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Visceral adiposity and metabolic syndrome after very high–fat and low-fat isocaloric diets: a randomized controlled trial[1,2]

Vivian L Veum,[3,4,6,8] Johnny Laupsa-Borge,[3,6–8] Øyvin Eng,[6] Espen Rostrup,[7] Terje H Larsen,[7] Jan Erik Nordrehaug,[3] Ottar K Nygård,[3,4,7] Jørn V Sagen,[3,4,6] Oddrun A Gudbrandsen,[5] Simon N Dankel,[3,4,6] and Gunnar Mellgren[3,4,6]**

[3]Department of Clinical Science, [4]KG Jebsen Centre for Diabetes Research, Department of Clinical Science, and [5]Department of Clinical Medicine, University of Bergen, Bergen, Norway; and [6]Hormone Laboratory and [7]Department of Heart Disease, Haukeland University Hospital, Bergen, Norway

ABSTRACT

Background: Different aspects of dietary pattern, including macronutrient and food profiles, may affect visceral fat mass and metabolic syndrome.

Objective: We hypothesized that consuming energy primarily from carbohydrate or fat in diets with similar food profiles would differentially affect the ability to reverse visceral adiposity and metabolic syndrome.

Design: Forty-six men (aged 30–50 y) with body mass index (in kg/m^2) >29 and waist circumference >98 cm were randomly assigned to a very high–fat, low-carbohydrate (VHFLC; 73% of energy fat and 10% of energy carbohydrate) or low-fat, high-carbohydrate (LFHC; 30% of energy fat and 53% of energy carbohydrate) diet for 12 wk. The diets were equal in energy (8750 kJ/d), protein (17% of energy), and food profile, emphasizing low-processed, lower-glycemic foods. Fat mass was quantified with computed tomography imaging.

Results: Recorded intake of carbohydrate and total and saturated fat in the LFHC and VHFLC groups were 51% and 11% of energy, 29% and 71% of energy, and 12% and 34% of energy, respectively, with no difference in protein and polyunsaturated fatty acids. Mean energy intake decreased by 22% and 14% in the LFHC and VHFLC groups. The diets similarly reduced waist circumference (11–13 cm), abdominal subcutaneous fat mass (1650–1850 cm^3), visceral fat mass (1350–1650 cm^3), and total body weight (11–12 kg). Both groups improved dyslipidemia, with reduced circulating triglycerides, but showed differential responses in total and low-density lipoprotein cholesterol (decreased in LFHC group only), and high-density lipoprotein cholesterol (increased in VHFLC group only). The groups showed similar reductions in insulin, insulin C-peptide, glycated hemoglobin, and homeostasis model assessment of insulin resistance. Notably, improvements in circulating metabolic markers in the VHFLC group mainly were observed first after 8 wk, in contrast to more acute and gradual effects in the LFHC group.

Conclusions: Consuming energy primarily as carbohydrate or fat for 3 mo did not differentially influence visceral fat and metabolic syndrome in a low-processed, lower-glycemic dietary context. Our data do not support the idea that dietary fat per se promotes ectopic adiposity and cardiometabolic syndrome in humans. This study was registered at clinicaltrials.gov as NCT01750021. *Am J Clin Nutr* doi: 10.3945/ajcn.115.123463.

Keywords: saturated fat, food profile, obesity, metabolic syndrome, computed tomography, CT

INTRODUCTION

Overweight and obesity cause major health problems worldwide by contributing to comorbidities such as fatty liver, type 2 diabetes (T2D)[9], cardiovascular disease (CVD), and certain cancers (1). Resistance to the hormone insulin characterizes metabolic syndrome and the associated risk of CVD and T2D (2, 3). Key components of metabolic syndrome are increased abdominal fat mass, elevated circulating triglyceride concentrations, reduced circulating concentration of HDL cholesterol, and elevated blood pressure (3). Fat accumulation, particularly in the intra-abdominal region (visceral adiposity), strongly associates with insulin resistance–related hyperglycemia and dyslipidemia (4, 5), with or without accompanying fatty liver (6). Fat storage depends on dietary habits that

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[2] Supplemental Tables 1–8 and Supplemental Figure 1 are available from the "Online Supporting Material" link in the online posting of the article and from the same link in the online table of contents at http://ajcn.nutrition.org.

[8] These authors were cofirst authors for this work.

*To whom correspondence should be addressed. E-mail: simon.dankel@uib.no (SN Dankel), gunnar.mellgren@uib.no (G Mellgren).

[9] Abbreviations used: CT, computed tomography; CVD, cardiovascular disease; HOMA2-IR, homeostasis model assessment of insulin resistance 2 index (computer model); HU, Hounsfield unit; INCP, insulin C-peptide; ITT, intention-to-treat; LFHC, low-fat, high-carbohydrate; LMEM, linear mixed-effects model; NEFA, nonesterified fatty acid; PP, per protocol; RER, respiratory exchange ratio; SAT, subcutaneous adipose tissue; TC, total cholesterol; T2D, type 2 diabetes; VAT, visceral adipose tissue; VHFLC, very high–fat, low-carbohydrate; WC, waist circumference; 95% BCa CI, bootstrapped (bias corrected and accelerated) 95% CI.

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favor excess energy intake relative to expenditure, largely from highly palatable foods that typically combine processed carbohydrates and fats (7).

Randomized controlled trials have shown that a sustained reduction in total energy, either by reducing fat or carbohydrate intake, can reduce body weight and metabolic syndrome in overweight and obese individuals (8, 9). Moreover, the health effects of nutrition partly depend on the types and sources of carbohydrates and fats, such as high- or low-glycemic–index foods (10–12), simple or complex sugars (13), and fructose or glucose (14). Low-carbohydrate or low-glycemic diets have gained popularity as part of a health-promoting lifestyle, partly because of the potential for temporary or long-term weight loss associated with reduced glycemic load and improved insulin sensitivity (10, 15). Yet, there is concern that an accompanying increase in the intake of total fat and SFAs may promote adverse health effects (16, 17), including visceral fat accumulation (18, 19) and nonalcoholic fatty liver disease (20). The suggested negative effects of SFAs have largely been attributed to a stimulatory effect on circulating cholesterol (21), in particular LDL cholesterol (22, 23), as well as inflammation and other mechanisms that interfere with insulin signaling (2, 24, 25). These purported adverse effects of SFAs have been suspected to persist even in the context of weight loss (26, 27).

Clinical (and animal) studies exploring the effects of high-fat diets often include energy-dense diets that also have a substantial amount of processed carbohydrates and poor overall quality (19), confounding the effect of a high intake of fat per se (22, 28). Most previous randomized controlled trials comparing low-carbohydrate and low-fat diets have been performed with a substantial energy restriction (outside a normal caloric intake of 8500–10,500 kJ/d) (29), or differing intake of carbohydrates, protein, and PUFAs, and/or without strict control of food profiles and quality.

In the present study (NCT01750021), we randomly assigned abdominally obese men to diets dominated by either fat or carbohydrate to evaluate possible differential effects on visceral fat mass and body composition (primary outcome measures), and biochemical and clinical variables, including components of metabolic syndrome (secondary outcome measures). The low-fat, high-carbohydrate (LFHC) and very high–fat, low-carbohydrate (VHFLC) diets were planned to be isocaloric and modestly energy-restrictive with an equal intake (percentage of total energy intake) of protein and PUFAs. To reduce bias from different food profiles, the experimental menus were based on the same food items in both diet groups.

METHODS

Participants and study design

The study was conducted according to the guidelines in the Declaration of Helsinki. All experimental procedures were approved by the Regional Ethics Committee (comparison of 2 weight-reducing diets in overweight men, 2011/2282/REK west). The study design, sample collection, and potential risks and benefits were carefully explained to each participant before they provided written informed consent.

We recruited 56 male subjects with abdominal obesity, normal fasting blood glucose <7 mmol/L, waist circumference (WC) >98 cm, and BMI (in kg/m^2) >29 or percentage of body fat ≥ 25 through a project description in a local newspaper. Exclusion criteria were severe diseases, including inflammatory bowel diseases, known food allergies, regular medication (except for alkalizing gastric buffers), attempts at systematic weight reduction over the previous 6 mo, and/or regularly consuming >2 alcohol units/wk. To further promote a homogeneous group, only men between 30 and 50 y of age were included.

The trial was performed in Bergen, Norway. From August 2012 to the intervention startup in January 2013, 2 prescreenings were performed to ensure that all participants met the inclusion criteria at the time of intervention. Six men did not meet the inclusion criteria, and 4 withdrew before random assignment, leaving 46 participants who were randomly assigned to a VHFLC or LFHC diet (parallel design). Random assignment was performed after the second prescreening ~ 8 wk before baseline. The list of prescreening appointments was used to randomly assign the participants by drawing ballots and blocking with block sizes of 2. Two additional men were lost before baseline assessment (1 in the LFHC group did not show up, and 1 in the VHFLC group withdrew for personal reasons), leaving a total of 44 men who started dieting. All participants were informed of group allocation after the baseline measurements and sampling. The nature of the trial required an open intervention with no blinding of the trial participants or the investigators.

The sample size was considered to be sufficient when assessed by the power in similar previous studies (16); the strong homogeneity with respect to age, sex, and abdominal adiposity; and the planned large difference in fat and carbohydrate intake. Post hoc power analysis for the primary outcome [(difference in change of visceral fat mass measured by computed tomography (CT)] indicated $\sim 40\%$ power for the per protocol analysis, and we could not rule out type II errors.

The body weight of all participants was stable ($<5\%$ change) before the intervention, and they were asked to refrain from changing their physical activity level during the intervention. To promote compliance, the participants were questioned about their ability to strictly follow a diet, and the importance of honesty and accuracy was emphasized during consultations.

To limit differences in individual micronutrient status that might influence energy metabolism, the participants were instructed to take a broad-spectrum vitamin and mineral supplement [Solaray Spektro without iron, provided by Au Naturel (UK)] for 8 wk before the start of the intervention.

Study visits

The study was carried out from January to May 2013. Each period of study visits (baseline and 4, 8, and 12 wk) was completed within ~ 3 wk. Four participants from the same diet group attended at any 1 d during the period of baseline visits, and the same participant order was used at follow-up visits. The participants arrived after fasting overnight or for ≥ 10 h, during which a small amount of water was permitted. An ~ 15 min conversation with a nutritionist in the presence of a physician took place at all visits. Measurements were performed in the following order: bioelectrical impedance analysis, indirect calorimetry, blood samples, and CT scan. To maximize retention and compliance to the diets, participants were followed up closely with regular contact between visits, and they were offered individual counseling as needed throughout the study.

Study diets

The planned macronutrient profile for the LFHC group was 53% of energy (275 g/d) carbohydrate, 17% of energy (90 g) protein, and 30% of energy (70 g) fat, in accordance with typical current Western dietary guidelines. The planned macronutrient profile for the VHFLC group was 10% of energy (50 g) carbohydrate, 17% of energy (90 g) protein, and 73% of energy (170 g) fat. Because our purpose was to compare responses to well-defined diets sharply contrasting in carbohydrate and fat within a normal caloric range, and not explicitly examining effects of energy restriction, all participants were asked to consume a total of 8750 kJ/d from carbohydrate, fat, and protein.

All men in both groups were instructed to avoid hydrogenated vegetable fat, sugar, and foods with added sugar, and to restrict the intake of highly processed foods and plant oils with high amounts of ω-6 fatty acids. Both groups were told to consume ≥500 g vegetables, berries, and fruits (emphasizing vegetables relative to fruits)/d, and to eat vegetables with every meal. Two fish dinners per week were recommended. To meet the energy requirement from carbohydrate on the LFHC diet, while avoiding excessive portion sizes of vegetables and rice, we chose to include juice as a regular carbohydrate source in this diet. To help participants choose standardized ingredients, they were provided with some products donated by food suppliers, including butter (Tine); coconut fat (Cocosa; Soma Nordic); erythritol and stevia as sugar substitutes (Sukrin+; Funksjonell Mat); and a special mix of, among other ingredients, almond flour and plant fibers to make low-carbohydrate breads and cakes (Funksjonell Mat).

To facilitate compliance and dietary recording during intervention, a comprehensive recipe booklet was customized for each diet group with precalculated nutrient content and clear instructions for meal and snack preparation. The recipe for each meal or snack was designed to comply with the daily intake of macronutrients (both in grams and percentage of energy) as well as the recommended food profile, emphasizing homemade meals and low-processed foods without added sugar. The booklet included 537 recipes (>225 for each diet and some common) that participants could choose from, in turn comprising 1092 ingredients or products. The booklet was developed with the software FileMaker Pro 12 Advanced, and was made available as an electronic application for iPhone and iPad, as well as in pdf format. Importantly, we obtained complete information on the content of total carbohydrate, fat, and protein for all predefined meals. Of the ingredients and products, 99 had missing data for dietary fiber, 228 for added sugar, 75 for fatty acid subtypes (saturated, monounsaturated, or polyunsaturated), and 175 for cholesterol. The food selections were carefully designed to minimize differences in food types consumed within and between groups. Before baseline, the participants completed a half-day course at which they received detailed instructions on how to implement the recipe booklet system.

Diet recording

Before starting the diet in January/February 2013, the participants completed baseline diet records for 5 consecutive days (including weekends). During the intervention, diet recording was carried out over 5 successive days (including weekends) each month, for a total of 15 d (17% of the entire intervention). The participants were asked to weigh and record the amount of all ingredients and products that were eaten at every meal (breakfast, lunch, dinner, supper, and snacks), together with the time of the meal. They submitted the consumed ingredients in an online dietary recording system (www.diett.no; operated by Dietika) linked to data on nutritional content derived from the official Norwegian Food Composition Table (www.matvaretabellen.no) and declaration by producers. When Norwegian data were not available, values from international databases (Danish, Finnish, or US food composition tables) were used. The recording system at www.diett.no directly calculated energy intake and amount of macronutrients and some micronutrients. The participants also provided additional comments on their food intake on this system, as well as records of physical activity, i.e., the frequency, duration, and intensity of different sport and daily life activities, which was converted to kilocalories per hour on the basis of estimated energy expenditure for the recorded activities (30). The data were transferred to a separate database (created by FileMaker Pro 12 Advanced) for further data processing.

During the intervention, the participants used www.diett.no to simply report the identifications of our recipes and dishes when following the predefined menus in the recipe booklet. Deviations from the menus were recorded in a template below the dish identification. When participants were traveling or dining out, they reported the amount of ingredients as standardized household measures and recorded them manually as for baseline records. To reduce biased diet recording, the participants were told that the selection of foods and recipes during these periods should reflect the actual food intake and the degree of deviation from the diet throughout the whole month. The online system also allowed interaction with participants during dietary recordings to help rectify missed recordings and obvious mistakes.

Blood samples

Venous blood (whole blood, plasma, and serum) was collected from fasting subjects between 0800 and 1130 and stored at −80°C after preparation. All analyses were performed at the Laboratory of Clinical Biochemistry and the Hormone Laboratory at Haukeland University Hospital according to standardized procedures, with the exception of serum nonesterified fatty acids (NEFAs), which were analyzed with the use of a NEFA FS kit from DiaSys (Diagnostics Systems) on a Cobas c111 Chemistry Analyzer (Roche Diagnostics). All samples from each single participant were measured simultaneously.

The homeostasis model assessment is reported as a surrogate measure of insulin sensitivity, with the use of the homeostasis model assessment calculator developed by the University of Oxford (http://www.dtu.ox.ac.uk/homacalculator/index.php) to estimate insulin resistance (HOMA2-IR) and sensitivity (homeostasis model assessment of insulin sensitivity index 2) on the basis of the updated computerized model (31, 32). We used glucose and insulin C-peptide (INCP) data for this.

Respiratory gas analyses

Respiratory gases were measured by an ergospirometer with a breath-to-breath analyzer (Schiller Cardiovit CS-200 Ergo-Spiro/13 Ganzhorn Power Cube; Schiller) (indirect calorimetry). The participants were told to avoid physical exertion and activity

for 2 d before the analysis to prevent interference by lactic acid and oxygen liabilities. Participants were placed in a prone position and rested for ~30 min during ventilation gas exchange measurement (20 min) to determine resting energy expenditure. The gas exchange from nutrient substrate catabolized for energy was assessed with the use of a respiratory exchange ratio (RER), i.e., the ratio of carbon dioxide produced to oxygen consumed. The RER for carbohydrate (glucose) oxidation produces an equal number of carbon dioxide molecules to oxygen molecules consumed, i.e., RER = 1. The RER for lipids range between 0.69 and 0.73, depending on the oxidized fatty acid carbon chain length (33). A Weir equation was used according to the exchange of gases to quantify the RER (34), and we used this quotient to verify dietary compliance.

Blood pressure was measured by a BP-200 plus oscillometric monitor (Schiller) during indirect calorimetry when the men were in a resting prone position (mean of 3 measurements).

Body composition

Body weight and composition were measured by a segmental multifrequency bioelectrical impedance measurement system (InBody 720; BioSpace). The participants were measured while barefoot and wearing light clothing, and were asked to use the restroom shortly before and to stand in an upright position for ≥5 min before measurement.

CT scan

Noncontrasted CT scans were performed to assess fat lipid accumulation in the abdominal area, i.e., visceral adipose tissue (VAT), subcutaneous adipose tissue (SAT), and liver. CT allows precise assessment of liver density measured in Hounsfield units (HUs), calculated as liver attenuation:spleen attenuation HU index and liver-spleen HU difference, which correlate strongly with liver triglycerides and liver macrosteatosis (35).

Volumetric adipose tissue imaging was performed on participants while they were lying in a supine position with the use of a 256-slice multidetector CT scanner (SOMATOM Definition Flash; Siemens). Participants had a mean of 70 contiguous 5-mm-thick cross-sectional abdominal images (120 peak kilovoltage; 180 milliampere; gantry rotation time, 500 ms). The cross-sectional abdominal images covered 350 mm from the level of L5/S1 to the upper right diaphragm. The mean radiation dose was 8.0 milligray.

A previously established method was used for measurement of the abdominal adipose tissue (36). SAT and VAT, as well as WC, were measured with an offline workstation (Aquarius iNtuition Edition version 4.4.7.85.5213; TeraRecon). WC (centimeters) was measured with the use of a single slice (5-mm thickness) at the umbilical level. HU values of pixels in CT correspond directly to the tissue property. A predefined image display setting with an image display window width of −195 to −45 HU was applied to identify pixels containing adipose tissue (37). To separate VAT from SAT, the abdominal muscular wall separating the 2 compartments was manually traced. SAT and VAT were measured across the total imaging volume and were calculated in cubic centimeters. The mean time for image analysis was 10 min/subject. Regrettably, data from 2 participants were lost during temporary storage and inadequate data transfer.

Statistics

We reported the results from a per protocol (PP) analysis for 38 participants who completed the study (**Tables 1–5**, **Figures 1–4**, **Supplemental Tables 1–5**) of the 46 who were initially enrolled. We also performed an intention-to-treat (ITT) analysis that included all randomly assigned participants except 2 cases that were lost before baseline (44 eligible participants in total). These 2 were unaware of the group allocation and did not provide any dietary or clinical data. Because the trial's objective was explanatory and we sought to determine the efficacy of the 2 diets, not primarily the effectiveness, we chose to report the PP analysis in the main manuscript (Tables 1–5, Figures 1–4), and to present the ITT analysis for our primary outcomes in **Supplemental Tables 6** and **7**. Notably, the results from the PP and ITT analyses did not differ in nominal significance for any of the primary or secondary outcomes.

Our missing data analysis (data not shown), including Little's missing completely at random test, indicated that the dropouts were missing (completely) at random, and we found no significant differences between dropouts and completers in baseline characteristics, except for significant higher diastolic blood pressure, total cholesterol (TC), and LDL cholesterol in completers. In the reported PP and ITT analyses, we used mixed models without imputation to handle missing data. However, we also conducted an ITT analysis based on a full dataset in which missing values in the original data were replaced by values from multiple imputation (data not shown). Differences in nominal significance between these ITT analyses are indicated in the footnotes of Supplemental Tables 6 and 7.

We tested for outliers and influential cases with the use of standardized and Studentized residuals (z score >3.29), boxplots, residual plots, and Cook's distance [$D > 4/(n - k - 1)$]. The assumption of normality, assessed by graphic tools and the Shapiro-Wilk test, was violated for some dietary and clinical variables. Therefore, both parametric and nonparametric significance tests, including robust methods with trimmed means, M-estimation, and/or bootstrapping, were conducted and compared, but no differences in nominal significance between methods were found. Here, we chose to report results from the parametric tests.

Group differences in score changes between different time points were analyzed by linear mixed-effects models (LMEMs) with the use of best-fitted variance and random structures, and diet, time, and diet × time as fixed effects factors with the categorical time variable defined by appropriate contrasts (**Supplemental Table 8**). In some analyses, means or score changes were compared between groups with the use of independent-samples t tests with Welch correction for unequal variances. If no significant group difference was found, we tested the score change from baseline to 12 wk for the combined groups (i.e., pooled data for all participants).

When testing for group differences in primary and secondary outcomes, the models were adjusted for age and covariates with potentially clinically important differences at baseline, i.e., intake of energy, carbohydrate, added sugar, and cholesterol (Table 1). The adjusted and unadjusted results did not differ in nominal significance, except for short-term responses between intermediate time points for some biochemical variables (Table 5). In separate analyses not shown, we also tested several other covariate

TABLE 1
Mean energy and macronutrient intake at baseline and during the intervention for the LFHC (n = 16) and VHFLC (n = 20) groups (per protocol analysis)[1]

Variable and diet	Planned[2]	Baseline	Intervention[3]	Score change[4]	VHFLC vs. LFHC[5]	Relative %[6]
Energy, kJ					+1024 (−469, 2518)	
LFHC	8750	11,984 ± 2017	9226 ± 711	−2759 (−3753, −1765)		−22.2 ± 14.8
VHFLC	8750	10,632 ± 2219	8898 ± 409	−1734 (−2838, −630)		−14.4 ± 22.1
Carbohydrate, g					−178 (−231, −124)***	
LFHC	275	306 ± 62.0	281 ± 23.5	−25.4 (−54.4, 3.60)		−6.9 ± 15.6
VHFLC	50	259 ± 94.1	56.1 ± 5.1***	−203 (−246, −160)		−77.1 ± 28.8
Protein, g					+6.8 (−10.6, 24.3)	
LFHC	90	117 ± 29.9	91.9 ± 7.8	−25.3 (−41.2, −9.41)		−19.7 ± 20.5
VHFLC	90	108 ± 18.5	89.3 ± 3.7	−18.5 (−26.9, −10.0)		−15.9 ± 16.5
Fat, g					+105 (85.3, 124)***	
LFHC	70	116 ± 28.0	71.9 ± 6.4	−43.6 (−58.2, −29.1)		−36.1 ± 22.1
VHFLC	170	106 ± 27.0	167 ± 8.0***	+61.1 (46.4, 75.8)		+63.1 ± 37.3
Fiber, g					−11.5 (−15.5, −7.47)***	
LFHC	no	23.5 ± 4.9	30.6 ± 3.4	+7.1 (4.46, 9.78)		+32.2 ± 23.1
VHFLC	no	21.5 ± 5.7	17.1 ± 5.3***	−4.4 (−7.58, −1.21)		−22.5 ± 37.5
Added sugar, g					+14.8 (−12.8, 42.6)	
LFHC	0	67.8 ± 32.3	4.8 ± 8.9	−63.0 (−81.3, −44.6)		−95.2 ± 73.6
VHFLC	0	48.8 ± 48.0	0.6 ± 0.9	−48.2 (−70.6, −25.7)		−97.6 ± 91.8
Alcohol, g					+1.0 (−13.7, 15.6)	
LFHC[7]	0	11.9 ± 24.8	1.0 ± 1.3	−11.0 (−24.6, 2.65)		−78.4 ± 78.3
VHFLC	0	10.6 ± 15.7	0.6 ± 0.9	−10.0 (−17.4, −2.63)		−89.8 ± 71.7
SFA, g					+55.6 (47.2, 63.9)***	
LFHC	no	47.6 ± 12.5	30.5 ± 4.4	−17.1 (−23.0, −11.2)		−34.3 ± 21.3
VHFLC	no	42.1 ± 10.5	80.6 ± 9.4***	+38.5 (31.9, 45.1)		+97.3 ± 40.0
MUFA, g					+37.9 (30.9, 44.9)***	
LFHC	no	37.3 ± 9.5	20.0 ± 3.1	−17.3 (−22.8, −11.9)		−45.5 ± 26.7
VHFLC	no	33.5 ± 9.7	54.0 ± 4.3***	+20.6 (15.3, 25.8)		+68.2 ± 41.3
PUFA, g					+2.1 (−2.99, 7.18)	
LFHC	no	17.1 ± 6.7	11.2 ± 2.1	−5.9 (−9.38, −2.39)		−30.1 ± 34.9
VHFLC	no	16.2 ± 7.7	12.4 ± 2.5	−3.8 (−7.78, 0.21)		−18.4 ± 36.9
Cholesterol, mg					+372 (233, 510)***	
LFHC	no	294 ± 86.4	371 ± 126	+76.9 (2.85, 151)		+24.6 ± 58.2
VHFLC	no	347 ± 163	795 ± 240***	+448 (342, 554)		+144 ± 66.7

[1] Values are raw unadjusted means ± SDs or mean score changes (95% CIs) and are based on values from all participants who provided diet records at baseline and from ≥1 time point (after 4, 8, and/or 12 wk) during the diet intervention. Values for the intervention period are the means of outcomes from dietary recordings after 4, 8 and 12 wk. CIs and P values are from 2-tailed analysis with the use of linear mixed-effects models (LMEMs) with best-fitted variance and random structures. Two participants in the LFHC group did not record their food intake. ***Significantly different: ***$P \leq 0.001$. LFHC, low-fat, high-carbohydrate; no, no specific recommendation; VHFLC, very high–fat, low-carbohydrate.

[2] Energy intake and macronutrient profile for the planned diets. Specified kiloJoules include the energy content of the planned amounts of the macronutrients carbohydrate, protein, and fat, and not fiber, for which there were no specific recommendations, or alcohol, for which the recommendation was no alcohol. Therefore, the actual energy content in the planned diets would be slightly higher.

[3] Asterisks designate a significantly different intake between groups during the intervention.

[4] Absolute mean score change from baseline to intervention, and 95% CI from an LMEM.

[5] Group difference in absolute score change from baseline to intervention, and 95% CI from a linear mixed-effects model. −: Greater reduction (or less increase) in the VHFLC group than in the LFHC group. +: Less reduction (or greater increase) in the VHFLC group. Asterisks designate a significant group difference.

[6] Relative percentage change from baseline to intervention calculated from the ln ratio. Log ratio = ln (follow-up value/baseline value). % = $(e^{\text{mean log ratio}} - 1) \times 100$.

[7] Extreme values for 3 participants in the LFHC group were excluded. These extreme values were due to very high alcohol intake at a single event during the entire intervention period.

scenarios, including a model adjusted for age and important factors that we intended to control for in our design, and that consequently should not vary between groups at baseline and during the intervention, i.e., activity level and intake of energy, protein, PUFAs, and alcohol. None of these adjusted models changed the nominal significance compared with the unadjusted models for variables reported in Tables 2–4 and Supplemental Tables 4–7.

Bivariate and partial correlation analyses with 95% bootstrapped (bias corrected and accelerated) CIs (95% BCa CIs) were used to determine associations between variables. Here, Pearson's r, 95% BCa CI, and P values are reported.

All data are presented as raw unadjusted means ± SDs or score changes (95% CIs) if not otherwise specified. P values < 0.05 were considered to be statistically significant when the 95% CI did not cross zero, and all P values are from 2-tailed analyses. The statistical analyses were performed with R, version 3.3.0 (https://www.R-project.org), and the LMEM analyses were conducted with the nlme package, version 3.1-128 (https://CRAN.R-project.org/package=nlme).

TABLE 2
Changes in anthropometric variables and blood pressure with the LFHC (n = 18) and VHFLC (n = 20) diets (per protocol analysis)[1]

Variable and diet	Baseline	4 wk[2]	8 wk[2]	12 wk[2,3]	Score change[4]	Time × group[5]
BMI, kg/m^2					−3.6 (−4.04, −3.18)***	0.571
LFHC	33.6 ± 3.6	31.5 ± 3.5	30.7 ± 3.4	29.9 ± 3.3		
VHFLC	34.1 ± 2.4	32.2 ± 2.2	31.3 ± 2.1	30.6 ± 1.9		
Body fat, %					−6.3 (−6.97, −5.56)***	0.234
LFHC	33.3 ± 5.4	30.5 ± 6.0	28.7 ± 6.3	26.7 ± 6.1		
VHFLC	34.0 ± 4.8	31.5 ± 5.2	30.1 ± 5.2	28.0 ± 5.3		
Body fat mass, kg					−10.3 (−11.5, −9.16)***	0.470
LFHC	37.5 ± 9.8	32.5 ± 9.7	29.5 ± 9.4	26.9 ± 8.7		
VHFLC	38.7 ± 6.8	33.9 ± 6.9	31.5 ± 6.7	28.6 ± 6.4		
Fat-free mass, kg					−1.7 (−2.22, −1.13)***	0.898
LFHC	73.5 ± 6.8	71.8 ± 6.2	71.9 ± 6.0	71.9 ± 6.0		
VHFLC	74.9 ± 7.3	73.4 ± 6.7	72.7 ± 6.3	73.1 ± 6.6		
Skeletal muscle mass, kg					−1.0 (−1.36, −0.73)***	0.850
LFHC	41.9 ± 3.9	40.9 ± 3.6	40.8 ± 3.5	40.9 ± 3.6		
VHFLC	42.7 ± 4.3	41.8 ± 3.8	41.4 ± 3.7	41.6 ± 3.9		
Waist circumference, cm					−11.7 (−13.0, −10.3)***	0.073
LFHC	117 ± 10.4	112 ± 10.2	107 ± 10.1	104 ± 10.4		
VHFLC	116 ± 6.8	112 ± 7.7	108 ± 7.9	105 ± 7.6		
Visceral fat area, cm^2					−57.2 (−65.2, −49.1)***	0.870
LFHC	198 ± 52.5	170 ± 48.5	153 ± 48.3	142 ± 45.2		
VHFLC	196 ± 48.0	171 ± 39.8	157 ± 40.3	139 ± 26.9		
Systolic BP, mm Hg					−14.5 (−18.7, −10.3)***	0.748
LFHC	133 ± 15.6	—	—	118 ± 11.7		
VHFLC	133 ± 13.7	—	—	119 ± 15.1		
Diastolic BP, mm Hg					−5.0 (−8.11, −1.87)**	0.130
LFHC	85.4 ± 7.7	—	—	77.4 ± 9.0		
VHFLC	84.7 ± 9.4	—	—	82.4 ± 7.4		

[1] Values are raw unadjusted means ± SDs or mean score changes (95% CIs). Body composition was measured with a segmental multifrequency bioelectrical impedance measurement system (InBody 720). CIs and P values are from 2-tailed analysis with the use of LMEMs with best-fitted variance and random structures, and diet, time, and diet × time as fixed-effects factors. **·***Significantly different: **$P < 0.01$, ***$P \leq 0.001$. BP, blood pressure; LFHC, low-fat, high-carbohydrate; LMEM, linear mixed-effects model; VHFLC, very high–fat, low-carbohydrate.

[2] LMEMs with time as a fixed effects factor coded by appropriate contrasts showed that the anthropometric variables changed significantly ($P < 0.001$) between different time points (from baseline to 4 wk, from 4 to 8 wk, and from 8 to 12 wk) within groups and for the combined groups (pooled data), except for fat-free mass and skeletal muscle mass (data not shown). Group differences in absolute score changes between intermediate time points were not significant for any of these variables.

[3] No significant difference between groups was found for any variable at 12 wk (data not shown).

[4] Absolute mean score change from baseline to 12 wk for pooled data, and 95% CI from an LMEM. Because there was no significant difference between the changes in the diet groups for any of the variables, we tested the changes from baseline to 12 wk for the combined groups. Within-group changes are shown in Supplemental Table 4.

[5] P value for change from baseline to 12 wk between groups (time × group interaction) from an LMEM adjusted for age and baseline intake of energy, carbohydrate, added sugar, and cholesterol (see main text). The adjusted and unadjusted (Supplemental Table 4) results did not differ in nominal significance, and were not significantly different from the intention-to-treat analysis (Supplemental Table 6).

RESULTS

Of the 46 randomly assigned participants, 2 withdrew before baseline visits, and 4 dropped out early during follow-up, leaving a total of 40 men who completed the intervention. Furthermore, data from 2 men in the VHFLC group were excluded from the analysis because of noncompliance based on diet records and data collected at study visits. The final numbers of analyzed participants were therefore 18 in the LFHC group (aged 40.2 ± 4.50 y) and 20 in the VHFLC group (aged 40.3 ± 5.53) (Figure 1A, **Supplemental Figure 1**). The results presented here originate from the PP analysis, which included these 38 participants, whereas data from the ITT analysis of all randomly assigned subjects (except the 2 lost before baseline) are not reported in this section because these results did not differ from the PP analysis in nominal significance.

Dietary intake and food profile

Detailed 5-d dietary records showed no differences at baseline in the intake of energy and macronutrients (Figure 1B, Table 1, Supplemental Table 1). As planned during the intervention, total energy and protein intake were equal in the 2 diet groups. The percentage of energy from fat and carbohydrate for the LFHC diet was 29% and 51%, respectively, and for the VHFLC diet, it was 71% and 11%, respectively (Figure 1B, Supplemental Table 1). The estimated contribution of SFAs to total energy intake was 12% of energy and 34% of energy for the LFHC and VHFLC diets, respectively (Supplemental Table 1). The dietary records also estimated an overall modest reduction in total energy intake for both diets (22% and 14% reduction for LFHC and VHFLC diets, respectively), but with notable interindividual variation (Figure 1B, Table 1).

TABLE 3
Changes in ectopic fat deposition analyzed by computed tomography imaging with the LFHC (n = 18) and VHFLC (n = 18) diets (per protocol analysis)[1]

Variable and diet	Baseline	12 wk[2]	Score change[3]	Time × group[4]
Total abdominal fat, cm^3			−3259 (−3663, −2854)***	0.135
LFHC	14,962 ± 4380	11,465 ± 4202		
VHFLC	15,202 ± 3132	12,181 ± 3165		
Subcutaneous fat, cm^3			−1769 (−2012, −1525)***	0.301
LFHC	8208 ± 2706	6352 ± 2562		
VHFLC	8420 ± 2482	6739 ± 2388		
Visceral fat, cm^3			−1490 (−1707, −1273)***	0.107
LFHC	6754 ± 2361	5113 ± 2133		
VHFLC	6781 ± 1749	5443 ± 1611		
Volume ratio, %			−0.23 (−0.77, 0.30)	0.112
LFHC	45.0 ± 7.9	44.4 ± 8.2		
VHFLC	45.0 ± 9.3	45.2 ± 9.5		
Density ratio (liver:spleen)			+0.13 (0.07, 0.18)***	0.544
LFHC	1.20 ± 0.23	1.30 ± 0.18		
VHFLC	1.11 ± 0.26	1.26 ± 0.11		
Pericardial fat, cm^3			−66.3 (−88.1, −44.6)***	0.408
LFHC	910 ± 126	839 ± 109		
VHFLC	930 ± 102	869 ± 79.1		

[1] Values are raw unadjusted means ± SDs or mean score changes (95% CIs). Volume ratio (%) = (visceral fat volume/total abdominal fat) × 100. Density ratio = liver HU:spleen HU. Density by HU was calculated from the mean of 3 measurements. CIs and P values are from 2-tailed analysis with the use of LMEMs with best-fitted variance and random structures, and diet, time, and diet × time as fixed effects factors. Values from 2 participants in the VHFLC group were lost during data transfer and storage. ***Significantly different: ***$P \leq 0.001$. HU, Hounsfield unit; LFHC, low-fat, high-carbohydrate; LMEM, linear mixed-effects model; VHFLC, very high–fat, low-carbohydrate.

[2] No significant difference between groups was found for any variable at 12 wk (data not shown).

[3] Absolute mean score change from baseline to 12 wk for pooled data and 95% CI from an LMEM. Because there was no significant difference between the changes in the diet groups for any of the variables, we tested the changes from baseline to 12 wk for the combined groups. Within-group changes are shown in Supplemental Table 4.

[4] P value for change from baseline to 12 wk between groups (time × group interaction) from an LMEM adjusted for age and baseline intake of energy, carbohydrate, added sugar, and cholesterol (see main text). The adjusted and unadjusted (Supplemental Table 4) results did not differ in nominal significance, and were not significantly different from the intention-to-treat analysis (Supplemental Table 7).

Compared with baseline, the absolute intake in grams of total fat, SFAs, and MUFAs increased in the VHFLC group by 63%, 97%, and 68%, respectively, whereas total and saturated fat were reduced by 34–36% with the LFHC diet (Table 1). The absolute intake of PUFAs was reduced by 30% and 18% in the LFHC and VHFLC groups, respectively. Importantly, there was no significant group difference in total PUFA intake during the diet intervention, and the percentage of energy of PUFAs at baseline remained unchanged for both interventions (Supplemental Table 1). The percentage of energy from protein remained unchanged from baseline on the 2 diets (after a 20% and 16% reduction in absolute intake in the LFHC and VHFLC groups, respectively), and did not differ between groups during the intervention (Table 1, Supplemental Table 1). Although the percentage of energy from carbohydrates increased on the LFHC diet in replacement of fat, the absolute intake of carbohydrate did not. Notably, the intake of added sugar was reduced in both groups by almost 100%, whereas the absolute intake of fiber increased in the LFHC group and decreased in the VHFLC group. However, the percentage of energy from fiber did not change significantly in the VHFLC group. Finally, cholesterol intake increased by 25% with the LFHC diet and more than doubled with the VHFLC diet compared with baseline (Table 1).

In line with the planned standardization of food profile, participants assigned to both diets consumed the same primary sources of fats, carbohydrates, and proteins (Supplemental Table 2). Of note, the LFHC group consumed almost twice the volume of food that the VHFLC group consumed (2126 compared with 1234 g/d), primarily because of a higher intake of vegetables, breads, rice, and juices (Supplemental Table 2). The most frequently chosen menus from the 2 diets are shown in Supplemental Table 3.

As an additional test of dietary adherence, we measured the RER based on indirect calorimetry, in which a decreased RER reflects enhanced oxidation of fatty acids relative to carbohydrate. Although the dietary records showed a marked difference in fat intake between groups, which is expected to differentially affect fat oxidation, we found no significant difference in RER between groups at follow-up (95% CI: −0.07, 0.01; P = 0.505) or in absolute score change from baseline (95% CI: −0.10, 0.01; P = 0.187) (Figure 2).

Physical activity level

The recorded levels of physical activity, expressed as kcal/h and based on the monthly recordings at www.diett.no, did not change from baseline to intervention (LFHC group—95% CI: −13.8, 57.1; P = 0.203; VHFLC group—95% CI: −5.38, 18.9; P = 0.285), and

TABLE 4
Changes in blood lipids and glycemic variables with the LFHC ($n = 18$) and VHFLC ($n = 20$) diets (per protocol analysis)[1]

Variable and diet	Baseline	4 wk	8 wk	12 wk[2]	Score change[3]	Time × group[4]
TGs, mmol/L						0.540
LFHC	1.45 ± 0.53	1.18 ± 0.54	1.12 ± 0.37	1.04 ± 0.38	−0.41 (−0.60, −0.21)	
VHFLC	1.52 ± 0.60	1.22 ± 0.48	1.26 ± 0.57	0.99 ± 0.51	−0.53 (−0.68, −0.37)	
NEFA, mmol/L						0.223
LFHC	0.45 ± 0.14	0.64 ± 0.20	0.71 ± 0.37	0.53 ± 0.18	+0.07 (−0.04, 0.18)	
VHFLC	0.45 ± 0.15	0.70 ± 0.19	0.66 ± 0.28	0.45 ± 0.16	−0.01 (−0.07, 0.06)	
TC, mmol/L						<0.001
LFHC	5.42 ± 1.14	4.60 ± 0.94	4.64 ± 0.95	4.46 ± 0.96	−0.96 (−1.23, −0.69)	
VHFLC	5.35 ± 1.17	5.56 ± 1.23	5.78 ± 1.22	5.48 ± 1.27	+0.13 (−0.29, 0.55)	
LDL-C, mmol/L						<0.001
LFHC	3.68 ± 1.07	2.99 ± 0.86	2.98 ± 0.89	2.90 ± 0.85	−0.78 (−1.08, −0.49)	
VHFLC	3.65 ± 1.14	3.99 ± 1.15	4.19 ± 1.18	3.91 ± 1.20	+0.26 (−0.08, 0.60)	
HDL-C, mmol/L						0.034
LFHC	1.23 ± 0.24	1.23 ± 0.29	1.23 ± 0.27	1.22 ± 0.29	−0.01 (−0.10, 0.07)	
VHFLC	1.05 ± 0.30	1.10 ± 0.21	1.13 ± 0.27	1.19 ± 0.29	+0.14 (0.06, 0.22)	
TC:HDL-C ratio						0.552
LFHC	4.54 ± 1.23	3.92 ± 1.23	3.88 ± 1.05	3.79 ± 1.03	−0.75 (−1.07, −0.43)	
VHFLC	5.47 ± 1.79	5.30 ± 1.59	5.41 ± 1.60	4.86 ± 1.53	−0.61 (−1.20, −0.02)	
TG:HDL-C ratio						0.066
LFHC	1.24 ± 0.59	1.06 ± 0.68	0.96 ± 0.44	0.91 ± 0.42	−0.33 (−0.54, −0.13)	
VHFLC	1.63 ± 0.86	1.21 ± 0.69	1.23 ± 0.74	0.92 ± 0.59	−0.70 (−0.96, −0.45)	
Glucose, mmol/L						0.013
LFHC	5.11 ± 0.46	5.00 ± 0.34	4.98 ± 0.33	4.78 ± 0.44	−0.33 (−0.53, −0.13)	
VHFLC[5]	4.94 ± 0.42	4.96 ± 0.42	5.12 ± 0.38	4.85 ± 0.42	−0.08 (−0.23, 0.06)	
Insulin, mU/L						0.886
LFHC	14.1 ± 5.47	10.5 ± 4.50	9.91 ± 5.13	8.56 ± 4.92	−5.52 (−7.72, −3.32)	
VHFLC[5]	13.3 ± 7.43	9.17 ± 6.72	11.9 ± 6.97	7.62 ± 4.31	−5.70 (−8.09, −3.31)	
INCP, nmol/L						0.308
LFHC	0.88 ± 0.22	0.73 ± 0.24	0.69 ± 0.24	0.62 ± 0.22	−0.26 (−0.35, −0.16)	
VHFLC[5]	0.89 ± 0.31	0.75 ± 0.36	0.87 ± 0.33	0.67 ± 0.29	−0.22 (−0.31, −0.12)	
HOMA2-IR						0.238
LFHC	1.95 ± 0.53	1.61 ± 0.52	1.52 ± 0.53	1.35 ± 0.51	−0.60 (−0.82, −0.37)	
VHFLC[5]	1.95 ± 0.69	1.65 ± 0.82	1.93 ± 0.81	1.47 ± 0.66	−0.48 (−0.71, −0.25)	
HOMA2-%S						0.330
LFHC	55.8 ± 19.1	70.8 ± 32.2	76.9 ± 35.1	85.4 ± 34.6	+29.6 (16.5, 42.7)	
VHFLC[5]	58.7 ± 24.1	76.1 ± 36.4	63.4 ± 33.1	82.9 ± 39.8	+24.2 (10.5, 37.9)	
HbA1c, %						0.822
LFHC	5.62 ± 0.7	—	—	5.08 ± 0.4	−0.54 (−0.75, −0.33)	
VHFLC	5.58 ± 0.5	—	—	5.06 ± 0.4	−0.52 (−0.74, −0.29)	
Glucagon, pmol/L						0.479
LFHC	30.1 ± 5.5	—	—	24.7 ± 5.6	−5.39 (−6.75, −4.04)	
VHFLC	32.3 ± 9.9	—	—	28.2 ± 7.5	−4.10 (−6.30, −1.90)	

[1] Values are raw unadjusted means ± SDs or mean score changes (95% CIs). CIs and P values are from 2-tailed analysis with the use of LMEMs with best-fitted variance and random structures, and diet, time, and diet × time as fixed-effects factors. HbA1C, glycated hemoglobin; HDL-C, HDL cholesterol; HOMA2-IR, homeostasis model assessment of insulin resistance index 2 (computer model); HOMA2-%S, homeostasis model assessment of insulin sensitivity index 2 (computer model); INCP, insulin C-peptide; LDL-C, LDL cholesterol; LFHC, low-fat, high-carbohydrate; LMEM, linear mixed-effects model; NEFA, nonesterified fatty acid; TC, total cholesterol; TG, triglyceride; VHFLC, very high–fat, low-carbohydrate.

[2] Significant group difference was found at 12 wk for TC (95% CI: 0.62, 2.01; $P = 0.001$), LDL-C (95% CI: 0.64, 1.92; $P < 0.001$), and TC/HDL-C (95% CI: 0.54, 2.36; $P = 0.003$) with the use of an adjusted LMEM (see below).

[3] Absolute mean score change from baseline to 12 wk within groups and 95% CI from an LMEM.

[4] P value for change from baseline to 12 wk between groups (time × group interaction) from an LMEM adjusted for age and baseline intake of energy, carbohydrate, added sugar, and cholesterol (see main text). The adjusted and unadjusted results did not differ in nominal significance (data not shown), except for TG:HDL-C ($P = 0.032$ in the unadjusted model). In addition, the results from the per protocol analysis were not significantly different from the intention-to-treat analysis (data not shown).

[5] One participant in the VHFLC group was identified as an influential outlier and excluded from the analyses of these variables.

TABLE 5
Differences between the LFHC (n = 18) and VHFLC (n = 20) groups in short-term responses between intermediate time points (per protocol analysis)[1]

	VHFLC vs. LFHC[2]				
Variable and diet	0–4 wk	0–8 wk	4–8 wk	8–12 wk	Score change 8–12 wk[3]
TGs, mmol/L	−0.04 (−0.29, 0.20)	+0.06 (−0.18, 0.31)	+0.10 (−0.14, 0.35)	−0.18 (−0.43, 0.06)	
LFHC					−0.08 (−0.28, 0.11)
VHFLC					−0.27 (−0.42, −0.11)
NEFA, mmol/L	+0.05 (−0.09, 0.19)	−0.06 (−0.27, 0.15)	−0.11 (−0.33, 0.12)	−0.02 (−0.24, 0.19)	
LFHC					−0.19 (−0.36, −0.01)
VHFLC					−0.21 (−0.31, −0.11)
TC, mmol/L	+1.03 (0.60, 1.47)***	+1.21 (0.68, 1.74)***	+0.18 (−0.14, 0.49)	−0.12 (−0.41, 0.17)	
LFHC					−0.18 (−0.45, 0.09)
VHFLC					−0.30 (−0.41, −0.18)
LDL-C, mmol/L	+1.03 (0.61, 1.45)***	+1.24 (0.74, 1.74)***	+0.21 (−0.10, 0.52)	−0.20 (−0.46, 0.06)	
LFHC					−0.08 (−0.36, 0.20)
VHFLC					−0.28 (−0.40, −0.15)
HDL-C, mmol/L	+0.05 (−0.08, 0.17)	+0.08 (−0.05, 0.20)	+0.03 (−0.06, 0.12)	+0.08 (−0.002, 0.15)*†	
LFHC					−0.01 (−0.10, 0.07)
VHFLC					+0.07 (0.02, 0.11)
TC:HDL-C ratio	+0.45 (−0.18, 1.07)	+0.60 (−0.10, 1.29)*†	+0.15 (−0.15, 0.46)*†	−0.45 (−0.75, −0.15)***	
LFHC					−0.09 (−0.41, 0.23)
VHFLC					−0.54 (−0.72, −0.37)
TG:HDL-C ratio	−0.23 (−0.56, 0.09)	−0.11 (−0.47, 0.24)	+0.12 (−0.14, 0.37)	−0.26 (−0.50, −0.02)*	
LFHC					−0.05 (−0.25, 0.16)
VHFLC					−0.31 (−0.45, −0.16)
Glucose, mmol/L	+0.14 (−0.11, 0.38)	+0.31 (0.07, 0.56)**	+0.18 (−0.07, 0.42)	−0.06 (−0.31, 0.18)	
LFHC					−0.20 (−0.40, 0.00)
VHFLC[4]					−0.26 (−0.41, −0.12)
Insulin, mU/L	−0.55 (−3.42, 2.31)	+2.77 (−1.06, 6.60)	+3.33 (0.66, 5.99)*	−2.95 (−5.85, −0.05)†	
LFHC					−1.34 (−3.55, 0.86)
VHFLC[4]					−4.29 (−6.45, −2.14)
INCP, nmol/L	+0.002 (−0.13, 0.14)	+0.17 (0.03, 0.30)**	+0.17 (0.03, 0.30)**	−0.13 (−0.26, 0.007)	
LFHC					−0.07 (−0.16, 0.03)
VHFLC[4]					−0.20 (−0.29, −0.10)
HOMA2-IR	+0.03 (−0.28, 0.35)	+0.41 (0.10, 0.73)**	+0.38 (0.07, 0.69)**	−0.30 (−0.61, 0.02)	
LFHC					−0.16 (−0.39, 0.06)
VHFLC[4]					−0.46 (−0.69, −0.23)
HOMA2-%S	+2.45 (−11.3, 16.2)	−16.3 (−31.7, −1.01)*	−18.8 (−34.8, −2.77)*	+11.0 (−8.77, 30.7)	
LFHC					+8.52 (−7.05, 24.1)
VHFLC[4]					+19.5 (6.82, 32.1)

[1] Values are mean score changes (95% CIs). CIs and P values are from 2-tailed analysis with the use of LMEMs with best-fitted variance and random structures, and diet, time, and diet × time as fixed effects factors. *·**·***Significantly different: *$P < 0.05$, **$P < 0.01$, ***$P \leq 0.001$. †Difference in nominal significance between adjusted and unadjusted models. HDL-C, HDL cholesterol; HOMA2-IR, homeostasis model assessment of insulin resistance index 2 (computer model); HOMA2-%S, homeostasis model assessment of insulin sensitivity index 2 (computer model); INCP, insulin C-peptide; LDL-C, LDL cholesterol; LFHC, low-fat, high-carbohydrate; LMEM, linear mixed-effects model; NEFA, nonesterified fatty acid; TC, total cholesterol; TG, triglyceride; VHFLC, very high–fat, low-carbohydrate.

[2] Group difference in absolute score change between intermediate time points and 95% CI from the unadjusted LMEM. −: greater reduction (or less increase) in the VHFLC group than in the LFHC group. +: less reduction (or greater increase) in the VHFLC group. The P value is derived from an LMEM adjusted for age and baseline intake of energy, carbohydrate, added sugar, and cholesterol (see main text). The adjusted and unadjusted results differed in nominal significance for HDL-C (8–12 wk: $P = 0.044$), insulin (8–12 wk: $P = 0.069$), and TC:HDL-C ratio (0–8 wk: $P = 0.039$; 4–8 wk: $P = 0.045$; P values from the adjusted models).

[3] Absolute mean score change from 8 to 12 wk within groups and 95% CI from an LMEM.

[4] One participant in the VHFLC group was identified as an influential outlier and excluded from the analyses of these variables.

were similar between groups at 12 wk (95% CI: −51.7, 20.3; $P = 0.753$), and in absolute score change from baseline (95% CI: −52.4, 22.6; $P = 0.401$ (data not shown).

Body composition and clinical variables

Total body weight decreased similarly on both diets, resulting in a 3.6–3.7 reduction in BMI points (Table 2, Figure 3A). Only 2 participants in each group lost <5% of their initial weight, and about one-half of them, i.e., 9 and 11 participants in the LFHC and VHFLC groups, respectively, lost ≥10%. Body composition variables were also assessed every 4 wk by bioimpedance. Because fat-free mass and skeletal muscle mass only decreased by 2–3% from baseline, the body weight reduction on both diets could largely be ascribed to a 26–30% decrease in body fat mass, related to a 9–11% decrease in WC and a 28–30% reduction in visceral fat area (Table 2, Supplemental Table 4). The participants also showed a highly significant reduction in blood pressure (Table 2).

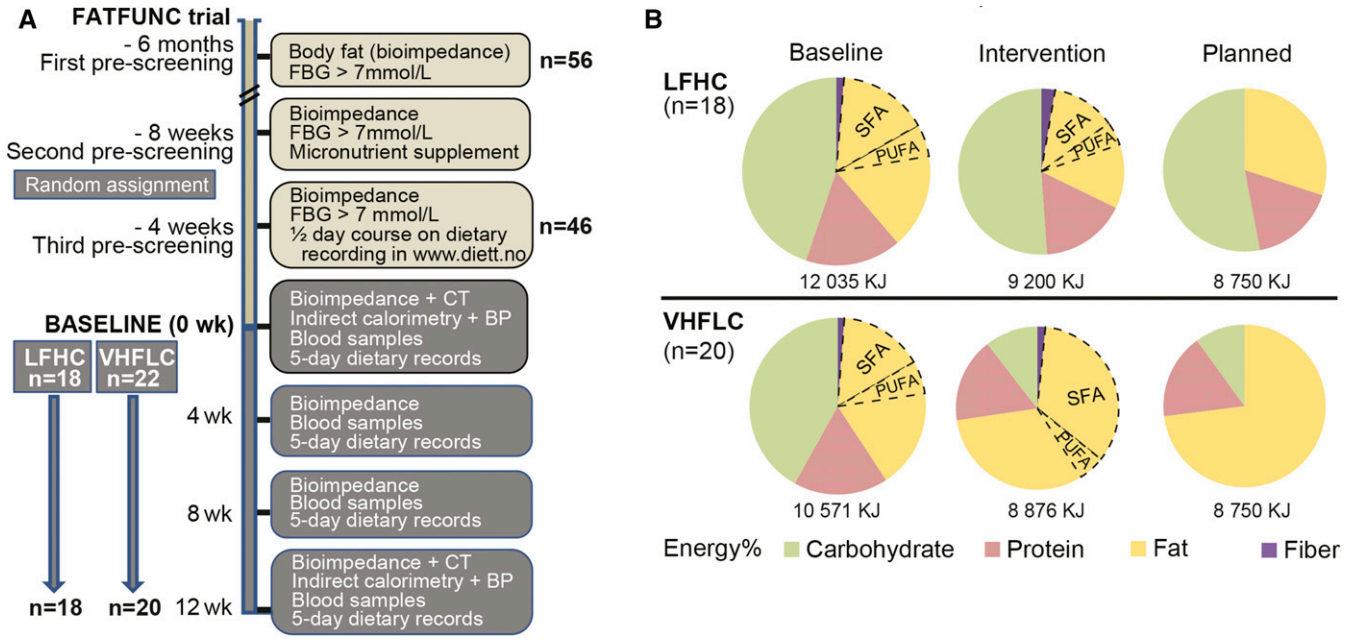

FIGURE 1 Overview of the study. Recruited candidates for the study (n = 56) were screened 6 mo before the intervention (A). Those qualifying for the study after the prescreenings (n = 46) were randomly assigned to an LFHC or VHFLC diet, and 38 completed the study. Macronutrient composition at baseline and during the intervention (B). BP, blood pressure; CT, computed tomography; Energy%, percentage of energy; FATFUNC, Dietary Fat and Carbohydrates and Fat Tissue Function in Abdominally Obese Men; FBG, fasting blood gluose; LFHC, low-fat, high-carbohydrate; VHFLC, very high–fat, low-carbohydrate.

We further quantified depot-specific changes in fat mass by CT imaging. The volume of total abdominal fat decreased on average by 21–26% with the respective diets, with no significant group differences in absolute or relative score changes from baseline to 12 wk (Table 3, Supplemental Table 4). Visceral fat and subcutaneous fat volume decreased on average by 21–27% and 21–25%, respectively (Figure 3B, Table 3, Supplemental Table 4). There were no significant group differences in absolute or relative score changes from baseline in the visceral-to-total abdominal fat volume ratio (Table 3, Supplemental Table 4). Finally, hepatic volume decreased equally with both diets (Figure 3C), as did pericardial fat volume (Table 3, Supplemental Table 4). The ITT analysis showed the same results as the PP analysis in terms of nominal significance for all anthropometric variables (Supplemental Tables 6, 7).

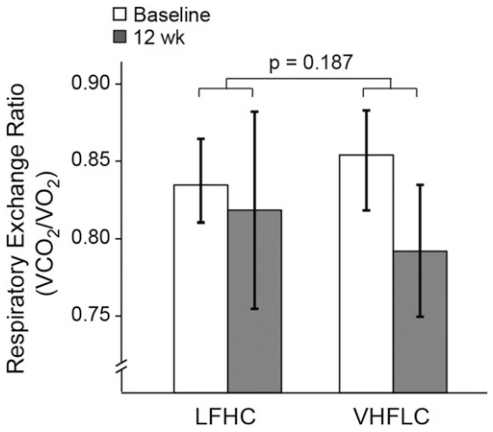

FIGURE 2 RER after 12 wk of intervention. The gas exchange from nutrient substrate catabolized for energy was assessed with the use of RER; ratio of CO_2 produced to O_2 consumed. RER for carbohydrate (glucose) oxidation produces an equal number of CO_2 molecules to O_2 molecules consumed, i.e., RER = 1. Generally, a value of 0.70 represents the RER for fat, with values ranging between 0.69 and 0.73, depending on the carbon-chain length. Data are presented as means ± SDs with error bars. The group difference in score changes from baseline was analyzed by a linear mixed-effects model adjusted for age and baseline intake of energy, carbohydrate, added sugar, and cholesterol (see main text). Within-group score changes (95% CI)—LFHC (n = 18): −0.02 (−0.05, 0.02); VHFLC (n = 20): −0.06 (−0.10, −0.03). LFHC, low-fat, high-carbohydrate; RER, respiratory exchange ratio; VCO_2, volume (L/min) carbon dioxide produced; VO_2, volume (L/min) oxygen consumed; VHFLC, very high–fat, low-carbohydrate.

To assess whether the diet-induced decreases in visceral fat might have been due to reduced energy intake, we correlated the relative change in total energy intake with the relative change in visceral fat volume. There were no significant correlations, either for the groups separately (Figure 3D) or for all participants combined (r = 0.14, 95% BCa CI: −0.29, 0.48; P = 0.433). This lack of correlation with energy intake was also observed for subcutaneous fat volume (r = 0.11, 95% BCa CI: −0.27, 0.50; P = 0.538), hepatic lipid content (liver-spleen ratio: r = −0.22, 95% BCa CI: −0.47, 0.09; P = 0.221) and pericardial fat volume (r = 0.08, 95% BCa CI: −0.36, 0.45; P = 0.658). Several of the participants recorded an almost unchanged or even an increased total energy intake compared with baseline, and still showed substantial reductions in body weight (data not shown), visceral fat volume (Figure 3D), and hepatic volume (data not shown). Of note, those with the highest hepatic lipid content at baseline experienced the largest reduction in hepatic fat on either diet, based on correlating liver-spleen ratio at baseline with the relative change during intervention (r = −0.86, 95% BCa CI: −0.92, −0.66; P < 0.001).

Biochemical variables

Fasting glucose concentrations showed a significant group difference in change from baseline to 12 wk, with a significant

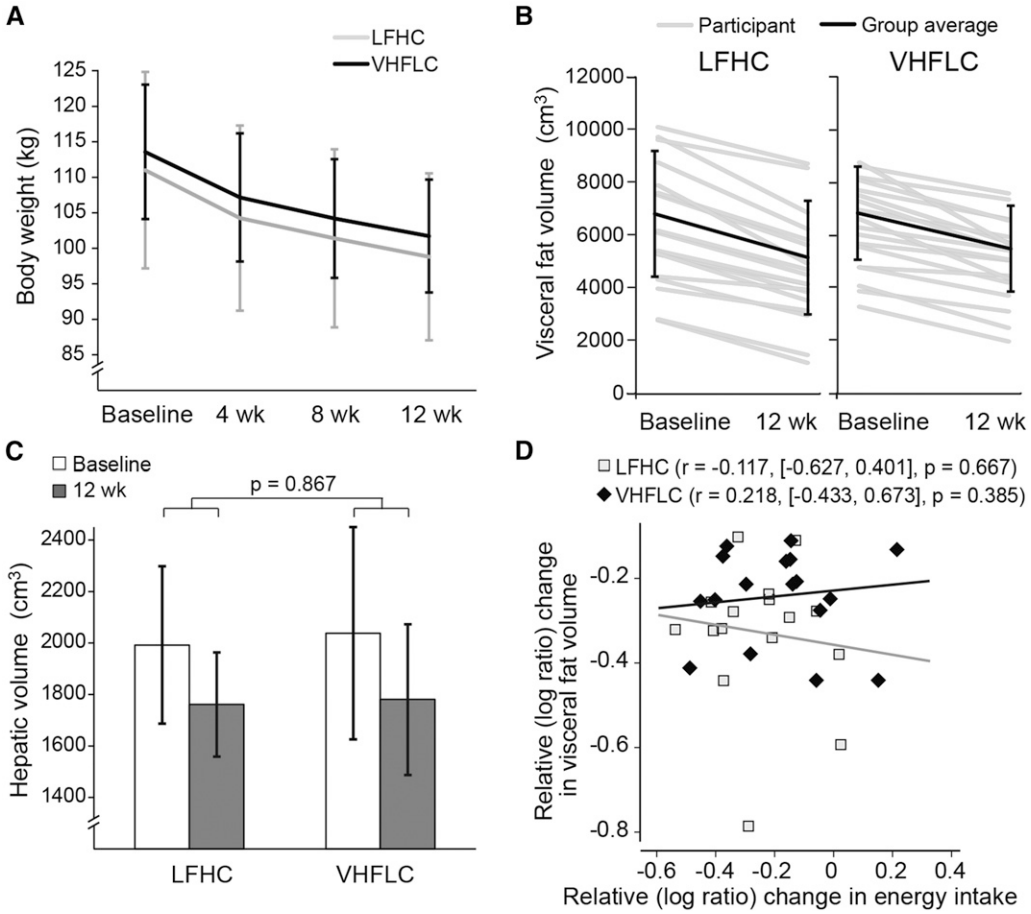

FIGURE 3 Diet-induced changes in body weight, visceral fat, and hepatic volume. Mean body weight reductions with the LFHC (n = 18) and VHFLC (n = 20) diets (A). Individual and group mean changes in visceral fat volume measured by CT imaging (B). The 2 groups showed similar reductions in hepatic volume measured by CT imaging and analyzed by a linear mixed-effects model adjusted for age and baseline intake of energy, carbohydrate, added sugar, and cholesterol (see main text) (C). The degree of change in total energy intake from baseline to 12 wk did not correlate with the percentage change in visceral fat volume measured by CT imaging (D). LFHC n = 18, VHFLC n = 18 (B–D). Means ± SDs with error bars are shown for the different time points (A–C). Within-group score changes are shown in Supplemental Table 4. CT, computed tomography; LFHC, low-fat, high-carbohydrate; VHFLC, very high–fat, low-carbohydrate.

reduction only in the LFHC group (Table 4, Figure 4A). Circulating concentrations of insulin, INCP, glycated hemoglobin, glucagon, and triglycerides, together with HOMA2-IR and homeostasis model assessment of insulin sensitivity index 2, were significantly reduced from baseline to 12 wk with both diets, with no significant group differences in change across time (Table 4, Figure 4B and C). Both diets also similarly affected circulating concentrations of NEFAs, with an initial significant increase from baseline to 4 wk, and a return to baseline concentrations after significant reductions between 8 and 12 wk (Table 4, Figure 4D).

Significant group differences were found in change from baseline to 12 wk for TC, LDL cholesterol (decreased in the LFHC group only), and HDL cholesterol (increased in the VHFLC group only) (Table 4, Figure 4E and F), but not for the TC-to–HDL cholesterol or triglyceride-to–HDL cholesterol ratios in the adjusted LMEMs when controlling for potentially clinically important differences at baseline in dietary intake, i.e., energy, carbohydrate, added sugar, and cholesterol (Table 1). These results did not differ in nominal significance from the unadjusted models, except for the triglyceride-to–HDL cholesterol ratio (95% CI: −0.71, −0.03; P = 0.032) because of a higher reduction in the VHFLC group.

In line with reduced liver fat, both groups showed significant reductions from baseline in circulating concentrations of alanine aminotransferase, aspartate aminotransferase, γ-glutamyltranspeptidase and lactate dehydrogenase, with no group differences in score changes from baseline for any of these or other measured variables related to liver function (Supplemental Table 5). No changes from baseline were observed for circulating albumin, bile acids, or creatine kinase, whereas alkaline phosphatase and bilirubin decreased significantly (groups combined) (Supplemental Table 5).

In summary, for all of the primary outcomes (i.e., anthropometric measures) and biochemical variables except glucose and cholesterol (TC, LDL cholesterol, and HDL cholesterol), we found no significant group differences in the overall score changes from baseline to 12 wk (time × diet interaction) (Tables 2–4, Supplemental Table 5).

Interestingly, although the anthropometric measurements typically showed a gradual decrease over time (Table 2), with no significant group differences for any of the 4-wk intervals (data not shown), many of the biochemical variables showed nonlinear changes throughout the intervention (Table 4). The groups showed significantly different short-term changes in TC and LDL cholesterol during the first half of the intervention (0–4 and 0–8 wk), in

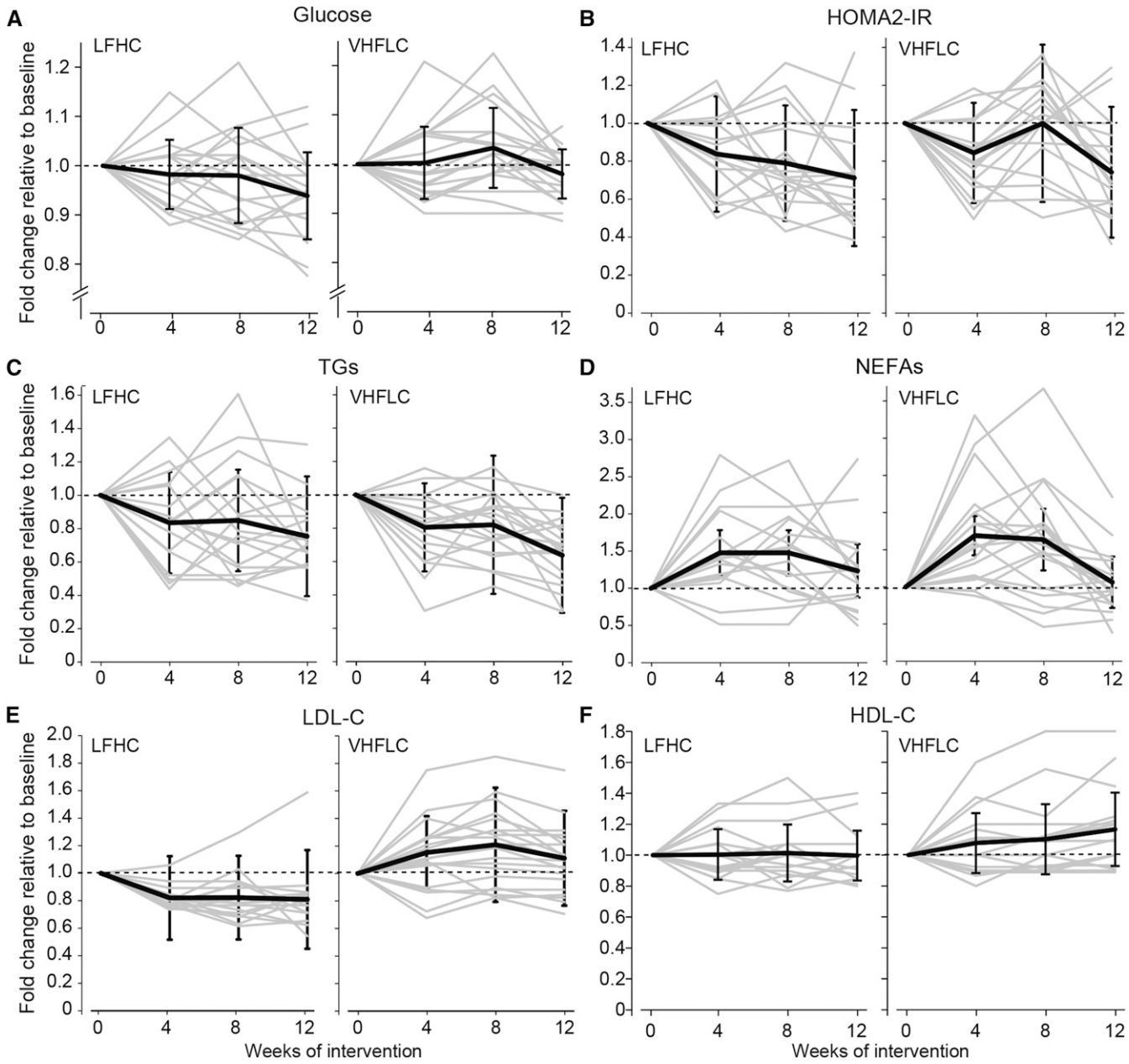

FIGURE 4 Individual changes in glucose (A), HOMA2-IR (B), TGs (C), NEFA (D), LDL-C (E), and HDL-C (F) with the diets relative to baseline. Values at each time point were normalized to the baseline value for each participant individually (light gray lines). The group mean fold change from baseline was in turn calculated. Thus, the figures do not reflect individual differences in absolute concentrations of the variables (variance shown in Table 4). Fasting glucose, TGs, NEFA, HDL-C, and LDL-C were measured in serum. HOMA2-IR, a surrogate measure of insulin resistance, was calculated on the basis of serum fasting insulin C-peptide and glucose. Data are shown as mean folds ± SDs. LFHC n = 18, VHFLC n = 20. P values for group differences in score changes are shown in Tables 4 and 5. HDL-C, HDL cholesterol; HOMA2-IR, homeostasis model assessment of insulin resistance index 2; LDL-C, LDL cholesterol; LFHC, low-fat, high-carbohydrate; NEFA, nonesterified fatty acid; TG, triglyceride; VHFLC, very high–fat, low-carbohydrate.

glucose and insulin and INCP during the middle (0–8 and 4–8 wk), and in HDL cholesterol during the last part (8–12 wk) (Tables 4 and 5). TC and LDL cholesterol showed an early decrease with the LFHC diet and an early increase with the VHFLC diet, whereas HDL cholesterol increased only with the VHFLC diet between 8 and 12 wk (Figure 4E and F, Tables 4 and 5). In contrast, the changes in triglyceride and NEFA concentrations were similar for the 2 groups across all time points (Figure 4C and D, Table 5). Interestingly, the TC-to–HDL cholesterol and triglyceride-to–HDL cholesterol ratios significantly improved only in the VHFLC group between 8 and 12 wk (Table 5) owing to simultaneous reductions in TC and triglycerides and an increase in HDL cholesterol in this group (Table 4). Moreover, glucose and insulin concentrations and HOMA2-IR, increased between 4 and 8 wk, followed by reductions between 8 and 12 wk (Figure 4E and F, Tables 4 and 5). Of note, all biochemical variables improved from 8 to 12 wk in the VHFLC group after increasing in ≥1 of the preceding intervals (Tables 4 and 5). Except for NEFAs (Figure 4D), this biochemical pattern was not found for any variable in the LFHC group (Tables 4 and 5). It should be

noted, however, that the responses of several biochemical markers varied between participants in both groups (Figure 4).

DISCUSSION

In the present dietary intervention study, we found a marked reduction in visceral fat mass and improvements in most of the measured clinical variables related to metabolic function, independent of a sharp dichotomy in the fat-to-carbohydrate intake ratio. Our results are in line with systematic reviews and meta-analyses of epidemiologic and dietary intervention studies, which overall do not support a causal connection between SFA intake per se and risk of metabolic syndrome, fatty liver, or CVD, regardless of the effects on LDL cholesterol (16, 21, 24, 28, 38–42).

Our study has some important characteristics. First, the top 10 energy-contributing foods were the same for both groups, varying mainly in quantity. As in animal diet studies, we believe this principle of controlling for food types should be standard in any comparison of diets. Second, we obtained complete information on the macronutrient content of every meal and food item the participants recorded during the intervention, which, along with our Internet-based meal planning system, helped to decrease uncertainty in the nutrient intake estimates. Moreover, a special feature of our randomized controlled study was the very high intake of total and saturated fat while controlling for intake of energy, protein, and PUFAs, as well as food types. Results of previous studies that replaced PUFAs with SFAs or vice versa (24, 43) partly might be ascribed to changes in PUFA intake. Controlled studies have shown that PUFAs modulate metabolic function, inflammatory response, and risk of CVD (44). Finally, the recorded daily energy intake at baseline and during the intervention period in the present study was substantially higher than in most similar dietary intervention trials (mostly 6500–7500 kJ/d for men) (45, 46).

Importantly, there was no significant correlation between relative change in energy intake and relative changes in subcutaneous, visceral, hepatic, or pericardial fat, and the overall reductions in body fat were greater in our trial than were those in previous studies with more energy-restricted diets (45, 47). Of note, the energy content of our high-fat, low-carbohydrate diet was likely higher than would have been the case with ad libitum consumption of such a diet (16, 45, 48), and it still resulted in marked metabolic improvements. Also, energy restriction cannot explain the nonlinear changes we observed in biochemical variables across the different 4-wk intervals, highlighting the involvement of macronutrient-dependent adaptive metabolic mechanisms beyond a static effect of energy intake. Importantly, loss of fat-free and skeletal muscle mass with both the VHFLC and LFHC diets was negligible (on average $<1.5\%$ of body weight at baseline). These results indicate that improving dietary pattern and food quality within a normal caloric intake of 8500–10500 kJ/d can lead to a substantial reduction in fat mass while avoiding excess loss of muscle mass, regardless of total and saturated fat intake.

Our results do not support the main finding in a recent metabolic ward study showing that a very low–fat diet (71% of energy carbohydrate and 8% of energy fat) promotes greater body fat loss after 6 d than an isoenergetic (1918 kcal) and isoproteinic (21% of energy protein) reduced-carbohydrate diet (29% of energy carbohydrate and 50% of energy fat) (49). The authors calculated that the mean 6-d cumulative fat loss differed between the groups by 218 g (i.e., 36 g/d), which would correspond to an $\sim$3 kg difference in body fat over the 12-wk duration of our study. However, we found no significant group differences in fat loss. The discrepancy may be related to, e.g., the degree of fat and energy restriction and different food profiles, as well as the uncertainty of linear predictions in estimating long-term responses.

An important aspect of our study was the repeated measurements after 4, 8, and 12 wk of intervention, providing insight into distinct short-term dynamics of adaptation to diets high in carbohydrate or fat. Previous observations have suggested that $\geq$3–6 wk of metabolic adaptation (keto-adaptation) may be needed when switching from carbohydrate to fat as the primary energy source (50, 51). In line with our results, studies of high-fat, low-carbohydrate diets that are $<$2 mo in duration therefore are likely to be misleading with respect to the actual health effects. Moreover, a study beyond 3 mo is needed to determine whether the high-fat, low-carbohydrate diet would be more beneficial for body composition than would the high-carbohydrate diet in the longer term. Ideally, future studies should be performed with a feeding component for better control of food intake.

The diet-specific temporal pattern we observed for glucose and insulin responses may reflect group-specific differences in postprandial and diurnal concentrations of NEFA, i.e., the NEFA fluxes, which we did not measure. It was shown previously that fasting concentrations of NEFA were not different between groups consuming a high-carbohydrate or a high-fat diet, whereas postprandial and diurnal NEFA concentrations were significantly higher in the high-fat diet group (52). NEFA fluxes may determine glucose and insulin concentrations, as well as insulin sensitivity (53–55).

A characteristic of our study is that the 2 groups consumed differing amounts of the same food types, unlike in many other studies, in which groups also differed with respect to the foods consumed. The similar food profiles in our study might explain at least some of the group similarity in metabolic benefits (e.g., by minimizing processed foods, improving micronutrient status, microbiota, or other factors), consistent with previous reports on effects of food profile beyond energy restriction (15, 56). For example, replacing processed foods with whole foods (e.g., replacing high-glycemic foods with lower-glycemic foods) can reduce the risk of coronary artery disease (57), promote weight loss and enhanced body composition (15), and improve key metabolic functions, such as insulin sensitivity (10, 58).

The marked improvements in most risk-related variables with both diets, on the one hand, and a lack of reduction in LDL cholesterol with the VHFLC diet on the other, raises the question of whether an isolated risk assessment based on LDL cholesterol alone is justified. Increased visceral fat remains a strong, independent risk factor for insulin resistance, T2D, and CVD, also after adjusting for circulating cholesterol concentrations (59, 60). In line with these data and the present results, there is compelling evidence against an important contribution of total and saturated fat intake per se to visceral and ectopic fat accumulation and risk of CVD (22, 24, 28, 42). Moreover, the dyslipidemia that is typical with obesity is characterized by increased concentration of triglycerides and low HDL cholesterol (61), the ratio of which is an independent CVD risk marker (62), and which clearly improved with the VHFLC diet. It should also be noted that the CVD risk associated with LDL cholesterol has a considerable genetic component (63). Finally, contrary to previous reports that a high intake of SFAs may increase LDL cholesterol (22, 23), and consistent with a feeding study showing a limited impact of dietary SFAs on plasma SFAs (64), we

observed no significant increase in LDL cholesterol with the VHFLC diet, which on average doubled SFA intake from baseline.

Limitations of the present study include a relatively low number of participants, no control group consuming their regular diet, an increased risk of false positives with a large number of statistical tests, and an open-label design that may have introduced some bias. Although great effort was made to obtain representative dietary records, uncertainty is expected, especially for the baseline estimates to which the interventions were compared. A larger number of participants might have revealed additional significant differences between the groups. The exclusion of 4 of the randomly assigned participants in our main analysis may have introduced some bias, whereas including these 4 in the ITT analysis increased the number of outliers, particularly in the VHFLC group. Nonetheless, the ITT and PP analyses showed similar and significant improvements from baseline to 12 wk for the trial's primary outcomes (visceral fat mass and body composition) and the secondary outcomes (e.g., biochemical variables). Furthermore, the present study was designed to compare short-term dynamics in response to the VHFLC and LFHC diets, and possible longer-term differential responses were not addressed. Finally, we studied a relatively homogenous group of middle-aged men without diabetes, which may have decreased interindividual variability, but may also limit the generalizability. For example, VHFLC diets may have better effect in more insulin-resistant individuals (65).

In summary, we found similar responses to highly standardized LFHC and VHFLC diets with respect to intra-abdominal fat mass, hepatic lipid content, pericardial fat volume, and components of metabolic syndrome. Our study cautions against extrapolating short-term (1–2 mo) metabolic responses to longer-term effects of macronutrients on cardiometabolic risk.

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The authors' responsibilities were as follows—VLV, JL-B, ER, OKN, JVS, OAG, SND, and GM: designed the trial; JL-B: designed all the recipes and developed the electronic recipe booklet and application for iPhone and iPad; VLV, JL-B, ØE, and ER: carried out visits and collected samples; OAG: analyzed the nonesterified fatty acids in the blood samples; ØE, THL, and JEN: collected and analyzed the computed tomography data; JVS, OAG, SND, and GM: supervised the trial; VLV, JL-B, and ØE: performed the data analyses; VLV, JL-B, and SND: wrote the manuscript with input from all authors; and all authors: read and approved the final manuscript. None of the authors reported a conflict of interest related to the study.

REFERENCES

1. Guh DP, Zhang W, Bansback N, Amarsi Z, Birmingham CL, Anis AH. The incidence of co-morbidities related to obesity and overweight: a systematic review and meta-analysis. BMC Public Health 2009;9:88.
2. Johnson AM, Olefsky JM. The origins and drivers of insulin resistance. Cell 2013;152:673–84.
3. Kim SH, Reaven G. Obesity and insulin resistance: an ongoing saga. Diabetes 2010;59:2105–6.
4. Kaess BM, Pedley A, Massaro JM, Murabito J, Hoffmann U, Fox CS. The ratio of visceral to subcutaneous fat, a metric of body fat distribution, is a unique correlate of cardiometabolic risk. Diabetologia 2012;55:2622–30.
5. Tchernof A, Després J-P. Pathophysiology of human visceral obesity: an update. Physiol Rev 2013;93:359–404.
6. Kotronen A, Yki-Järvinen H, Sevastianova K, Bergholm R, Hakkarainen A, Pietiläinen KH, Juurinen L, Lundbom N, Sørensen TIA. Comparison of the relative contributions of intra-abdominal and liver fat to components of the metabolic syndrome. Obesity (Silver Spring) 2011;19:23–8.
7. Swinburn BA, Sacks G, Hall KD, McPherson K, Finegood DT, Moodie ML, Gortmaker SL. The global obesity pandemic: shaped by global drivers and local environments. Lancet 2011;378:804–14.
8. Hu T, Mills KT, Yao L, Demanelis K, Eloustaz M, Yancy WS, Kelly TN, He J, Bazzano LA. Effects of low-carbohydrate diets versus low-fat diets on metabolic risk factors: a meta-analysis of randomized controlled clinical trials. Am J Epidemiol 2012;176:S44–54.
9. Johnston BC, Kanters S, Bandayrel K, Wu P, Naji F, Siemieniuk RA, Ball GD, Busse JW, Thorlund K, Guyatt G, et al. Comparison of weight loss among named diet programs in overweight and obese adults: a meta-analysis. JAMA 2014;312:923–33.
10. Bhupathiraju SN, Tobias DK, Malik VS, Pan A, Hruby A, Manson JE, Willett WC, Hu FB. Glycemic index, glycemic load, and risk of type 2 diabetes: results from 3 large US cohorts and an updated meta-analysis. Am J Clin Nutr 2014;100:218–32.
11. Juanola-Falgarona M, Salas-Salvadó J, Ibarrola-Jurado N, Rabassa-Soler A, Díaz-López A, Guasch-Ferré M, Hernández-Alonso P, Balanza R, Bulló M. Effect of the glycemic index of the diet on weight loss, modulation of satiety, inflammation, and other metabolic risk factors: a randomized controlled trial. Am J Clin Nutr 2014;100:27–35.
12. Raben A. Glycemic index and metabolic risks: how strong is the evidence? Am J Clin Nutr 2014;100:1–3.
13. Cummings JH, Stephen AM. Carbohydrate terminology and classification. Eur J Clin Nutr 2007;61:S5–18.
14. Stanhope KL, Havel PJ. Fructose consumption: considerations for future research on its effects on adipose distribution, lipid metabolism, and insulin sensitivity in humans. J Nutr 2009;139:1236S–41S.
15. Thomas DE, Elliott EJ, Baur L. Low glycaemic index or low glycaemic load diets for overweight and obesity. Cochrane Database Syst Rev 2007;(3):CD005105.
16. Noakes M, Foster P, Keogh J, James A, Mamo J, Clifton P. Comparison of isocaloric very low carbohydrate/high saturated fat and high carbohydrate/low saturated fat diets on body composition and cardiovascular risk. Nutr Metab (Lond) 2006;3:7.
17. Noto H, Goto A, Tsujimoto T, Noda M. Low-carbohydrate diets and all-cause mortality: a systematic review and meta-analysis of observational studies. PLoS One 2013;8:e55030.
18. Bray GA, Krauss RM. Overfeeding of polyunsaturated versus saturated fatty acids reduces ectopic fat. Diabetes 2014;63:2222–4.
19. Rosqvist F, Iggman D, Kullberg J, Cedernaes J, Johansson HE, Larsson A, Johansson L, Ahlstrom H, Arner P, Dahlman I, et al. Overfeeding polyunsaturated and saturated fat causes distinct effects on liver and visceral fat accumulation in humans. Diabetes 2014;63:2356–68.
20. Leamy AK, Egnatchik RA, Young JD. Molecular mechanisms and the role of saturated fatty acids in the progression of non-alcoholic fatty liver disease. Prog Lipid Res 2013;52:165–74.
21. Michas G, Micha R, Zampelas A. Dietary fats and cardiovascular disease: putting together the pieces of a complicated puzzle. Atherosclerosis 2014;234:320–8.
22. Mozaffarian D. Saturated fatty acids and type 2 diabetes: more evidence to re-invent dietary guidelines. Lancet Diabetes Endocrinol 2014;2:770–2.
23. Poudyal H, Brown L. Should the pharmacological actions of dietary fatty acids in cardiometabolic disorders be classified based on biological or chemical function? Prog Lipid Res 2015;59:172–200.
24. Micha R, Mozaffarian D. Saturated fat and cardiometabolic risk factors, coronary heart disease, stroke, and diabetes: a fresh look at the evidence. Lipids 2010;45:893–905.
25. Snel M, Jonker JT, Schoones J, Lamb H, de Roos A, Pijl H, Smit JWA, Meinders AE, Jazet IM. Ectopic fat and insulin resistance: pathophysiology and effect of diet and lifestyle interventions. Int J Endocrinol 2012;2012:983814.
26. Schugar RC, Crawford PA. Low-carbohydrate ketogenic diets, glucose homeostasis, and nonalcoholic fatty liver disease. Curr Opin Clin Nutr Metab Care 2012;15:374–80.
27. Wang CCL, Adochio RL, Leitner JW, Abeyta IM, Draznin B, Cornier M-A. Acute effects of different diet compositions on skeletal muscle insulin signalling in obese individuals during caloric restriction. Metabolism 2013;62:595–603.
28. Siri-Tarino PW, Chiu S, Bergeron N, Krauss RM. Saturated fats versus polyunsaturated fats versus carbohydrates for cardiovascular disease prevention and treatment. Annu Rev Nutr 2015;35:517–43.

29. Krieger JW, Sitren HS, Daniels MJ, Langkamp-Henken B. Effects of variation in protein and carbohydrate intake on body mass and composition during energy restriction: a meta-regression. Am J Clin Nutr 2006;83:260–74.
30. McArdle WD, Katch FI, Katch VL. Exercise physiology: nutrition, energy, and human performance. 8th ed. Philadelphia: Lippincott Williams & Wilkins; 2015. (Appendix F: Energy expenditure in household, occupational, recreational, and sport activities.).
31. Levy JC, Matthews DR, Hermans MP. Correct homeostasis model assessment (HOMA) evaluation uses the computer program. Diabetes Care 1998;21:2191–2.
32. Wallace TM, Levy JC, Matthews DR. Use and abuse of HOMA modeling. Diabetes Care 2004;27:1487–95.
33. DeLany JP, Windhauser MM, Champagne CM, Bray GA. Differential oxidation of individual dietary fatty acids in humans. Am J Clin Nutr 2000;72:905–11.
34. Matarese LE. Indirect calorimetry: technical aspects. J Am Diet Assoc 1997;97:S154–60.
35. Shores NJ, Link K, Fernandez A, Geisinger KR, Davis M, Nguyen T, Sawyer J, Rudel L. Non-contrasted computed tomography for the accurate measurement of liver steatosis in obese patients. Dig Dis Sci 2011;56:2145–51.
36. Maurovich-Horvat P, Massaro J, Fox CS, Moselewski F, O'Donnell CJ, Hoffmann U. Comparison of anthropometric, area- and volume-based assessment of abdominal subcutaneous and visceral adipose tissue volumes using multi-detector computed tomography. Int J Obes (Lond) 2007;31:500–6.
37. Kvist H, Chowdhury B, Sjöström L, Tylén U, Cederblad A. Adipose tissue volume determination in males by computed tomography and 40K. Int J Obes 1988;12:249–66.
38. Chowdhury R, Warnakula S, Kunutsor S, Crowe F, Ward HA, Johnson L, Franco OH, Butterworth AS, Forouhi NG, Thompson SG, et al. Association of dietary, circulating, and supplement fatty acids with coronary risk: a systematic review and meta-analysis. Ann Intern Med 2014;160:398–406.
39. de Souza RJ, Mente A, Maroleanu A, Cozma AI, Ha V, Kishibe T, Uleryk E, Budylowski P, Schünemann H, Beyene J, et al. Intake of saturated and trans unsaturated fatty acids and risk of all cause mortality, cardiovascular disease, and type 2 diabetes: systematic review and meta-analysis of observational studies. BMJ 2015;351:h3978.
40. Mensink RP, Zock PL, Kester AD, Katan MB. Effects of dietary fatty acids and carbohydrates on the ratio of serum total to HDL cholesterol and on serum lipids and apolipoproteins: a meta-analysis of 60 controlled trials. Am J Clin Nutr 2003;77:1146–55.
41. Puaschitz NG, Strand E, Norekvål TM, Dierkes J, Dahl L, Svingen GFT, Assmus J, Schartum-Hansen H, Øyen J, Pedersen EKR, et al. Dietary intake of saturated fat is not associated with risk of coronary events or mortality in patients with established coronary artery disease. J Nutr 2015;145:299–305.
42. Siri-Tarino PW, Sun Q, Hu FB, Krauss RM. Meta-analysis of prospective cohort studies evaluating the association of saturated fat with cardiovascular disease. Am J Clin Nutr 2010;91:535–46.
43. Sofi F, Casini A. Mediterranean diet and non-alcoholic fatty liver disease: new therapeutic option around the corner? World J Gastroenterol 2014;20:7339–46.
44. Buckley JD, Howe PRC. Long-chain omega-3 polyunsaturated fatty acids may be beneficial for reducing obesity: a review. Nutrients 2010; 2:1212–30.
45. Bazzano LA, Hu T, Reynolds K, Yao L, Bunol C, Liu Y, Chen C-S, Klag MJ, Whelton PK, He J. Effects of low-carbohydrate and low-fat diets: a randomized trial. Ann Intern Med 2014;161:309–18.
46. Brinkworth GD, Noakes M, Buckley JD, Keogh JB, Clifton PM. Long-term effects of a very-low-carbohydrate weight loss diet compared with an isocaloric low-fat diet after 12 mo. Am J Clin Nutr 2009;90:23–32.
47. Chaston TB, Dixon JB. Factors associated with percent change in visceral versus subcutaneous abdominal fat during weight loss: findings from a systematic review. Int J Obes (Lond) 2008;32:619–28.
48. Guldbrand H, Dizdar B, Bunjaku B, Lindström T, Bachrach-Lindström M, Fredrikson M, Östgren CJ, Nystrom FH. In type 2 diabetes, randomisation to advice to follow a low-carbohydrate diet transiently improves glycaemic control compared with advice to follow a low-fat diet producing a similar weight loss. Diabetologia 2012;55:2118–27.
49. Hall KD, Bemis T, Brychta R, Chen KY, Courville A, Crayner EJ, Goodwin S, Guo J, Howard L, Knuth ND, et al. Calorie for calorie, dietary fat restriction results in more body fat loss than carbohydrate restriction in people with obesity. Cell Metab 2015;22:427–36.
50. Phinney SD. Ketogenic diets and physical performance. Nutr Metab (Lond) 2004;1:2.
51. Simi B, Sempore B, Mayet MH, Favier RJ. Additive effects of training and high-fat diet on energy metabolism during exercise. J Appl Physiol (1985) 1991;71:197–203.
52. Hernandez TL, Sutherland JP, Wolfe P, Allian-Sauer M, Capell WH, Talley ND, Wyatt HR, Foster GD, Hill JO, Eckel RH. Lack of suppression of circulating free fatty acids and hypercholesterolemia during weight loss on a high-fat, low-carbohydrate diet. Am J Clin Nutr 2010; 91:578–85.
53. Girousse A, Tavernier G, Valle C, Moro C, Mejhert N, Dinel A-L, Houssier M, Roussel B, Besse-Patin A, Combes M, et al. Partial inhibition of adipose tissue lipolysis improves glucose metabolism and insulin sensitivity without alteration of fat mass. PLoS Biol 2013;11: e1001485.
54. Karpe F, Dickmann JR, Frayn KN. Fatty acids, obesity, and insulin resistance: time for a reevaluation. Diabetes 2011;60:2441–9.
55. Perry RJ, Camporez JP, Kursawe R, Titchenell PM, Zhang D, Perry CJ, Jurczak MJ, Abudukadier A, Han MS, Zhang XM, et al. Hepatic acetyl CoA links adipose tissue inflammation to hepatic insulin resistance and type 2 diabetes. Cell 2015;160:745–58.
56. Naude CE, Schoonees A, Senekal M, Young T, Garner P, Volmink J. Low carbohydrate versus isoenergetic balanced diets for reducing weight and cardiovascular risk: a systematic review and meta-analysis. PLoS One 2014;9:e100652.
57. Dong J-Y, Zhang Y-H, Wang P, Qin L-Q. Meta-analysis of dietary glycemic load and glycemic index in relation to risk of coronary heart disease. Am J Cardiol 2012;109:1608–13.
58. Karschin J, Lagerpusch M, Enderle J, Eggeling B, Müller MJ, Bosy-Westphal A. Endocrine determinants of changes in insulin sensitivity and insulin secretion during a weight cycle in healthy men. PLoS One 2015;10:e0117865.
59. Després JP. Is visceral obesity the cause of the metabolic syndrome? Ann Med 2006;38:52–63.
60. Pouliot M-C, Després J-P, Nadeau A, Moorjani S. Prud'Homme D, Lupien PJ, Tremblay A, Bouchard C. Visceral obesity in men. Associations with glucose tolerance, plasma insulin, and lipoprotein levels. Diabetes 1992;41:826–34.
61. Giannini C, Santoro N, Caprio S, Kim G, Lartaud D, Shaw M, Pierpont B, Weiss R. The triglyceride-to-HDL cholesterol ratio: association with insulin resistance in obese youths of different ethnic backgrounds. Diabetes Care 2011;34:1869–74.
62. Wan K, Zhao J, Huang H, Zhang Q, Chen X, Zeng Z, Zhang L, Chen Y. The association between triglyceride/high-density lipoprotein cholesterol ratio and all-cause mortality in acute coronary syndrome after coronary revascularization. PLoS One 2015;10:e0123521.
63. Ridker PM. LDL cholesterol: controversies and future therapeutic directions. Lancet 2014;384:607–17.
64. Forsythe CE, Phinney SD, Feinman RD, Volk BM, Freidenreich D, Quann E, Ballard K, Puglisi MJ, Maresh CM, Kraemer WJ, et al. Limited effect of dietary saturated fat on plasma saturated fat in the context of a low carbohydrate diet. Lipids 2010;45:947–62.
65. Cornier MA, Donahoo WT, Pereira R, Gurevich I, Westergren R, Enerback S, Eckel PJ, Goalstone ML, Hill JO, Eckel RH, et al. Insulin sensitivity determines the effectiveness of dietary macronutrient composition on weight loss in obese women. Obes Res 2005;13:703–9.