EFFECT OF MILLED RICE AMYLOSE CONTENT ON IN-VITRO DIGESTIBILITY AND GLUCOSE RESPONSES IN RATS

RIZK, LAILA F.1, HANAA. A. DOSS1,
A. A. EL-HISSEWY1 AND M. M. WASIF2

1 Food Technology Research Institute, ARC.
2 Rice Technology Training centre. ARC.
3 Nutrition Institute, Cairo.

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Abstract

Two rice varieties Giza 175 (high amylase) and Giza 181 (low amylase) were used in this investigation. The in vitro and in vivo digestibilities of raw, cooked milled rice and cooked parboiled rice of the two varieties were studied.

The hydrolysis rates with α amylase of high amylase rice (179.14-180.03) were lower than of those with low amylase rice (188.73-209.85).

Weight gains of rats differed significantly between groups. High amylase rice caused increase in feed efficiency in diabetic rats.

Rice with a higher amylase content, decreased serum glucose levels from 321 to 213-228 mg/100ml than did low amylase rice (255-269 mg/100ml), the same trend was observed for serum cholesterol.

For cooked high amylase rice, it lowered the serum glucose to 213 against 269 mg/100ml for cooked rice with low amylase, while in cooked parboiled rice, their values were 226 against 255 mg/100ml.

INTRODUCTION

Rice is the major staple food and source of calories for over half the world population. It is nutritious cereal grain whose variation in composition and form can be used to optimize carbohydrate absorption. Starchy endosperm is the single major constituent of rice grains (85-90%), varying ratios of amylase versus amylopectin have (recently) become an issue of nutritional concern.

Yoshizawa et al., (1981) showed that digestibility of starch by amylase at an early stage of digestion was inclined to be higher in the starch containing less amylase. Gee and Johnson (1985) showed that the rate of hydrolysis of starch appears to be the most important factor governing the human glycemic response to food. Miller et al., (1992) determined the glycemic (GI) and insulin index (II) values for 12 rice products using eight healthy subjects. The high amylase rice gave a lower GI and II than did
the normal amyllose and waxy rice varieties. They indicated that many varieties of rice whether white, brown, or parboiled should be classified as high GI food, only high amyllose varieties gave low GI diets.

Oxus and Behall (1993) studied the effect of varying ratios of amyllose versus amylpectin in foods. High amyllose (HA) starches from corn and barley had been fed to chicks and rats and to human subjects in short and long term studies, the goals of these studies were investigating the effects of HA foods, to decrease blood insulin, glucose, cholesterol and triglycerides in normal subjects and those of higher than normal insulin response to a glucose tolerance test.

Eggum et al., (1993) studied the digestible energy and protein content of raw and cooked milled rice and of cooked parboiled high amyllose rice. In 5 rice varieties, differing in amyllose content, the in-vitro and in vivo resistant starch levels were low values and were positively correlated with amyllose content. High resistant starch levels were found in cooked and parboiled cooked rice than in raw rice. Values for undigestible energy were similar in raw rice but increased with processing, particularly in the higher amyllose rice. The resistant starch content of cooked rice was increased by 1% with parboiling. Also, casiraghi et al., (1993) found that percentage of starch digested were lower in parboiled and quick cooking parboiled rice than in polished rice.

Yokovame (1994) found that the rate of glucose absorption was affected by amyllose content, form, milling and processing conditions. Rice varieties higher in amyllose reported to produce relatively low serum glucose responses in human subjects.

Rani and Bhattacharya (1995) investigated microscopy of rice starch granules during cooking and found that low amyllose rice starch showed total granules disintegration after 60min of cooking at 95°C, but high amyllose granules showed only marginal disorganization. Also, low amyllose starch granules are weak, fragile, so swell and easily disintegrated, while high amyllose rice starch was relatively strong rigid and can resist swelling as well as disintegration.

Ong and Blanshard (1995) found that hard cooking rice had a higher amyllose content (amyllose/amylpectin ratio) and longer chain amylpectin than soft cooking rice. Longer chain amylpectin may encourage greater intra and/or inter molecular interaction with other rice grain components, giving firm texture. Also, different amylpectin structures may explain the different textural properties of rice with similar amyllose content.
Crosby (1998) studied the properties of high amylose corn starch, the amylose molecule can form strong intra and inter molecular network and was the predominant gel forming polymer, producing stronger, more thermally stable gels.

The aim of this study was to clarify the response of high amylose rice to plasma glucose and cholesterol.

**MATERIALS AND METHODS**

Raw and cooked milled rice and cooked parboiled milled rice of two varieties (high amylose and low amylose) were used in this study to investigate the effect of varying ratio of amylose versus amylpectin in rice to decrease blood glucose in-vitro and in-vivo studies.

The milled rice varieties Giza 175 (high amylose) and Giza 181 (low amylose) were obtained from the rice Breeding section, field crops research Institute. The same two varieties of white rice after parboiling were obtained from Rice Technology Training Center, Alexandria, Egypt (RTTC).

**Processing:**

Rice were tested in three forms:

1. Raw rice (uncooked white rice)
2. Cooked rice
3. Cooked parboiled rice

**Cooking:**

Each sample was cooked for the predetermined length of time in excess water, then the sample was semidried at room temp., followed by drying in an air oven.

**Chemical analysis**

A portion of each sample was ground using universal type laboratory rice mill. The contents of moisture, protein and carbohydrates were determined as reported in A.O.A.C (1990). The procedure of Juliano (1971) was used for the determination of amylose content in milled rice.
**In-vitro Digestion**

The rice was tested in-vitro (raw, cooked and cooked parboiled rice) to investigate their susceptibility to amylase using the method of Michalinos, (1962), then ferrocyanide number was determined according to the procedure given by Kerr and Severson (1943).

**In vivo study:**

In the present study, the effect of high amylase rice as antidiabetic agent was investigated. Male albino rats Sprague Dawley Strain (forty eight) weighing between 140 and 185g. provided from Nutrition Institute, were housed individually in wire cages with screen bottom. The experimental animals were fed for one week on standard diet.

The standard diet was consisted of Casein 10%, cotton seed oil 10%, cellulose 5%. Salt mixture 4% (Hegsted et al, 1941), vitamin mixture 1% (Campbell 1961), corn starch 69.8% and choline chloride 0.2%. After that period rats were divided into two groups, the first group of 6 rats was fed on standard diet and was considered as control A.

The second group (42 rats) was injected with 150mg/kg body weight of recrystallized alloxan to induce hyperglycemia after fasting over night according to Lazzarow and Palay, (1954).

To ensure occurrence of diabetes in rats, blood sample was withdrawn 48 hour after alloxan injection and glucose (mg/100) was determined according to Asatorr and King (1954).

One group (6 rats) from diabetic rats were fed on standard diet and kept as positive control (B). The other diabetic groups were divided into six groups, (6 rats each) and fed on experimental diet.

The first group was fed with raw rice, the second with cooked rice and the third with cooked parboiled rice all contain high amylase. The three other groups were fed on the rice variety (Giza 181) low amylase.

Each rat was weighed every two days and its food consumption was determined.

Blood samples were obtained from each rat from orbital vinous plexuses each 7th day after feeding on different rice varieties.
The experiment was terminated when the glucose level in blood showed constant
values. At the end of the experimental period, animals were fasted overnight and sacri-
fied under ether anaesthesia. Blood was collected from hepatic portal, serum was sepa-
rated and kept in plastic vial at -20°C until analysis.

Total cholesterol (Abell et al., 1985) and glucose were determined in blood ser-
um of the experimental rats.

RESULTS AND DISCUSSION

The chemical composition of two rice varieties Giza 175 and Giza 181 is report-
ed in (Table 1). The results show that, the average protein content was 9.30 and
7.84%; Lipid 0.526 and 0.436%, ash 0.55 and 0.72%; total carbohydrates 85.35 and
86.96%. The amylose content 28.1 and 19.5 for Giza 175 and Giza 181 resp.

In-vitro study

The in vitro digestibilities of low and high amylose rice varieties are shown in (Ta-
ble 2). The results showed that the hydrolysis rates of high amylose rice variety
(179.14-180.53) were lower than those of low amylose rice variety (169.73-209.85).

This agreed with Yoshizawa et al., (1981) who found that digestibility of starch
by amylase was higher in starch containing low amylose. Also, Bjorek et al., (1990) re-
ported that with increasing amylose content, a decrease occurred in-vitro digestibility.
Digestibility of starch was negatively correlated with amylose content. Table 3, shows
that a significant decrease of average (gain) in body weight for rats injected with allox-
an. Diabetic rats fed on low amylose rice gave non significant increase in body weight
gain (18 Vs 19-24) while a significant increase was found on feeding with high amylose
rice (18 Vs 28-32).

It could be said that, high amylose rice showed glucose lowering effect when
compared with low amylose rice, consequently, weight gains of rats differed significantly
(p<0.05 between groups). This may attributed to the differences observed in blood
glucose responses.

Regarding feed efficiency (Table 3), it is decreased from 0.1997 in healthy rats
to 0.037 in diabetic rats. Feeding rice with low amylose to diabetic rats increased the
feed efficiency (from 0.042 to 0.048) while feeding rice with high amylose increase it
(from 0.0587 to 0.0686). This observation means that high amylose rice caused an in-
crease in feed efficiency in diabetic rats.
The results in (Table 4) reveal that rice with a higher amylase content decreased serum glucose level significantly in diabetic rats from 321 control Vs 213-227mg/100ml than did the low amylase rice variety 321 control Vs 255-269mg/100ml. These results agree with Goddard et al., (1984) who tested rice with different amylase contents (0%, 14-17 and 25%) and found that rice with the highest amylase content elicited the lowest glycemic and insulunemic responses. These authors attributed the observed effects to the formation (during cooking) of amylase complexes based on the constitutive lipids. Amylose lipid complexes were reported to be resistant to α-amylase in-vitro, Eliasson and Krog (1985) as well as in-vivo, Holm et al., (1983).

Also, (Table 4) shows that the cholesterol in alloxanized rats was highly increased comparing with healthy rats, the results revealed that rice with a higher amylase content decreased cholesterol level in diabetic rats (349 Vs 287-303 mg/100ml) than did the low amylase rice variety (349 Vs 306-329 mg/100ml). Similar results have been reported by Amelisvoort and Weststrate (1992) who found that high amylase maize starch showed positive lowering effect on blood glucose and insulin levels, as well as triglycerides and cholesterol in humans. Goddard et al., (1984) observed similar responses from high amylase rice. Behall et al., (1989) concluded that long term intake of high amylase starch may benefit individuals with elevated glucose and insulin levels and apparent insulin resistance.

For cooked rice, it was found that high amylase (28% amylase) lowered the serum glucose to 213 against 269 mg/100ml for cooked low amylase, while in cooked parboiled rice it was 226 against 255mg/100ml.

Also, the same trend was observed for the serum cholesterol. Akerberg et al., (1998) investigated boiled rice made from varieties with different amylase/amylpectin ratios, they found that the high amylase rice (28% amylase) gave significantly lower GI=64 than ordinary GI=83 and GI=87 for (20% amylase) and GI=88 for waxy (0-2% amylase).

The results indicate that, only varieties of rice with a high amylase content (parboiled or not) said to be potentially useful with diabetes, whether where they decreased availability of glucose or lowered the rate of digestion. These varieties provide slow source of carbohydrate for modulation of postprandial glucose and insulin plasma level.

High amylase rice is a relative newcomer to cereal science investigation, applications and may have future industrial applications.
Table 1. Chemical composition of rice varieties.

<table>
<thead>
<tr>
<th>Rice variety</th>
<th>Amylose %</th>
<th>Protein %</th>
<th>Lipid %</th>
<th>Ash %</th>
<th>Total carbohydrates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Giza 181</td>
<td>19.5</td>
<td>7.84</td>
<td>0.436</td>
<td>0.72</td>
<td>86.96</td>
</tr>
<tr>
<td>Giza 175</td>
<td>28.1</td>
<td>9.3</td>
<td>0.526</td>
<td>0.55</td>
<td>85.35</td>
</tr>
</tbody>
</table>

Table 2. Ferricyanide Number for rice varieties before and after hydrolysis with α-Amylase.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Before hydrolysis with α-Amylase</th>
<th>After hydrolysis with α-Amylase</th>
<th>Increase</th>
</tr>
</thead>
<tbody>
<tr>
<td>Giza 181</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Raw Rice</td>
<td>5.52</td>
<td>215.37</td>
<td>209.85</td>
</tr>
<tr>
<td>Cooked Rice</td>
<td>6.58</td>
<td>204.07</td>
<td>197.49</td>
</tr>
<tr>
<td>Cooked parboiled rice</td>
<td>6.01</td>
<td>195.74</td>
<td>189.73</td>
</tr>
<tr>
<td>Giza 175</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Raw Rice</td>
<td>4.93</td>
<td>185.33</td>
<td>180.40</td>
</tr>
<tr>
<td>Cooked Rice</td>
<td>7.64</td>
<td>186.78</td>
<td>179.14</td>
</tr>
<tr>
<td>Cooked parboiled rice</td>
<td>7.61</td>
<td>186.14</td>
<td>180.53</td>
</tr>
</tbody>
</table>

Giza 175 = high amylose.
Giza 181 = low amylose.
Table 3. Food efficiency and gain in body weight for alloxanized rats given (low and high amylase rice varieties).

<table>
<thead>
<tr>
<th>Groups</th>
<th>Gain in body wt (g)</th>
<th>Food intake (g)</th>
<th>Food efficiency</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control A (negative)</td>
<td>91.8±7.9</td>
<td>464.00±38.9</td>
<td>0.1997±0.031</td>
</tr>
<tr>
<td>Control B (positive)</td>
<td>18.0±7.7</td>
<td>478.17±31.4</td>
<td>0.0370±0.016</td>
</tr>
<tr>
<td>Raw low amylase rice</td>
<td>19.8±6.6</td>
<td>460.20±33.7</td>
<td>0.0420±0.012</td>
</tr>
<tr>
<td>Cooked low amylase rice.</td>
<td>20.5±7.5</td>
<td>477.80±35.1</td>
<td>0.0425±0.015</td>
</tr>
<tr>
<td>Cooked parboiled low amylase rice.</td>
<td>24.5±11.8</td>
<td>495.70±41.0</td>
<td>0.0480±0.020</td>
</tr>
<tr>
<td>Raw high amylase rice</td>
<td>29.8±13.3</td>
<td>455.30±26.5</td>
<td>0.0660±0.073</td>
</tr>
<tr>
<td>Cooked high amylase rice.</td>
<td>32.0±9.0</td>
<td>472.60±43.0</td>
<td>0.0666±0.012</td>
</tr>
<tr>
<td>Cooked parboiled high amylase rice.</td>
<td>28.5±8.3</td>
<td>477.20±44.9</td>
<td>0.0587±0.013</td>
</tr>
</tbody>
</table>

Table 4. Means of serum glucose and cholesterol for alloxanized rats fed low and high amylase varieties.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Serum Glucose mg/100ml</th>
<th>Serum cholesterol mg/100ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control A (negative)</td>
<td>99.0±5.1</td>
<td>173.3±22.7</td>
</tr>
<tr>
<td>Control B (positive)</td>
<td>321.7±34.5</td>
<td>349.7±35.6</td>
</tr>
<tr>
<td>Raw low amylase rice</td>
<td>264.0±37.4</td>
<td>329.5±34.3</td>
</tr>
<tr>
<td>Cooked low amylase rice.</td>
<td>269.2±26.9</td>
<td>316.5±34.4</td>
</tr>
<tr>
<td>Cooked parboiled low amylase rice.</td>
<td>255.0±22.8</td>
<td>308.5±23.0</td>
</tr>
<tr>
<td>Raw high amylase rice</td>
<td>227.0±20.9</td>
<td>303.0±29.0</td>
</tr>
<tr>
<td>Cooked high amylase rice.</td>
<td>213.3±22.7</td>
<td>287.7±14.0</td>
</tr>
<tr>
<td>Cooked parboiled high amylase rice.</td>
<td>226.7±19.7</td>
<td>298.0±20.8</td>
</tr>
</tbody>
</table>
REFERENCES


تأثير محتوى الأميلوز في الأرز الأبيض على مستوى الهضم معمولاً وعلي مستوي الجلوكوكون في دم الفئران

علي فهمي رزق 1, هناء مرزوق دوس 2, أحمد المصعيدي 3, ماهر مرجان 2

1 معهد بحوث تكنولوجيا الغذاء
2 مركز تدريب تكنولوجيا الأرز بالأسكندرية
3 معهد التغذية

استخدم في هذا البحث من الفصيلة الأرز الأبيض، جزيرة 181 (عالي الأميلوز) جزيرة 181 (منخفض الأميلوز). تم تقدير نسبة الهضم بواسطة أ-أميلوز وأيضاً تم تقدير نسبة الجلوكوكون في الدم للفئران المحتجزة لكل من الصنفين (أرز غير مطهي - أرز مطهي - أرز بعد القلي).

وكانت معدل الهضم بصورة أ-أميلوز للصنف جزيرة 175 (عالي الأميلوز) يتراوح بين 1.4-1.79. في حين أن الفصيلة منخفضته فصيلة أ-أميلوز يتراوح بين 1.59-1.78. في تجربة التغذية وجد فروق معنوية بين مجموعات الفصيلة في النسبة في الوزن بالنسبة للأرز المرتفع في نسبة الأميلوز وأيضاً زيادة في الكفاءة الغذائية للبروتين المضادة بعرض السكر.

أيضاً الأرز المرتفع في مستوي الأميلوز أي إلى انخفاض مستوي الجلوكوكون في الدم من 221 والأرز مطهي إلى انخفاض جلوكوكون في الدم من 221 إلى 213 مجم/0.01 مل. وصناعات الأشبة بالفروق في نسبة الأميلوز من 0.49 إلى 0.80 مجم/0.01 مل.

بالنسبة للأوز الأبيض العالي في نسبة الأميلوز أي إلى انخفاض نسبة السكر إلى 0.01 مجم/0.01 مل بالمقارنة بالأوز طبيعي المنخفض في نسبة الأميلوز 38 مجم/0.01 مل. بينما في الأوز الفصيلة المنخفض في نسبة الأميلوز أي إلى انخفاض 38 بالمقارنة المنخفض في نسبة الأميلوز 38 مجم/0.01 مل وذلك لاندماج بالنسبة الكوليسترول.