

Paleolithic Diet is Associated With Unfavorable Changes to Blood Lipids in Healthy
Subjects

Honors Research Thesis

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The Ohio State University, May 2013

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Abstract

Background: The Paleolithic (Paleo) diet is one modeled after the perceived food consumption of early human ancestors of the Paleolithic Era, consisting of mainly meat, fish, fruit, vegetables, eggs, and nuts. The purpose of this study was to examine the effects of a Paleo diet on blood lipids, including high-density lipoprotein (HDL), low-density lipoprotein (LDL), non-HDL cholesterol, triglycerides (TG), total cholesterol (TC), and the ratio between TC and HDL (TC/HDL) in a healthy population. **Methods:** Subjects of both genders (23 males, 20 females) with no history of diabetes, heart disease, dyslipidemia, or other metabolic disease were asked to eat an *ad libitum* Paleo diet for 10 weeks. Throughout the intervention, subjects participated in a CrossFit-based, high-intensity circuit training exercise program. Prior to the intervention, body weight, body fat percentage (BF%), maximal oxygen consumption (VO₂max), TC, TG, HDL, and LDL were measured. These measurements were repeated following 10 weeks of a Paleo diet. **Results:** As a whole, there was a significant increase in non-HDL (107.1 ± 6.0 mg/dL to 120.2 ± 6.5 mg/dL; $P < 0.01$), LDL (93.1 ± 5.4 mg/dL to 105.6 ± 6.1 mg/dL; $P < 0.01$), TC/HDL (3.0 ± 0.2 to 3.3 ± 0.2 ; $P < 0.05$), and TC (168.8 ± 5.4 mg/dL to 178.9 ± 6.6 mg/dL; $P < 0.05$) in healthy subjects following a Paleo diet. When stratified into groups based on initial blood lipid levels, deleterious changes were found in those with optimal HDL (82.1 ± 3.2 mg/dL to 68.6 ± 4.8 mg/dL; $P < 0.05$), non-HDL (86.6 ± 3.9 mg/dL to 101.4 ± 4.8 mg/dL; $P < 0.01$), TC (157.2 ± 0.7 to 168.2 ± 0.9 mg/dL; $P < 0.05$), TC/HDL (2.5 ± 0.1 to 2.7 ± 0.1 ; $P < 0.05$), and LDL (69.1 ± 3.1 mg/dL to 83.5 ± 4.1 mg/dL; $P < 0.01$), whereas those within sub-optimal stratifications showed no significant changes. Subjects also decreased body weight (177.6 ± 5.8 lbs to 170.6 ± 5.3 lbs; $P <$

0.001) and BF% ($24.3 \pm 1.2\%$ to $20.7 \pm 1.2\%$; $P < 0.05$), while increasing VO_2max (3.18 ± 0.14 L/min to 3.46 ± 0.15 L/min; $P < 0.001$). **Conclusions:** Our results demonstrate that an *ad libitum* Paleo diet intervention is associated with deleterious changes to blood lipids in healthy subjects, despite concurrent improvements in body composition and cardiorespiratory fitness. Future research should focus on determining recommendations that embrace the positive aspects of the Paleo diet, while minimizing any deleterious impact on blood lipids in a healthy population.

Background

The Paleolithic (Paleo) diet is modeled after the perceived food consumption of early human ancestors of the Paleolithic Era. Since the origin of this dietary concept, which dates back to the late 1970s [1], numerous books and articles have claimed that increasing rates of “diseases of civilization”, including cardiovascular disease, metabolic syndrome, and type 2 diabetes, among others [2, 3], can be largely attributed to the deviation of the modern diet from the diet consumed during the majority of the evolutionary history of humans [1, 3, 4]. Many prominent CrossFit athletes advocate adherence to the Paleo diet, which has contributed to a recent boost in the popularity of the diet. The common belief is that the Paleolithic Era pre-dated the advent of the cultivation and processing of plants and domestication of animals [2], though this idea has recently been challenged [5, 6]. Researchers have attempted to reconstruct the diet of the early human based on archaeological evidence and studies involving modern hunter-gatherer societies, leading to a proposed early-human diet comprised of wild animal-

source foods and uncultivated plant-source foods, devoid of dairy, legumes, and cereal grains [2].

The body of existing literature pertaining to the Paleo diet is characterized by inconsistency. Because the diet is defined by the avoidance of particular food sources rather than a specific macronutrient distribution, there is a large degree of variation in the macronutrient composition of various Paleo diet interventions. One of the earliest peer-reviewed papers on the Paleolithic diet suggests that the diet of the late Paleolithic Era was roughly 33% protein, 46% carbohydrate, and 21% fat [3]. Recent interventions, which place more emphasis on food source than macronutrient distribution, have not replicated these suggested intakes. In two studies in which food rations were controlled, one diet derived 31.9% of its caloric content from fat and 36.9% from carbohydrate [2], while the other derived 16% of calories from fat and 57% from carbohydrate [7].

Some degree of variation in macronutrient intake is to be expected in Paleo studies featuring free-eating subjects, but even the dietary guidelines enforced in such studies have been inconsistent. One study imposed limits on intakes of eggs, nuts, potatoes, and oils [8], one limited dried fruit intake along with the aforementioned foods [9], and another added fatty meats, honey, cured meats, and salted seafood to the list of limited foods, without any mention of egg intake [10]. While previous interventions have utilized varying macronutrient distributions and somewhat inconsistent guidelines, the studies uniformly reinforce the basic characteristics of a Paleolithic diet: A diet based on lean meat, fish, eggs, nuts, fruit, and vegetables, and devoid of cereal grains, dairy, legumes, and processed sugars.

A number of studies have shown Paleo diets to have a positive influence on health, including improved maintenance of blood glucose [2, 8, 9], decreased body mass [8, 9], and improved blood lipids [2, 9]. The studies reporting these improvements have used subjects that had an average body mass index (BMI) greater than 27 kg/m² [2, 8, 9] and were either sedentary [2], had diabetes [9], or had heart disease [8]. Furthermore, these studies have compared the Paleo diet to another diet containing greater total calories [8, 9], carbohydrates [2, 8, 9], and saturated fats [2, 8, 9], along with a higher carbohydrate to protein ratio [2, 8, 9]. It is important to note that lower carbohydrate to protein ratios have been shown to be independently effective in improving glycemic control [11-13] and blood lipid profile [14]. For these reasons, it is hard to determine if the results of these studies are applicable to active, healthy populations, and whether or not the effectiveness of the Paleo diet in these studies can simply be attributed to lower intakes of calories, carbohydrates, saturated fats, and a more ideal carbohydrate to protein ratio rather than the avoidance of certain food sources. While much research has been done on the Paleo diet and the effects it has on overweight/obese and disease populations, there is a lack of research pertaining to the application of the Paleo diet in healthy, active populations.

Authors of previous Paleo interventions have embraced the potential of the *ad libitum* Paleo diet. Authors of a 2006 study wrote that contemporary groups of hunter-gatherers “Stay lean and apparently reap health benefits similar to those induced by food restriction despite *ad libitum* availability of food.”[7] Another group of researchers found that a Paleo diet intervention yielded an improvement in glucose tolerance that appeared to be independent of energy intake and macronutrient distribution, prompting them to

conclude that "...avoiding Western foods is more important than counting calories, fat, carbohydrate or protein." [8] Although previous trials with free-eating subjects have recognized food sources like meat, eggs, nuts, potatoes, and certain oils as part of the Paleolithic diet, such trials have imposed guidelines that restrict the consumption of one or more of the aforementioned foods [8-10].

A truly *ad libitum* application of the Paleo diet, without restrictions on intakes of fatty meat, eggs, nuts, and oils, could potentially be conducive to high intakes of total fat, saturated fat, and cholesterol. This style of dieting, without regard for daily intakes of total fat, saturated fat, or cholesterol, neglects guidelines set by the American Heart Association (AHA) regarding a heart-healthy diet aimed at lowering disease risk and supporting a healthy blood lipid profile [15]. For this reason, the current study aims to investigate the effects of a practical, *ad libitum* application of the Paleo diet on blood lipids in a sample of healthy, active subjects. To accomplish this objective, we recruited healthy, active adults and measured body composition and blood lipid markers before and after a 10-week Paleo diet. We hypothesized that a Paleo dietary intervention made without specific recommendations regarding macronutrient intake would be associated with an increase in low density lipoprotein (LDL), non-high density lipoprotein (n-HDL), and total cholesterol (TC), but no change in high density lipoprotein (HDL). Furthermore, we hypothesized that this diet would be associated with a decrease in body weight and body fat percentage.

Methods

This study investigated the effect of a 10-week Paleo diet on body composition and blood lipids in healthy adults. Body composition using air displacement plethysmography and fasted state blood draws were performed in all subjects in the morning (7:30 a.m. to 11:30 a.m.) over a five-day period preceding the onset of the intervention. Measurements were obtained following an overnight fast, and subjects refrained from exercise, alcohol, and caffeine for the 24 hours prior to testing. A total of 43 subjects completed the dietary intervention and returned for the post-training assessment of changes in the dependent variables of body composition and blood lipids. All returning subjects were assessed at the same time of day as the pre-training measures over a five-day period following the completion of the intervention. During the dietary intervention, all subjects regularly participated in a CrossFit-based, high-intensity circuit training program.

Subjects

A total of 43 (23 males, 20 females) subjects fully completed the dietary intervention and returned for follow-up testing. All of the subjects provided written informed consent and all study methods and protocols were approved in advance by the Institutional Review Board at The Ohio State University.

Procedures

Dietary Intervention

A Paleolithic diet, as first described by Eaton and Konner, was implemented for all study participants [16]. Subjects were advised to increase their consumption of lean meat, fish,

eggs, nuts, fruit, and vegetables and were instructed to strictly avoid all grains, dairy products, and legumes. All modern, processed foods including any form of processed sugar, soft drinks, and coffees were also excluded from the diets of the subjects. No specific macronutrient recommendations were made, as the study design wanted to closely mimic a real world model that would incorporate food choices made by the average consumer. Intake of specific proportion of food categories (e.g. animal vs. plant foods) was also not given. Dietary recall logs were distributed to all subjects during the last week of the dietary intervention and subjects were asked to record all food and liquids over three days, including a weekend day.

Blood Lipid Analysis

For all subjects, a finger-stick blood sample from a sterilized site was collected into a lithium heparin coated capillary tube by one trained investigator. Samples were immediately transferred into a point of care desktop lipid analyzer (Cholestech LDX, San Diego, CA). This method has previously been shown to be within standards of accuracy and precision set forth by the National Cholesterol Education Program (NCEP) [17]. High-density lipoprotein (HDL), total cholesterol (TC), and triglycerides (TG) were measured directly, while low density lipoprotein (LDL) was estimated using the Friedewald equation [18].

Body Composition

Percentage body fat was calculated using the Bod Pod air-displacement plethysmography device (Life Measurements Instruments, Concord, CA), which is shown to be an accurate

method for assessing body composition in adults [19]. Prior to measurement, the system was calibrated for volume using a cylinder of a known volume (50.312 L) and for mass using two 10 kg weights. Fasting-state body weight was measured to the nearest 0.1 kg and subjects entered the Bod Pod chamber wearing only a tight fitting swimsuit and swim cap. Body volume measurements were taken in duplicate and repeated if measures were not within 150 mL of each other [20]. Body density was calculated as mass/body volume and body fat percentage was calculated by using Siri's formula [21]. Body mass index (BMI) was calculated as kg body mass divided by height in meters squared.

Statistical Analyses

Changes in body composition and blood lipids were tested using a two-tailed, paired t-test. These values were tested as an entire group, and also in subsets that were stratified by initial values of blood lipids for each subject. Participants were stratified into subsets based on cutoffs (Table 2) published by the NCEP Adult Treatment Panel III (ATP III) [22]. Two-tailed, paired t-tests were then used to test differences between pre- and post-intervention values of stratified blood lipids. Data are reported as mean \pm SEM.

Statistical analysis was performed using STATA (version 11.1, College Station, TX).

Statistical significance was defined *a priori* as the critical α -level of $P < 0.05$.

Results

Characteristics of all the subjects who volunteered for the study are presented in Table 1.

A total of 43 subjects participated in the study (23 males, 20 females). The average age of study participants was 33.5 ± 0.3 yr and 31.2 ± 0.3 yr for men and women, respectively.

Following the dietary intervention, n-HDL (107.1 ± 6.0 mg/dL to 120.2 ± 6.5 mg/dL; $P < 0.01$), LDL (93.1 ± 5.4 mg/dL to 105.6 ± 6.1 mg/dL; $P < 0.01$), and TC (168.8 ± 5.4 mg/dL to 178.9 ± 6.6 mg/dL; $P < 0.05$) increased significantly from baseline in all participants (Figure 1). Further, TC/HDL increased to a statistically significant degree (3.0 ± 0.2 to 3.3 ± 0.2 ; $P < 0.05$). Body fat percentage decreased significantly ($24.3 \pm 1.2\%$ to $20.7 \pm 1.2\%$; $P < 0.05$) compared with baseline values (Figure 1), as did body weight (177.6 ± 5.8 lbs to 170.6 ± 5.3 lbs; $P < 0.01$). When stratified by initial blood lipid levels based on the ATP III, there was a significant decrease of HDL among subjects with the highest initial HDL (82.1 ± 3.2 mg/dL to 68.6 ± 4.8 mg/dL; $P < 0.05$; Figure 2) and significant increases of n-HDL among subjects with the lowest levels of n-HDL (86.6 ± 3.9 mg/dL to 101.4 ± 4.8 mg/dL; $P < 0.01$; Figure 3). Furthermore, significant increases of LDL (69.1 ± 3.1 mg/dL to 83.5 ± 4.1 mg/dL; $P < 0.01$; Figure 4), TC (157.2 ± 0.7 mg/dL to 168.2 ± 0.9 mg/dL; $P < 0.05$), and TC/HDL (2.5 ± 0.1 to 2.7 ± 0.1 ; $P < 0.05$; Figure 5) were observed among subjects with the most optimal initial levels of each respective outcome variable. Relative maximal oxygen consumption also improved during the study (39.8 ± 1.2 ml/kg/min to 44.9 ± 8.2 ml/kg/min; $P < 0.001$; Table 3), presumably resulting from the concurrent regular participation in a CrossFit-based, high-intensity circuit training program, along with a decrease in body weight. Despite the weight loss observed in study participants, absolute oxygen consumption also increased (3.18 ± 0.14 L/min to 3.46 ± 0.15 L/min; $P < 0.001$), which indicates an overall concurrent improvement of aerobic fitness.

Table 1. Subject characteristics

	Males (n=24)	Females (n=20)	Range
Age (years)	33.5 ± 0.3	31.2 ± 0.3	21.0 – 48.0
Height (in)	70.6 ± 0.1	64.8 ± 0.1	60.0 – 77.0
Weight (kg)	90.71 ± 0.25	68.02 ± 0.31	44.54 – 118.18
BMI (kg/m ²)	28.1 ± 0.1	25.1 ± 0.2	19.1 – 37.4
Body fat (%)	22.2 ± 0.3	26.6 ± 0.4	10.7 – 46.1
Lean mass (kg)	70.25 ± 0.37	49.00 ± 0.25	36.35 – 82.17
TC (mg/dL)	171.7 ± 8.7	164.7 ± 6.3	110.0 – 258.0
HDL (mg/dL)	50.6 ± 2.3	72.8 ± 3.8	35.0 – 100.0
LDL (mg/dL)	105.6 ± 7.8	76.8 ± 6.3	33.0 – 191.0
n-HDL (mg/dL)	118.0 ± 9.1	91.9 ± 6.7	40.0 – 204.0
TG (mg/dL)	72.6 ± 9.0	71.8 ± 6.9	45.0 – 226.0
TC/HDL	3.5 ± 0.2	2.3 ± 0.1	1.4 – 5.0

BMI = body mass index; in = inches; kg = kilograms; VO₂max = maximal oxygen consumption. All data are resting values and is presented as mean ± SEM.

Table 2. Blood lipid risk stratification

	HDL (mg/dL)	LDL (mg/dL)	n-HDL (mg/dL)	TC/HDL	
Low	<40	Optimal	<100	Low <130	Optimal <3.4
Normal	40-60	Near optimal	100-129	Normal 130-189	Above optimal ≥3.5
High	≥60	Borderline High	130-159 ≥160	High ≥190	

HDL = high density lipoprotein cholesterol; LDL = low density lipoprotein cholesterol; n-HDL = non-high density lipoprotein cholesterol; TC/HDL = ratio of total cholesterol to high density lipoprotein.

Table 3. Select changes observed after 10-week Paleo intervention

Changes observed after 10-Week Paleo Intervention						
	Relative VO ₂ max (mL/kg/min)	Absolute VO ₂ max (L/min)	Body Fat (%)	TC (mg/dL)	LDL (mg/dL)	n-HDL (mg/dL)
Pre- test	39.82 ± 7.72	3.18 ± 0.14	24.32 ± 7.63	168.8 ± 5.4	93.1 ± 5.4	107.1 ± 6.0
Post- test	44.90 ± 8.20**	3.46 ± 0.15**	20.65 ± 7.99**	178.9 ± 6.6*	105.6 ± 6.1**	120.2 ± 6.5**

VO₂max = maximal oxygen consumption; TC = total cholesterol; LDL = low density lipoprotein; n-HDL = non-high density lipoprotein. * P < 0.05, ** P < 0.01.

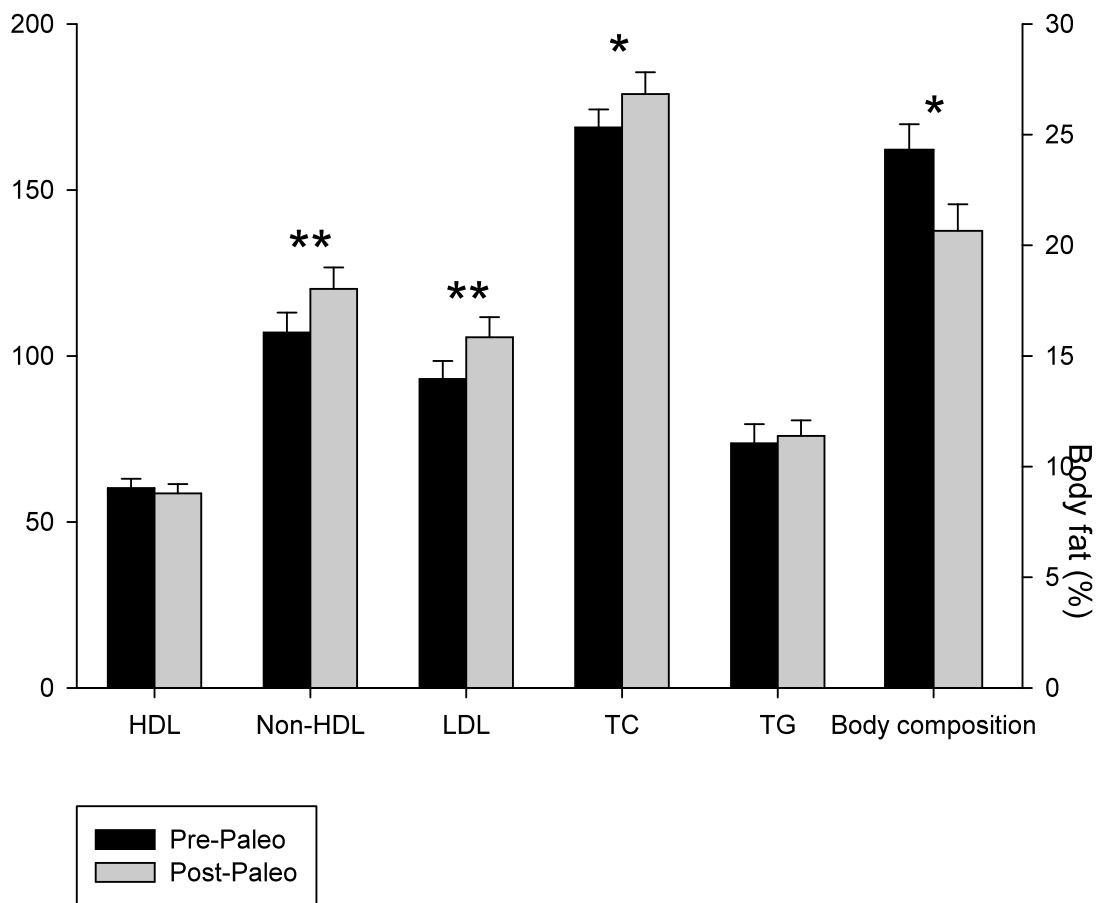


Figure 1. Blood lipids in healthy volunteer prior to and following a 10 week Paleolithic diet intervention. Non- high density lipoprotein (n-HDL), low density lipoprotein (LDL) and total cholesterol (TC) increased significantly from baseline, whereas no changes were observed with regard to high density lipoprotein (HDL) and triglycerides (TG). A significant decrease in body composition was observed compared to baseline. * $P < 0.05$; ** $P < 0.01$.

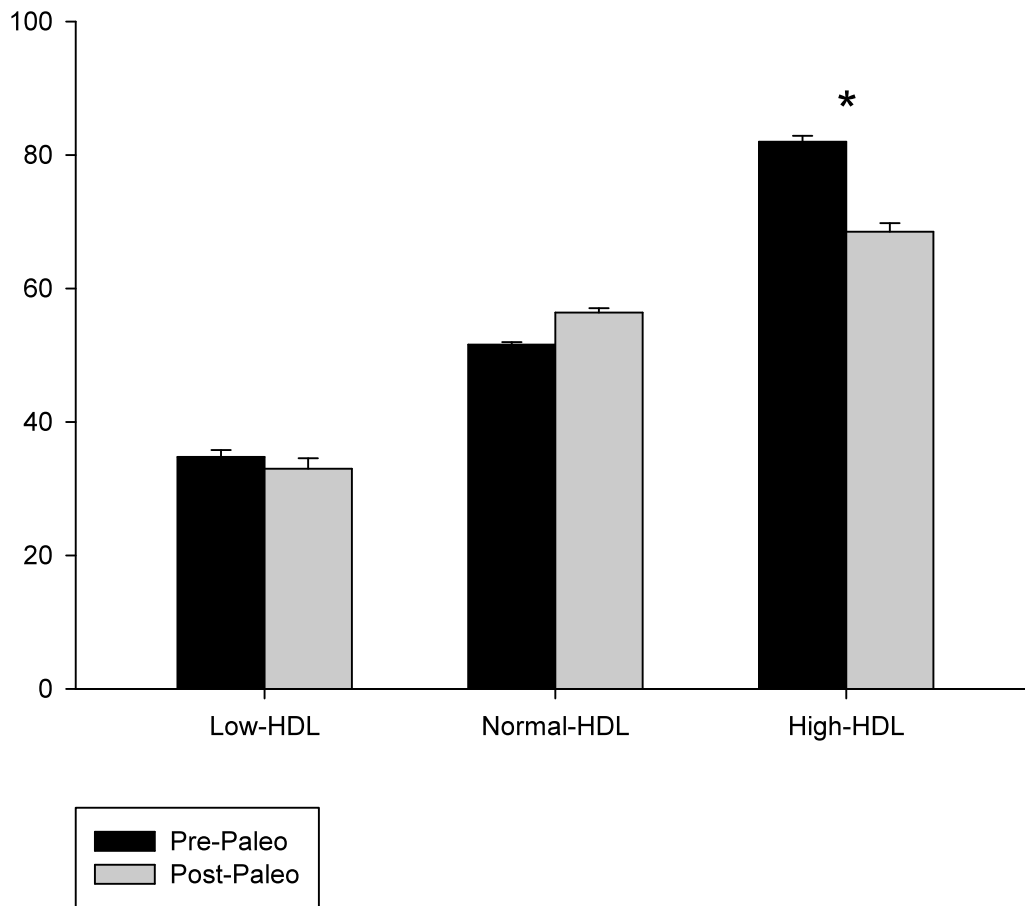


Figure 2. High density lipoprotein (HDL) levels in healthy volunteers before and following a 10 week Paleolithic dietary intervention. When stratified by initial HDL levels, only subjects who presented with “High-HDL” were observed to have a significant decrease in HDL following a Paleolithic diet. * $P < 0.05$.

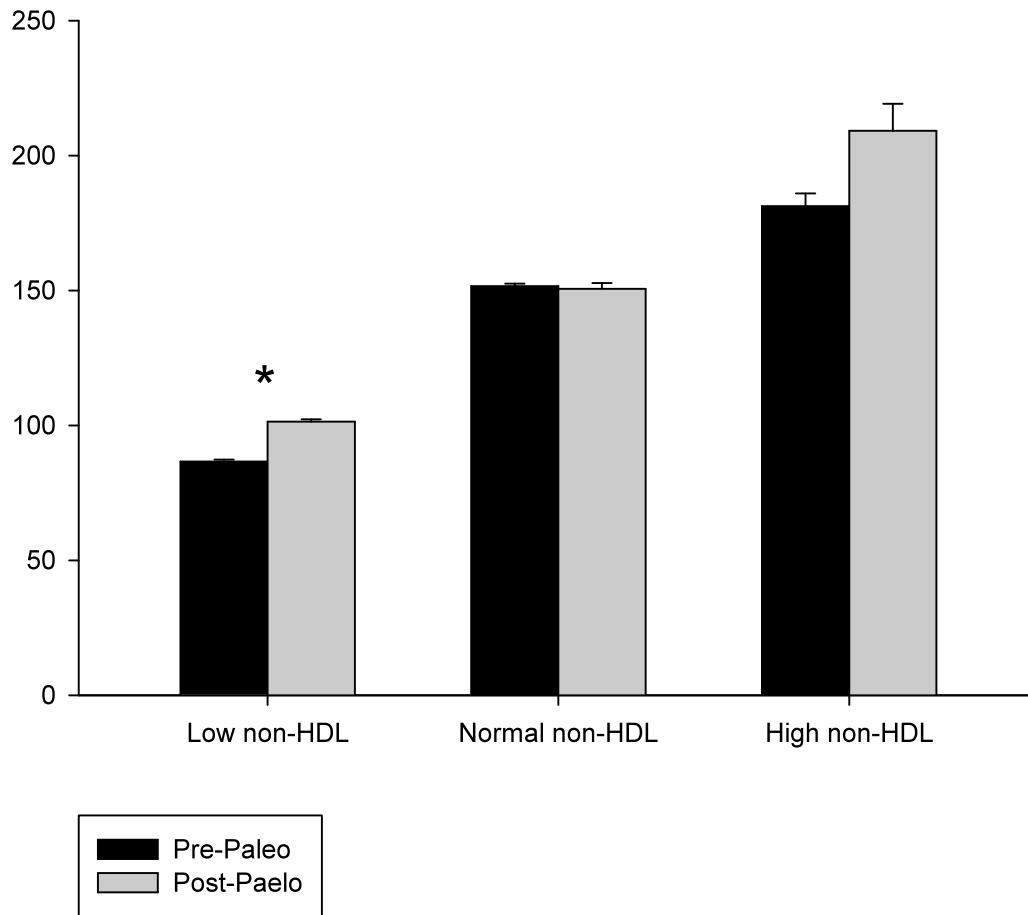


Figure 3. Non- high density lipoprotein (n-HDL) levels in healthy volunteers prior to and following a Paleolithic dietary intervention. When stratified by initial levels of n-HDL, only subjects with n-HDL considered to be “low” were measured to have a significant increase of n-HDL following 10 weeks of a Paleolithic diet. * $P < 0.05$.

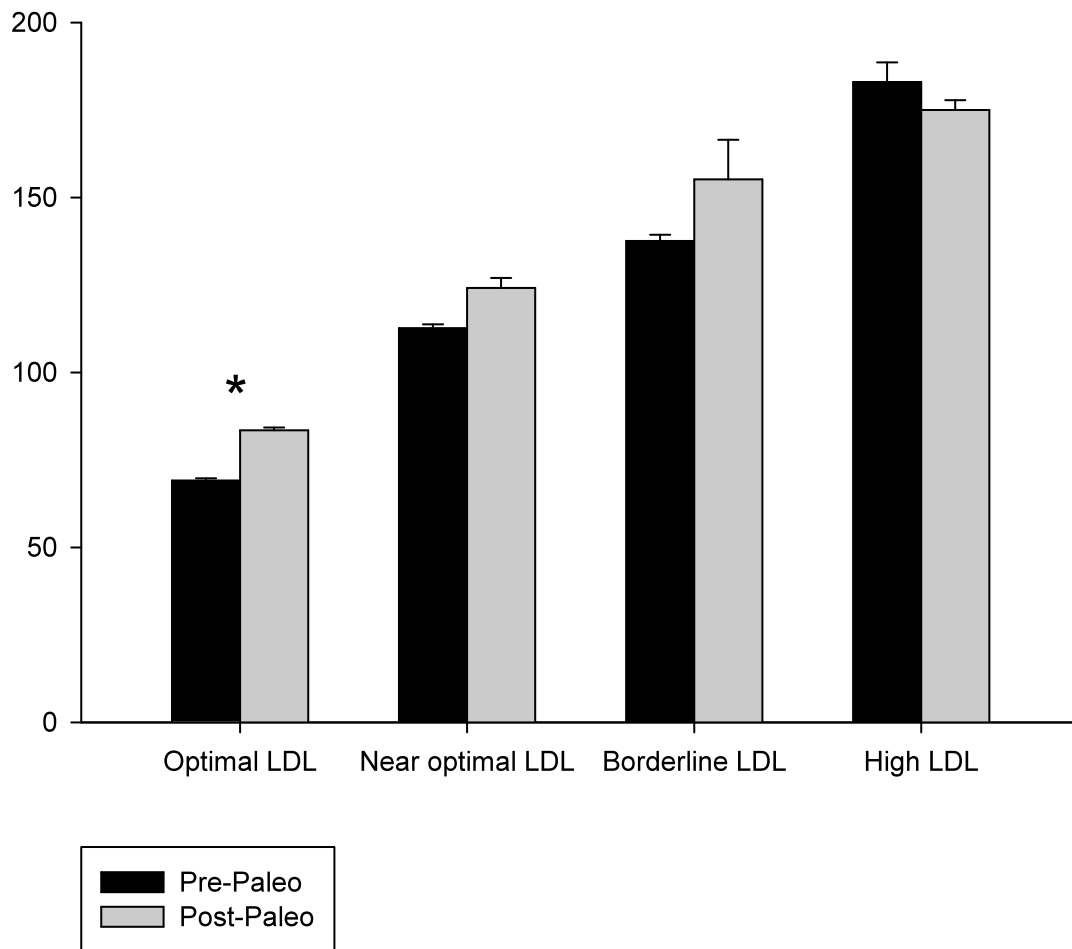


Figure 4. Low density lipoprotein (LDL) levels in healthy volunteers prior to and following a Paleolithic dietary intervention. When stratified by initial levels of LDL, only subjects with optimal LDL were measured to have a significant increase of LDL following 10 weeks of a Paleolithic diet. * $P < 0.05$.

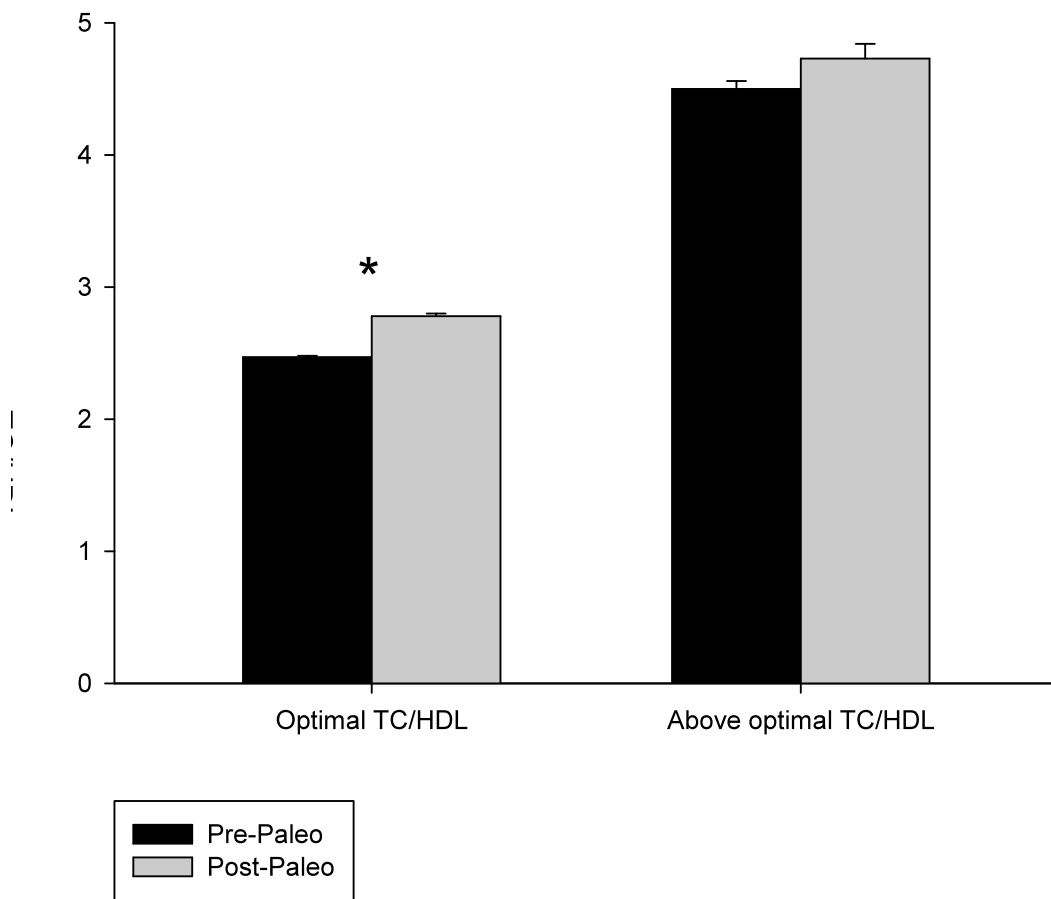


Figure 5. Total cholesterol to high density lipoprotein ratio (TC/HDL) in healthy volunteers prior to and following a Paleolithic dietary intervention. When stratified by initial levels of TC/HDL, only subjects with optimal TC/HDL were measured to have a significant increase of TC/HDL following 10 weeks of a Paleolithic diet. * $P < 0.05$.

Discussion

Our study featured a large number ($n = 43$) of healthy, active subjects that followed an *ad libitum* Paleo diet while participating in a CrossFit-based circuit training program. As hypothesized, we observed negative impacts on non-HDL, LDL, TC, and TC/HDL, no significant change in HDL, and a decrease in body weight and body fat percentage.

Furthermore, when stratified by pre-test blood lipid values, the statistically significant effects primarily came from deleterious changes in the most healthy subjects; subjects within optimal pre-test blood lipid stratifications saw statistically significant negative changes in HDL, non-HDL, LDL, TC, and TC/HDL. The Law of Initial Values might explain why these deleterious effects were so pronounced in subjects with the healthiest blood lipid profiles. Subjects with optimal initial blood lipids were unable to maintain their ideal blood lipid values after adhering to the Paleo diet for 10 weeks, whereas a similar effect would not likely be seen in subjects with poor initial blood lipid values. Simply limiting food sources to the list of Paleo-approved foods could effectively improve the diet of very unhealthy populations, but the results of our study indicate that it may be difficult to maintain an ideal blood lipid profile while adhering to the commonly followed Paleo guidelines used in this intervention.

While some approaches to the Paleo diet provide guidelines that serve to limit the intake of total fat and saturated fat, such guidelines may not accurately reflect a truly *ad libitum* approach. Indeed, it is likely that many people who adhere to the Paleo diet merely limit themselves to an approved list of food sources and consume those accepted foods *ad libitum*. While previously conducted Paleo studies have utilized restrictive guidelines that help to limit total fat and saturated fat intakes [8-10], there is little data

pertaining to the application of the less restrictive, more practical Paleo guidelines that do not impose limits on the intake of any approved food sources. Our study, which utilized the less restrictive guidelines that appear to be more commonly followed by many Paleo dieters, found deleterious effects on blood lipids following a 10-week Paleo intervention.

Analysis of 3-day diet recalls revealed that subjects were eating an average of 49.75% of daily caloric intake from fat, 13.1% of daily caloric intake from saturated fat, 672.7 mg of dietary cholesterol, and only 25 g of fiber per day. The dietary recommendations from the AHA for maintaining a healthy blood lipid profile include limiting total fat intake to 35% of total caloric intake, limiting saturated fat intake to 7% of total caloric intake, consuming <300 mg of dietary cholesterol daily, and consuming a diet high in fiber [15]. The marked deviations from the AHA recommendations demonstrated by our subjects may explain the deleterious changes to blood lipid values despite their high level of physical activity.

Previous studies have shown positive effects of Paleo interventions on a number of health markers [2, 7-10], and in some cases blood lipids [2, 9]. However, these studies used more restrictive Paleo guidelines, and many used subjects that were initially overweight [2, 8, 9] and were either sedentary [2], had coronary artery disease [8], or had type 2 diabetes [9]. There is a general lack of literature pertaining to Paleo interventions in healthy, highly active populations, and there is reason to question whether previous Paleo studies accurately replicate the actual dietary intake of the free-living Paleo dieter. Diet log analysis revealed that subjects in our study reported higher intakes of total fat, saturated fat, and cholesterol than any of the previously mentioned human interventions [2, 8-10]. While our subjects reported daily intakes of 115 g total fat, 29 g saturated fat,

and 673 mg cholesterol, the four previously mentioned studies averaged drastically lower daily intakes of total fat (67 g), saturated fat (15g), and cholesterol (426 mg) [2, 8-10].

Throughout the intervention, subjects completed a CrossFit-based exercise program while adhering to the Paleo diet, resulting in significant decreases in body weight and body fat percentage while significantly increasing maximal oxygen consumption, a common measure of cardiorespiratory fitness. In previous interventions utilizing diet and/or exercise, improvements in body composition and cardiorespiratory fitness have been accompanied by improvements in one or more blood lipid measurements, including TG [23, 24], TC [23, 24], LDL [23-25], HDL [23, 26], and TC/HDL [23], although it should be noted that some studies have shown HDL levels to drop during active weight loss, before increasing after weight stabilization is achieved [27]. Additionally, cardiorespiratory fitness [28-31] and weight loss [27] have both been independently associated with improved blood lipids. Previous literature would indicate that a combination of Paleo and vigorous exercise that improves cardiorespiratory fitness and body composition should also improve blood lipid measurements. However, while our subjects improved cardiorespiratory fitness and body composition, all blood lipid measurements were either unchanged or negatively impacted. Though the lack of HDL improvement could potentially be attributed to the possibility that many subjects had yet to sufficiently stabilize at their post-test body weight, this does not explain the increases in n-HDL, LDL, TC/HDL, or TC experienced by the group, nor does it explain the absence of TG improvement. Our data indicate that the Paleo diet's deleterious impact on blood lipids was not only significant, but substantial enough to counteract the blood lipid improvements commonly seen with improved fitness and body composition.

It should be acknowledged that the Paleo diet has many positive characteristics. The guidelines allow dieters to consume plenty of high-protein foods and fruit, which have been shown to be highly satiating food sources [32]. Direct comparison of the Mediterranean and Paleo diets has shown that subjects report higher levels of satiety per calorie from the Paleo diet [33]. The approved food list includes an abundance of foods with high micronutrient density but low energy density. For an individual with obesity, merely switching to Paleo-approved foods with low energy density would likely be enough of an improvement from their normal diet to induce positive effects on their health. Additionally, Paleo guidelines do not require dieters to track their calorie intake, a tedious task that many people are unwilling to do. Finally, studies have shown that the low carbohydrate to protein ratio commonly associated with the Paleo diet improves glycemic control and body composition [2, 8, 11-14].

With high levels of reported satiety, the freedom to eat *ad libitum*, absence of calorie tracking, and improvements in glycemic control and body composition, the Paleo diet appears to be an effective way to improve health in populations with obesity or type 2 diabetes. However, the results of our study indicate that Paleo dieting without macronutrient intake guidelines can have deleterious effects on the blood lipids of healthy, active subjects. These results suggest that when adhering to the Paleo diet, the maintenance of a healthy blood lipid profile requires that special attention be paid to dietary variables that may affect blood lipids, including total fat, saturated fat, fiber, and dietary cholesterol.

The Paleo diet encourages the consumption of foods that are satiating, full of micronutrients, and often great sources of fiber, essential fatty acids, and essential amino

acids. Likewise, this diet restricts the consumption of a number of foods that are calorically dense and low in micronutrient content. Replacing highly processed, energy-dense foods with Paleo-approved foods is highly beneficial and achievable for many individuals. However, the diet unnecessarily restricts the consumption of foods that can contribute to a healthy diet, including dairy, whole grains, and legumes. Numerous studies have shown high dairy intake to be associated with decreased risk of all-cause mortality, ischemic heart disease, stroke, and diabetes [34], and studies have shown an inverse relationship between high dairy or calcium intake and bodyweight, body fat, and BMI [35]. In clinical trials, dairy has been shown to be more effective in terms of attenuating fat deposition [36] and increasing tibial cortical thickness [37] than equal amounts of supplementary calcium, prompting researchers to suggest that it is difficult to sufficiently replace the nutrients provided by dairy in the diet with supplements and non-dairy food sources [38, 39]. Whole grains are associated with improved blood lipids and reduced risk of coronary artery disease, cardiovascular disease, type II diabetes, obesity, and some cancers [40-43]. Whole grains also contain a number of phytochemicals and bioactive compounds that may contribute to chronic disease prevention [41, 42]. Some research has shown legumes to be inversely associated with coronary heart disease and cardiovascular disease, and soy protein is known to improve blood lipids by a small but significant degree [43]. While those with Celiac Disease, gluten sensitivity, or lactose intolerance might wisely avoid gluten or lactose, a large body of literature indicates that dairy, legumes, and whole grains can contribute to a healthy, well-rounded diet.

Our study does have some limitations. A low number of diet logs were returned and deemed sufficient for analysis ($n = 8$), which calls into question how accurately the

returned logs portray the dietary intake of the entire group. Also, our study did not impose a high level of control over our subjects— meals were not prepared for subjects, nor did the subjects stay in a metabolic ward for the duration of the study. Despite these limitations, the study used a large sample (n = 43), utilized a sample containing healthier, fitter subjects than previous Paleo interventions, featured a practical application of the Paleo diet that may represent the normal dietary habits of Paleo dieters more accurately than previous studies, and stratified the subjects to examine the diet's effects on the blood lipid profiles of subjects of varying pre-test blood lipid categories. Future studies with more controlled conditions will help to identify the exact dietary adjustments subjects must make to ensure the maintenance of a healthy blood lipid profile while adhering to this popular, practical application of the Paleo diet.

Conclusion:

In conclusion, our study shows that the Paleo diet was significantly deleterious to blood lipid profiles in healthy subjects concurrently participating in a CrossFit-based, high-intensity circuit training program. Specifically, subjects with optimal initial blood lipid values demonstrated the greatest increase in LDL, TC/HDL, TC, and n-HDL values, along with the greatest decline in HDL values, following the 10-week Paleo diet intervention. Despite concurrent improvements in aerobic capacity and body composition noted in these subjects, the Paleo diet may have negated the positive effects of exercise on blood lipids.

Competing Interests:

The authors declare that they have no competing interests.

Authors' contributions:

ETT participated in data collection, literature review, statistical analysis, and helped draft the manuscript. MMS participated in the design of the study, performed the statistical analysis, carried out the blood draws and analysis, and helped draft the manuscript. AJS participated in data collection, statistical analysis, and helped draft the manuscript. BES participated in data collection, statistical analysis, and helped draft the manuscript. STD conceived of the study, participated in its design and coordination and helped to draft the manuscript.

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