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## **The Effect of Fiber and Omega-6:Omega-3 Fatty Acid Ratio on Asthma and Asthma-related Symptoms in an Adult U.S. Population**

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Epidemiology: Infectious Disease

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### **Abstract**

Asthma is one of the most common diseases in the world affecting more than 10% of adults in many westernized countries and 339 million globally. Studies have shown a positive association between increased dietary fiber and improvement in asthma-related inflammation. Omega-3 fatty acids have also been shown to help mitigate inflammation in general. No identified studies have looked at the synergistic effect of omega-3 and omega-6 fatty acids alone and as a ratio with fiber on asthma, lung function, and respiratory morbidities. A positive association could mean that dietary interventions of fiber and fatty acids could significantly improve asthma and asthma-related symptoms. Dietary interventions are especially attractive because they are inexpensive and can be easy to incorporate into daily routines. Using data from NHANES from year cycles 2007-2012, analysis through logistic regression was performed to find the association of the synergistic effect of fiber and the omega-6:omega-3 fatty acid ratio on asthma and asthma-related symptoms in an adult U.S. population. Intake of fiber was found to be associated with cough ( $p$ -value=0.02), but

that there was no significant association of omega-3 or omega-6 fatty acids with asthma or asthma-related symptoms either alone or as a ratio. The role of omega-6 fatty acids in inflammation, whether pro- or anti-inflammatory, is still not fully understood, but even less is understood about the complex relationship between omega-3 fatty acids and omega-6 fatty acids. The understanding of the interaction of fiber and omega fatty acids on asthma and asthma-related symptoms requires further study.

## **Background**

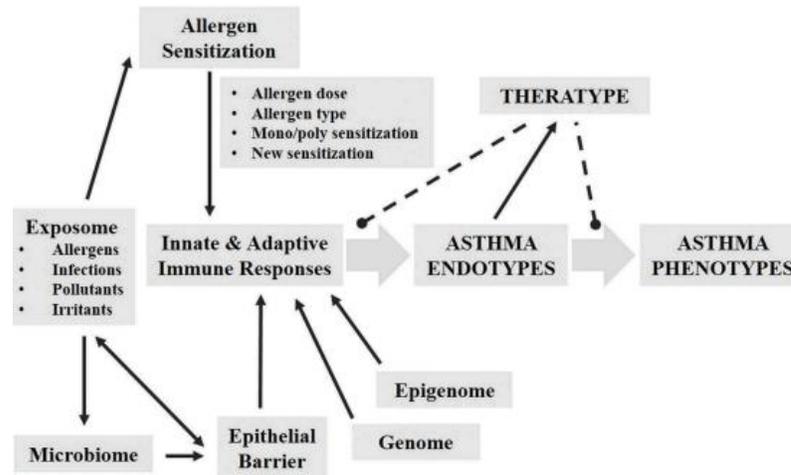
Asthma is a chronic inflammatory disease of the airways. Symptoms include wheezing, breathlessness, chest tightness, sputum production, coughing (that varies over time and intensity), and a variable expiratory airflow limitation (via a spirometry test).<sup>1,2</sup> Asthma is one of the most common diseases in the world affecting more than 10% of adults in many westernized countries and 339 million globally.<sup>3,4,5</sup> In the U.S., asthma prevalence is 8.3%.<sup>4</sup>

The total economic burden of asthma in the U.S., in 2008-2013, was \$81.9 billion and includes costs for both children and adults comprising medical costs, absenteeism, and mortality costs.<sup>6</sup> In fact, 1.8 additional work days and 2.3 additional school days were missed due to asthma during 2008-2013.<sup>6</sup> The annual incremental medical cost of asthma per-person is projected to be \$4,699.<sup>7</sup> In addition to economic losses, there are negative effects to quality of life related to asthma, demonstrated through significant quality of life year (QALY) losses. It is estimated that both adolescents and adults will lose 15.46 million QALY in the next 20 years, which is a 4.2% increase of loss, due to

uncontrolled asthma.<sup>7</sup> Because of these substantial losses, prevention, intervention, and treatment of asthma is critical.

There are many phenotypes of asthma, rendering it a complex syndrome rather than a single disease.<sup>8</sup> Some of these phenotypes include inflammatory (allergic versus non), immunological (atopic versus non), age of onset (child versus adult), related to genetic factors, related to comorbidities, related to triggering factors (like exercise), and clinical (episodic versus persistent).<sup>5</sup> Allergic (inflammatory) and atopic (immunological) are the most common phenotypes, both often called type 2 asthma. In fact, half of adult asthma patients have allergic asthma.<sup>2</sup> Although prevention of asthma is important, it is difficult for many reasons, to include a wide range of risk factors, many of which are unmodifiable. In addition to this, asthma is often under- and over-diagnosed.<sup>2,5</sup>

Although there is no cure for asthma, knowing the phenotype of asthma can help guide the decision-making process for the most appropriate and effective intervention. Interventions such as pharmacological, immunotherapy, allergen avoidance, dietary, or a combination, can help lessen the severity and mitigate symptoms (see Figure 1).<sup>5</sup> For example, the presence of immunoglobulin E (IgE), which has a fundamental role in the pathophysiology of asthma, is one of the most valuable indicators for allergic asthma. Once identified, understanding the allergens to which the patient is allergic to will help with effective avoidance, thus mitigating asthma symptoms.<sup>2</sup> In another example, a mother being overweight during pregnancy has been associated with childhood asthma.<sup>9</sup> A dietary intervention of fish oil for pregnant women from 20 weeks of gestation until delivery showed a reduction in asthma outcomes at one year for



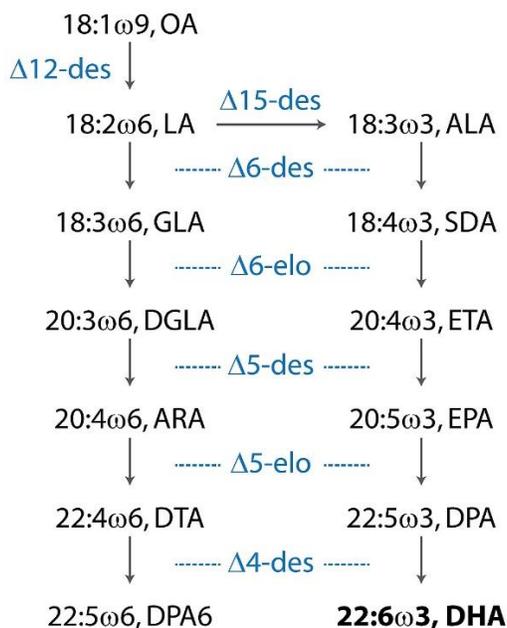
**Figure 1.** Factors influencing asthma endotypes and phenotypes. The interactions between immune responses and exposome (a measure of all exposures in an individual from birth until current time), microbiome, epigenome and genomes, allergen sensitization, and epithelial barrier all affect the asthma endotypes and phenotypes.

**Source:** Ozdemir C, Kucuksezer UC, Akdis M, Akdis CA. The concepts of asthma endotypes and phenotypes to guide current and novel treatment strategies. *Expert Rev Respir Med.* 2018;12(9):733-43.

children.<sup>5</sup> Additionally, diet changes in those who already have asthma have a strong association with improvement of symptoms.<sup>4,5</sup> Dietary interventions are especially attractive because they are inexpensive and easy to incorporate into daily routines.<sup>5</sup>

A review by Calder exploring marine omega-3 fatty acids (FA) in inflammation, to include asthma, found that the intake of marine omega-3 fatty acids, i.e., eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), can help control the effects of inflammation. When inflammation occurs, elevated levels of leukocytes concentrate at the site (here, we focus on the lungs) and release lipid-derived mediators, peptide mediators, reactive oxygen species, amino acid derivatives, and enzymes. This influx of cells at the site of inflammation, and subsequent mediators, generate the cardinal signs of inflammation: redness, swelling, heat, pain, and loss of function.<sup>10</sup> Due to its prevalence in phospholipids, arachidonic acid (ARA), an omega-6

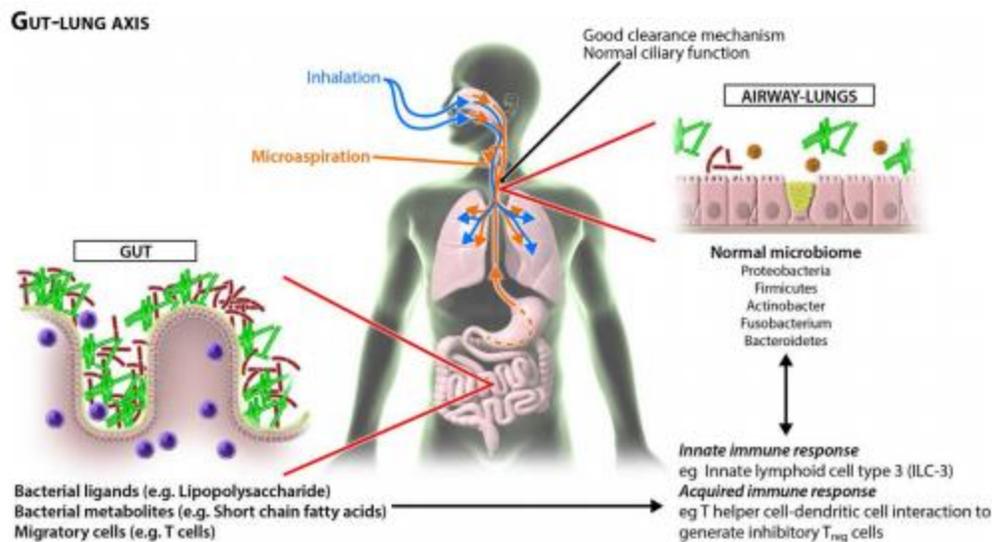
FA, is often the substrate for eicosanoids.<sup>10</sup> These eicosanoids, derived from ARA, are involved in determining the allergic predisposition to asthma, as well mediators involved in the inflammatory response.<sup>10</sup> An increase in omega-6 FA (specifically ARA) and a reduction in omega-3 FA, has been correlated to increased incidence of asthma.<sup>11</sup> Thus, inhibiting ARA conversion from linoleic acid via competition of omega-3 FA could potentially improve inflammation related to asthma (see Figure 2). Additionally, both EPA and DHA have been shown to improve lung function by reducing eosinophil infiltration and giving rise to resolvins, which are anti-inflammatory and inflammation resolving.<sup>10</sup>



**Figure 2.** The parallel pathways of omega-3 and omega-6 and shared enzymes. ‘Des’ denotes desaturase and ‘elo’ denotes elongase. OA, oleic acid; LA, linoleic acid; GLA,  $\gamma$ -linolenic acid; DGLA, dihomo- $\gamma$ -linolenic acid; ARA, arachidonic acid; DTA, docosatetraenoic acid; DPA6,  $\omega$ 6 docosapentaenoic acid; ALA,  $\alpha$ -linolenic acid; SDA, stearidonic acid; ETA, eicosatetraenoic acid; EPA, eicosapentaenoic acid; DPA,  $\omega$ 3 docosapentaenoic acid.

**Source:** Petrie JR, Shrestha P, Belide S, Kennedy Y, Lester G, Liu Q, Divi UK, Mulder RJ, Mansour MP, Nichols PD, Singh SP. Metabolic engineering *Camelina sativa* with fish oil-like levels of DHA. *PLoS One*. 2014; 9(1):e85061.

Recent studies have shown that dietary fiber exhibits both anti-inflammatory and antioxidant properties.<sup>12</sup> Specifically, high-fiber has a protective role in asthma, respiratory morbidity (wheeze, cough, and phlegm), and systemic inflammation. It has also been shown that dietary fiber changes the composition of the gut microbiome, particularly altering the ratio of *Firmicutes* and *Bacteroidetes*, thus increasing the concentration of short-chain fatty acids (SCFA). These SCFA, or the by-products of fiber fermentation in the gastrointestinal tract by the gut microbiota, can be found in the systemic circulation and provide a protective effect on the lungs via regulation of neutrophils, attenuation of pulmonary inflammation, and epithelial-based protection against bacterial infections (see Figure 3).<sup>12</sup>



**Figure 3.** The gut-lung axis. The gut microbiota can influence the lung microbiota by modulating lung immunity through bacterial ligands, bacterial metabolites, and migratory cells that then circulate systemically to the lungs. The lung microbiota is responsible for maintaining a healthy immune response. It is also possible that the immune response could regulate the lung microbiota, as indicated by the *double-ended arrows*.

**Source:** Chung KF. Airway microbial dysbiosis in asthmatic patients: A target for prevention and treatment? *J Allergy Clin Immunol.* 2017;139(4):1071-81.

The recommended dietary requirement of fiber is 20-30 g/d, with positive results seen in asthma by Saeed et al. at levels above 21.2 g/d.<sup>4</sup> An effective dose for EPA and DHA is recommended to be 2 g/d, but the required dose of the omega-3 and omega-6 FA ratio to be effective in asthma is still unclear.<sup>10</sup> Additionally, the interaction between fiber and these FA, and their role in asthma, need further exploration. In fact, no identified studies have looked at the synergistic effect of omega-3 and omega-6 FA with fiber on asthma, lung function, and respiratory morbidities. We believe that although fiber and omega-3 FA are protective separately, their combined effect could be greater together and significantly improve asthma and asthma-related symptoms.

## **Aims**

No identified studies have looked at the synergistic effect of both omega-6 and omega-3 FA with fiber on asthma, lung function, and respiratory morbidities. This study explored the effect of fiber and the omega-6:omega-3 FA ratio on asthma and asthma-related symptoms in an adult U.S. population. We accomplished this objective through the following aim.

Aim: Determine the moderating role of fiber intake: on the relationship of omega-6 and omega-3 FA with asthma, and asthma-related respiratory symptoms. *We hypothesize that a lower omega-6:omega-3 FA ratio and increased fiber intake are associated with a decreased odds of asthma and asthma-related symptoms compared to those with a higher omega-6:omega-3 FA ratio and lower fiber intake.*

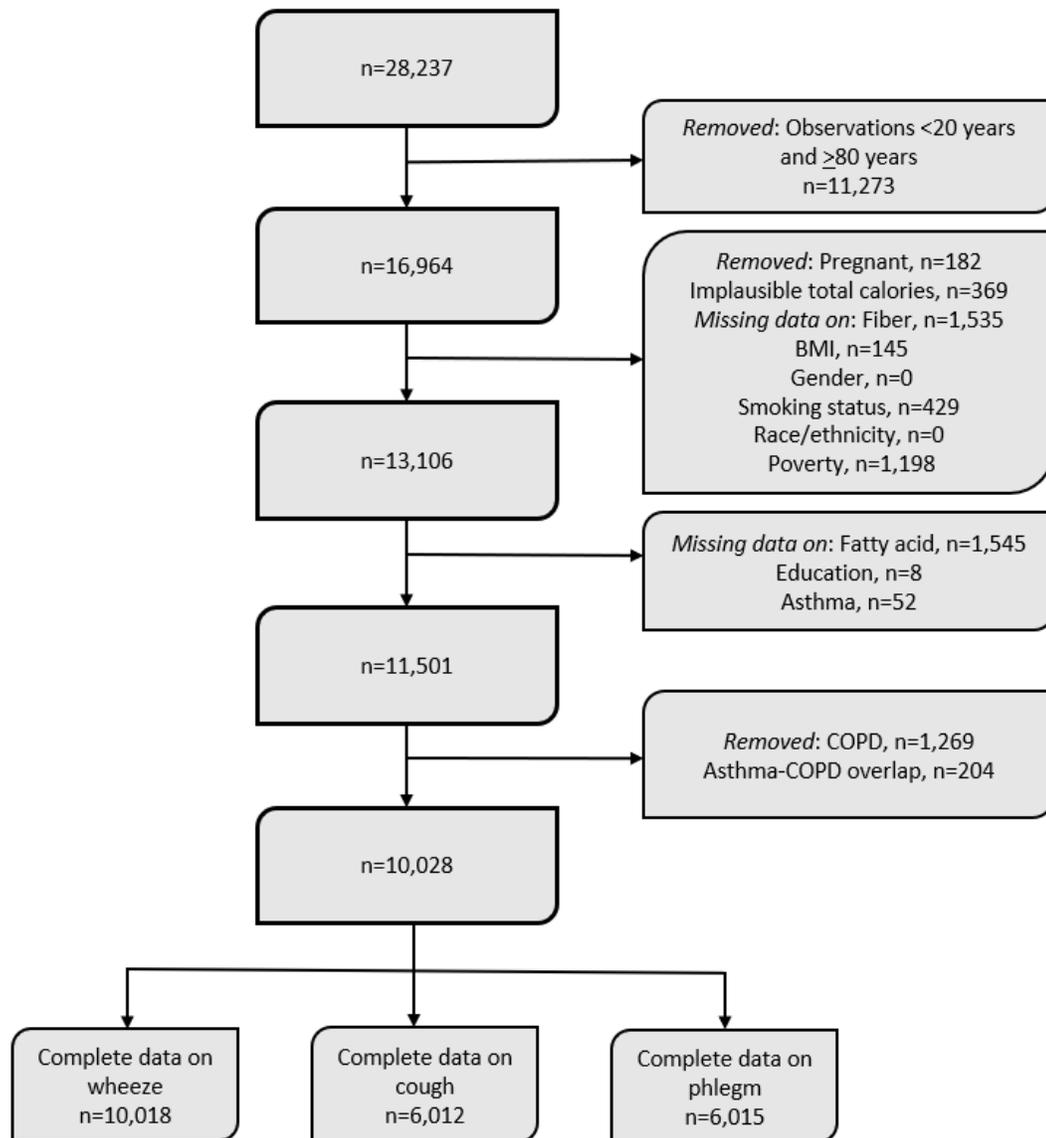
## **Methods**

### *Study population*

The National Center for Health Statistics of the Center for Disease Control and Prevention (CDC) conducts the National Health and Nutrition Examination Survey (NHANES).<sup>13,14</sup> This is an ongoing cross-sectional survey conducted nationwide designed to assess the health and nutritional status of the U.S. population that is not institutionalized. Study participants are selected by using stratified multistage probability sampling, including oversampling of minorities, low-income, and elderly subjects to increase statistical power for data analysis and to represent the U.S. population as a whole.

### *Subjects*

We analyzed data from adults 20-79 years old with complete information on asthma, age, gender, race/ethnicity, education status, body mass index (BMI), smoking status, income to poverty ratio, fiber intake, and omega FA intake (Figure 4). Age (19 years or younger and 80 years or older), pregnancy status (pregnant), and chronic obstructive pulmonary disease (COPD)/asthma-COPD Overlap Syndrome (ACOS) (the presence of) were used as exclusion criteria. Implausible caloric intake was deemed to be <600 or >6,000 kcal/d for women and <800 or >8,000 kcal/d for men.<sup>4,15</sup> Those missing data on asthma, age, gender, race/ethnicity, education status, BMI, smoking status, income to poverty ratio, fiber intake, or omega FA intake were also excluded.



**Figure 4.** Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) diagram. COPD is defined as 40 years of age or older, having smoked at least 100 cigarettes in lifetime, having a FEV1/FVC ratio of <0.7, and no asthma. Those with asthma-COPD overlap syndrome (ACOS) had the characteristics of someone with COPD, as previously stated, but also had answered yes to currently having asthma. Those with missing data, as shown above, were removed along with those out of the age range (<20 years and ≥80 years), pregnant, having implausible total calories, or having COPD or ACOS.

Both asthma and COPD are common chronic lung diseases seen worldwide.

These diseases, however, differ in pathogenesis, disease progression, prognosis, and

treatment options.<sup>16</sup> Asthma affects the small and large airways and is often accompanied by allergies. It typically develops in childhood, but may develop in adulthood for a small group of people. Generalized airway obstruction causes wheezing, coughing, chest tightness, and breathlessness. This airway obstruction is caused by smooth-muscle spasm and is reversible after the attack. Bronchial hyperresponsiveness, a key feature of asthma (but not sufficient for a firm diagnosis), is caused by inhaled stimuli.<sup>17</sup> COPD affects mainly the small airways and is primarily caused by smoking. Due to its link to smoking, COPD is often seen in adults over 40 years old and has poor life expectancy. Airway obstruction causes wheezing, chronic cough, breathlessness, and phlegm. This obstruction is caused by smooth-muscle contraction, airway mucus, or tissue breakdown, leading to a loss of lung elastic recoil that inhibits reversibility.<sup>17</sup> Although heterogeneous diseases, similar symptoms shared between the two create overlap, causing distinction between the two diseases to be difficult.<sup>16,17</sup> These shared symptoms include shortness of breath, cough, and sputum.<sup>16</sup> Additionally, some patients with asthma may experience irreversible airway damage and those with COPD reversible damage, contradicting what is typically seen.<sup>16,17</sup> Thus, relevant variables to consider for diagnosis of either asthma or COPD are: age of onset, pattern and time course of symptoms, personal/family history, variable/persistent airflow limitation, lung function between symptoms, and severe hyperinflation.<sup>17</sup> If three or more features of either disease (asthma or COPD) is present, then it is commonly diagnosed as that disease.<sup>17</sup> If there are similar number of features in a patient for both COPD and asthma, it is suggestive of ACOS.

### *Asthma and Respiratory Symptoms*

Current asthma was defined as “yes” if participants answered positively to two questions: “Has a doctor or other health professional ever told you that you have asthma?” and “Do you still have asthma?” Participants were controls if they answered “no” to either question. Current wheeze was defined as a “yes” to the question: “In the past 12 months, have you had wheezing or whistling in your chest?” Chronic cough was defined as answering “yes” to the question: “Do you usually cough on most days for three consecutive months or more during the year?” Chronic phlegm was defined as answering “yes” to the question: “Do you bring up phlegm on most days for three consecutive months or more during the year?”

### *Fiber and Omega Fatty Acid Intake*

Intake of both fiber and omega FA was obtained via the NHANES 24-hour diet recall interview, using the average of day one and day two for both diet variables. The first dietary interview was collected in-person in the mobile examination center (MEC) using the Automated Multiple Pass Method from the U.S. Department of Agriculture.<sup>18</sup> At the end of the MEC dietary interview, the interviewers schedule the participants for a phone follow-up interview. A follow-up dietary interview is conducted by telephone 3-10 days later. Detailed descriptions of dietary interview and data processing procedures can be found under the dietary interview components on the NHANES website.<sup>15</sup> In this study, the goal is to rank individuals with respect to fiber and FA intake, rather than to make accurate estimates of absolute intake. Diet variable

quartiles were based on the population. Fiber was categorized into quartiles (Q1 <11.15 g/d; Q2 11.15-<15.7 g/d; Q3 15.7-<21.95 g/d; Q4  $\geq$ 21.95 g/d). Omega FA was defined as intake of omega-3 and omega-6 or as a ratio (omega-6:omega-3). Omega-3 in this study was defined as intake of octadecatrienoic acid (18:3 ALA), octadecatetraenoic acid (18:4 SDA), EPA (20:5), docosapentanoic acid (22:5 DPA), and DHA (22:6). Omega-6 was defined as intake of octadecadienoic acid (18:2 LA) and ARA (20:4). NHANES did not define what isomers ALA, DPA, and ARA are, thus determining whether omega-3 or omega-6. Thus, based on previous research, they were categorized in this study as stated above.<sup>19-27</sup> Fatty acid intake was categorized into quartiles (Quartiles for total omega-3 are: Q1 <1.07 g/d; Q2 1.07-<1.54 g/d; Q3 1.54-<2.22 g/d; Q4  $\geq$ 2.22 g/d. Quartiles for omega-6 are: Q1 <10.07 g/d; Q2 10.07-<14.53 g/d; Q3 14.53-<20.35 g/d; Q4  $\geq$ 20.35 g/d. Quartiles for omega-6:omega-3 ratio are: Q1 <7.77; Q2 7.77-<9.16; Q3 9.16-<10.95; Q4  $\geq$ 10.95).

### *Additional Covariates*

Covariates included in our adjusted models are age, gender, race/ethnicity, income to poverty ratio, BMI, smoking status, and education. These covariates were defined from previous literature.<sup>4</sup> The correlation between diet and physical activity was examined in the study population and there was not a significant correlation found. Additionally, use of physical activity as a covariate would have brought the study population to 2,469 participants. Age was categorized as: 20-39 years, 40-59 years, and 60+ years. Race/ethnicity was categorized as: non-Hispanic white, non-Hispanic black, Mexican American and other Hispanic, and Other (to include multiracial). Socio-

economic status was determined by the poverty to income ratio, which was categorized as: <1(poverty), 1-2.9, and 3-5+. BMI status was categorized as: underweight (<18.5 kg/m<sup>2</sup>), normal (18.5-24.9 kg/m<sup>2</sup>), overweight (25-29.9 kg/m<sup>2</sup>), and obese (30-40 kg/m<sup>2</sup>). Smoking status was defined as: never, former (smoked>100 cigarettes in a lifetime, but does not currently smoke), and current (smoked>100 cigarettes in a lifetime and currently smokes). Lastly, education was grouped into three categories: less than high school graduate, high school graduate/GED, and college (to include some college or AA and college graduate or above).

### *Statistical Analysis*

Statistical analysis was done in SAS 9.4. Descriptive statistics (frequency, mean, median, and standard deviation) were performed on demographic variables of age, gender, race/ethnicity, income to poverty ratio, BMI, smoking status, and education. Bivariate analysis was used to determine the association between fiber, omega FA, and demographics variables. Chi-square test was used to assess associations between outcome variables ([yes/no]: asthma, wheeze, cough, phlegm) and exposure variables (fiber and omega FA quartiles, and FA ratio). PROC SURVEYMEANS and PROC SURVEYFREQ were used for descriptive analysis of the population and associations for continuous and categorical variables, respectively. Finally, multivariable logistic regression was applied to examine the relationship of fiber, omega FA ratio, and FA quartiles with the odds of developing asthma, wheeze, cough, and phlegm using PROC SURVEYLOGISTIC after controlling for age, gender, race/ethnicity, income to poverty ratio, BMI, total energy intake (continuous), and smoking status. Fiber was tested as an

interaction term with omega-3 FA, omega-6 FA, and the FA ratio.  $P_{\text{trend}}$  was determined by using PROC SURVEYLOGISTIC and setting the diet variable (either fiber, omega-3 FA, or omega-6 FA) as continuous, rather than categorical. For all tests, significance is set at two-sided  $p\text{-value} < 0.05$ .

## Results

After exclusion criteria were applied to the NHANES population ( $n=28,237$ ) our study population included 10,028 individuals (see Figure 4). The mean age was  $44.2 \pm 0.4$  (SD) years with 53.3% female and 46.7% male (see Table 1). The mean income to poverty ratio was  $3.3 \pm 0.04$  (SD) and the majority of subjects were non-Hispanic white (69%), had at least some college (63%), and never smokers (63%). Nearly 70% of all subjects were either overweight or obese with a mean BMI of  $28.97 \text{ kg/m}^2 \pm 0.12$  (SD). When compared to controls (no asthma), those with asthma tended to be female (65.3% versus 52.4%), in poverty (22.5% versus 13.9%), and obese (48.0% versus 35.8%).

The association of intake of fiber and omega FA with asthma was determined by bivariate analysis. Intake of fiber was associated with lower odds of asthma ( $p\text{-value}=0.0016$ ). This association, however, was lost in multivariable analysis. Although not statistically significant, those with asthma were more likely to have lower intake of fiber than those with no asthma, with over 54% of asthmatics consuming fiber in the lower two quartiles, while 48% of those with no asthma had similar intake levels (mean for those with asthma compared to no asthma,  $16.22 \pm 0.52$  g/d and  $17.47 \pm 0.22$  g/d,

**Table 1.** Characteristics of NHANES (2007-2012) study population stratified by asthma status

Demographic	Total n(%)	Asthma n(%)	No Asthma n(%)	p-value
	10028	719(7.02)	9309(92.98)	
<b>Age, years</b>				0.0551
20-39	4009(41.67)	314(44.81)	3695(41.44)	
40-59	3425(38.48)	216(32.97)	3209(38.89)	
60+	294(19.85)	189(22.22)	2405(19.67)	
<b>Gender</b>				<.0001
Male	4648(46.66)	237(34.70)	4411(47.57)	
Female	5380(53.34)	482(65.30)	4898(52.43)	
<b>Race/Ethnicity</b>				0.0045
Non-Hispanic white	4377(68.99)	353(71.24)	4024(68.82)	
Non-Hispanic black	2166(11.10)	187(13.90)	1979(10.89)	
Mexican American and other Hispanic	2654(13.76)	133(9.78)	2521(14.06)	
Other, including multiracial	831(6.15)	46(5.08)	785(6.23)	
<b>Education</b>				0.5177
Less than high school grad	2350(15.34)	155(13.81)	2195(15.45)	
High school grad/GED equivalent	2188(21.26)	165(22.04)	2023(21.21)	
College (some/AA degree to graduate)	5490(63.40)	399(64.15)	5091(63.34)	
<b>Ratio of income to poverty*</b>				<.0001
<1	2183(14.47)	228(22.50)	1955(13.87)	
1-2.9	4011(34.16)	264(31.90)	3747(34.33)	
3-5+	3834(51.37)	227(45.60)	3607(51.80)	
<b>BMI, kg/m<sup>2</sup></b>				<.0001
Underweight, <18.5	129(1.36)	2(0.16)	127(1.45)	
Normal, 18.5-24.9	2643(28.49)	141(22.43)	2502(28.95)	
Overweight, 25-29.9	3303(33.45)	193(29.38)	3110(33.75)	
Obese, 30-40+	3953(36.70)	383(48.03)	3570(35.85)	
<b>Smoking Status</b>				0.1316
Never smoker	6265(62.76)	424(61.15)	5841(62.88)	
Former Smoker	1927(20.04)	129(18.16)	1798(20.18)	
Current Smoker	1836(17.20)	166(20.69)	1670(16.94)	
<b>Wheeze</b>				<.0001
yes	1217(11.48)	428(58.82)	789(7.90)	
no	8801(88.52)	291(41.18)	8510(92.10)	
<b>Chronic Cough</b>				<.0001
yes	512(8.41)	113(27.23)	399(7.07)	
no	5500(91.59)	291(72.77)	5209(92.93)	
<b>Chronic Phlegm</b>				<.0001
yes	423(5.97)	86(17.90)	337(5.12)	
no	5592(94.03)	318(82.10)	5274(94.88)	

\*Under one means in poverty; BMI= body mass index. PROC SURVEYFREQ used for analysis. Wheeze (total=10,018), Cough (total=6,012), Phlegm (total=6,015).

**Table 2.** Association of intake of fiber and omega fatty acids with asthma

Diet variable	Total	Asthma	No Asthma	p-value
	10028	719	9309	
<b>Fiber</b>				0.0016
Q1 (<11.15 g/d)	2657(23.82)	246(31.16)	2411(23.27)	
Q2 (11.15-<15.7 g/d)	2546(25.25)	176(23.77)	2370(25.36)	
Q3 (15.7-<21.95 g/d)	2465(25.64)	169(23.51)	2296(25.80)	
Q4 ( $\geq$ 21.95 g/d)	2360(25.29)	128(21.56)	2232(25.57)	
Mean $\pm$ stnd err	17.57 $\pm$ 0.203	16.22 $\pm$ 0.52	17.48 $\pm$ 0.22	
<b>Total omega-3</b>				0.1702
Q1 (<1.07 g/d)	2713(24.23)	221(26.90)	2492(24.03)	
Q2 (1.07-<1.54 g/d)	2541(25.27)	187(27.48)	2354(25.10)	
Q3 (1.54-<2.22 g/d)	2412(24.83)	168(23.54)	2244(24.93)	
Q4 ( $\geq$ 2.22 g/d)	2362(25.67)	143(22.08)	2219(25.94)	
Mean $\pm$ stnd err	1.78 $\pm$ 0.014	1.68 $\pm$ 0.06	1.78 $\pm$ 0.01	
<b>Total omega-6</b>				0.4054
Q1 (<10.07 g/d)	2822(24.27)	215(25.74)	2607(24.15)	
Q2 (10.07-<14.53 g/d)	2492(25.00)	183(26.90)	2308(24.85)	
Q3 (14.53-<20.35 g/d)	2426(25.26)	167(24.94)	2259(25.29)	
Q4 ( $\geq$ 20.35 g/d)	2289(25.47)	154(22.42)	2135(25.71)	
Mean $\pm$ stnd err	16.25 $\pm$ 0.12	15.23 $\pm$ 0.41	16.27 $\pm$ 0.13	
<b>Omega ratio n6:n3</b>				0.6362
Q1 (<7.77)	2672(24.81)	176(24.01)	2496(24.88)	
Q2 (7.77-<9.16)	2443(24.63)	182(25.64)	2261(24.55)	
Q3 (9.16-<10.95)	2427(24.86)	160(22.56)	2267(25.03)	
Q4 ( $\geq$ 10.95)	2486(25.70)	201(27.79)	2285(25.54)	
Mean $\pm$ stnd err	9.79 $\pm$ 0.05	9.68 $\pm$ 0.14	9.76 $\pm$ 0.06	

PROC SURVEYFREQ and PROC SURVEYMEANS used for analysis.

respectively) (see Table 2). This same trend was seen with omega-3 FA intake as well. Those with asthma tended to have less omega-3 FA intake than those with no asthma with over 54% of asthmatics consuming omega-3 FA in the lower two quartiles, while

49% of those with no asthma had similar intake values (mean for those with asthma compared to no asthma,  $1.68 \pm 0.06$  g/d and  $1.78 \pm 0.01$  g/d, respectively). Unlike fiber and omega-3 FA, lower levels of omega-6 FA are desirable since higher levels are thought to contribute to inflammatory diseases such as asthma.<sup>11,28</sup> When comparing the two higher quartiles of omega-6 FA (Q3 14.53-<20.35 g/d; Q4  $\geq 20.35$  g/d) between those with asthma and those with no asthma, surprisingly, those with asthma consumed less omega-6 FA than those with no asthma, 47.4% versus 51%, respectively (mean of  $15.23 \pm 0.41$  g/d versus  $16.27 \pm 0.13$  g/d). When comparing the two higher quartiles of the FA ratios (Q3 9.16-<10.95; Q4  $\geq 10.95$ ) between those with asthma and those with no asthma, there were nearly identical amounts of subjects in these higher quartiles, 50.3% versus 50.6%, with a mean of  $9.68 \pm 0.14$  versus  $9.76 \pm 0.06$ , respectively. Although notable, the association of asthma with omega FA and FA ratio was not statistically significant.

Multivariable analysis did not show any significant association between diet variables and respiratory outcomes except for fiber intake and cough (see Table 3). Lower fiber intake (Q1) showed increased odds of cough (OR=1.8, 95% CI 1.21-2.77, p-value=0.005) when compared to higher fiber intake (Q4). A linear trend was also observed for fiber intake and cough ( $p_{\text{trend}}=0.008$ ). Although not significant, a trend between fiber intake and phlegm was also seen, with higher odds of experiencing phlegm with lower fiber intake (Q1 OR=1.5, CI 1.00-2.39; Q2 OR=1.3, CI 0.82-2.15; Q3 OR=1.1, CI 0.68-1.77).

**Table 3.** Relationship of fiber and omega fatty acid quartiles with asthma and respiratory symptoms

Diet variable	Asthma	Phlegm	Cough	Wheeze
	10028	6015	6012	10018
<b>Fiber</b>				
Q1 (<11.15 g/d)	1.2(0.86-1.57)	1.5(1.00-2.39)	1.8(1.21-2.77)	1.2(0.92-1.51)
Q2 (11.15-<15.7 g/d)	0.9(0.70-1.15)	1.3(0.82-2.15)	1.6(1.17-2.23)	1.2(0.96-1.55)
Q3 (15.7-<21.95 g/d)	0.9(0.70-1.27)	1.1(0.68-1.77)	1.3(1.06-1.74)	1.1(0.86-1.44)
Q4 (≥21.95 g/d)	1	1	1	1
Overall p-value	0.15	0.1	0.02	0.39
Q1 vs. Q4 p-value	0.32	0.05	0.005	0.18
P trend	0.92	0.17	0.008	0.11
<b>Total omega-3</b>				
Q1 (<1.07 g/d)	1.0(0.72-1.36)	1.0(0.61-1.75)	0.8(0.58-1.14)	1.0(0.73-1.38)
Q2 (1.07-<1.54 g/d)	1.1(0.81-1.46)	1.0(0.63-1.57)	0.9(0.64-1.32)	1.1(0.88-1.38)
Q3 (1.54-<2.22 g/d)	1.0(0.77-1.32)	0.9(0.66-1.50)	0.7(0.55-1.10)	0.9(0.82-1.21)
Q4 (≥2.22 g/d)	1	1	1	1
Overall p-value	0.87	0.99	0.53	0.8
Q1 vs. Q4 p-value	0.95	0.9	0.23	0.99
P trend	0.64	0.86	0.68	0.91
<b>Total omega-6</b>				
Q1 (<10.07 g/d)	1	1	1	1
Q2 (10.07-<14.53 g/d)	1.2(0.87-1.53)	1.1(0.74-1.69)	0.9(0.68-1.4)	1.1(0.85-1.53)
Q3 (14.53-<20.35 g/d)	1.1(0.80-1.57)	1.0(0.63-1.73)	1.1(0.75-1.65)	1.0(0.82-1.33)
Q4 (≥20.35 g/d)	1.1(0.79-1.56)	1.2(0.61-2.17)	1.3(0.79-2.11)	1.1(0.77-1.44)
Overall p-value	0.81	0.93	0.64	0.83
Q1 vs. Q4 p-value	0.55	0.67	0.31	0.74
P trend	0.98	0.99	0.57	0.4
<b>Omega ratio n6:n3</b>				
Q1 (<7.77)	1	1	1	1
Q2 (7.77-<9.16)	1.0(0.76-1.40)	1.4(0.92-2.07)	1.4(1.00-2.07)	1.0(0.78-1.31)
Q3 (9.16-<10.95)	0.9(0.66-1.18)	1.3(0.81-2.04)	1.0(0.69-1.49)	0.8(0.65-1.08)
Q4 (≥10.95)	1.1(0.81-1.43)	0.9(0.57-1.34)	1.1(0.74-1.50)	1.0(0.84-1.25)
Overall p-value	0.68	0.22	0.06	0.35
Q1 vs. Q4 p-value	0.61	0.52	0.78	0.82
P trend	0.92	0.41	0.79	0.36

PROC SURVEYLOGISTIC used for analysis. Adjusted for age (20-39 years, 40-59 years, and 60+ years), gender, race/ethnicity (non-Hispanic white, non-Hispanic black, Mexican American and other Hispanic, and Other), income to poverty ratio (<1, 1-2.9, and 3-5+), BMI (underweight, <18.5 kg/m<sup>2</sup>; normal, 18.5-24.9 kg/m<sup>2</sup>; overweight, 25-29.9 kg/m<sup>2</sup>; and obese, 30-40 kg/m<sup>2</sup>), total energy intake (continuous), and smoking status (never, former, and current).

## Discussion

We explored the association of fiber, omega-3, and omega-6 FA intake with asthma and asthma-related symptoms in a population-based study of 10,028 individuals from NHANES (2007-2012). In the bivariate analysis, we found that higher intake of fiber was associated with decreased odds of asthma; however, only the relationship between fiber and cough was significant in multivariable analysis. There was no significant association of omega-3 or omega-6 FA with asthma or asthma-related symptoms.

Alwarith et al. noted that diets comprised of high fat and low fiber are associated with airway inflammation and decreased lung function in asthmatics.<sup>11</sup> In support of this, previous research has shown that higher levels of fiber intake are correlated with lower odds of experiencing an exacerbation of asthma and respiratory symptoms.<sup>3,4</sup> While we did not observe a strong association between fiber and cough, phlegm or wheeze in bivariate analysis, an inverse association was seen between fiber and cough in the multivariable analysis. These results are slightly different than our previous study by Saeed et al. where we found that lower levels of fiber increased the odds of asthma and asthma-related symptoms (wheeze, cough, phlegm, and chronic bronchitis).<sup>4</sup> While the results seem contradictory, the NHANES population used in the two studies differed. Both studies used the definition of asthma; however, Saeed et al. included individuals with asthma as well as those with COPD in their study population. The current study removed individuals with COPD as defined as having a  $FEV_1/FVC < 0.7$  and were current or former smokers. Asthma is epidemiologically difficult to study because of its heterogeneous nature and lack of standard definition or gold standard diagnostic test.

To compound this challenge, clinical characteristics of asthma and COPD sometimes overlap (thus, diagnosed as ACOS) or are experienced concomitantly. Those experiencing COPD, either alone or as ACOS, often have behavioral differences and notably different demographic characteristics when compared to those experiencing asthma alone. Specifically, those with COPD tend to be smokers (current or former), over the age of 40, and male.<sup>16,17,21</sup> Eliminating those with COPD from the current study population ensured we did not skew our analysis or spuriously attribute certain characteristics to our defined groups of asthma and no asthma. It is likely that Saeed et al. saw significant associations between fiber, asthma, and respiratory symptoms because their population contained subjects with COPD and/or ACOS.

The National Institutes for Health (NIH) state that optimum levels of fiber intake are 38 g/d for males and 25 g/d for females.<sup>29</sup> The NIH also noted that in 2010 the U.S. average intake for fiber was only 14 g/d.<sup>29</sup> In our study, no subjects met the fiber recommendations set by the NIH, but nearly 32% had a fiber intake of >20g/d. These fiber intake values are similar in the Saeed study where only 33% of their subjects met their cited recommendation of 20 g/d.<sup>4</sup>

Omega-3 FA have been shown to have anti-inflammatory properties and thought to reduce the risk of cardiovascular disease, improve infant health and neurodevelopment, prevent cancer, and positively affect Alzheimer's disease, dementia, cognitive function, age-related macular degeneration, dry eye disease, and rheumatoid arthritis.<sup>19,21,30</sup> We did not, however, observe an association of omega-3 FA with asthma or asthma-related symptoms. These results are similar to those found by Kumar et al. where they found in a Dutch population-based study that omega-3 FA did not protect

against asthma.<sup>31</sup> However, another population-based study in Northern Europe found that a minimum weekly intake of fish high in omega-3 FA was protective for asthma.<sup>31</sup> The inconsistencies found in these studies, as well as our own, may be due to inaccuracies inherent to dietary recall methods used in population-based studies.<sup>31</sup>

While there is no set recommendation for daily intake for any omega-3 FA, the NIH has suggested adequate intake (AI) (or the intake level that is assumed to be nutritionally adequate) for ALA as 1.6 g/d for adult males and 1.1 g/d for adult females.<sup>32</sup> In our study, 24% of males and 33% of females met the recommended AI for ALA. Because only ALA is considered essential, there are no AIs stated for any other omega-3, to include EPA or DHA.<sup>32</sup> Calder et al., however, found that resolution can be seen in inflammation when EPA and DHA intake levels are at 2 g/d and positive effects of omega-3 FA are seen on asthma when intake levels of EPA and DHA were above 2 g/d.<sup>10</sup> Intake of  $\geq 2$  g/d of EPA and DHA was only seen in 0.2% of our population. It should be noted that while EPA and DHA can be converted from ALA, the conversion rate is incredibly poor in humans.<sup>10</sup>

The role of omega-6 FA in inflammation and respiratory outcomes, like asthma, is unclear. While many believe these FA to be pro-inflammatory, studies have shown that increasing intake of omega-6 FA did not actually affect many inflammatory biomarkers.<sup>33</sup> In fact, some epidemiological studies have suggested that omega-6 FA can even reduce inflammation.<sup>33</sup> Additionally, there is evidence that omega-6 FA inhibit the anti-inflammatory effects of omega-3 FA, creating a complex interaction between the two FA.<sup>33</sup> Western diets are laden with omega-6 FA; thus, a deficiency of omega-6 FA is rarely a problem.<sup>11</sup> Like ALA, LA is the omega-6 precursor to ARA, and is

considered an essential FA.<sup>32</sup> The NIH recommends that daily intake for adults are 17 g/d for males and 12 g/d for females.<sup>34</sup> Many agree, however, that it is more important to increase omega-3 intake than it is to decrease omega-6 levels.<sup>32</sup> In our study, 23% of males and 29% of females consumed levels of LA at or above those recommended by the NIH. However, when stratified by asthmatic status, those with no asthma consumed less omega-6 FA compared to those with asthma (mean 15.23 g/d versus 16.27 g/d, respectively). A study by McKeever et al. showed similar results where they looked at omega FA intake in 13,820 adults and its association with asthma symptoms. The researchers found that higher levels of omega-6 FA, not omega-3 FA, were associated with a higher FEV<sub>1</sub>.<sup>35</sup>

In this study, dietary omega-3 FA was determined based on intake of ALA, SDA, EPA, DPA, and DHA. Omega-6 FA was defined as ARA and LA intake. Some studies, such as that from Lemoine et al., used the most basic formula with only EPA and DHA representing omega-3 FA and ARA representing omega-6 FA.<sup>21</sup> While there is a simplicity to this profile, it does not account for the omega-3 FA that people consume in the greatest amount: ALA.<sup>32</sup> Additionally, many people consume a variety of foods and, thus, not just one type of omega-3 or omega-6 FA. The definitions we used in this study are similar to that of Dong et al. and Chen et al., and we suggest this allowed for a more complete assessment of omega FA intake.<sup>20,23</sup> To support this approach, we found that the omega-6:omega-3 ratio was much lower when using the most basic FA profile (ARA: EPA+DHA, 5.48) compared to the profile ultimately used in our study (ALA+SDA+EPA+DPA+DHA:ARA+LA, 9.79). Although the lower ratio is nutritionally desired, we suggest it is a false depiction of omega-3 and omega-6 intake since it does

not account for ALA. Many western diets have an omega-6:omega-3 ratio of 15:1 to 17:1.<sup>19</sup> Some may be as low as 10:1, representing a healthier western diet, but still above a ratio considered to be beneficial.<sup>36</sup> Although it is believed that at one point our diets contained the “ancestral” ratio of 1:1, beneficial ratios have shown to range from 4:1 to 7:1.<sup>19,28</sup> These ratios, specifically 4:1, have shown to protect against diabetes and emotional and cognitive behavior impairment.<sup>19,20,27</sup> Thus, a smaller a ratio, or a ratio closest to 4:1, is the desired metric. In our study, over 75% of the individuals had a ratio of 7:1 or more.

Because our main aim was to test fiber’s potential synergistic role on omega FA, we statistically tested the interaction of fiber with omega FA on respiratory effects. Despite our hypothesis of synergism, we found that the fiber and omega FA interactions to be nonsignificant. A study by Navarro et al. looked at the effect of the interaction of fiber and omega-3 FA (EPA and DHA) on the risk of colorectal cancer in women.<sup>37</sup> They found the interaction to be nonsignificant, with only high levels of fiber intake having protective effects.<sup>37</sup> Interestingly, Navarro et al. noted that at low fiber levels an interaction with the omega-3 FA was detected, and weakened as fiber levels increased. This interaction, however, was no longer significant after correcting for multiple testing.<sup>37</sup> We hypothesize that it is likely we did not find significant associations when comparing diet variables to asthma and respiratory symptoms due to the complex interactions of the omega FA with one another and fiber.

Our study has the distinct advantage in that it addresses the interaction of fiber and omega FA intake in relation to asthma. Despite our results not supporting our hypothesis of seeing a lower prevalence of asthma and respiratory outcomes with

higher intake of fiber and omega-3 FA, and lower intake of omega-6 FA, our results and the complexity of the diet variables with one another support the need for more research. Our study has a few limitations. First, the race/ethnicity profile of our study population for those with asthma does not match the prevalence seen in the U.S. Those with asthma in our population were predominantly non-Hispanic white, while asthma in the U.S. is most prevalent in non-Hispanic blacks with a prevalence of 10.6% (versus 7.6% for non-Hispanic whites).<sup>11,29</sup> Other demographics predominantly found in our study population such as gender (female), socioeconomic status (in poverty), and BMI (overweight/obese) were similar to those of adult asthmatics in the U.S.<sup>6,8,11,29,38</sup>

Second, our dietary intakes were measured by dietary recall. NHANES relies on 24-hour dietary recall interviews in-person for day one and via phone interviews for day two. Because these interviews are not substantiated with laboratory diagnostics, there is inherent bias and potential for inaccuracy.<sup>31</sup> Third, despite much care taken in determining dietary intake of FA, there may be misclassification due to lack of isomer specification in NHANES, as noted by Chen et al.<sup>23</sup> It should also be noted that once a FA is consumed, FA metabolism may differ among subjects, thus altering outcomes specific to that subject.<sup>21</sup> It is suggested that a randomized control trial be designed and conducted to compare actual intake versus minimal intake versus levels high intake (defined as enough to confer a positive effect on respiratory outcomes). Additionally, it may be beneficial to measure clinical biomarkers and their correlation with dietary intake.

## **Conclusion**

In the general U.S. population we found that intake of fiber was associated with asthma in bivariate analysis and with cough in multivariable analysis; however, no significant association was observed between omega-3 or omega-6 FA intake and asthma or asthma-related symptoms. The role of omega-6 FA in inflammation, whether pro- or anti-inflammatory, is still not fully understood, but even less is understood about the complex relationship between omega-3 and omega-6 FA. The understanding of the interaction of fiber and omega FA on asthma and asthma-related symptoms require further study.

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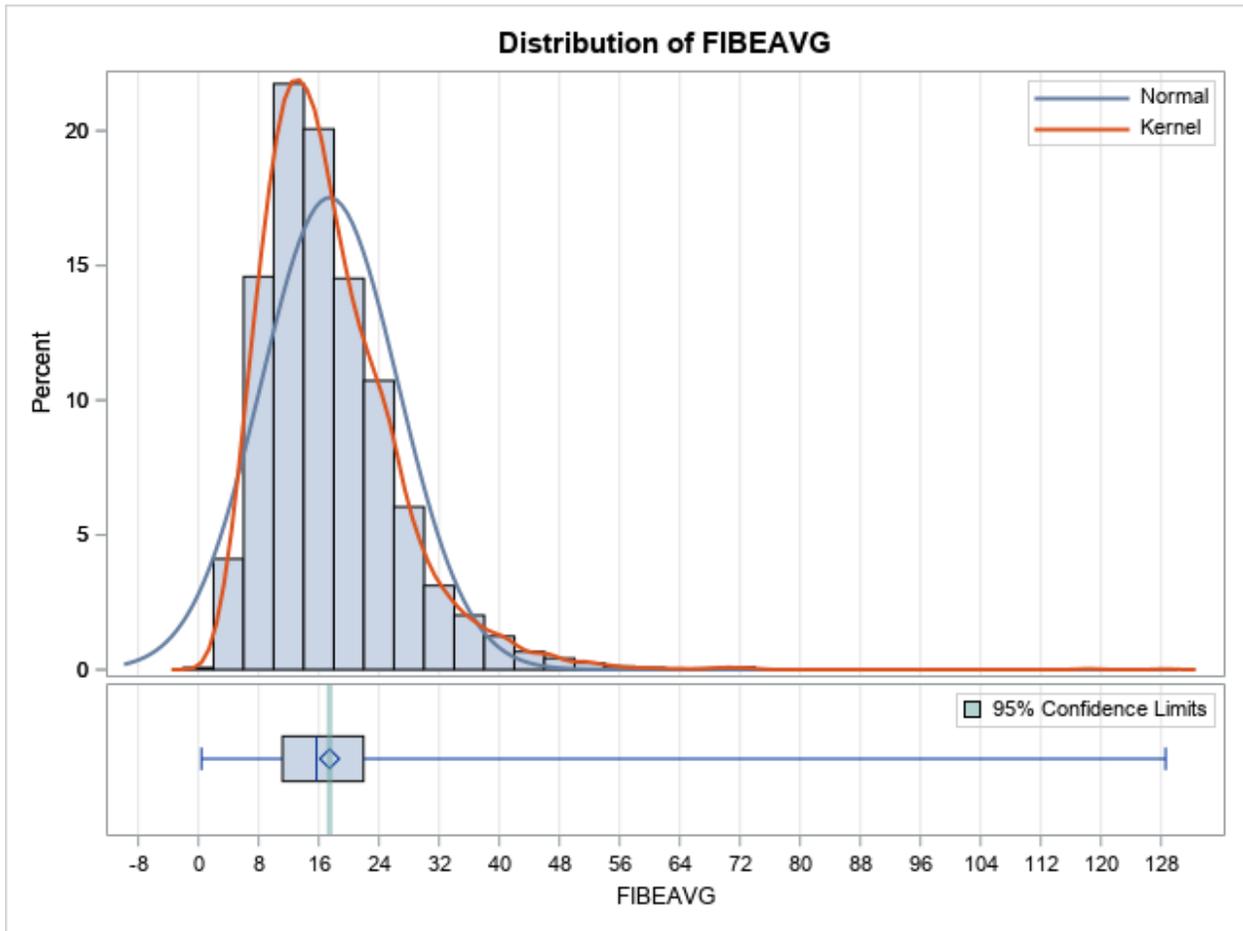
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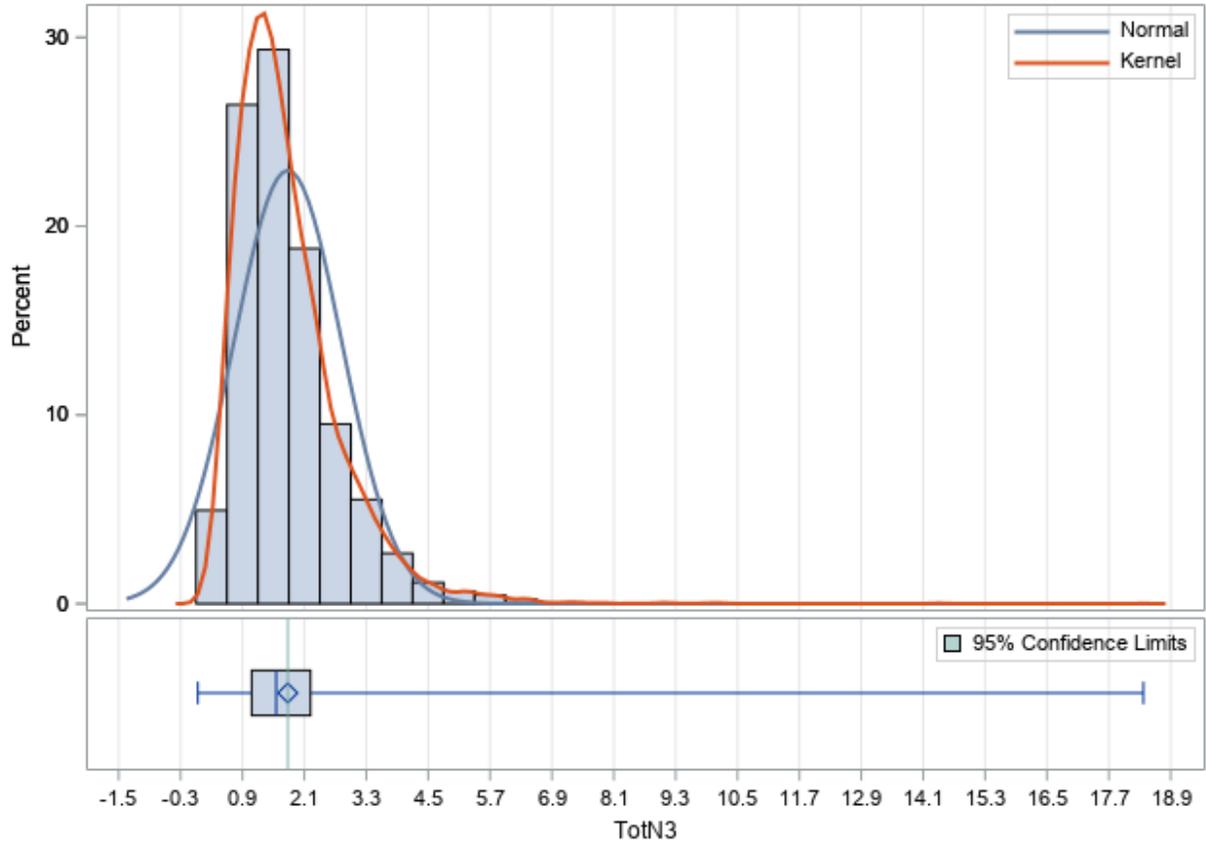
### **IRB Approval**

No IRB approval is required since the data used was obtained from the public database NHANES.

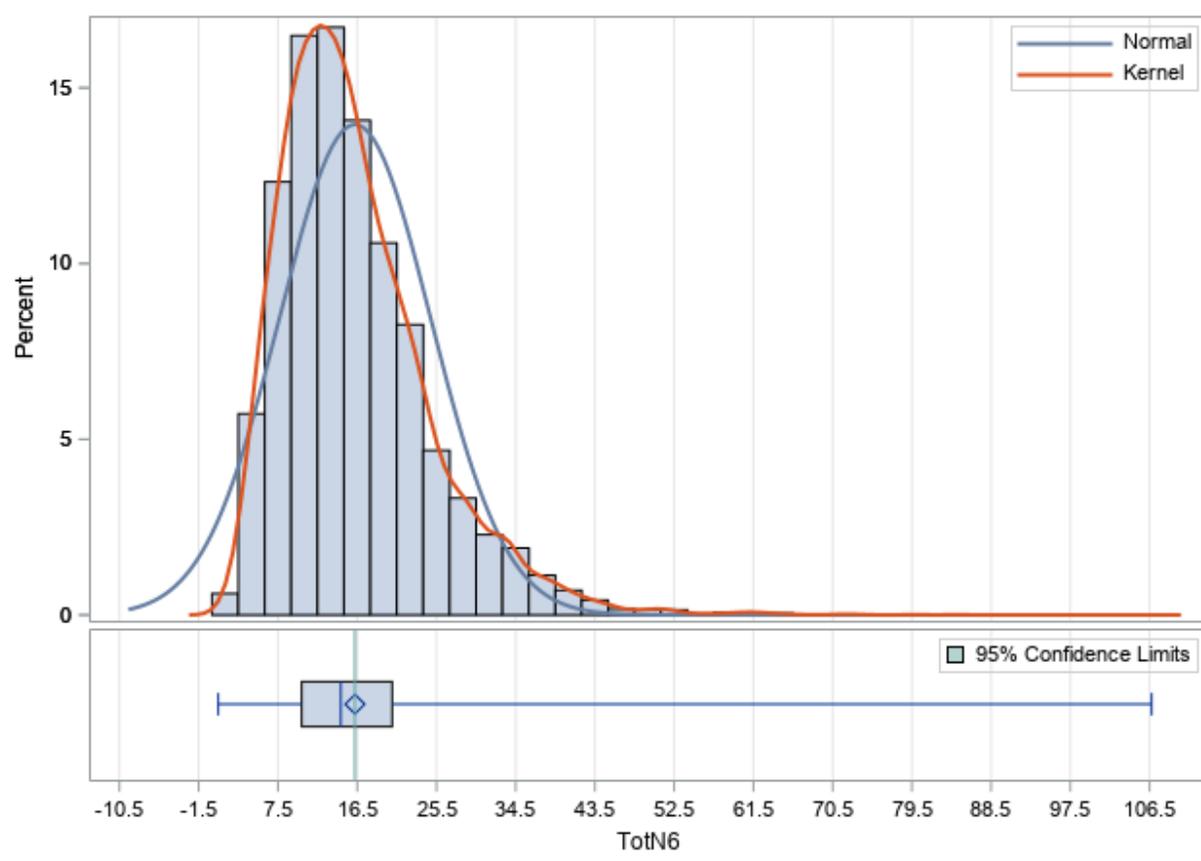
# APPENDIX A: Distribution of Diet Variables



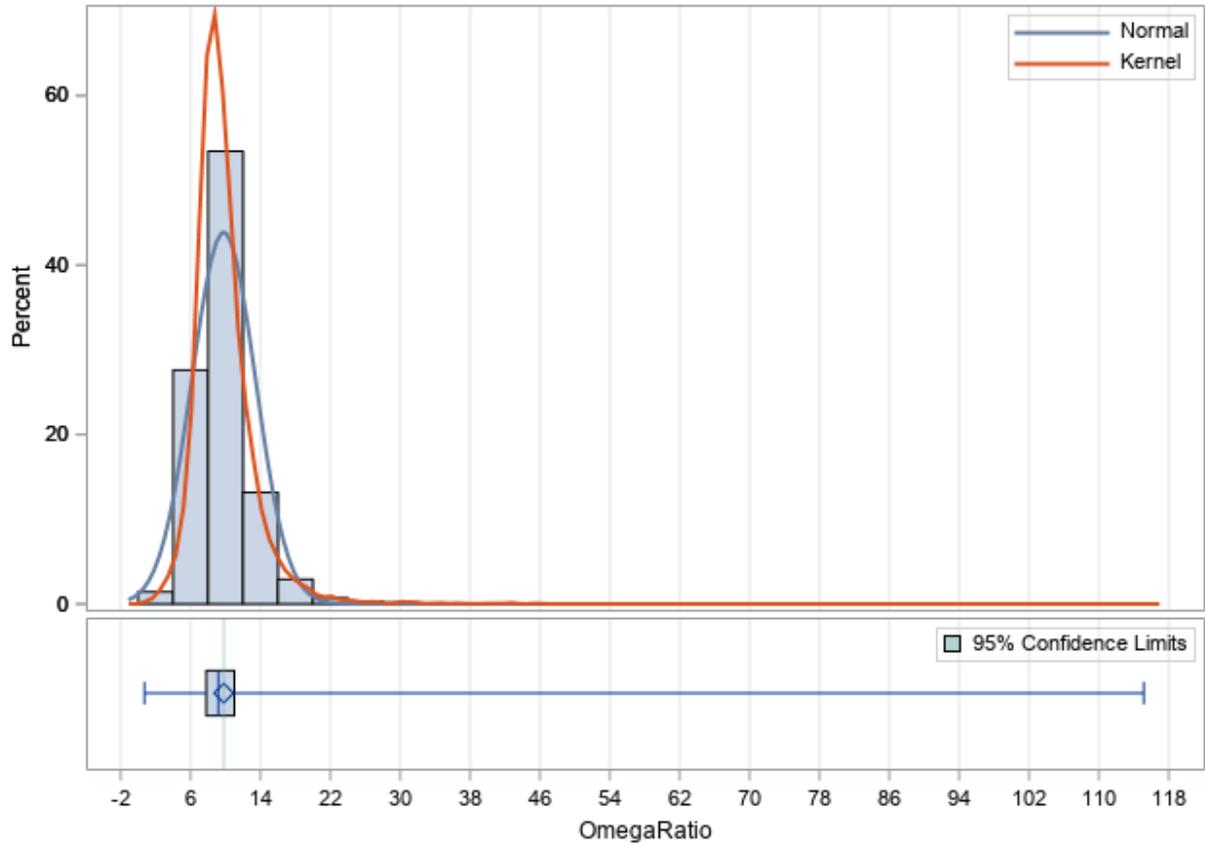
### Distribution of TotN3



Distribution of TotN6



Distribution of OmegaRatio



## **APPENDIX B: Biography and CV**

Korie A.S. Nelson is a Master of Public Health student at the University of Nebraska Medical Center. Her focus has been in Epidemiology: Infectious Disease. Ms. Nelson completed her summer internship at the Garrett County Health Department, Oakland, Maryland. While she does not currently hold a position in the Public Health field, she is excited to apply her newly acquired knowledge and other experiences to this area. Some of these experiences include being a Girl Scout leader, working as a vet tech, and serving six years in the military, with two overseas deployments. Ms. Nelson also holds a Master of Science in Dairy Nutrition from Utah State University and a Bachelor of Science in Microbiology from Colorado State University.

# Korie (Korinne) Ann Snyder Nelson

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## EDUCATION

### Master of Public Health- Epidemiology

*University of Nebraska-Medical Center*

Courses: Epidemiology, Planning and Evaluation, Leadership and Advocacy, Biostats I, Biostats II, Epidemiological Methods, Intro to SAS Programming, Emergency Preparedness: Response and Recovery, Applied Epidemiology, Chronic Epidemiology, Infectious Disease Epidemiology, Human Health and Disease, Epidemiological Methods in Infectious Disease Outbreak Investigation  
Capstone: The Effect of Fiber and Omega-6:Omega-6 Fatty Acid Ratio on Asthma and Asthma-related Symptoms in an Adult U.S. Population

Online

Expected May 2021

### Master of Science-Dairy Nutrition

*Utah State University*

Thesis: Enhancing the Proportions of Healthy Fatty Acids in Milk from Dairy Cows

Logan, UT

Dec. 2008

### Bachelor of Science-Microbiology

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May 2005

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## PUBLICATIONS

Nelson, K.A.S., Martini, S. (2009). Increasing omega fatty acid content in cow's milk through diet manipulation: Effect on milk flavor. *J. Dairy Sci.* 92, 1378-1386. doi:10.3168/jds.2008-1780.

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## PROFESSIONAL EXPERIENCE

### Applied Experience – Internship

*Garrett County Health Department- Education and Outreach Department*

-created template for annual report, assisted with community COVID testing, utilized data from Maryland Public Opinion Survey and created a PowerPoint presentation, compiled information for social media

May-July 2020

### Correspondence Clerk

*Internal Revenue Service*

-date stamped and sorted correspondence and money

Jan.-March 2019

### Associate and Cashier

*Old Navy*

-assisted customers, organized the store

Oct. 2008-Jan. 2010

### Veterinarian Technician/Receptionist

*Animal Care Veterinarian Hospital-Roy*

-assisted vets with medical procedures, helped clients

Sep. 2005-Oct. 2007

### Staff Sergeant/Fuels System Mechanic

*Colorado Air National Guard*

-deployed twice to Middle East for Operation Iraqi Freedom and Operation Enduring Freedom

Aug. 2000-Aug. 2006

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## VOLUNTEER AND LEADERSHIP EXPERIENCE

### Parent Reader and First Lego League Mentor

*Northeast Elementary, Cumberland, MD*

-read to second and fourth graders weekly, attend weekly FLL meetings, assist teachers

Sept. 2019-current

### Head Room Mom/Parent Volunteer

*Plain City Elementary, Plain City, UT*

Oct. 2013-Dec. 2018

-plan and hold holiday parties for multiple classes, assist teachers

**Daisy Leader**

*Troop 57 Ogden, UT*

July 2015-June 2017

-planned and led meetings for daisies, chaperoned fieldtrips, helped entire troop earn a special badge