Supplemental Material

Urinary Glyphosate Concentrations among Pregnant Participants in a Randomized, Crossover Trial of Organic and Conventional Diets

Carly Hyland, Meredith Spivak, Lianne Sheppard, Bruce P. Lanphear, Michael Antoniou, Maria Ospina, Antonia M. Calafat, and Cynthia L. Curl

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Food Log – Week 1

We ask that you try to eat your meals and snacks from the groceries that were delivered to you, however we understand that is not always realistic. Please fill out the following form for each day:

Date	Approximately how much of the food you ate today was from the groceries provided by the study? (Check one box for each day):	Please make note of any food or drinks you ate or drank that were not from the groceries provided by the study, including any meals from restaurants:
Tuesday June 15	☐ **Everything** I ate was from the study ☐ **Most** of what I ate was from the study ☐ **About half** of what I ate was from the study ☐ **A little bit** of what I ate was from the study ☐ **None** of what I ate was from the study	
Wednesday June 16	☐ **Everything** I ate was from the study ☐ **Most** of what I ate was from the study ☐ **About half** of what I ate was from the study ☐ **A little bit** of what I ate was from the study ☐ **None** of what I ate was from the study	
Thursday June 17	☐ **Everything** I ate was from the study ☐ **Most** of what I ate was from the study ☐ **About half** of what I ate was from the study ☐ **A little bit** of what I ate was from the study ☐ **None** of what I ate was from the study	
Friday June 18	☐ **Everything** I ate was from the study ☐ **Most** of what I ate was from the study ☐ **About half** of what I ate was from the study ☐ **A little bit** of what I ate was from the study	

	☐ **None** of what I ate was from the study	
Saturday June 19	☐ **Everything** I ate was from the study ☐ **Most** of what I ate was from the study ☐ **About half** of what I ate was from the study ☐ **A little bit** of what I ate was from the study ☐ **None** of what I ate was from the study	
Sunday June 20	☐ **Everything** I ate was from the study ☐ **Most** of what I ate was from the study ☐ **About half** of what I ate was from the study ☐ **A little bit** of what I ate was from the study ☐ **None** of what I ate was from the study	
Monday June 21	☐ **Everything** I ate was from the study ☐ **Most** of what I ate was from the study ☐ **About half** of what I ate was from the study ☐ **A little bit** of what I ate was from the study ☐ **None** of what I ate was from the study	

Protocol for Scoring Participants' Self-Reported Dietary Compliance Records

Participants were asked to complete a brief form describing their compliance each day of the dietary intervention, including a) self-rating the proportion of food they consumed each day that was provided by the study on a 5-point Likert Scale and b) writing down the specific foods they consumed each day that were not provided by the study. This protocol describes the process for a) recording scores from the Likert Scale and b) systematically quantifying and scoring the amount of food that the participants indicated they ate from outside of that which was provided by the study, in order to assess each participant's compliance with the organic diet intervention.

Key Definitions

Conventional Foods: Food items that do not bear the label "USDA organic." Participants received one week of conventional food from the study.

Food Series: A group of foods that was determined to have been consumed together based on the use of commas or other punctuation and/or is differentiated from other foods by white space and/or lines.

Meal: A Food Series that has been eaten for breakfast, lunch or dinner; contains at least three distinct food groups and/or a central protein or carbohydrate.

Organic Food: Food items bearing the label "USDA Organic." Participants received one week of organic food from the study.

Snack: A Food Series that is not eaten as a full meal; contains fewer than three food groups (often only one food group) and is frequently energy dense and/or quick to eat.

Introduction

The goal of this protocol is to describe a method for quantifying self-reported compliance with the dietary intervention during the week that participants received organic food. This protocol provides a series of instructions to record and sum participants' self-reported scores of their own compliance using a 5-point Likert Scale, as well as to quantify the amount of conventional food that participants reported having consumed. This study was designed as a randomized crossover trial, with each participant receiving one week of organic food and one week of conventional food, randomized to order. A study inclusion criterion was that participants typically ate conventional diets; thus, the following instructions will only be applied to the week in which participants received organic food. It is assumed that participants ate 100% conventional food during the conventional week, whether or not that food was provided by the study. However, in order to train study staff to conduct this scoring, this protocol was applied first to the conventional forms; subsequently, following protocol development, it is used for the organic forms.

This protocol describes a system in which participants are assigned a maximum number of points if they report eating only food from the study (or if they note that any food consumed from outside the study was organic). In this system, points are subtracted for each meal (2 points) or snack (1 point) the participants report during the organic week that was not designated as

organic (i.e., if the participant indicated the meal/snack was conventional, or if they did not include a designation). This system employs an assumption that participants consumed three meals and two snacks per day, as has been recommended for pregnant women (Misan et al., 2019; Siega-Riz et al., 2001), and we will characterize each Food Series listed by a participant as either a "meal" or a "snack", based on the criteria provided below. Participants were instructed to prioritize purchasing "primary" items they anticipated eating for meals and snacks, and to secondarily purchase beverages (e.g., water, soda, tea, milk, coffee), condiments, and spices if they had enough money from the study available. Beverages, spices, and condiments will be excluded from the scoring because not all participants purchased these items as part of the study, and because the mass of these items is expected to be minimal in comparison with the mass of the food consumed. If no food items are reported to have been sourced from outside the study, or if any items consumed from outside the study are described as organic, the participant will be assigned 8 points for that day ([3 meals x 2 points] + [2 snacks x 1 point]). We will sum participants' total daily scores over the organic week, for a maximum score of 56 points.

Step One: Recording Scores from the Likert Scale (Self-Reported Compliance with the Dietary Intervention)

The dietary compliance form contains a single sheet of paper with two columns for each day: 1) a Likert Scale, where participants self-rated approximately how much of the food they ate that day was from the groceries provided by the study, and 2) space for participants to write in any food they consumed each day that was not provided by the study. Raters should begin with the self-reported scores from the 5-point Likert Scale found on the left side of the form. Raters should use the Excel scoring sheet to record all scores (link provided *here*), according to the following instructions:

1. Record the participant ID at the top of the scoring box.
2. Record the participant's self-reported score from the Likert Scale:
   - If the option "Everything I ate was from the study" was circled, assign a score of 5.
   - If the option "Most of what I ate was from the study" was circled, assign a score of 4.
   - If the option "About half of what I ate was from the study" was circled, assign a score of 3.
   - If the option "A little bit of what I ate was from the study" was circled, assign a score of 2.
   - If the option "None of what I ate was from the study" was circled, assign a score of 1.
   - It is possible that a participant did not circle any of the Likert Scale options. If this occurs, the rater should write "NA" in the corresponding cell for that day.
   - It is possible that a participant will have selected two Likert Scale options, corresponding to two scores (e.g., a participant circled 3 and 4). If this scenario occurs, the average of the two scores should be calculated and that number should be documented in the Likert Scale scoring column for that day.

3. Begin by scoring Day 1, then Day 2 and so on until Day 7 is completed. Day 1 will correspond to June 15th if the participant was assigned to receive organic food during the first week of the dietary intervention, or to June 23rd if the participant was assigned to receive organic food the second week of the dietary intervention. <u>It is important to note that the first day listed on the form for participants who were assigned to the organic week during the second week of the dietary intervention is June 22nd. However,</u> **raters should ignore June 22nd** <u>and begin with June 23rd, as June 22nd was a wash-out day.</u>

4. Sum the daily scores and record the total score in the "Totals" cell at the bottom of the Likert Scale column on the scoring sheet for each participant. If "NA" is written in <u>any</u> of the daily cells, then the rater should write "NA" in the "Totals" cell at the bottom of the Likert Scale column on scoring sheet for that participant.

Step Two: Determine which foods were consumed together

First, determine which foods recorded by the participants in the cells of the second column were consumed together. Several different foods that were consumed together will be referred to hereafter as a Food Series. Raters will determine whether or not these foods were consumed together based on the observed use of lines, white space and commas. A list of foods that is connected by commas and/or separated from other foods by white space and/or a line on the page will be analyzed together as a Food Series. The list of foods that raters determine to constitute a Food Series may be written either horizontally or vertically. Participants who consumed multiple meals or snacks that were not provided by the study should have several Food Series written in. It is important to note that participants may write in just one food, or even a restaurant name, in the cell of the second column, or separated from other foods by white space, commas or lines. These single foods and/or restaurant names should still be considered a Food Series, even though they consist of only one food or restaurant name.

Step Three: Determine whether a Food Series was organic or conventional

The next step is to determine if a given Food Series was organic or conventional. If participants wrote down a Food Series and specified that it was organic, the Food Series should be documented in the "Excluded Items" column of the scoring sheet and no points will be deducted. Participants may have also written down food without indicating if it was organic or conventional. In these cases, it will be assumed that the Food Series is conventional because of the aforementioned study inclusion criterion.

Step Four: Determine whether a Food Series constitutes a meal or a snack

Differentiating between a meal and a snack can be subjective, but the goal of this section is to provide instructions to make this distinction as consistent as possible. Studies that have examined relationships between snacking and health have noted that snacking generally occurs between meals, with the consumption of somewhat smaller portions of different types of foods than meals (Wadhera & Capaldi, 2012; Barnes et al., 2015; Hampl, Heaton & Taylor, 2003). A food that is eaten on its own may be considered a snack, while this same food may be considered

part of a meal when eaten with multiple foods or food groups. Thus, it is crucial to have established which foods go together as part of a Food Series.

The FDA has different qualifications regarding which food groups must be consumed in the same sitting for a meal to constitute breakfast versus lunch/dinner. Specifically, a meal that constitutes breakfast must contain food from the following three food groups: dairy (e.g. yogurt, cheese), vegetable and/or fruit, and grains (e.g., rice, bread, cereal, pasta). A meal that constitutes lunch or dinner must contain food from the following five food groups: dairy, meat/meat alternatives (e.g., chicken, beef, tofu, legumes), vegetable, fruit, and grains.

It should be noted that participants were not asked to clarify which Food Series corresponded to which meal (breakfast, lunch or dinner); they were only instructed to write in all foods they consumed that were not part of the study. Thus, for scoring purposes, a Food Series will be classified as either a meal or a snack by combining the FDA guidelines for breakfast and dinner (see bulleted list below). The type of meal (breakfast, lunch or dinner) will not be determined. The following guidelines provide the criteria to classify a Food Series as either a meal or a snack.

For a Food Series to be classified as a meal, any <u>one</u> of the following criteria must be met:

1. The Food Series contains at least three of the five food groups outlined by the FDA: dairy (e.g., cheese, yogurt), meat/meat alternatives (e.g., bacon, eggs, chicken, steak, legumes), vegetable, fruit, or grains (e.g., oatmeal, bread, rice, pasta).
2. The Food Series is carbohydrate-centered, meaning that the Food Series includes a central carbohydrate such as oatmeal, bread, rice or pasta.
3. The Food Series is protein-centered, meaning that the Food Series includes a central protein, such as eggs, bacon, chicken, steak or legumes.
4. The Food Series consists solely of a restaurant name, such as McDonald's or Applebee's, under the assumption that if the participant went to a restaurant, it was to consume a meal rather than a snack. However, if other foods preface or follow the restaurant name, those foods should be analyzed for the presence of a Food Series per Step Two and whether that Food Series constitutes a meal or a snack, using the criteria in this step.

For a Food Series to be classified as a snack, any <u>one</u> of the following criteria must be met:

1. The Food Series consists of a single, prepackaged item (e.g., applesauce)
2. The Food Series consists of a single protein-based item (e.g., beef jerky)
3. The Food Series consists of a single, dairy-based item (e.g., yogurt or cheese)
4. The Food Series consists of a single carbohydrate-based item (e.g., granola bar)
5. The Food Series consists of an energy-dense food item (meaning that food contains a high amount of calories, salt, fat or sugar; e.g., trail mix, potato chips or cookies)
6. The Food Series consists of a single fruit or vegetable (e.g., apple, carrots)
7. The Food Series consists of a single ingredient that is part of, but does not on its own constitute, a meal (e.g., pork meat or rice)
8. The Food Series does not meet any of the criteria for a meal

Step Five: Scoring Self-Report of Foods Consumed from Outside of the Study

Raters should again use the Excel scoring sheet to record all scores (link provided *here*). Scoring should then proceed as follows:

1. Each participant will start each day with eight points. For each non-organic meal the participant reported, subtract two points for that day. For each non-organic snack the participant reported, subtract one point for that day. Beverages, including, but not limited to, water, milk, juice, coffee and soda, should be documented in the "Excluded Items" column on the scoring sheet and should not be scored. Any spices and condiments should also be documented in the "Excluded Items" column and should not be scored. Any organic Food Series will have already been documented in the "Excluded Items" column in Step Three and should not be scored.
   a. Theoretically, a participant could report so many conventional (or presumed conventional) meals and snacks that they end up with a negative score for a given day (e.g., three non-organic meals and three non-organic snacks, which would result in a subtraction of 9 points, and would thus receive a score of -1 for that day). In this event, record the true score (e.g., -1) in the "True Score" column on the Excel sheet. Additionally, record a "Truncated Score", with a minimum of 0, in the "Truncated Score" column on the Excel sheet (i.e., for any participants who receive a score of ≤ 0 for that day).
2. Sum the daily scores and record the total score in the "Totals" cell at the bottom of the "Truncated Score" column. True scores will *not* be added together and totaled; they will simply be recorded.
3. Provide a thorough explanation of why any points were subtracted in the notes section. Each cell in the notes column already has a structure for recording notes for a single Food Series in place, but if there are multiple Food Series, then the rater should simply copy and paste the notes structure below the first set of notes. To complete the notes column:
   a. List the foods determined to be in the Food Series
   b. Note whether the Food Series was determined to be a meal or snack
   c. Provide the criteria that was met by the Food Series that was used to make the designation of either a meal or a snack
4. Any beverages, spices, condiments or organic Food Series that the participant wrote down should be documented in the Excluded Items column.
5. The highest and lowest possible scores are explained below:
   a. The highest possible score a participant can receive for the week is 56 points. A participant who receives 56 points would have received 8 points per day for each of the seven days; thus, they would have only eaten food provided by the study (or would have noted that any other food consumed was organic).
   b. The lowest possible score a participant can receive is 0 points. A participant who ate three meals and two snacks (or more) that were not provided by the study for

each of the seven days of the intervention would receive daily totals of 0 points, and thus a weekly total of 0 points.

Step Six: Comparing Scores between Raters

There will be two raters for each dietary compliance form, who will separately rate each participant-day and calculate total scores for each participant. After the raters have completed the scoring for an initial subset of the conventional week's forms for 10 participants, we will assess score consistency between raters and the potential need for modifications and additional clarity to the protocol. After a discussion of any initial scoring differences and possible indications for protocol modifications, the remainder of the conventional week's forms will be scored. Again, we will assess consistency in scoring and clarity of the protocol. In the event that any scores are discordant, raters will compare their notes section to determine how each rater selected the specific foods that constituted the Food Series for which they subtracted points, and whether that Food Series was a meal or a snack, in order to reach a conclusion. It is also possible that raters will have selected the same type of food series (meal/snack), but will have selected different criteria justifying their decision. If this scenario occurs, raters should discuss the differences, but do not need to agree on a final criterion, as it is possible that a food series may meet more than one criterion. If necessary, modifications to the protocol will be made for any systematic differences in scoring between the two raters. Finally, the two raters will score the dietary compliance forms for the weeks when organic food was provided to the participants. Interrater reliability will then be assessed for the organic dietary compliance forms. Scores do not need to be perfectly concordant, as scores will be used to place participants in categories for sensitivity analyses. However, we aim to achieve an interrater reliability that is ≥ 80%, which will be assessed using both percent agreement and the kappa statistic.

Table S1. Change in urinary glyphosate concentrations from conventional diet to organic diet in secondary analyses to restricted to participants who complied with intervention based on *a priori* criteria,[1] stratified by far (> 5 km) vs. near (≤ 0.5 km) field residential location

A priori exclusion	Median (IQR) Percent Change from Conventional to Organic Diet			Wilcoxon signed rank test *p*-value[2]		
	All participants	Far field	Near field	All participants	Far field	Near field
Didn't turn in Food Log *and* didn't write anything for food consumed from outside of study (*n*=33; 18 near-field and 15 far-field)[3]	-18.4 (-36.0, 17.5)	-24.2 (-37.4, -16.6)	1.4 (-36.0, 30.2)	0.12	0.01	0.91
Didn't turn in Food Log *and* didn't write anything for food consumed from outside of study *and* had a Likert scale <22 (*n*=24; 11 near-field and 14 far-field)[4]	-18.4 (-42.5, 15.7)	-24.2 (-37.4, -17.0)	13.8 (-47.7, 30.2)	0.11	0.02	0.86
Didn't turn in Food Log *and* didn't write anything for food consumed from outside of study *and* scored <36 on writing foods down on Food Log (*n*=27; 12 near-field and 15 far-field)[5]	-20.3 (-37.4, 13.8)	-29.1 (-37.4, -17.0)	1.4 (-41.9, 43.0)	0.06	0.01	0.94
Didn't turn in Food Log *and* who had a Likert scale <22 (*n*=27; 12 near-field and 15 far-field)[6]	-20.3 (-48.8, 13.8)	-29.1 (-48.8, -17.0)	1.4 (-49.4, 30.0)	0.03	0.01	0.53
Didn't turn in Food Log *and* missing ≥ 4 samples *and* <22 on Likert (*n*=26; 12 near-field and 14 far-field)[7]	-19.5 (-48.8, 13.8)	-27.9 (-48.8, -17.0)	1.4 (-48.4, 30.0)	0.03	0.02	0.53

[1]Detailed calculation of each criterion available in Food Log Protocol.
[2]*p* value comparing glyphosate concentrations from conventional to organic week
[3]3 participants missing Food Log; 3 participants didn't write down outside food
[4]3 participants missing Food Log; 3 participants didn't write down outside food; 9 participants had Likert scale <22
[5]3 participants missing Food Log; 3 participants didn't write down outside food; 6 participants had score <36 on Food Log
[6]3 participants missing Food Log; 9 participants had Likert scale <22
[7]1 participant missing ≥ 4 samples; 3 participants missing Food Log; 9 participants had Likert scale <22

Table S2. Urinary glyphosate concentrations from NHANES and previous studies of pregnant women (μg/L)

Study	Population	Mean/GM	Median	IQR	Range
Current study[1]	39 pregnant women in Idaho (conventional and organic diet phases) (recruited 2021)	0.18	0.19	0.11-0.27	0.05-0.99
Lesseur et al., 2021 (TIDES Cohort)[1]	94 pregnant women from four US university-based medical centers (recruited 2010-2012)	0.33	0.22	0.12-0.52	0.01-1.90
Parvez et al., 2018[2]	71 pregnant women in central Indiana (recruited 2015-2016)	3.40	3.25	2.84-3.91	0.50-7.20
Silver et al., 2021 (PROTECT Cohort)[1]	247 pregnant women from case-control study in Puerto Rico (recruited 2011-2017)	0.49	0.50	0.31-0.82	0.22-2.73[3]
2013-2014 NHANES[2,4]	2,309 samples from individuals ages 6 years and older from NHANES	0.42	0.41	0.27-0.68	0.14-8.13
2015-2016 NHANES[2,4]	2,428 samples from individuals ages 3 years and older from NHANES	0.37	0.35	0.14-0.65	0.14-8.21

Abbreviations: GM, Geometric Mean; IQR, Interquartile Range (25th-75th percentile); NHANES, National Health and Nutrition Examination Survey; PROTECT, Puerto Rico Testsite for Exploring Contamination Threats; TIDES, The Infant Development and the Environment Study
[1]Specific gravity-adjusted values; values <LOD included
[2]Uncorrected; values <LOD included
[3]10th percentile reported (rather than minimum value)

Table S3. Specific gravity-adjusted urinary glyphosate concentrations on log-log scale and percent change between conventional and organic week among 39 pregnant participants in a randomized crossover conventional vs organic dietary intervention trial. Data used for Figures 3 and 4.

Concentration Conventional Week[1]	Concentration Organic Week[1]	Field within 0.5 km of home[2]	Exclusion criteria[3]	Percent Change from Conventional to Organic
0.114184	0.104135	0	0	-8.80
0.096134	0.084422	1	0	-12.18
0.107994	0.096134	1	0	-10.98
0.110141	0.113138	0	0	2.72
0.339404	0.230633	0	0	-32.05
0.191795	0.160042	0	0	-16.56
0.101078	0.046981	0	0	-53.52
0.119649	0.186439	1	0	55.82
0.079196	0.056187	0	1	-29.05
0.1638	0.07629	0	0	-53.43
0.150147	0.284308	0	1	89.35
0.1087	0.086621	0	0	-20.31
0.73445	0.564713	1	0	-23.11
0.182768	0.095582	1	0	-47.70
0.22045	0.112896	0	0	-48.79
0.368618	0.09783	0	0	-73.46
0.287173	0.234383	1	0	-18.38
0.087188	0.152524	1	0	74.94
0.206815	0.31701	0	1	53.28
0.237002	0.308589	1	0	30.21
0.188161	0.142586	0	0	-24.22
0.392773	0.153561	1	0	-60.90
0.19289	0.250429	1	0	29.83

0.314754	0.215386	0	0	-31.57
0.301379	0.147637	1	0	-51.01
0.114113	0.214329	1	1	87.82
0.54048	0.381261	0	0	-29.46
0.143482	0.278127	1	0	93.84
0.09783	0.210519	0	0	115.19
0.236416	0.30091	1	0	27.28
0.128604	0.104534	0	0	-18.72
0.315304	0.261756	0	0	-16.98
0.412208	0.986857	1	0	139.41
0.189734	0.216	1	0	13.84
0.166311	0.195449	1	0	17.52
0.232165	0.089899	1	0	-61.28
0.187833	0.1176	0	0	-37.39
0.586155	0.26638	1	0	-54.55
0.12089	0.077354	1	0	-36.01

[1]Specific gravity adjusted urinary glyphosate concentrations with values <LOD imputed as $\frac{LOD}{\sqrt{2}}$

[2]0=no field within 0.5 km of home; 1=field within 0.5 km/met exclusion criteria

[3]0=did not meet exclusion criteria; 1=did meet exclusion criteria (missing $\geq$ 4 samples or Food Log from either week)