JMBFS
Journal of Microbiology, Biotechnology and Food Sciences
International peer-reviewed scientific online journal
Published by Faculty of Biotechnology and Food Sciences

BIOLOGICAL ACTIVITY OF PSILOCYBIN EXTRACTS FROM THE FRUITING BODIES OF *PSILOCYBE CUBENSIS* ANTIOXIDANT AND ANTIMICROBIAL POTENTIAL

Shimal Y. Abdul-Hadi[1][1], Noor Alaubidi*[1], Walla H. Shuker[2], Sanabel[1] A. Abd Almajeed*

Address(es): Noor Alaubidi,
[1] Department of Biology, College of Education for Pure Sciences, University of Mosul, Mosul, Iraq.
[2] Department of sciences. College of Basic Education. University of Mosul, Mosul, Iraq.

*Corresponding author: noorameeralaubidi@uomosul.edu.iq

https://doi.org/10.55251/jmbfs.12097

ARTICLE INFO

Received 23. 11. 2024
Revised 4. 3. 2025
Accepted 13. 3. 2025
Published xx.xx.201x

Regular article

OPEN ACCESS

ABSTRACT

Several fruiting bodies from Mosul forests were collected and were diagnosed using the Polymerase Chain Technique based on ITS region. It was proved they belong to Psilocybe cubensis mushroom, and was registered in the GenBank, NCBI in the serial number OR140556.1. The fruiting body powder was used in order to extract the alkaloid compound "Psilocybe", which was analyzed using HPLC technique. As a result, a single peak for this compound appeared, at a retention time of 19.73 which is very close to the standard compound's retention time of 19.98. Also, alkaloid compound "Psilocybe" showed the antioxidant activity by using DPPH test, and it was found out that "Psilocybe" exhibits antioxidant activity of 40% at a concentration of 40 µg/ml with a significant value compared to the standard antioxidant "Ascorbic acid". In addition, the compound "Psilocybe" had activity against different types of pathogenic bacteria, and the highest diameter of the growth inhibitor towards Escherichia coli bacteria at a concentration of 50 mg/ml and reached 25.50 mm.

Keywords: *Psilocybe cubensis*, Psilocybe, Antioxidant, Pathogenic Bacteria, Molecular diagnosis,HPLC analysis

INTRODUCTION

Norbaeocystin, Aeruginascin, Psilocin, Baeocystin, and Psilocybin are considered similar compounds to the Tryptamines compound, which is known as pharmacologically active substance, and it is used in curing some psychological diseases, **(Ziff *et al.*, 2022, Yuan *et al.*, 2024).** Psilocybe mushroom is called the hallucination or magical mushroom, and it is part of Strophariaceae family, which belong to basidiomycota mushroom (**Sommano *et al.*, 2022; Gotvaldova *et al.*, 2021**).
Psilocybe mushroom has long been used since the ancient times due to its therapeutic characteristics. The Psilocybin compound (Phosphoryloxy-N, N-dimethyltry ptamine) has recently been discovered in the Psilocybe (**Serreau *et al.*, 2022; Goff *et al.*,2024**). This mushroom is considered one of the non-toxic types, and the dose used for treatment ranges between (3.5.1) gram of the dried mushroom. Between 10-15 grams of fresh mushrooms (**Dinis-Oliveira, 2017**). Based on research, no cases of poisoning from mushroom consumption have been proven, but toxicity may occur if combined with other medications. Generally, no substance is free from side effects, and consuming this mushroom increases heart rate and blood pressure **(Archer *et al.*, 2017)**. Despite this, the mushroom holds significant medical benefits, used to treat various organic and psychiatric diseases (**Prochazkova *et al.*, 2018**; **Lowe *et al.*, 2021**). Chemically, numerous analytical studies focused on its composition reveal Indole alkaloid compounds (**Nkadimeng, et al., 2020a**) that play a crucial medical role. Among these alkaloids is Psilocybin with the chemical structure (C12H17N204P), alongside Psilocin and terpenoid and phenolic compounds (**Van Amsterdam *et al.*, 2011**). Iraq's environment is rich in diverse types of these large fungi found in agricultural fields and forests (**Al-Khesraji and Suliaman, 2019**). Our survey study represents the first of its kind in Iraq, identifying the presence of Psilocybe cubensis mushrooms, highlighting the active role of alkaloid compounds as antimicrobials and antioxidants. The current study aimed to extract active Psilocybin from dried fruiting bodies powder, identified by high-performance liquid chromatography (HPLC), evaluating its antioxidant and antibacterial activities (**Nkadimeng *et al.*, 2020b**).

MATERIAL AND METHODS

Sample collection

During a scientific field trip at the forests of Mosul, located on the banks of the Tigris River, on 22/03/2023, fruiting bodies of different sizes were collected. These fruiting bodies belong to one type of mushroom that were spread on the grass and the fallen leaves of the forest shown in Figure 1

Figure 1 Fruiting body for *Psilocybe cubensis*

DNA extraction

The fungal isolate was activated on the obtained PDA medium, and the DNA was extracted from the fungal hyphae using the extraction kit prepared by the company (Geneaid). The same steps according to the company special protocol were followed in the amplification of the DNA, which includes denaturation, annealing and finishing processes. The result of the DNA amplification was sent to the Korean company "Macrogene". A month later, the nucleotide sequences from the Korean company were received and were compared with reference sequences stored in the global NCBI database website (National center for Biotechnology information). Then, an alignment of the nucleotide sequence was conducted to get an accurate diagnosis of the isolate in terms of species level as well as the degree of conformity, in addition to comparing the isolate with other diagnosed and registered reference isolates with serial numbers in the GenBank. Based on the received nucleotide sequence from the mentioned company, which gives the gene ITS1 (TCCGTAGGTGAACCTGCGG), ITS4(TCCTCCGTTATTGATATGC) Primers both forward and reverse types, selected to avoid primer dimers and serve as an initiation sites for elongation, the genetic tree was drawn using Mega 6 program (**Abdul hadi *et al.*, 2020**).

Psilocybin extraction

10 gram of the fruiting body powder was taken and put into a 250 ml glass beaker. Then, 100 ml (10%) of Acetic acid was added. After that, the mixture was subjected to ultrasound for 10 minutes (**Fernandes,*et al*.,2022**), and the mixture was placed in a centrifuge device at a rotation speed of 6000 round per minute for 10 minutes. The supernatant layer was taken and equalized with sodium bicarbonate. In addition, the mixture was poured into a separating funnel, and chloroform with the same value was added in order to release the alkaloid compounds. The organic layer was added taken and concentrated by using Rotary Evaporator at a temperature of 60 °C. The obtained extract was kept in opaque glass bottles in the refrigerator to be diagnosed (**Sarwar and Mcdonald, 2003**).

Psilocybin diagnosis extracted from the fungal isolate using High-performance liquid chromatography (HPLC) technique

The alkaloid compound "Psilocybin" that is extracted from the fungal isolate was diagnosed using HPLC technique and by using C_{18} column, in which 1 microliter of the extract was injected at a flow speed rate of 1 ml per minute that represent the mobile phase (79:2:0.1) CAN: Glacial acetic: Triethylamine, thus, the response was shown by measuring the absorbance at a wave-length of 284 nanometer.

Biological antioxidant activity

The biological antioxidant activity was estimated based on the method suggested by (**Da Silva,*et al*.,2024**) for the Psilocybe compound that is purified from *Psilocybe cubensis* mushroom by capturing the free radicals of 2.2-diphenyl-1-picrylhydrazyl (DPPH). Also, the Ascorbic acid (vitamin C) was used as a standard antioxidant.

Biological activity of Psilocybin compound against some pathogenic bacterial types to humans

In order to determine the effect of Psilocybin extract in some types of pathogenic bacteria, an inhibitory activity test of Psilocybin extract was conducted on four pathogenic bacterial types. Two of which were gram- negative: *Salmonella typhi, Escherichia coli,* and the other two were gram positive: *Bacillus cereus, Staphylococcus aureus.*(The source of the bacteria from Dr.Walid Ahmed,Molecular Biology Laboratory,Plant Protection Department,College of Agriculture Tikrit University) Based on the agar well diffusion method to examine the inhibitory activity of Psilocybin compound, and in concentration of 50 mg/ml, on the pathogenic bacteria growth inside the culture medium cabinet under sterile conditions, sterilized petri dishes containing 20 ml Mueller- Hinton medium were prepared. Also, a 6 mm cork borer was used after sterilizing it with alcohol flaming, three holes in the dish was made in which the distance was equal among the holes (**Hidayathulla,*et al*.,2018**). Moreover, by using cotton swab, the bacterial suspension of 0.1 ml, aged 18 hours, which is growing on N.B medium, by wiping the dish surface with wiping motion of one direction. By using the micropipette, 40 microliter of Psilocybin extract was taken and put in the holes. Also, Gentamycin was used at a concentration of 10 µg/ml as a positive control. The dishes were left for 15 minutes in the culture cabinet to stabilize, and was incubated at a temperature of 37 °C. After incubation for 24 hours, the inhibition diameters around the holes were measured (**Karthiyayini,*et al*., 2024**).

RESULTS AND DISSCUSION

Molecular diagnosis of fungal

After collecting fruiting bodies and cultivating them on the PDA medium, pure colonies were obtained and DNA was extracted from fungal hyphae of the pure colonies. The amplification results revealed one distinct and clear band, and within the amplification range, in which the molecular size reached 650 base pairs in comparison with the marker ladder shown in Figure 2. The results were in conformity with the special data for the primer design of the free program in the International Center of Biotechnology Information (NCBI) multi- purpose website. One of the important principles to the success of the process is mainly relying on the purity and quality of the extracted DNA, in addition to the absence of the DNA polymerase inhibitors. This ensures that PCR targets the DNA piece. Some of the indicators that confirm the reaction is the time period and temperature, and components' concentration of the reaction mixture.

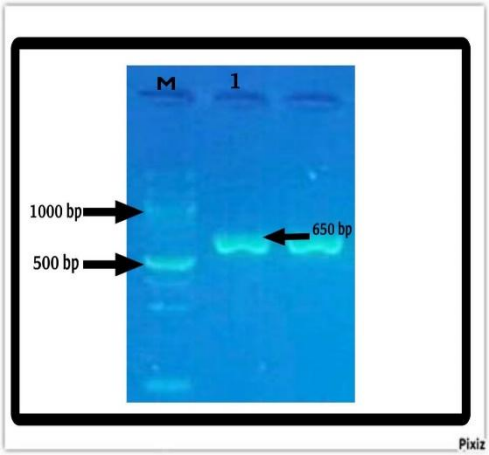

Figure 2 DNA amplification of the fungal isolate on agarose gel Path M represents: size marker, 1-2 DNA amplification of fungal isolate

Based on a study conducted by the scientist(Alexander *et al*., 2022), 18 species-type that belong to Psilocybe genus were identified by using PCR. In another study by (Dörner *et al*., 2022), it was possible to identify five species- type that belong to Psilocybe genus based on rDNA and amplifying the gene region ITS.

The nucleotide sequence of fungal isolate

The nucleotide sequences were identified by using the Sequencing technique for the result of the DNA amplification after sending them to the Korean company "Macrogen", in which the results were sent as a Fasta file for the DNA bundle as shown in Figure 3.

```
CGTGGTTGTAGCTGGCCCTCTCGGCGGCATGTGCTCGCC<
CGTCATCTTTATATTTCCACCTGTGCACTTTTTGTAGATC
ATTGTTATTGGAAGCTGGATTGAAGTCAGAGATTACTCTC
TGATGAATTGAAGGCTTTCTCAATGATGGTCTACGTTTTC
ATATACTCCAATGAATGTAACAGAATGTATCTATATGGCC
TTGTGCCTATAAAACAATATACAACTTTCAGCAACGGATC
TCTTGGCTCTCGCATCGATGAAGAACGCAGCGAAATGCGA
TAAGTAATGTGAATTGCAGAATTCAGTGAATCATCGAATC
TTTGAACGCACCTTGCGCTCCTTGGTATTCCGAGGAGCAT
GCCTGTTTGAGTGTCATTAAATTCTCAACCTTACCAGCTT
TTGTTAGCTTGTGTAATGGCTTGGACTTGGGGGTTTATTT
TGCCGGCTTCTTACCAAGTCAGCTCCCCTTAAATGCATTA
GCCGGCTGCCCGCTGTGGACCGTCTATTGGTGTGATAATT
ATCTACGCCGTGGATGTCTACTATTAATGGGTTGAAGCTG
CTTCAAACCGTCTGTTTACTCAGACAATTAATGACAATTT
GACCTCAAATCAGGTAGGACTACCCGCTGAACTTAAGCAT
                                ATCAATAAGGCGGA
```

Figure 3 The nucleotide sequence of fungal isolate

The sequence analysis of the nucleotide sequences for the *Psilocybe cubensis* genus was conducted via Blast program which belongs to (www.ncbi.gov/Blast//:http). Also, by using the copy/ paste feature to the nucleotide sequence which is used by the Korean company, *Psilocybe cubensis* type was registered for the first time as an Iraqi local isolate in the GenBank in the serial number OR140556.1 as shown in Figure 4 and Table 1.

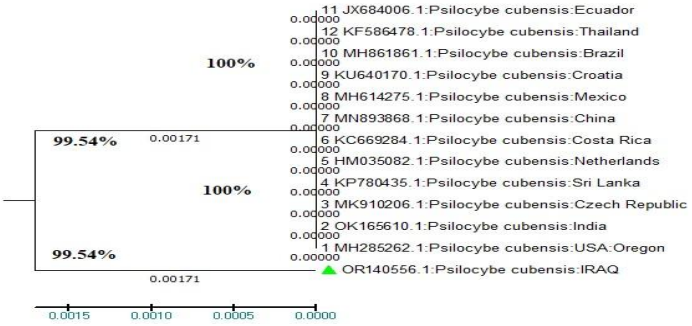

Figure 4 Genetic tree of *Psilocybe cubensis* mushroom, and other global refernce types

Table 1 Shows the percentage of the genetic similarity of *Psilocybe cubensis* mushroom with other global types

	Source: *Psilocybe cubensis*			
	Accession	**Country**	**Isolation Source**	**Compatibility**
1.	ID: MH285262.1	USA:Oregon	Jaime Cultivated Portland, Oregon	99%
2.	ID: OK165610.1	India	Mushroom	99%
3.	ID: MK910206.1	Czech Republic	Sporocarp	99%
4.	ID: KP780435.1	Sri Lanka	----------	99%
5.	ID: HM035082.1	Netherlands	----------	99%
6.	ID: KC669284.1	Costa Rica	----------	99%
7.	ID: MN893868.1	China	----------	99%
8.	ID: MH614275.1	Mexico	On cow manure	99%
9.	ID: KU640170.1	Croatia	----------	99%
10	ID: MH861861.1	Brazil	----------	99%
11	ID: JX684006.1	Ecuador	forest floor-saprophyte	99%
12	ID: KF586478.1	Thailand	----------	99%

In regard to this context, previous research was able to identify several types belong to Psilocybe genus. There wereTow new *Psilocybe* species ,*P.maluti* and *P.ingeli*,are described from southern Africa (**Van *et al.*, 2024**). It was based on the genetic tree results among the homogenous species that have the same serial number specified for the gene (1MH285262.1).

Identifying Psilocybin compound by HPLC for the fungal isolate

The alkaloid compound "Psilocybin" that is extracted from the fruiting body of the fungal isolate "*Psilocybe cubensis"* was diagnosed by HPLC technique in terms of quality and quantity. The diagnosis is conducted via comparing Chromatograms resulted from the analysis by this technique for the extract "Psilocybin" with the standard compound imported from Sigma. In order to conduct the qualitative analysis of this compound as a model, and to verify it is the same as the standard compound, the detention time has to be identical for both of the standard substance and the model extracted. Figure 5 shows the appearance of one peak for the standard compound at a retention time of 19.98 which was close to the retention time of Psilocybin compound extracted from the fungus at a retention time of 19.73. This ensures that the extracted substance is very pure because there is no other materials or substances that interferes with the model bundle, and it is free of impurities.

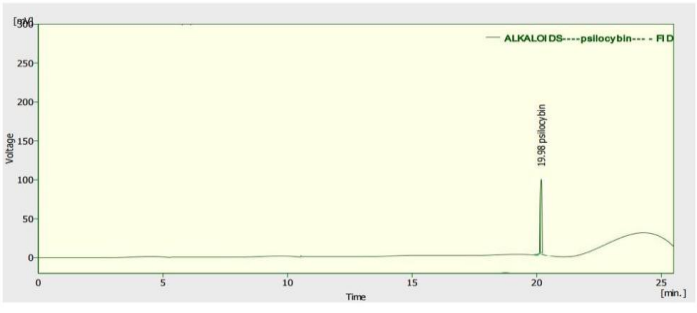

Figure 5 HPLC of the standard compound Psilocybin

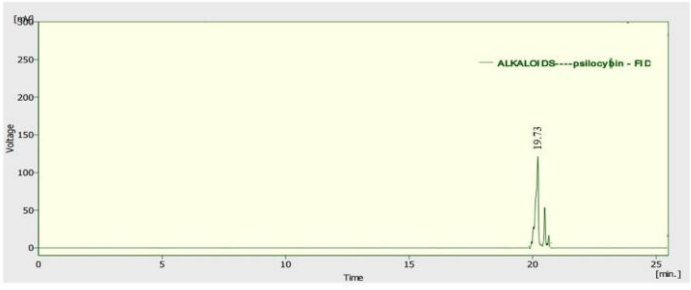

Figure 6 HPLC of the Psilocybin compound extracted and purified form the fungal isolate *Psilocybe cubensis*

In a study conducted by (**Samuelsson *et al.*, 2021**), Psilocybin compound was identified from the fungal isolate Psilocybe Mexicana, and one peak has appeared at a retention time of 21.8.

Evaluation of the biological antioxidant activity of Psilocybin compound extracted from the fungal isolate "Psilocybe cubensis"

Figure 7 illustrates that Psilocybin has antioxidant activity by DPPH test, and according to the used concentrations. The lowest concentration at a 12.5 mircrogram/ ml gave antioxidant activity that reached 13% compared with the standard Ascorbic acid which gave 21% in the same concentration. There was a gradual increase in the antioxidant activity that reached 39% at a concentration of 50 µg/ml, a significant difference than the standard antioxidant 45%. Whereas the increase was not significant for both concentrations 100 and 200 µg/ml. In the last few years, there was an increased interest in the free radicals, and the possibility of getting some diseases. These free radicals have a vital role as a mediator or a stimulant for their occurrence. For the previous results, it was noted that the Psilocybin compound in rich is electrons which can provide electrons to the free radicals, which are in an unbalanced state. Therefore, this compound works as electrons donor, which can neutralize free radicals and recreate balance again, and this process is called "oxidation". In this regard, it is worthy to refer the findings of (**Nkadimeng *et al.*, 2020**) as he mentioned that Psilocybin compound extracted from Psilocybe natalensis has antioxidant activity with the two concentrations (50, 100) µg/ml and gave (40%, 80%) respectively.

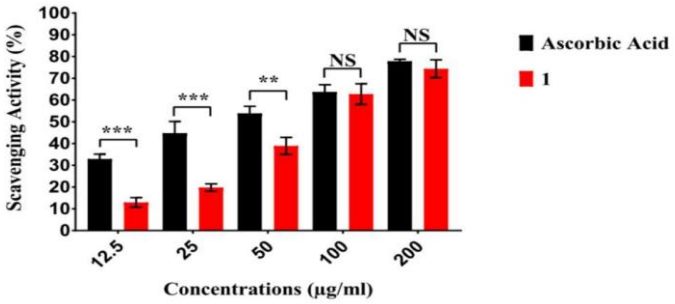

Figure 7 Biological antioxidant activity of Psilocybin compound extracted from the fungal isolate *Psilocybe cubensis* via DPPH

Biological activity of Psilocybin against some types of pathogenic bacteria

Table (2) and Figure (8) illustrate that Psilocybin extract in a concentration of 50 mg/ ml has the inhibitory activity for the growth of laboratory bacterial species that is under experiment with varying in effectiveness in which it gave the highest inhibition diameter towards bacteria *E. coli* which reached 25.50 ml. This has outperformed the commercial antioxidant which gave inhibitory activity of 14.50 ml and the inhibitory diameter towards bacteria *B. cereus* reached 22.50 ml, in addition to both *S. aureus*, and *S. typhi* bacteria, in which the diameter inhibitory reached 20.50, 18.50 respectively.

Table 2 Activity of Psilocybin extracted from *Psilocybe cubensis* against some types of pathogenic bacteria to humans

Negative control	Inhibitory region (ml) (50 mg/ml)	Inhibitory region (ml) Gentomycin	Bacteria type
-	20.50	18.00	*S.typhimurium*
-	18.50	9.50	*S.aureus*
-	22.50	19.00	*B.cereus*
-	25.50	14.50	*E.coli*

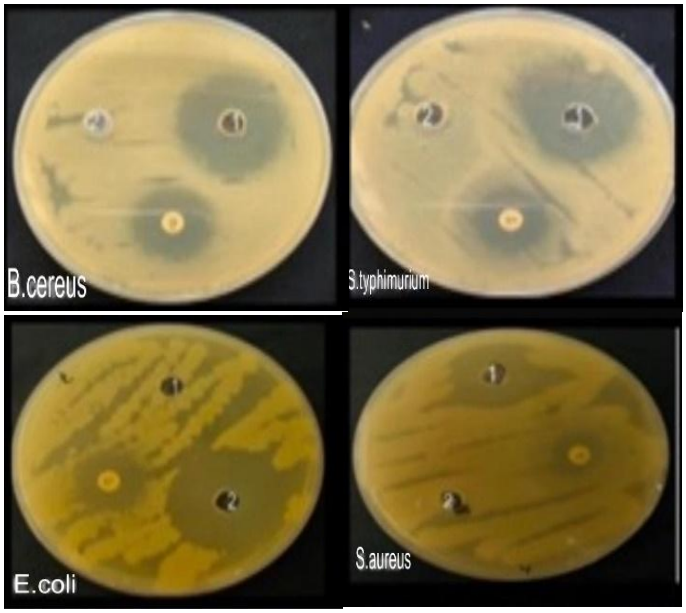

Figure 8 Inhibitory region of Psilocyben extract (ml) from *Psilocybe cubensis* fungus

Recent studies have proved that Psilocybin extract has anti-bacterial and anti-virus activity that are pathogenic to humans, and as well as in various types of cancer treatment (**Khan *et al*., 2022**). Researcher **Kelly *et al*. (2023)** have also stated that Psilocybin extract have inhibitory effect on the growth of different types of pathogenic bacteria to humans particularly towards *E. coli* bacteria.

CONCLUSION

Results of current research state that *Psilocybe cubensis* possesses a very important base compound named Psilocybin extracted using HPLC technique. this compound is a very important source of anti-oxidant as well as antibiologocal effect on many sickening bacteria.

Acknowledgments: This research is supported by laboratories of College of Education for Pure sciences / Biology department, many thanks for Dr. Waleed Ahmed from department of preserving plants / College of Agriculture and Forestery / University of Tikrit

Conflict of interest: The authors declare no conflicts of interest associated with this manuscript

REFERENCES

Abdul-Hadi, S. Y., Owaid, M. N., Rabeea, M. A., Aziz, A. A., & Jameel, M. S. (2020). Rapid mycosynthesis and characterization of phenols-capped crystal gold nanoparticles from *Ganoderma applanatum*, Ganodermataceae. *Biocatalysis and Agricultural Biotechnology*, ***27***,101683. https://doi.org/10.1016/j.bcab.2020.101683

Al-Khesraji, T. O., & Suliaman, S. Q. (2019). New taxa records for macromycota of Iraq from Salahadin Governorate. *Journal of Research on the Lepidoptera*, ***50***(3),125-35. https://doi.org/10.36872/lepi/v50i3/201032

Archer, C. R., Robinson, E. L., Drawnel, F. M., & Roderick, H. L. (2017). Endothelin-1 promotes hypertrophic remodelling of cardiac myocytes by activating sustained signalling and transcription downstream of endothelin type A receptors. *Cellular signalling*, ***36***,240-254. https://doi.org/10.1016/j.cellsig.2017.04.010

Bradshaw, A. J., Backman, T. A., Ramírez-Cruz, V., Forrister, D. L., Winter, J. M., Guzmán-Dávalos, L., Furci, G., Stamets, P., & Dentinger, B. T. (2022). DNA authentication and chemical analysis of Psilocybe mushrooms reveal widespread misdeterminations in Fungaria and inconsistencies in metabolites. *Applied and Environmental Microbiology*, ***88***(24), e0149822. https://doi.org/10.1128/aem.01498-22

Da Silva, D. F., Cunha, L. S., de Menezes Filho, A. C. P., Melo, A. F., Sharma, P., Machado, T. H. L., ... & da Rocha, E. N. (2024). Phytochemical screening, phenolic and flavonoid contents, psilocybin, antioxidant, and acetylcholinesterase inhibition activities of the aqueous extract from the fungi Cyathus striatus, Laternea dringii, and Marasmius haematocephalus. Brazilian Journal of Science, 3(11), 39-50. https://doi.org/10.14295/bjs.v3i11.694

Dinis-Oliveira, R. J. (2017). Metabolism of psilocybin and psilocin: clinical and forensic toxicological relevance. *Drug metabolism reviews*, ***49***(1), 84-91. https://doi.org/10.1080/03602532.2016.1278228

Dörner, S., Rogge, K., Fricke, J., Schäfer, T., Wurlitzer, J. M., Gressler, M., Pham, D. N., Manke, D. R., Chadeayne, A. R., & Hoffmeister, D. (2022). Genetic survey of Psilocybe natural products. *Chembiochem*, ***23***(14), e202200249. https://doi.org/10.1002/cbic.202200249

Fernandes, J. M. C., Brito-da-Costa, A. M., Marin-Bruzos, M., Saayman, J., Sanders, D., & Dinis-Oliveira, R. J. (2022). Production and extraction of psilocybin and psilocin from Psilocybe spp. mushrooms. RevSALUS-Revista Científica Internacional da Rede Académica das Ciências da Saúde da Lusofonia, 4(Sup), 219-219. https://doi.org/10.51126/revsalus.v4iSup.456

Goff, R., Smith, M., Islam, S., Sisley, S., Ferguson, J., Kuzdzal, S., ... & Schug, K. A. (2024). Determination of psilocybin and psilocin content in multiple Psilocybe cubensis mushroom strains using liquid chromatography–tandem mass spectrometry. Analytica Chimica Acta, 1288, 342161. https://doi.org/10.1016/j.aca.2023.342161

Gotvaldová, K., Hájková, K., Borovička, J., Jurok, R., Cihlářová, P., & Kuchař, M. (2021). Stability of psilocybin and its four analogs in the biomass of the psychotropic mushroom *Psilocybe cubensis*. *Drug testing and analysis* ,***13***(2),439-446. https://doi.org/10.1002/dta.2950

Hidayathulla, S., Shahat, A. A., Alsaid, M. S., & Al-Mishari, A. A. (2018). Optimization of physicochemical parameters of tannase post-purification and its versatile bioactivity. FEMS Microbiology Letters, 365(12), fny051. https://doi.org/10.1093/femsle/fny051

Karthiyayini, B., Kalyani, N. N., Gowdhami, B., Muthuselvam, M., & Dharumadurai, D. (2024). Characterization and Identification of an Antimicrobial Compound Psilocybin from Psychedelic Mushroom. Indian Journal of Microbiology, 1-13 https://doi.org/10.1007/s12088-024-01396-2

Kelly, J. R., Clarke, G., Harkin, A., Corr, S. C., Galvin, S., Pradeep, V., Cryan, J. F., O'Keane, V., & Dinan, T. G. (2023). Seeking the Psilocybiome: Psychedelics meet the microbiota-gut-brain axis. *International Journal of Clinical and Health Psychology*,***23***(2),100349. https://doi.org/10.1016/j.ijchp.2022.100349.

Khan, F. I., Hassan, F., & Lai, D. (2022). In silico studies on psilocybin drug derivatives against SARS-CoV-2 and cytokine storm of human interleukin-6 receptor. *Frontiers in Immunology*,***12***,794780. https://doi.org/10.3389/fimmu.2021.794780

Lowe, H., Toyang, N., Steele, B., Valentine, H., Grant, J., Ali, A., Ngwa, W., & Gordon, L. (2021). The therapeutic potential of psilocybin. *Molecules*, ***26***(10), 2948. https://doi.org/10.3390/molecules26102948

Nkadimeng, S. M., Nabatanzi, A., Steinmann, C. M., & Eloff, J. N. (2020a). Phytochemical, cytotoxicity, antioxidant and anti-inflammatory effects of *Psilocybe natalensis* magic mushroom. *Plants*, ***9***(9),1127. https://doi.org/10.3390/plants9091127

Nkadimeng, S. M., Steinmann, C. M., & Eloff, J. N. (2020b). Effects and safety of *Psilocybe cubensis* and *Panaeolus cyanescens* magic mushroom extracts on endothelin-1-induced hypertrophy and cell injury in cardiomyocytes. *Scientific Reports*, ***10***(1), 22314. https://doi.org/10.1038/s41598-020-79328-5

Prochazkova, L., Lippelt, D. P., Colzato, L. S., Kuchar, M., Sjoerds, Z., & Hommel, B. (2018). Exploring the effect of microdosing psychedelics on creativity in an open-label natural setting. *Psychopharmacology*, ***235***,3401-3413. https://doi.org/10.1007/s00213-018-5049-7

Samuelsson, A., Janusson, E., Shah, S., & Roggen, M. (2021). Rapid quantification of Psilocybin with reversed-phase HPLC and single-wavelength detection. Available from https://chemrxiv.org/engage/chemrxiv/article-details/617755bc913a74cab06a8a2d

Sarwar, M., & McDonald, J. L. (2003). Technical note: A rapid extraction and GC/MS methodology for the identification of psilocyn in mushroom/chocolate concoctions. *Microgram J.*,***1***(3-4), 177-183.

Serreau, R., Amirouche, A., Benyamina, A., & Berteina-Raboin, S. (2022). A Review of synthetic access to therapeutic compounds extracted from Psilocybe. Pharmaceuticals, ***16***(1),40. https://doi.org/10.3390/ph16010040

Sommano, S. R., Suksathan, R., Sombat, T., Seehanam, P., Sirilun, S., Ruksiriwanich, W., Wangtueai, S., & Leksawasdi, N. (2022). Novel perspective of medicinal mushroom cultivations: A Review case for 'magic' mushrooms. *Agronomy*, ***12***(12),3185. https://doi.org/10.3390/agronomy12123185

Sulkowska-Ziaja, K., Trepa, M., Olechowska-Jarząb, A., Nowak, P., Ziaja, M., Kała, K., & Muszyńska, B. (2023). Natural compounds of fungal origin with antimicrobial activity-Potential cosmetics applications. *Pharmaceuticals*, ***16***(9), 1200. https://doi.org/10.3390/ph16091200

Van Amsterdam, J., Opperhuizen, A., & van den Brink, W. (2011). Harm potential of magic mushroom use: a review. *Regulatory toxicology and pharmacology*, ***59***(3), 423-429. https://doi.org/10.1016/j.yrtph.2011.01.006

Van Der Merwe, B., Rockefeller, A., Kilian, A., Clark, C., Sethathi, M., Moult, T., & Jacobs, K. (2024). A description of two novel Psilocybe species from southern Africa and some notes on African traditional hallucinogenic mushroom use. Mycologia, 116(5), 821-834. https://doi.org/10.1080/00275514.2024.2363137

Yao, Y., Guo, D., Lu, T. S., Liu, F. L., Huang, S. H., Diao, M. Q., ... & Han, Y. (2024). Efficacy and safety of psychedelics for the treatment of mental disorders: A systematic review and meta-analysis. Psychiatry Research, 115886. https://doi.org/10.1016/j.psychres.2024.115886

Ziff, S., Stern, B., Lewis, G., Majeed, M., & Gorantla, V. R. (2022). Analysis of Psilocybin-Assisted Therapy in Medicine: A Narrative Review. *Cureus*, ***14***(2). https://doi.org/10.7759/cureus.21944