Physical, chemical and sensory characteristics of cookies made of pumpkin seed flour (PSF) and baru seed flour (BSF) for celiac disease

Abstract

The objective was to use pumpkin seed flour (PSF) and baru seed flour (BSF) in the manufacture of cookies for celiac disease, evaluating their physical, chemical, physicochemical and sensory characteristics. Three types of cookies were prepared by standard formulation modification (P), with the starch flour partial substitution by PSF (A) and BSF (B) by 10%. The biscuits physical analysis was determined by the 10-50D method of the American Association of Cereal Chemists procedures. The chemical and physicochemical characteristics followed methodologies proposed by Adolfo Lutz Institute. The sensory evaluation was performed by two tests: the acceptability one (affective test) with a 9 points hedonic scale, and the multiple comparisons one (discriminatory test), in a randomized balanced complete block design. Data were subjected to variance analysis by Tukey’s and Dunnett’s test at 5% significance level. It was found that cookie B differed (p <0.05) in the control formula, for some physical characteristics such as weight before and after cooking, thickness, length and width after cooking. Cookies differed (p <0.05) from the control formulation in terms of yield, the volume expansion ratio and also showed higher value for moisture, ash, fat and fiber. Cookie B had good acceptance in all attributes. In the discriminatory test, cookie B got better grades. It is concluded that PSF and BSF partial addition improves the cookies nutritional value, increases the dietary fiber, protein, minerals and lipids however, sensorially the BSF showed better results, demonstrating the possibility of using such flour cookies formulations for celiac at domestic and industrial level.

Key words: Cookies. Pumpkin. Baru. Nutritional Value.
Introduction

Celiac disease (CD) is a permanent intolerance to gluten, a protein contained in some cereals such as wheat, rye, barley and oats. The mortality rate of this disease worldwide is two times higher than the mortality from other causes, with an increase that takes place predominantly during the first year, after the disease diagnosis. Death occurs mainly due to the presence of intestinal malignancies such as lymphoma.

Patients with celiac disease show symptoms such as diarrhea, anorexia, malnutrition, abdominal bloating, and weight loss. The disease may be associated to many others, such as Dermatitis Herpetiformis, osteoporosis, epilepsy and diabetes mellitus type 1. The diagnosis is based on clinical characteristics, serological tests for specific antibodies and intestinal biopsy. The celiac patients treatment is gluten exclusion from their diet for life, correcting the different malnutrition, anorexia, dehydration degrees, food intolerances, vitamin and mineral needs.

To access to products made with wheat flour substitutes that have nutritional value and pleasant sensory characteristics are difficulties encountered by coeliacs and by the food industry. Cookies are among the most consumed products by the society in general, which is justified by the consuming facility and the affordable cost. Despite the significant biscuits production in Brazil, the supply of such products gluten-free is very limited.

There are numerous unconventional foods that are generally not used for human consumption, but they have lots of nutrients. The minimally processed food waste as well as fruits and vegetables waste, used in the food industry, are generally despised and could be used to enrich foods. An example is the pumpkin seed that is despised, however studies have reported many nutrients found in them, such as minerals, proteins, lipids, fiber and antioxidants, allowing its use in the food fortification and increasing the products nutritional value.

Another food that could be used to enrich products is baru almonds, due to its functional properties, such as phytosterols, antioxidants and dietary fibers. They are good protein, lipids sources, being mostly monounsaturated and polyunsaturated fatty acids, which are related to a reduced risk of cardiovascular diseases. They also have considerable content of several minerals, specifically iron, calcium, zinc and selenium, the first two being important in the prevention of nutritional deficiencies of Public Health.
relevance, and zinc and selenium, for the enzymatic and regulatory functions, as part of the body antioxidant defense system. Another positive factor is the pleasant taste, being similar to peanuts which increases the products acceptability.\textsuperscript{6,7}

Studies have shown that both pumpkin and baru seeds have anti-nutritional factors that affect the nutrients bioavailability. However, these factors could have their action inhibited through an appropriate heat treatment such as roasting, in the form of flour, which further contributes to improving the taste and texture.\textsuperscript{5,7}

From the available evidence regarding the pumpkin and baru seeds nutritional benefits, cookies for celiac partially replacing the sweet flour by pumpkin and baru seeds were elaborated, aiming to add nutritional value to the product, evaluating as well the adding flour effects into the physical, chemical, physicochemical and sensory biscuits properties.

**Material and Methods**

**Material**

The baru and pumpkin flours were obtained by roasting the seeds, as described in flowcharts 1 and 2. At the end of the process, flours were obtained, the baru one being thinner, and both with a light color and odor similar to peanuts. The baru seed was provided by Belverde manufacturer, based in Diamantina Garden, Goiânia, Goiás state.
Figure 1. Flowchart of obtaining baru seed flour.
Figure 2. Flowchart of obtaining pumpkin seed flour.

The formulation of cookies type of biscuits

Three types of cookies by modification of standard formulation were confected, with partial substitution of the starch sweet by the pumpkin seed flour (PSF) and baru seed flour (BSF) in 10%. The cookies produced were named A and B respectively. A basic formulation was prepared without the flour addition, called pattern (P). Chart 1 shows the ingredients proportion used to prepare the formulations. The seeds used in the cookies formulation were obtained commercially in Rio de Janeiro (pumpkin seeds) and Goiânia (baru seeds). They were roasted for 7-10 minutes and three minutes, respectively, to obtain the golden color. Afterwards the seeds were crushed in a blender and sieved for further use in the formulations described in Chart 1. The dry ingredients were mixed and margarine
gradually added until obtaining a homogeneous mass. Then, the cookies were molded with the aid of shapes and baked at a moderate temperature of 150-180°C oven for 15 minutes. The cookies were cooled to room temperature for the physical, physicochemical and chemical analysis.


<table>
<thead>
<tr>
<th>Ingredients (g%)</th>
<th>Control</th>
<th>A</th>
<th>B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brown sugar</td>
<td>11</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>Sweet starch</td>
<td>30</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>Arrowroot</td>
<td>26</td>
<td>26</td>
<td>26</td>
</tr>
<tr>
<td>Margarine</td>
<td>27</td>
<td>27</td>
<td>27</td>
</tr>
<tr>
<td>Baking powder</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Cocoa powder</td>
<td>5</td>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td>PSF</td>
<td>-</td>
<td>10</td>
<td>-</td>
</tr>
<tr>
<td>BSF</td>
<td>-</td>
<td>-</td>
<td>10</td>
</tr>
</tbody>
</table>

A - cookie with PSF; B - cookie with BSF.

**Physical characterization of the cookies**

The physical analyzes of the cookies (P) control and experimental A and B comprised procedures described in 10-50D macro method of the *American Association of Cereal Chemists* (AACC),\(^8\) to determine weight, thickness, length and width before and after cooking. The cookies were weighed in digital balance, ICEL brand. Analyzes were conducted with ten cookies from the same batch, randomly sampled, after they were cooled to room temperature. The cookies thickness, length and width were determined by a millimeter scale ruler. The specific volume was determined by the displacement method of millet seeds, followed by three repetitions for each formulation. The specific volume was calculated according to equation 1 below:

\[
\text{Specific volume (cm}^3/\text{g}) = \frac{\text{Cookie volume (cm}^3)}{\text{cookie weight (g)}}
\]

\[
\text{Cookie volume} = \text{Volume without cookie (cm}^3) - \text{Volume with cookie}
\]
From the weight of pre and post-cooking cookies the product yield was also calculated (after cooking), as per equation 2:

\[(\text{Weight after cooking/Weight before cooking}) \times 100\]

and the thermal factor according to equation 3:

\[\text{Weight after cooking/Weight before cooking}\]

The expansion factor was determined by the ratio between the cookies diameter and thickness values as per the AACC 10-50D method.

**Cookies chemical and physicochemical analysis**

The chemical and physicochemical characteristics were determined by the following procedures: moisture at 105 °C until constant weight, ash by incineration at 550 °C; lipids by the solvent extraction method (Soxleht method); nitrogen determined by the Kjeldahl method and converted to crude protein by a 6.25 factor, according to AOAC, insoluble fiber by the enzymatic-gravimetric method, carbohydrate (NIFEXT difference), titratable acidity and pH electrometric determination, as per the methodologies proposed by Adolfo Lutz Institute. The moisture, titratable acidity and pH determination analysis were performed in triplicate, and the remaining ones, in duplicate.

**Cookies sensory analysis**

This research has its project evaluated and approved by the Research Ethics Committee of the Federal University of Rio de Janeiro (numbers 1014/07 and 160). The cookies were submitted to sensory evaluation through two tests: an acceptability one (affective test) and a multiple comparison one (discriminatory test), applied in a randomized complete balanced block design. The tasters profile was also carried out (Figure 3).

The team consisted of 62 untrained, randomly selected tasters, belonging to a UFRJ team with their consent. The first test performed was the affective one, where evaluators reported how they liked or disliked each formulation preparation for color, aroma, texture and flavor attributes using a nine points hedonic scale, ranging from “like very much” (maximum score) to “dislike very much” (minimum score). The second test performed was the discriminating one, in which evaluators compared the formulations A and B with the
overall appearance control formulation, giving a scale ranging from 9 (extremely better than the standard) to 1 (extremely worse than the standard).10

**Consumer profile**

Name:

Sex: ( ) M ( ) F

Age: ( ) < 18 years ( ) 18 to 25 years ( ) 26 to 40 years ( ) above 40

Educational level: ( ) Basic ( ) Medium ( ) Superior ( ) Graduate

Consumption frequency:

<table>
<thead>
<tr>
<th>Foods</th>
<th>Never</th>
<th>Daily</th>
<th>Weekly</th>
<th>Monthly</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soda</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cake</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cookie</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fruits</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Acceptability Test**

Please, taste and evaluate, from left to right, how much you liked or disliked the sample, using the scale below:

9- Liked immensely
8- Liked very much
7- Liked moderately
6- Liked slightly
5- Didn’t like/nor dislike
6- Dislike slightly
3- Dislike moderately
2- Dislike very much
1- Dislike immensely

Sample: Sample:

Color: Color:

Aroma: Aroma:

Texture: Texture:

Flavor: Flavor:

**Multiple Comparison test**

Name: Date:

You are receiving this standard sample (P) and 2 codified samples. Compare each sample, from left to right, with the pattern and identify if it is better, equal or worse that the standard, giving the following notes:

9- Extremely better than standard
8- Much better than standard
7- Moderately better than standard
6- Slightly better than standard
5- Equal to standard
4- Slightly worse than standard
3- Moderately worse than standard
2- Much worse than standard
1- Extremely worse than standard

Sample: Sample:

**Figure 3.** Customer identification sheet and its affective and discriminative tests.
Statistical Analysis

The physical, chemical, physicochemical characterization and sensory tests results were analyzed using the variance analysis and the average Tukey’s and Dunnett tests, specific to each sensory analysis at 5% significance level, with the Statistica program use.

Results and Discussion

Cookies physical characterization

In chart 2 are the cookies physical analysis results of the different formulations produced with partial substitution of sweet starch by the PSF or BSF.


<table>
<thead>
<tr>
<th>Determinations</th>
<th>Control</th>
<th>A</th>
<th>B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Weight before cooking (g)</td>
<td>5,91a</td>
<td>6,07a</td>
<td>7,02b</td>
</tr>
<tr>
<td>After cooking</td>
<td>5,4a</td>
<td>5,75a</td>
<td>6,74b</td>
</tr>
<tr>
<td>Height before cooking (cm)</td>
<td>0,63a</td>
<td>0,66a</td>
<td>0,73a</td>
</tr>
<tr>
<td>After cooking</td>
<td>0,69a</td>
<td>0,79ab</td>
<td>0,88b</td>
</tr>
<tr>
<td>Length before cooking (cm)</td>
<td>2,71ab</td>
<td>2,68a</td>
<td>2,76b</td>
</tr>
<tr>
<td>After cooking</td>
<td>2,66a</td>
<td>2,69a</td>
<td>2,83b</td>
</tr>
<tr>
<td>Width before cooking (cm)</td>
<td>2,71ab</td>
<td>2,67a</td>
<td>2,75b</td>
</tr>
<tr>
<td>After cooking</td>
<td>2,65a</td>
<td>2,69a</td>
<td>2,79b</td>
</tr>
<tr>
<td>Thermal factor</td>
<td>0,91a</td>
<td>0,95b</td>
<td>0,96b</td>
</tr>
<tr>
<td>Yield (%)</td>
<td>91,4a</td>
<td>94,7b</td>
<td>96,3b</td>
</tr>
<tr>
<td>Volume (mL/g)</td>
<td>1,69a</td>
<td>4,90b</td>
<td>3,77b</td>
</tr>
<tr>
<td>Expansion factor</td>
<td>106,9a</td>
<td>124b</td>
<td>127,6b</td>
</tr>
</tbody>
</table>

Averages followed horizontally by the same letter do not differ. p>0.05
A - cookie with PSF; B - cookie with BSF.
The B cookie differed (p <0.05) in the control formula for the weight measurements before and after cooking as well as thickness, length and width after cooking, while the A cookie did not differ in these aspects.

The highest weight variation observed in B cookie may indicate a higher water holding capacity by the cookie dough components after adding BSF. It was also noted that after cooking, the BSF cookie showed increased thickness, width and length, indicating that its addition may have interfered with the cookie physical characteristics, making the dough to suffer a greater spacing during cooking, consequently increasing its yield.

The expansion factor was greater (p <0.05) for cookies A and B relative to cookie P control (Chart 2). The expansion factor is related to the ingredients ability to absorb water and is used as a quality indicator. The cookies with high fiber content usually have decreased expansion factor, which did not occur in this work. The same result was found in Moura & Spier study.4

Importantly, A and B cookies did not differ (p> 0.05) regarding yield and volume, but differed (p <0.05) in the control formulation, indicating that when partially replacing the sweet starch by these flours, better results are obtained than the related ones to yield, which is considered a positive aspect for its use in the food industry.

Cookies chemical and physicochemical characterization

The cookies chemical composition is shown in Chart 3.

**Chart 3.** Average of chemical and physicochemical characteristics of cookies preparation. Rio de Janeiro-RJ, 2014.

<table>
<thead>
<tr>
<th>Determinations (g%)</th>
<th>Cookies</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>A</td>
</tr>
<tr>
<td>Humidity</td>
<td>3,82a</td>
<td>4,86b</td>
</tr>
<tr>
<td>Ashes</td>
<td>1,51a</td>
<td>1,93c</td>
</tr>
<tr>
<td>Lipids</td>
<td>23,79a</td>
<td>27,30b</td>
</tr>
<tr>
<td>Proteins</td>
<td>1,3a</td>
<td>3,92b</td>
</tr>
<tr>
<td>Carbohydrates</td>
<td>67,89a</td>
<td>57,09b</td>
</tr>
</tbody>
</table>
The cookies moisture content was less than 6.50%, being consistent with the standard set by CNNPA,\textsuperscript{11} which must be less than 14%. Cookies A and B showed higher moisture content, ashes, fat and fiber, compared to the cookie control. The A and B cookies larger lipid content is related to the large amount of \(\omega-6\) polyunsaturated fatty acid (linoleic acid) present in pumpkin and baru seeds, besides \(\omega-9\) monounsaturated fatty acid (oleic), which also comprises lipid fractions of baru seed.

This mono and polyunsaturated fatty acids composition is important for health, since these acids contribute to the low-density lipoprotein fraction reduction (LDL) and very low density (VLDL), responsible for the serum cholesterol increase.\textsuperscript{4,6} The amount of ash was higher in cookies made with pumpkin seed, due to the high content of minerals present in it.\textsuperscript{4} There was a significant increase in the fiber content in cookies A and B compared to the control one, which is directly related to the fiber high content of these flours.

As per Freitas & Naves,\textsuperscript{6} the baru seed is a good source of dietary fiber, predominantly insoluble fibers. Considerable insoluble fibers amounts contribute to the stool increase and prevent intestinal problems, further enhancing these foods in promoting health.

Several studies show that pumpkin seed also has high dietary fiber content, as well as a laxative effect, provided by the insoluble fibers present in it.\textsuperscript{12} Both A and B products are considered dietary fiber sources according to ANVISA.\textsuperscript{13}

As for physical and chemical analysis, in Chart 3, all prepared cookies were very similar in acidity, with no significant difference between them. The pH values found showed that cookie A had higher (p <0.05) value, when compared to control and there was no difference between cookie A and B for such determination.

The pH and titratable acidity provide information on the flour quality, for the lower the pH and the higher the titratable acidity value, the greater the conversion of long chain fatty acids into short-chain organic acids, which give unpleasant taste and odor to products.\textsuperscript{14} Therefore, the pumpkin and the baru seed flours are suitable for the cookies preparation.

### Chart 3

<table>
<thead>
<tr>
<th>Determinations (g%)</th>
<th>Control</th>
<th>A</th>
<th>B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fibers</td>
<td>1.69\textsuperscript{a}</td>
<td>4.90\textsuperscript{b}</td>
<td>3.77\textsuperscript{b}</td>
</tr>
<tr>
<td>Titratable Acidity Titratable (mL.g\textsuperscript{-1})</td>
<td>2.79\textsuperscript{a}</td>
<td>3.22\textsuperscript{a}</td>
<td>2.42\textsuperscript{a}</td>
</tr>
<tr>
<td>pH</td>
<td>6.35\textsuperscript{a}</td>
<td>6.87\textsuperscript{b}</td>
<td>6.77\textsuperscript{ab}</td>
</tr>
</tbody>
</table>

Averages followed by the same letter horizontally do not differ. p>0.05

A- cookie with PSF; B- cookie with BSF.
Sensory Analysis

Tasters profile

Figure 4 states that 74.19% of the evaluators are female and 25.80% male, predominantly adult, aged between 18-25 years. The dominant level of education was higher education. Regarding the frequency of food consumption the attributes “never”, “daily”, “weekly” and “monthly” were considered. In this group, the weekly consumption frequency prevailed for both the cookie and the other industrial products surveyed (soda and cake).

Therefore the results indicate that these products are often present in the tasters’ diet. However, for the fruits consumption the frequency of daily consumption prevailed.
Cookies sensory tests

The tasters’ distribution per hedonic values for the experimental products is shown in Figure 5.

Figure 5. Tasters’ distribution hedonic values for evaluated products.

A = pumpkin seed flour and B = baru seed flour
The tasters’ scores average in the affective test hedonic scale to cookie formulations regarding sensorial preference of texture, aroma, flavor and color attributes are shown in Chart 4. The B cookie had the highest average for the flavor attribute, and regarding the other attributes A and B cookies did not differ. This can also be seen in Figure 5 that shows the tasters preference for the four evaluated attributes. In general, one can say that more than 62% of the tasters assigned grades higher than 5 (didn’t like/or dislike) for all attributes for cookie B. However, this did not occur with cookie A, where only 48% of the tasters attributed grades above 5 for the “flavor” attribute, indicating that this attribute was one that most differentiated the cookies, revealing that cookie B had better flavor than cookie A.

**Chart 4.** Affective test averages - hedonic scale for evaluated cookies. Rio de Janeiro-RJ, 2014.

<table>
<thead>
<tr>
<th>Attributes</th>
<th>A</th>
<th>B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Texture</td>
<td>6,18&lt;sup&gt;a&lt;/sup&gt;</td>
<td>6,64&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Flavor</td>
<td>5,14&lt;sup&gt;a&lt;/sup&gt;</td>
<td>6,53&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Color</td>
<td>6,93&lt;sup&gt;a&lt;/sup&gt;</td>
<td>6,94&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Aroma</td>
<td>6,37&lt;sup&gt;a&lt;/sup&gt;</td>
<td>6,55&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Averages followed by the same letter horizontally do not differ. p>0.05
A= pumpkin seed flour; B= baru seed flour.

Chart 5 presents the scores average values given in the discriminatory test for the overall appearance of cookies A and B compared to standard, where cookie B had the highest average.


<table>
<thead>
<tr>
<th>Attributes</th>
<th>A</th>
<th>B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aspect Global</td>
<td>5,23&lt;sup&gt;a&lt;/sup&gt;</td>
<td>6,63&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Averages followed by the same letter horizontally do not differ. p>0.05
A= pumpkin seed flour; B= baru seed flour.
The acceptability rate of cookies A and B are shown in Chart 6. The B cookie showed good acceptance in all evaluated attributes. However cookie A didn’t reach the minimum acceptance ratio of 70% for texture and flavor attributes.

**Chart 6.** Acceptability rates (%) for evaluated cookies. Rio de Janeiro-RJ, 2014.

<table>
<thead>
<tr>
<th>Attributes</th>
<th>A</th>
<th>B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Texture</td>
<td>68.63a</td>
<td>73.77a</td>
</tr>
<tr>
<td>Flavor</td>
<td>57.16a</td>
<td>72.55b</td>
</tr>
<tr>
<td>Color</td>
<td>77.06a</td>
<td>77.11a</td>
</tr>
<tr>
<td>Aroma</td>
<td>70.78a</td>
<td>72.77a</td>
</tr>
</tbody>
</table>

Averages followed by the same letter horizontally do not differ. p>0.05
A= pumpkin seed flour; B= baru seed flour.

Overall, the results showed that tasters found cookie B better than cookie A.

**Conclusion**

We conclude that the addition of PSF and BSF, partially substituting the sweet starch, improve the cookies nutritional value for celiac for they increase the dietary fiber content, as well as proteins, minerals and lipids but, sensory, cookie B containing BSF showed better results, showing that this flour can partially substitute the traditional one in cookies formulations for celiac at a domestic level as for the industrial one.

**References**


