Dear Dr. Loder,

Thank you for the fast re-review and the opportunity to respond to several additional issues. We corresponded via email about how to address issues related to the Per Protocol analysis (Point #1) and energy intake analysis (Point #17), as described below. On most other points, we have made the requested change to the manuscript.

We've also made a few polishes to the text for clarity and consistency, all very minor, as shown in Track Changes. (Please let us know if you'd like a Track Changes version of the Supplement.) Formatting guidelines have been addressed in the prior revision.

Thank you,
David

The paper describes a carefully designed, executed and described clinical trial testing the effect of dietary composition on total energy expenditure following weight loss. I have some comments on the study design, analysis, presentation and interpretation.

1.   The Abstract has a focus on the per protocol analysis results that I mentioned and deprecated in my original notes on the paper. I presume the authors include them because the between-group contrasts are larger than for ITT, though not more significant. For me the ITT results should be the gold standard, and it weakens the impact of the paper to have to rely in the Abstract on PP results. Which leads to my second point.

We agree that a PP analysis may not be appropriate to include in the abstract of an effectiveness study, where non-compliance reflects real-life translation. However, for our mechanistically oriented study, the PP may offer a more precise estimate of the true effects of nutrients on biology (eliminating bias resulting from objectively defined non-compliance). For that reason, and as discussed in our email correspondence, we would like to keep the PP in the abstract. However, as previously requested, all results reporting (with the one exception, See Point #17) lead with the ITT analysis.

2.   The results in the Abstract are presented as the contrasts HI - MED and HI - LO. But the research question involves the effect of CHO% on TEE, and as such the linear trend in TEE across the three CHO% groups would be both more relevant and more parsimonious than the group contrasts. The authors point out that they include the comparison of Hi vs LO in Table 3, and state that it is algebraically equivalent to the linear trend. This would be true if the HI vs LO contrast were fitted using all the data, but not if it involved only the HI and LO groups. This needs to be confirmed.

In further discussion with our statistician, we confirmed that the HI vs. LO contrast was constructed using parameters fitted to all the data and therefore algebraically equivalent to a test for linear trend, as clarified in the text by adding the phrase "From parameters of the fitted model, taking account of all data ..." (page 12, para 1). We further clarify this point by adding the following: "The HI vs. LO comparison was equivalent to a test for linear trend across the three diets according to their equally spaced carbohydrate content" (also page 12, para 1).

We have also included data on the linear trend (by ITT analysis) to the abstract.

*3.	The authors do not give the linear trend any emphasis, but I should like to see the results presented as this trend. Normally it is hard to describe what is effectively an interaction in a clear way, but the concept is already mentioned in the sample size calculation: "a predicted effect of +50 kcal/d per 10% decrease in the contribution of dietary carbohydrate to total energy intake". The advantage of this approach is that the research question is tested with a single degree of freedom rather than two, which can be presented as an effect and 95% CI in the form above, and from Table 3 it is highly significant even for ITT.*

Linear trends and CIs are now included in the revised Table 3 for all endpoints. The linear trend for the primary outcome is now included in the abstract, per Point #2.

*4.	Omitting PP from the Abstract would save some words, allowing the Results to be expanded. The sentence "Main outcomes were pre-specified" is redundant, and would be better replaced with a list of other outcomes, including ghrelin and insulin secretion which appear later in the Abstract.*

We address the issue of the PP above (Point #1). We have deleted mention of pre-specification. We would defer to the editor as to whether ghrelin and leptin should be listed prior to presentation later in the abstract.

*5.	Some more minor points. The four time points in the study are labelled BSL, PWL, MID and END (Figure 1). However PWL looks (for me) like pre-weight-loss, which is confusing. I'd like to suggest instead labelling them PRE, START, MID and END, which should be less ambiguous.*

We agree and have made this change systematically throughout the manuscript, including Figure 1 showing study design.

*6.	Where the Abstract refers to "+91 kcal/d greater" it would be better without the '+'.*

Done

*7.	Page 9 What does "a physical activity factor of 1.5" mean?*

We have now included an explanation of this factor and how it was used (page 9, para 2), as follows: "We determined individual energy needs based on resting requirements, estimated using a regression equation, multiplied by a physical activity factor of 1.5 (which corresponds to a light activity lifestyle)."

*8.	Page 12 line 22 lists the design factors of study site, cohort and enrolment wave. However the Supplementary Methods also refer to obesity and Hispanic ethnicity – the two lists should match.*

In the Methods (page 12, para 1), we specify the design factors (study site, cohort, enrollment wave) that were included as covariates in the statistical analyses. In the Supplementary Online Content (*Implementation of Randomization*), we specify the stratification variables for randomization (feeding site, sex, ethnicity-race, age, BMI).

9.   Page 12 line 35 refers to weeks 10 and 20, which would be more consistent as MID and END.

Done

10.   Page 12 last line mentions 'hormone levels' without saying which they are.

We now specifically mention ghrelin and leptin at first mention of hormones in this section.

11.   Page 13 describes the log transformation of outcomes for analysis and back-transformation for reporting. It's a well-kept secret that the back-transformation is both unnecessary and over-complicated – see my paper https://urldefense.proofpoint.com/v2/url?u=https-3A__doi.org_10.1136_bmj.j3683&d=DwIFaQ&c=qS4goWBT7poplM69zy_3xhKwEW14JZMSdi oCoppxeFU&r=HzbHdpS-3grwpLx9r6E-9VusIayHmpyiieQcL7MXKaNp4MapU5y_XVbBHBTjdVvH&m=Bvjfk1CnL_DnsqBgnogBpmF9 2g4udFCKA6pr6GJvQpg&s=7B_W0iEfgidNnnS8Ko_UIYoR4X63I2jOLWmhgPOEJH8&e=. Multiplying the natural-log-transformed results by 100 they can be viewed as percentages – see for example the footnote to eTable 6 which laboriously concludes that -0.0101 on the log scale is "almost exactly -1%", and my paper shows it is a generalisable conclusion.

We followed the suggested approach exactly, as previously indicated in Table 3 (albeit perhaps a bit too briefly). We have clarified that footnote (g) and now include following passage in the text (page 13, para 1): "Concentrations … were log-transformed for analysis. For reporting, the adjusted mean and standard error were retransformed to the original units (exp(mean log) ± exp(mean log)×(exp(SE log)–1)), and changes were expressed in percentage units (100% × (exp(change in log)–1))."

12.   Which version of SAS?

The version is 9.4, as now indicated (page 13, para 3).

13.   Page 15 "at START, body weight did not differ across groups". Was this also true of weight loss?

Yes, as now indicated, with a *P*=0.65 (page 15, para 2)

14.   Give n = 120 (PP) as a percentage of ITT (n = 164 or 162).

We have now provided the requested information on page 15, para 2: 74% of the 162 included in the ITT for the primary outcome.

15.   Page 16 line 8 "all pre-specified covariates". Might be worth listing them here.

We have now provided the requested information (page 16, para 1): "... all pre-specified covariates (sex, ethnicity, race, and age; PRE values for BMI, percentage lean mass, and TEE; and weight loss from PRE to START): ..."

16.   Line 24 "the ... group effect retained statistical significance". Better to avoid mentioning significance at all.

Agree. We now simply say (page 16, para 1): "The relative insensitivity of TEE to the assumed value of FQ is shown in eTable 6, and the robustness of the observed effect of diet on TEE to substantial non-compliance is demonstrated in eTable 7"

17.   Page 17 top line implies that dietary intake was not measured in the ITT non-PP participants, which I'm sure was not the case.

We did obtain intake data on all participants, but analyzed these data only for the PP participants for reasons summarized in our prior Response Letter. To redo this analysis for the full ITT cohort would involve going back to the raw data and determining:

1. Calories served to the participants at meals and provided as snacks
2. Calories not consumed at the meals (from plate waste)
3. Calories not consumed in the snacks
4. Measures of non-compliance from the daily logs (i.e., meals not completely consumed at home, or food consumed off protocol)

These streams of information are in different files and would require significant effort and time to analyze. We were motivated to do so for the PP after the first round of review, because those data seemed to be relevant and informative. However, we question the relevance of those data for participants not in the PP (i.e., not maintaining weight loss) and by definition not in energy balance. Because these individuals were either gaining or losing weight rapidly, interpretation of their energy intake would be quite problematic.

Per our email exchange, and in the interests of avoiding delay in the resubmission, we understand it will be acceptable not to conduct this analysis for the manuscript.

18.   Page 18 the first 3 sentences need moving to the Methods.

To save space in print, we moved this material to the Supplemental Methods (*Study Outcomes – Additional Details*).

19.   The precision of numbers in the tables needs attention. My guidelines on this are here:
https://urldefense.proofpoint.com/v2/url?u=https-
3A__adc.bmj.com_content_100_7_608&d=DwIFaQ&c=qS4goWBT7poplM69zy_3xhKwEW14J
ZMSdioCoppxeFU&r=HzbHdpS-3grwpLx9r6E-

9VusIayHmpyiieQcL7MXKaNp4MapU5y_XVbBHBTjdVvH&m=Bvjfk1CnL_DnsqBgnogBpmF9 2g4udFCKA6pr6GJvQpg&s=Jk9nUTZFffaNo-qvx-1-z0hik0pOR-QA_FHJRrtf7tg&e=. *In Table 1 % appears inconsistently in the body of the table. I doubt that CHO is accurate to 0.1g and suggest rounding. Food quotient would be better than FQ, and why the extraneous 3 columns? If the values are averages per 2000 kcal, why are the kcal 2001?*

We have revised data presentation per the preferred style. Macronutrients are expressed in gram amounts (to the nearest integer) and as percent of energy (one decimal place, for consistency, as some values are <10%).

We now clarify in the title that data are calculated daily averages (for a 2,000-kcal target). The footnote specifies that we performed the calculations using Food Processor Nutrition Analysis Software (ESHA Research Inc., Salem, OR).

20.   Table 2 would be easier to read with two significant digits for the percentages, i.e. rounded for ≥10% and one decimal place for <10%. The same applies to eTable 4.

Done

21.   Table 3, give full names rather than acronyms for the outcome measures. Replace the final column with results for the linear trend (effect and 95% CI), I suggest per 10% change in CHO.

Done

22.   In eTable 7 fat would be better rounded.

Done

23.   eFigure 1 can be simplified by combining the ITT and PP graphs and highlighting (using colour or linetype) the individuals who are ITT but not PP. It would also make it easier to see if such individuals were a biased sample.

We attempted to produce a combined figure as suggested but were disappointed to find that it gave a cluttered appearance and did not clarify the issue of biased exclusion as hoped. We believe that the question of biased exclusion is answered more satisfactorily by the statistical analysis cited in eTable 4, where we demonstrate a similar distribution of covariates in excluded and included subjects, and by a newly added comparison of weight change across diet groups in both the ITT and PP analyses (page 15, para 2). For purposes of illustration, we think the adjacent display offers a better format for comparison and contrast.

24.   Figure 4 and eFigures 2-3 deserve more informative labels on the x-axis.

Done. We've also included a new Supplement figure on insulin resistance which provides additional insights on effect modification.