Proximate Composition and Glycaemic Index of Destarched Rice

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ABSTRACT

There is strong evidence linking low glycaemic diets and reduced risk of developing diabetes mellitus (DM). This study was designed to develop low glycaemic index (GI) food using rice. Rice was subjected to enzymatic treatment at 40 °C for 8 mins for destarching. After the incubation, the proximate composition and GI of the processed rice were determined. Proximate analysis was carried out using the standard methods of AOAC, whereas GI was assessed using a postprandial oral glucose test. The results showed a significant reduction ($p < 0.05$) in percentage carbohydrates in the processed rice compared with the unprocessed control. Other parameters such as % moisture and lipids were also reduced in the processed rice when compared with the unprocessed control. Furthermore, the percentage of proteins and fibres were significantly increased in the processed rice compared to the unprocessed control. The GI of the processed rice was 69.86% as against the 86.43% of unprocessed rice, representing a 16.57% reduction. Overall, the results suggest that the enzymatic treatment of the rice have the potential of reducing both the starch content and GI of the rice. This can be explored in the development of a diabetic diet.

Keywords: Alpha-amylase, Destarching, Glycaemic index, Proximate composition, Rice

INTRODUCTION

Foods, which provide nutrients for normal body functions have also been identified to play key roles in the prevention and management of chronic diseases such as diabetes, hypertension and cardiovascular diseases. Due to the high cost and multiple side effects that are associated with conventional drugs, there have been many efforts in search of alternative options from natural sources that could be cheap, affordable and with fewer side effects. Therapeutic strategy using nutrition plays an essential role in the management of type 2 diabetes and associated complications (Schwingshackl *et al*., 2018).

Reports indicate that low-glycaemic index (GI) diets may be valuable in glycaemic control and could be effective in mitigating clinical sequelae of diabetes mellitus (DM) (Dong *et al*., 2011; Livesey *et al*., 2019; Zafar *et al*., 2019). Glycaemic response is a normal physiological process that relies on the amount of glucose that enters circulation, the amount absorbed and tissue uptake as well as hepatic regulation of the release of glucose (Triplitt, 2012; Vega-López *et al*., 2018). In DM, these processes are impaired and hinder proper glucose utilization, leading to hyperglycaemia. Carbohydrate diets have a variety of effects on glycaemic response. While some result in a rapid increase accompanied by a rapid fall in blood glucose level, others show a lengthy rise and slow extended decrease in glucose concentration (Vega-López *et al.,* 2018). A systematic review and meta-analysis indicated that fructose, a low GI sugar lowered peak postprandial blood glucose and insulin in people with prediabetes, type 1 and type 2 diabetes (Evans *et al.,* 2017). Low-GI diets have been reported to improve body weight, body mass index, low-density lipoprotein and total cholesterol in obese subjects (Zafar *et al.,* 2019). With existing evidence of therapeutic benefits of low-GI diets, using available local

foods to reduce their GI would play a significant role in the development of cheap and affordable food for DM.

Rice (Oryza sativa) is an edible starchy cereal grain and the grass plant of the family, Poaceae. The rice kernel (paddy) is enclosed by the hull or husk and during the milling process, the hull and bran layers of the kernel are usually removed. Apart from carbohydrates, nutrients such as proteins, thiamine, niacin, riboflavin, iron and calcium are present. Rice is an economically important food crop in several developing nations, including some developed countries where its consumption is at an increase (Ajala & Gana, 2015). In Nigeria, rice is a principal staple food and its cultivation serves as an important source of income for people in rural and urban communities. Therefore, harnessing this food crop for the management of DM would help in mitigating the menace and burden associated with the disease. Therefore, this study was designed to reduce the GI of rice with a view of developing a low GI rice that could be used as diabetic food.

MATERIALS AD METHODS

Chemicals

All chemicals used in this study were of analytical grade.

Collection of Sample and Identification

The white rice was obtained from Kofan Doya in the Sokoto metropolis. Its botanical identity was confirmed at the Herbarium section of the Botany Unit, Department of Biological Sciences, Usmanu Danfodiyo University, Sokoto, Nigeria.

Extraction of Alpha-amylase

Alpha-amylase was extracted as previously described (McCleary et al., 2002). Baker's yeast was crushed with mortar and pestle. Thereafter, 10 g of the crushed baker's yeast was measured and dissolved in 200 ml of distilled

water and mixed properly. The mixture was then filtered to obtain the enzyme extract. This extraction was performed on ice to minimize the denaturation of the extracted enzyme.

Determination of Optimum pH and Temperature of Alpha-amylase

The alpha-amylase activity was determined as previously described (Gillard *et al.,* 1977). The assay relies on the substrate, p-nitrophenol alpha –maltoside, which is hydrolysed by alpha-amylase to a chromogenic product, p-nitrophenol and the absorbance was measured using Optima SP-300 spectrophotometer at 400 nm. To study the effect of temperature and pH on the crude alpha-amylase, these factors were varied and the activity of the enzyme was assayed. The temperature range used was between 30- 55 °C, whereas the pH range was between 2 and 10. A graph of absorbance was plotted against varied pH (Figure 1a) and temperature (Figure 1b) values and optimum values of these variables were determined.

Destarching of Rice

A quantity (40 ml) of boiled phosphate buffer (pH 6) was added to 20 g each of the rice samples in conical flasks. The medium was allowed to cool to 40 °C (alpha-amylase optimum temperature). Then,.50 µl of a-amylase was added to the sample. The sample was suspended in a water bath set at 40 °C and heated under continuous stirring for 8 mins. After heating, the liquid part was removed and the solid residues were washed several times with distilled water and then dried at 50°C in an oven for 6 h.

Determination of Proximate Composition

The proximate analysis of rice was determined in processed and unprocessed. Percentage moisture, ash, crude protein, crude fibre, crude lipids and carbohydrate contents were determined according to the standard methods of the Association of Official Analytical Chemists, (2005). Moisture content was determined gravimetrically by drying the samples in an oven at 100 °C to a constant weight. Crude lipid was measured using Soxhlet apparatus with n-hexane as the extracting agent (40–60 °C), whereas crude protein was determined according to the Kjeldahl method. Ash content was assayed by incinerating the samples in a muffle furnace at 550 °C. The crude fibre was determined using sequential hot digestion with acid and alkaline solution of the defated sample, which was followed by thorough washing with boiling water and drying. Available carbohydrate was determined by difference (% carbohydrate = 100 - % (ash + moisture +crude protein + crude lipid + crude fibre).

Experimental Animals

Eighteen (18) rats were used in this study. The animals were purchased from the Department of Biological Science, Usmanu Danfodiyo University, Sokoto. They were kept in the Animal House and allowed access to water and feed *ad libitum* before and throughout the experimental time. The rats were allowed to acclimatize to standard laboratory conditions (temperature of 25 ± 2°C and relative humidity of 70 ± 5% for one week before the experiment and these laboratory conditions were maintained during the experimental period.

Grouping of Animals

Animals were randomized into three groups of six animals per group.
Group 1: Standard (glucose)
Group 2 Control (non-processed rice)
Group 3: Test (processed rice)

Administration of Test Sample and Glucose Estimation

Two (2) grams each of processed and non-processed rice was made into a slurry paste with 20 ml of boiled distilled water and administered orally to rats at a dose of 0.71 g/kg. The glucose (0.71 g/kg) was also given to the animals, which served as the standard.

Determination of Blood Glucose

Blood was collected from the tail of the rats every 30 mins for glucose estimation using a glucometer. The tail was punctured with a lancet and the blood was allowed to drop on the glucometer strip, the value of glucose concentration was displayed on the glucometer screen and the readings were recorded.

Determination of the Glycemic Index

The glycemic index was determined from a plot of glucose concentration against time using the incremental area under the 2 h blood glucose response curve (AUC) following 12 h of fast and oral administration of the test food and standard glucose. GI was calculated from the incremental area under the curve after the test food was given divided by the incremental area under the curve of glucose and multiplied by 100 (Dodd et al. 2011). The average glycemic index value was calculated from the data collected from 6 rats.

Statistical Analysis

Data are expressed as mean ± standard deviation. The mean was used to plot a graph of concentration in (mmol/l) against time (min). The area under the curve was determined using GraphPad Prism (version 6) statistical software. The proximate composition of processed rice was compared with control using a t-test. A $p < 0.05$ was set as a significant level.

RESULTS

The results of optimal pH and temperature of the isolated alpha-amylase are depicted in Figures 1a and b. The optimum pH and temperature were 6.0 and 40 °C., respectively.

The results of the percentage proximate composition of processed rice are presented in Table 1. Incubation of rice with alpha-amylase at 40 °C for 8 mins showed a significant reduction (p <0.05) in carbohydrate contents when compared with unprocessed rice. The moisture and crude lipid contents were also reduced following incubation but the differences were not statistically significant compared to control. Furthermore, the percentage of ash, crude fibre and protein were significantly increased after treatment.

The eight (8) mins for incubation was the optimum time and this was selected based on the result of optimization.

The results of glucose from the 2 h postprandial test is presented in Figures 2 a and b. The results showed the incremental area under the curve of the processed rice was lower than that of the unprocessed rice. The GI of processed rice (69.86%) was less than that of unprocessed rice (86.43%).

(a)   (b)
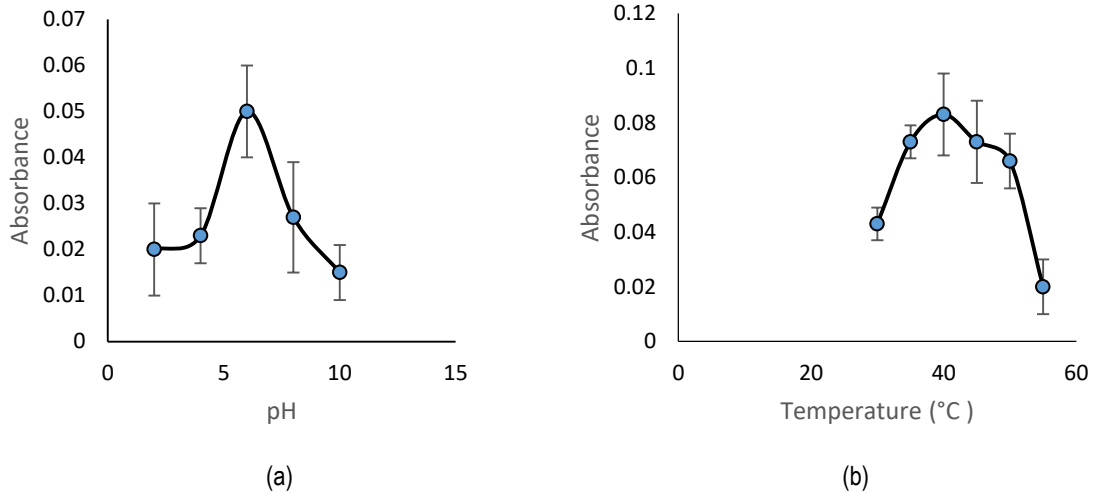
Figure 1: Optimum pH (a) and temperature (b) of the isolated alpha-amylase
Values are mean ± SD of three replicates from two independent experiments

(a)   **(b)**
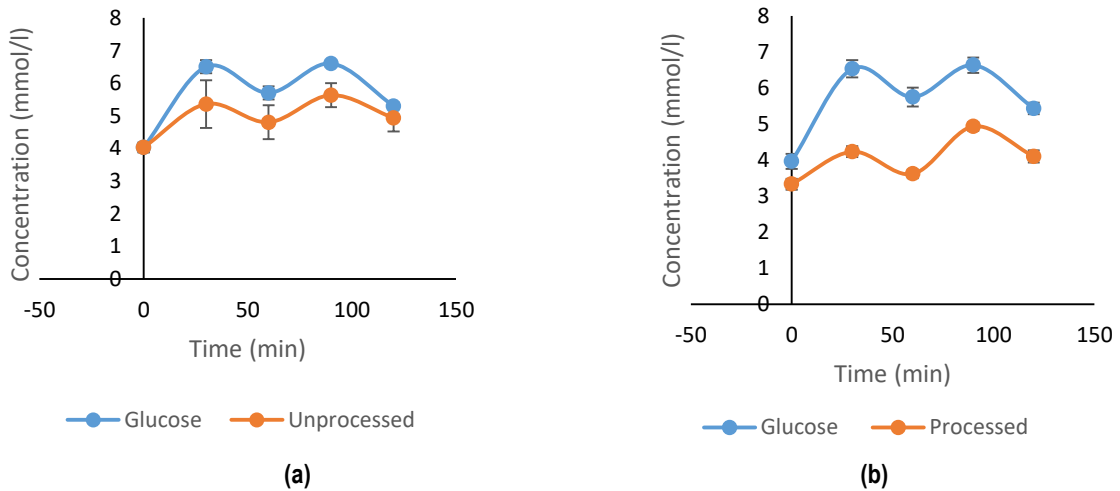
Figure 2: Oral glucose tolerance tests of processed and unprocessed rice
Incremental area under the curve of unprocessed (a) and processed (b) rice. Values are mean ±SD of six rats per group

Table 1: Proximate composition of rice treated with alpha-amylase

PARAMETER (%)	UNPROCESSED RICE	PROCESSED RICE
Carbohydrate	80.10±0.09	72.46±1.42*
Moisture	12.08±0.08	11.16±0.76
Crude fibre	0.19±0.01	1.16±0.28*
Ash	0.37±0.02	7.16±0.57*
Crude lipid	1.25±0.02	0.83±0.28
Crude protein	5.97±0.01	7.17±0.08*

Rice was treated with alpha-amylase at 40 °C for 8 mins
Values are mean ±SD of three replicates from two independent experiments
Rows with an asterisk are significant when compared with control using t-test

DISCUSSION

DM and its associated complications are major causes of morbidity and mortality. Low GI diets seem to be an alternative option for postprandial glycaemic control in individuals suffering from DM. In this study, we investigated how local staple food (rice) can be explored for the management of DM. There were improvements in the contents of protein, crude fibre and ash, whereas carbohydrates, lipids and moisture were decreased in the processed rice when compared with the unprocessed rice. Improvement in the protein and fibre and decrease in carbohydrates suggests that the method adopted in reducing the starch content of the rice was effective. Amylase is an enzyme that hydrolyses starch and when starch is heated with water, the granules absorb the hot water to swell and burst, releasing the starch into the water. Therefore, using amylase along with heating resulted in the reduction of the carbohydrates content of the rice. It has been shown that parboiling of rice before milling caused starch retrogradation which increased the amylopectin and amylose crystallization and hence, reduced the carbohydrate content and glycaemic response (Kalita *et al.* 2021). The increase in fibre content may be due to less susceptibility of the fibre, especially the insoluble ones to hydrolysis during the processing. Similarly, the increase in protein content may be attributed to the loss of dry matter largely carbohydrates after the processing. This observation is consistent with the previous report that the treatment of rice with carbohydrate-hydrolysing enzymes increased the protein content (Shih & Daigle, 1997). Furthermore, Kalita *et al.* (2021) have demonstrated that parboiled milled rice had high fibre content than unparboiled milled rice. The decrease in lipid content after processing may be attributed to an increase in lipolysis, resulting in the release of fatty acid.

The role of dietary fibre in the prevention of chronic diseases such as diabetes and cardiovascular diseases have been well documented in the literature. A meta-analysis of 44 trials on the role of high-dietary fibre diets on mortality and glycaemic control and other risk factors of cardiometabolic diseases in adults with diabetes indicated an improvement in glycaemic control, blood lipids, body weight and inflammation, resulting in a reduction in premature deaths (Reynolds *et al.*, 2020). The significant improvement in the fibre content of the rice observed after enzymatic treatment is an indication that it can be explored for glycaemic control in DM. As a means of glycaemic control in individuals with DM, protein-rich diets are usually recommended. From our results, a significant improvement of protein was observed, suggesting that the rice obtained can serve as a source of protein with a low percentage of carbohydrates for DM subjects. A reduction in lipids content was also observed, signifying that the rice may be suitable for DM patients as they are recommended to limit their lipid intake. One of the common features of diabetes is dyslipidaemia and there is a strong connection between atherosclerotic cardiovascular disease and serum cholesterol and triglycerides levels in subjects with type 1 and 2 diabetes (Schofield *et al.*, 2016; Warraich & Rana, 2017).

To validate our results whether the reduction in carbohydrate, lipid and moisture contents and increase in amounts of proteins, fibres and ash could translate into a reduction in GI, the postprandial glucose levels in rats was tested. The GI of the processed rice was found to be 69.86% compared to 86.43% of the unprocessed rice. Low GI food has been reported to improve clinical sequelae of DM (Grant *et al.*, 2011; Zafar et al., 2019). These studies reported improvements in glycaemic control, blood glucose, glycated haemoglobin, body mass index and cholesterol levels after intervention with low GI diets in these studies. Furthermore, a meta-analysis of 11 randomized trials involving 402 participants showed that low GI improved glycaemic control in diabetes without compromising hypoglycaemic events (Thomas & Elliott, 2009). This strategy of using low GI may be effective in the prevention and management of DM and its associated complications. Going forward, exploring local food materials for DM would provide a cheap and affordable therapy in glycaemic control to combat the menace posed by DM. If this is achieved in an unbiased manner, it would provide a cheap option with minimal side effects for DM prevention and management.

CONCLUSION

This study provides an insight into the exploration of rice for the production of low GI food. Our findings showed that

treatment of rice with alpha-amylase results in a reduction in percentage carbohydrates and GI and improved fibre and protein contents. Hence, this study provides evidence that alpha-amylase can be used to lower the GI of carbohydrate-rich foods under suitable treatment conditions.

CONFLICTS OF INTEREST
The authors declared that they have no conflicts of interest.

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