International Journal of Research in Medical Sciences
Dipnaik K et al. Int J Res Med Sci. 2017 Oct;5(10):4502-4505
www.msjonline.org
pISSN 2320-6071 | eISSN 2320-6012
DOI: http://dx.doi.org/10.18203/2320-6012.ijrms20174585

Original Research Article

Ratio of Amylose and Amylopectin as indicators of glycaemic index and in vitro enzymatic hydrolysis of starches of long, medium and short grain rice

Kavita Dipnaik[1]*, Praneeta Kokare[2]

[1]Department of Biochemistry, LTM Medical College and General Hospital, Sion, Mumbai, Maharashtra, India
[2]Department of Biochemistry, Preventive Laboratory, Sanpada, Navi Mumbai, Maharashtra, India

Received: 02 August 2017
Accepted: 28 August 2017

***Correspondence:**
Dr. Kavita Dipnaik,
E-mail: kavita.dipnaik@yahoo.com

ABSTRACT

Background: Rice (Oryzasativa L.) is the staple food of over half the world's population. The major carbohydrate of rice is starch, which is about 72 to 75%. Rice can be classified into three different types: long-grain, medium-grain and short-grain rice based upon their length as compared to their width. Aim of the study was to predict the glycemic index of long, medium and short grain rice based on amylose, amylopectin ratio and to study in vitro hydrolysis of starch of long, medium and short grain rice by salivary and pancreatic amylases and formulate the right type of rice to be consumed by diabetics.
Methods: Starches were isolated from long, medium and short grain rice. Amylose and amylopectin from the isolated starches were estimated. Starches isolated were subjected to enzymatic hydrolysis by salivary amylase and pancreatic amylase under optimum in vitro conditions and reducing sugars released after hydrolysis and incubation period of 0, 5, 10, 15, 20 and 30 minutes were estimated by Folin-Wu method. Results were analysed using unpaired t-test and statistical significance was established.
Results: Long grain rice was found to have high amylose and low amylopectin content as compared to medium and short grain rice. Long grain rice showed slow release of reducing sugars as compared to medium and short grain rice.
Conclusions: Long grain rice because of its high amylose content is a low glycemic food and can be consumed by diabetics. Sustained slow release of reducing sugars given by long grain rice is desirable in diabetics.

Keywords: Amylases, Amylopectin, Amylose, Glycemic index (GI), Starch

INTRODUCTION

Rice (Oryzasativa L.) is the most important cereal crop in the developing world and is the staple food of over half the world's population. The major carbohydrate of rice is starch, which is about 72 to 75%. Rice can be classified into three different types: long-grain, medium-grain and short-grain rice. The three different types of rice grains are categorized based upon their length as compared to their width.[1] If the length of rice grain is four times more than its width then it is considered to be a long grain rice. Short-grain rice is short and fat. It often looks round in shape because the width is close in size to the length. If the length of rice grain is three times more than its width then it is considered to be medium grain rice. Starch consists of two components: Amylose which is a long, unbranched chain with 200-1000 D-glucose units held by alpha (1-4) glyosidic linkages. It is water soluble in nature. White rice with low amylose content has high GI value as amylose content has a negative relationship with GI value.[2] Amylopectin is a branched chain with alpha (1-6) glyosidic bonds at the branching points.

Amylopectin molecule containing thousand glucose units looks like a branched tree (20-30 glucose units per branch). The amylopectin chains are 120-400 nm long and their relative molecular masses may reach 15-30 million.[3] Amylase is an enzyme belonging to the class of "hydrolases" (E.C.3.2.1.1) which catalyses the breakdown of starch. In human body α-amylase is found primarily in saliva and pancreas.[4]

The digestion of starch begins in the mouth. Saliva contains α-amylase, which randomly hydrolyzes all the α(1-4) glucosidic bonds of starch. By the time thoroughly chewed food reaches the stomach, where the acidity inactivates amylase, the average chain length of starch has been reduced from several thousand to fewer than eight glucose units. Starch digestion continues in the small intestine under the influence of pancreatic amylase. This enzyme degrades starch to a mixture of the disaccharide maltose, the trisacharride maltotriose, which contains three α(1-4) linked glucose residues, and oligosaccharides known as dextrins that contain the α (1-6) branches. These oligosaccharides are hydrolyzed to their component monosaccharides by specific enzymes contained in the brush border membranes of the intestinal mucosa: an glucosidase, which removes one glucose residue at a time from oligosaccharides, an dextrinase or debranching enzyme, which hydrolyses α (1-6) and α (1-4) bonds.[5-7] The resulting monosaccharides are absorbed by intestine and transported to the bloodstream. Slow release of reducing sugars is desirable in diabetics.[8] Starch is fractionated into its structural components amylose and amylopectin by McCready and Hassid method.[9,10] Starch can be subjected to action of salivary and pancreatic amylases in vitro and the resulting reducing sugars are estimated by Folin-Wu's method.[11]

METHODS

Starches were isolated from long, medium and short grain rice which were procured from local grocery shop. Starch is a homo-polysaccharide composed of amylose and amylopectin fractions. Amylose and amylopectin from the isolated starches were estimated by Mc-Cready and Hassid method. Starches isolated from long, medium and short grain rice were subjected to enzymatic hydrolysis by salivary amylase and pancreatic amylase under optimum in vitro conditions. Source of salivary amylase was natural saliva and pancreatic amylase was in the form of synthetic sample.

Complete hydrolysis of starch by the action of salivary and pancreatic amylases resulted in the formation of reducing sugars. Starches were subjected to hydrolysis for different incubation period of 0, 5, 10, 15, 20 and 30 minutes and the reducing sugars released were estimated by Folin-Wu method in which alkaline copper sulphate is reduced by reducing sugars to cuprous oxide which in turn reduces phosphomolybdic acid to form reduced phosphomolybdic acid which is estimated colorimetrically.

RESULTS

Starch is a polysaccharide composed of amylose and amylopectin. These components were obtained on fractionation in all the three varieties of rice viz. long grain, medium grain and short grain rice.

Table 1 depicts amylose and amylopectin in long, medium and short grain rice starches. Amylose was found to be highest for long grain rice as compared to medium and short grain varieties. Long grain rice had high amylose and low amylopectin content due to which it is a low glycemic food.

Table 1: Concentration of amylose and amylopectin in long grain, medium grain and short grain rice.

Rice type	Amylose in gram %	Amylopectin in gram %
Long grain	10.6	9.4
Medium grain	6.8	13.2
Short grain	6.6	13.4

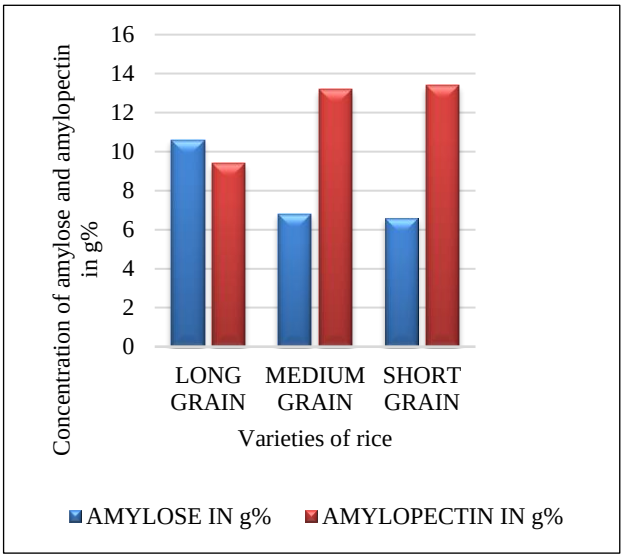

Figure 1: Graphical representation of Amylose and Amylopectin in long grain, medium grain and short grain rice.

The Amylose and Amylopectin ratio is indicator of glycemic index of rice amylose content has negative relationship with glycemic index value Table 2 depicts the ratio of Amylose and Amylopectin in long grain, medium grain and short grain rice.

Amylose and amylopectin ratio was found to be highest in long grain rice as compared to medium and short grain rice.

The action of salivary and pancreatic amylases results in the hydrolysis of starch to form reducing sugars. These reducing sugars were estimated in the time interval of 0-

5, 5-10,10-15, 15-20 and 20-30 minutes by Folin-Wu method.

Table 2: Ratio of Amylose and Amylopectin in long grain, medium grain and short grain rice.

Varieties of rice	Ratio of amylose and amylopectin
Long grain	1.127±0.11
Medium grain	0.515±0.02
Short grain	0.4925±0.01

Table 3 depicts the slow release of reducing sugars as a result of salivary and pancreatic amylases hydrolysis of starch shown by long grain rice as compared to medium and short grain rice. The reducing sugars released by medium and short grain starches were lower in the time interval of zero to five minutes, but as time progressed they released higher quantities of reducing sugars.

Table 3: Reducing sugars released in mg% by long, medium and short grain rice in the following time intervals.

Time interval	Reducing sugars released in mg% by		
	Long grain	Medium grain	Short grain
0-5	22.37	11.07	9.28
5-10	0.56	12.0	10.8
10-15	3.00	3.0	2.1
15-20	6.96	0.3	1.0
20-30	3.01	6.6	7.0

Statistical analysis of ratio of amylose and amylopectin from starches of three types of rice was carried out by students unpaired t-test and statistical significance was established.

Table 4: Statistical analysis of ratio of amylose and amylopectin from starches of following types of rice.

Ratio of Amylose and Amylopectin	Calculated 't'	Table 't' value	Significance
Long versus short grain	109.132	2.78	S
Short versus medium grain	-5.196	2.78	NS
Long versus medium grain	103.93	2.78	S

Table 5: Statistical analysis of reducing sugars released by starches of following types of rice.

Reducing sugars released by starches of rice	Calculated 't'	Table 't' value	Significance
Long versus short grain	0.453	2.23	NS
Short versus medium grain	0.051	2.23	NS
Long versus medium grain	0.978	2.23	NS

Statistical analysis of reducing sugars released by starches of three types of rice was carried out by students unpaired t-test and statistical significance was established

DISCUSSION

Starch is a homo-polymer of glucose made of two components, unbranched amylose and a branched amylopectin. Starches from short, medium and long grain rice samples were subjected to fractionation into its components amylose and amylopectin. Table 1 depicts the starch composition with respect to amylose and amylopectin fractions in short, medium and long grain rice. Long grain rice showed highest amylose and low amylopectin content when compared with short and medium grain rice.

Starches with higher amylose content will be less susceptible to gelatinization that is breaking down into glucose hence giving a low GI.[9,12] Therefore long grain rice has a low GI as compared to short and medium grain rice. Diabetics can consume long grain rice in their diets because of a comparative low GI.

Figure 1 depicts the concentration of amylose and amylopectin in long, medium and short grain rice.

Amylose and Amylopectin ratio can predict the glycemic index of rice. A high value of amylose amylopectin ratio indicates low glycemic index. Long grain rice because of its high value of ratio is a low glycemic food which can be consumed by diabetics. Table 2 depicts the ratio of amylose and amylopectin in short, medium and long grain rice. There is no research data available in support of these three types of rice samples in this respect in the literature.

Starches isolated from short, medium and long grain rice were subjected to salivary and pancreatic amylases hydrolysis in vitro. The sustained release of reducing sugars was found to be comparatively lower in case of long grain rice as compared to short and medium grain rice. This is clearly evident from Table 3. Short grain and medium grain rice showed high amylopectin content as compared to long grain rice, hence showed rapid hydrolysis of starch. The rate of hydrolysis of starch containing high amount of amylopectin was found to be rapid by M. Frei et al.[9] This proves the rapid rate of

hydrolysis seen in short and medium grain rice. There are no research findings in the literature on three types of rice studied namely short, medium and long grain.

Unpaired t-test was used to find significant difference between means and p-value was calculated and statistical significance was established.[13]

Table 4 shows the ratio of amylose and amylopectin for long versus short grains and long versus medium grains showed statistically significant results.

Table5 showed the statistical analysis of reducing sugars released by starches of long, medium and short grain rice. It can be clearly seen that the results obtained which were statistically not significant.

The limitation of the study was it was limited to long, medium and short grain rice which was procured from local grocery shop. Varieties of rice with different amylose and amylopectin contents should be used.

CONCLUSION

Amylose and Amylopectin ratio can predict the glycemic index of rice. Lower amylose content will have higher glycemic index. Starches with higher amylose content will be less susceptible to gelatinization. These starches with higher amylose content do not easily hydrolyse to glucose, this is the reason for low glycemic index of starches with higher amylose content. Long grain was found to have high amylose and low amylopectin content due to which it is a low glycemic food. The long grain rice can be consumed by diabetics because of its comparative low GI. The long grain rice shows slow release of reducing sugars as compared to medium and short grain rice. Sustained slow release of reducing sugars is desirable in diabetics. Long grain rice is the best option as a staple diet when compared with medium and short grain rice.

Funding: No funding sources
Conflict of interest: None declared
Ethical approval: The study was approved by the Institutional Ethics Committee

REFERENCES

1. Julian BO. Rice in human nutrition. International Rice Research institute. Food and agriculture organization of the United Nations; 1993. Available at: http://books.irri.org/9251031495_content.pdf
2. Jeevetha S, Barakatun-Nisak MY, Ngan H, Ismail A, Azlan A. Relationship between amylose content and glycemic index of commonly consumed white rice. IOSR-JAVS. 2014;7(9):12-8.
3. Aarathi A, Urooj A, Puttaraj S. In vitro starch digestibility and nutritionally important starch fractions in cereals and their mixtures. Starch-Stärke. 2003;55(2):94-9.
4. P.M Day, J.B.Harborne, Methods in Plant Biochemistry, Volume 3, Enzymes of Primary Metabolism. U.S edition Published by Academic Press, 1990:105-413.
5. Satyanarayana U, Chakrapani U. Biochemistry, Third revised edition, 2006;166-792.
6. Snow P, O'Dea K. Factors affecting the rate of hydrolysis of starch in food. Am J Clin Nutrition. 1987;34(12):2721-7.
7. Yoon JH, Thompson LU, Jenkins DJ. The effect of phytic acid on in vitro starch digestibility and blood glucose response. Am J Clinical Nutrition. 1983;38:835-42.
8. Voet D, Voet JG. Sugars and polysaccharides. Biochemistry. 4th edition. United States of America, John Wiley and Sons, Inc; 2004:370.
9. Frei M, Siddhuraju P, Backer K. Studies on the in vitro starch digestibility and the glycemic index of six different indigenious rice cultivars from the Phillppines. J Food Chemistry. 2003;83(3):395-402.
10. Kingman, Hudson, John H. Measurement of starch fractions. Br J Nutrition. 1996;75:749-55.
11. Chakrabarathy TK, Dwarkanath KR, Bhatt PB. Studies on physicochemical properties of some varieties of rice. J Food Sci Tech. 1972;9:140-3.
12. James W, Bird A. The effect of a brief salivary alpha amylase exposure during chewing on subsequent in vitro starch digestion curve profiles, Int J Molecular Sci. 2010;11(8):2780-90.
13. Mahajan BK. Methods in biostatistics for medical students and research workers. Jaypee Brothers. 2000;5:83.

Cite this article as: Dipnaik K, Kokare P. Ratio of Amylose and Amylopectin as indicators of glycaemic index and in vitro enzymatic hydrolysis of starches of long, medium and short grain rice. Int J Res Med Sci 2017;5:4502-5.