The Effect of *Wolffia globosa* Mankai, a Green Aquatic Plant, on Postprandial Glycemic Response: A Randomized Crossover Controlled Trial

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OBJECTIVE

To compare the postprandial and overnight glycemic response using a novel green aquatic plant thought to provide a dietary source for high-quality protein, with an iso-carbohydrate/protein/caloric dairy shake.

RESEARCH DESIGN AND METHODS

This is a randomized controlled crossover trial among 20 abdominally obese participants (age 51.4 years; fasting plasma glucose 110.9 mg/dL), who were allocated to replace dinner with either, first, a green shake containing *Wolffia globosa* duckweed (Mankai: specific-strain) or an iso-carbohydrate/protein/calorie yogurt shake. A 2-week flash glucose-monitoring system was used to assess postmeal glucose dynamics (6 net administration days; 97 observation days in total). We further obtained from each participant dietary/daily activity/satiety scale/sleep logs. Participants were recruited from the green-Mediterranean diet arm of the 18-month Dietary Intervention Randomized Controlled Trial-Polyphenols Unprocessed (DIRECT-PLUS) study.

RESULTS

***Wolffia globosa* Mankai elicited a lower postprandial glucose peak compared with yogurt (△peak = 13.4 ± 9.2 vs. 19.3 ± 15.1 mg/dL; $P = 0.044$), which occurred later (77.5 ± 29.2 vs. 59.2 ± 28.4 min; $P = 0.037$) and returned faster to baseline glucose levels (135.8 ± 53.1 vs. 197.5 ± 70.2 min; $P = 0.012$). The mean post–net incremental area under the curve (netAUC) was lower with *Wolffia globosa* up to 60 and 180 min (netAUC 60 min: 185.1 ± 340.1 vs. 441.4 ± 336.5 mg/dL/min, $P = 0.005$; netAUC 180 min: 707.9 ± 1,428.5 vs. 1,576.6 ± 1,810.1 mg/dL/min, $P = 0.037$). A *Wolffia globosa*–based shake replacing dinner resulted in lower next-morning fasting glucose levels (83.2 ± 0.8 vs. 86.6 ± 13 mg/dL; $P = 0.041$). Overall, postprandial glucose levels from the shake administration until the next morning were lower in the *Wolffia globosa* Mankai green shake compared with the yogurt shake ($P < 0.001$). Overnight sleep duration was similar (378.2 ± 22.4 vs. 375.9 ± 28.4 min; $P = 0.72$), and satiety rank was slightly higher for the *Wolffia globosa* shake compared with the yogurt shake (7.5 vs. 6.5; $P = 0.035$).**

CONCLUSIONS

***Wolffia globosa* Mankai duckweed may serve as an emerging alternative plant protein source with potential beneficial postprandial glycemic effects.**

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Health-related concerns (1–4), sustainability (5), and ideological reasons are common motivations to search for plant-based alternatives for animal protein sources. The Academy of Nutrition and Dietetics recommends a vegetarian, plant-based diet for primary prevention of obesity, cardiovascular disease, and type 2 diabetes (6). To date, the short-term effects of vegetarian, plant-based sources rich in polyphenols, on postprandial glycemia have not been fully characterized. Studies that have measured the acute glycemic response of plants rich in polyphenols suggest a beneficial effect in improving glycemic profile, with a lower glucose peak (7,8).

Wolffia globosa duckweed (Mankai strain), an emerging edible aquatic plant, is rich in protein content (>45% of the dry matter) and includes all nine essential and six conditional amino acids. Recently, we reported that, among humans, the bioavailability of the essential amino acids in *Wolffia globosa* was similar to the well-established animal (soft cheese) and plant (peas) iso-protein sources (9), highlighting this plant as a high-quality protein source. Moreover, in rat models, iron derived from *Wolffia globosa* was found to be bioavailable and efficient in treating iron deficiency anemia (10). In the current study, the *Wolffia globosa* Mankai green shake was designed to provide 12% of the daily recommended intake of protein, 4.3% of carbohydrates, and 1.8% of fat for men, as well as a relatively high dietary fiber content, accounting for 13.5% of the daily recommended intake. In addition, this aquatic plant is highly rich in polyphenols, mainly phenolic acids and flavonoids (including catechins). The aim of this crossover trial was to assess the postprandial and overnight glycemic response after *Wolffia globosa* Mankai administration compared with an iso-carbohydrate/protein/caloric dairy protein shake (yogurt) among abdominally obese adults.

RESEARCH DESIGN AND METHODS

Study Design

This crossover trial is a substudy of the Dietary Intervention Randomized Controlled Trial-Polyphenols Unprocessed (DIRECT-PLUS) (clinical trial reg. no. NCT03020186, ClinicalTrials.gov) study that began in May 2017. The DIRECT-PLUS study included 294 participants, who were enrolled from an isolated workplace (Nuclear Research Center Negev, Dimona, Israel). Inclusion criteria for the DIRECT-PLUS study were age >30 years with abdominal obesity (waist circumference: men >102 cm; women >88 cm) and/or dyslipidemia (triglycerides >150 mg/dL; HDL cholesterol: men ≤40 mg/dL; women ≤50 mg/dL). Exclusion criteria for the DIRECT-PLUS study were the inability to take part in physical activity (PA) in the gym, serum creatinine ≥2 mg/dL, disturbed liver function tests, major illness that might require hospitalization, pregnancy, the presence of active cancer, receiving or having received chemotherapy in the previous 3 years, participation in another trial, treatment with coumadin (warfarin), or possessing a pacemaker or metal implant. Among randomized participants assigned to one of the intervention groups (the PA+green-Mediterranean diet group), we selected individuals capable of using a glucose-monitoring system. Specific exclusion criteria for the crossover trial were lactose intolerance or milk allergy and the use of antihyperglycemic medications and/or insulin. Participants were defined as having type 2 diabetes if their fasting plasma glucose (FPG) concentration was ≥126 mg/dL or HbA_{1c} ≥6.5% (48 mmol/mol). Prediabetes was defined as FPG between 100 and 125 mg/dL or HbA_{1c} levels between 5.7% and 6.4% (39–47 mmol/mol) (11).

Intervention

This 2-week crossover substudy was performed in the initial phase of the DIRECT-PLUS study (Supplementary Fig. 1) in one phase. A flash glucose-monitoring system device (Freestyle Libre; Abbott Diabetes Care, Witney, Oxon, U.K.) was used (Supplementary Data). The two shakes, *Wolffia globosa* duckweed (Mankai) and yogurt, were consumed by each participant at 7:00 P.M., instead of dinner. The shake-type order was randomized (simple randomization and random allocation), and the shakes were given at the same time at the specific hour in the evening (Supplementary Fig. 1). The participants were requested to refrain from food and drink intake 1 h before shake consumption and from shake consumption until the next morning. The participants were instructed to avoid strenuous PA 5 h prior to the shake consumption, without further instructions for a specific sequence or timing of exercise. The 2 weeks of glucose monitoring included a total of 6 administration days, 3 days for each shake. Shakes were not administered during weekends, holidays, and on the first day after inserting the sensor, although the sensors were used continuously for 14 days. Of the 23 initial participants, 3 dropped out for technical reasons. We excluded 23 observation days in which preadministration glucose levels were measured at >100 mg/dL. This was done in order to standardize the initial levels before the test.

The *Wolffia globosa* Mankai green shake included three frozen cubes of the plant (25 g each), while the yogurt shake included 100 g low-fat yogurt with no added sweetener. Both shakes included 28 g walnuts and one medium-sized banana. The two shakes were isocaloric (366 kcal in the *Wolffia globosa* Mankai shake; 351 kcal in the yogurt shake) and equivalent in terms of macronutrient content (carbohydrates: 35 g in both the *Wolffia globosa* and yogurt shakes; protein: 12 g in the *Wolffia globosa* shake; 11 g in the yogurt shake; fat: 20 g in the *Wolffia globosa* shake; 19 g in the yogurt shake). Regardless of the equal degree of carbohydrate content, the *Wolffia globosa* plant shake naturally included 9 g dietary fibers, compared with the natural yogurt shake with 5 g dietary fibers. We did not add any further supplements or components to the shakes.

Adherence to the intervention protocol was assessed using a detailed dietary (including time and portion size), daily activity, satiety scale (rank 1–10) and sleep recall during these 2 weeks. Text messages were sent on a daily basis as a reminder for maintaining study protocol instructions. The Soroka University Medical Center Medical Ethics Board and Helsinki Committee approved the trial protocol. All participants provided written informed consent and received no financial compensation or gifts. The Mankai and walnuts were provided free of charge.

Statistical Analysis

This is a crossover trial as part of the 18-month DIRECT-PLUS trial. Although the primary aim of the long-term DIRECT-PLUS trial was to assess changes in abdominal and hepatic fat, our specific

aim here was to specifically assess the acute glycemic effects of a *Wolffia globosa*–based versus yogurt-based dinner shake, measured by a continuous glucose monitoring system. The power calculation for the postprandial glycemic effect is based on a randomized crossover trial that compared the effect of berries as a polyphenol source in comparison with a similar sucrose load among 12 healthy subjects (8). The trial demonstrated a significant difference in the glycemic profiles (division of time [min] during which the plasma glucose was above the fasting concentration with the incremental peak value [mmol/L]) between berries and the control meals (49.9 ± 21.1 vs. 24.7 ± 9.3 min/(mmol/L), P = 0.003). Therefore, the calculated power for our study was 90.9%. Continuous variables are presented as the mean ± SD, unless specified otherwise. Nominal variables are expressed as numbers and percentages. Variables were tested for normal distribution using a Kolmogorov-Smirnov test. Baseline characteristics are presented for each group and for the entire study population. Differences in baseline characteristics were tested by Mann-Whitney test. We assessed the differences within subjects by measuring the mean net incremental area under the curve (netAUC) (12,13) of glucose at 60 and 180 min after the administration of the shake, and overnight glucose profile. In addition, we calculated the following personal glycemic parameters: glucose time to peak, postprandial glucose peaks, time until glucose levels returned to baseline, next-morning fasting glucose, sleep duration, and satiety rank using the Wilcoxon test. Observation day was recorded based on the time from shake administration until the next morning. Additionally, we performed a mixed-model analysis over time (3 days, 15-min intervals on average) as a within-subject test and the shake meal group as a between-subject factor, with the primary outcome being the glucose level. We used exploratory analysis to determine the differences between time points that were expressed as absolute values unless specified otherwise. Glucose trajectory similarities were assessed using dynamic time warping (DTW); all 97 postprandial 180-min observations were used to create a distance matrix, and interindividual and intraindividual distances were subsequently extracted and compared by Mann-Whitney test. Statistical significance was set at $P < 0.05$ (two sided). Statistical analysis was performed using SPSS (version 22.0) software. Power calculations were performed using WinPepi software, version 11.6. Graphs were constructed using GraphPad Prism 7.

RESULTS

Baseline Characteristics

Baseline characteristics of all participants and baseline characteristics of participants by sequence of shake administration are shown in Table 1. We randomized 23 participants in a crossover design to first consume either a *Wolffia globosa* (Mankai) shake (n = 12) or a yogurt shake (n = 11). After installation of the flash glucose-monitoring system device, and prior to shake administration, the sensor was detached in three participants; thus, there were no data of glycemic response to the shakes for these three participants. The initial randomization table is shown in Supplementary Table 3. The 20 participants were randomized to Mankai shake first (n = 10) or yogurt shake first (n = 10) groups and collected a total of 97 observation days. Of the 20 participants who completed the intervention, 13 participants had prediabetes and 1 participant had type 2 diabetes. Eighteen participants were men, the mean age was 51.4 ± 11.2 years, and the mean weight was 91.1 ± 15.3 kg. The mean FPG level was 110.9 ± 16.2 mg/dL, with a mean HbA_{1c} of 5.5 ± 0.7% (36.8 ± 7.3 mmol/mol). Baseline parameters were similarly distributed between the groups.

Glycemic Response

The glucose trajectory overnight after *Wolffia globosa* (Mankai) shake and yogurt shake intakes are presented in Fig. 1, and the glycemic indices are presented in Supplementary Table 4. The consumption of the *Wolffia globosa* shake elicited lower postprandial glucose peaks (Δ = 13.4 ± 9.2 mg/dL) compared with the yogurt shake (Δ = 19.3 ± 15.1 mg/dL; P = 0.044). *Wolffia globosa* shake consumption induced a longer time to

Table 1—Baseline characteristics of the study population across groups in the continuous glucose crossover trial (n = 20)

	Group 1 order (n = 10; 45 observation days)*	Group 2 order (n = 10; 52 observation days)†	Entire (n = 20)	P between groups
Age, year	51.1 ± 9.3	51.7 ± 13.3	51.4 ± 11.2	0.97
Weight, kg	91.3 ± 14.2	90.8 ± 17.1	91.1 ± 15.3	0.58
WC, cm	109.2 ± 9.3	107.5 ± 10.4	108.4 ± 9.6	0.97
FPG, mg/dL	113.7 ± 20.6	107.9 ± 9.8	110.9 ± 16.2	0.66
HbA_{1c}				
%	5.8 ± 0.8	5.3 ± 0.4	5.5 ± 0.7	0.08
mmol/mol	39.7 ± 8.7	33.9 ± 4.3	36.8 ± 7.3	0.08
Insulin, μIU/mL	16.5 ± 9.5	14.5 ± 8.7	15.5 ± 8.9	0.63
Systolic blood pressure, mmHg	129.3 ± 13.2	137.0 ± 17.4	133.1 ± 15.6	0.22
Diastolic blood pressure, mmHg	82.6 ± 13.9	79.6 ± 12.2	81.1 ± 12.8	0.91
Triglycerides, mg/dL	131.6 ± 37.8	124.2 ± 66.7	127.9 ± 52.9	0.44
HDL, mg/dL	43.7 ± 8.6	49.5 ± 16.7	46.6 ± 13.2	0.53
ALT, units/L	35.0 ± 15.4	30.4 ± 17.6	32.7 ± 16.3	0.44
AST, units/L	27.1 ± 8.4	23.7 ± 8.3	25.4 ± 8.3	0.63

Values are presented as the mean ± SD for continuous variables. P value according to a Mann-Whitney test for continuous variables. ALT, alanine aminotransferase; AST, aspartate transaminase; IU, international units; WC, waist circumference. *Group 1 started with a yogurt shake. †Group 2 started with a Mankai shake.

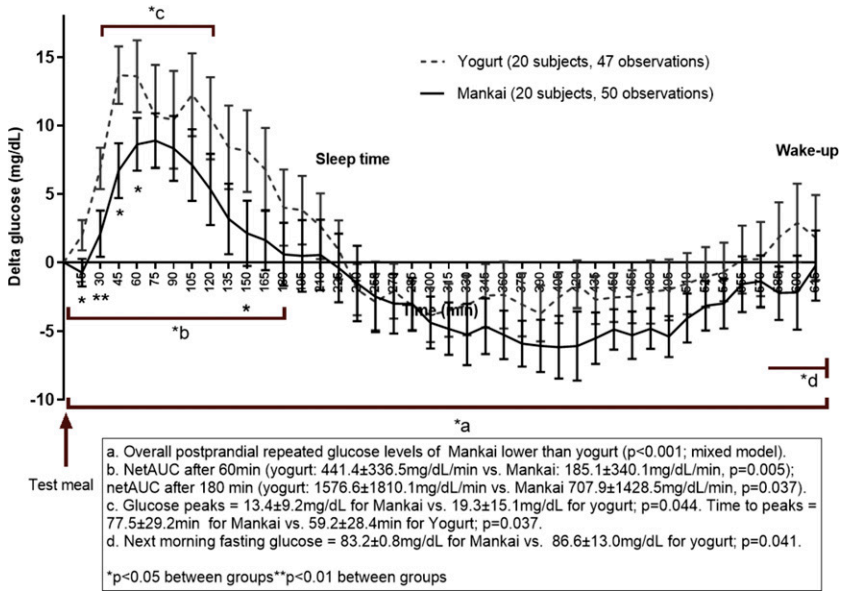

Figure 1—Glucose trajectory overnight after *Wolffia globosa* (Mankai) and yogurt intake. *$P <$ 0.05 differences between groups, **$P < 0.01$ differences between groups. a shows that the overall, postprandial repeated glucose levels from the administration of the shake until the next morning were significantly lower in the Mankai shake group compared with the yogurt shake group ($P <$ 0.001). Differences were analyzed by mixed model over time as a within-subject test, and the shake meal group as a between-subject factor. b shows a significant difference in the mean netAUC (0–60 and 0–180 min). The netAUC after 60 min (yogurt 441.4 ± 336.5 mg/dL/min; Mankai 185.1 ± 340.1 mg/dL/min; P = 0.005); the netAUC after 180 min (yogurt 1,576.6 ± 1,810.1 mg/dL/min; Mankai 707.9 ± 1,428.5 mg/dL/min; P = 0.037). Differences within subjects were analyzed by Wilcoxon test. c shows a significant difference in glucose peaks (13.4 ± 9.2 vs. 19.3 ± 15.1 mg/dL; P = 0.044) and a significant difference in time to peaks (77.5 ± 29.2 vs. 59.2 ± 28.4 min; P = 0.037). Differences within subjects were analyzed by Wilcoxon test. d shows a significant difference in next-morning fasting glucose concentration (83.2 ± 0.8 vs. 86.6 ± 13.0 mg/dL; P = 0.041). The difference within subjects was analyzed by Wilcoxon test.

reach peak glucose levels compared with the yogurt shake (77.5 ± 29.2 vs. 59.2 ± 28.4 min, respectively; P = 0.037) and a lower mean netAUC after 60 min (*Wolffia globosa* 185.1 ± 340.1 mg/dL/min; yogurt 441.4 ± 336.5 mg/dL/min; P = 0.005) and after 180 min (*Wolffia globosa* 707.9 ± 1,428.5 mg/dL/min; yogurt 1,576.6 ± 1,810.1 mg/dL/min; P = 0.037). The overnight mean netAUC was not significantly different after the consumption of a *Wolffia globosa* shake versus a yogurt shake (743.9 ± 5,141.8 vs. 2,733.1 ± 5,589.3 mg/dL/min, respectively; P = 0.069). *Wolffia globosa* consumption was associated with lower next-morning fasting glucose concentration (83.2 ± 0.8 vs. 86.6 ± 13 mg/dL; P = 0.041). The *Wolffia globosa* Mankai shake resulted in significantly lower postprandial glucose levels when measured 15, 30, 45, 60, and 150 min following the ingestion, compared with the yogurt shake ($P <$ 0.05 for all). Glucose levels returned to baseline values faster after the consumption of the *Wolffia globosa* shake (mean 135.8 ± 53.1 min) than after consumption of the yogurt shake (mean 197.5 ± 70.2 min; P = 0.012). Participants with dysglycemic FPG levels at baseline (FPG ≥100; n = 14) also exhibited a significantly lower next-morning fasting glucose concentration in the *Wolffia globosa* compared with the yogurt shake (84.0 ± 12.2 vs. 88.1 ± 14.5 mg/dL, respectively; P = 0.028). Overall, postprandial glucose levels, from the time of administration of the shake replacing dinner until the following morning, were significantly lower after the *Wolffia globosa* Mankai shake than the yogurt shake ($P <$ 0.001). The order of intervention group allocation did not affect the results ($P >$ 0.05). A secondary sensitivity analysis that included all ob`-servations (i.e., without exclusion of individual measurements, as described above) revealed similarly significant differences between groups, as observed in the original analysis. This included the following parameters: postprandial glucose peak; mean netAUC after 60 and 180 min; postprandial glucose levels after 15, 30, 45, and 150 min; next-morning fasting glucose level; and postprandial repeated glucose levels from the time of administration until the following morning. However, differences in the time to peak glucose level and in the postprandial glucose level after 60 min lost their statistical significance. Nevertheless, in this inclusive analysis of all measurements, the overnight mean netAUC and other overnight time points became significantly lower in the Mankai intervention arm compared with the yogurt intervention arm (Supplementary Fig. 5 and Supplementary Table 5).

Logs

According to participant's records, the median post-shake satiety rank for *Wolffia globosa* Mankai was 7.5, while for yogurt it was 6.5 (P = 0.035). There was no significant difference in the sleep duration after *Wolffia globosa* Mankai consumption compared with a yogurt-based meal (378.2 ± 22.4 vs. 375.9 ± 28.4 min; P = 0.72). All participants reported refraining from strenuous PA for at least 5 h prior to shake consumption.

Individual Patterns

The individual continuous blood glucose levels are presented in Fig. 2. We compared intraindividual (within persons–between treatments) Mankai-yogurt 180-min glucose excursions with interindividual (between persons–within treatments) Mankai-Mankai and yogurt-yogurt 180-min glucose excursions using DTW distances and accounting for all recorded observations. The intraindividual responses were significantly closer in DTW measure than the interindividual responses (intraindividual responses 43.3 ± 2.4; interindividual responses 48.6 ± 0.5; P = 0.004), suggesting that the glucose trajectory response of each participant was personally characterized with similar patterns for either intervention.

CONCLUSIONS

In this crossover randomized controlled trial comparing glucose excursions in

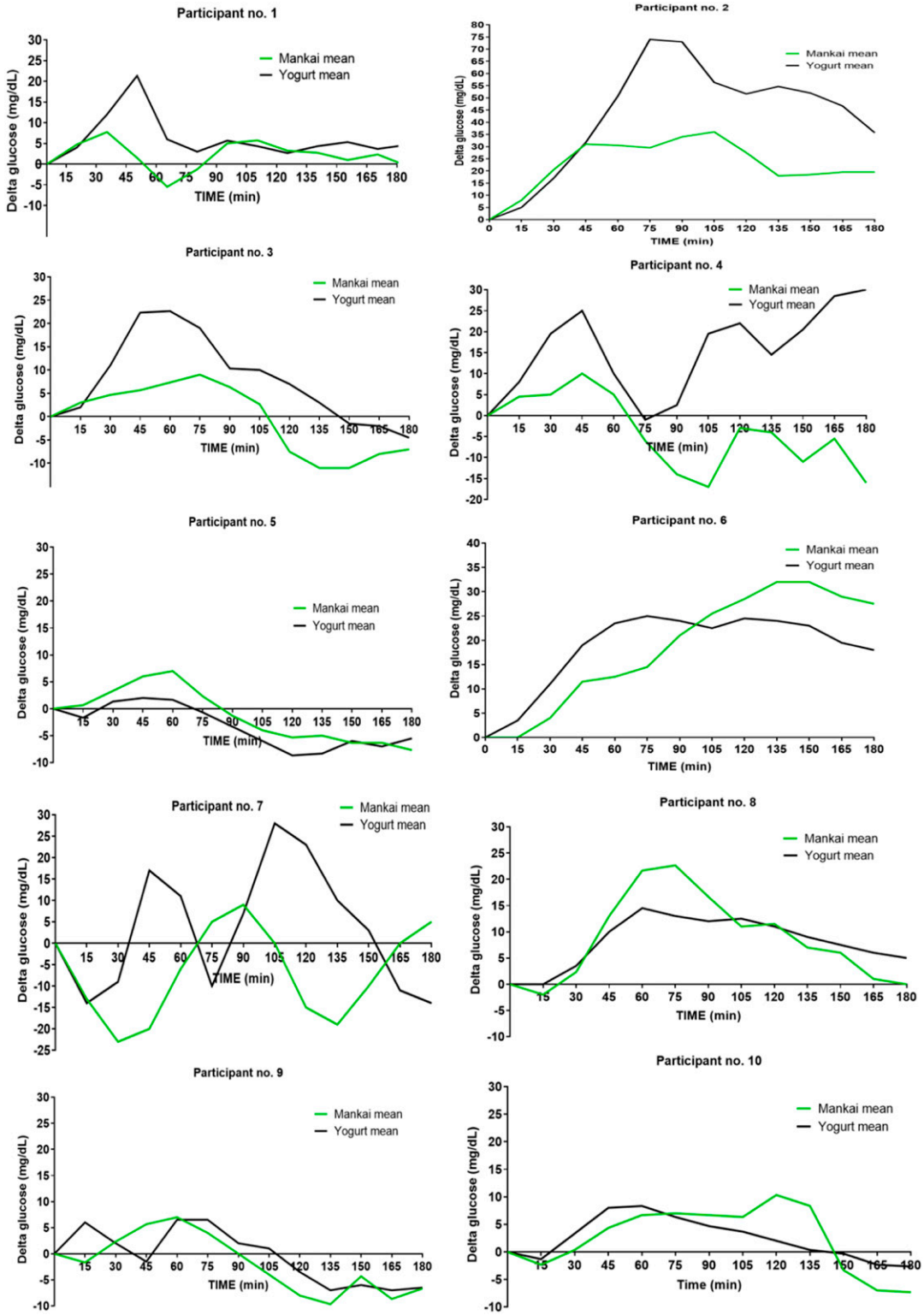

Figure 2—Individual glucose responses to both shake meals. The mean glucose distribution of each individual response among 20 participants in the crossover trial. DTW distance of intraindividual response (43.3 ± 2.4) vs. DTW of interindividual responses (48.6 ± 0.5, $P = 0.004$). All 97 postprandial 180-min observations were used to create a distance matrix; interindividual and intraindividual distances were subsequently extracted and compared by Mann-Whitney test.

response to iso-carbohydrate/caloric meals of *Wolffia globosa* (Mankai) versus yogurt shakes, the glucose response to the *Wolffia globosa*–containing shake was apparently slightly favorable, with the effect being discernable from the time of the meal (i.e., at dinner) until the following morning. Furthermore, glucose peaks were lower, times to glucose peaks were delayed, and the netAUCs of the first hour and after 3 hours were smaller. These results suggest that *Wolffia*

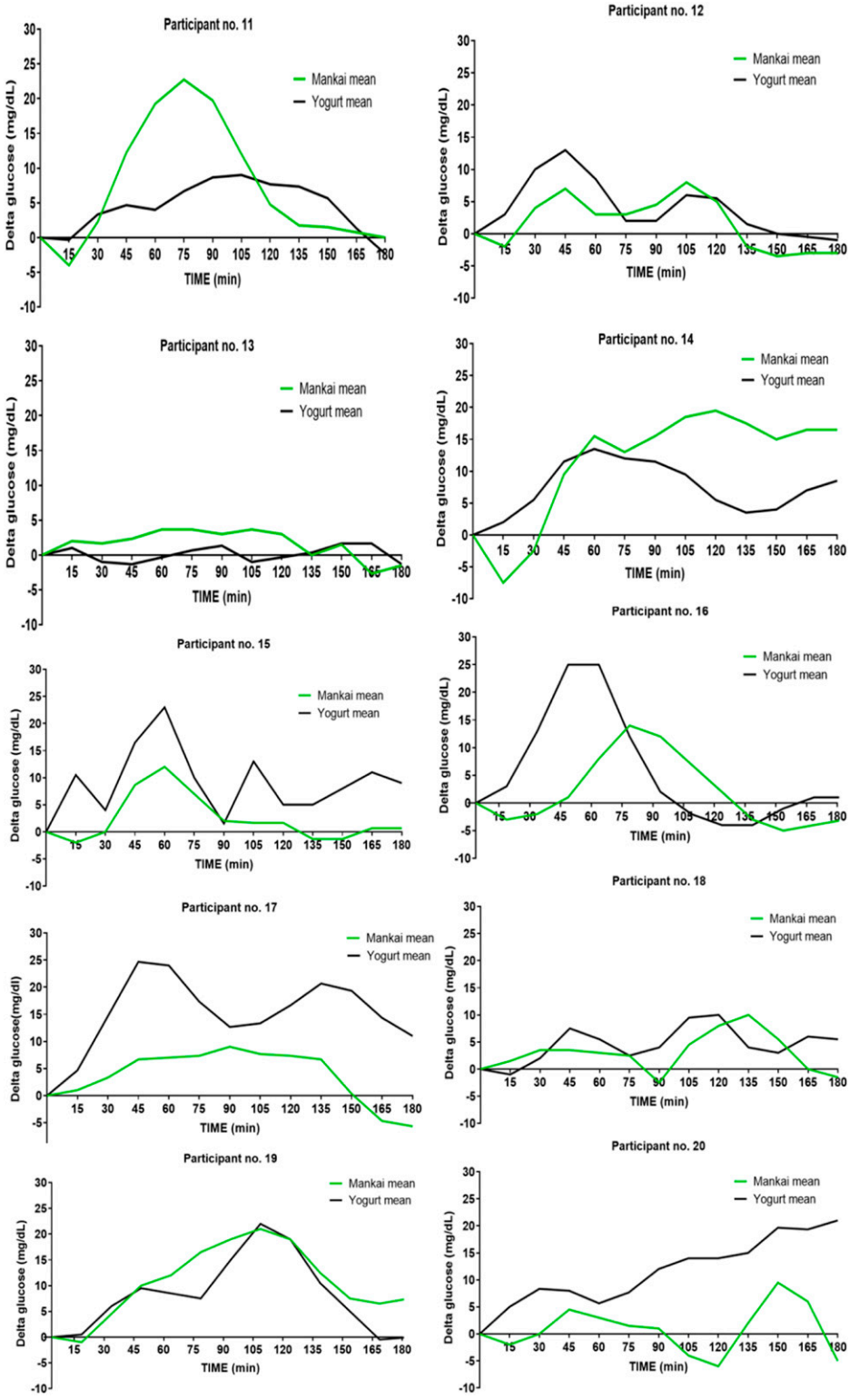

Figure 2—Continued.

globosa, which is naturally rich in dietary fibers and polyphenols, may constitute an alternative iso-protein/carbohydrate shake with a beneficial glycemic response.

Several limitations should be considered. This specific substudy included individuals with abdominal obesity but mostly with normal FPG levels or prediabetes. Thus, our findings cannot be generalized to patients with diabetes. Furthermore, the participants were recruited from the PA+green-Mediterranean dietary intervention arm of the DIRECT-PLUS study, with an energy

restriction and elevated PA, and do not represent the general population. Although we monitored exercise performance 5 h prior to shake administration, data regarding the timing of the exercise with respect to meals was only available regarding this time frame. Nevertheless, as this is a crossover trial, and comparisons are between interventions within the same subjects, it is unlikely that participants altered their daily routine pattern of exercise when switching between the two types of shakes. Moreover, as a consequence of the sex profile of workers in the workplace setting studied, the majority were men. Finally, we randomized only 20 participants. Nevertheless, this relatively small sample provided sufficient statistical power to detect a significant difference between the Mankai and yogurt groups over nearly 100 repeated days of observations in a crossover design. Study strengths included its one-phase randomized controlled design, in which all participants started intervention on the same day, a strict and detailed lifestyle log (dietary, including time frames and portion size, daily activity, and sleep and arising recall), and that the two intervention arms were identical in the energetic and macronutrient content.

The glucose excursion following the consumption of *Wolffia globosa* was apparently favorable compared with that of yogurt. This effect could be explained by the unique nutritional properties of *Wolffia globosa,* such as the high fiber content and polyphenol levels that are absent in yogurt. These components may minimize postprandial glucose peaks (7,8,14,15) as well as lower the glycemic index beyond the impact associated with carbohydrate quantity. Previous studies (16–18) demonstrated a beneficial effect of dietary fibers on insulin resistance among patients with type 2 diabetes and subjects with impaired glucose tolerance, and showed that those dietary fibers can lower the risk for type 2 diabetes and cardiovascular disease. Moreover, in our study, we found that the *Wolffia globosa* shake was more satiating compared with the yogurt shake, an effect potentially attributable to the difference in fiber content. Furthermore, polyphenols, which are abundant in the *Wolffia globosa*, can exert anti-inflammatory effects and may influence glucose metabolism through different mechanisms, including by inhibiting glucose absorption in the gut, decreasing fasting insulin levels (19–21), and reducing insulin resistance (22,23). Some hypoglycemic effects were observed with polyphenols ingested shortly before glucose consumption (23,24) and have been associated with a reduced incidence of type 2 diabetes (25–27).

Our previous study (9) aimed to evaluate the bioavailability of essential amino acids in Mankai compared with a dairy iso-protein (soft cheese). We found that the increase in plasma branched-chain amino acids (leucine/isoleucine and valine) concentrations occurred relatively faster in the cheese group compared with the Mankai group (9). Possibly, these results may relate to recent evidence suggesting that plasma branched-chain amino acids, which are common in dairy products, are associated with insulin resistance and type 2 diabetes (28). We further analyzed the postprandial glucose and insulin blood levels in the bioavailability test (9) (Supplementary Fig. 6) and found that although the Mankai dish included, in the overall recipe, much higher content of carbohydrates than the iso-protein cheese meal, the insulin levels induced after Mankai consumption were significantly lower than those induced after cheese consumption, whereas glucose levels similarly decreased in all time points.

The initial postprandial glucose response (0–30 min) during a glucose tolerance test might mainly reflect the suppression of hepatic glucose production (29). The higher initial rise in glucose levels reflects greater hepatic insulin resistance or deficient pancreatic β-cell function (29). After ~120–180 min, hepatic glucose production is maximally suppressed (30). Thus, the glucose levels declining to the nadir indicate glucose uptake by peripheral tissues, specifically, muscle. We observed a lower glucose peak, a smaller netAUC after 60 and 180 min, and a faster decline from the glucose peak level back to baseline after green shake consumption compared with yogurt shake consumption. Our findings suggest that the beneficial glucose excursion in response to the *Wolffia globosa* Mankai green shake may represent a combined effect of a lower glycemic index and a better insulin response.

Although the Mankai group demonstrated a beneficial glycemic response compared with the yogurt group, we observed different patterns of glucose response between subjects. Indeed, we found that the individual pattern is more pronounced than the intervention itself. A previous study (31) showed a wide variety between glucose responses in subjects who consumed the exact same meal. Since our trial was a crossover study, we could compare the shakes in our study group beyond the interpersonal differences.

In conclusion, our study suggests that *Wolffia globosa* (Mankai) may serve as a new alternative protein source with potential beneficial postprandial glycemic effects.

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Duality of Interest. I.Sha. advises the nutritional committee of Hinoman Ltd. No other potential conflicts of interest relevant to this article were reported.

Author Contributions. H.Z., A.K., A.Y.M., G.T., E.R., I.She., A.T., D.B., E.P., L.Q., J.T., M.S., N.K., M.v.B., U.C., M.B., M.J.S., and I.Sha. had full access to all of the data in the study, take responsibility for the integrity of the data and accuracy of the data analysis, and read and approved the final manuscript. H.Z., A.K., J.T., M.S., N.K., M.v.B., U.C., and M.B. analyzed the data. H.Z., A.K., and I.Sha. designed the research, conducted the study, wrote the manuscript, and are responsible for the final content. A.Y.M., G.T., E.R., D.B., and E.P. conducted the study. I.She., A.T., and L.Q. reviewed and edited the manuscript. M.J.S. designed the research, and reviewed and edited the manuscript. H.Z. and A.K. are the guarantors of this work and, as such, had full access to all the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

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