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Effect of hot and cooled carbohydrate diet on glycemic response in healthy individuals: a cross over study

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ABSTRACT

Background: Cooling of starch after cooking is known to cause starch retrogradation which increases resistant starch content. Resistant starch cannot be digested in the gut and acts as dietary fiber. The study aimsed to determine the effect of cooling of carbohydrate rich diet on glycemic response on healthy adults.

Methods: The present study was a randomized, single blind, crossover study where 20 healthy subjects were selected. Two rice preparations were used, one freshly prepared hot, second, cooked and cooled at 4°C for 12 hours. All subjects were evaluated after giving both rice preparations separately with a crossover period of 7 days. Glycemic response was checked over a period of 2 hours at various time intervals using ACCU-CHEK® Active glucometer.

Results: Glycemic response with cooled white rice was better in comparison to freshly prepared hot white rice at all time points (mean±SD, 121.9±17.4 vs 128.0± 22.1 mg/dl). However, the difference in means at 30 mins was maximum and statistically significant (p<0.001).

Conclusions: Cooled white rice yields better glycemic response when consumed by healthy individuals possibly due to formation of resistant starch.

Keywords: Cooled, Glycemic response, Retrogradation, Resistant starch

INTRODUCTION

For the past few decades diabetes is on the rise especially in countries like India. Diet is a modifiable risk factor for various life style disorders. Strict dietary control is thus one of the important component of Holistic management of diabetes.

Rice, wheat and maize are used as staple diets in many parts of India. Rice is consumed fresh as well as after being reheated thereby providing varying amounts of starch. Starch is composed of 2 structural components namely Amylose (linear polymer) and amylopectin (branched polymer). It is the ratio of amylose: amylopectin which determines the digestibility of starch in the intestines. The higher the amylose:amylopectin

ratio the higher the indigestibility of starch thus making it resistant. 1,2 Resistant starch is classified under nondigestible starch fractions as it is indigestible by body enzymes and extensive studies have proved them to have functions similar to that of dietary fibre.3 Resistant starch classified into four subtypes based on its physiochemical properties.^{4,5} Type 1 (RS1) is physically inaccessible form of starch granules, i.e. seeds, partly milled grains.

Type 2 (RS2) is naturally occurring granular starch, such as found in potato and banana. Type 3 (RS3) is retrograded starch made by cooking/cooling of starchy materials. Type 4 (RS4) is chemically modified starch. Foods containing RS3 have relatively increased dietary fiber content and reduced digestible carbohydrate

content.4 Cooling of starch after cooking is known to cause starch retrogradation which increases resistant starch content (RS3). Sonia et al. reported that higher amylose-amylopectin ratio and storage at 1-25°C led to increased retrogradation and formation of RS.^{6,7} Resistant starch cannot be digested in the stomach and small intestine by the pancreatic amylase and enters into the large intestine, where it may be fermented by colonic microbiota to produce short chain fatty acids such as acetate, butyrate and propionate which reduce the pH.8-11 These short chain fatty acids (SCFAs) like acetate, butyrate, propionate etc. offer many benefits including lowering of glycemic response and improving insulin sensitivity(Cummings 1987).¹² RS has shown these beneficial effects in both healthy and those with metabolic syndrome.¹³ However, efficacy of RS in individuals with T2DM has not been investigated in depth.14

The number of studies conducted on this topic are almost negligible. The present study was aimed to assess the effect of the freshly cooked white rice and yesterday's cooled then reheated white rice on the glycemic response in healthy subjects.

METHODS

The present study had a randomised, single blind, crossover study design.

Prior permission from Institutional Ethics Committee, Government Medical College, Jammu (IECGMCJ), and approval number IEC/Thesis/research/I4C/2017/436 was obtained before initiation of study. Written informed consent was taken from every participant before participating in the study. For selection of subjects, list of municipal wards was procured from Municipal corporation Jammu .Out of which one urban census enumeration tract was selected randomly. List of all households was prepared and 80 households were selected randomly using simple random sampling. Potential participants were individually contacted by the Investigator and informed about the study.

Inclusion criteria

Healthy individuals with no Co-morbid conditions, age between 20-70 years, fasting plasma glucose $\leq \! 100$ mg/dL, non smoker, non alcoholic and who eats breakfast regularly and on time daily were included in the study.

Exclusion criteria

Pregnancy/ breastfeeding, significant current/ past medical history (Hypertension, T2 DM etc., BMI≥30 kg/m², under any medication likely to interfere with glucose metabolism, history of any known white rice allergy or any food allergy, history of any malabsorption syndromes were excluded from the study.

Rice preparation

An Initial survey regarding the most common variety of rice consumed locally was done. The rice chosen for this study was grown in RS Pura Belt of Jammu region which is consumed by majority of the population here. This variety of rice is medium grain basmati. Rice was prepared in an open top vessel in which ratio of rice to water was maintained at 1:1.5, over a gas stove for about 20-25 minutes covered with a lid. No water was strained out of the vessel.

For the second session i.e cooled rice, same procedure of cooking the rice was followed, however in this case rice was cooled in a refrigerator at 4°C for 12 hours overnight maintaining the temperature at a constant throughout. Reheating of the rice was done before the session begun, rice was reheated in a microwave oven to maintain a uniform temperature for reheating. It was reheated at 80 percent power for a time period of 3 minutes. Reheating was done separately for each subject for uniformity.

Study

All the initially selected subjects were pre-informed about the interview a night before and were asked to keep a fasting state till the interview which was conducted around 8-9 AM. During the interview subjects were asked to fill a questionnaire based on daily routine dietary practices, type of rice used, level of physical activity, health history to confirm the health status and history of any current/past drug intake.15 At the same time fasting blood glucose was measured using ACCU-CHEK® Active glucometer (Manufactured by-Roche diabetes care, Sandhofer Strasse 116, Germany) to ensure subjects did not have impaired fasting blood glucose. Of these subjects interviewed, 40 were finally selected to participate in the study. Before the first study visit, Subjects were instructed to have dinner before 8 PM the night before the study. Subjects were allowed to have meal of their choice, preferably less carbohydrate rich diet. And proper record of the food and beverages eaten was kept. A fasting of at least 12 hours by the subjects was desirable. Subjects were told to avoid unusual vigorous physical activity on the day of each study visit.

Two types of rice preparations were used in the study, hot freshly prepared rice and yesterday's cooled then reheated rice. Subjects had to attend two breakfast sessions, one with hot and other with cooled then reheated rice. The subjects were not informed of which type of rice was being served. The crossover was done between the two sessions which were separated by 7 days to allow complete washout. The subjects were advised to maintain normal daily routine diet in between the crossover and same precautions aforementioned to be followed before the second session.

The breakfast session started at 8:00 AM to 8:30 AM. At each session subjects were given 200 g of cooked white

rice with a standard portion of 50 g of cauliflower curry and a glass of water. Subjects were asked to finish in no more than 15 minutes. Freshly cooked or reheated rice was served warm, immediately after preparation. As some of the subjects were vegetarians among the other non-vegetarian subjects, In order to maintain the uniformity of the study cauliflower was given to all as it is widely acceptable to people and eaten in the region throughout the year. Blood glucose measurements were conducted using ACCU-CHEK® Active glucometer at time 0 (before the first bite) and 15, 30, 45, 60, 90,120 minutes.

Statistical analysis

Data was entered in MS Excel. Continuous parametric variables were reported as Mean±SD. Difference in mean was evaluated using Mixed method ANOVA with repeated measures (within subjects) using SPSS version 20.0 (IBM). A p value of less than <0.05 was considered

as statistically significant. All p values reported are two tailed.

RESULTS

A total of 40 healthy subjects were recruited in the study. Out of which, 18 were females and 22 were males. Mean age of subjects was 42.6 ± 14.9 years and Age Range was between 24-68 years. Baseline BMI was 24.6 ± 3.2 kg/m². Mean fasting plasma glucose before intervention was 88.3 ± 4.6 mg/dl (Table 1).

Table 1: Distribution of subjects according to baseline characteristics.

| Characteristic | Mean± SD |
|-------------------------------|------------|
| Age (years) | 42.6 ±14.9 |
| Body mass Index (kg/m²) | 24.6±3.2 |
| Fasting plasma glucose(mg/dl) | 88.3±4.6 |

SD- Standard deviation

Table 2: Blood glucose measurements at different time intervals.

| Type of food | Time (in minutes) | Mean blood glucose levels(mg/dl) | Standard Deviation | Mixed method repeated measures ANOVA |
|--------------------------|-------------------|-------------------------------------|-----------------------|--------------------------------------|
| | 0 | 89.8 | 6.2 | F (1,39) = 14.08 p<0.01 |
| | 15 | 137.2 | 18.2 | |
| | 30 | 156 | 29.9 | |
| Hot food | 45 | 146.6 | 38.7 | |
| | 60 | 132.6 | 39.5 | |
| | 90 | 122.7 | 24.9 | |
| | 120 | 111.8 | 16.6 | |
| | 0 | 90.1 | 5.9 | |
| | 15 | 125.4 | 16.9 | |
| Cooled and reheated food | 30 | 142.7 | 23.4 | |
| | 45 | 137.3 | 31.2 | |
| | 60 | 128.9 | 31.5 | |
| | 90 | 118.4 | 24.6 | |
| | 120 | 111 | 12.8 | |

Blood glucose levels at 0 min, 120 min did not differ significantly between two sessions. However, at 15 min, 30 min, 45 min, 60 min and 90 min, mean blood glucose levels of subjects served cooled reheated rice was significantly lower than the subjects served hot rice. It reached maximum level at 30 minutes in both groups. There was no statistical difference between the glycemic indices of two treatments. Mean blood glucose levels differed significantly between different time points and within subjects. (F(1,39) = 14.08 p < 0.01) (Table 2).

Profile plot depicted that overall means of blood glucose levels at various observed times were higher in case of freshly cooked hot food as compared to ingestion of cooled and then reheated food. Peak of mean blood glucose levels were seen at 30 minutes in both groups (Figure 1).

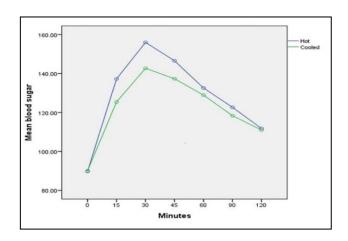


Figure 1: Profile plot depicting variations in blood glucose levels at different time intervals.

DISCUSSION

In the present study 40 subjects participated in both the sessions, which was more than the study by Sonia et al. (n=15), and by Chiu et al. (n=21).7,15 No study has been yet conducted in healthy individuals with a larger sample size. The mean age of subjects in our study was 42.6±14.9 years which was higher than mean age of participants in study by Sonia et al. while Chiu et al. reported mean age of 29.3 years. In our study blood glucose levels at 15, 30, 45, 60, 90 min were significantly lower after ingestion of cooled then reheated rice compared with freshly prepared hot rice. The difference was maximum at around 30 min which corresponds to the time when the blood sugar levels are expected to be at the peak following a carbohydrate rich meal. Blood glucose levels did not differ at 0.120 mins. Ananda et al. also reported lower blood glucose levels at 45 through 120 min after ingestion of cold cooked white rice (cooled for 10 hours at 3°C) compared with warm cooked white rice. 16 Dewi and Isnawati found lower postprandial blood glucose levels after ingestion of vesterday rice (cooled for 24 hours at 4°C and then reheated) compared with freshly made rice, although the differences were found to be not statistically significant.17

Rice has historically been considered having low dietary fibre, Chiu YT et al, reported resistant starch content varies with the cooking method (steamed, boiled, strained or pressure cooked), It has been reported that the variety of rice, determines significantly the RS content, and rice with high RS content will result in a lower postprandial glycemic response. Walter et al, also reported similar results. However in this clinical study we focused on using one variety of rice which was consumed the most locally. The resistant starch content in the rice variety used however, was not measured due to limited resources.

White rice is considered a high GI food with a mean GI to be 73 as reported by Atkinson et al. however, the GI and the digestibility varies significantly between the different varieties of rice depending upon the amylose content and other physiochemical properties like cooling, gelatinization, particle size. ¹⁹⁻²¹

Insulin resistance leading to Type 2 Diabetes Mellitus lifestyle and pharmacological requires diet. modifications. Of these, the dietary modifications are the least explored aspects. Increased saturated fats and decreased amount of fibre in diet is linked to decreased insulin sensitivity.²² Increasing consumption of non viscous fibres i.e RS has shown beneficial effects on postprandial blood glucose and increased insulin concentrations in people with normal or impaired blood glucose. Yamada et al. reported that single ingestion of bread containing 6 g RS significantly postprandial glucose and improved insulin responses in subjects with fasting blood glucose >110 mg/dl. The

treatment had no effect on subjects with fasting glucose $<110 \text{ mg/dl.}^{23}$

Bodinham et al. showed that RS improves glucose and insulin metabolism through increased postprandial GLP1 secretion i.e. Incretin due to stimulation of colonic enteroendocrine cells. This can result in improved insulin secretion. These effects observed however have been seen after long term RS consumption. Also, Johnston et al. reported that RS may alter insulin sensitivity by changes in adipokines arising from adipose tissue remodeling in addition to acute changes in rate of adipose tissue lipolysis. However one should keep in mind that these conclusions were drawn based on animal based studies and human trials to deduce the same are further required for conclusive evidence.

RS being indigestible in the small intestine, interacts with the colonic microbiota and ferments into short chain fatty acids (SCFA's) which lowers the pH of the colon. SCFA's are rapidly absorbed from the large bowel stimulating sodium and water reabsorption thus playing a role in maintaining homeostasis. In addition, colonic epithelial cells utilize SCFA's as a fuel especially butyrate which has regulatory effects on nucleic acid metabolism. ^{12,26,27} However no effect on serum fasting levels of SCFA is seen.

Chronic consumption of RS is hypothesized to improve insulin sensitivity and insulin secretion. The beneficial effects of RS may however be only evident with long term intake. Resistant starch could be used as a modality of dietary modification in form of dietary supplements or incorporated in daily diet in people with T2DM, metabolic syndrome or even healthy individuals. More studies and large scale interventions are required before any dietary recommendations could be proposed. However, based on this clinical study a practice/habit of consuming carbohydrate rich diet (like white rice, pasta, potatoes etc) on cooling rather than consuming immediately when its hot could be proposed.

Limitations

Due to the small sample size, generalizations cannot be made on a larger scale. Also, this study focused only on white rice as the carbohydrate source, which might not be the popular choice in other parts of the world. Due to the limited amount of resources, the resistant starch content could not be measured in the variety of rice used.

CONCLUSION

Ingestion of cooked white rice cooled at 4°C for 12 hours then reheated yields better glycemic response than freshly cooked white rice in same portion size when consumed by healthy individuals due to formation of resistant starch.

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Conflict of interest: None declared

Ethical approval: The study was approved by the

Institutional Ethics Committee

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