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Influence of an intervention targeting a reduction in sugary beverage intake on the $\delta^{13}\text{C}$ sugar intake biomarker in a predominantly obese, health-disparate sample

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Abstract

Objective—Controversy exists surrounding the health effects of added sugar (AS) and sugar-sweetened beverage (SSB) intakes, primarily due to a reliance on self-reported dietary intake. The purpose of the current investigation was to determine if a 6-month intervention targeting reduced SSB intake would impact $\delta^{13}\text{C}$ AS intake biomarker values.

Design—A randomized controlled intervention trial. At baseline and at 6 months, participants underwent assessments of anthropometrics and dietary intake. Fasting fingerstick blood samples were obtained and analysed for $\delta^{13}\text{C}$ value using natural abundance stable isotope MS. Statistical analysis included descriptive statistics, correlational analyses and multilevel mixed-effects linear regression analysis using an intention-to-treat approach.

Setting—Rural Southwest Virginia, USA.

Subjects—Adults aged 18 years who consumed 200 kcal SSB/d (837 kJ/d) were randomly assigned to either the intervention (n 155) or a matched-contact group (n 146). Participants (mean age 42.1 (SD 13.4) years) were primarily female and overweight (21.5 %) or obese (57.0 %).

Results—A significant group by time difference in $\delta^{13}\text{C}$ value was detected ($P < 0.001$), with mean (SD) $\delta^{13}\text{C}$ value decreasing in the intervention group (pre: -18.92 (0.65) ‰, post: -18.97 (0.65) ‰) and no change in the comparison group (pre: -18.94 (0.72) ‰, post: -18.92 (0.73) ‰). Significant group differences in weight and BMI change were also detected. Changes in biomarker $\delta^{13}\text{C}$ values were consistent with changes in self-reported AS and SSB intakes.

Conclusions—The $\delta^{13}\text{C}$ sugar intake biomarker assessed using fingerstick blood samples shows promise as an objective indicator of AS and SSB intakes which could be feasibly included in community-based research trials.

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Keywords

Added sugar; Sugar-sweetened beverage; Dietary biomarker; Isotope; Dietary assessment

In spite of a large body of evidence linking sugar-sweetened beverage (SSB) intake with adverse health outcomes, including increased risk of type 2 diabetes^(1,2), weight gain^(1,3) and obesity^(1,2), significant controversy exists surrounding the health effects of added sugar (AS) and SSB^(1,4,5) due primarily to a reliance on self-reported dietary intake methodologies^(5–7). This controversy has recently extended into the health and economic policy arena (e.g. references 8–10). Public health guidelines have recommended that consumers limit AS intake and replace SSB with water⁽¹⁾, yet critics of these guidelines cite the use of research utilizing memory-based dietary recall methods as a fatal flaw, with significant public health consequences⁽⁸⁾. The availability of an objective indicator of dietary intake, such as an AS intake biomarker, could overcome this research limitation⁽¹⁰⁾.

Because corn and cane plants employ the rare C₄ photosynthetic pathway, their sugars naturally contain a high concentration of ¹³C (the heavy stable isotope of carbon) relative to ¹²C⁽¹¹⁾. After digested food is absorbed from the intestine into the bloodstream, the carbon within food becomes carbon within tissues. Thus, a high ¹³C:¹²C (measured as ‘δ¹³C’) in human blood may reflect a high ¹³C:¹²C in the diet, with corn and cane sugars as important contributors⁽¹¹⁾. Because approximately half of AS is consumed in the form of SSB⁽¹²⁾ and the majority of the AS consumed is ultimately derived from corn and cane plants⁽¹¹⁾, the δ¹³C value of human blood may be affected by SSB intake^(11,13,14).

Talking Health is a randomized controlled trial which aimed to reduce SSB intake among residents of rural, health-disparate communities⁽¹⁵⁾. Participants were randomly assigned to either a 6-month intervention targeting a reduction in SSB intake (SIPsmartER) or a matched-contact comparison group (MoveMore). This trial represents the first randomized controlled trial directly targeting SSB intake reduction to include assessments of the δ¹³C biomarker, using fingerstick blood samples⁽¹³⁾. Our objective was to determine if group differences in biomarker δ¹³C values were evident over the 6-month intervention period.

Materials and methods

Detailed design and methods for the Talking Health trial have been reported previously⁽¹⁵⁾. Briefly, eligible individuals (English-speaking adults ≥ 18 years of age, who consumed ≥ 200 kcal SSB/d (837 kJ/d) and self-reported no contraindications for physical activity) were recruited and enrolled from eight counties/cohorts in Southwest Virginia, USA, each spaced approximately 3 months apart. Within each cohort, participants were randomly assigned to SIPsmartER (*n* 155) or MoveMore (*n* 146). For randomization, equal numbers of envelopes were prepared containing the name of each study condition; participants selected a sealed envelope to determine their assigned condition. SIPsmartER targeted decreasing SSB consumption, with the primary goal of ≤ 8 fluid ounces (237 ml) per day. MoveMore targeted physical activity promotion and did not contain content related to SSB or other dietary factors. Conditions were matched in terms of contact (i.e. three small-group classes, one live teach-back call, eleven interactive voice response calls) and structure⁽¹⁵⁾.

Participants were compensated with a gift card for completing assessments (baseline, \$US 25; month 6, \$US 50).

At baseline and at 6 months, participants underwent assessments of height and weight, measured in light clothing without shoes (scale model 310GS; Tanita, Tokyo, Japan). AS sugar and SSB intakes were assessed using three 24 h recalls, obtained using the five-step multiple pass method⁽¹⁶⁾. Recalls were collected within a two-week baseline testing period. Recalls were analysed using nutritional analysis software (Nutrition Data System for Research (NDS-R 2011), University of Minnesota, Minneapolis, MN, USA). The NDS-R database primarily utilizes the US Department of Agriculture's Nutrient Data Laboratory for its food composition data, and is supplemented by information from food manufacturers and data available in the scientific literature. Imputation procedures are applied to minimize missing values. The database is 100 % complete for AS⁽¹⁷⁾.

Fasting blood samples were obtained via routine fingersticks (One Touch Fine Point Lancet; Johnson & Johnson Company, New Brunswick, NJ, USA). Blood samples were blotted on to sterilized binder-free glass microfibre filters (Whatman, type GF/D, 2.5 cm diameter; Whatman, Inc., Piscataway, NJ, USA), air-dried and then analysed for $\delta^{13}\text{C}$ value using natural abundance stable isotope MS, as per our previous work^(13,14). Samples were analysed in triplicate; the analytical error associated with each measurement in the current investigation was 0.039 ‰. Stable isotope values are reported using standard δ -notation in units of 'per mil' (‰) relative to international standards (Vienna Pee Dee Belemnite (VPDB)). The unit 'per mil' is standard within stable isotope reporting and refers to the number of units out of 1000; similar to how 'per cent' refers to the number of units out of 100. Human blood samples have lower $\delta^{13}\text{C}$ values than the VPDB standard, therefore the $\delta^{13}\text{C}$ values presented here are less than 0. A $\delta^{13}\text{C}$ value of human blood that is closer to 0, representing a higher $^{13}\text{C}:^{12}\text{C}$ in the diet, corresponds to a higher AS or SSB intake^(11,14,18). Alanine was used as an internal laboratory standard for carbon. A more detailed description of this biomarker technique, which includes the $\delta^{13}\text{C}$ values of several dietary sources of AS and SSB, as well as foods with 'low' $\delta^{13}\text{C}$ values which may contribute to total sugar intake (e.g. fruit), has been previously published⁽¹⁸⁾.

Five women who were pregnant were excluded from the present analysis (n 4, SIPsmartER; n 1, MoveMore). Descriptive statistics were used to summarize baseline demographic characteristics. The t test was used to compare group means; the χ^2 test was used to compare proportions across groups. Multilevel mixed-effects linear regression analyses were performed using the statistical software package Stata version 13 (2013) to account for clustering of individuals within cohorts. Results of intention-to-treat are presented. The models included controls for the following baseline covariates: age, gender, race/ethnicity, income, education level, health literacy level, employment status, number of children, smoking status and BMI. Correlational analyses were also performed.

Results

Demographic characteristics of participants (n 296) are presented in Table 1. Participants (mean age 42.1 (SD 13.4) years) were primarily female and Caucasian, and almost half of

the sample (43 %) reported an annual household income of \$US 14 999. Most participants were overweight (21.5 %) or obese (57.0 %). There were no significant demographic differences between groups. At month 6, the overall retention rate was 74 %, which was not statistically different between conditions. Group differences over time (both $P < 0.05$) were noted in BMI and weight, which decreased in SIPsmartER (-0.21 (95 % CI $-0.35, -0.06$) kg/m^2 , -0.5 (95 % CI $-0.9, 0.0$) kg) but did not change in MoveMore (0.10 (95 % CI $-0.09, 0.30$) kg/m^2 , 1.0 (95 % CI $-0.2, 0.4$) kg).

Changes in biomarker $\delta^{13}\text{C}$ values and self-reported dietary intake over the 6-month intervention period are presented in Table 2. A significant group by time difference in $\delta^{13}\text{C}$ value was detected, with mean $\delta^{13}\text{C}$ value decreasing (i.e. reflecting a reduction in AS and SSB intakes) in the intervention group (Table 2). Group changes in biomarker $\delta^{13}\text{C}$ values over time were largely consistent with changes in self-reported AS and SSB intakes. Total sugar intake declined in the intervention group and there was a significant group difference over time in total sugar intake. Significant correlations were noted between self-reported SSB intake and $\delta^{13}\text{C}$ values at baseline ($r = 0.259$, $P < 0.001$) and at month 6 ($r = 0.280$, $P < 0.001$). Changes in self-reported SSB intake and $\delta^{13}\text{C}$ values were not significantly associated ($r = 0.066$, $P = 0.261$), nor were changes in self-reported AS intake and $\delta^{13}\text{C}$ values ($r = 0.041$, $P = 0.487$). The correlation between changes in percentage of total energy from AS and $\delta^{13}\text{C}$ value was not statistically significant ($r = 0.101$, $P = 0.084$). As expected, changes in total sugar intake and $\delta^{13}\text{C}$ values were not significantly associated (total sugar, percentage of total energy: $r = 0.099$; total sugar, grams: $r = 0.040$; both $P = 0.05$).

To determine if weight loss impacted our findings related to changes in biomarker values, we developed a separate model to predict change in $\delta^{13}\text{C}$ value for each group and included changes in weight, AS and SSB intakes as predictors. The model for the MoveMore comparison group was not significant ($P = 0.4695$). The model for the SIPsmartER group was significant ($P = 0.008$) and the only significant predictor of change in $\delta^{13}\text{C}$ value was change in SSB kcal ($P = 0.019$). This suggests that weight loss does not impact change in $\delta^{13}\text{C}$ values when holding SSB and AS intake changes at the same level.

Discussion

The Institute of Medicine and others have highlighted the need for novel methods to objectively assess dietary intake, including biomarkers of food and nutrient intakes^(10,19). Common limitations of existing biomarker techniques include cost, participant burden and degree of invasiveness⁽²⁰⁾. The current investigation describes results from the first randomized controlled trial evaluating an SSB intake reduction intervention to include assessment of the $\delta^{13}\text{C}$ sugar intake biomarker with particular application to SSB. Group differences in biomarker $\delta^{13}\text{C}$ values and in self-reported AS and SSB intakes were detected over the intervention period, and in weight and BMI. Importantly, this biomarker was evaluated using minimally invasive fingerstick blood samples obtained in a field research setting. These findings suggest that the $\delta^{13}\text{C}$ value of fingerstick blood shows promise as a biomarker of AS –and by extension – SSB intake, which could be feasibly included in large-scale, community-based research trials.

Mean $\delta^{13}\text{C}$ value in this sample was higher than that reported in a university community⁽¹⁴⁾, which is expected given the high SSB consumption reported by participants (~400–500 kcal/d; 1674–2092 kJ/d). Although the significant correlations between biomarker values and self-reported SSB intake at baseline and at month 6 were considered modest (i.e. $r \sim 0.3$)⁽²¹⁾, these correlations may underestimate biomarker validity due to dietary intake under-reporting⁽²²⁾, which is a particular problem when reporting intake of socially undesirable foods such as SSB^(10,23).

Strengths of the current investigation include a large sample size, the low degree of invasiveness for sample collection and the investigation of changes in biomarker values in response to an intervention targeting a reduction in SSB consumption. Limitations include the use of self-reported AS and SSB intakes as the method for biomarker comparison and the inability of the $\delta^{13}\text{C}$ biomarker to detect some forms of AS such as beet sugar, honey and maple syrups. These represent minor contributors of AS to the US diet (i.e. 22 %) compared with corn- and cane-derived sweeteners (i.e. 78 %)⁽¹¹⁾; however, we did not assess these forms of AS in this sample.

The availability of an objective indicator of AS and SSB intake could overcome a commonly cited methodological limitation of research investigating the health effects of AS and SSB consumption^(5–7,10). Additional research is warranted to investigate the validity of the $\delta^{13}\text{C}$ sugar intake biomarker in children and adolescents, who consume high amounts of AS, and in controlled feeding trials.

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Table 1
 Baseline participant characteristics in the full sample, and in the SIP-smartER intervention and MoveMore matched-contact comparison groups, rural Southwest Virginia, USA

| Characteristic | Full sample (n 296) | | SIP-smartER (n 151) | | MoveMore (n 145) | | Test statistic, P value [†] |
|-----------------------------------------|---------------------|---------|---------------------|---------|------------------|---------|--------------------------------------|
| | Mean or n | SD or % | Mean or n | SD or % | Mean or n | SD or % | |
| Age (years), mean and SD | 42.1 | 13.4 | 41.7 | 13.4 | 42.4 | 13.3 | <i>t</i> = -0.41 <i>P</i> = 0.68 |
| Gender, <i>n</i> and % | | | | | | | |
| Male | 56 | 19 | 30 | 20 | 26 | 18 | $\chi^2 = 0.18$ |
| Female | 240 | 81 | 121 | 80 | 119 | 82 | <i>P</i> = 0.67 |
| Race, <i>n</i> and % | | | | | | | |
| Caucasian | 275 | 93 | 137 | 91 | 138 | 95 | $\chi^2 = 2.22$ |
| African American | 13 | 4 | 10 | 6 | 3 | 2 | <i>P</i> = 0.14 |
| More than one race | 7 | 2.5 | 3 | 2 | 4 | 3 | |
| Other | 1 | 0.5 | 1 | 1 | 0 | 0 | |
| Ethnicity, <i>n</i> and % | | | | | | | |
| Hispanic/Latina | 3 | 1 | 2 | 1 | 1 | 0.5 | $\chi^2 = 0.48$ <i>P</i> = 0.79 |
| Education level, <i>n</i> and % | | | | | | | |
| High-school graduate | 93 | 31 | 49 | 32.5 | 44 | 30 | $\chi^2 = 0.15$ |
| Some college or greater | 203 | 69 | 102 | 67.5 | 101 | 70 | <i>P</i> = 0.70 |
| Annual household income, <i>n</i> and % | | | | | | | |
| \$US 14 999 | 126 | 43 | 69 | 46 | 57 | 39 | $\chi^2 = 6.89$ |
| \$US 15 000–34 999 | 94 | 32 | 52 | 35 | 42 | 29 | <i>P</i> = 0.08 |
| \$US 35 000–54 999 | 39 | 13 | 18 | 12 | 21 | 15 | |
| \$US 55 000 | 37 | 12 | 12 | 8 | 25 | 17 | |
| Weight status | | | | | | | |
| Weight (kg), mean and SD | 90.6 | 25.4 | 90.7 | 26.4 | 90.4 | 24.3 | <i>t</i> = 0.09 <i>P</i> = 0.93 |
| BMI (kg/m ²), mean and SD | 33.0 | 9.1 | 33.3 | 9.3 | 32.7 | 9.0 | <i>t</i> = 0.49 <i>P</i> = 0.62 |

[†]The *t* test was used to compare group means; the χ^2 test was used to compare proportions across groups.

Table 2
 $\delta^{13}\text{C}$ sugar intake biomarker values and self-reported dietary intake (*n* 296) at baseline and after the 6-month intervention in the SIPsmartER intervention and MoveMore matched-contact comparison groups, rural Southwest Virginia, USA

| Variable | Group | Baseline [†] | | Month 6 [†] | | Adjusted change, baseline to month 6 [‡] | | P value, group by time [§] |
|------------------------------------|------------|-----------------------|------|----------------------|------|---------------------------------------------------|-------------|-------------------------------------|
| | | Mean | SD | Mean | SD | Mean | 95% CI | |
| $\delta^{13}\text{C}$ (‰) | SIPsmartER | -18.92 | 0.65 | -18.97 | 0.65 | -0.05 | -0.10, 0.01 | <0.001 |
| | MoveMore | -18.94 | 0.72 | -18.92 | 0.73 | 0.02 | -0.04, 0.08 | |
| SSB (kcal/d) | SIPsmartER | 496 | 374 | 268 | 297 | -227 ^{***} | -326, -127 | <0.001 |
| | MoveMore | 377 | 287 | 325 | 319 | -53 ^{**} | -88, -17 | |
| SSB (fluid ounces/d) | SIPsmartER | 43 | 31 | 24 | 24 | -19 ^{***} | -28, -10 | <0.01 |
| | MoveMore | 33 | 24 | 28 | 27 | -5 ^{***} | -7, -2 | |
| AS (% of total energy) | SIPsmartER | 22 | 12 | 17 | 12 | -5 ^{***} | -7, -3 | <0.001 |
| | MoveMore | 21 | 11 | 20 | 12 | -1 | -2, 0 | |
| AS (g/d) | SIPsmartER | 108 | 92 | 74 | 88 | -34 ^{***} | -46, -22 | <0.001 |
| | MoveMore | 95 | 66 | 89 | 70 | -6 | -14, 2 | |
| TS (% of total energy) | SIPsmartER | 26 | 12 | 22 | 13 | -5 ^{***} | -7, -2 | <0.01 |
| | MoveMore | 26 | 11 | 25 | 11 | -1 | -3, 1 | |
| TS (g/d) | SIPsmartER | 130 | 98 | 94 | 97 | -36 ^{***} | -49, -23 | <0.001 |
| | MoveMore | 117 | 67 | 110 | 75 | -8 | -17, 1 | |
| Energy (kcal/d) | SIPsmartER | 1975 | 1100 | 1690 | 1099 | -285 ^{***} | -434, -136 | <0.05 |
| | MoveMore | 1766 | 640 | 1723 | 682 | -44 | -136, 49 | |
| Protein (% of energy) | SIPsmartER | 14.6 | 4.2 | 16.4 | 5.0 | 1.8 ^{***} | 0.9, 2.8 | NS |
| | MoveMore | 15.2 | 4.3 | 15.9 | 4.4 | 0.7 | -0.02, 1.4 | |
| Fat (% of energy) | SIPsmartER | 33.6 | 7.9 | 35.2 | 9.4 | 1.6 [*] | 0.3, 2.9 | NS |
| | MoveMore | 33.6 | 6.7 | 34.6 | 7.9 | 0.9 | -0.1, 1.9 | |
| Carbohydrate (% of energy) | SIPsmartER | 51.0 | 10.4 | 47.6 | 11.3 | -3 ^{***} | -5, -0.04 | NS |
| | MoveMore | 50.4 | 9.0 | 48.9 | 9.9 | -1.5 ^{***} | -2.5, -0.4 | |

SSB, sugar-sweetened beverages; AS, added sugar; TS, total sugar.

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[†] Means and standard deviations are not adjusted for covariates.

[‡] Within-group statistical significance indicated by asterisks:

* $P < 0.05$,

** $P < 0.01$,

*** $P < 0.001$.

[§] Models control for baseline covariates including age, gender, race/ethnicity, income, education level, health literacy level, employment status, number of children, smoking status and BMI.

// 1 kcal = 4.184 kJ.

¶ 1 fluid ounce = 29.57 ml.