Title

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Journal

Current Developments in Nutrition, 6(5)

ISSN

2475-2991

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Publication Date

2022-05-01

DOI

10.1093/cdn/nzac083

Peer reviewed

ORIGINAL RESEARCH

CURRENT DEVELOPMENTS IN NUTRITION

Nutrition in Health and Disease

Waking Salivary Cortisol Associated with Magnitude of Cholesterol Reduction in Women Fed a Healthy Whole-Food Diet for 8 Weeks

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ABSTRACT

Background: Diet and cortisol are independently linked to cardiometabolic function and health, but underlying alterations in circulating cortisol may influence beneficial cardiometabolic effects of consuming a healthy diet.

Objective: This study was a secondary analysis to examine whether baseline concentrations of waking salivary cortisol interacted with 8-wk whole-food diet interventions to affect cardiometabolic outcomes.

Methods: A randomized, double-blind, controlled 8-wk diet intervention was conducted in 44 participants. The trial was conducted at the Western Human Nutrition Research Center in Davis, California. Participants were overweight or obese women aged 20–64 y, minimally active, and insulin resistant and/or dyslipidemic. Diets were randomly assigned and based on the 2010 Dietary Guidelines for Americans (DGA) or a typical American diet (TAD). Cardiometabolic risk factors and salivary cortisol were assessed at baseline and at 8 wk. Primary outcome measures included 8-wk change in overnight fasted cardiometabolic risk factors, including blood pressure, BMI, and circulating triglycerides, cholesterol, glycated hemoglobin (HbA1c), nonesterified fatty acids, and high-sensitivity C-reactive protein. This trial was approved by the University of California, Davis, Institutional Review Board.

Results: Baseline waking cortisol concentrations interacted ($P = 0.0474$) with diet to affect 8-wk changes in fasting total cholesterol. Compared with a TAD, a DGA diet was associated with 8-wk decreases in total cholesterol in participants with low (10th percentile of all participants; 2.76 nmol/L) or average (7.76 nmol/L) but not higher (90th percentile of all participants; 13.44 nmol/L) baseline waking cortisol. Consistent with this finding, there was a DGA-specific positive association ($P = 0.0047$; b: 2.88 ± 0.94) between baseline waking cortisol and 8-wk increases in total cholesterol.

Conclusions: The underlying status of waking cortisol may explain interindividual variability in total cholesterol responses to whole-food diets. This trial was registered at www.clinicaltrials.gov (https://clinicaltrials.gov/ct2/show/NCT02298725) as NCT02298725. *Curr Dev Nutr* 2022;6:nzac083.

Keywords: whole food diet intervention, cholesterol response, awakening cortisol phenotype, randomized control trial, women

Manuscript received September 30, 2021. Initial review completed March 16, 2022. Revision accepted April 14, 2022. Published online May 3, 2022.
Supported by funding from USDA Agricultural Research Service project 2032-51530-022-00-D, the National Dairy Council, and the Campbell Soup Company.
Author disclosures: The authors report no conflicts of interest. The National Dairy Council and the Campbell Soup Company did not participate in the design, implementation, analysis, or interpretation of the data.
The contents of this article are solely the responsibility of the authors and do not necessarily represent the official view of the USDA. The USDA is an equal opportunity provider and employer.Address correspondence to KDL (e-mail: kevin.laugero@usda.gov).

Abbreviations used: ACR, awakening cortisol response; CAD, coronary artery disease; DGA, Dietary Guidelines for Americans; GLM, general linear model; HbA1c, glycated hemoglobin; HPA, hypothalamic-pituitary-adrenal; NEFA, nonesterified fatty acid; TAD, typical American diet; TSST, Trier Social Stress Test; WHNRC, Western Human Nutrition Research Center.

Introduction

Phenotypic differences in endocrine and neuroendocrine system activity may explain interindividual variability in some cardiometabolic responses to whole-food diets and, therefore, may partly determine the beneficial or detrimental effects of diet. Little is known about whether the steroid hormone cortisol can diminish the benefits of a healthy diet on cardiometabolic risk. Increases in basal and stress-associated glucocorticoid (1) activity can directly alter liver, pancreatic, and adipose tissue metabolism and function; inflammation; and other hormones that affect cardiovascular and metabolic function. Alterations in basal cortisol and cortisol reactivity have been linked to high BMI (2, 3), elevated blood pressure (1, 2), high total cholesterol (1–3), high LDL cholesterol, low HDL cholesterol (2, 3), increases in circulating triglycerides (1), elevated glycated hemoglobin (HbA1c) (2), abdominal obesity (4), insulin resistance (1, 3, 4) and low maximal volume uptake of oxygen, VO_2max (2). We previously reported that differences in meal-induced cortisol at baseline explained variable body fat loss to a diet intervention aimed at reducing body weight (5). In that study, we found that the diet intervention only led to significant weight loss in women displaying a more normal cortisol response to a mixed meal.

Together, knowing differences in cortisol activity or status may help explain variable metabolic responsiveness to diet interventions or changes in diet. Basal and stress-associated hyper- and hypocortisolemia are associated with very distinct disease types and subtypes. Therefore, we hypothesized that distinct cortisol "signatures" help predict susceptibility or resistance to diet-induced improvements in cardiometabolic health. To better understand the potential for

phenotypic variation in basal and stress-induced cortisol status to moderate metabolic risk outcomes in response to whole-food diets, we tested whether baseline differences in basal circulating cortisol and stress-induced cortisol responsiveness explained variation in the cardiometabolic response to an 8-wk whole-food diet intervention. A primary report on the effect of the diet intervention on clinical risk factors for type 2 diabetes and cardiovascular disease was published (6). Therefore, in this paper, we report on a secondary analysis of that diet intervention.

Methods

Subjects

Women from Davis, California, and the surrounding area were recruited for the study. To be included in the study, women had to be overweight or obese, 20–64 y old, have a BMI (in kg/m^2) of 25–39.9, did not meet the minimal physical activity guidelines of 150 min/wk, and present with either insulin resistance and/or dyslipidemia. A telephone screening questionnaire was first conducted by study coordinators to determine general study eligibility. If eligible, women were asked to visit the Western Human Nutrition Research Center (WHNRC) for more detailed screening. Exclusion criteria included presence of metabolic diseases, including gastrointestinal disorders, cancer, or other serious chronic disease; being pregnant or lactating; current tobacco use; taking prescribed or over-the-counter weight-loss medications during the 6 mo before enrollment into the study; participating in moderate or strenuous physical activity >30 min/d on ≥5 d/wk; experiencing weight change of >5% of body weight within 6 mo of entry into the study; having a resting blood pressure >140/90 mm Hg, hemoglobin <11.5 g/dL, total cholesterol >300 mg/dL, LDL cholesterol >189 mg/dL, triglycerides >400 mg/dL, and clinically abnormal thyroid or liver function; working "graveyard" shifts or being forced to stay awake all night; having dietary restrictions that would interfere with consuming the intervention foods; or use of corticosteroids and medications for elevated lipids or glucose. Individuals considered vulnerable populations, including adults unable to consent, infants, children, and prisoners, were deemed ineligible for the study. No participants declared a diagnosis of Cushing syndrome.

Subjects provided written informed consent for screening and again, if selected, prior to participation in the intervention study. Consent included authorization for all future uses of data in published research. No adverse events were reported by intervention participants. Subjects were not compensated for screening but were compensated for participation in the intervention study. Data were thoroughly anonymized during this study and managed by the study director. Data were collected on forms and questionnaires using only a participant identification code, which was stored separately from any identifiable data. There were no direct benefits to the subjects; however, it is possible that provision of calorie-controlled diets contributed to health benefits. Results of blood analyses (clinical chemistry panels, lipid panels) were shared with subjects upon completion of the study.

Study design

This report is a secondary analysis based on women who participated in a primary study reported by Krishnan et al. (6) examining the effects of whole-food diets on cardiometabolic risk factors. The study was a randomized, double-blind, controlled 8-wk intervention conducted in overweight and obese women randomly assigned to either *1*) a diet based on the 2010 Dietary Guidelines for Americans (DGA) or *2*) a diet based on a typical American diet (TAD). The TAD diet was developed based on dietary intake data from the USDA's "What We Eat in America" survey or the dietary component of the NHANES, while the DGA diet was developed from food-group recommendations in the USDA's DGA (6). Both DGA and TAD diets were developed to offer commonly available foods and beverages while maintaining body weight over the 8-wk trial. The diets varied based on specific guidelines, with the DGA diet containing more whole grains, low-fat dairy, polyunsaturated fat, vegetables, and fruit and less added sugars, refined grains, sodium, and solid fats. The study published by Krishnan et al. (6) provides a specific nutrient breakdown, sample menus, and key products used for meal preparation. Compliance to the prescribed diet in individuals who completed the study was assessed through diet logs along with surveillance of weight and returned packaging of offered meals. The trial was conducted at the WHNRC in Davis, California.

Prior to participation, subjects were given an in-person orientation to the study and informed that the research was investigating a broad set of factors that may contribute to understanding dietary effects on the body. Following orientation, subjects were instructed to consume their typical diets for a 1-wk baseline (pre-intervention) period. Following this period, they were either provided DGA- or TAD-based intervention diets for 8 wk. At the first test visit, physical activity, usual diet, energy requirements, body composition, cardiometabolic risk markers, and markers of psychological and physiological stress were measured. These measurements were repeated at the end of the 8-wk intervention. Details of the study timeline, baseline characteristics, and measurements can be found in the report published by Krishnan et al. (6). This trial was approved by the University of California, Davis, Institutional Review Board and is registered (NCT02298725) at clinicaltrials.gov. All participants provided written informed consent for participating in the study.

Measurements

To assess 8-wk changes in metabolic-related risk, fasting blood samples were taken at test weeks 0 (baseline) and 8 (final) to assess standard risk factors. These measurements included plasma concentrations of total cholesterol, HDL cholesterol, LDL cholesterol, triglycerides, nonesterified fatty acids (NEFAs), HbA1c, and high-sensitivity C-reactive protein.

Fasting blood samples were collected using venipuncture into Vacutainers (Becton Dickinson VACUTAINER Systems) and then prepared for serum and plasma (EDTA). Serum Vacutainers were allowed to clot at room temperature for 30 min prior to centrifugation at 1300 × *g* at 4°C for 10 min; plasma Vacutainers were kept on ice directly after collection and centrifuged at 1300 × *g* at 4°C for 10 min. Prior to centrifugation, aliquots of whole blood were also collected and stored with serum and plasma aliquots at −80°C for analysis. Samples were sent to the UC Davis Health Department of Pathology and Laboratory Medicine for analysis. Analysis of serum lipids, including HDL, LDL, and total cholesterol, was conducted using the Beckman Coulter DxC 600/800 platform using Beckman Coulter reagent kits. Triacylglycerides and NEFAs were measured using standard laboratory

methods and a clinical chemistry analyzer (Hitachi 902; Roche Diagnostics). Plasma C-reactive protein was analyzed on an MSD Sector Imager 2400 using the Vascular Injury Panel 2 reagent kit (Meso Scale Discovery). Whole-blood HbA1c was analyzed on a Cobas Integra 400 Plus Analyzer (Roche Diagnostics).

Salivary cortisol measurements

Alterations in basal and stress- or challenge-induced cortisol responsiveness have all been linked to disturbances in behavior, metabolism, and chronic diseases. Therefore, salivary cortisol status was assessed basally at home and during a test visit where we could evaluate the responsiveness of this glucocorticoid hormone under controlled experimental conditions. Morning cortisol upon waking (waking cortisol), awakening cortisol response (ACR; difference between morning cortisol upon waking and 30 min after waking), and the diurnal fluctuation in cortisol (difference between morning cortisol and bedtime cortisol) were used as indicators of basal cortisol activity and status. During the test visit, the acute cortisol response to a mental stress test, Trier Social Stress Test (TSST), was used as an indicator of challenge-induced cortisol responsiveness. The TSST was administered later in the afternoon, 2 h after a standard lunch at each test visit during test weeks 0 (baseline) and 8 (final 8-wk post-intervention testing). The TSST is composed of a speech task and mental arithmetic task that have been shown to induce psychological stress and subsequent increases in plasma and salivary cortisol (7). Tasks were facilitated by 2 judges who were unfamiliar to the subjects and conducted in front of a video camera. Two hours following consumption of a standard lunch, participants were led to a room and introduced to 2 judges, who briefed them on the activity they would complete in the next 20 min. Judges informed subjects that they would be filmed and that videos would be used by research staff to assess their behavior during the activity. Each subject was then informed that they would be allocated 5 min to prepare a 5-min speech, which they would present to the judges. Directly upon completion of the speech, a 5-min question-and-answer session commenced. During this time, subjects were asked to describe past work experience and their interactions with managers and co-workers. After this session, subjects were asked to accurately count backwards in odd steps from a prime number (e.g., start at 1022 and count backwards by 13) as rapidly as possible for 5 min. Every time mistakes were observed, the judges instructed the participants to start over from the beginning.

Saliva samples were collected using Salimetrics Oral Swabs. Samples were collected directly before, immediately after (~30 min), and 60, 90, and 120 min after induction of the TSST. The maximum cortisol response was taken as the difference (**Δ**) between the highest cortisol value during the post-TSST period and the pre-TSST value. Participants placed the swab in their mouth for 1–2 min and then deposited the swab into provided sample tubes. To measure ACR, participants were asked to use the provided collection materials to collect their saliva upon waking and 30 min post-waking on a study day preceding test days. Participants were asked to store their samples in a home refrigerator prior to returning the samples to the WHNRC for analysis. Saliva was also collected up to 11 times associated with each test day to measure diurnal cortisol variations and response to stress tests. Swab samples were immediately centrifuged to collect and aliquot saliva, and then stored at −80°C until being assayed for cortisol at the WHNRC. ELISA (expanded-range high-sensitivity salivary cortisol kit; Salimetrics) was used to determine salivary cortisol concentrations. This assay can detect cortisol concentrations ranging from 0.193 to 82.77 nmol/L (0.007–3.0 μg/dL) and has a typical intra- and interassay CV of 3.5% and 5.1%, respectively. The standard reference range indicated by Salimetrics for morning cortisol in all adults is 0.094–1.551 μg/dL or 2.593–42.786 nmol/L.

Physical measurements

Height, weight, and waist-to-hip ratio measurements were taken during test weeks 0 and 8. Subjects were weighed wearing lightweight surgical scrubs using a calibrated electronic scale (Tanita BWB-627A Class III electronic scale; Toledo Scale) to the nearest 0.1 kg. A wall-mounted stadiometer (model S100; Ayrton Corporation) was used to measure height to the nearest 0.1 cm. BMI was calculated as kg/m^2. An anthropometric tape was used to measure waist circumference, which was measured as the minimum circumference between the iliac crest and the rib cage, and hip circumference, which was measured at the maximum protuberance of the buttocks. These measurements were completed in duplicate to ensure accuracy. A standard blood pressure cuff (GE DINAMAP vitals monitor; GE Healthcare) was used on 1 arm to evaluate resting blood pressure at the beginning of the test visit while in a fasted state.

Statistical analysis

Statistical analyses were conducted using SAS for Windows, release 9.4 (SAS Institute). Details about the primary study sample size and randomization sequence can be found in Krishnan et al. (6). For this secondary study, analyses were conducted on 37 participants (DGA, 21; TAD, 16) who had a complete cortisol and metabolic risk factor dataset (**Figure 1**). The 8-wk change in each metabolic risk factor was used as the dependent variable in statistical models (Proc GLM) that evaluated the interactions between the diet group (DGA vs. TAD) and each of the salivary cortisol variables. The cortisol variables assessed were as follows: morning cortisol upon waking ("waking cortisol"), ACR (difference between morning cortisol upon waking and 30 min after waking), the diurnal fluctuation in cortisol (difference between morning cortisol and bedtime cortisol), and the stress-induced cortisol response [max difference (**Δ**) between the highest cortisol value during the 2-h post-TSST period and the value immediately before the TSST]. All statistical models included the independent variables of diet group and baseline (pre-intervention) age, BMI, education, and the relevant cortisol parameter (e.g., waking cortisol, ACR).

Also, the relevant baseline metabolic risk factors were included as independent variables in each of the relevant statistical models. For example, if the tested dependent variable was 8-wk change in fasting triglycerides, the baseline (0 week) triglyceride variable was included as an independent variable in that model. To better decipher the nature of significant interactions between diet and the cortisol variables, we used the "lsmeans at" option in the same general linear model (GLM) model to estimate the expected value of the relevant 8-wk **Δ** risk factor (e.g., 8-wk change in total cholesterol) for each diet group at selected levels of the relevant baseline cortisol indicator. The levels chosen for this evaluation were derived using all the subjects' data and included the average and 90th and 10th percentile levels of the relevant baseline stress or cortisol indicator. In addition, to further help interpret the nature of significant interactions between diet group and the baseline cortisol

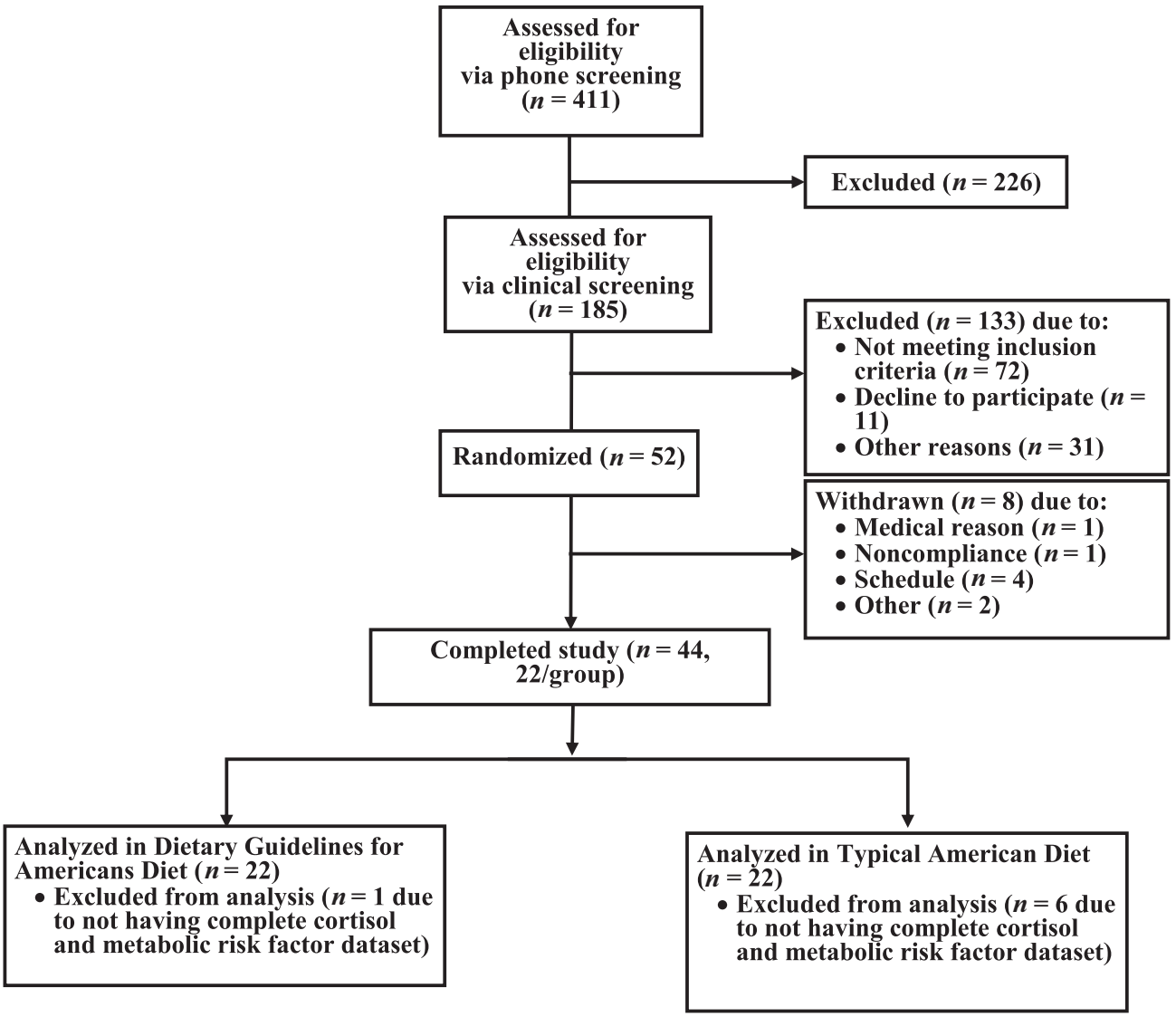

FIGURE 1 Participant flow diagram. Of the 411 individuals screened for phone and clinical eligibility, 52 were randomly assigned and 44 completed the study. Of these subjects, 22 individuals consumed each of the TAD and DGA diets for 8 wk. For this analysis, 1 individual was excluded from the DGA group and 6 were excluded from the TAD group due to an incomplete cortisol and metabolic risk factor dataset. DGA, Dietary Guidelines for Americans; TAD, typical American diet.

variables, from the same GLM model, we checked for statistically significant associations between the metabolic outcome/dependent variable (e.g., 8-wk change in total cholesterol) and the cortisol variable using the "estimate" option [*t* test; slope (association constant) different from 0, $P < 0.05$]. In all cases, a *P* value ≤ 0.05 was taken to indicate statistical significance.

Results

In this randomized controlled diet intervention, we found that waking salivary cortisol concentrations at baseline and diet group interacted to significantly ($P = 0.0046$) affect 8-wk change in fasting total cholesterol. To understand the nature of this interaction, we estimated differences in the 8-wk change in total cholesterol between the DGA and TAD groups at low (10th percentile; 2.76 nmol/L), average (7.76 nmol/L), and high (90th percentile; 13.44 nmol/L) levels of baseline waking cortisol concentrations. **Table 1** shows the estimated differences in total cholesterol change between the DGA and TAD groups at low, average, and high concentrations of baseline waking salivary cortisol. We also included LDL-cholesterol data in Table 1 since there was a tendency towards a significant ($P = 0.0542$) interaction for Δ LDL cholesterol. **Figures 2** and **3** further demonstrate the nature of this interaction between diet group and pre-intervention waking cortisol on Δ total fasting cholesterol. Compared with the TAD group, we found that the DGA diet group had a significantly lower Δ total fasting cholesterol when baseline morning cortisol concentrations were low ($P = 0.0005$) and average ($P = 0.0172$) but not high ($P = 0.3724$) (Figure 2). In corroboration with

TABLE 1 Interaction effects for waking salivary cortisol and diet group[1]

Diet group[2]	Waking cortisol level[3]	Least-square means	SE	Difference by diet, *P*
Δ Fasting total cholesterol: overall diet × waking cortisol interaction ($P = 0.0046$)				
DGA	Low	−15.7835	6.566096	0.0005
TAD	Low	13.18842	6.064751	
DGA	Average	−1.3816	4.55089	0.0172
TAD	Average	10.72444	5.352503	
DGA	High	14.98192	6.962218	0.3724
TAD	High	7.924848	6.455387	
Δ Fasting LDL cholesterol: overall diet × waking cortisol interaction ($P = 0.0542$)				
DGA	Low	−12.5327	6.673675	0.0031
TAD	Low	11.84816	6.186113	
DGA	Average	−2.39367	4.65019	0.0156
TAD	Average	10.50114	5.406354	
DGA	High	9.126306	7.321172	0.9851
TAD	High	8.970652	6.520664	

[1]Dietary Guidelines for Americans; TAD, typical American diet.
[2]$n = 21$ for DGA and 16 for TAD.
[3]Levels of waking salivary cortisol represent the 10th percentile (low, 2.76 nmol/L), average (7.76 nmol/L), and 90th percentile (high, 13.44 nmol/L) of all participants ($n = 37$).

these observations, we found a significant ($P = 0.0047$; b: 2.88 ± 0.94) and positive association between baseline waking cortisol concentrations and D total fasting cholesterol in the DGA group but not the TAD group ($P = 0.4227$; b: −0.49 ± 0.60) (Figure 3). For Δ LDL cholesterol, compared with the TAD group, we found that the DGA diet group had a significantly lower Δ LDL cholesterol when baseline waking cortisol concentrations were low ($P = 0.0031$) and average ($P = 0.0156$) but not high ($P = 0.9851$). Furthermore, we found a significant and positive association between pre-intervention waking cortisol concentrations and Δ LDL cholesterol in the DGA group ($P = 0.0474$; b: 2.02 ± 0.98) but

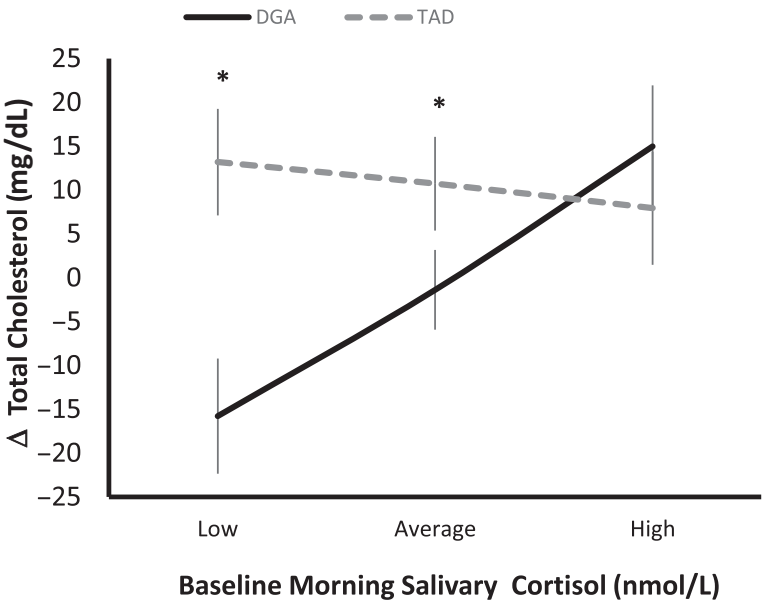

FIGURE 2 Eight-week change in fasting total cholesterol significantly differed (indicated by asterisk) between the DGA and TAD diet groups but only at low (10th percentile; $P = 0.0005$) or average ($P = 0.0172$) pre-intervention waking cortisol concentrations. No differences between the DGA and TAD groups were observed when evaluated at high (90th percentile; $P = 0.3724$) pre-intervention waking salivary cortisol concentrations. Error bars represent SEs. Change in cholesterol was adjusted for age and educational level ($n = 21$ for DGA and 16 for TAD). The 8-wk change in each metabolic risk factor was used as the dependent variable in statistical models (Proc GLM) that evaluated the interactions between the diet group (DGA vs. TAD) and each of the salivary cortisol variables. All statistical models included the independent variables of diet group and baseline (pre-intervention) age, BMI, education, and the relevant cortisol parameter (e.g., waking cortisol, ACR). ACR, awakening cortisol response; DGA, Dietary Guidelines for Americans; TAD, typical American diet.

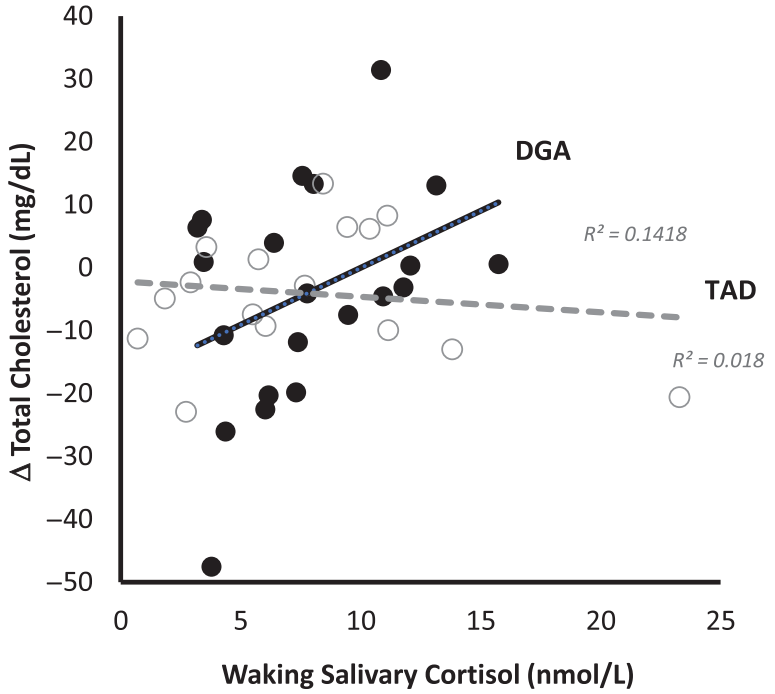

FIGURE 3 The association between pre-intervention waking cortisol concentrations and 8-wk change in fasting total cholesterol was significant ($P = 0.0047$) in the DGA group but not in the TAD group ($P = 0.4227$). The change in total cholesterol was adjusted for age and educational level, and the adjusted values for total cholesterol were regressed against pre-intervention waking salivary cortisol. The 8-wk change in each metabolic risk factor was used as the dependent variable in statistical models (Proc GLM) that evaluated the interactions between the diet group (DGA vs. TAD) and each of the salivary cortisol variables. All statistical models included the independent variables of diet group and baseline (pre-intervention) age, BMI, education, and the relevant cortisol parameter (e.g., waking cortisol). DGA (Dietary Guidelines for Americans), filled circles; TAD (typical American diet), open circles.

not the TAD group ($P = 0.6688$; b: -0.27 ± 0.62). We did not find statistically significant interactions between diet and ACR, diurnal cortisol, or the stress-induced cortisol response. For all other metabolic risk factors tested, we failed to find a statistically significant interaction between diet and any of the selected baseline cortisol variables.

Discussion

To our knowledge, this is the first report showing the association between cortisol status at baseline and the comparative effects of a DGA- and TAD-based intervention on longitudinal cardiometabolic risk outcomes. Our previous work showed that whole-food diets can influence biological and qualitative measures of chronic stress and acute stress reactivity (8, 9). The current findings are a logical extension of our prior work and demonstrated how cortisol status upon waking in the morning might influence cardiometabolic responses to whole-food diets. Our general findings support our hypothesis and suggest that baseline phenotypic differences in waking salivary cortisol influence the ability of a DGA-based diet to lower total and possibly LDL cholesterol relative to levels of these cardiovascular risk factors observed in individuals consuming the TAD diet. We found that participants having higher concentrations of salivary cortisol just after waking in the morning had significantly less 8-wk improvement in circulating cholesterol in response to the DGA diet. In support of this observation, we also found a significant and positive association between morning salivary cortisol and circulating total and LDL cholesterol, but only in the participants consuming the DGA diet. Thus, it appears that the cholesterol-lowering effect of a DGA diet may be linked to variations in waking cortisol status or some other factor associated with waking cortisol. However, it is also evident that the apparent detrimental effects of a less-healthy diet like the TAD quell the influence of factors potentially linked to pre-intervention phenotypic differences in morning cortisol.

We previously reported that phenotypic differences in meal-induced cortisol at baseline explained variable body fat loss to a diet intervention aimed at reducing body weight (5). In that study, we found that the diet intervention only led to significant weight loss in women displaying a more normal cortisol response to a mixed meal. Thus, given those previous results and our current findings, it is not surprising that pre-intervention differences in cortisol may influence a person's metabolic response to whole-food diets. In the present study, we did not find stress-induced cortisol or the ACR to be associated with any of the cardiometabolic outcomes measured in this study. Thus, it appears that status or regulation in basal (waking) cortisol, but not cortisol in response to stress or awakening, is linked in some way to a DGA-based diet-induced shift in total cholesterol. Future work is needed to determine possible

mechanistic factors that uniquely link certain cardiometabolic or other functional responses to basal cortisol versus cortisol responsiveness. Together, future studies are needed to better clarify specific mechanisms linking cortisol phenotypes or signatures and susceptibility to beneficial or detrimental effects of whole-food diets.

Whether our present findings reveal a direct effect of morning cortisol concentration status on diet-induced changes in cholesterol cannot be determined from this study. However, glucocorticoids, such as cortisol, have profound effects on cardiovascular and metabolic functions, including carbohydrate, protein, and lipid and lipoprotein metabolism (10). Glucocorticoids, such as cortisol, also affect the postprandial metabolic response to consuming a mixed meal (11), so it is not surprising that we found variations in cortisol concentrations to be associated with the metabolic response to diet. Subtle disturbances in hypothalamic-pituitary-adrenal (HPA) function and augmented tissue sensitivity to glucocorticoids have been observed in patients with metabolic syndrome (12). Cortisol and cholesterol are positively associated in individuals who had active coronary artery disease (13). Previous chronic exposure to glucocorticoids has also been associated with central fat deposition, abnormal blood lipid concentrations, muscle wasting, diabetic symptoms, and other symptoms that suggest high metabolic risk (11). Persistent exposure to high levels of glucocorticoids can lead to cardiovascular and metabolic dysfunction, and pharmacologic strategies for mitigating these glucocorticoid effects have been considered (14–16). Treating patients with corticosteroids has been associated with elevations in total cholesterol (17). More relevant to our findings, higher morning cortisol was reported to be associated with cardiovascular disease risk (18). Moreover, higher waking cortisol was reported to be associated with higher circulating LDL cholesterol in youth, and this association was independent from any differences in age, BMI, sex, insulin sensitivity, and puberty (19). The link between cortisol and circulating cholesterol concentrations is thought to result, in part, from the actions of this steroid hormone to increase lipid synthesis and turnover, as well as stimulate hepatic synthesis of VLDL and accumulation of fatty acids (20, 21). These glucocorticoid effects on lipoprotein metabolism may arise indirectly through glucocorticoid-induced hepatic and peripheral insulin resistance (22).

While elevations in cortisol induce insulin resistance, abnormally low cortisol via cortisol withdrawal increases insulin sensitivity and reduces glucose production (10). Thus, in our study, it is possible that pre-intervention waking levels of cortisol are indicative of overall cortisol tone and exposure, which may have mitigated (high cortisol) or permitted (low cortisol) the cholesterol-lowering effects of consuming a DGA diet. Of course, it is also possible that some other factors or characteristics directly or indirectly related to waking cortisol explain our findings of an association between elevated waking cortisol and inhibited diet-associated reductions in circulating cholesterol concentrations. For example, differences in baseline waking cortisol may reflect alterations in other related systems, such as the HPA axis, autonomic nervous system, and brain systems that regulate both adrenocortical activity and metabolism (23, 24). Differences in the regulation of and activity in these systems are linked to behavioral and metabolic disturbances. Therefore, baseline waking cortisol in our study may reflect underlying disturbances to other systems and mediators that would be expected to influence the beneficial effects of whole-food diets on metabolism.

It is interesting that, in participants consuming the TAD, 8-wk change in circulating cholesterol failed to fluctuate regardless of whether baseline waking cortisol was low (10th percentile) or high (90th percentile) in those participants. Compared with the DGA diet, the TAD used in this study was higher in added sugars and saturated fat, which have been linked to increases in inflammation and elevated cholesterol (25–27). It is possible that the competing effects of consuming a TAD on lipid and lipoprotein metabolism in already at-risk persons explained a lack of association between varying waking cortisol levels and the cholesterol response to diet. There also exists significant interindividual variation in lipid responsiveness to glucocorticoid treatment (28, 29). Positive associations between waking cortisol and cholesterol were reported in patients with mild to significant coronary artery disease (CAD) but not in persons without CAD. In that same study, positive associations between cortisol administration and cholesterol also depended on a specific behavioral subtype (13). Our results further suggest that the link between cortisol status and lipoprotein metabolism may also be related to dietary habits.

Mechanistic studies are warranted to test whether cortisol status physiologically mediates diet-induced changes in cholesterol or other cardiometabolic outcomes. Our work adds to the expanding appreciation for precision nutrition and the need to better understand interindividual differences when developing nutritional and other therapeutic strategies for preventing or treating chronic disease.

To our knowledge, while this is the first controlled-feeding trial assessing a moderating effect of pre-intervention cortisol status on metabolic risk factor response to diets based on the DGA and TAD, this study is not without limitations, as previously reported (9). The 8-wk duration of the study is relatively short and may not reflect outcomes extending beyond the 8-wk intervention period. Furthermore, the sample size of the study was relatively small. Finally, this study was conducted in women at metabolic risk, and outcomes may not be generalizable to men or to individuals with normal glucose or lipid values. In addition, the study did not exclude participants who were taking birth control, antidepressant, or other medications that may affect cortisol binding in the bloodstream, and thus cortisol expression in the saliva. However, given that the data analyzed include 8-wk differences, including 2 full 4-wk birth control cycles, the authors do not believe that it is likely that the results observed are attributable to these factors. Finally, while we did not identify any individuals who declared having a disease or taking medications that would interfere with cortisol assessment during screening, there is a possibility that participants had undiagnosed Cushing syndrome. However, there were no significant outliers to the cortisol values that suggest that this was the case. Although we provide some speculation about possible mechanisms, it is important to re-emphasize that we cannot draw any conclusions about causality from our results.

Acknowledgments

We thank Jan Peerson for her expert advice on statistical analysis. The authors' responsibilities were as follows—KDL and NLK: contributed to project design, development of the overall research plan, and study oversight; HS and KDL: analyzed data and performed statistical analysis; and all authors: contributed to conducting the research, wrote the manuscript and contributed to its final content, and read and approved the final manuscript.

Data Availability

The data underlying this article will be shared on reasonable request to the corresponding author.

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