE	Pearson Correlation	.293**	1.000	.112	.196*
	Sig. (2-tailed)	.002		.250	.042
	N	108	108.000	108	108
AGE BEGAN	Pearson Correlation	-.079	.112	1.000	-.216*
	Sig. (2-tailed)	.418	.250		.025
	N	108	108	108.000	108
SMOKE PER DAY	Pearson Correlation	.403**	.196*	-.216*	1.000
	Sig. (2-tailed)	.000	.042	.025	
	N	108	108	108	108.000

**. Correlation is significant at the 0.01 level (2-tailed).

*. Correlation is significant at the 0.05 level (2-tailed).

Race and Smoking Status

Kendall's tau b was used to examine the correlation between race and cigarettes per day. In general, race was inversely correlated with cigarettes per day (p=.045) and positively correlated with baseline cotinine (p=.004). This indicates that African American subjects smoked fewer cigarettes per day and had higher baseline cotinine levels compared to their Caucasian counterparts (See Table 9).

Table 9: Correlation between level of smoking and race (N=108)

Correlations

			BASELINE COTININE	CIGS PER DAY	RACE
Kendall's tau b	BASELINE COTININE	Correlation Coefficient	1.000	.282**	.227**
		Sig. (2-tailed)	.	.000	.004
		N	108	108	108
	CIGS PER DAY	Correlation Coefficient	.282**	1.000	-.171*
		Sig. (2-tailed)	.000	.	.045
		N	108	108	108
	RACE	Correlation Coefficient	.227**	-.171*	1.000
		Sig. (2-tailed)	.004	.045	.
		N	108	108	108

**. Correlation is significant at the 0.01 level (2-tailed).

*. Correlation is significant at the 0.05 level (2-tailed).

Sociodemographic Variables and Nutrition (omega-3, niacin, pyridoxine, and tryptophan)

Kendal's tau b was used to examine the relationship between sociodemographics variables (gender, age, race, and level of education) and nutritional intake. Gender was significantly inversely correlated with niacin, vitamin B6, and tryptophan, indicating that males consumed higher levels of these nutrients than females. In addition, age was significantly positively correlated with omega-3 intake, indicating that omega-3 consumption increased with age (See Table 10).

Table 10: Correlation between sociodemographics and selected nutrients

Correlations

			gender	AGE	race	edu	NIACIN	OMEGA3	TRYPTOPHAN	VITB6
Kendall's tau_b	gender	Correlation Coefficient	1.000	-.096	-.046	-.010	-.300**	-.141	-.267**	-.299**
		Sig. (2-tailed)	.	.239	.637	.907	.000	.076	.001	.000
		N	108	108	108	108	108	108	108	108
	AGE	Correlation Coefficient	-.096	1.000	.326**	.072	.038	.152*	.044	.021
		Sig. (2-tailed)	.239	.	.000	.317	.565	.022	.513	.757
		N	108	108	108	108	108	108	108	108
	race	Correlation Coefficient	-.046	.326**	1.000	-.262**	-.012	.054	-.054	-.087
		Sig. (2-tailed)	.637	.000	.	.002	.877	.496	.496	.274
		N	108	108	108	108	108	108	108	108
	edu	Correlation Coefficient	-.010	.072	-.262**	1.000	-.102	-.086	-.044	-.015
		Sig. (2-tailed)	.907	.317	.002	.	.147	.225	.530	.827
		N	108	108	108	108	108	108	108	108
	NIACIN	Correlation Coefficient	-.300**	.038	-.012	-.102	1.000	.556**	.751**	.792**
		Sig. (2-tailed)	.000	.565	.877	.147	.	.000	.000	.000
		N	108	108	108	108	108	108	108	108
	OMEGA3	Correlation Coefficient	-.141	.152*	.054	-.086	.556**	1.000	.591**	.501**
		Sig. (2-tailed)	.076	.022	.496	.225	.000	.	.000	.000
		N	108	108	108	108	108	108	108	108
	TRYPTOPHAN	Correlation Coefficient	-.267**	.044	-.054	-.044	.751**	.591**	1.000	.697**
		Sig. (2-tailed)	.001	.513	.496	.530	.000	.000	.	.000
		N	108	108	108	108	108	108	108	108
	VITB6	Correlation Coefficient	-.299**	.021	-.087	-.015	.792**	.501**	.697**	1.000
		Sig. (2-tailed)	.000	.757	.274	.827	.000	.000	.000	.
		N	108	108	108	108	108	108	108	108

**. Correlation is significant at the 0.01 level (2-tailed).

*. Correlation is significant at the 0.05 level (2-tailed).

IV. Discussion and Conclusions

In summary, Pearson's correlations among the 108 African American and Caucasian male and female cigarette smokers indicated a non-significant inverse relationship between smoking and tryptophan and smoking and omega-3 intake, and there was no significant relationship between smoking, depressive symptoms, and the selected nutrients. Neither nutrient intake nor smoking could predict depressive symptoms. Less education was correlated with higher CES-D scores (p=.046) and Caucasians had higher levels of education (p=.001). Females had higher body mass indexes (p=.034). Older subjects had higher baseline cotinine levels (p=.002) and smoked more cigarettes per day (p=.042). African Americans had higher baseline cotinine levels (p=.004), and smoked more cigarettes per day (p=.045).

Tryptophan and Depressive Symptoms

Previous studies have indicated an inverse correlation between tryptophan levels and depression, with smoking exacerbating this relationship (Booij et al, 2005; Spring et al, 2007; Pergadia et al, 2004). The results from this study are similar to these previous studies in identifying this potential inverse relationship, implying that increased depressive symptoms are associated with low intake of tryptophan; however, statistics indicate no significant relationship (p=.265) among the 108 subjects. Despite these insignificant findings, the potential relationship still remains biologically plausible due to the fact that serotonin production is dependent upon tryptophan and niacin intake. All previous studies have examined this inverse relationship between tryptophan and depressive symptoms through the use of acute tryptophan depletion (Booij et al, 2005; Spring et al, 2007; Pergadia et al, 2004), which was not an option in this current study. In addition, all previous studies have controlled tryptophan intake, while this study examined the subjects' self-reported intake over the past 90 days via the Vio Food Frequency Questionnaire.

Smoking and Depressive Symptoms

This study also shows no relationship between smoking, defined as baseline cotinine level and cigarettes per day, and depression, despite the results of numerous studies that indicate the high correlation among these variables (Son et al, 1997; Brown et al, 2000; Kenny et al, 2006). Although the CES-D score was the selected tool for many of the previous studies that examined the relationship between smoking and depressive symptoms, many of the reviewed studies used a nicotine dependence questionnaire in addition to baseline cotinine levels to define smoking level. When nicotine dependence was taken into consideration in these studies, the dependent smokers displayed significantly higher CES-D scores (Son et al, 1997; Brown et al, 2000; Kenny et al, 2006).

In addition, the subjects' CES-D scores range from 3 to 43, with a score of 16 indicating depressive symptoms. Many studies have established the high correlation between smoking and depression; however, in order to examine this relationship with other variables, it is important to have depressed subjects. Of the 108 participants, 69 subjects (63.9% of the sample), had reported CES-D scores less than 16. This emphasizes the notion that the 108 participants may not adequately

represent the true incidence of depressive symptoms in the smoking population. In addition, previous studies have compared large samples of both smokers and nonsmokers when establishing this relationship. For example, a total of 671 women with no history of major depressive disorders were studied by researchers at the University of Melbourne. Of the women who were smokers, 15% went on to develop major depressive disorder. However, among non-smokers, only 6.5% developed major depressive disorder during a decade of follow-up (University of Melbourne, 2008).

Omega-3 PUFAs and Depressive Symptoms

The relationship observed between omega-3 PUFAs and depressive symptoms among the 108 subjects was not statistically significant (p=.881). Previous studies have indicated an inverse relationship between serum levels of PUFAs and depressive symptoms. Despite the statistical insignificance observed in this study, the correlation between these two variables remains biologically plausible due to the fact that fatty acids inhibit pro-inflammatory cytokine synthesis, which is associated with depression, by suppressing inflammation and immune reactivity markers (Mamalakis et al, 2006). While the Vio Food Frequency Questionnaire is a validated approach for estimating types and quantities of foods consumed over the past 90 days (Patterson et al, 1999), no previous studies were found that used self report in examining the relationship between omega-3 PUFAs and depressive symptoms. Instead, previous studies have used adipose tissue aspiration and serum levels, which provides for a more exact estimation of omega-3 intake (Mamalakis et al, 2006; Keicolt-Glaser et al, 2007).

Smoking and Selected Nutrients

Results from this study also imply no relationship between omega-3, tryptophan, niacin, vitamin B6 and smoking level. Previous studies demonstrated an inverse relationship between smoking status and omega-3 fatty acid intake because cigarette smoke negatively affects omega-3 fatty acid synthesis. In addition cigarette smoke decreases PUFA synthesis because of the free radicals that result from smoking (Pawlosky et al, 2007; Marangoni et al, 2004). In this study, all

nutrient variables were measured via the Food Frequency Questionnaire; therefore serum nutrient levels were not available. It was hypothesized that the 108 participants would exhibit decreased intake of the selected nutrients because the most comprehensive study that has examined the relationship between smoking and nutrition indicated that smokers have less than adequate dietary intakes of a variety of nutrients when compared to nonsmokers (Dyer et al, 2003). While the reported levels of nutrients could not be compared to nonsmokers, the Vio Food Frequency Questionnaires of all 108 subjects indicated adequate tryptophan, omega-3, vitamin B6, and niacin intake among all the 108 subjects as defined by the Food and Nutrition Information Center of the United States Department of Agriculture National Agricultural Library (United States Department of Agriculture, 2002).

Predictors of Depression

Although previous research indicated associations between smoking and nutrition, and smoking and depressive symptoms, no research was found that integrated all three of these variables. Of the three hypothesized predictors of depression, omega-3 intake, tryptophan intake, and baseline cotinine level, there was no significant data supporting that these variables could adequately predict increased depressive symptoms in the sample of the 108 participants. Because the correlations were low among the hypothesized variables, the regression data was likely to be insignificant. Again, serum nutrient levels would have provided better indication of nutrient intake.

Stress, Nicotine Dependence, Depression and Nutrition

Results indicated that nicotine dependent smokers, the smokers who smoked their first cigarette within 30 minutes of awakening, had higher perceived stress ($p=.058$) and lower tryptophan intake ($p=.018$). From a biological perspective, serotonin is a neurotransmitter that is synthesized from tryptophan, an essential amino acid that must be ingested. The Booij et al (2005) study supports this notion that low levels of tryptophan ultimately deplete serotonin and diminished serotonin levels are highly associated with depression. However, the Booij et al study looked at this

relationship by purposefully depleting tryptophan in the participants. The Spring, et al (2007) study also depleted tryptophan levels in their participants, but they added the smoking variable. Researchers reported that smoking magnified the depressive symptoms in subjects with low tryptophan levels.

This study is the first known study to look at the differences between tryptophan intake, perceived stress, and depressive symptoms among nicotine dependent and non-nicotine dependent smokers. While significant differences were found, there are some limitations to this data. Tryptophan intake was assessed via self-report and the PSS and CES-D are also subjective tools. In addition, a majority of the subjects smoked a cigarette within 30 minutes of awakening (n=87), so non-parametric statistics were used to assess these differences.

Sociodemographics, smoking level, BMI, depressive symptoms, and nutrition

A secondary aim of this study was to explore the relationship between sociodemographics, smoking level, body mass index, depressive symptoms, and nutrition. Among the sociodemographic data, level of education was the only variable that was significantly correlated with CES-D score ($r = -.171$, $p = .076$). The negative, low correlation implies an inverse relationship between education level and CES-D scores, indicating that subjects with lower levels of education exhibited higher scores on the CES-D. Years of education was also significantly correlated with race ($r = -.290$, $p = .002$). The inverse correlation implies that the Caucasian participants generally had higher levels of education. African Americans typically had lower education levels; however, there was no correlation of depression and race. Despite this, the association of education level and depressive symptoms could be attributed to a variety of factors, including the economic stressors that may result from having a lower level of education. These findings are congruent with a recent study that showed that smoking and low education levels were predictors of depression (Rubio, et al., 2008). Despite this, further research regarding the relationship between level of education and depression needs to be completed in order to further investigate factors associated with less education that contribute to depressive symptoms in adults, specifically adults who smoke.

Another area of significance was the relationship between BMI and gender. The low positive correlation between body mass index and gender implies that the female participants had higher body mass indexes than male participants. Despite this, gender was significantly inversely correlated with niacin, vitamin B6, and tryptophan, indicating that males consumed more of these nutrients than females. Further research is needed to assess these relationships; however, the higher BMI in females can be indicative of severe health problems, such as cardiovascular disease and diabetes mellitus. The American Heart Association recommends that all adults increase their consumption of omega-3 fatty acids because of the increased risk of developing coronary artery disease that is associated with omega-3 deficiency (He et al, 2004). These preliminary results indicate that further research is needed to investigate this risk among the female smoking population.

In addition, analyses indicate that baseline cotinine was positively correlated with age ($r=.293$, $p=.002$). In addition, age was significantly correlated with number of cigarettes per day ($r=.196$, $p=.042$). The moderate correlation between number of cigarettes per day and baseline cotinine ($r=.403$, $p=.001$) validates the use of baseline cotinine in measuring level of smoking. To further validate the relationship between age and level of smoking, the age at which the subject began smoking was inversely correlated with number of cigarettes smoked per day, indicating that the sooner the subjects began smoking, the more cigarettes they currently smoke per day. This is particularly significant because serum cotinine level has been found to be a predictor of risk of lung cancer among smokers (Boffetta et al, 2006). This means that higher baseline cotinine levels put people at a higher risk of developing lung cancers. Because cigarette smoking is highly addictive, the older participants are more vulnerable when it comes to health status.

Another area of significance was the relationship between race and smoking level. Race was inversely correlated with cigarettes per day and positively correlated with baseline cotinine. This implies that African American subjects smoked fewer cigarettes per day, but yet had higher baseline cotinine levels. This data indicates that the African American participants in the study had more cotinine, a nicotine metabolite, in their system despite the fact that they smoke fewer cigarettes per

day. These findings are consistent with existing criterion in which African American subjects smoke fewer cigarettes per day and have higher baseline cotinine levels.

Conclusions

In summary, the findings of this study do not completely support the proposed hypothesis regarding the relationship between smoking level, depressive symptoms and the selected nutrients. It was hypothesized that smokers with increased depressive symptoms would have lower than the recommended levels of tryptophan and omega-3 fatty acids in their diets and that nicotine dependent smokers would have greater depressive symptoms, higher perceived stress, and lower than the recommended dietary intake of tryptophan and omega-3 fatty acids. Smokers with increased depressive symptoms did not have significantly lower than the recommended levels of tryptophan and omega-3 fatty acids in their diets; however, nicotine dependent smokers had higher perceived stress, greater depressive symptoms, and lower tryptophan intake.

Despite this, more research is needed in order to further assess this relationship. The small sample size and secondary nature of this study limit the applicability of the findings. The self-report method of identifying food intake via the Food Frequency Questionnaire was a major limitation in this study as well as the fact that the population was not compared to nonsmokers. Serum nutrient levels would allow researchers to accurately identify systemic nutrient levels.

Previous studies indicated that smokers have less healthful diets than nonsmokers, are at a higher risk for developing depressive symptoms with low dietary intake of omega-3 PUFAs and tryptophan, and are at a higher risk of depressive symptoms with increased nicotine dependence and cotinine level. This was the first study to examine the relationship among all three variables and warrants further examination. Nursing interventions in smokers need to focus on smoking cessation as well as improving diets in the effort to reduce cancer and cardiovascular disease risks. Screening for depression among smokers is also an area of importance because of the high incidence of Major Depressive Disorder among the smoking population. This screening becomes especially important in

smokers who are attempting smoking cessation because smoking cessation may be less successful in people who exhibit depressive symptoms (Perez, et al., 2008).

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