Prolonged Glycemic Adaptation Following Transition From a Low- to High-Carbohydrate Diet: A Randomized Controlled Feeding Trial

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OBJECTIVE

Consuming ≥150 g/day carbohydrate is recommended for 3 days before an oral glucose tolerance test (OGTT) for diabetes diagnosis. For evaluation of this recommendation, time courses of glycemic changes following transition from a very-low-carbohydrate (VLC) to high-carbohydrate diet were assessed with continuous glucose monitoring (CGM).

RESEARCH DESIGN AND METHODS

After achieving a weight loss target of 15% (±3%) on the run-in VLC diet, participants (18–50 years old, BMI ≥27 kg/m^2) were randomly assigned for 10 weeks to one of three isoenergetic diets: VLC (5% carbohydrate and 77% fat); high carbohydrate, high starch (HC-Starch) (57% carbohydrate and 25% fat, including 20% refined grains); and high carbohydrate, high sugar (HC-Sugar) (57% carbohydrate and 25% fat, including 20% sugar). CGM was done throughout the trial (n = 64) and OGTT at start and end (n = 41). All food was prepared in a metabolic kitchen and consumed under observation.

RESULTS

Glucose metrics continued to decline after week 1 in the HC-Starch and HC-Sugar groups (P < 0.05) but not VLC. During weeks 2–5, fasting and 2-h glucose (millimoles per liter per week) decreased in HC-Starch (fasting −0.10, P = 0.001; 2 h −0.10, P = 0.04). During weeks 6–9, 2-h glucose decreased in HC-Starch (−0.07, P = 0.01) and fasting and 2-h glucose decreased in HC-Sugar (fasting −0.09, P = 0.001; 2 h −0.09, P = 0.003). The number of participants with abnormal glucose tolerance by OGTT remained 10 (of 16) in VLC at start and end but decreased from 17 to 9 (of 25) in both high-carbohydrate groups.

CONCLUSIONS

Physiological adaptation from a low- to high-carbohydrate diet may require many weeks, with implications for the accuracy of diabetes tests, interpretation of macronutrient trials, and risks of periodic planned deviations from a VLC diet.

Preceding carbohydrate intake affects glucose tolerance, as demonstrated by research dating to the early 20th century (1–6). For this reason, diabetes screening

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protocols recommend an unrestricted diet containing ≥150 g/day carbohydrate for 3 days prior to an oral glucose tolerance test (OGTT) (7). However, the adequacy of this 3-day preparatory period specifically and the process of physiological adaptation to changes in macronutrients in general have not been well characterized.

Recent evidence indicates that the process of adaptation from a conventional high-carbohydrate diet to a low-carbohydrate diet may require several weeks to months (8–10), raising the question as to whether the reverse process—that is, adaptation from a low-carbohydrate to a high-carbohydrate diet—may also be prolonged. This question has special relevance today, in view of the increased popularity of low-carbohydrate diets for weight loss (11) and diabetes management.

Changes in dietary macronutrients necessitate alternations in myriad biochemical pathways related to energy homeostasis, as elicited in large part by the postprandial secretion patterns of insulin and glucagon (12). Although no single clinical variable defines physiological adaptation to a diet, temporal changes in glycemia provide a convenient and robust measure. Blood glucose concentrations, fasting and in response to defined meals, reflect a dynamic balance between β-cell function and tissue insulin sensitivity in key organs (especially liver, muscle, and adipose). With use of state-of-the-art technology for continuous glucose monitoring (CGM), interstitial glucose concentration can be accurately assessed on a long-term basis.

The aim of this study was to examine changes in glycemia among participants habituated to a very-low-carbohydrate (VLC) diet and then randomized for 10 weeks to one of three isoenergetic diets: *1*) VLC; *2*) high carbohydrate, high starch (HC-Starch); and *3*) high carbohydrate, high sugar (HC-Sugar).

RESEARCH DESIGN AND METHODS

Study Design

The randomized controlled feeding trial comprised introductory, run-in, and residential phases (Fig. 1). During the introductory phase, pre–weight loss data were collected while participants were eating their usual diets. During the run-in phase (14–15 weeks), energy intake was restricted to promote a weight loss target of 15% (±3%), relative to baseline body weight, on a VLC diet. During the residential phase (13 weeks), participants who achieved the target weight loss were housed in Ashland, MA, with supervision of food intake. Participants received a eucaloric VLC diet for the first 3 weeks of this phase and then were randomly assigned to one of three isocaloric test diets for 10 weeks: VLC, HC-Starch, or HC-Sugar. Methods for parallel random assignment and power calculations are presented in the preanalysis plan (13). The primary outcome for the main trial was body fat mass. The exploratory analyses presented herein address the issue of physiological adaptation to macronutrient change following randomization. The trial was conducted from May 2018 to May 2020, with participants recruited in cohorts to begin the run-in phase in May or October of each year (2018 and 2019). The study was stopped early due to the coronavirus disease 2019 (COVID-19) pandemic. The protocol amendment history is presented in Supplementary Data. The main study was registered at clinicaltrials.gov, NCT03394664.

Eligibility Criteria

Eligible participants included men and women aged 18–50 years with pre–weight loss BMI ≥27 kg/m^2 and no known cardiovascular disease or diabetes. Additional eligibility criteria are listed in Supplementary Table 1. Demographic information including sex, date of birth, ethnicity, and race was collected at time of enrollment. The study was approved by the institutional review board at Boston Children's Hospital, and all participants provided written informed consent.

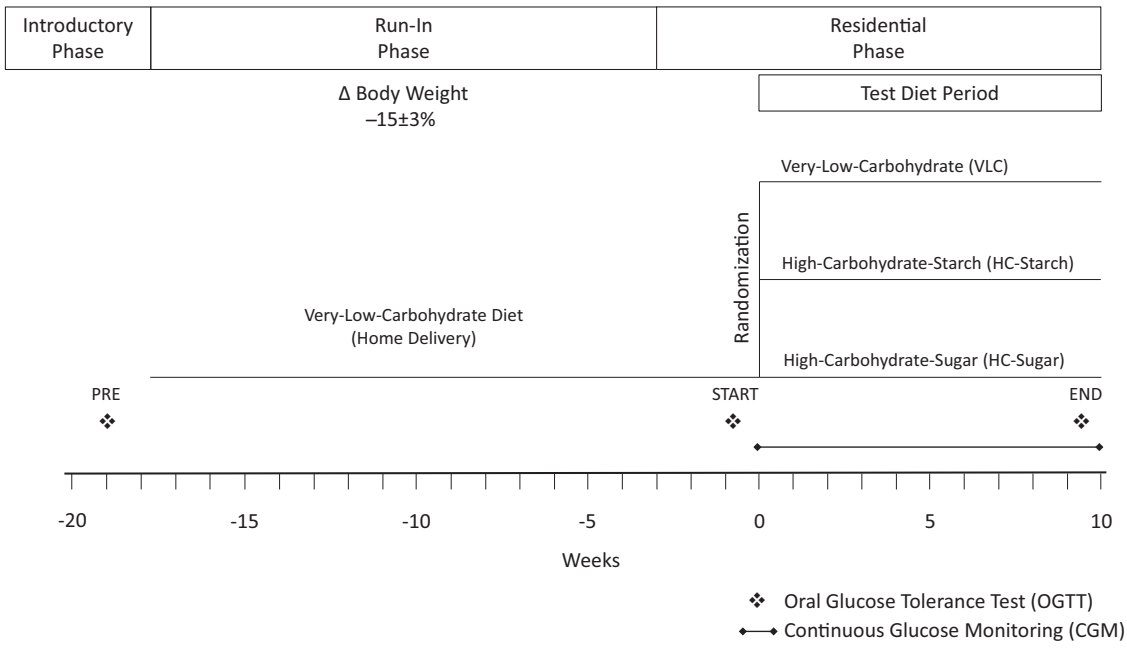

Figure 1—Study design.

Dietary Interventions

Run-in Diet

A ketogenic VLC diet (7.5% of total energy from carbohydrate, 67.5% from fat, 25% from protein) was used to promote weight loss during the run-in phase. Individual energy needs were estimated based on resting energy expenditure, calculated with a regression equation (14), multiplied by a physical activity factor of 1.5 (assuming a lifestyle with light-intensity physical activity) (15). Energy intake was restricted to 60% of estimated needs. Run-in phase meals were prepared in a central facility (Metabolic Meals, St. Louis, MO) and delivered to participant homes on a weekly basis. When the weight loss target was achieved, energy intake on the VLC was increased to 100% of estimated needs for weight loss maintenance. Prior to random assignment, energy intake was "locked" at a level corresponding to weight loss maintenance on the VLC run-in diet.

Test Diets

The VLC test diet contained 5% of energy from carbohydrate, 77% from fat, and 18% from protein. The two high-carbohydrate test diets were controlled for macronutrient composition (57% of energy from carbohydrate, 25% from fat, 18% from protein) and whole grains (25% of total energy) but differentiated by starch and sugar content (20% of total energy either from refined grains or sugar). Timing of meals followed a daily schedule with breakfast at 7:30 A.M., lunch at 1:00 P.M., and dinner at 7:00 P.M. All meals during the residential phase were prepared by an on-site professional hospitality service (FLIK Hospitality Group, Rye Brook, NY) under supervision of a research dietitian and were consumed in a group setting, monitored by study staff. Designated participant support personnel conducted weekly in-person check-ins to ensure participant well-being. For all diets, energy was distributed throughout the day: 30% for breakfast, 35% for lunch, and 35% for dinner, and the macronutrient composition of every meal reflected the composition of each respective diet (Supplementary Table 2).

Measurements

Anthropometrics and glucose tolerance were assessed at the following time points: pre–weight loss (PRE), before randomization at the start of the trial (START), and at the end of the residential phase (END) (Fig. 1). Body weight and height were measured with a calibrated scale (model BVB-800; Tanita, Arlington Heights, IL) and stadiometer (model PE-AIM-101; Perspective Enterprises, Portage, MI), respectively. Research personnel assessing these outcomes were blinded to random group assignment.

OGTTs were administered following a 12-h overnight fast, and arterialized venous blood samples (16) were collected at −10, −5, 0, 10, 20, 30, 60, 90, and 120 min relative to the time of dextrose (Trutol) consumption (75 g). Plasma samples were processed and stored at −80°C until analysis for glucose and insulin. Glucose was measured with the enzymatic hexokinase method (Roche Diagnostics, Indianapolis, IN), and insulin was measured with electrochemiluminescence immunoassay (Roche Diagnostics). HbA_{1c} in fasting blood samples was measured by turbidimetric immunoinhibition (Roche Diagnostics). For the OGTT, variables of interest for this report included fasting (mean of values obtained at −10, −5, and 0 min) and 2-h postprandial glucose concentrations.

The FreeStyle Libre Pro Flash Glucose Monitoring System (Abbott Diabetes Care, Alameda, CA) was used for CGM throughout the test diet period of the residential phase (Fig. 1), with participants blinded to readings. Interstitial glucose was measured at 15-min intervals, processed, and stored in the device memory for subsequent download. Devices were applied on the Thursday of week 3 of the residential phase (prior to random assignment) and replaced every second Thursday between breakfast and lunch, in accordance with instructions from the manufacturer for a 2-week sensor cycle. Two researchers (L.T.J. and D.S.L.) reviewed all CGM data to determine meal start times using daily mealtime schedules and logs for verification.

This report focuses on glucose dynamics pertaining to breakfast in light of evidence for diurnal variation in glucose metabolism in individuals without a diagnosis of diabetes. Higher β-cell responsiveness and glucose tolerance in the morning, compared with other times of day, may comprise a metabolic milieu for a more pronounced effect of dietary composition on outcomes of interest (17). Example breakfasts are presented in Supplementary Table 3. Fasting glucose was calculated as the average of the five measurements immediately preceding the first mealtime glycemic response. Peak glucose was defined as the highest glycemic value in the 2-h postprandial time frame. Values at the 2-h postprandial timestamp were used as a measure of meal glucose tolerance. Data from weeks 1–9 of the test diet period were used in analyses because during week 10 CGM was affected by assessments for the main study (e.g., prolonged fasting prior to measurements, consumption of dextrose for OGTT, removal of CGM device for evaluation of body composition). For weeks 1–9, a mean was calculated with use of data from the first 3 days of each week, except when missing or in the case of visually implausible data (suggesting technical error) or when mealtime deviated substantially from schedule. In these cases, the first 3 days with valid data were selected.

Statistical Analyses

Participant characteristics at PRE and START were summarized with descriptive statistics. Mean (SD) was calculated for continuous variables, and frequency (percentage) was calculated for categorical variables.

In studying the effects of dietary interventions on glycemic control, four different constructs could be relevant: *1*) typical glucose metabolism, *2*) achievable glucose metabolism, *3*) glucose metabolism in a narrowly defined time interval, and *4*) projected glucose metabolism after achievement of steady state on a prescribed regimen. A particular measurement strategy may be biased or influenced by measurement artifact for one construct but not another. If transitioning from one diet to the next produces acute effects that are not indicative of long-term steady-state glucose metabolism, such a measurement strategy might be biased as a result of measurement artifact. That is, measurement error would be correlated with the intervention (18). In contrast, if one were fundamentally interested in the effects of transition from one diet to the next on acute-term glucose metabolism, this concern would not apply. The focus of our study and choice of analytic approach, described below,

are premised on steady-state effects (construct 4).

Following visual inspection, linear trend analysis was used to evaluate slopes for the specified CGM metrics from weeks 2–9 of the test diet period. We excluded data from week 1 in the analysis to test the hypothesis of prolonged adaptation beyond the commonly recommended 3-day adaptation window (2,19). Time in weeks (continuous variable), diet group, and time × diet group interaction were modeled as fixed effects. Coefficients for interaction terms and main effects were used to estimate the slopes for time within diet group.

To further examine patterns of adaptation for each CGM metric within diet group, a segmented mixed-model procedure with random change points (20) was used to estimate the change point—also called the break point, transition point, or knot (in spline terminology). The change point (κ) was defined as time in weeks corresponding to a change in slope, and the model output SE was used to calculate a normal-based 95% CI ($\kappa \pm 1.96$ SE). As a check of internal consistency, bootstrap resampling also was conducted, and percentile bootstrap CIs were estimated for the change points.

Then, a piecewise linear mixed-model procedure was used to assess patterns of change within each diet (slope estimates) before and after a time-indexed change point common to CIs for all metrics. Pairwise contrasts for each interval (before and after the change point) were used to assess differences in patterns of change between the two high-carbohydrate diet groups (HC-Starch − HC-Sugar) and between each high-carbohydrate diet group and the VLC group, which served as negative control (HC-Starch − VLC, HC-Sugar − VLC).

To explore implications of adaptation on results from clinical tests commonly used to screen for diabetes, we calculated frequency of laboratory values corresponding to abnormal HbA_{1c} (≥39 mmol/mol, ≥5.7%), fasting glucose (≥5.6 mmol/L, ≥100 mg/dL), and 2-h glucose (≥7.8 mmol/L, ≥140 mg/dL) at START and END within diet groups.

All models were adjusted for diet group and cohort. Residual plots were examined for assessment of the assumptions of normality in residuals, homogeneity of residual variance, and independence. Additionally, influence diagnostic tests were done. Analyses were conducted with SAS 9.4 (SAS Institute, Cary, NC) or the R platform (21). No imputation was done for missing data, considering the exploratory nature of the study, overall low occurrence of missing CGM data, and plausibility that data are, for practical purposes, missing completely at random. A priori significance levels were set at two-tailed $\alpha = 0.05$, exact *P* values are reported, and data are presented as mean (95% CI).

Data and Resource Availability

Data presented in this article—along with the analytic code and code book—is publicly available on Open Science Framework (https://osf.io/m6v73/).

RESULTS

Study Participants

Participant flow is presented in Supplementary Fig. 1. At the end of the run-in phase, 77 participants were randomized to a diet group, and 70 were retained through the residential phase. Among the retained participants, CGM data were available for 64 participants (because a different device, yielding incomparable data, was used for 6 participants in the initial cohort) and OGTT data were available for 41 participants (due to elimination of the OGTT at END for 29 participants in the final cohort, as part of risk mitigation in response to COVID-19). Participant characteristics at PRE and START are presented in Table 1 and Supplementary Fig. 2. All results were reproduced and verified by an independent statistician. Assumption checking conducted independently by two statisticians suggested no violation of model assumptions.

Linear Trends

Changes in CGM metrics (fasting, peak, and 2-h glucose) during the test diet period are presented in Table 2 and Supplementary Fig. 3. Visual inspection suggested ongoing adaptation beyond 1 week in both high-carbohydrate diet groups. Linear trend analyses for glucose metrics from weeks 2–9 of the test diet period detected significant negative slopes in HC-Starch and HC-Sugar but not in VLC. In HC-Starch, slopes were significant for fasting glucose (mean estimate −0.04 mmol/L per week [95% CI −0.07, −0.01]), peak glucose (−0.09 mmol/L per week [−0.12, −0.05]), and 2-h glucose (−0.07 mmol/L per week [−0.10, −0.03]). In HC-Sugar, slopes were significant for fasting glucose (−0.03 mmol/L per week [−0.06, −0.00]) and peak glucose (−0.07 mmol/L per week [−0.11, −0.03]).

Change Points During Glycemic Adaptation

Segmented regression modeling of slope dynamics from weeks 2–9 are presented in Fig. 2. In both high-carbohydrate diet groups, change point estimates were identified for fasting glucose (HC-Starch mean estimate 5.2 weeks [95% CI 4.1, 6.3] and HC-Sugar 4.6 weeks [3.6, 5.6]) and 2-h glucose (HC-Starch 5.3 weeks [3.2, 7.3] and HC-Sugar 5.7 weeks [3.8, 7.7]). Due to linearity of the data profiles for peak glucose, the segmented regression procedure failed to converge, and change points were not detected for this variable in either HC-Starch or HC-Sugar. No change points were detected in VLC for any of the specified CGM metrics. Further percentile bootstrap resampling to assess internal consistency indicated that the change point estimates for fasting glucose and 2-h glucose were consistent with asymptotic estimates based on regression modeling assumptions (Supplementary Table 4).

Patterns of Glycemic Adaptation

Inspection of data from the segmented regression modeling indicated that normal-based CIs for fasting and 2-h glucose all included 5 weeks in HC-Starch and HC-Sugar. Thus, 5 weeks was used as the time index for piecewise linear mixed modeling. Slope dynamics before (weeks 2–5) and after (weeks 6–9) the change point for fasting and 2-h glucose are presented in Supplementary Tables 5 and 6.

From week 2 to week 5, fasting glucose decreased in HC-Starch (mean estimate −0.10 mmol/L per week [95% CI −0.17, −0.04]) but did not change significantly in HC-Sugar (0.04 mmol/L per week [−0.03, 0.11]). From week 6 to week 9, fasting glucose did not change in HC-Starch (−0.00 mmol/L per week [−0.05, 0.04]) but decreased in HC-Sugar (−0.09 mmol/L per week [−0.14, −0.04]). Slopes before and after the

Table 1—Characteristics of study participants

	Completers*	CGM analyses†	OGTT analysis‡
Participants, *n*	70	64	41
Sex			
Female	29 (41.4)	24 (37.5)	27 (65.9)
Male	41 (58.6)	40 (62.5)	14 (34.2)
Race			
White	55 (78.6)	51 (79.7)	30 (73.2)
Black	7 (10.0)	6 (9.4)	6 (14.6)
Other or no response	8 (11.4)	7 (10.9)	5 (12.2)
Ethnicity			
Non-Hispanic	51 (72.9)	47 (73.4)	27 (65.9)
Hispanic	19 (27.1)	17 (26.6)	14 (34.1)
Cohort			
1	6 (8.6)	0 (0)	6 (14.6)
2	13 (18.6)	13 (20.3)	13 (31.7)
3	22 (31.4)	22 (34.4)	22 (53.7)
4	29 (41.4)	29 (45.3)	0 (0.0)
Measurements at PRE, mean (SD)			
Age, years	34.0 (9.2)	34.2 (9.1)	35.6 (9.2)
Height, cm	172.6 (9.3)	173.5 (9.1)	169.9 (8.7)
Weight, kg	101.2 (16.7)	102.2 (16.8)	95.5 (13.6)
BMI, kg/m^2	34.0 (5.1)	34.0 (5.2)	33.1 (4.2)
Fasting glucose, mg/dL	101.1 (8.0)	101.3 (7.5)	100.1 (7.7)
Fasting glucose, mmol/L	5.6 (0.4)	5.6 (0.4)	5.6 (0.4)
2-h glucose, mg/dL§	139.5 (38.8)	137.9 (39.2)	145.0 (42.4)
2-h glucose, mmol/L§	7.7 (2.2)	7.7 (2.2)	8.0 (2.4)
Fasting insulin, μIU/mL	16.0 (10.1)	15.7 (10.0)	15.1 (9.4)
Fasting insulin, pmol/L	95.8 (60.8)	94.4 (60.2)	90.9 (56.5)
Measurements at START, mean (SD)			
Weight, kg	85.8 (14.1)	86.6 (14.2)	81.4 (12.0)
BMI, kg/m^2	28.8 (4.4)	28.8 (4.5)	28.2 (3.8)
Fasting glucose, mg/dL	81.5 (7.7)	81.5 (7.9)	81.0 (6.3)
Fasting glucose, mmol/L	4.5 (0.4)	4.5 (0.4)	4.5 (0.4)
2-h glucose, mg/dL§	148.4 (40.5)	146.1 (41.3)	156.0 (42.5)
2-h glucose, mmol/L§	8.2 (2.2)	8.1 (2.3)	8.7 (2.4)
Fasting insulin, μIU/mL	5.9 (3.7)	5.7 (3.7)	5.6 (3.3)
Fasting Insulin, pmol/L	35.5 (22.2)	34.5 (22.1)	33.7 (19.6)

Data are *n* (%) unless otherwise indicated. *Of the 77 randomized participants, 70 were retained at END (completers). †CGM data were available for 64 retained participants (because a different device, yielding incomparable data, was used for 6 participants in the initial cohort). ‡OGTT data were available for 41 retained participants (due to elimination of the OGTT at END for 29 participants in the final cohort as part of risk mitigation in response to COVID-19). §OGTT curves for glucose and insulin at PRE and START are presented in Supplementary Fig. 2.

Table 2—Linear trends in glucose dynamics measured by CGM from week 2 to week 9 of the test diet period*

	VLC (*n* = 23)		HC-Starch (*n* = 22)		HC-Sugar (*n* = 19)	
Outcome	Estimate (95% CI)	*P*	Estimate (95% CI)	*P*	Estimate (95% CI)	*P*
Fasting glucose						
mg/dL, per week	−0.0 (−0.5, 0.5)	0.981	−0.7 (−1.2, −0.2)	0.004	−0.6 (−1.1, −0.1)	0.029
mmol/L, per week	−0.00 (−0.03, 0.03)		−0.04 (−0.07, −0.01)		−0.03 (−0.06, −0.00)	
Peak glucose						
mg/dL, per week	−0.4 (−1.1, 0.3)	0.223	−1.6 (−2.3, −0.9)	0.000	−1.2 (−1.9, −0.5)	0.002
mmol/L, per week	−0.02 (−0.06, 0.01)		−0.09 (−0.12, −0.05)		−0.07 (−0.11, −0.03)	
2-h glucose						
mg/dL, per week	−0.2 (−0.8, 0.4)	0.537	−1.2 (−1.9, −0.6)	0.000	−0.5 (−1.2, 0.2)	0.191
mmol/L, per week	−0.01 (−0.05, 0.02)		−0.07 (−0.10, −0.03)		−0.03 (−0.06, 0.01)	

Data are mean estimate (95% CI). Null hypothesis, slope = 0. *Within–diet group estimates of slopes derived from linear regression model of weekly means from week 2 to week 9 of the test diet period.

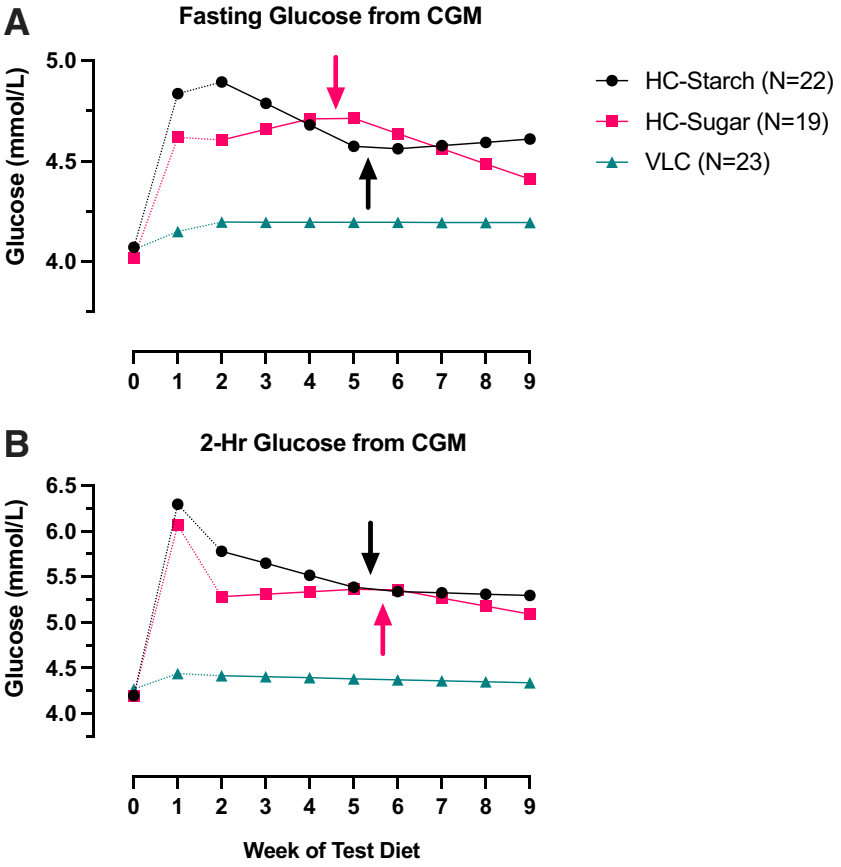

Figure 2—Segmented regression modeling of CGM slope dynamics from week 2 to week 9 of the test diet period. Data are depicted as estimate means from the models for fasting (*A*) and 2-h (*B*) glucose. Estimated change points are indicated by arrows. Data points for week 0 (last week of run-in diet) and week 1 (first week of test diet) are raw means to illustrate the full time course of changes.

change point for fasting glucose were significantly different between HC-Starch and HC-Sugar ($P < 0.05$).

From week 2 to week 5, 2-h glucose decreased significantly in HC-Starch (mean estimate −0.10 mmol/L per week [95% CI −0.19, −0.01]) but did not change in HC-Sugar (0.04 mmol/L per week [−0.07, 0.14]). From week 6 to week 9, 2-h glucose decreased significantly in HC-Starch (−0.07 mmol/L per week [−0.12, −0.01]) and HC-Sugar (−0.09 mmol/L per week [−0.15, −0.03]). However, slopes before and after the change point for 2-h glucose were not significantly different between HC-Starch and HC-Sugar.

HbA_{1c} and OGTT Metrics

HbA_{1c} and OGTT metrics at START are presented in Supplementary Table 7. The OGTT curves for glucose and insulin at START and END by diet group are depicted in Supplementary Fig. 4. Data for frequency of abnormal diabetes screening tests at START and END are presented in Table 3. At START, no participants in any of the diet groups had abnormal HbA_{1c} or fasting glucose. At END, HbA_{1c} remained within normal limits for all participants, but one participant in HC-Starch and four participants in HC-Sugar had abnormal fasting glucose. Data from the OGTT indicate that more than one-half of the participants had abnormal 2-h glucose at START (VLC, $n = 10$ of 16; HC-Starch, $n = 9$ of 13; HC-Sugar, $n = 8$ of 12). At END, the frequency remained at $n = 10$ for VLC and decreased to $n = 7$ in HC-Starch and $n = 2$ in HC-Sugar. Among the 17 abnormal tests in both high-carbohydrate groups at START, approximately one-half ($n = 8$) normalized at END.

CONCLUSIONS

In this exploratory study of data from a large, controlled feeding trial, we found evidence of a prolonged physiological process of adaptation following transition from a low- to high-carbohydrate diet in adults without diabetes. If 3 days of consuming ≥150 g/day carbohydrate were adequate preparation for diabetes screening, then no changes in measures

Table 3—Frequency of abnormal diabetes screening tests at START and END by diet group*

	VLC ($n = 16$)		HC-Starch ($n = 13$)		HC-Sugar ($n = 12$)	
Diagnosis	START	END	START	END	START	END
Abnormal HbA_{1c}						
≥5.7%	0	0	0	0	0	0
≥39 mmol/mol						
Abnormal fasting glucose						
≥100 mg/dL	0	0	0	1	0	4
≥5.6 mmol/L						
Abnormal glucose tolerance (2-h glucose)						
≥140 mg/dL	10	10	9	7	8	2
≥7.8 mmol/L						

*Frequency of participants presenting with abnormal diabetes screening tests at START (when all were habituated to a VLC prior to randomization) and END (during week 10 of test diet period).

of glucose homeostasis would be expected after the first week following transition to the high-carbohydrate diets. However, data from CGM indicate significant downward trends from week 2 to week 9 for several common measures, including fasting and 2-h glucose, with contrasting temporal patterns according to carbohydrate type (starch vs. sugar). Moreover, both amount and type of carbohydrate may influence OGTT metrics. As expected, based on previous studies (1,5–7), participants on a low-carbohydrate diet (all participants at START and those consuming the VLC test diet at END) frequently exhibited abnormal glucose tolerance during an OGTT, even with normal HbA_{1c}.

Data are limited regarding duration of adaptation to different macronutrient diets, particularly when transitioning from a low- to high-carbohydrate diet. In 1919, Hamman and Hirschman (22) first described improved carbohydrate tolerance with repeated ingestion of glucose. This phenomenon was subsequently confirmed by Staub (23), Traugott (24), and Foster (25). In 1963, Hales and Randle (4) reported that, among five healthy men previously given a low-carbohydrate diet ($<$50 g/day carbohydrate) for 5 days, glucose tolerance remained abnormal for several weeks after a return to a high-carbohydrate diet. More recently, Bonuccelli et al. (26) identified key mechanisms for the so-called Staub-Traugott effect, including potentiation of insulin secretion from pancreatic β-cells and increased hepatic suppression of glucose production. Beyond islet-specific mechanisms, prolonged adaptation might also be mediated by changes in tissue-specific insulin sensitivity, as affected by growth hormone, thyroxin, catecholamines, or other hormones regulated by diet and by central autonomic tone.

The different patterns of adaptation between the two high-carbohydrate diets suggest the existence of additional mechanisms related specifically to fructose. The chemical structure of starch (glucose polymer) in refined grains contrasts with that of sugar (a disaccharide comprising glucose and fructose). Whereas high fructose intake has adverse metabolic effects in the gastrointestinal tract and liver (27,28), moderate amounts may have a "catalytic" effect on β-cell function. Studies indicate that fructose increases hepatic glucose uptake during the postprandial period when consumed in small amounts (29,30) and may be particularly beneficial in individuals with impaired glucose tolerance (31), perhaps analogous to our participants following consumption of the low-carbohydrate diet. Interestingly, substitution of sugar for refined grains (starch) in a high-carbohydrate diet tended to decrease the likelihood of abnormal glucose tolerance, as assessed at END, although this comparison is underpowered.

These findings have implications for dietary research, clinical care, and public health. First, many macronutrient feeding trials are short in duration, typically $<$1 month (8). Such trials may yield misleading findings about long-term dietary effects if the process of physiological adaptation to changes in macronutrients continues during data collection (8). Indeed, glycemia may comprise a gross biomarker of adaptation, relatively less sensitive to insulin action than lipolysis (32). Thus, ongoing adaptive changes in short-term trials could plausibly affect adipocyte biology, with relevance to substrate partitioning, energy expenditure, and body weight control. Second, regarding clinical protocols to prepare for an OGTT, the recommended 3-day period (with $\geq$150 g/day carbohydrate) (2,3,7,19) may be inadequate, giving rise to false-positive diagnoses of diabetes among people habitually consuming a low-carbohydrate diet. Third, the period immediately following transition to a high-carbohydrate diet among our participants may recapitulate planned deviations from a strict diet. Although a common strategy to enhance motivation and long-term adherence (33), such deviations when following a low-carbohydrate diet may result in marked postprandial hyperglycemia, with detrimental effects on endothelial function and oxidative stress (34,35).

This trial has several strengths. A structured feeding protocol, implemented with direct observation in a residential setting, ensured a high level of adherence to dietary interventions and appropriate differentiation in consumption of specified nutrients across diet groups. The test diets were controlled for dietary protein and energy intake, such that the analyses of CGM data were not confounded by the recognized effects of these variables on glucose levels (36). Collection of CGM data for more than a single 2-week sensor cycle is a notably longer protocol compared with commonly used protocols, particularly in populations without diabetes (37,38). High retention rate, likely reflecting intensive participant support, reduced the possibility of bias from missing data. However, despite lack of plausible mechanisms relating missingness to the outcomes of interest, we cannot say with absolute certainty that data are missing completely at random.

Several design limitations warrant consideration. The temporal glycemic trends within the high-carbohydrate diets could be affected by time-varying confounding, such as changing accuracy of the CGM system. However, the monitors were used according to manufacturer specification (e.g., before expiration date), and simultaneous collection of data from participants randomized to remain on a VLC diet serves as a relevant negative control. Also, regarding CGM, the FreeStyle Libre Pro system is well suited for evaluating patterns of change over time and absolute differences between sensor readings, but the device overestimates hypoglycemic events, suggesting that those values must be interpreted cautiously (39). Lack of frequently sampled insulin concentrations in the postprandial period limits mechanistic interpretation of results, especially related to β-cell function. With regard to generalizability, the participants were young to middle-aged adults, with overweight or obesity but otherwise healthy, who were able to adhere to the rigors of a feeding study to achieve substantial weight loss prior to randomization; as such, results cannot be directly translated to other populations. In addition, both high-carbohydrate diets contained high–glycemic index foods (refined grains or sugar); therefore, results may not be applicable to diets containing alternative sources of carbohydrate with lower glycemic indexes. Nevertheless, a high-carbohydrate diet containing 20% of total energy from refined grains (and 25% from whole grains) is consistent with the public health recommendation that less than one-half of all grains consumed be refined grains (40). Furthermore, the sample size was impacted by COVID-19 mitigations, including elimination of the OGTT for participants enrolled in the residential phase at the start of the pandemic, and then early study shutdown.

In conclusion, adaptation following transition from a low- to high-carbohydrate diet begins within 1 week but continues for several weeks to months thereafter, with implications for the conduct of dietary trials, the clinical diagnosis of diabetes, and the significance of planned deviations from popular low-carbohydrate diets. More research is needed to evaluate reproducibility and generalizability of the results from this exploratory study, including differences in patterns of adaptation with consumption of starch versus sugar and mechanisms underlying the adaptive process.

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