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Parboiled Brown Rice Product Reduces Postprandial Plasma Glucose Response in Men

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Abstract

A staple crop such as rice provides an ideal starch source for creating a functional starch ingredient. Functional starch fractions can act as a functional ingredient by controlling glucose and insulin levels with application for glucose control for health in addition diabetes mellitus. The objective of this human study was to investigate the effect of a parboiled brown rice flour pudding on postprandial plasma glucose and insulin levels. *Wells* brown rice was parboiled at 120°C for 20 min and ground into flour, and *in vitro* nutritional starch fractions were measured. A randomized-crossover design was used to observe plasma glucose and insulin responses from 14 healthy, male subjects. Compared with the control, significant reductions after consumption of parboiled brown rice pudding in mean glucose levels at 15, 30, 45, 60, 75, and 90 minutes were observed ($P < 0.05$). Mean glucose incremental Area Under the Curve (iAUC) were also significantly lower (3795 ± 602 mg/dL) than the control solution (5880 ± 658 mg/dL) ($P<0.05$). Plasma insulin mean incremental response reduced also from 3066 ± 525 μU/L iAUC to 2219 ± 715 μU/L iAUC of the control and rice pudding treatments, respectively. Results suggest optimal parboiling of brown rice provided in a flour application could assist in managing plasma glucose levels for individuals, and with additional research functional starch fractions may help in the prevention of diabetes and obesity.

Keywords: Postprandial plasma glucose; Plasma insulin; Slowly-digestible starch; Resistant starch; Diabetes

Introduction

Glucose and insulin control is crucial to the health of both healthy and diabetic individuals. Diabetes has increased in the U.S. within the last two decades, and 25.8 million people or 8% of the population suffers from diabetes mellitus, a trend which is expected to continue to rise along with obesity [1]. Diet modifications are one of the most effective ways to maintain a healthy weight and also prevent or control diabetes. Starches and sugars are responsible for the sharp increases in blood glucose levels; however, certain carbohydrates can also allow for a slow release of glucose or maintain homeostasis levels. Differences in starch structure or source influence the starch hydrolysis, and consequently, those structural variations allow for a controlled release of glucose [2]. Results from a study conducted with maize-based starches and fibers displayed a strong relationship of reduced glycemic and insulinemic responses from both male and female subjects, while other studies have focused strictly on enzymatic digestion [2-4].

Starch digestion has prompted many nutritional applications for a variety of starch types based on structure and digestion composition. Starch digestion is represented by three enzymatic digestion rates and is identified by three fractions: rapidly-digestible starch (RDS), slowly-digestible starch (SDS), and resistant starch (RS) [5]. Additionally, structural differences such as amylose and amylopectin content correlates with digestion rate and starch fractions [6-9]. Increasing SDS and RS contents, or functional starch fractions, in starches has been studied in addition improved starch fraction analysis techniques [6,10-11]. Starch fractions have been investigated thoroughly with human and animal studies, particularly focusing on SDS and RS fractions [12-14]. Based on the properties and presence in most diets, [15,16] functional starch fractions can act as functional ingredients. Novel starches containing increased SDS and RS content from various starch sources have been linked to reduced glucose and insulin responses in both healthy human subjects and diabetic subjects [17-19]. Overall, the RS starch fraction presents a two-fold nutritional benefit: escaping digestion in the small intestine and providing substrate for fermentation in the lower intestine [20-22]. There are five distinctive types of RS which also have a biological impact based on starch process [23,24].

One of the most widely consumed starch sources which are consumed as a cooked whole grain or in a variety of products is rice. Previous research shows nutritionally important starch fractions such as RDS, SDS, and RS in rice can be modified based on type of cultivar, environmental conditions, or processing steps [25]. Parboiling is a major processing method which has effectively increased functional starch fractions in rice and storage is also another important factor for increasing retrograded amylose content and decreasing digestibility [7]. Increasing functional starch fractions by such methods in rice can reduce the starch enzymatic digestibility and thereby providing a controlled glucose delivery, and improving nutritional benefits [26]. Brown rice is a model starch source based on intrinsic starch structure properties and has shown to provide multiple health benefits as blood glucose lowering effects [27,28]. Previously, cooked whole white and brown rice and its effect on glucose levels has been researched, however, no studies have reported the anti-diabetic effect and glucose control of parboiled brown rice. This study aims to investigate the effects on plasma glucose and insulin responses of healthy men after the consumption of parboiled brown rice flour pudding.

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Materials and Methods

Treatment materials

Wells cultivar brown rice grown in Arkansas was provided by the University of Arkansas Rice Processing Program. Autoclaving and milling preparation of the brown rice was carried out by the University of Arkansas Department of Food Science Carbohydrate lab as well as initial material analysis [7]. Materials were selected based on recent research investigating both optimal parboiling conditions and cultivars for increasing functional starch fractions [7]. Comparing starch digestibility across hybrid and pureline cultivars exposed to three parboiling conditions identified optimal parboiling conditions for cultivars tested. Results indicated the *Wells* cultivar parboiled at 120°C for 20 min and stored at room temperature for 24 h (cycle 2 treatment) exhibited optimal starch digestibility of *in vitro* analysis [7]. Lemon extract and sucralose used in pudding formula were purchased from a local grocery store.

Starch analysis

Total Starch (TS) was quantified using Megazyme Total Starch kit (Megazyme, Inc., Wicklow, Ireland) and means taken for initial rice flour and final pudding product. Total starch amount was analyzed for parboiled-brown rice flour, and flour amount per serving was calculated based on total starch content. Rapidly-digestible starch (RDS), slowly-digestible starch (SDS), and resistant-starch (RS) were analyzed for both parboiled-brown rice flour and pudding using a modified Englyst method [5] based on initial research [7]. 20 mL of sodium acetate (0.1M, 5.2 pH) was vortexed with 800 mg sample, and 5 mL of enzyme solution (450 mg pancreatin, 6mL amylocosidase) was added to each sample tube in addition a blank and 25 mg/mL glucose control. After 20 min and 100 min of enzymatic digestion, 0.5 mL was taken at both time points and added to 20 mL of 80% ethanol. After deactivating enzymes, 0.1 mL of solution was carried out in the glucose assay (GOPOD). The starch fractions RDS and SDS were observed at 20 min and 120 min, respectively. RS was determined by the difference of RDS and SDS.The parboiled brown rice flour had 30.6% amylose content as determined by iodine method after drying overnight and defatting for 5 h with hexane [29]. Pancreatin and amyloglucosidase enzymes from Sigma-Aldrich (St. Louis, MO) were used for *in vitro* starch analysis. All other reagents used for *in vitro* analysis were of analytical grade.

Experimental design and sample preparation

A human study was approved by the Institute of Research Board (IRB) at the University of Arkansas and conducted to investigate plasma glucose and insulin responses. A randomized-crossover design was implemented and responses were analyzed after two 3 h periods over 2 wks. Fourteen healthy, nonsmoking male subjects with age range of 18-45 y not taking medication were recruited to participate in the study. Healthy male subjects were recruited to minimize metabolism variability in addition all subjects' fasting blood glucose levels were <100 mg/dL. Participants were randomly divided into three separate cohorts and each cohort included 4 or 5 subjects. A one-week washout period was conducted between treatments. After fasting 10-12 h, subjects consumed one serving of parboiled brown rice pudding containing 50 g of starch or one 273 mL bottle of 50 g glucose reference drink, Fisher brand SUN-DEX® from Fisher Diagnostics, LLC (Middletown, VA) along with 200 mL of water. Subjects were not allowed to drink additional water during testing. The parboiled brown rice pudding product contained 140 g water, 59 g parboiled *Wells* brown rice flour, 2 g lemon extract, and 1.2 g sucralose-artificial sweetener and ingredients were mixed immediate before consumption. The pudding did not receive any heating treatment. Based on total starch analysis, 59 g of parboiled brown rice flour contained 50 g of available starch. The 50 g glucose reference beverage was chosen because it did not receive cooking treatment and also consumed in the same amount of time and manner as the parboiled brown rice flour pudding.

Postprandial plasma-glucose-and-insulin concentration analysis

After 10-12 h fasting, ~0.4 mL blood sample was collected as a baseline measurement 15 min prior to each treatment as a reference. Subjects consumed treatments within 2 min and blood samples were taken at 0, 15, 30, 45, 60, 75, 90, 120, and 180 min increments. Lancets (Roche Diagnostics, Indianapolis, IN) were used to obtain whole blood samples and collected with Fisher brand microhemocrit capillary tubes (Middletown, VA). Whole blood samples were collected in 0.6 mL sterile, centrifuge tubes and centrifuged at 7000 rpm for 10 min at 4°C. Plasma was pipetted and transferred to labeled 0.6 mL sterile, centrifuge tubes and stored at -20°C until analysis. Plasma glucose concentrations were measured using ACE® Glucose Reagent from Alfa Wassermann Diagnostic Technologies, LLC with Alfa Wassermann Clinical Analyzer (West Caldwell, NJ). Plasma insulin concentrations were measured using the Human Ultrasensitive Insulin ELISA kit from Mercodia, Inc. (Uppsala, Sweden). Incremental AUC was calculated by the trapezoidal rule [30] for each individual and averaged for treatment responses from the group.

Statistical analysis

Incremental plasma-glucose-and insulin changes based on differences after the baseline measurement were averaged and means in addition incremental AUCs were analyzed using analyses of variance (ANOVA) with 9.2 SAS (Cary, NC). Mean differences at each time point and iAUC were evaluated by a *t*-test using Tukey's adjustment with a significance level at $p<0.05$.

Results and Discussion

Nutritional starch fraction analysis

Significant findings from previous research indicated parboiling process variables, feedstock, and storage influenced final outcome of brown or milled rice [7]. Identifying storage treatment as increasing SDS formation was also significant in addition the decrease of RDS for parboiled rice samples [7]. Findings were consistent with previous research [7,31] and showed nutritional starch fractions can be influenced by cultivar and parboiling conditions. Also, research notes more RS content for cooked brown rice in comparison to cooked white rice, in addition cooked parboiled white rice also reports greater RS content compared to cooked white rice [32]. Total Starch (TS) analysis and RDS, SDS, and RS fractions were consistent for the parboiled brown rice flour and rice pudding as shown in Table 1. Average TS content varied 0.1% between flour and pudding samples. Starch fractions were overall consistent; however, RS content of pudding increased 1.5%

Material	TS (%)	RDS (%)	SDS (%)	RS (%)
Flour	82.0 ± 0.5	55.9 ± 2.2	11.8 ± 2.5	14.2 ± 2.9
Pudding product	81.9 ± 0.1	58.1 ± 1.9	7.9 ± 2.4	15.9 ± 2.2

*RDS, SDS, RS values represent mean ± SEM percent per total starch content (n= 4 per sample)

Table 1: *In vitro* mean Total Starch (TS) analysis and Rapidly Digestible (RDS), Slowly Digestible (SDS), and Resistant (RS) starch fractions for parboiled brown rice flour and pudding product.

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Subject Group Number (n)	14
Ethnicity	African/African American (n=2) Asian/Asian American (n=6) Caucasian (n=6)
Age (y)	26.8 ± 4.9
Body Mass Index (kg/m²)	26.5 ± 3.4
Fasting Blood Glucose (mg/dL)	83.1 ± 7.9

Table 2: Male participant information including ethnicity, age, body mass index, and screened fasting blood glucose.

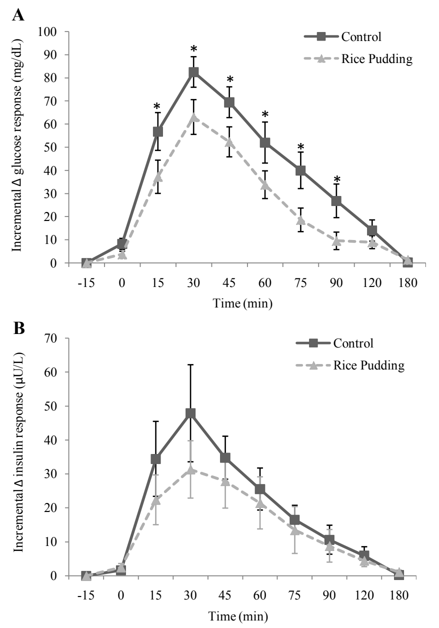

Figure 1: Mean incremental plasma glucose response (A), 15 min before consumption to 180 min after consumption (Control n= 14, Rice Pudding n=14). Mean incremental plasma insulin response (B) from 0 min to 180 min after consumption displays reduced response. (Control n=12, Rice pudding n=10). Each value represents the mean ± SEM. * indicates significant difference P<0.05.

compared to flour while 3.9% SDS content increased in the flour. A 2.2% increase of RDS in the pudding samples may is due to the starch granule swelling from water and slight modifications of starch digestibility in the pudding. Overall, rice flour and rice pudding enzymatic starch analysis of RDS, SDS, and RS fractions did not significantly change, and the rice pudding did not receive heat treatment before consumption. The control glucose reference drink provided a good reference of digestion to the pudding because it also did not undergo heat treatment and consumed in a similar manner.

Participant profile and postprandial glucose responses

Table 2 illustrates the participant profile of the study group. Fourteen participates identified themselves as either: African/African American, Asian/Asian American, or Caucasian, and participates represented either the normal or overweight BMI (Body Mass Index) category. Analysis of incremental glucose responses based on BMI group was observed, however, no significant difference in responses was determined between the groups (data not shown). Incremental glucose response of the parboiled brown rice pudding was significantly lower at 15, 30, 45, 60, 75, and 90 minutes compared to the control glucose reference drink (P<0.05) (Figure 1A). Also, while observing the group response, the mean incremental AUC for the control was significantly different at 5880 ± 658 mg/dL compared to the rice pudding incremental AUC of 3795 ± 602 mg/dL as shown in Figure 2A (P<0.05). Previous research with studying the glucose response rapidly available starch from Englyst and others [33] demonstrated the reduction in rapidly available starch had a profound impact on postprandial glucose response. Although the SDS and RS fractions were targeted by parboiling the brown riceflour, the indirect decrease of RDS was also an effect. A study conducting by Panlasigui and Thompson [34] observed a reduction in blood glucose for both normal and diabetic subjects. Brown rice compared with milled rice had a nearly 20% reduction in glycemic area in healthy subjects and 35% reduction in diabetic subjects. Previous research has indicated brown rice does have a lower starch digestion in addition glycemic response, yet some results report no changes observed as well [27,35]. Conflicting results may have been due to amylose content as previous *in vitro* starch digestion research has noted [14], but also physiochemical properties such as the degree of gelatinization of the rice starch content from heating conditions may also influence digestibility [27]. Perhaps with a larger participant group differences in glucose response would be more pronounced and investigating additional rice cultivars which have shown high SDS and RS contents.

Postprandial insulin responses

A reduction in plasma insulin concentrations was also observed in 12 participants for the rice pudding treatment. Due to limited sample volume, only 12 subjects' insulin samples were analyzed for the control glucose treatment, and the same 10 subjects' samples were available for the pudding treatment. Although no specific time interval was significantly different (Figure 1B), mean incremental AUCs for treatments reflected a strong, similar trend as observed in the glucose response for the participant group. Figure 2B shows incremental AUC response to the control glucose treatment was 3066 ± 525 µU/L compared to 2219 ± 715 µU/L of the parboiled brown rice pudding, an average 28% less response compared to the control treatment. Although the mean insulin responses did not significantly differ at time intervals as in the glucose responses, increasing group size would

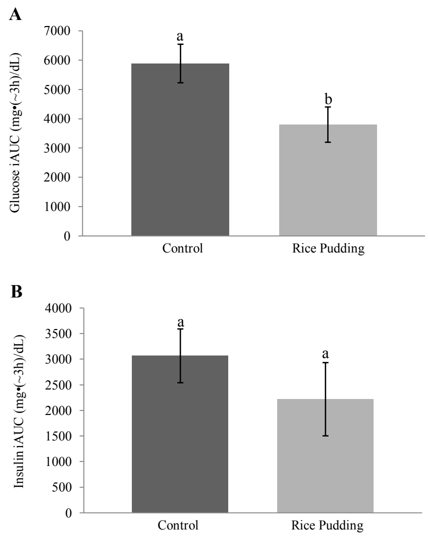

Figure 2: Mean iAUC (incremental Area Under the Curve) plasma glucose response with SEM (A) (Control n= 14, Rice Pudding n=14). Mean iAUC plasma insulin response with SEM (B) (Control n=12, Rice pudding n=10). Different letters indicates significant difference P<0.05.

perhaps show significant differences at time intervals. Also, additional studies investigating long-term effects of regular consumption of starch products with the parboiled brown rice flour or similar starch types may offer benefits for insulinemic control.

Conclusion

The results of this study show that consumption of the parboiled brown rice pudding reduced the postprandial plasma glucose to 36% and insulin to 28% compared with the control treatment. Our study suggests parboiled brown rice has a potential for use as functional food ingredient to improve human health such as lower blood glucose, decreased insulin release, and weight control.

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