Shelf-life evaluation of cheese bread added of residue from *Agaricus blazei* Murrill extration

Avaliação da vida de prateleira de pão de queijo adicionado de resíduo do extrato do cogumelo *Agaricus blazei* Murrill

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Abstract

The aim of this work was to determine the composition of the A. blazei residue obtained after β-glucans aqueous extraction and the shelf-life of cheese bread elaborated with this residue. The residue showed a high content of crude protein (308,7 g kg-1), fiber (147,9 g kg-1) and phosphorus (4,1 g kg-1) and low fat content (48,3 g kg-1) on a dry weight basis. The results of sensory analysis showed that there was no significant difference ($p \leq 0.05$) among the samples regarding to texture and flavor attributes. Concerning appearance attribute, only the sample with 14 days of storage was significantly different ($p \leq 0.05$) from the sample with 21 days of storage. The appearance ordination total for the sample with 14 days of storage was bigger than sample with 21 days, which indicates that sample with 14 days was less preferred regarding this attribute. Agaricus blazei Murrill residue is an excellent source of nutrients. Its addition to the cheese bread formulation was favorable because it did not produce significant alterations in the visual aspect product. For all evaluated attributes, the sample with the largest time of storage

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had good acceptance, which shows that it is possible to storage the product for approximately 30 days without great flavor, texture and appearance negative changes.

Key words: *Agaricus blazei;* sensory acceptance; physicochemical analyses; mushrooms.

Resumo

O objetivo deste trabalho foi determinar a composição química do resíduo de *A. blazei* obtido após extração aquosa das β-glucanas e analisar a vida de prateleira de um pão de queijo elaborado com esse subproduto. O resíduo apresentou um alto conteúdo de proteína bruta (308,7 g kg^{-1}), fibras (147,9 g kg^{-1}) e fósforo (4,1 g kg^{-1}) e baixo conteúdo de lipídios (48,3 g kg^{-1}) na base seca. O resultado da análise sensorial mostrou que não há diferença significativa ($p \leq 0,05$) entre as amostras em relação aos atributos textura e sabor. Considerando o atributo aparência, apenas a amostra com 14 dias de armazenamento foi significativamente diferente ($p \leq 0,05$) da amostra com 21 dias de armazenamento. O total de ordenação para a aparência da amostra com 14 dias foi maior que o da amostra com 21 dias de armazenamento, o que indica que a amostra com 14 dias foi menos preferida em relação a esse atributo. O resíduo de *Agaricus blazei* Murrill é uma excelente fonte de nutrientes. A adição do resíduo do cogumelo na formulação do pão de queijo não provocou alterações significativas no aspecto visual do produto, favorecendo a utilização desse subproduto. Para todos os atributos avaliados a amostra com o maior tempo de armazenamento teve boa aceitação sensorial, o que mostra ser possível armazenar o produto por aproximadamente 30 dias sem grandes mudanças negativas no sabor, na textura e na aparência.

Palavras-chave: *Agaricus blazei*; aceitação sensorial; análises físico-químicas; cogumelos.

Introduction

The *Agaricus blazei* Murrill mushroom is a species native to Brazil, commonly named “mushroom of the sun” in Brazil or “Himematsutake” in Japan. It was discovered in the Piedade city, São Paulo State, Brazil, and sent to Japan with the aim of to study its medicinal properties (DIAS et al., 2004).

According to Vilela (2007), the mushrooms production in Brazil does not have official statistics, but it is known that the biggest producer zone is placed in Alto Tietê, São Paulo. In Brazil, edible mushrooms consumption have been growing significantly due to the recognition of its high nutritional value and due to its increasing on supply, which makes the product more popular and accessible (EIRA and MINHONI, 1997).

The greatest obstacles found on mushrooms commercialization are related to the popular beliefs about its toxicity, price, alimentary habit and low productivity cultivation. One of the main factors that have been contributed to the increasing on mushroom consumption in Brazil is a bigger demand for "natural" foods without agricultural defenses, with a smaller fat content and higher protein content, beyond preventive and/or curative therapeutic properties (SHIBATA and DEMIATE, 2003).

The mushroom nutritive value is superior to several vegetables. It has high protein content, reaching 1,5 to 6% on wet weigh basis, according to differences between species (BRAGA et al., 1998).

Mushrooms are source of fibers and vitamins B complex. Besides, on wet weigh basis, they have high moisture content (90%) and low fat (0,5%) and calorie (35 kcal in 100 g) content. It is used to make tea, juice or extracts, taking advantage of some present substances like b-glucans. These substances are recognized for improve the immune system, acting like a supporting on alternative treatments against cancer (MANTOVANI, 2005; CHEN et al., 2004; MATSUI et al., 2003; DELMANTO et al., 2001; EGUCHI et al., 1999; MIZUNO et al., 1990). After the active substances extraction the mushrooms remain whole and the nutritional value is kept, although of being usually discarded due to unpleasant flavor.

Generally, scientific works found in this area only deal with the centesimal composition of different kinds of mushrooms without to show a product easy application. The residue obtained after the extraction of b-glucans comes like an opportunity to use it in new products formulation or products that already exist at the market. Therefore, the investigations about the mushrooms nutritional value are not the only important issue, being also necessary searching in the field of the consumer acceptance of new mushroom-based products.

It is difficult to determinate how much the food sensorial properties influence on the consumer choice. First of all, the external appearance is probably the quality attribute that determines the product commercial value (VASQUES et al., 2006). In the case of cheese bread, the color is the first evaluated quality attribute.

Food products quality changes by the storage time due to several biochemical and microbiological transformations. The shelf-life of these products depends of the package protection against the humidity changes on the storage place besides other factors (VASQUES et al., 2006).

Thus, the aims of this work were to perform physicochemical analyses of the *A. blazei* residue, to test a technological application of the residue by adding it in a cheese bread formulation and to evaluate the shelf-life of this product.

Material and Methods

Residue preparation

The Agaricus blazei humid residue is obtained after b-glucans aqueous extraction of fresh mushroom. The samples of residue were supplied by a small industry from Maringá city – PR, Brazil.

The dehydration of A. blazei residue was carried out in an oven drying at 50 ± 5 ºC until it reached constant mass. Subsequently, it was ground and divided into two parts. One of these parts was sent to physicochemical analyses and the other one was used to make cheese bread. All samples were stored in hermetic flasks under refrigeration at 8±2 ˚C until its utilization.

Physicochemical analyses

The physicochemical analyses of the residue were carried out on the Water and Food Laboratory of the Chemical Department – State University of Maringá/PR, Brazil. Moisture, crude protein, total fat content, crude fiber, potassium, calcium and phosphorus were determined according to the methods of AOAC (1995). The conversion factor of 4,38 was used to calculate the protein content (BREENE, 1990; FURLANI and GODOY, 2007; TSAI et al., 2008).

Cheese bread formulation

In the cheese bread formulation were used eggs, potatoes, grated parmesan cheese, soybean oil, integral milk, salt and acid cassava flour acquired at the local market.

In the preparation of the cheese bread the potatoes were cooked and mixed with eggs, grated parmesan cheese, soybean oil, milk and salt in an industrial blender for two minutes to become the dough homogeneous. Thereafter, acid cassava flower and 4 g of *A. blazei* dehydrated residue were added. The dough was separated into small quantities and shaped after ingredients mixture. Although care in keeping lots uniformity, the samples showed a small size variation.

The cheese bread samples were produced in the Laboratory of Transformation Technology and Agricultural Products Conservation of the Agronomy Department – Universidade Estadual de Maringa, Parana, Brazil.

Shelf-life

For the bread cheese shelf-life analysis 5 samples were prepared: the fresh product with no storage time and samples stored in intervals of 7, 14, 21 and 29 days at 18°C in sterilized and hermetic plastic packages.

The sensory evaluation was performed to compare flavor, texture and appearance attributes of the samples without storage and with 7, 14, 21 and 29 days of storage. For the sensory analysis the bread cheese samples were baked at 200 ± 5 ºC for approximately 40 minutes.

It was accomplished an Ordination Test with 20 trained judges familiar with mushroom-based products. They had already been trained in sensory evaluation of mushroom-based products at the Sensory Analysis Laboratory of State University of Maringá. Each judge received 5 cheese bread samples randomically distributed and codified with 3-digit numbers. Concerning the preference, the volunteers were asked to ordinate the samples by using flavor, texture and appearance attributes.

Results were evaluated by Friedman Test with confidence of 95%. It was used the Newel's and MacFarlane table, which indicates the critical difference between ordination totals, according to the number of tested treatments and the number of obtained judgments (DUTCOSKY, 1996).

Results and Discussion

The results of the physicochemical analyses of *A. blazei* residue, on a dry weigh basis, are shown in table 1.

Tsai et al. (2008) reported that *A. blazei* has a protein content of 267,4 g kg^{-1} (dry basis) and Menezes et al. (2008) reported proteins levels from 264,9 g kg^{-1} to 375,8 g kg^{-1} (dry basis) in six different strains of dehydrated powdered *A. blazei* fruiting bodies. The crude protein level of the

Table 1. Physicochemical characterization of dehydrated A. blazei residue obtained after b-glucans aqueous extraction

Analysis	A. blazei residue (g kg^{-1})
Moisture (105 ºC)	34,70 ± 0,13
Crude Protein (N x 4,38)	243,60 ± 0,50
Total Fat	50,0 ± 0,20
Crude Fiber	153,2 ± 1,30
Potassium (K)	4,518 ± 0,36
Calcium (Ca)	0,415 ± 0,15
Phosphorus (P)	4,237 ± 0,92

A. blazei residue found in this study (243,6 g kg^{-1}) was smaller than that aforementioned, which might be due to the difference in the strains used. However, according to Crisan and Sands (1978) mushrooms are a good source of protein, and their protein contents range from 190 g kg^{-1} to 350 g kg^{-1} on a dry weight basis. Besides, previous studies found protein levels ranging from 134,0 g kg^{-1} to 350,0 g kg^{-1} in different kinds of mushrooms (FURLANI and GODOY, 2007; MANZI et al., 2001; YANG et al., 2001; LONGVAH and DEOSTHALE, 1998; CHANG and MILLES, 1989).

Regarding the crude fiber content, the result was smaller (153,2 g kg^{-1}) than that reported for Tsai et al. (2008) (183,1 g kg^{-1}), but was within the range from 30 g kg^{-1} to 320 g kg^{-1} usually found in mushrooms (CRISAN and SANDS, 1978). Furlani and Godoy (2007) found 419,2 g kg^{-1} of crude fiber in shiitake and another authors reported fiber content of champignon within the range from 80,0 g kg^{-1} to 182,0 g kg^{-1} (CHEUNG, 1996; CHANG and MILLES, 1989).

The total fat content of the *A. blazei* residue was low (50 g kg^{-1}), but was higher than those found by Tsai et al. (2008) (26,2 g kg^{-1}) and by Menezes et al. (2008) (15,2 g kg^{-1} to 27,4 g kg^{-1}) in the same mushroom. Crisan and Sands (1978) reported values of fat content from 11 g kg^{-1} to 83 g kg^{-1} for the most mushrooms and Chang and Miles (1989) found values from 17,0 g kg^{-1} to 80,0 g kg^{-1} for champignons, shiitake e shimeji, which shows that the fat content found in this study was in the normal range.

The analyzed residue showed a high phosphorus content (4,2 g kg^{-1}), which was higher than the values found in the literature (0,73 g kg^{-1} a 1,1 g kg^{-1}) for shiitake, shimeji and champignon mushrooms. Among these three mushrooms the champignon showed the highest phosphorus content. (FURLANI and GODOY, 2007; MATTILA et al., 2001). With regard to the calcium content of *A. blazei*, Menezes et al. (2008) reported values ranging from 0,40 g kg^{-1} to 1 g kg^{-1} and Oliveira et al. (1999) found 0,7 g kg^{-1}, which is in agreement with calcium content found in this work (0,41 g kg^{-1}). Potassium content (4,52 g kg^{-1}) was smaller than that reported by Oliveira et al. (1999) (23,4 g kg^{-1}). The contents of phosphorus, calcium and potassium in the residue were significant, which also contributes to its good nutritional value.

Moisture content was significantly low (34,70 g kg^{-1}), which was expectable since the *A. blazei* residue was dehydrated in an oven drying at 50 ± 5 ºC until constant mass was achieved. Considering dried mushrooms of *A. blazei*, Tsai et al. (2008) reported a moisture content of 108,6 g kg^{-1} and Oliveira et al. (1999) found a moisture content of 96,7 g kg^{-1}, but the authors did not specify the drying conditions.

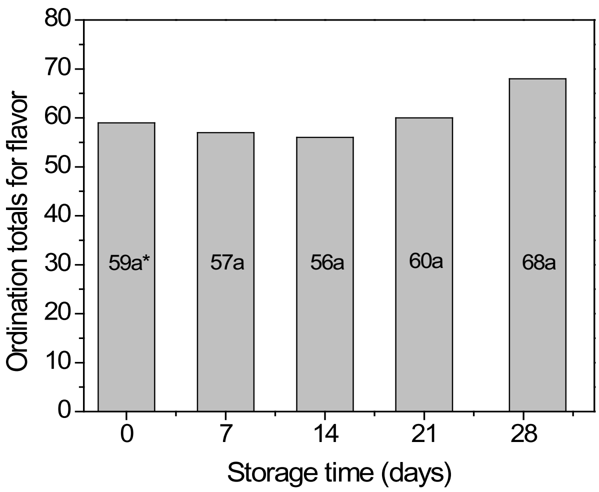
Differences found in the chemical composition of the residue may be due to several factors, since the mushrooms have a varied composition when cultivated in different regions or countries. The substrate type used in the cultivation influences the mushrooms chemical composition, especially concerning fiber, proteins and minerals content (STURION and OETTEREE, 1995).

The physicochemical results have shown that the mushroom residue had a good nutritional quality and can justify its utilization as an ingredient in food products.

One of the greatest obstacles about the use of "mushroom of the sun" residue in human alimentation is its dark brown color developed after the extraction of active principles. This negative change becomes difficult the residue application in food products. The discarding of this byproduct is a waste of nutrients, whereas after b-glucans extraction the amount of remain nutrients is significant. Therefore, studies about the reuse of this residue are very important.

The mushroom residue addition to the cheese bread formulation did not produce significant alterations in the visual aspect of the product, favoring its food products utilization.

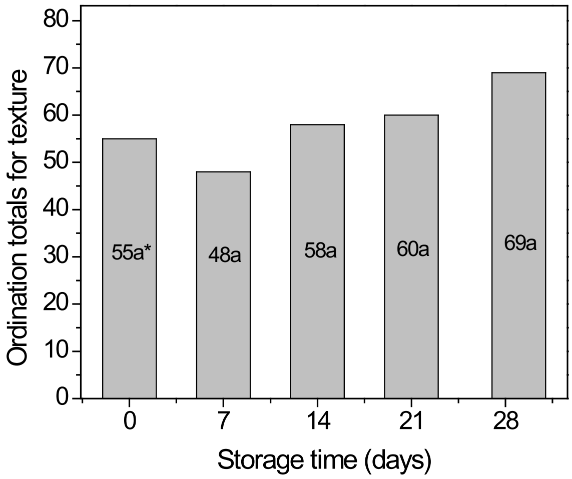
Flavor, texture and appearance attributes were evaluated in the sensory analysis of the fresh sample and of the samples stored in the rage of 7, 14, 21 and 29 days at 18 °C, totalizing 5 samples of cheese bread. These samples were analyzed by 20 trained tasters and the ordination totals for flavor, texture and appearance are shown in Figures 1, 2 and 3, respectively. Considering that samples were putted in order from more preferred (number 1) to less preferred



** Ordination totals labelled with the same letter are similar according to a 5% least significant difference, according to Newell and Mac Farlane table for 5 samples and 20 tasters: (5;20) = minimum significant difference of 28.*

Source: The authors

Figure 1. Ordination totals obtained for flavor attribute in the shelf-life sensory evaluation of cheese bread added of *A. blazei* residue



**Ordination totals labelled with the same letter are similar according to a 5% least significant difference, according to Newell and Mac Farlane table for 5 samples and 20 tasters: (5;20) = minimum significant difference of 28.*

Source: The authors

Figure 2. Ordination totals obtained for texture attribute in the shelf-life sensory evaluation of cheese bread added of *A. blazei* residue

(number 5), high values represent samples with smaller preference.

The Newell and Mac Farlane table indicates that the critical difference between the ordination totals at 5% significance level is 28 points for 5 samples and 20 tasters. Thus, as can be seen in Figures 1, 2 and 3, all samples that have a difference of 28 points or more are significantly different ($p \leq 0,05$) (DUTCOSKY, 1996).

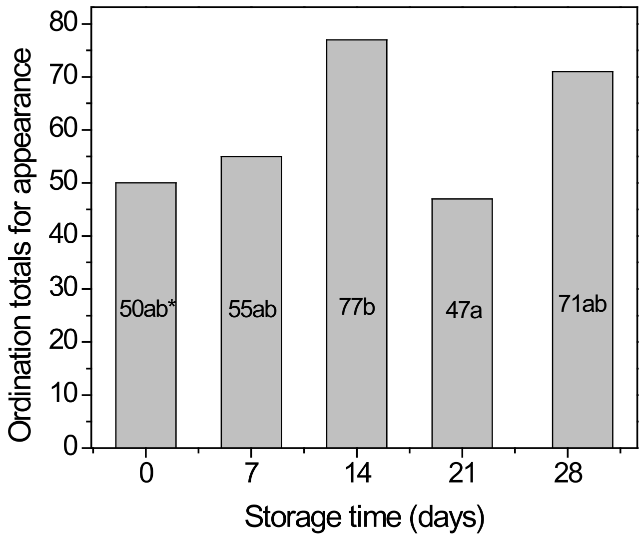
The results showed that there was no significant difference ($p \leq 0.05$) among the samples in the sensory evaluation of texture and flavor attributes (Figures 1 and 2). Concerning the appearance attribute, only the sample with 14 days of storage was significantly different ($p \leq 0.05$) of the sample with 21 days of storage (Figure 3). The appearance ordination total for the sample with 14 days was bigger than that for the sample with 21 days of storage, which indicates that the sample with 14 days was less preferred. This may be explained by the fact that samples were manually modeled, contributing to the lack of uniformity between them.

For all evaluated attributes the sample with larger time of storage (29 days) had good acceptance, showing that is possible the product storage for approximately 30 days without large flavor, texture and appearance negative changes.

Conclusion

The residue of *Agaricus blazei* Murrill is a food of excellent nutritive value for the reason that it has high protein, fiber and mineral content and low fat content.

The stored product (*A. blazei* residue) with storage time of approximately 30 days exhibited good sensory characteristics,



**Ordination totals labelled with the same letter are similar according to a 5% least significant difference, according to Newell and Mac Farlane table for 5 samples and 20 tasters: (5;20) = minimum significant difference of 28.*

Source: The authors

Figure 3. Ordination totals obtained for appearance attribute in the shelf-life sensory evaluation of cheese bread added of *A. blazei* residue

showing that it is possible to use the mushroom residue as a food, avoiding its discarding after the removal of the pharmaceutical extract. These data are an encouragement to the utilization of *A. blazei* Murrill residue in the human diet.

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