PBPC ISSN 2674-9432
PBPC ISSN 2674-9432
Qualis A3 CAPES 2021-2024 | DOI - Crossref | Latindex | Indexado no Google Acadêmico

Quantification of beta glucans and phenolic compounds present in extract of Lion's Mane mushroom (Hericium erinaceus) from Brazil.

David Lucas Nunes de Souza Cruz[1]; Carolina Pereira de Morais Faria[1]; Erna Elisabeth Bach[2].

https://doi.org/10.36557/2674-9432.2026v5n2p156-170
Artigo recebido em 2 de Fevereiro e publicado em 2 de Abril de 2026

ARTIGO ORIGINAL

RESUMO

O cogumelo Juba de Leão (*Hericium erinaceus*) é um basidiomiceto comestível com propriedades medicinais reconhecidas, incluindo efeitos anticancerígenos, neuroprotetores e imunomoduladores. Ele contém compostos biológicos e bioativos, como beta-glucanas, compostos fenólicos, proteínas e terpenoides. Este estudo enfoca a extração e quantificação de beta-glucanas e compostos fenólicos de *H. erinaceus* cultivado no Brasil, utilizando métodos de extração aquosa. Os resultados demonstram que o aumento da temperatura da água acima de 90°C eleva a concentração de beta-glucanas e proteínas, confirmando maior solubilidade. Todos os extratos apresentaram compostos fenólicos como ácido clorogênico, ácido cafeico, ácido ferúlico, ácido p-cumárico e kaempferol, importantes para manter o efeito antioxidante. A precipitação com etanol (até 20%) separa efetivamente os compostos bioativos, destacando diferenças nas concentrações de fenóis extraídos. Os resultados enfatizam a necessidade de técnicas de extração personalizadas dependendo da aplicação pretendida, seja para uso alimentar, farmacêutico ou medicinal.

Palavras-chave: cogumelo juba de leão, beta-glucanas, proteínas e compostos fenólicos

ABSTRACT

The Lion's Mane mushroom (*Hericium erinaceus*) is an edible basidiomycete with recognized medicinal properties, including anticancer, neuroprotective, and immunomodulatory effects. It contains biological and bioactive compounds such as beta-glucans, phenolic compounds, proteins, and terpenoids. This study focuses on the extraction and quantification of beta-glucans and phenolic compounds from *H. erinaceus* cultivated in Brazil using aqueous extraction methods. The results demonstrate that increasing water temperature above 90°C enhances the concentration of beta-glucans and proteins, confirming greater solubility. All extracts presented phenolic compounds as chlorogenic acid, caffeic acid, ferulic acid, p-coumaric acid and kaempferol that are important to maintain the antioxidant effect. Ethanol precipitation (up to 20%) effectively separates bioactive compounds, highlighting differences in extracted phenolic concentrations. The findings emphasize the need for tailored extraction techniques depending on the intended application, whether for food, pharmaceutical, or medicinal purposes.

Keywords: Lion's Mane mushroom, beta-glucans, proteins and phenolic compounds

Instituição afiliada – David Lucas Nunes de Souza Cruz[1]; Carolina Pereira de Morais Faria[1]; Erna Elisabeth Bach[2].
[1]Pos-graduated master student, Fisheries Institute/APTA/SAA—Postgraduation Program; [2] Researcher associated, Institution: Fisheries Institute/APTA/SAA

Autor correspondente: *ErnaE Bach* ernabach@gmail.com

1. INTRODUCTION

The lion's mane mushroom (*Hericium erinaceus* (Bull.: Fr. Pers.)) is an edible basidiomycete of Chinese origin, widely used in traditional Chinese medicine. It belongs to the class Basidiomycetes, subclass Holobasidiomycetidae, order Hericiales, and family Hericiaceae (Musulmanbekovich & Ayaulym, 2021; Thongbai et al., 2015; Wojewoda, 1998). Lion's mane is also known by various names worldwide, including its Japanese name (Yamabushitake), Chinese name (Houtou), and other common names such as Monkey's Mushroom, Bear's Head, Hog's Head Fungus, White Beard, Old Man's Beard, Pom Pom, and Bearded Tooth (He et al., 2017).

H. erinaceus, considered a saprotroph or weak parasite, primarily grows on dead wood but can also be found in knotholes or cracks of living hardwoods. It is cultivated at the greenhouse level using a substrate. The mature fruiting body is fleshy, semi-spherical, and whitish, gradually changing to a yellowish or brownish hue with age (Boddy et al., 2004). In Brazil, it is cultivated in substrate-containing blocks within greenhouses, where its production has been increasing annually due to its medicinal properties.

This mushroom is well known for its diverse medicinal properties, including anticancer, antioxidant, antimicrobial, hypolipidemic, neuroprotective, immunomodulatory, and anti-aging effects (Chaiyasut & Sivamaruthi, 2017; Chakraborty et al., 2021; Ghosh et al., 2020; Khan et al., 2013, Roda et al, 2021). Robinson et al. (2015) described its potential in treating cognitive impairment and mental disorders such as Alzheimer's disease in mice.

Recently, modern science has become increasingly interested in the bioactive compounds present in lion's mane, particularly beta-glucan, a polysaccharide with recognized immunomodulatory and antitumor properties. Beta-glucan is a type of soluble fiber found in the cell walls of bacteria, fungi, yeast, and certain cereals such as oats and barley. This compound is known for its ability to modulate the immune system, acting as a bioactive agent that can positively influence immune responses. In lion's mane, beta-glucan is one of the primary components responsible for its therapeutic effects (Chakraborty et al., 2021; Ghosh et al., 2020; Parada et al., 2015).

The purpose of the present study is to introduce a methodology for the extraction and preparation of aqueous lion's mane extracts and the detection of bioactive compounds. Aqueous extraction is one of the most commonly used methods for obtaining bioactive compounds from mushrooms due to its simplicity and efficiency. Moreover, the precise extraction and determination of beta-glucan concentration is crucial for assessing the quality and therapeutic potential of the obtained extracts.

2. MATERIALS AND METHODS

2.1. Origin, Extraction and biochemical analyses of *H. erinaceus*

The dried mushrooms were sent by the company Grupo Alta Villa, a producer of edible mushrooms located in São Roque, São Paulo, Brazil. Then were finely milled in a hammer mill equipped with a 1mm mesh stainless steel sieve. The sieve powder was stored dry until further use (Figure 1).

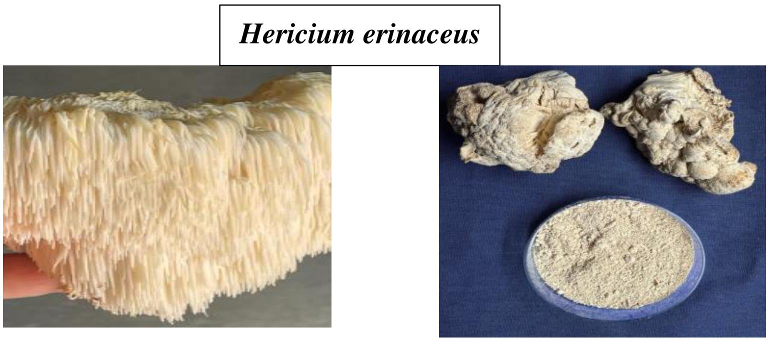


Figure 1: Aspect from fresh, dried mushrooms and powder.

Two gram of dry mushroom was submitted to four methods for extracting with water that are: 1) mass homogenizing in 50mL of water and maintained in cool for one hour that´s (JCollW); 2) mass homogenate in 50mL of water, but kept at 60C for one hour (JW60); 3) mass homogenate in 50mL of water, but kept at 90C for one hour (JW90); 4) mass homogenate in 50mL of water, but kept at 120C for 10 minutes in autoclave (JW120). All extracts were filtered with paper filter (Whatmann n.1). For analysis by HPLC the extracts were also filtered in millipore 0.45um.

Another sample was two grams of dry mushroom extracted with 50mL of water, kept to 90C for one hour and then precipitated sugar with alcohol 96% until volume final was 20% hidroalcoholic. The precipitated was perform over night at 4C and centrifuged to obtain Sobrenadant (Sn) and also Sediment (Sd). The sediment was restaured with 10mL of water. Only Sn was analysed in HPLC and filtered in millipore 0.45um. With Sd was make biochemical analyses but don´t HPLC.

All extracts were repeated three times and analyzed proteins (Lowry et al., 1951), phenols (Swain and Hillis, 1959); beta-glucan (Lever, 1972, Van Hoof et al. 1991, using enzyme beta-glucosidase Sigma); anthrone (Yemm and Willis, 1954) and antioxidants (Rufino et al., 2007). For Standart in proteins was used serum albumin bovin (SBA mg), for phenols (chlorogenic acid mg), for anthrone (glicose mM) and for antioxidants was Trolox expressed as Trolox equivalents (µmol trolox/min/1mL /1g mushroom).

2.2. ***HPLC***

The phenolic compounds present in the extracts were separated and identified by HPLC (high pressure liquid chromatograph) equipment (Yong YL 9300), equipped with a quaternary pump, UV-vis detector and YL9330 column. The column used was Kinetex C18 (4.6mm x 250mm diameter and 5um) with a wavelength of 254nm. The elution was 1mL/min at 35°C. Phase A consisted of methanol and phase B of 0.1% acetic acid in water, with the following program: 0-1 min (20% B); 1.01-1.5 min (5% B); 1.51-8 min (4% B). The volume injected into the column was 20uL [41]. The compounds used as standards are from Sigma (coumaric acid, ferulic acid, caffeic acid, rutin, quercetin, kaempferol, chlorogenic acid; benzoic acid) and dissolved in HPLC grade solvent. The identification of the compounds was done by the Clarity software involving retention time and peak height, compared with the standards at the time of the run and the corresponding quantification in micrograms of each standard.

3. RESULTS

3.1. Extract and biochemical analyses

Aqueous extracts from the lion's mane mushroom were subjected to different temperatures and biochemically analyzed. High temperatures resulted in the breakdown of the polysaccharide-protein complex structure and demonstrated great ability to extract beta-glucan, protein and exhibit antioxidant capacity (Table 1).

As for the free sugars of both alpha and beta bonds, as the beta glucan increases, there are fewer broken sugars. Undoubtedly, the higher the temperature, the greater the quantity of phenolic compounds may appear, although this quantity is small.

Observing the same Table 1, the test involving anthrone indicates the amount of glucose in mM (milimol). Higher temperature, demonstrated greater total glucose concentration. Regarding the aqueous extract at 90°C, followed by the precipitation of beta-glucan with alcohol, it confirms that the sediment contains a higher amount of beta-glucan, total sugar in mM, and antioxidant activity when compared to the supernatant. However, when analyzing the phenol content, it was higher in the supernatant.

3.2. HPLC

HPLC-UV-Vis system was used to identify and quantify phenolic compounds in all extracts from mushroom. The quantification and identification was made by analysing spectra comparison with Standart compounds and retention time by software Clarity. Figure 2 demonstrating the peak chromatograms with height peak and retention time from three times of extraction.

Table 1: Quantification of beta-glucan, sugar free alfa or beta, proteins, phenols and antioxidant activity present in extracts of mushroom with 1gram submitted in different temperatures.

Extracts	mg glucose ligation beta	mg glucose free, alfa or beta	mg protein (SAB)	mg phenol (ác clorog)	Antioxidant (uM trolox/min/1mL)	conc glucose total mM
JCollW	13,72+/-1,25*	201,68+/-0,87	1,257+/-0,74	0,237+/-0,64	71,01+/-1,07	12,29+/-1,04
JW60	162,26+/-5,62	212,02+/-0,27	2,554+/-0,98	0,466+/-0,62	155,98+/-2,77	22,39+/-1,06
JW90	181,68+/-2,37	188,34+/-3,09	2,588+/-0,85	0,554+/-0,58	152,81+/-2,57	24,14+/-1,28
JW120	183,60+/-1,84	186,86+/-2,22	3,848+/-0,32	0,677+/-0,24	152,48+/-2,67	30,32+/-0,58
JW90 ppt Sn	20,23+/-0,47	172,24+/-0,96	0,620+/-0,07	0,892+/-0,67	55,48+/-1,08	12,85+/-0,26
JW90 ppt Sd	134,70+/-4,67	48,14+/-0,57	1,320+/-0,08	0,231+/-0,18	102,40+/-1,27	23,46+/-0,34

JCollW=mass homogenizing in water and maintained in cool for one hour ; JW60= mass homogenate in water, but kept at 60C for one hour; JW90= mass homogenate in water, but kept at 90C for one hour; JW120= mass homogenate in water, but kept at 120C for 10 minutes in autoclave.
JW90 ppt Sn= mass homogenate in water, kept at 90C for one hour, precipitate with alcohol and centrifugate, Sobrenadant (Sn). JW90 ppt Sn= mass homogenate in water, kept at 90C for one hour, precipitate with alcohol and centrifugate, Sediment (Sd).
*Mean from three repetitions +/- Standard Error

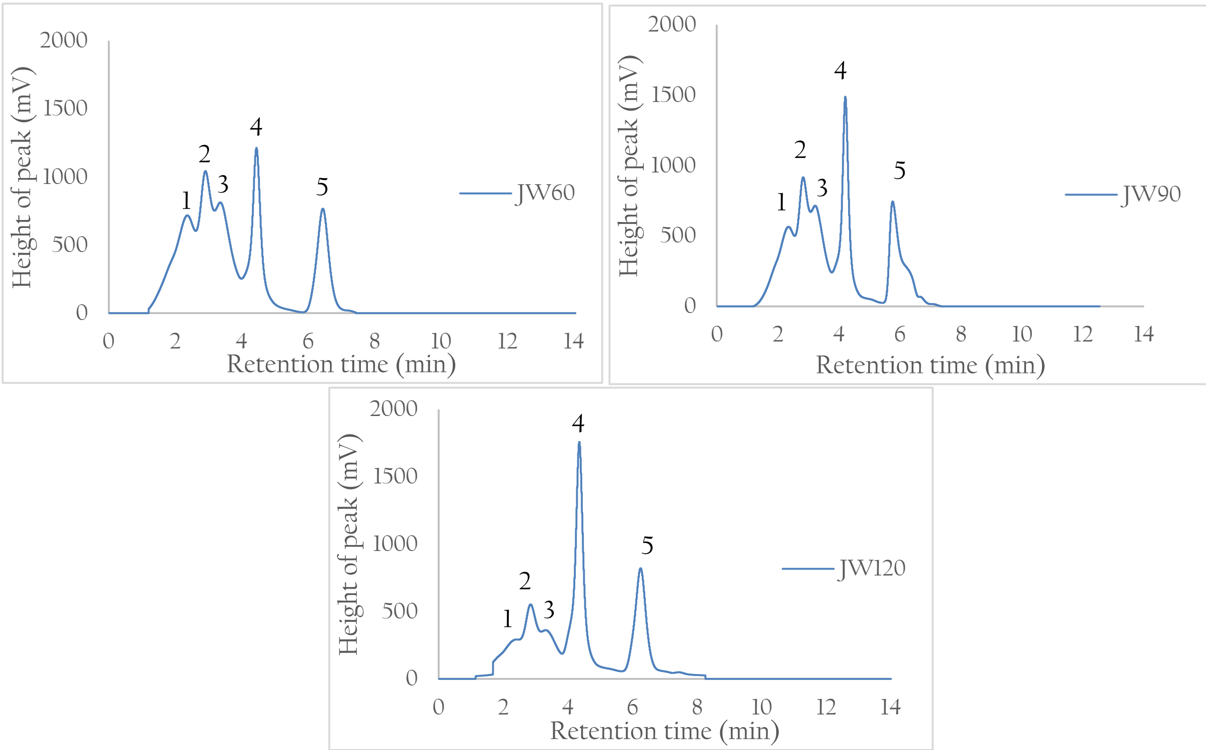


Figure 2. HPLC-UV-Vis chromatograms of phenolic compounds from three types of extraction. The compounds were in 1=chlorogenic acid; 2= cafeic acid; 3= ferulic acid; 4=p-coumaric acid; 5= Kaempferol. OBS: JW60= mass homogenate in water, but kept at 60C for one hour; JW90= mass homogenate in water, but kept at 90C for one hour; JW120= mass homogenate in water, but kept at 120C for 10 minutes in autoclave.

In Table 2 observed the quantification in micrograms of phenolic compounds obtained by Standart curve. With the extract JCollW we have trace of compounds in HPLC and have not chromatogram. It was possible to observe that for chlorogenic, caffeic and ferulic acids the quantity decreased with increasing temperature. For P-coumaric acid this compound increased with increasing temperature. For kampferol the quantity increases from 60°C to 120°C, remaining slightly below 90°C.

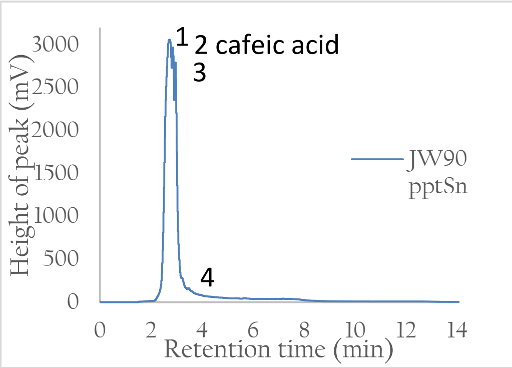
Table 2: Quantification in micrograms of each phenolic compounds present in extracts of mushroom with 1gram submitted in different temperatures.

Compounds (ug)	JW60*	JW90	JW120
1.Chlorogenic acid	798,620	558,060	301,720
2.Cafeic acid	634,620	517,290	316,010
3.Ferulic acid	4610,04	4025,08	2024,09
4.p-coumaric acid	679,940	817,710	960,510

5.Kaempferol	1496,240	1376,050	1574,980

*JW60= mass homogenate in water, but kept at 60C for one hour; JW90= mass homogenate in water, but kept at 90C for one hour; JW120= mass homogenate in water, but kept at 120C for 10 minutes in autoclave.

When the mushroom was heated to 90°C and then precipitated with ethanol, resulting in 20% at the end as hydroalcoholic, the supernatant exhibited 3 phenolic compounds that were unknown according to the standards used in the HPLC test (Figure 3)



Compound	Retention time [min]	Height [mV]	ug
1. unknow	2,720	3052,67	X
2. cafeic acid	2,837	2942,60	1756,15
3 unknow	2,937	2783,51	X
4. unknow	3,333	180,75	X

Figure 3. HPLC-UV-Vis chromatograms of phenolic compounds from mushroom heated, precipitated with etanol and centrifugation obtained Sn. The compounds were in 1, 3 and 4 =unknow; and 2= cafeic acid.

4. DISCUSSION

There are several methodologies described for the extraction and purification of bioactive compounds from mushrooms. The most widely adopted method for extracting polysaccharides is to pulverize the mushroom powder in heated water for several hours, which is very time-consuming (Sun et al, 2022). Wadt et al (2015) working with Ganoderma show that extraction from beta glucan can be made with hot water for one hour and also parts with water-alcohol .

In the case of *H. erinaceus*, Ookushi et al (2009) demonstrated the ability to extract in water using a microwave oven at 140◦C for 5 min, which is equivalent to extraction in water at 100◦C for 6 h. However, a microwave oven works for small quantities, and the extracted polysaccharide is rich in β(1-3) bonds, while in normal water with further heating it is β(1-6) bonds. Another process involved green solvents using super-critical CO_2 extraction that was used to obtain soluble fractions from *H. erinaceus* (He et al, 2017; Wang et al, 2019). That process resulted in a dissolution of more than 40% of *H.*

erinaceus (Parada et al ,2015). Another process that has been reported involves the use of enzymes to characterize polysaccharides in the purification process (Zhu et al, 2014).

Thongbai et al. (2015) in his review described the presence of glucan, proteins, phenols, lectins, and terpenoids in fruiting bodies of *H. erinaceus*, which are considered as most dominant bioactive compounds of this mushrooms. Friedmann (2015) confirmed in his review the presence of glucan, proteins, phenols, lectins, and terpenoids in fruiting bodies of *H. erinaceus*, which are considered as most dominant bioactive compounds of this mushrooms. In the same work confirmed in the presence of five classes of organic compounds (erinacines, aromatic compounds, steroids,alkaloids, and lactones) isolated from the fruit bodies. However, Shirokikha et al (2020)isolated polysacharides from *H. erinaceous* by extraction with hot water (70C), followed by precipitation with 96% ethanol. Residues of rhamnose, fructose, xylose, arabinose, mannose, glucose, and galactose in the ratio of 7: 8:1:19:14:26:27, respectively, were identified in the composition of the polysaccharide. This have a cryoprotective activity (James et al, 2024).

Based on the articles, water extraction was performed, according to Wadt et al (2015) but, taking into account Shirokikha et al (2020) and Ookushi et al (2009), working with other temperatures. Mori et al (1998) was working with hot water extraction and purification of beta-glucans that have antitumor activity Against Sarcoma 180. This polysaccharide were composed of a backbone of *β*-(l → 6)-linked D-glucopyranosyl residues, and had *β*-(1 → 3) and *β*-(l → 6) glucosidic linkages.

Thus, partial results obtained here demonstrated that the extraction of polysaccharides, mainly beta-glucan, was only extracted in greater concentration when the water temperature was elevated, proving greater solubility. The bioactivities of polysaccharides, including β-glucans, can be influenced by factors such as chain conformation and the introduction of suitable ionic groups with an appropriate degree of substitution. In the case of extraction above 90°C, it was possible to verify a greater concentration of beta glucan, confirming the well-closed structural conformation and only releasing after heating.

It was also possible to verify the amount of free alpha or beta glucose in the extracts, verifying a variation from 186 to 212 mg/1 g of dry mushroom. Regarding the concentration of total phenols, as the extraction temperature increased, there was an increase in the amount of phenols. Even so, it was below that found by Wadt et al (2015) working with *Ganoderma lucidum.*

The anthrone test, which determines the concentration in mM of total sugar, demonstrated that in extracts subjected to different temperatures, a higher extraction temperature presented a greater quantity of sugar, ranging from 12.29 to 30.32 mM of glucose. However, overall, β-glucans from mushrooms contribute significantly to promoting health and preventing various diseases but its necessary other bioactive compounds such proteins, flavonoids, phenolic compounds, ergosterol, and others (James, 2024; González et al, 2020; Xu et al., 2011; Wang et al., 2019; Zhang et al., 2012; Zhu et al, 2015).

The presence of protein was verified in the extracts, and as the water temperature increased, the protein concentration also increased. This indicated that the protein must be strongly bound to the polysaccharide and, when this beta-glucan was solubilized, the protein also came out. The process released the polysaccharide-protein complex. According to Zan et al., 2015; Zhang et al., 2017 and Younis, 2017, the presence of the complex was important for presenting antiproliferative activity against various cancer cells, highlighting their potential as bioactive compounds with therapeutic implications.

However, Deshmukh et al. (2021) have shown that in addition to functional macromolecules such as polysaccharides and proteins, *H. erinaceus* also contains small molecular active components such as terpenoids, cerebrosides, phenols (phenolic acids, flavonols) and sterols. Polyphenols act as antioxidants by inhibiting oxidative changes, a fundamental mechanism in the development of endothelial lesions associated with conditions like atherosclerosis and others diseases, including cardiovascular disease, osteoporosis, neurodegenerative diseases, cancer, and diabetes mellitus. Zhu et al (2014) described that carbohydrates are the most abundant fraction in *H. erinaceus*, but the water and alcoholic extractives have phenolic and flavonoid componentes. However, Nagem et al. (1992) and Ozyürek et al. (2014) said that the presence of ferulic acid in the mushroom demonstrate its importance for the human health, because possesses antioxidant action with activity of protection against the free radicals and related diseases.

Partial results demonstrated that extracts presented phenolic compounds as chlorogenic acid, caffeic acid, ferulic acid, p-coumaric acid and kaempferol in all extracts. Concentration of acids as chlorogenic, caffeic and ferulic, the quantity decreased with increasing temperature, as if it were oxidizing, however the function of antioxidant activity was maintained. As for p-coumaric acid, this compound increased with increasing temperature. Kaempferol only increased in extracts from 60°C and 120°C, remaining slightly below at 90°C. However, presence of phenolic compounds are important to maintain the antioxidant effect.

Li et al (2012) working with methanolic extract of *H. erinceous* and identified phenolic acids chlorogenic acid, p-hydroxybezoic acid, vanillic acid, ferulic acid, sinapic acid, syringic acid and p-anisic acid and flavonoids (catechin, epicatechin, rutin, myricetin, hesperidin and quercetin). In the present work with aqueous extract and confirmed with the work of Li et al (2012) was the presence of ferulic acid and chlorogenic acid.

According to Zhu et al (2015) and Thongbai et al (2014), hericenones and erinacines were isolated from *H. erinaceum*. In this case, a dual extract that combines the extraction with alcohol or hot water can be important. In the present study, aqueous extraction was performed at 90°C and then precipitated with 96% ethanol (up to 20%) and the supernatant was separated from the sediment. In the sediment, beta-glucan was quantified at 134.70 mg, protein 1.32 mg, phenol 0.231 mg, and total sugar concentration was 23.46 mM. However, in Sn, more free sugars with beta and alpha bonds were found, a low amount of beta-glucan, but phenol in high concentration (0.892 mg) compared to other extracts. In the HPLC analysis, it was possible to visualize 3 unidentified compounds and one, identified as caffeic acid in high concentration. This demonstrates that aqueous and then alcoholic extracts extract different phenolic compounds.

Qiu et al (2024) analyzed in their review the bioactive compounds with biological activity of *H.erinaceus*. The compounds involve polysaccharides, hericenones, erinacines, proteins and peptides. For each case involving cancer or neuroprotection, the extraction must be different because it will involve different compounds. And then, after being determined as food or medicinal, the industry will be able to adjust its extract.

5. CONCLUSION

In this context, what has been verified so far in the work carried out is that beta-glucan is important, the concentration of phenols should be low but there should also be phenolic compounds (chlorogenic and caffeic acid) as well as proteins, among other compounds, but it should be verified what is most important for each disease or treatment. However, when used water can be use temperature above 90C for one hour and can mixture with alcohol until 20%. This study base will be used in the future to develop feed incorporating this mushroom (extract) as food for fish.

ACKNOWLEDGMENTS

The authors would like to thank the Fisheries Institut, São Paulo, Brazil for infrastructural, as well Coordenação de Aperfeiçoamento de Pessoa de Nível Superior [CAPES] for Master's Scholarship. Also thanks to Angela Vanelli Weschenfelder from the company Grupo Alta Villa, located in São Roque, SP, Brazil, for having sent dried lion's mane mushroom.

This work was supported by Master's Scholarship by CAPES (Coordenação de Aperfeiçoamento de Pessoa de Nível Superior) under Grant (88887.902344/2023-00).

ORCID

David Lucas Nunes de Souza Cruz http://orcid.org/0009-0003-5442-0086; Carolina Pereira de Morais Faria http://orcid.org/0000-0002-0169-2905; Erna Elisabeth Bach http://orcid.org/0000-0003-3238-673x

REFERENCES

BODDY L., WALD P.M., PARFITT D., ROGERS H.J. Preliminary Ecological Investigation of Four Wood-Inhabiting Fungi of Conservation Concern: Oak Polypore Piptoporus Quercinus (=*Buglossoporus Pulvinus*) and the Tooth Fungi *Hericium/Creolophus* spp., **English Nature**, Peterborough, pp. 188–189, 2004.

CHAIYASUT, C., AND SIVAMARUTHI B. S. Anti-hyperglycemic property of *Hericium erinaceus*—A mini review. **Asian Pacific Journal of Tropical Biomedicine**, *7*:1036–1040. https://doi.org/10.1016/j. apjtb.2017.09.024, 2017.

CHAKRABORTY N., BANERJEE A., SARKAR A., GHOSH S., ACHARYA K. Mushroom polysaccharides: A potent immune-modulator. **Biointerface Research in Applied Chemistry**, 11:8915–8930. https://doi.org/10.33263/ BRIAC 112.89158930, 2021.

DESHMUKH S., SRIDHAR K., GUPTA M. *Hericium erinaceus*-a rich source of diverse bioactive metabolites**.** **Fungal Biotec** 1(2):10-38. https://doi. org/10.5943/FunBiotec/1/2/2, 2021.

FRIEDMAN M. Chemistry, Nutrition, and Health-Promoting Properties of *Hericium erinaceus* (Lion's Mane) Mushroom Fruiting Bodies and Mycelia and Their Bioactive Compounds. **J. Agric. Food Chem**. 63:7108−7123, 2015.

GHOSH S., CHATTERJEE T., SARDAR A., CHATTERJEE I., BOSE A., DASGUPTA A., MALITHA A., ACHARYA K. Antioxidant properties and phytochemical screening of infusion and decoction obtained from three cultivated *Pleurotus* species: A comparative study. **Jordan Journal of Pharmaceutical Science** *13*: 121–129, 2020.

GONZÁLEZ A., CRUZ M., LOSOYA C., NOBRE C., LOREDO A., RODRÍGUEZ R., CONTRERAS J., BELMARES R. Edible mushrooms as a novel protein source for functional foods. **Food & function** 11 (9):7400-7414, 2020.

HE X., WANG X., FANG J., CHANGA Y., NINGA N., GUOA H., HUANGA L., HUANGA X., ZHAO Z. Structures, biological activities, and industrial applications of the polysaccharides from *Hericium erinaceus* (Lion's Mane) mushroom: A review. **International Journal of Biological Macromolecules** 97:228–237, 2017.

JAMES J.A.K. Medicinal Mushrooms: The Extraction and Analysis of Bioactive Compounds from *Hericium erinaceus* and *Ganoderma lucidum* Mushrooms. **Electronic Theses and Dissertations.** 9145, 2024. https://scholar.uwindsor.ca/etd/9145.

KHAN M. A., TANIA M., LIU R., RAHMAN M. M. *Hericium erinaceus*: An edible mushroom with medicinal values. **Journal of Complementary and Integrative Medicine** 10:253–258, 2013. https://doi.org/10.1515/jcim-2013-0001

LEVER M. A new reaction for colorimetric determination of carbohydrates. **Analytical Biochemistry**, Academic Press 47: 273-279, 1979.

LI H., PARK S., MOON B., YOO Y.B., LEE Y.W., LEE C. Targeted phenolic analysis in *Hericium erinaceum* and its antioxidant activities. **Food Science and Biotechnology**, 21, 881e888, 2012.

LOWRY O.H., ROSEBROUGH N.J., FARR A.L., RANDALL R.J. Protein measurement with the Folin phenol reagent. *Journal of Biological Chemistry* 193, 265–275, 1951.

LUQUIS F., SILVA A.A.O., WADT N.S.Y., BACH HI E.M., MARTINS A.M.C.; BACH E.E. *Arctium lappa* L.: Chemical composition, antioxidants, phytochemical compounds and use for healing activity. **South Florida Journal of Development**, Miami, 2 (2):2063-2071, 2021.

MORI H., AIZAWA K., INAKUMAT., ICHII A., YAMAUCHII R., KATO K. Structural analysis of the β-D-glucan from the fruit-body of Hericium erinaceum. **Journal of Applied Glycoscience** 45 (4): 361-365, 1998.

MUSULMANBEKOVICH A.B., AYAULYM K.A. Medicinal properties of *Hericium erinaceus*. **Encyklopedia biologiczna**. Opress, Kraków. 1-6, 2021.

NAGEN T. J., ALBUQUERQUE T.T.O., MIRANDA L.C.G. Ácidos fenólicos em cultivares de soja: Ação antioxidante. **Arq. Biol. Tecnol**. 35: 129-138, 1992.

OOKUSHI Y., SAKAMOTO M., AZUMA J. Effects of microwave irradiation on water-soluble polysaccharides of the fruiting body of *Hericium erinaceum*. **J.Appl. Glycosci** 56: 153–157, 2009.

OZYÜREK M., BENER M., GUCLU K., APAK R. Antioxidant/antiradical Properties of microwave-assisted extracts of three wild edible mushrooms. **Food Chemistry**, 157: 323-331, 2014.

PARADA M., RODRÍGUEZ-BLANCO A., MAGÁN A.F.F., DOMÍNGUEZ H. Sequential extraction of *Hericium erinaceus* using green solvents, LWT – **FoodSci. Technol**. 64: 397–404, 2015.

QIU Y., LIN G., LIU W., ZHANG F., LINHARDT R.J., WANG X., ZHANG A. Bioactive compounds in *Hericium erinaceus* and their biological properties: a review. **Food**

Science and Human Wellness 13:1825-1844, 2024.

ROBINSON L., TANG E., TAYLOR J.P. Dementia: timely diagnosis and early intervention. **British Medical Journal**. 350: article h3029, 2015.

RODA E., PRIOR E.C., RATTO D. Neuroprotective metabolites of *Hericium erinaceus* promote neuro-healthy aging. **Int. J. Mol. Sci.** 22(12): 6379. https://doi.org/10.3390/ijms22126379, 2021.

RUFINO M.S.M., ALVES R.E., BRITO E.S., MORAIS S.M., SAMPAIO C.G., PÉREZ-JIMÉNEZ J., SAURA-CALIXTO F.D. Metodologia Científica: Determinação da Atividade Antioxidante Total em Frutas pela Captura do Radical Livre ABTS.+. **Embrapa Agroindústria Tropical**. Comunicado técnico 128 Embrapa 128: 1-6, 2007.

SHIROKIKH I.G., POLEZHAEV T.V., SHIROKIKH A.A., KHUDYAKOV A.N., SERGUSHKINA M.I., NAZAROVA J.I., PATUROVA I.G. Cryoprotective Properties of the Polysaccharide Fraction of the Mushroom *Hericium erinaceus* BP 16. **Biology Bulletin** 47(1): 1–6, 2020.

SOMOGY M. A new reagente for determination of sugars. **A new Sugar Reagent**, may, p.61-68, 1945.

SUN Y., HE H., WANG Q., YANG X., JIANG S., WANG D. A Review of Development and Utilization for Edible Fungal Polysaccharides: Extraction, Chemical Characteristics, and Bioactivities. **Polymers** 14:4454-4490, 2022.

SWAIN R., HILLIS W. E. The phenolic constituents of Prunus domestica. I. The quantitative analysis of phenolic constituents. **Journal of the Science of Food and Agriculture**, Oxford. 10: 63-68, 1959.

THONGBAI B., RAPIOR S., HYDE K.D., WITTSTEIN K., STADLER M. *Hericium erinaceus*, an amazing medicinal mushroom. **Mycological Progress**. 14:91. https://doi.org/10.1007/s1155 7-015-1105-4, 2015.

VAN HOOF A.; LEYMAM J.; SCHEFFER H. J.; WALTON J. D. A single beta-1,3-glucanase secreted by the maize pathogen *C. carbonum* acts by an exolytic mechanism. **Physiological and Molecular Plant Pathology** 39:259-267, 1991.

WADT N.S.Y., OKAMOTO M.K.H., BACH HI E.M., BACH E.E. Chemical, toxicological, anti-inflammatory and antimicrobial evaluation of *Ganoderma lucidum* extracts. **Emirates Journal of Food and Agriculture**. 27(7): 577-584, 2015.

WANG X.Y., ZHANG D.D., YIN, J.Y., NIE S.P.; XIE M.Y. Recent developments in *Hericium erinaceus* polysaccharides: extraction, purification, structural characteristics and biological activities. **Crit. Rev. Food Sci. Nutr.** 59:S96-S115. https://doi.org/10.1080/10408398.2018.1521370, 2019.

WOJEWODA W. **Encyklopedia biologiczna**, Opress, Kraków pp. 162–163, 1998.

Xu X., Yan H., Chen J., Zhang X. Bioactive proteins from mushrooms. **Biotechnology advances** 29 (6):667-674, 2011.

YEMM E. W., WILLIS A. J. The estimation of carbohydrates in plant extracts by anthrone. **Biochemical Journal**. 57:508-514, 1954.

YOUNIS A. Anticancer potential of *Hericium erinaceus* extracts against particular human cancer cell lines. **Microbial Biosystems** 2 (1):9–20, 2017.

ZAN X., CUI F., LI Y., YANG Y., WU D., SUN W., PING L. *Hericium erinaceus* polysaccharide-protein HEG-5 inhibits SGC-7901 cell growth via cell cycle arrest and apoptosis. **International Journal of Biological Macromolecules**. 76:242–53, 2015.

ZHANG A.Q., FU L., SUN P.L., ZHANG J.S., PAN Y.J. Isolation and structural characterization of a water-soluble polysaccharide from *Hericium erinaceus*.

Mycosystema 29:911–917, 2010.

ZHANG C., LI J., HU C., WANG J., ZHANG J., REN Z., SONG X., JIA L. Antihyperglycaemic and organic protective effects on pancreas, liver and kidney by polysaccharides from *Hericium erinaceus* SG-02 in streptozotocin-induced diabetic mice. **Scientific Reports** 7 (1): 10847–60, 2017.

ZHU F., DU B., BIAN Z., XU B. 2015. Beta-glucans from edible and medicinal mushrooms: Characteristics, physicochemical and biological activities. **Journal of Food Composition and Analysis** 41:165-173, 2015.

ZHU Y., LI Q., MAO G.H., ZOU Y., FENG W.W., ZHENG D.H., WANG W., ZHOU L.L., ZHANG T.X.,YANG J., YANG L.Q., WU X.Y. Optimization of enzyme-assisted extraction and characterization of polysaccharides from *Hericium erinaceus*. **Carbohydr. Polym**. 101:606–613, 2014.