Revisión

Mushrooms of the genus *Agaricus* as functional foods

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Abstract

Mushrooms of the genus *Agaricus* are noted for their pharmacological and culinary properties. In this study, it was performed a critical literature review, focusing primarily on aspects of the chemical composition of these mushrooms whose pharmacological properties and nutritional composition characterize them as functional foods. It was also discussed articles conducted in vitro and in vivo proving the high antioxidant potential of the Agaricaceae family, in addition to articles which emphasize the toxicity characteristics and safety for its use in therapy or in human nutrition. These mushrooms exhibit numerous bioactive substances as well as safety regarding toxicity, which characterize them as functional foods. Despite the countless beneficial effects on human health, mushrooms of the genus *Agaricus* are little known by the population, making it necessary partnership and combined efforts among producers, industries and researchers in order to disseminate, research and consumption of these foods.

DOI:10.3305/nh.2012.27.4.5841

Key words: Agaricaceae. Health. Medicinal foods.

Introduction

Edible mushrooms belong to the Funghi group, which can grow in the wild or be cultivated, and after properly prepared, will be suitable for use as food.¹
In accordance with Resolution RDC no 272/05 of the Anvisa (National Health Surveillance Agency), edible mushrooms are classified as products obtained from species of edible fungi, traditionally used as food, and can be prepared in different ways such as dried, whole, fragmented, ground or preserved, subject to drying, smoked, cooked, salted, fermented or any other technical process deemed safe for food production.1

The term functional food attributed to edible mushrooms is due to its rich nutritional value and therapeutic properties described by several researchers, but regulation is permits only after proof of its healthy physiological effects. To be classified as functional foods they should be included in daily eating habits, providing consumers with specific physiological benefits, thanks to its components capable of causing physiological sound effects.2

To be considered functional food, conditions of use and nutritional value, chemical composition or molecular characterization or the product formulation must be registered. Biochemical, nutritional and/or physiological, and/or toxicological tests in experimental animals should also be submitted, further to epidemiological studies, clinical trials, and comprehensive evidence of scientific literature; accredited by international health organizations and international laws recognized under properties and characteristics of the product; proven to be of traditional use by the population having no association with adverse health effects.3

The study of functional foods is very important, since they have beneficial results for the increase in life expectancy of the population. Often times there are cases of chronic diseases such as obesity, atherosclerosis, hypertension, osteoporosis, diabetes and cancer. These ailments have been of great concern both for the population as well as public agencies related to health, are part of their agenda to discuss solutions for better eating habits.4

According to Araújo,5 health-conscious consumers are increasingly looking for foods that help control their own health and well-being. This growing search for a balanced diet in maintaining health has contributed to encourage research into new biologically active substances and safety for toxicity, which characterize them as functional foods. Some species of the genus Agaricus have shown chemical and nutritional composition suitable for human consumption, as well as a flavor much appreciated for culinary purposes.

In 2007 the Brazilian production of mushrooms of the genus Agaricus reached around 40 tons of dehydrated mushrooms, 95% of which destined for export to the Japanese market. In order to increase their profits, many businessmen and farmers started looking for these mushrooms as a new alternative source of income. For this reason, several companies and cooperatives have produced and marketed the inoculum (seed or spawn) of A. blazei or the colonized compost itself. But little is known about the origin and genetic variability of these products.6

To talk about A. sylvaticus is the same as to talk about A. blazei. When there are small differences in morphology, it does not justify creating a new species. Therefore, mushrooms A. sylvaticus and A. brasiliensis are synonyms of A. blazei.7

In a study conducted by Tominazawa et al., the authors investigated nine isolates of A. blazei obtained from different regions in Brazil (São Paulo, Espírito Santo, Minas Gerais, Rio Grande do Sul), through the use of molecular markers to assess genetic similarity among them. The authors concluded that six of the nine isolates showed high genetic similarity and are considered the same origin or clones.

A. sylvaticus mushroom is a Brazilian fungus found natively in the countryside in Brazil. Its popular name is “Sun Mushroom”. This mushroom is ranked as Eukaryotic superkingdom, Fungi kingdom, Metazoa group, Phylum Basidiomycota, class Hymenomycetes, subclass Homobasidiomycetes, order Agaricales, family Agaricaceae.8

Materials and methods

A review of articles published in Data Bases Medline, Lilacs, PubMed, from 1990 to 2012 was done, crossing data between the descriptors in Health Sciences: mushrooms, functional foods, Agaricaceae, in Portuguese, English and Spanish.

Results and discussion

It was found 60 papers and given the reduced number of articles, all of them have been selected in this review. The mushrooms showed numerous bioactive substances and safety for toxicity, which characterize them as functional foods. Some species of the genus Agaricus have shown chemical and nutritional composition suitable for human consumption, as well as a flavor much appreciated for culinary purposes.

In 2007 the Brazilian production of mushrooms of the genus Agaricus reached around 40 tons of dehydrated mushrooms, 95% of which destined for export to the Japanese market. In order to increase their profits, many businessmen and farmers started looking for these mushrooms as a new alternative source of income. For this reason, several companies and cooperatives have produced and marketed the inoculum (seed or spawn) of A. blazei or the colonized compost itself. But little is known about the origin and genetic variability of these products.9

The identification and classification of species of Agaricus mushrooms have been based on morphological and physiological characteristics or by genetic methods, molecular and biochemical. The genetic variability of the genus Agaricus, native or cultivated throughout the world is enormous. Generally these differences are in color, shape and size of microscopic structures and fruiting bodies (spores, plates, and cystides).10

To talk about A. sylvaticus is the same as to talk about A. blazei. When there are small differences in morphology, it does not justify creating a new species. Therefore, mushrooms A. sylvaticus and A. brasiliensis are synonyms of A. blazei.11

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Chemical composition of mushrooms of the genus Agaricus

Through knowledge of the chemical composition of a product, it is possible to recognize its nutritional value and perform analysis of the proportion of homogeneous groups of substances in 100 g of food analyzed. The homogeneous groups of substances considered are those present in all foods, such as water, lipids, protein, fiber, minerals and sugars.

Determination of the chemical composition of mushrooms shows the nutritional value of the food under consideration and can be used as a source of information for nutritional tables on the labels, since several companies that commercialize mushrooms do not display the chemical composition on the Nutrition Facts label of their product.

The high water content in fresh commercialized mushrooms, limits its nutritional value when analyzing a portion of 15 g commonly used on labels. Information on food composition is critical to assess their quality.

There are several factors which directly influence the bromatological characteristics of mushrooms. Among these, species, lineage, post-harvest processing, development stage of the basidiome, the part of the basidiome analyzed and substrate, in addition to genetic factors, environmental characteristics, intrinsic attributes, season and growing conditions, substrate composition, handling, storage and transportation.

According to Braga et al., other determinants for the characteristics of mushrooms, especially when measured protein content are: age, environment and area of cultivation. This fact can be observed when analyzing young mushrooms, which have higher protein content than the more mature ones. According to Shibata et al., larger mushrooms are higher protein content than the more mature ones. According to Tsai et al., mushrooms of the genus Agaricus may have their antioxidant properties associated with a high concentration of tocopherols.

Composition and health benefits

For a food to be considered functional it should have beneficial effects; reach one or more functions or actions in the human body. It should also provide well-being, quality of life, health, and reduce the risk of disease as in the case of chronic degenerative diseases.

Only with the development of more accurate techniques for isolation and purification of chemicals, was it possible to prove scientifically the therapeutic action of some mushrooms, isolating both antibacterial and antitumoral substances.

Agaricales mushrooms and other medicinal fungi exert essential nutritional and pharmacological effects, which can be used as adjuvant in cancer therapy. The mechanisms of action of bioactive substances present in mushrooms are not yet completely understood. But there seems to be clear scientific evidence suggesting that these substances contribute to modulate both the initiation and promotion/progression stages of carcinogenesis, thus propitiating benefits to individuals with various cancers, mainly by immunostimulatory activity.

Several studies have also revealed that A. sylvaticus mushroom potentially reduces tumor growth, stimulates the immune system and even contributes to a better prognosis of these patients improving their quality of life.

In folk medicine the A. brasiliensis mushroom has been used to fight physical and emotional stress, treat and prevent illnesses such as diabetes, osteoporosis and gastric ulcer, digestive and circulatory problems in addition to reducing cholesterol.

The main group of inhibitory agents of carcinogenesis is represented by antioxidant and free radicals blockers; substances capable of slowing oxidation rate. In this way, they inhibit free radicals and prevent diseases, hence contributing to longevity, helping maintain the essential balance between free radicals and antioxidant defense system of the body.

Antioxidant activity

In a study by Costa et al., observation noted that the alcoholic extract of the mushroom A. sylvaticus has great antioxidant potential (74.6%), suggesting that most of the antioxidant compounds present in mushrooms can be diluted more easily by alcohol. However, aqueous and ether fractions showed reduced antioxidant potential (14.6% each) when compared to the alcoholic fraction, since they were less able to hijack the DPPH (2, 2-diphenyl-1-picrylhydrazyl) radical after 20 minutes reaction.

On the other hand the antioxidant potential of different extracts of the A. blazei mushroom, through the DPPH method by Silva et al., showed a higher antioxidant activity (28.6%) in methanol extracts: aqueous (1:1).

According to Percário et al., mushrooms of the genus Agaricus may have their antioxidant properties associated with a high concentration of tocopherols.

The authors suggested that the antioxidant activity of A. sylvaticus mushroom is attributable to the number of molecules present, not to a specific component, and these molecules are easily degraded when exposed to industrial processes, which reduces its antioxidant capacity.
In vitro studies

In a study by Angeli et al., the authors suggested that -glucan present in A. blazei has no genotoxic or mutagenic effect, but protects the damaged DNA (Deoxyribonucleic acid) caused by benzopyrene in test protocols. Results indicate that the beta-glucan works through a link with benzopyrene by capturing free radicals during their activation.

In the clastogenicity test performed by Mantovani et al., the authors discovered that concentrations of 0.2% and 0.4% of A. brasiliensis mushroom were not damage-inducing, unlike a higher concentration of (0.6%). On the genotoxic treatments in SCGE (single cell gel electrophoresis), the concentration of 0.2% of the mushroom extract showed no genotoxic activity, as opposed to concentrations of 0.4% and 0.6% that proved to be effective DNA damage-inducing. Anti-

opposed to concentrations of 0.4% and 0.6% of mushroom extract showed no genotoxic activity, as opposed to concentrations of 0.4% and 0.6% that proved to be effective DNA damage-inducing. Anti-

In a study by Fortes et al., the authors found that dietary supplementation with A. sylvaticus can provide metabolic benefits when analyzing biochemical, enzymatic and blood pressure of patients with colorectal cancer in post-operative phase.

Carvalho et al., aiming at verifying the antinociceptive and anti-inflammatory activity of A. blazei Murill in Wistar rats, through modified formalin test, found results showing that A. blazei acts on nociceptive response and in acute inflammation, because rats treated with this mushroom made fewer movements with paws during phase III, this most likely being related to pain caused by mediators of acute-phase inflammation.

Ishii et al. demonstrated in their studies that A. blazei mushroom has no genotoxic activity but, rather, anti-genotoxic activity. Results derived from these data propose that A. blazei may act as a functional food capable of promoting immunomodulation which can account for the destruction of cells with DNA alterations correlated with the development of cancer. Therefore, supplementation with A. blazei mushroom can be an effective method for the prevention of cancer as well as being an important co-adjuvant treatment in chemotherapy.

In works carried out by Fortes et al., the authors suggested that dietary supplementation with A. sylvaticus mushroom showed to be beneficial in improving well-being and quality of life of patients with colorectal cancer in post-surgery phase.

In a study by Padilha et al., the authors studied the action of A. blazei extract on chronic inflammatory diseases in male albino Wistar rats. Results found indicated that A. blazei extract was active in experimental animals, this response is consistent, since the D-glucan compound is present in the extract.

Fortes et al. conducted a study to assess the effects of dietary supplementation with A. sylvaticus in the lipid profile of patients with colorectal cancer in post-surgery phase. The experiment revealed that dietary supplementation with A. sylvaticus fungi is capable of reducing total cholesterol, LDL-C (low-density lipoprotein cholesterol) and triglycerides, with beneficial outcome on lipid metabolism and, consequently, the prognosis of these patients.

Fortes et al. also found that dietary supplementation with A. sylvaticus fungi acts in regulating fasting blood glucose levels of patients after colorectal cancer surgery. A dietary supplementation with these fungi was found to be successful in reducing blood sugar levels of patients in post-surgery phase, providing beneficial effects on the carbohydrate metabolism of these patients. However, the authors emphasize the importance of studying other clinical conditions to determine the benefits of using A. sylvaticus.

Hi et al. with the purpose of assessing the effects of A. sylvaticus extract in supplemented mice inoculated with pristane (2,6,10,14-tetrametilpentadecano), attested the carcinogen nature of this drug and that the extract of A. sylvaticus mushroom has immunomodulatory activity, without producing toxic effects in test animals.

Hsu et al. obtained results that indicate the potential benefits of supplementation with A. blazei Murill fungus to normalize liver function in patients with hepatitis B after 12 months of clinical observations.

Taveira et al. conducted a study to determine the effects of A. sylvaticus extract on anaemia and C-reactive protein (CRP) levels in rats inoculated with Walker 256 solid tumor. Results suggest that treatment with A. sylvaticus mushroom has positive outcome in animals with Walker 256 tumor. Observation noted that the fungus is capable of reducing anaemia in animals, obtaining results close to those obtained for healthy pets.

Hsu et al. observed in their studies that supplementation with A. Murill blazei improves insulin resistance in patients with type 2 diabetes. The beneficial effects assessed were due to increase in AdipoQ (adiponectin) concentration from adipose tissue with anti-inflammatory and antiteratogenic effect after ingestion of the mushroom for 12 weeks.

Bernardshaw et al. observed an increase in the concentrations of cytokines MIP-2 (macrophage inflammatory protein 2) and TNF-α (tumor necrosis factor alaph) in the serum of mice supplemented with A. blazei extract, resulting in protection against systemic infection by Streptococcus pneumoniae owing to involvement of the innate immune system.
Miglinski\textsuperscript{41} intending to evaluate the immunomodulatory effect of dry \textit{A. blazei} Murill extract on the growth and differentiation of hematopoietic precursors of granulocytes-macrophage (CFU-GM), in bone marrow and spleen of BALB/c mice infected with \textit{Lysteria monocytogenes}, obtained results demonstrating that \textit{A. Murill blazei} has potent immunomodulatory activity able to increase survival of animals infected with a lethal dose of \textit{L. monocytogenes}, likely due to the ability of this extract to restore marrow and spleen hematopoiesis.

In a study by Verçoza-Junior et al.\textsuperscript{42} whose purpose was to evaluate the use of \textit{A. blazei} in the form of filtered and full aqueous suspension (10 mg/animal) in the treatment of mice bearing Ehrlich solid tumor testing its anti-cancer activity, the authors found that animals treated daily with \textit{A. blazei} showed higher values of haematological parameters (erythrogram and leukogram), and final relative spleen weight compared to the control group (distilled water), but with no significant difference ($p > 0.05$).

In works carried out by Ferreira et al.\textsuperscript{43} whose purpose was to evaluate the use of \textit{A. blazei} in rats, assayed its effect on clastogenicity induced by cyclophosphamide. Results derived from this study suggest that in some circumstances \textit{A. blazei} exhibits antimutagenic activity that probably contributes to the anticarcinogenic effects observed.

Takaku et al.\textsuperscript{44} observed the action of ergosterol isolated from the lipid fraction of \textit{A. blazei} as being responsible for antitumor action against sarcoma 180 in mice. According to the authors, tumor regression activity may be related to direct inhibition of angiogenesis, resulting in death of tumor cells.

**Studies on the addition of mushrooms in functional foods**

Bassan et al.\textsuperscript{45} developed a gluten-free cake, sponge like, with \textit{A. brasiliensis} mushroom. The authors obtained positive results in this study because the product reached a high level of acceptance (83.22\%).

Mesono et al.\textsuperscript{46} determined the chemical composition of \textit{A. blazei} residue obtained after aqueous extraction of $\beta$-glucans and analyzed the shelf life of cheese bread made with this byproduct. Observation revealed that \textit{A. blazei} Murill residue is an excellent source of nutrients and its addition in the cheese bread formulation did not cause significant changes in the visual aspect of the product. For all attributes evaluated by the authors, the sample with the largest storage time had good sensory acceptance, which shows the product can be stored for about 30 days without major changes in taste, texture and appearance.

Escouto et al.\textsuperscript{47} noted that there is a diversity of studies on the \textit{A. brasiliensis} mushroom, but realized that there are no literature accounts on the use of this mushroom as food appreciated for its sensory characteristics, nor studies to assess its acceptance. Therefore, we conducted a survey of the acceptance of this mushroom taking a rice dish as reference for developing preparation techniques to boost its use in food. The global average grade obtained in the hedonic scale was 6.14 (liked slightly) and global acceptance rate was 68.3%.

Lemos\textsuperscript{48} developed and characterized a product similar to burger based on the \textit{A. brasiliensis} mushroom and compared their characteristics with a control

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**Eating habits and use of mushrooms**

Among the characteristics necessary for food to be framed as functional food, is that these should be conventional foods consumed in normal and usual diet.\textsuperscript{49}

In Brazil, mushrooms are not part of the diet of most people, being restricted to economic and cultural groups most favored.\textsuperscript{50} According to Shibata et al.,\textsuperscript{41} the greatest barriers to the use of mushrooms in Brazil are linked to popular belief in their poisonous nature, expensive, eating habits and poor availability of product on the market.

The low consumption of mushrooms can also be explained by its recent cultivation in the country, still low productivity compared to its commercialization potential. With the development of new cultivation techniques, the market for these products has become an expensive culture, and their popularity depends on reducing the selling price. This could be achieved through increased production or imports, particularly from countries like China.\textsuperscript{47}

According to Ishii et al.,\textsuperscript{51} further researches must be carried out on the functional characteristics of the genus \textit{Agaricus} mushrooms. Brazil should also pursue a policy of effective use of these foods; enable their consumption by a new target public in the quest for continuous improvement of quality of life and prevention of diseases, mainly cancer.

In research performed by Lemos,\textsuperscript{48} the author concluded that different ways of consumption most used with mushrooms are in sauces, followed by fresh or dry form in soup. Mushroom sauté, pickled, on pizzas, pastas and risottos was also mentioned. However, due to its nutraceutical characteristics, the \textit{A. blazei} mushroom can also be consumed as tea or in capsules containing lyophilized extract.\textsuperscript{51}

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nutr. hosp. 2012;27(4):1017-1024

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formulation in which the mushroom was replaced with ground beef and commercialized products: one with bovine meat and another one with vegetable protein. The sensory analysis showed that the mushroom-based product was well accepted by consumers when their attitude and intention to purchase were tested. The formulation that had 12% of mushroom stood out among the others, presenting high protein content (20.31%), carbohydrates (27.84%), dietary fiber (24.47%) and ash (6.12%), higher than the commercial burgers also evaluated in the work, and lipid content (1.60%) was much lower.

In another study headed up by Miller, it was found that tomato sauces with A. brasiliensis mushroom had higher amounts of polyphenols in relation to sauces without the extract. The results obtained by the author indicated that A. brasiliensis contributed to increase polyphenols in tomato sauces. Glucan complex, lycopene, β-carotene present in this mushroom, meant that when added to tomato sauce they present β-glucan and increased levels of carotenoids and lycopene.

A study was developed by Silva et al., aiming at assessing the antioxidant activity of different extracts of mushroom A. blazei, as well as the oxidative stability of soybean oil added with mushroom extract. Results demonstrated that mushroom extract is effective in preserving the oil, and could be considered a promising natural potential antioxidant ingredient. The authors concluded that further research on its role at different concentrations is fundamental so that mushrooms might be more competitive in the food market.

Toxicity of mushrooms

Despite the fact that mushrooms are considered a functional food, they may also present some type of toxicity. However, for a food to be considered functional, there should be no risk or toxic effects for the consumer.

The substrate exerts direct influence on the chemical composition of mushroom, because nutrients are removed by hyphae which are in direct contact with this material. Consequently, they absorb essential elements, but together with these they can accumulate toxic metals such as lead, mercury, cadmium, arsenic and others. In this sense, some species of mushrooms have been used as bioindicators of environmental pollution. Knowing that chemical composition of mushrooms may be related to the substrate, it stands to reason that a polluted region will produce mushrooms with high levels of metals. This fact was observed by Kalac et al. when they presented different species of mushrooms such as A. sylvaticus, with high levels of accumulated cadmium.

In a study performed by Moura it was detected the presence of arsenic in mushrooms of the genus Agaricus. But this fact was not considered indicative of risk to human health, since the concentration of this element in the samples analyzed by the author was rather low.

Bellini et al. observed that the methanolic fractions of A. blazei tested in their study did not provide chemical protection, being potentially mutagenic according to results in HGPRT test. For the authors, the methanol extracts of this mushroom should not be used widely by individuals because of the possibility of their genotoxicity. Therefore, care must be taken in the use of A. blazei by the population as long as a comprehensive assessment of the biochemical characterization of this fungus is not complete.

In a study conducted by Sugui, the outcome indicates no mutagenic, genotoxic or carcinogenic effects on rats tested with the aqueous solution of the A. brasiliensis. Nevertheless, an antimutagenic effect against the mutagenicity of ENU (N-ethyl-N-nitrosourea) was observed in bone marrow cells, in addition to a significant reduction in the number of aberrant crypts per focus (4-6 crypts/focus) induced by DMH (1,2-dimethylhydrazine) in the colon of animals post-treated with the aqueous solution of the mushroom. In this context, results suggest that the aqueous solution of A. brasiliensis possesses compounds that can significantly reduce the frequency of micronucleated cells from bone marrow of rats, and that they can act at a later stage of carcinogenesis initiation.

In study carried out by Singi et al. results revealed that the concentration of 1.25 mg/kg of A. blazei mushroom did not cause significant changes in mean arterial pressure (MAP) or heart rate (HR). The concentration of 2.50 mg/kg of mushroom caused decreased MAP to 15s (p < 0.01) and HR to 30s (p < 0.001) and of 5.00 mg/kg decreased MBP to 15s (p < 0.001) and HR at 15 and 30s (p < 0.001).

Costa et al. aiming at evaluating the possible protective effects of A. blazei tea against the urethane genotoxic action in somatic cells of Drosophila melanogaster, noted that no increase was statistically significant in the frequency of mutant spots in larvae exposed to A. blazei tea. However, when this mushroom was associated with urethane, we observed a reduction statistically significant in the frequency of mutant spots. The results imply that A. blazei is not genotoxic and has a protective effect against the genotoxicity of urethane.

With the intent of investigating effects of acute toxicity of A. sylvaticus aqueous extract by clinical, biochemical and histopathological on healthy mice, Novaes et al. verified that both the administration of the aqueous extract as well as the placebo, caused a temporary rise of apathy, piloerection and respiratory changes, which were slightly more persistent in the group treated with the fungus. Biochemical and histopathological changes were not statistically significant between groups. The authors determined that administration of A. sylvaticus aqueous extract showed very low toxicity.

In a study by Ishii et al. the researchers concluded that the Agaricus blazei mushroom offers no genotoxic
consequences, but made it possible to visualize the anti-
genotoxic effects. The results suggested that the fungus
acted as functional food, capable of promoting immuno-
modulation when the destruction of cells with DNA
damage correlated with cancer development was
observed. Therefore, the Sun mushroom had a preven-
tive effect against colorectal neoplastic lesions assessed.

Orsine et al. observed that A. *sylvaticus* extract has
no toxicity proving to be safe for human use.

### Conclusions

To be included in the group of functional foods,
mushrooms should bring benefits to human health, do
not present themselves toxic and be included in the
daily eating habits. Thus, the benefits of eating mush-
rooms of the genus *Agaricus* are shown in several
papers. Currently there are many researchers working
in order to spread the advantages of the consumption of
mushrooms of the genus *Agaricus*.

It has been shown in some studies the rich nutritional
composition of mushrooms of the genus *Agaricus*, and
the presence of substances that act on the human body,
being widely used in therapy against cancer. Also low
toxicity was observed in different studies using diffe-
rent toxicological methods evaluation.

Despite the countless beneficial effects on human
health, mushrooms of the genus *Agaricus* are little
known by the population, making it necessary part-
nership and combined efforts among producers, indus-
tries and researchers in order to disseminate, research
and consumption of these foods.

### Acknowledgments

This paper was made possible by the supported and
assistance of Fundação de Ensino e Pesquisa e Ciências
da Saúde-FEPECS.

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