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BOLD (BEEF IN AN OPTIMAL LEAN DIET) EFFECTS ON METABOLIC SYNDROME
(BOLD-X), PHASE 2: A 12-MONTH FOLLOW-UP STUDY

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ABSTRACT

In the United States, the obesity epidemic has been on the rise over the past couple of decades. Overweight and obesity are risk factors for several comorbidities, including type 2 diabetes, hypertension, and cardiovascular disease (CVD). Even a modest reduction in weight can significantly decrease an individual's risk for these diseases. Weight loss is a very challenging process, but maintaining one's weight loss over a long period of time can be even more difficult. The BOLD-X study investigated the effects of three diets (BOLD, BOLD⁺ and modified-DASH) that are low in saturated and total fat but contain variable amounts of total, plant, and animal protein on short-term controlled weight maintenance and weight loss, and free living weight loss. The total duration of the BOLD-X study was 6 months long. This thesis describes results from a one-year follow-up study which assessed the effects of each diet on weight loss maintenance and lipid profiles. Eighteen individuals participated in the one-year follow-up. The results found that for all participants, low-density lipoprotein cholesterol (LDL-C), systolic blood pressure (SBP) levels, and sodium intake increased significantly from the weight loss period to the one-year follow-up ($p \leq 0.05$). Even though weight increased in the one year follow up for two of the diet treatments (not in the BOLD diet treatment), energy expenditure also increased at the one-year follow-up for each diet. Further research is needed to determine the specific factors that predict weight regain in individuals and a continuation of the BOLD-X one-year follow-up study should be performed in order to include a larger population size and achieve more predictive results.

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**BOLD (Beef in an Optimal Lean Diet) Effects on Metabolic Syndrome (BOLD-X), Phase 2:
A 12 Month Follow-Up Study**

I. BACKGROUND AND SIGNIFICANCE

A. OBESITY AND CARDIOVASCULAR DISEASE

The obesity epidemic in the United States is growing at an alarming rate. Nearly two-thirds of Americans are overweight (BMI ≥ 25) or obese (BMI ≥ 30). Overweight and obesity are risk factors for several comorbidities, including type II diabetes, hypertension, and cardiovascular disease (CVD). According to the NHANES study (National Health and Examination Survey) in 2007-2008, the prevalence of overweight and obesity combined is 68.0% (95% CI, 66.3%-69.8%) in the United States (Flegal et al., 2010). However, a simple weight loss of 5-10% of an individual's body weight can significantly decrease the risk for these health issues (Kraschnewski et al., 2010). There has been an association between overweight and overall mortality. In a 30 year follow-up of men in the Framingham Heart Study, the group with the lowest mortality rates was those individuals in the desirable weight range (100%-109% of baseline weight). Nonsmoking men who were overweight ($>110\%$ baseline weight) had a 3.9 times greater risk of mortality than men of desirable weight (Garrison & Castelli, 1985). Given the current obesity statistics, it is necessary to examine and better understand how individuals who are successful at weight loss are able to maintain their reduced body weight.

B. LONG TERM WEIGHT LOSS MAINTENANCE

Much of the research surrounding weight loss interventions has shown the effectiveness of weight loss through dietary modification, yet there appears to be a high prevalence of weight regain following reductions in energy intake. Following a weight loss intervention, most

individuals observe a return to their baseline or original weight (Wadden et al., 1989). In a recent meta-analysis, Franz and colleagues evaluated the results of 59 clinical weight loss trials. Interventions that included food and meal planning strategies (diet alone, diet and exercise, and meal replacements) elicited a mean initial weight loss of approximately 5% to 9% from starting weight during the first 6 months. Weight loss plateaued at approximately 6 months and stabilized at 4.8% to 8% at 12 months (Franz et al., 2007). These findings are consistent with the American Dietetic Association's (ADA) Adult Weight Management Evidence-Based Nutrition Practice Guidelines, which recommends that weight loss and weight-maintenance therapy consist of a comprehensive program that includes diet, physical activity, and behavioral therapy. Further, weight loss interventions should last at least 6 months, at which time the continuation of professional support appears to be most effective in preventing weight regain (ADA 2006).

The American Dietetic Association's position on weight management attributes the success of weight management to many different variables. These include the amount of initial weight loss, comorbid conditions, presence of depression, perception of weight loss success, level of self-monitoring, level of physical activity, type of intervention (including frequency of contact), coping style, and stressful life events. However, the ADA also recognizes that knowledge and research on weight maintenance must consider many issues, including the amount of weight loss, weight loss duration, time between weight loss and evaluation of weight maintenance, and minimum length of weight maintenance (Seagle et al., 2009).

Maintaining weight loss over a long time period, from months to years, is an arduous struggle for most individuals, and it is now known that weight loss is compromised for most individuals beyond 6 months. Yet the duration of the majority of weight loss research studies is limited to a period of 6 months. Since hypertension, Metabolic Syndrome (MetSyn), and

diabetes are often life-long, chronic conditions, long-term management of an individual's weight is important as it contributes to the development and exacerbation of these conditions.

According to Wing and Phelan, only 20% of individuals who lost 10% of their initial body weight were successful in maintaining their loss for at least 12 months (Wing & Phelan, 2005).

The Diabetes Prevention Program Research Group found that 57% of individuals who had participated in a lifestyle intervention program had lost 7% of their initial body weight after 6 months, but this number decreased to 38% by 12 months (Diabetes Prevention Program Research Group, 2002). The emphasis of a weight-management trial should therefore progress from a focus on weight loss to weight loss with weight maintenance. That is, participants must be able to implement lifestyle changes that will support dietary and physical activity behaviors to prevent weight regain.

Little is known as to why certain individuals are successful at maintaining weight loss. However, there is some evidence that the larger the greater the initial weight loss, the more likely that an individual will be able to maintain their weight in the long-term. This is consistent with the results of a meta-analysis by Astrup and Rossner, which found a correlation between a greater the initial weight loss (usually at 2 to 4 weeks of treatment) and long-term weight maintenance at a 1 to 5 year follow-up (Astrup & Rossner, 2000).

Studies show that those individuals who maintained weight loss were those who incorporated physical exercise and self-monitoring. One of the largest studies of weight loss and maintenance is the National Weight Control Registry (NWCR). This is a longitudinal study in which participants included those who have lost at least 30 pounds and maintained this loss for at least one year. In this particular study, 90% (714 of 793 subjects) of participants who had reached their weight loss goal at the one-year follow-up to the beginning of the program were examined.

At the one-year follow-up, nearly 35% had gained at least five pounds or more. These “gainers” all had similar characteristics compared to those who had maintained their weight. They reported being cycling dieters (many constant attempts at dieting and losing weight) several times throughout their life. Gainers also weighed more at their maximum life weight, had lost a greater percentage of that maximum life weight more than 30%), and weighed heavier at the initiation of the study. Behavioral and psychological characteristics of gainers include decreases in restraint, increases in hunger, dietary disinhibition, and binge eating. Overall, the study found that those who had maintained their weight loss for 2-5 years were 50% less likely to subsequently regain the weight (McGuire et al., 1999). In other studies, it is quite common that 30-35% of the weight that a person loses will be regained within one year after treatment (Turk et al., 2009).

In a study conducted by Klem et al., 629 women and 129 men who had participated in the NWCR were examined. These individuals had lost at least 30 kg and maintained a minimum weight loss of 13.6 kg over five years. The subjects were asked to rate the difficulty of the weight loss compared to the difficulty of maintaining the weight loss. Nearly a third reported that the weight loss and weight loss maintenance were equally difficult or easy and a quarter found that weight maintenance was more difficult than weight loss (Klem et al., 1997).

In another study, researchers examined weight loss at 6 months and at 12 months in obese individuals assigned to either a low carbohydrate or conventional diet. The low carbohydrate diet consumed less than 30 grams of carbohydrate per day and the conventional diet group consumed less than 500 calories per day (with less than 30% of calories from fat). The study consisted of 79 severely obese women (average BMI of 43) with 39% having diabetes and 43% with Metabolic Syndrome. At six months, those on a low carbohydrate diet lost more weight

than those on a conventional low fat diet (mean [\pm SD], -5.8 ± 8.6 kg vs. -1.9 ± 4.2 kg) and also had greater decreases in triglycerides ($-20\% \pm 43$ vs. $-4\% \pm 31$). At the one-year follow-up to the study, the conventional diet group continued to lose weight more than the low-carbohydrate group, but these results were not statistically significant. By one year, however, those in the low carbohydrate group had a greater decrease in triglycerides and less of a decrease in HDL cholesterol (Stern et al., 2004). This is an example of a study that successfully compared weight and metabolic changes in results between six months and at a one-year follow-up.

Several studies have investigated the efficacy of different macronutrient profiles on weight loss (Kansinger et al., 2005, Gardner et al., 2007, Sacks et al., 2009). These trials generally conclude that weight loss is most successful in participants who follow a dietary plan that includes a reduction in calories. Indeed the most recent and largest intervention trial to investigate this to date (Sacks et al., 2009) concluded that macronutrient differences are less important than an overall hypocaloric diet. A plateau in weight loss was observed at 6 months with weight regain after 12 months. However macronutrient variances elicited different blood pressure (BP), lipid and lipoprotein lowering effects, which followed a similar pattern to those of weight stable dietary interventions (Appel et al., 2005). These findings are of interest as they suggest that heart healthy diets can elicit beneficial cardiovascular (CV) effects beyond weight loss, and emphasize the importance of finding a dietary intervention that can be implemented over an extended period of time (i.e. longer than 12 months). Higher protein diets have also shown to be more effective for loss of fat mass and improvements in body composition during initial weight loss and long-term weight maintenance (Layman et al., 2009).

Delbridge and colleagues also investigated the macronutrient profile of weight loss diets. The study consisted of two phases with 70 male and 71 female subjects. The first phase was a

three month very-low-energy diet. The second phase was a 12 month weight management phase in which the participants were randomly assigned to either a high protein (HP) diet or high carbohydrate (HC) diet. At the end of the 12 months, both diet groups lost significant weight, with no statistically significant difference between the two groups. There was a difference, however, in blood pressure. From the baseline of the study, the mean decrease in systolic blood pressure for the HP group was 14.3 ± 2.4 mm Hg and 7.7 ± 2.2 mm Hg for the HC group (Delbridge et al., 2009). Thus, even though both diet groups maintained similar weight loss, the high protein group had a greater decrease in systolic blood pressure.

Diet adherence has also been found to be a factor in predicting weight regain after weight loss. In a study by Corral et al., 116 healthy weight (BMI <20-25) postmenopausal women who had recently lost 12.2 ± 0.27 kg on a low-calorie diet of 800 kcal/day were examined and studied. The study consisted of a one-year and two-year follow-up. During the first year of follow-up, the women received dietary education classes. The researchers determined the participants dietary adherence based on changes in body composition determined by DXA before and after the weight loss intervention, as well as total energy expenditure (TEE) determined by doubly labeled water. The women were placed in either a lower (low adherence of $1,573 \pm 33$ kcal/day) or higher (high adherence of 644 ± 74 kcal/day) tertile. Results showed that percent weight regain at the one-year follow-up was greater in the low adherence group ($31.0 \pm 5.0\%$ vs. $68.7 \pm 6.4\%$). And at the two-year follow-up, weight regain was greater for both groups, but those in the low adherence group gained significantly more weight ($51.6 \pm 9.5\%$ vs. $99.0 \pm 12.3\%$) (Corral et al, 2010). This study shows that weight regain after a weight loss intervention is correlated with dietary adherence of participants. Because there was greater weight regain at

the two-year follow-up than at the one-year follow-up, this shows that weight regain is also correlated with the amount of time after the intervention.

C. EFFECTS OF BLOOD LIPIDS AND CLINICAL RISK FACTORS ON CVD RISK

LDL-C has shown to promote early atherosclerosis and may be one of the biggest predictors of CVD risk. According to a study by Cannon et al, for every 1.8 mg/dL decrease in LDL-C, risk of a cardiovascular event decreases by 1%. Saturated fatty acids (SFA) are the major source of LDL-C in the westernized diet, the major sources being red meat, full-fat dairy, and tropical oils (palm, palm kern, and coconut oils). It is advised to replace SFA with unsaturated fatty acids because of its lowering effects on LDL-C, TC, and TC:HDL ratio (Katcher et al., 2009).

In the Seven Countries Study conducted between 1958 and 1964, dietary characteristics were collected from individuals in sixteen cohorts in seven different countries. The population included 12,763 men ages 40-59. A 25 year follow up of this study was performed in 1987, and found that there was a significant positive association between SFA intake from lauric, myristic, palmitic, and stearic acid and 25-year mortality (Gardner et al, 2007).

Lower HDL-C has also been associated with greater incidence of CVD and cardiovascular events. In the Lipid Research Clinics Prevalence Study conducted from 1972-1976, a follow up of 8,825 male and female participants were examined. After control allowing for age, low LDL-C, BMI, TG, SBP, and smoking, an inverse relationship was found between high HDL-C and CVD mortality. The mortality rate ratio for men was a decrease in 10 mg/dL increments for HDL-C and an increase in 30 mg/dL increments for LDL-C (Jacobs et al., 1990).

Another well-accepted CVD risk factor is hypertension or high blood pressure. There is a positive linear relationship between blood pressure levels and CVD risk of myocardial

infarction, heart failure, and stroke. High blood pressure is a common epidemic across the United States, with nearly 55 million individuals diagnosed with hypertension. According to the Seventh Report of the Joint National Committee (JNC VII), high systolic blood pressure is more of a CVD risk factor than diastolic blood pressure. Also, starting at 115/75 mm Hg, CVD risk is doubled with every 20/10 mm Hg increment increase (Chobanian et al., 2003).

In another study, 436 participants with high total cholesterol (mean 260 mg/dL) were assigned to either a control diet or the DASH diet (high amounts of fruit and vegetables, low-fat dairy, low saturated and total fat). The diets were administered for eight weeks while weight was maintained. The result of the diet treatments was the DASH diet significantly lowered total cholesterol, LDL, and HDL levels, without a significant change in triacylglycerol levels (Obarzanek et al., 2001). Therefore, the study concluded that the DASH diet had beneficial effects on blood lipids (except for the HDL lowering mechanism) and reduced the risk for CVD.

D. EFFECT OF PHYSICAL ACTIVITY ON WEIGHT MAINTENANCE

Another important factor in assessing an individual's likelihood of weight loss maintenance in the long term is the amount of participation in physical activity and exercise. Meaning that, those individuals who participate in regular physical activity, maintain their weight more successfully than those who do not participate in any physical activity. There are current physical activity recommendations from the American College of Sports Medicine and the American Heart Association for healthy adults, aged 18-65, in order to maintain a healthy weight, prevent chronic diseases, and improve physical fitness. These recommendations include moderate-intensity aerobic activity 5 days/week for a minimum of 30 min/day or vigorous-intensity aerobic activity 3 days/week for a minimum of 20 min/day. This can be achieved in short 10 minute bouts throughout the day. Moderate-intensity physical activity includes any

activity that accelerates heart rate and can be equivalent to a brisk 2 mile walk. Vigorous-intensity physical activity is any activity that causes rapid breathing and increased heart rate, such as jogging. It is also recommended that adults perform exercises to improve muscular strength and endurance a minimum of 2 days/week (Haskell et al., 2007).

As stated earlier, an example of a moderate-intensity physical activity is brisk walking. One study attempted to show the effects of walking training on 82 premenopausal women, ages 30-45 with a BMI between 30-45. The study consisted of a 12-week weight loss phase in which the women consumed a very-low-energy diet. Then the women were separated into three groups for a 40-week maintenance period: one group who participated in no exercise, a walk-1 group who was calculated to expend 4.2 MJ/wk (approximately 2-3 hrs/wk), and a walk-2 group who was calculated to expend 8.4 MJ/wk (approximately 4-6 hrs/wk). The women were to walk within 50-60% of their maximum heart rate. At the end of the maintenance period, the control group gained around 2 kg, whereas the walking groups' weight remained stable (0.6-0.7 kg loss). The results showed at the one- and two-year follow-up, that the walk-1 group regained less weight and had a less increase in waist circumference than the control group, but the walk-2 group did not change significantly from the control group. Also, at the start of the study, 27 participants had the characteristics to define them as having Metabolic Syndrome. After the weight loss phase, only 11 participants had Metabolic Syndrome (Fogelholm et al., 2000). This indicates that even a moderate-intensity walking regimen reaps many significant health benefits.

In a study done at Boston University Medical Center, 160 male participants were assigned to one of four diet treatments: a balanced calorie-deficit diet (1000 kcal balanced diet), a protein sparing-modified fast (1000 kcal, no carbohydrate and high biologic protein sources), as well as a liquid form of each. The individuals in each diet treatment were assigned to an

exercise and non-exercise group. The non-exercise groups regained 60-92% of their baseline weight at the 6 and 18-month follow up. There were no significant differences between the diet groups. However, resting heart rate and systolic and diastolic blood pressure did not significantly change in the exercise group, but the values in the non-exercise group all returned to their baseline values. HDL-C in the exercise group also significantly increased. This study concluded that individuals, who live an active lifestyle (exercising three times per week in order to expend 1500 kcal/week) are the most successful at maintaining weight loss (Pvalov et al., 1989).

E. THE BOLD-X STUDY & PREVIOUS RESEARCH BY PRINCIPLE INVESTIGATOR

The current BOLD-X trial investigates the effects of three diets (called BOLD, BOLD⁺ and modified-DASH) that are low in saturated and total fat but contain variable amounts of total, plant and animal protein on short-term controlled weight maintenance and weight loss, and free living weight loss. The total duration of the BOLD-X study was 6 months and therefore fails to evaluate the long-term efficacy of these diets. The purpose of the current one-year follow-up study was to evaluate the long term effects of each of these diets. This follow-up assessment will include data on current dietary intake patterns to assess each participant's ability to adapt each of the test diets to the free living environment while preserving a sufficient caloric reduction to maintain weight loss.

Dr. Kris-Etherton's research group has conducted numerous controlled feeding studies assessing the effects of different diets on CVD risk factors. Previous collaborative studies from Dr. Kris-Etherton and other's laboratories show that cholesterol-lowering diets higher in total fat from unsaturated fat and low in SFA, trans fat and dietary cholesterol (10), and higher in protein (9), improve lipid levels. These results suggest that a diet high in unsaturated fatty acids and

other cardioprotective nutrients, such as fiber and plant sterols/stanols, can accommodate the inclusion of lean beef, and may provide additional benefits for weight loss. Dr Kris-Etherton is the PI on the BOLD (Beef in an Optimal Lean Diet) grant which investigates whether this dietary pattern can lower multiple CVD risk factors and, hence significantly improve CVD risk.

My personal role in the BOLD-X one-year follow-up study was to contact the participants who had completed the study and had consented to participate in the follow-up. I then scheduled appointments at the General Clinical Research Center (GCRC) to measure participants' weight status, collect and process blood samples for analysis, and administer 3-day food and physical activity diaries. I then analyzed the food diaries by entering the data into a food processor software program. I also researched current literature for scholarly information to incorporate in this thesis and wrote the undergraduate thesis according to the requirements of the Schreyer Honors College at Penn State.

II. HYPOTHESIS

A. PRIMARY HYPOTHESIS:

Individuals who consume diets that include lean beef as the primary source of protein with average (18% BOLD) or moderate-high (28% BOLD⁺) total protein intake will maintain or improve their weight loss at 12 months compared to individuals who consume a modified-DASH diet.

B. SECONDARY HYPOTHESIS:

Individuals who consume diets that include lean beef as the primary source of protein with average (18% BOLD) or moderate-high (28% BOLD⁺) total protein intake will show similar or greater reductions in CVD risk as determined by reductions in select lipids and lipoproteins, and BP at 12 months compared to individuals who consume a modified-DASH diet.

III. OBJECTIVE

To determine whether individuals who consumed one of three heart-healthy diets that provided protein from predominantly lean beef (either 5 oz or 7 oz per day, referred to as BOLD or BOLD⁺, respectively) or plants (modified-DASH diet), continued their assigned diet and maintained their weight and lipid profile levels 12 months after beginning the BOLD-X study.

IV. METHODS

A. SUBJECTS AND ELIGIBILITY

All procedures were approved by the Institutional Review Board of the Pennsylvania State University (IRB #29336). To determine eligibility for the BOLD-X study, each participant completed a phone interview followed by a clinical screening appointment. All subjects reviewed and signed an Informed Consent document. All participants who had completed the BOLD-X study and agreed to being contacted for future studies were eligible to participate. Inclusion criteria were obese male and female subjects (BMI 27-42 kg/m²) aged 30-60 years with three of the follow risk factors (defined as having Metabolic Syndrome):

- Abdominal obesity [waist circumference >102 cm (40 inches) in men and >88 cm (35 inches) in women]
- Elevated blood glucose [>110mg/dl (5.6 mmol/L)]
- Elevated TG [150 mg/dl (1.7 mmol/L)]
- Low HDL-C [\leq 40 mg/dl (1.03 mmol/L) in men and \leq 50 mg/dL (1.29 mmol/L) in women]
- Hypertension (SBP >130 mmHg or DBP >85 mmHg)

Additional exclusion criteria for all subjects included:

- A history of myocardial infarction, stroke, diabetes mellitus, liver disease, kidney disease, and thyroid disease (unless controlled on medication)
- Lactation, pregnancy, or desire to become pregnant during the study
- Intake of putative cholesterol-lowering supplements (psyllium, fish oil capsules, soy lecithin, niacin, fiber, flax, and phytoestrogens, stanol/sterol supplemented foods)

- High alcohol consumption (≥ 14 drinks/week)
- Participation in regular physical activity (>1 formal/session/week)
- Lipid, BP, or glucose lowering medications*

*Subjects were removed from lipid and glucose lowering medications prior to beginning the study in consultation with their physician.

All subjects who completed the BOLD-X study were eligible for participation in the one-year follow-up study. Subjects were contacted by telephone or email. Of the 36 individuals who had completed the BOLD-X study, 18 successfully completed the one-year follow-up study.

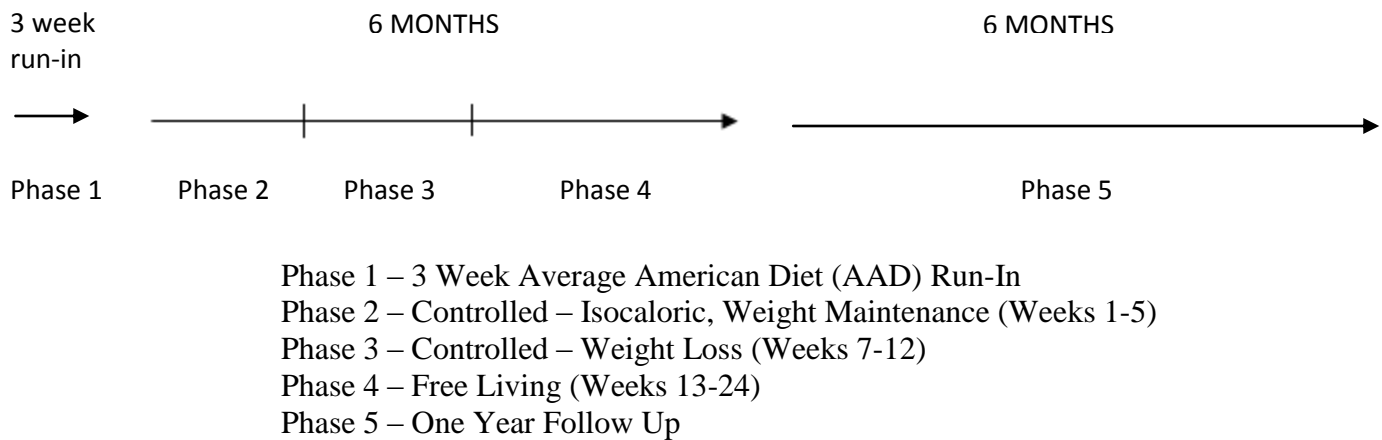
B. EXPERIMENTAL DESIGN

Subjects were asked to complete two consecutive clinical appointments that were scheduled at least 12 months from their start date in the BOLD-X study. The clinical appointments were conducted at the General Clinical Research Center (GCRC) at the Pennsylvania State University campus in University Park, PA.

The design of the Bold-X Study with the one year follow up is described in Figure 1. The Bold-X study consisted of a three week run-in of controlled feeding of an Average American Diet (AAD) to establish standardized baseline values. Phase two was 5-week isocaloric weight maintenance phase in which subjects were randomly assigned to one of three diets (BOLD, BOLD+, or modified-DASH). This was a controlled feeding phase in which participants received precisely measured and weighed meals through the Metabolic Diet Study Center in accordance with their assigned diet and calorie level needed. The third phase was a six week weight loss phase and incorporated exercise along with the control diets. Participants received physical activity and dietary counseling in accordance with their specific intervention diet. During the fourth phase, subjects were placed in a free living environment. During this

phase, participants no longer received meals through the Metabolic Diet Study Center and no longer received dietary and physical activity counseling. And finally, the last phase was the follow-up time point one year from the start date of the study (or six months after phase 4).

Figure 1. BOLD-X Study with One-Year Follow-Up Design



C. CLINICAL MEASUREMENTS

Anthropometric measures (weight, waist circumference) were taken at the clinical follow up appointments. The participant's weight was measured on a digital scale on two consecutive days. The average of the two measurements was used as the data for weight. Waist circumference was taken on day one of the follow up time point. Two separate measurements were taken and the average of the two was calculated for data. Participants' height had already been taken during their first clinical appointment at the baseline time point at the beginning of the BOLD-X study.

On each consecutive day of the clinical follow up appointments, a fasting blood draw (~25 ml per day) was taken by trained professional nurses at the GCRC. The fast before each blood draw consisted of subjects refraining from the consumption of food (except water).

Subjects also were asked to avoid strenuous or vigorous exercise twelve hours prior to the appointment, and to refrain from consuming alcohol forty-eight hours before the appointment. Subjects were denied a blood draw if they were taken certain medications or did not comply with these conditions.

After the blood was collected, it was sent to be analyzed by the diagnostic testing laboratory, Quest Diagnostics, in Pittsburgh, PA for total cholesterol (TC), low density lipoprotein cholesterol (LDL-C), high density lipoprotein cholesterol (HDL-C), triglycerides (TG), and glucose levels.

Blood pressure was taken by the trained nurses at the GCRC on day one. Participants were asked to sit without talking and legs uncrossed for five minutes in order to get an accurate reading. A standard blood pressure cuff was used to measure systolic and diastolic pressure. Three different measurements were taken at least one minute apart and the average of the last two readings was used as data. Additional blood samples were stored and frozen for potential use in future studies.

D. DIET AND PHYSICAL ACTIVITY DIARIES

In the week prior to their appointment, subjects were asked to complete two forms and questionnaires regarding their current diet and physical activity level. These included a 3-day food record and a 3-day physical activity record.

The 3-day food records were completed on two weekdays and one weekend day. Participants were asked to complete a 3-day food record during both the 6 month free living and one year follow up phases. However, the diet data that was used for this study during the weight loss period was calculated based on the specific type of diet treatment that was prescribed and calorie level for a particular individual. For each of the three days, subjects were asked to

indicate the date/day of the week, record each food item consumed, the time of day the food item was consumed, the specific amount or portion size, how the food was prepared, the type of cooking fat, and where the food was eaten. The data was entered into SAS to be statistically analyzed.

The 3-day physical activity record was adapted from a study by Bouchard et. al, who developed a procedure for assessing energy expenditure in population studies. The physical activity diaries consisted of recording all physical activities throughout a twenty-four hour time period for three days (one weekday and one weekend day). Each twenty-four hour time period was broken down into fifteen minute intervals. For each fifteen minute interval, subjects recorded any type of physical activity, the purpose of the activity, and the intensity of the activity. Intensity was rated on a scale from 1 to 10, 1 indicating rest or sleep and 10 indicating high intensity activities and manual work. This scale correlates to and amounts of MET's from 1-7.8. Physical working capacity was shown to be more significant in correlating with energy expenditure because of the variations in weight status and size. For the range of intensity of physical activity, the mean energy expenditure was calculated for kcal/kg/15 min (Bouchard et al., 1983). This procedure and formula is what was used to calculate energy expenditure for the participants in this study.

E. STATISTICAL ANALYSIS

A *proc* mixed analysis in the Statistical Analysis Software (SAS) for WINDOWS was used in order to test the differences between the five study periods, the differences between the three diet treatments, and the interactions of period by diet treatment effects. The p-values for the differences of least square means were calculated using a Tukey-Kramer adjustment. Significance was defined as $p \leq 0.05$.

V. RESULTS

Eighteen subjects participated in the follow up study. Baseline characteristics for all 18 participants were measured at screening, or entrance into the study, as shown in Table 1.

Table 1. Mean Baseline Characteristics for all Participants at Screening

Variable	Screening Value
Age (years)	48
Weight (lbs)	217
BMI (kg/m ²)	33
Waist Circumference (cm)	111
TC (mg/dL)	209
LDL-C (mg/dL)	129
HDL-C (mg/dL)	43
TG* (mg/dL)	180
Glucose* (mg/dL)	105
SBP* (mmHg)	125
DBP (mmHg)	86

*Values were log transformed

Table 2. Mean Baseline Characteristics For Each Diet Treatment at end ofAAD

Variables	DASH	BOLD	BOLD-X	P-Value
Participants (n)	6	4	8	
Age (years)	47.0	48.3	48.8	0.836
Weight (lbs)	217.4	219.2	215.9	
BMI (kg/m ²)	33.6	34.1	32.9	0.679
Waist Circumference (cm)	111.8	107.2	110.3	0.682
TC (mg/dL)	204.2	214.4	184.8	0.240
LDL-C (mg/dL)	133.3	136.3	108.6	0.145
HDL-C (mg/dL)	36.9	39.8	38.9	0.649
TG* (mg/dL)	165.5	189.6	176.5	0.570
Glucose* (mg/dL)	98.5	105.4	98.4	0.792
SBP* (mmHg)	128.9	115.7	125.8	0.209
DBP (mmHg)	84.2	81.5	85.6	0.571

*Values were log transformed.

Table 2 provides the baseline characteristics of all participants at the end of the AAD period. A Scheffe test was performed in order to determine the differences between the three diet groups at baseline. Because none of the p-values for any of the variables were statistically significant ($p \leq 0.05$), this shows that the participants in each of the three diet groups were not statistically different from one another at the start of the intervention treatment.

Table 3. Significant Effect of Each Variable During Study Periods ($p \leq 0.05$)

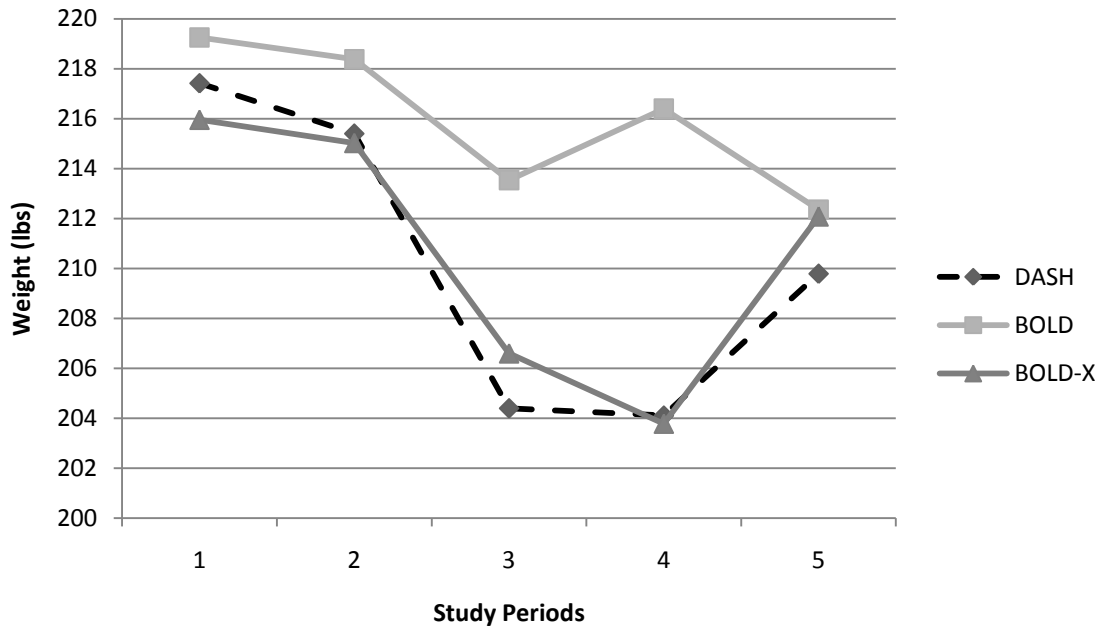
Variable	Period 1 (AAD)	Period 2 (Weight Maintenance)	Period 3 (Weight Loss)	Period 4 (Free Living 6 Months)	Period 5 (1 Year Follow-Up)
Weight (lbs)	222.4 ^a	221.5	213.3	212.5 ^a	217.2
BMI (kg/m ²)	33.6 ^B	33.4 ^c	33.2 ^b	32.1 ^{b,c}	32.9
Waist Circumference (cm)	110.2 ^D	110.5 ^E	107.1 ^{d, e}	107.4 ^{d, e}	107.7
TC (mg/dL)	195.6 ^f	185.0	173.8 ^F	196.8 ^f	204.0 ^f
LDL-C (mg/dL)	122.1	113.2 ^h	109.8 ^{g, h}	123.7 ^g	130.2 ^H
HDL-C (mg/dL)	37.5 ^{I, J}	33.1 ^{I, J}	36.4 ^{I, J}	41.0 ^J	42.3 ^J
TG* (mg/dL)	174.6 ^k	178.9 ^k	133.2 ^K	146.1	145.9
Glucose* (mg/dL)	100.6	105.3	100.6	99.9	103.1
SBP* (mmHg)	124.2 ^l	121.5	117.2 ^L	120.0	124.4 ^l
DBP (mmHg)	83.9	82.9	80.4	82.4	82.1

*These values were log transformed.

Mean values within a row sharing a common letter are statistically different, $p \leq 0.05$. An uppercase letter indicates that it is statistically different from its lowercase letter form.

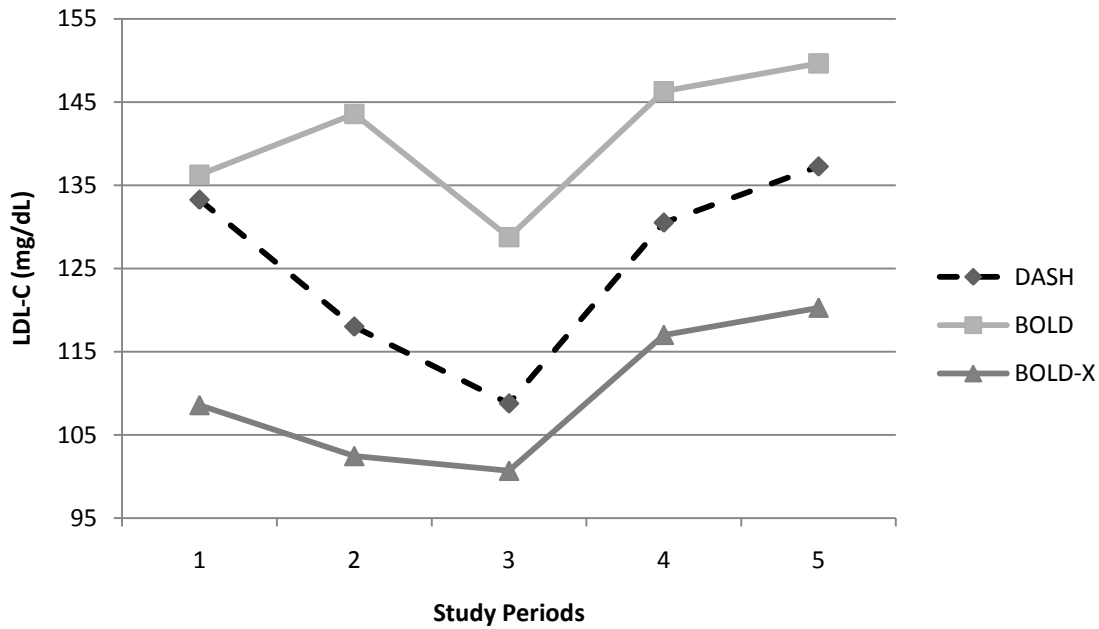
Table 3 represents the mean values for all participants during each of the five periods of the study for each variable. This table shows where there were significant differences ($p \leq 0.05$) from one period to another. Of particular interest to the one-year follow-up are weight status, TC, LDL-C, HDL-C, TG, and SBP values, which are explained below.

Figure 2. Mean Weight Status for Each Diet Treatment



The mean weight status of the participants in each diet group throughout all five periods of the study is illustrated in Figure 2. There were no period by treatment effects, but a significant main effect of period was observed at period 4 ($p=0.009$). Post-hoc analysis showed that body weight at period 4 was significantly lower than body weight at period 1 ($p=0.034$). A test for the differences of least squares means also found a trend toward a significant difference between the end of period 1 and period 3 ($p=0.06$). In general, weight status increased from period 4 to the one-year follow-up for the DASH and BOLD-X diet treatments. However, weight status decreased from period 4 to the one-year follow-up for the BOLD diet treatment.

Figure 3. Mean Serum LDL-C Levels for Each Diet Treatment



Overall, there was a steady decrease in LDL-C in each of the three diet groups from period 1 to period 3, and then a steady increase from the period 3 to period 5 (Figure 3). Post-hoc analysis showed that LDL-C was significantly lower at period 3 than at period 5 ($p=0.0006$) and LDL-C was also significantly lower at period 2 than at period 5 ($p=0.006$). LDL-C was also significantly lower at period 3 than at period 1 ($p=0.0379$).

Figure 4. Mean Serum HDL-C Levels for Each Diet Treatment

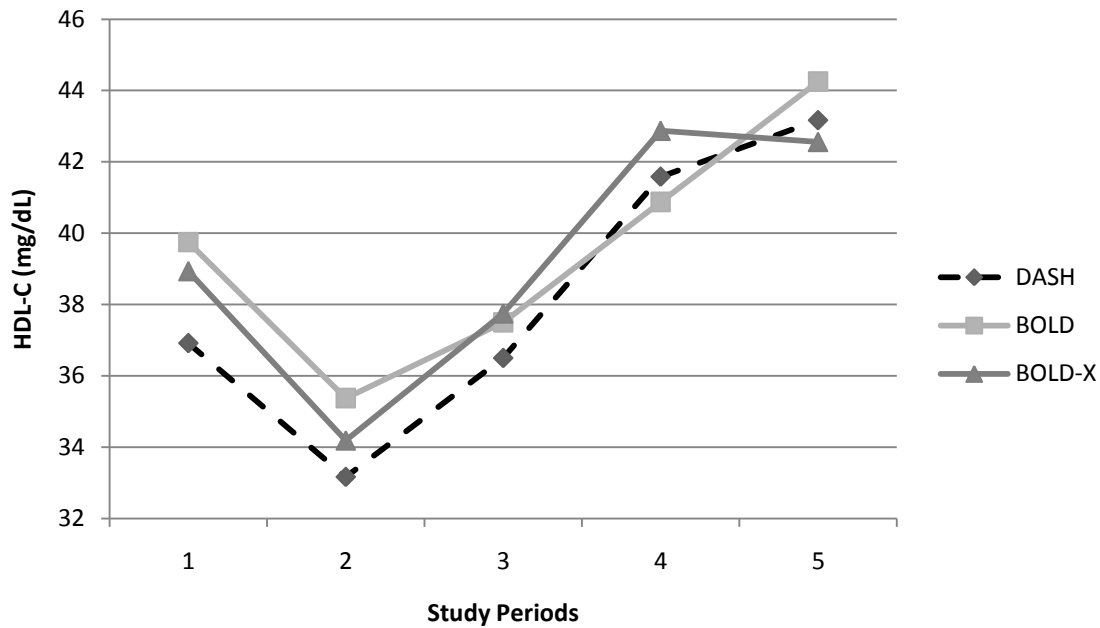
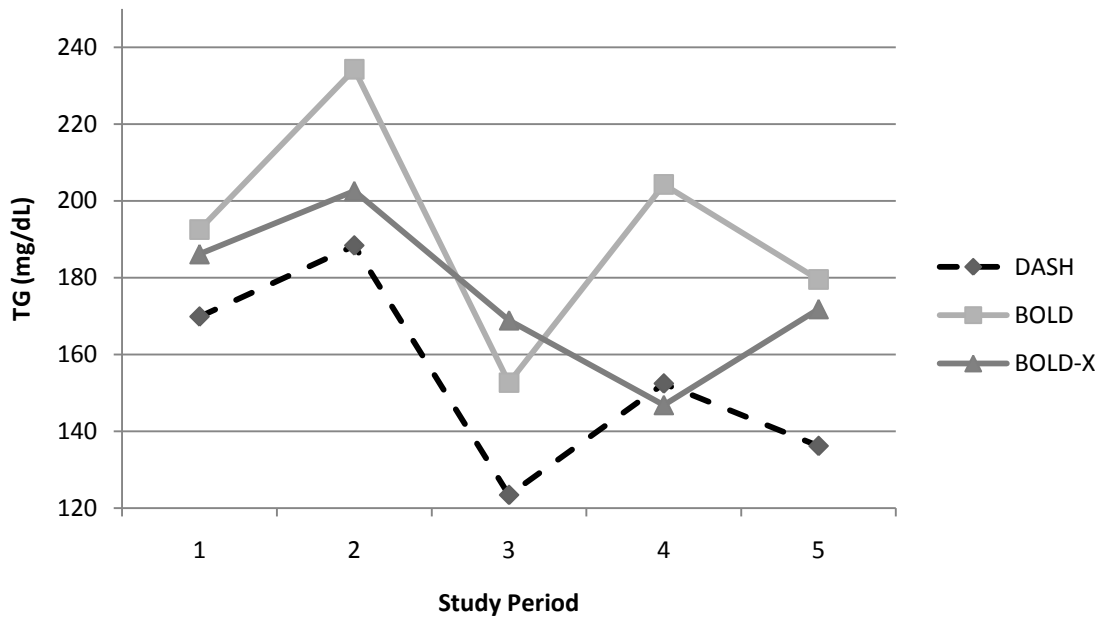


Figure 4 illustrates that, overall, there was a steady increase in HDL-C levels from period 2 to period 5. The significant increase in HDL-C levels for the entire population was observed during the fourth period of the study ($p < 0.0001$). Post-hoc analysis revealed that HDL-C levels were significantly higher at period 5 than at periods 2 and 3 ($p < 0.0001$).

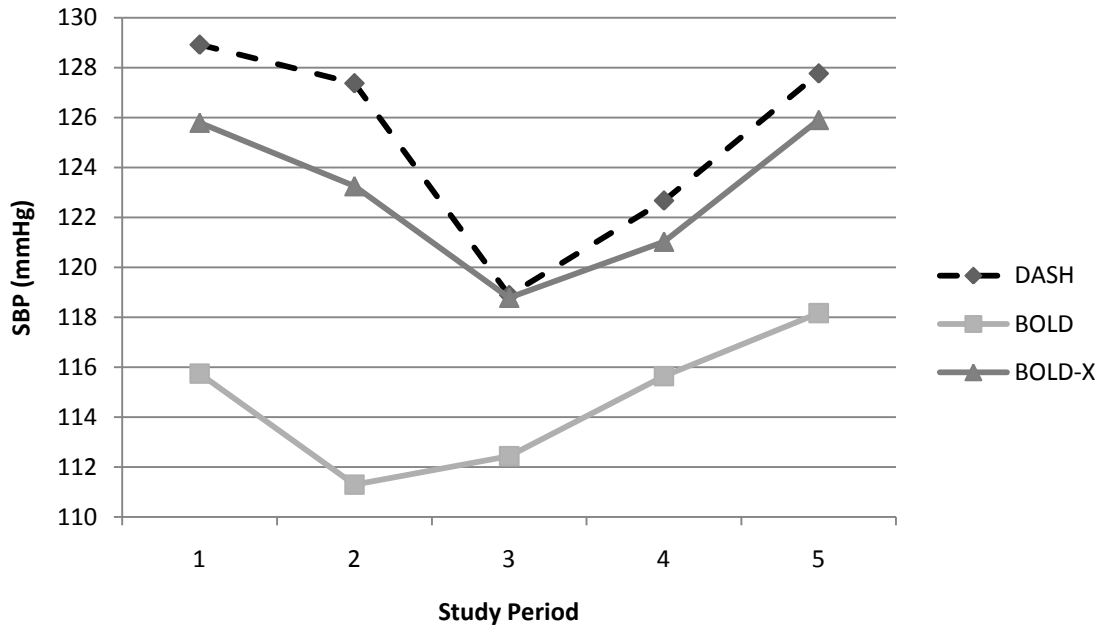
Figure 5. Mean Serum TG Levels for Each Diet Treatment*



*These values were log transformed.

Figure 5 shows that, for each diet treatment, there was an increase in serum TG levels from period 1 to the period 2, and then a significant decrease between period 2 and period 3 ($p=0.0022$). There was also a significant decrease in serum TG levels between periods 1 and 3 ($p=0.006$). Between periods 3 and 4, the DASH and BOLD treatments had a slight increase in serum TG levels, whereas the BOLD-X diet experienced a slight decrease. Between periods 4 and 5, the DASH and BOLD treatments both experienced a slight decrease in serum TG levels, whereas the BOLD-X treatment had a slight increase in TG levels.

Figure 6. Mean SBP Levels for Each Diet Treatment*



*Values were log transformed.

Figure 6 shows that, for each of the diet treatments, there was a significant decrease from period 1 to period 3 ($p=0.0073$) and a significant steady increase from period 3 to period 5 ($p=0.005$). Both the BOLD and BOLD-X treatment groups had higher SBP levels at the one year follow up than when the study began at the AAD period, but these values were not significant. The DASH treatment group had a slightly lower SBP level at the one-year follow-up than at the AAD period, but these values were not significant.

Table 4. Nutrient Intake Significant Differences Between Periods

Nutrient	Mean Intake by Period			Period Significant Difference (P-value \leq 0.05)		
	3 (Weight loss period)	4 (Free living period)	5 (One year follow up)	3 to 4	4 to 5	3 to 5
PRO (g)*	103.4	82.1	92.3	0.0200		
Total Fat (kcal)	568.1	770.1	723.1	0.0056		0.0272
Fiber (g)*	38.8	25.0	24.3	0.0005		0.0003
SFA (g)	14.36	26.43	21.57	<0.0001	0.0461	0.0015
Chol (g)*	133.7	203.0	234.3	0.0331		0.0098
Calcium (mg)	1322.9	957.3	1088.4	0.0052		
Magnesium (mg)	437.9	274.0	356.0	0.0002		0.0397
Potassium (mg)	4021.0	3122.7	2398.5	0.0209		<0.0001
Sodium (mg)*	2482.1	3058.2	3098.4			0.0092

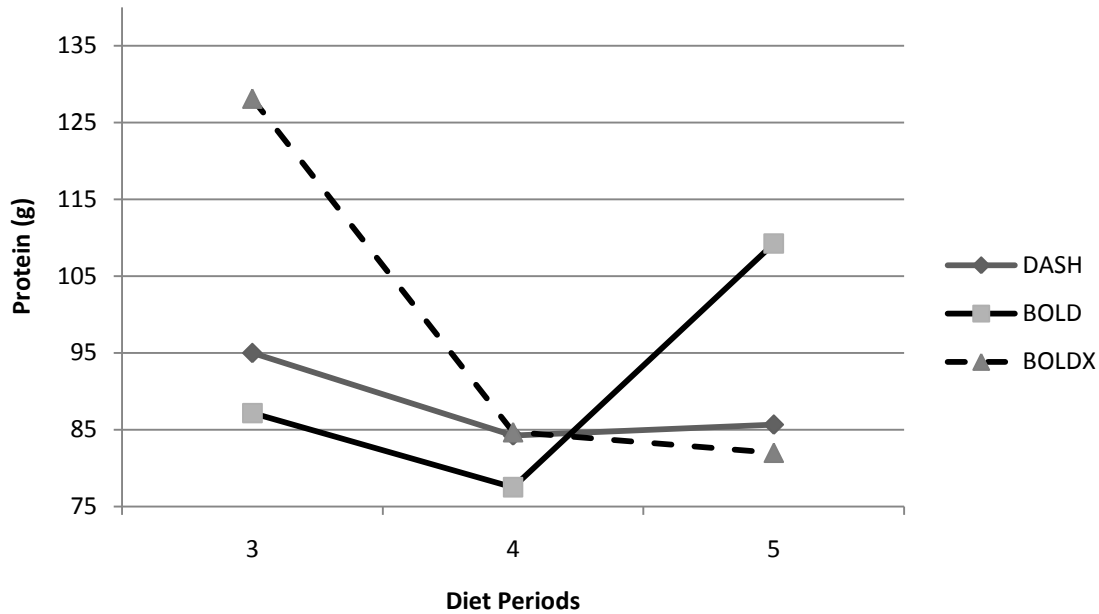
*Values were log transformed.

Table 4 represents the nutrient intakes of all participants that exhibited a significant difference between periods 3 to 4 or between periods 3 to 5. Protein intake decreased between period 3 and 4 ($p=0.02$) and then slightly increased again between periods 4 and 5. Total fat kcal intake increased between period 3 and 4 ($p=0.0056$) and then decreased slightly between periods 4 and 5. There was a significant difference between the increase in total fat kcals from period 3 to period 5 ($p=0.0272$). Fiber intake decreased from period 3 to period 4 ($p=0.0005$) and then slightly decreased from periods 4 to 5. There was also a significant decrease in total fiber intake from period 3 to period 5 ($p=0.0003$).

SFA intake significantly increased from period 3 to period 4 ($p<0.0001$). SFA was the only nutrient in this study that observed a significant decrease from period 4 to period 5

($p=0.0461$). The mean SFA intake at period 5 was still higher, however, than SFA intake at period 3 and these values were also statistically significant ($p=0.0015$). Total cholesterol intake increased between periods 3 and 4 ($p=0.0331$) and then increased yet again between periods 4 and 5 ($p=0.0098$). There was a significant increase in cholesterol between periods 3 and 5 ($p=0.0098$). In terms of calcium, there was a significant decrease in intake between periods 3 and 4 ($p=0.0052$), and a slight increase again between periods 4 and 5. Calcium intake at period 5, however, did not reach the calcium intake that was being consumed at period 3. Magnesium intake decreased significantly between periods 3 and 4 ($p=0.0002$) and slightly increased from periods 4 to 5, and significantly decreased between periods 3 and 5 ($p=0.0397$). Potassium intake decreased significantly between periods 3 and 4 ($p=0.0209$) and intake decreased yet again from period 4 to period 5. The decrease in potassium between periods 3 and 5 was significant ($p<0.0001$). Sodium intake increased slightly, without statistical significance, from period 3 to period 4 as well as between period 4 and period 5. However, the increase between period 3 and period 5 was statistically significant ($p=0.0092$).

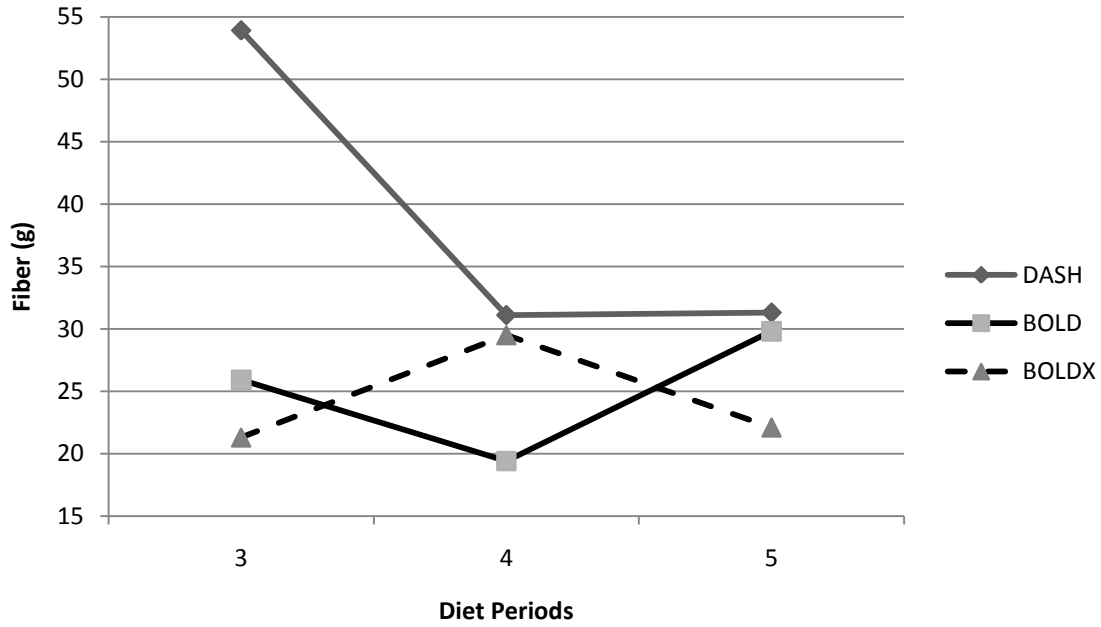
Figure 7. Protein Intake During Periods 3, 4, and 5 for Each Diet Treatment



*Values were log transformed.

Figure 7 represents protein intake, which exhibited significant period by treatment effects. The protein intake of the BOLD-X treatment during period 3 (BOLDX 3) was significantly higher than that of the BOLD treatment during period 3 (BOLD 3) ($p=0.0394$) and DASH 3 ($p=0.0309$). BOLD X 3 was significantly higher than BOLD X 4 ($p=0.0016$), DASH 4 ($p=0.0028$), BOLD X 5 ($p=0.0004$), and DASH 5 ($p=0.0048$).

Figure 8. Fiber Intake During Periods 3, 4, and 5 for Each Diet Treatment*



*Values were log transformed.

Figure 8 represents fiber intake, which exhibited significant period by treatment effects. The fiber intake of the DASH diet at period 3 was significantly higher than that of BOLD X 4 ($p=0.0204$), DASH 4 ($p=0.0055$), BOLD X 5 ($p=0.0083$), and DASH 5 ($p=0.0303$).

Figure 9. SFA Intake During Periods 3, 4, and 5 for Each Diet Treatment

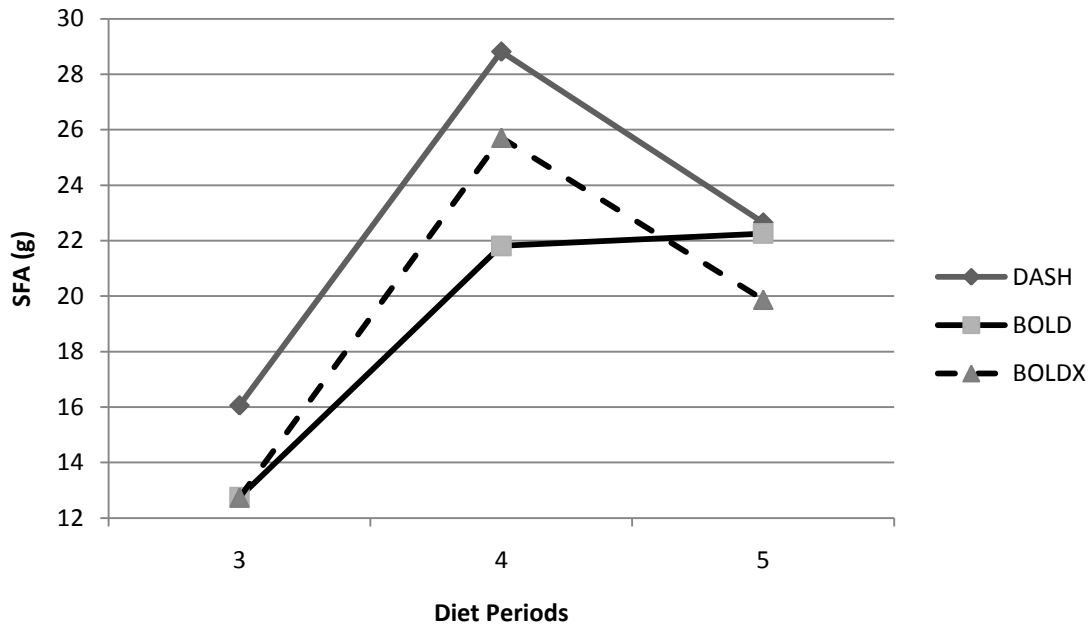
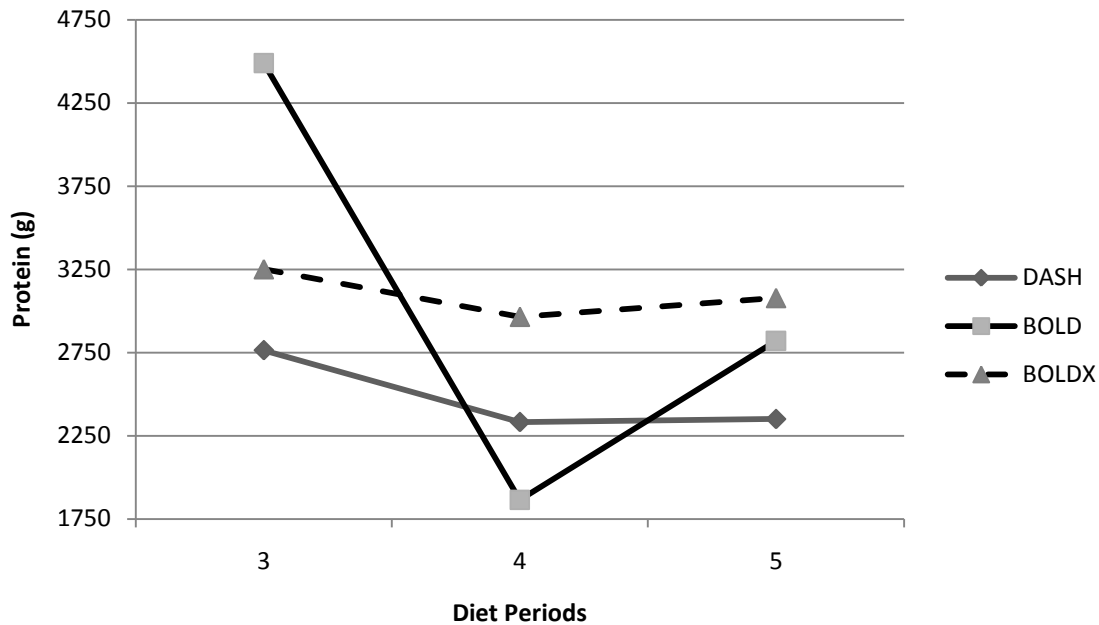


Figure 9 represents SFA intake for each diet treatment. Even though there were no significant period by diet treatment effects, there were, however, significant effects between periods. For the entire study population, SFA intake significantly increased from period 3 to period 4 ($p < 0.0001$). There was also a significant increase in SFA from period 3 to period 5 ($p = 0.0015$). SFA was also the only nutrient in this study that observed a significant decrease from period 4 to period 5 ($p = 0.0461$). For the DASH and BOLD-X diets, SFA intake decreased from period 4 to period 5 (-6.2 and -5.8, respectively). In the BOLD group, however, SFA slightly increased from period 4 to period 5 (+0.447).

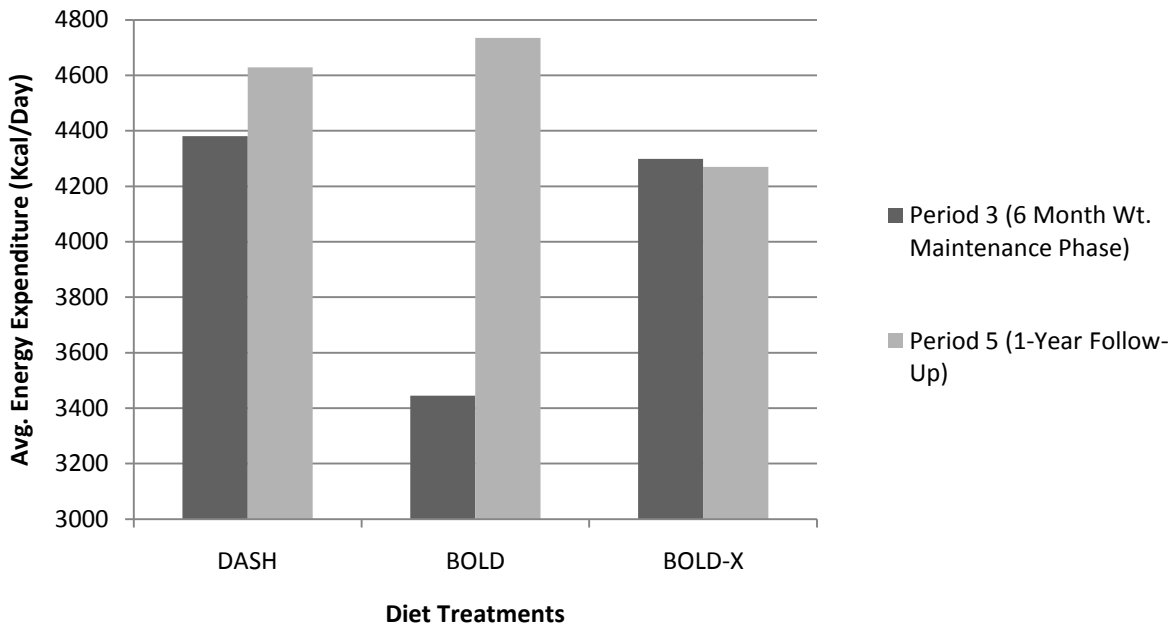
Figure 10. Sodium Intake During Periods 3, 4, and 5 for Each Diet Treatment*



*Values were log transformed.

Figure 10 represents sodium intake, which exhibited significant period by treatment effects. The sodium intake of the DASH diet during period 4 was significantly lower than that of BOLD 3 ($p=0.0116$), BOLD X 3 ($p=0.0127$), and DASH 3 ($p=0.0346$), but was significantly higher than that of BOLD 4 (0.0041). BOLD 4 was also significantly lower than BOLD X 5 ($p=0.0303$).

Figure 11. Three-day Avg. Energy Expenditure for Each Diet Treatment (Periods 3 & 5)



The results of the mean energy expenditure for the three diet treatments at the weight maintenance (6 month) and follow-up (1 year) periods are shown in Figure 11. Out of the 18 participants, 17 completed the 3-day physical activity diary. 4 out of 17 participants did not complete the form at the wt. maintenance period and 3 out of 17 participants did not complete the form at the one-year follow-up period. Thus, these values were not included in the total mean for each diet group. The DASH diet treatment group experienced a slight increase in energy expenditure from the 6 month phase (4380.3 kcals/day) to the one-year follow-up time point (4628.2 kcals/day). The BOLD diet treatment experienced a large increase in physical activity from the 6 month phase (3445.2 kcals/day) to the one-year follow-up time point (4734.6 kcals/day). However, the BOLD-X diet treatment experienced the opposite effect and had a lower energy expenditure at the one-year follow-up (4270.1 kcals/day) than at the 6 month phase (4298.8 kcals/day).

VI. DISCUSSION

In this study, it was expected that individuals who consumed diets that include lean beef as the primary source of protein with average (18% BOLD) or moderate-high (28% BOLD-X) total protein intake would maintain or improve their weight loss at 12 months compared to individuals who consumed a modified-DASH diet. It was also expected that individuals who consumed diets that included lean beef as the primary source of protein with average (18% BOLD) or moderate-high (28% BOLD-X) total protein intake would show similar or greater reductions in CVD risk as determined by reductions in select lipids and lipoproteins, and BP at 12 months compared to individuals who consumed a modified-DASH diet.

However, the results of the follow up study found that the BOLD diet group maintained their weight loss more successfully than the DASH and BOLD-X diet groups. The BOLD diet also had the least increase in TG and SBP, and the greatest increase in HDL-C levels from period 3 to period 5 compared to the BOLD-X and DASH diet groups. In terms of LDL-C levels, the BOLD and BOLD-X diet groups had similar increases from period 3 to period 5 (+20.875 mg/dL and +19.597 mg/dL, respectively), whereas the DASH diet had the greatest increase in LDL-C (+28.5 mg/dL). The DASH diet also had the lowest sodium intake (2350.4 mg) compared to the BOLD and BOLD-X diet groups (2820.0 mg and 3078.2 mg, respectively) at period 5.

Further research is needed to determine the specific factors that predict weight regain in individuals and a continuation of the BOLD-X one-year follow-up study should be performed in order to include a larger population size and achieve more predictive results.

A. OBSERVED CHANGES IN BLOOD LIPIDS

The three diets had variable effects on blood lipid and lipoprotein profiles of the participants in the study. LDL-C was significantly higher at period 5 than at periods 2 ($p=0.006$)

and 3 ($p=0.0006$). This rise in LDL-C at period 5 was consistent for all three diet treatments. Because of the vast amount of scientific research specifying that higher SFA intake correlates with higher LDL-C levels, this suggests that the participants in each diet treatment should have consumed greater amounts of SFA at period 5 than at period 3. The results of this study were not consistent with these findings because, according to the 3-day food diaries that were reported at these periods, SFA intake was significantly lower at period 5 than at period 3 ($p=0.0015$). Total cholesterol levels also significantly increased from period 3 to periods 4 ($p=0.0016$) and 5 ($p<0.0001$) for all participants in the study.

HDL-C levels generally increased, for each of the diet treatment, from period 3 to period 5. HDL-C levels were significantly higher at period 5 than at periods 2 and 3 ($p=<0.0001$). There may be a few explanations as to why HDL-C levels were lowest at the weight maintenance phase, and then gradually increased through periods 3, 4, and 5. Weight loss has been shown to increase HDL-C levels. In a meta-analysis of over 70 studies by Datillo and Kris-Etherton, a correlation was found for every 1 kg decrease in body weight, HDL-C decreased by 0.09 mmol/L in individual's actively losing weight and 0.007 mmol/L increase for individuals at a stabilized, reduced weight (Datillo & Kris-Etherton, 1992). These results are consistent with the data collected in this study because HDL-C levels were significantly lower during the weight maintenance and weight loss phase than during the one-year follow-up.

Another reason for the decrease in HDL-C levels is the macronutrient profile of the diets of the participants. In the current study, total fat intake significantly increased from period 3 to period 4 and then decreased from period 4 to period 5. In another study by Pelkham et al., a lower fat diet was shown to decrease HDL-C levels. The participants consumed either a low fat diet (17% of energy) or a moderate fat diet (33% of energy) during a six-week weight loss

period, and then followed by a four-week weight maintenance period. During the weight loss period, the low fat diet group experienced a 12% decrease in HDL whereas the moderate fat diet group did not experience a significant change in HDL. During the weight maintenance period, neither diet group experienced any significant changes in HDL. However, HDL levels at the end of the weight maintenance period were lower than at baseline for the low fat group (Pelkham et al., 2004). These studies indicate that a combination of weight loss and low fat diets are associated with a decrease in HDL-C levels. These results are similar to the results of this study, because total fat intake significantly increased from period 3 to period 4 ($p=0.0056$) and period 5 ($p=0.0272$). Thus, as participants consumed higher amounts of total fat in their diet, HDL-C levels also significantly increased.

Another interesting finding was a significant relationship between HDL-C and age for each diet treatment ($p<0.001$). A linear relationship ($HDL = 16.673 + 0.4656*age$) showed a steady increase in HDL-C as age increased. In terms of TG levels, there was no significant period by diet treatment differences. However, for all three diet treatments, there was a slight observed increase in TG from periods 3 to 5. Also, a significant relationship was observed between TG levels and age for each diet treatment ($p<0.001$). These results showed a linear decrease ($Tg_{log} = 2.4491 - 0.0052*age$) in TG levels as the age of participants increased.

These findings are consistent with a one year weight loss study of men ages 35-59 years. The results of the study found that participants, who lost an average of 4-7 kg, had also experienced a significant increase in HDL-C and a significant decrease in TG (Wood et al., 1988). In another cross-sectional study of elderly men and women, HDL-C increased with age in men, but not with women (Ferrara et al., 1997).

B. OBSERVED INCREASE IN SBP

For all participants in the study, there was a significant decrease from period 1 to period 3 ($p=0.0073$) and a significant steady increase from period 3 to period 5 ($p=0.005$). Both the BOLD and BOLD-X treatment groups had higher SBP levels at period 5 than when the study began at period 1. This was not the same for the DASH diet group, which experienced lower SBP at period 5 than at period 1.

Sodium intake has been positively associated with increased SBP (Sacks et al., 2009, Law et al., 1991). The National Institutes of Health reports that one of the primary lifestyle changes in reducing blood pressure and hypertension is decreasing sodium intake. In fact, reducing dietary sodium intake to 2.4 g of sodium or 6 g of sodium chloride can reduce SBP approximately 2-8 mm Hg (Chobavian et al., 2003). These statistics are consistent with the results of the present BOLD-X one-year follow-up study because there was a significant increase in SBP ($p=0.005$) as well as a significant increase in sodium intake ($p=0.0092$) for the entire population from period 3 to period 5. Therefore, results of this study are consistent in that SBP increases as sodium intake increases.

This indicates that the weight loss intervention, regardless of diet treatment, was successful at lowering SBP. At period 5, however, the BOLD and BOLD-X treatments seemed to have even higher SBP than at the entrance into the study. The DASH diet group still experienced some increase in SBP between periods 3 and 5, but not as to the extent of their SBP at the start of the study. Therefore, the DASH diet group was more successful at maintaining baseline SBP levels than the BOLD and BOLD-X diet groups.

C. OBSERVED CHANGES IN PHYSICAL ACTIVITY

One of the most interesting findings from the follow up study was the observed changes in physical activity. For the DASH and BOLD groups, participants reported greater physical

activity levels at the one-year follow-up than at period 3. The BOLD-X group did experience a slight decrease in physical activity at period 5, but this decrease was minimal. Regular physical activity helps individuals lose or maintain one's weight status. And according to many studies mentioned above, physical activity may be one of the most important factors in maintaining one's weight loss over a long period of time. The results of this study conflict with this notion because even though self-reported physical activity levels increased at period 5, the participant's mean weight status was greater at period 5 than at period 3.

There may be a few explanations for this discrepancy. Physical activity measurements were based on self-reported data from each participant. The participants may have inaccurately under- or over-reported physical activity levels in three different ways: type, intensity, and duration of the activity. Any of these factors could have been misreported by a participant, thus representing incorrect data.

VII. CONCLUSIONS

The expected outcome of the BOLD-X one-year follow-up study was that individuals who consumed diets that included lean beef as the primary source of protein (the BOLD and BOLD-X diets) would maintain or improve their weight loss at 12 months compared to a modified-DASH diet. Another expected outcome was that BOLD and BOLD-X diets would show greater reductions in CVD risk in terms of reduced lipid levels, lipoproteins, and blood pressure, compared to the modified-DASH diet at 12 months. However, the BOLD diet group maintained their weight loss more successfully than the DASH and BOLD-X diet groups. The BOLD diet group also had the least increase in TG and SBP, and the greatest increase in HDL-C levels from period 3 to period 5 compared to the BOLD-X and DASH diet groups.

LDL-C levels were significantly higher for each diet treatment at the one-year follow-up compared to the weight loss phase. Interestingly however, SFA intake was significantly lower at the one-year follow-up compared to the weight loss phase. Another interesting finding was that HDL-C levels were significantly higher at the one-year follow-up than during the weight loss phase. When adjusted for age, a linear relationship showed a steady increase in HDL-C as age increased.

SBP levels were significantly higher at the one-year follow-up than at the weight loss phase. Sodium intake was also significantly higher at the one-year follow-up than at the weight loss phase. In terms of physical activity levels, the DASH and BOLD groups reported greater physical activity levels at the one-year follow-up than at the weight loss phase, the BOLD-X group experienced a minimal decrease at the one-year follow-up.

Even though this study was designed very carefully, there are a few ways in which future studies could improve and modify the methods of the research. The study could include a greater

number of participants in order to obtain results that were more representative of the population at large. Self-reported 3-day food and physical activity diaries can facilitate collective better diet and physical activity data. Food diaries can be challenging for individuals to keep and maintain because of lack of knowledge about portion sizes, food preparation techniques of everything they ate, as well as the burden of recording all foods consumed. It can be difficult for researchers to analyze the data because of these factors, which results in incomplete and inaccurate dietary data. Physical activity diaries are also difficult for participants to accurately report the type, intensity, and duration of the activity. Future studies could incorporate more accurate methods of recording food intake and physical activity, such as food frequency questionnaires, 24-hour recalls, and international physical activity questionnaires (IPAQ), as well as using new technologies to collect diet and physical activity data.

Furthermore, future research is needed in order to determine the comprehensive effects of animal protein on weight status and cardiovascular disease risk factors. The current study showed that the BOLD diet that included a moderate amount of lean beef beneficially effected weight status and CVD risk factors compared with experimental diets that were very low or very high in lean beef. Therefore, nutrition education is needed in order to teach the general public about planning a heart healthy diet that incorporates lean beef.

VIII. REFERENCES

- American Dietetic Association (ADA). Adult weight management evidence based nutrition practice guideline. American Dietetic Association (ADA); 2006 May. Various p.
- Appel LJ, Sacks FM, Carey VJ, et al. OmniHeart Collaborative Research Group. Effects of Protein, Monounsaturated Fat, and Carbohydrate Intake on Blood Pressure and Serum Lipids: Results of the OmniHeart Randomized Trial. *JAMA*. 294 (19):2455-2464, 2005.
- Astrup A, Rössner S. Lessons from obesity management programmes: greater initial weight loss improves long-term maintenance. *Obes Rev*. 1: 17–19, 2000.
- Bouchard, et al. A method to assess energy expenditure in children and adults. *Am J Clin Nutr*. 37; 461-467, 1983.
- Chobanian AV, et al. The seventh report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure: the JNC 7 report. *JAMA*, 289(19):2560-72, 2003.
- Corral PD, et al. Dietary adherence during weight loss predicts weight regain. *Obesity*, 2010 Dec 16.
- Dansinger ML, Gleason JA, Griffith JL, et al. Comparison of the Atkins, Ornish, Weight Watchers, and Zone Diets for Weight Loss and Heart Disease Risk Reduction: A Randomized Trial. *JAMA*. 293:43-53, 2005.
- Delbridge EA, Prendergast LA, Pritchard JE, Proietto J. One year weight maintenance after significant weight loss in healthy overweight and obese subjects: does diet composition matter? *Am J Clin Nutr*. 90(5) 1203-1214, 2009.
- Diabetes Prevention Program Research Group. Reduction in the incidence of type 2 diabetes with lifestyle intervention or metformin. *N Engl J Med*. 346:393–403, 2002.

Datillo AM, Kris-Etherton PM. Effects of weight reduction on blood lipids and lipoproteins: a meta-analysis. *Am J Clin Nutr.* 56:320-8, 1992.

Ferrara A, Barrett-Connor E, Shan J. Total, LDL, and HDL cholesterol decrease with age in older men and women. *Circulation,* 96:37-43, 1997.

Flegal KM, Carroll MD, Ogden CL, & Curtin LR. Prevalence and trends in obesity among U.S. adults, 1999-2008. *JAMA* 303(3):235-241, 13 Jan 2010.

Fogelholm M, Kukkonen-Harjula K, Nenonen A, Pasanen M. Effects of walking training on weight maintenance after a very-low-density diet in premenopausal women. *Arch Int Med,* 160:2177-2184, 2000.

Franz MJ, VanWormer JJ, Crain AL, et al. Weight-Loss Outcomes: A Systematic Review and Meta-Analysis of Weight-Loss Clinical Trials with a Minimum 1-Year Follow-Up. *J Am Diet Assoc.* 107:1755-1767, 2007.

Gardner CD, Kiazand A, Alhassan S, et al. Comparison of the Atkins, Zone, Ornish, and LEARN Diets for Change in Weight and Related Risk Factors Among Overweight Premenopausal Women: The A TO Z Weight Loss Study: A Randomized Trial. *JAMA.* 297:969-977, 2007.

Garrison RJ, Castelli WP. Weight and thirty-year mortality of men in the Framingham study. *Annals of Int Med.* 103(6):1006-1009, 1985.

Haskell WL, et al. Physical activity and public health: updated recommendation for adults from the American College of Sports Medicine and the American Heart Association. *Circulation,* 116(9):1081, 2007.

Jacobs DR, Mebane IL, Bangdiwala SI, Criqui MH, Tyroler HA. High density lipoprotein cholesterol as a predictor for cardiovascular disease mortality in men and women the

follow-up study of the lipid research clinics prevalence study. *Am J Epid*, 131(1); 32-47, 1990.

Katcher HI, Hill AM, Lanford J, Yoo JS, & Kris-Etherton PM. Lifestyle approaches and dietary strategies to lower LDL-cholesterol and triglycerides and raise HDL-cholesterol. *Endocrinology and metabolism clinics of North America*, 38(1); 45 -78, 2009.

Klem ML, Wing RR, McGuire MT, et al. A descriptive study of individuals at long-term maintenance of substantial weight loss. *Am J Clin Nutr*. 66(2) 239-246, 1997.

Kraschnewski JL, Boan J, Esposito J, et al. Long-term weight loss management in the United States. *Inter J of Obesity*. 34;1644-1654, 2010.

Kromhout D, et al. Dietary saturated and transfatty acids and cholesterol and 25-year mortality from coronary heart disease: the seven countries study. *Prev Med*, 24(3);308-315, 1995.

Law MR, Frost CD, Wald NJ. By how much does dietary salt reduction lower blood pressure? Analysis of observational data among populations. *BMJ*. 302(6780):815-818, 1991.

Layman DK, Evans EM, Erickson D, et al. A moderate-protein diet produces sustained weight loss and long-term changes in body composition and blood lipids in obese adults. *J Nutr*. 139(3):514-521, 2009.

McGuire MT, Wing RR, Klem ML, Lang W, Hill JO. What predicts weight gain in a group of successful weight losers? *J Cons Clin Psych*. 67(2) 177-185, 1999.

Obarzanek E, et al. Effects of blood lipids of a blood pressure-lowering diet: the Dietary Approaches to Stop Hypertension (DASH) trial. *Am J Clin Nutr*. 74(1);80-89, 2001.

Pelkman CL, Fishell VK, Maddox DH, et al. Effects of moderate-fat (from monounsaturated fat) and low-fat weight-loss diets on the serum lipid profile in overweight and obese men and women. *Am J Clin Nutr*. 79: 204-212, 2004.

Pelkman CL, Fishell VK, Maddox DH, Pearson TA, Mauger DT, Kris-Etherton PM. Effects of moderate-fat (from monounsaturated fat) and low-fat weight-loss diets on the serum lipid profile in overweight and obese men and women. *Am J Clin Nutr.* 79;204-12, 2004.

Pvalov KN, Drey X, Steffee WP. Exercise as an adjunct to weight loss and maintenance in moderately obese subjects. *Am J Clin Nutr.* 49;1115S-23S, 1989.

Sacks FM, Bray GA, Carey VJ, et al. Comparison of Weight Loss Diets with Different Compositions of Fat, Protein, and Carbohydrates. *N Engl J Med.* 360:859-873, 2009.

Seagle HM, Strain GW, Makris A, Reeves RS, American Dietetic Association. Position of the American Dietetic Association: weight management. *J of Am Dietetic Assoc.* 109(2) 330-346, 2009.

Turk MW, Yang K, Hravnak M, Sereika SM, Ewing LJ, Borke LE. Randomized clinical trials of weight loss maintenance: a review. *J Cardiovascular Nurs.* 24(1) 58-80, 2009.

Wadden TA, Sternberg JA, Letizia KA, Stunkard AJ, & Foster GD. Treatment of obesity by very low calorie diet, behavior therapy, and their combination: a five year perspective. *International Journal of Obesity,* 13; 39-46, 1989.

Wing RR, Phelan S. Long Term Weight Loss Maintenance. *Am J Clin Nutr.* 82 (1 Suppl):222s-225s, 2005.

Wood PD, et al. Changes in plasma lipids and lipoproteins in overweight men during weight loss through dieting as compared with exercise. *N Eng J Med.* 319(18);1173-9, 1988.

Academic Vita

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Education

Pennsylvania State University, University Park, PA
Schreyer's Honors College, University Park, PA
B.S. in Nutritional Sciences
Honors in Nutritional Sciences
Graduation Date: May 2011
Thesis Title: BOLD (Beef In An Optimal Lean Diet) Effects on Metabolic Syndrome (BOLD-X), PHASE 2: A 12 Month Follow-Up Study
Thesis Supervisor: Penny Kris-Etherton, Distinguished Professor of Nutritional Sciences

International Education:

International Program in Nutrition Summer 2010
The Pantheon Institute, Rome, Italy

- Completed nine credits in Historical, Cultural and Scientific Aspects of the Mediterranean Diet, Program Planning, and Italian Architecture and Urban Form
- Attended lectures at the Food and Agriculture Organization in Rome

Experience

Undergraduate Research Assistant, Kris-Etherton Research Laboratory, Penn State University

- Jan. 2010 - May 2011, Organized and conducted a follow-up to the BOLD-X study at Penn State under the supervision of Penny Kris-Etherton. The BOLD-X study assessed the effects of consumption of lean red beef on cardiovascular disease and overall health. The purpose of the follow-up was to assess which control group maintained their weight and lipid status. Research was used as data for completion of an undergraduate honors thesis.

Tutor for Adult English as a Second Language Learner, CIU #10 Development Center for Adults, Center County, PA

- Jan-May 2010, Prepared lesson plans each week for a Korean-speaking ESL learner. Built confidence in the student's ability to speak, read, and write the English language.

Honors and Awards

Dean's List, All semesters
Pittsburgh Tribune-Review Outstanding Young Citizen Award, 2007

Professional Memberships

American Dietetic Association

Pennsylvania Dietetic Association

Scholarships and Grants

Schreyer Ambassador Travel Grant for study in Italy, 2010

Activities

Peer Educator, HealthWorks

- Promoting health and wellness initiatives on the Penn State Campus

Secretary, Student Nutrition Association, Sept. 2010-May 2011

Member, Penn State Dance Marathon (THON), Rules and Regulations Committee

Certifications

ServSafe Certification, Nov. 2010-Nov. 2015