













Biological efficiency and evaluation of bioactive compounds of wild mexican strains of *Hericium erinaceus*



Eficiencia biológica y evaluación de los compuestos bioactivos de cepas silvestres mexicanas de *Hericium erinaceus*

Eficiência biológica e avaliação de compostos bioativos de cepas selvagens mexicanas de *Hericium erinaceus*

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Crop production

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Abstract

The basidiomycete *Hericium erinaceus* is one of the most consumed edibles and medicinal mushrooms in the world and appreciated in traditional Chinese medicine. In Mexico it is known as lion's mane. The biological efficiency of wild strains of *H. erinaceus* cultivated in different substrates in the Durango region, mainly agave bagasse, a waste from mezcal production, was evaluated. The CCH003 strain from Espinazo del diablo showed the highest biological efficiency of 42.33 % and a productivity rate of 0.47 %, with a total cultivation period of 90 days and three harvests. Regarding the evaluation of bioactive compounds, the same strain obtained significant differences compared to the others, it presented the highest values for all determinations; 60 ± 0.018 mg EAG.g ES⁻¹ in phenolic content, 4.21 ± 0.013 mg EQ.g ES⁻¹ for flavonoids, 71.16 ± 0.002 mg EAA.g ES⁻¹ in CAT, 0.0012 ± 0.001 mg AA.g ES⁻¹ for by ABTS and 121 ± 0.107 μg EAG.mL⁻¹ by DPPH. The variability of the results in the tests carried out provides information on how the type of substrate, climatological and geographical conditions and stage of maturity influence the development of the fungus, including the production of secondary metabolites, even if it is the same species. It is expected that this information will be useful to promote the use of agave bagasse as a substrate in the cultivation of *H. erinaceus* and thereby diversify rural activities in the region, and in the future generate new studies on the effect of conditions on the production of bioactive compounds.

Resumen

El basidiomiceto *Hericium erinaceus* es uno de los hongos comestibles y medicinales más consumido en el mundo y apreciado en la medicina tradicional China, en México es conocido como melena de león. Se evaluó la eficiencia biológica de cepas silvestres de *H. erinaceus* cultivado en distintos sustratos de la región de Durango, principalmente bagazo de agave, un desecho de la producción de mezcal. La cepa CCH003 proveniente del Espinazo del diablo mostró la mayor eficiencia biológica de 42,33 % y una tasa de productividad de 0,47 %, con un periodo total de cultivo de 90 días y tres cosechas. Con respecto a la evaluación de los compuestos bioactivos, la misma cepa obtuvo diferencias significativas en comparación con las demás, presentó los mayores valores para todas las determinaciones; $60 \pm 0,018$ mg EAG.g ES⁻¹ en contenido fenólico, $4,21 \pm 0,013$ mg EQ.g ES⁻¹ para flavonoides, $71,16 \pm 0,002$ mg EAA.g ES⁻¹ en CAT, $157 \pm 0,089$ µg EAG.mL⁻¹ por ABTS y $121 \pm 0,107$ µg EAG.mL⁻¹ por DPPH. La variabilidad de los resultados en los ensayos realizados aporta información sobre cómo el tipo de sustrato, las condiciones climatológicas y geográficas, y el estado de madurez influyen el desarrollo del hongo incluyendo la producción de metabolitos secundarios, aun tratándose de la misma especie. Se espera que esta información sea útil para promover el aprovechamiento del bagazo de agave como sustrato en el cultivo de *H. erinaceus* y con ello, diversificar las actividades rurales de la región, y en un futuro generar nuevos estudios sobre el efecto de las condiciones en la producción de compuestos bioactivos.

Palabras clave: bagazo de agave, antioxidantes, hongos medicinales.

Resumo

O basidiomiceto *Hericium erinaceus* é um dos cogumelos comestíveis e medicinais mais consumidos no mundo e apreciado na medicina tradicional chinesa, sendo conhecido no México como juba de leão. Foi avaliada a eficiência biológica de cepas selvagens de *H. erinaceus* cultivadas em diferentes substratos na região de Durango, principalmente bagaço de agave, resíduo da produção de mezcal. A cepa CCH003 de Espinazo del diablo apresentou a maior eficiência biológica de 42,33 % e produtividade de 0,47 %, com período total de cultivo de 90 dias e três colheitas. Quanto à avaliação de compostos bioativos, a mesma cepa obteve diferenças significativas em relação às demais, apresentou os maiores valores para todas as determinações; $60 \pm 0,018$ mg EAG.g ES⁻¹ em conteúdo fenólico, $4,21 \pm 0,013$ mg EQ.g ES⁻¹ para flavonóides, $71,16 \pm 0,002$ mg EAA.g ES⁻¹ em CAT, $157 \pm 0,089$ µg EAG.mL⁻¹ pela ABTS e $121 \pm 0,107$ µg EAG.mL⁻¹ pela DPPH. A variabilidade dos resultados nos testes realizados fornece informações sobre como o tipo de substrato, as condições climatológicas e geográficas e o estágio de maturidade influenciam o desenvolvimento do fungo, incluindo a produção de metabolitos secundários, mesmo tratando-se da mesma espécie. Espera-se que estas informações sejam úteis para promover a utilização do bagaço de agave como substrato no cultivo de *H. erinaceus* e assim diversificar as atividades rurais na região, e futuramente gerar novos estudos sobre o efeito das condições na produção de compostos bioativos.

Palavras-chave: bagaço de agave, antioxidante, cogumelos medicinais.

Introduction

White rot fungi are basidiomycetes that can be cultivated on lignocellulosic substrates; within the cultivable fungi, there are edible genera such as fungi of the genus *Pleurotus* and also medicinal ones, such as *Hericium erinaceus* (Bull.) Pers., known in Mexico as lion's mane mushroom; it has been reported in Oaxaca, Chihuahua and Durango (Quiñónez-Martínez *et al.*, 2014; Páez-Olivan *et al.*, 2022). *H. erinaceus* was domesticated in Shanghai, and from there the cultivation technique was spread to other places, currently it is cultivated on a wide variety of substrates such as: oak sawdust, corn tazole and wheat straw, sometimes these substrates are supplemented with wheat bran, sucrose and gypsum (Sobieralski *et al.*, 2009). In China, 2,800 tons of *H. erinaceus* production were estimated for 1998 and 9,547 tons for 2001. It was also established that the developmental stage of the fungus influences its biological activity, and it was mentioned that there are differences in the composition of basidiomes with respect to the substrate that is administered to them (Nieto & Chegwin, 2010). This fungus is one of the most appreciated in traditional Chinese medicine, as it is attributed multiple properties thanks to the bioactive compounds it possesses (Kozarski *et al.*, 2015). In Mexico, there are studies on the evaluation of the biological efficiency and bioactive compounds of commercial strains of *H. erinaceus*, but not of wild strains; hence the importance of this work, whose objective was to evaluate the biological efficiency and the potential of bioactive compounds of *H. erinaceus* cultivated on different substrates in the Durango region, mainly on agave bagasse, which is a waste product of mezcal production. With the development of this research, we seek to diversify the productive activities of the forest and, at the same time, promote the cultivation of this fungus in Durango, Mexico.

Materials and methods

Collection and identification of biological material

Specimens were collected at sites belonging to the region of El Salto, Pueblo Nuevo, Durango, Mexico; La Peña (CCH005), Coscomate (CCH008), La Gallina (CCH007), Los Túneles (CCH001), Puenteillas (CCH002), Espinazo del diablo (CCH003) and Puenteillas M (CCH006). The collected specimens of *Hericium* spp. were herborized for preservation and deposited in the Mycological collection of the Herbario Micológico del Instituto de Botánica, Universidad de Guadalajara, Jalisco, México (IBUG) and the strains in the Laboratorio de Biotecnología de Hongos del CIIDIR-IPN-DGO (CLBH-CIIDIR-DGO). The taxonomic and molecular identification of the collected specimens is shown in the previous work published by Páez-Olivan *et al.* (2022).

Isolation of strains

Isolation of wild strains was carried out by cloning from live tissue samples (Stamets & Chilton, 1983). In culture medium malt agar and 1 % oak sawdust at acid pH (4.7), it was incubated for ten days at 24 ± 2 °C.

Cultivation of the fungus *Hericium erinaceus*

Preparation of inoculum and substrate

Sorghum seed was used for the inoculum, prepared according to the methodology proposed by Omarini *et al.* (2009) for each of the previously isolated strains. Subsequently, they were incubated for 21 days, in darkness, at a temperature of 23 ± 2 °C. Ten 5 kg (wet weight) bags of substrate were prepared for each treatment with a mixture of

oak sawdust, corn tazole and agave bagasse (previously washed and crushed to a size of 3 to 5 cm) at different concentrations. Treatments A, B and C were composed individually (100 %) of oak sawdust, corn stover and agave bagasse, respectively. Treatment D was a mixture of 50 % oak sawdust, 25 % corn tazole and 25 % agave bagasse, treatment E contained 25 % oak sawdust, 50 % corn tazole and 25 % agave bagasse. Treatment F was composed of 25 % oak sawdust, 25 % corn tazole and 50 % agave bagasse.

Seeding and incubation

The bags with each of the treatments were inoculated with 500 g of fungus inoculum and subsequently incubated at a temperature of 23 ± 2 °C. The bags with more than 80 % of the substrate colonized by mycelium were transferred to the production area.

Induction and harvesting

During the induction period, a temperature of 16 °C and a relative humidity of more than 80 % were maintained. Two automated sprinkler irrigations were carried out every 12 hours, the lighting conditions were 10 h.day⁻¹ using white light LED tubes (16 W).

Calculation of biological efficiency and productivity

The calculation of biological efficiency and total productivity was performed using the formulas published by Beelman *et al.* (2003), taking as total production period, between 60 and 90 days depending on the capacity of each strain. Data were analyzed with a one-way ANOVA and Tukey's test with 95 % reliability in GraphPad Prism V5 software, ©2014.

$$BE = \text{Biological efficiency} = \frac{\text{kg mushrooms (fresh weight)}}{\text{kg substrate (dry weight)}} \times 100$$

$$PT = \text{Total productivity} = \frac{\text{Biological efficiency}}{\text{Days of incubation} + \text{production}}$$

Preparation of ethanolic extracts of the wild *H. erinaceus* strains

Three hundred grams of dehydrated basidioma from each of the specimens from which the wild *H. erinaceus* strains used for the culture were obtained were macerated separately for five days with 1.5 L of solvent (ethanol). The ethanolic phase of each extract was recovered and concentrated in a rotary evaporator at a temperature of 30 °C (this process was repeated three times), the final concentrate was lyophilized for five days.

Quantification of bioactive compounds

Quantification of total phenols

Quantification of total phenols was performed using the Folin-Ciocalteu method (Nurmi *et al.* 1996). The reading was performed in a spectrophotometer (Thermo Scientific Genesys 105 UV-VIS), at a wavelength of 760 nm and a correlation coefficient $r = 0.9997$, the calibration curve of gallic acid (GA) (Sigma-Aldrich) with a purity of 99.9 %, was prepared from a solution with 0.01 mg and the content of total phenols was expressed in milligram equivalents of GA per gram of dry sample (DM) (mg EAG.g MS⁻¹).

Quantification of total flavonoids

Quantification of total flavonoids was performed by the technique proposed by Woisky & Salatino (1998). Using a quercetin standard curve constructed with aliquots from 1 to 140 µL.mL⁻¹ ($r = 0.9999$), readings were performed in a spectrophotometer (Thermo Scientific Genesys 105 UV-VIS), at an absorbance of 420 nm and a correlation coefficient $r = 0.9993$. Total flavonoid content was expressed as milligram quercetin equivalents (QE) per gram of dry sample (mg QE.g MS⁻¹).

Total antioxidant capacity

Total antioxidant capacity was determined using the phosphomolybdenum reagent following the technique proposed by Prieto & Aguilar (1999), the absorbance of each sample was measured at 695 nm ($r = 0.9999$). Ascorbic acid (AA, 1 mg.mL⁻¹) was used as a reference to construct the ascorbic acid calibration curve (Sigma-Aldrich) using six concentrations between 0.1 and 1 mg AA.mL⁻¹ with a correlation coefficient $r = 0.9921$. Sample results were expressed as mg ascorbic acid equivalents per gram of dry extract (mg EAA.g ES⁻¹).

Determination of 1,1-diphenyl-2-picrylhydrazyl free radical (DPPH) blocking activity

The inhibitory capacity of DPPH of *H. erinaceus* extracts was measured using the method described by Xu & Chang (2007). The absorbance was measured at 517 nm using a microplate spectrophotometer (Multiskan Go Thermo Scientific). A calibration curve was prepared with ascorbic acid (Sigma-Aldrich) at 99.9 % purity with concentrations from 1.4 to 10.4 µg.mL⁻¹. The experiments were performed in triplicate. The obtained EC50 values were expressed in µg EAG.mL⁻¹.

Analysis of the selective scavenging activity of the ABTS radical

The protocol specifications proposed by Re *et al.* (1999) were used for the evaluation of the ABTS radical. The reading was performed on a microplate spectrophotometer (Multiskan Go Thermo Scientific) at an absorbance of 734 nm using gallic acid as standard ($y = 3.6338x - 0.0805$, $r = 0.9997$) and the concentrations of the results were expressed as µg EAG.mL⁻¹.

Statistical analysis for antioxidants

The determinations of bioactive compounds were processed in the XLSTAT software, a one-way ANOVA and a Tukey test with 95 % confidence were performed.

Results and discussion

The total culture period (incubation plus production) of *H. erinaceus* was 60 days except for strains CCH002, CCH003, CCH007, which covered 90 days, during which time a maximum of three harvests were performed. The three substrates evaluated individually for basidiome growth showed a low biological efficiency and productivity rate (treatment A and C) except for the corn tazole substrate (B), which did not show fruiting body development. Table 1 shows the values obtained for the biological efficiency and productivity rate of the strains analyzed. Strain CCH003 showed significant differences with all strains and treatments ($P < 0.01$); it also presented the highest values of biological efficiency for all treatments, the best being treatment D with 42.33 % and a productivity rate of 0.47 %. In contrast, strain CCH006 showed the lowest biological efficiency values of 0.46 % and a productivity rate of 0.01 % (treatment E). It has been reported that the lion's mane fungus grows mainly on the bark of coniferous trees. Hassan (2007) mentioned that the most economical substrate for growing *H. erinaceus* is coniferous sawdust and yields of 29.3 % are obtained. Thus, a great diversity of substrates and mixtures of substrates have been reported for the cultivation of *H. erinaceus*; Karadžić (2006) obtained an EB of 52 % with a mixture of beech sawdust enriched with wheat bran (20 %), rye grain (25 %) and soybean meal (7 %); Hassan (2007) reported yields of 50.3 % using oak sawdust, 20 % wheat bran, 1 % CaCO₃ + 1 % sugar as substrate. In this study, a BS of 42.3 % was obtained using a mixture

of 50 % oak sawdust, 25 % agave bagasse and 25 % corn tazole. Thus, the highest EB percentages were obtained in the treatments where an additional carbon source is added, as is the case of the different types of flour; in this case, agave bagasse has high percentages of lignin and hemicellulose in its composition (Tello-Balderas & García-Moya, 1988), compounds that are more accessible as a substrate for the fungus, in addition to the addition of molasses, which provides an extra carbon source. In the case of humic acids, these compounds present modified lignins (Vaca-Paulín *et al.* 2006) that are easier to use as substrate for fungi; this could explain the high values of EB for the fungus *H. erinaceus*. Strains CCH003 and CCH007 represent the best option to promote the cultivation of *H. erinaceus* in the region, since being the fastest strains to colonize the substrate, the risk of contamination would be reduced by presenting a greater competition of the mycelium with respect to the contaminating microorganisms that could be present in the substrate during sowing, as mentioned by Colavolpe *et al.* (2014), added to the fact that they were the strains with the highest biological efficiency. Treatment F was able to sustain the growth and production of all strains, compared to the other substrates.

Table 2 shows the results for the determination of bioactive compounds in the wild basidiomata of *H. erinaceus*. The highest concentrations of antioxidants were found in the specimen collected at the Espinazo del Diablo site (CCH003), and the lowest in the specimen belonging to the Puentecillas M. collection site (CCH006).

Total phenolic content

Specimen CCH003 showed significant statistical differences ($P < 0.01$) compared to the other samples. As shown in table 2, the phenolic content of the fruiting body of the fungus *H. erinaceus* varies according to the place of collection, even in the same site. The phenolic content is very variable, which coincides with that reported by Almaraz-Abarca *et al.* (2013), who mentions that the phenolic content in fungi varies due to the type of nutrients that make up the substrate and the climatic conditions where they develop, thus influencing their metabolism to generate a wide variety of compounds. The highest total phenolic content was obtained from the mature fruiting body collected in the Espinazo del Diablo site (60.00 mg EAG.g ES⁻¹), a value lower than those reported in India by Puttaraju *et al.* (2006) with 290.4 mg EAG.g ES⁻¹ for *Lentinula edodes*, considered as a medicinal mushroom. The mushroom collected at the Puentecillas M site (young specimen) yielded the lowest phenolic content (20.13 mg EAG.g ES⁻¹) (table 2), a value similar to that estimated by Li *et al.* (2012) for *H. erinaceus* (19.08 mg EAG.g ES⁻¹), using n-hexane as solvent. Similar values were also reported for *Sparassis crispa* (19.0 mg EAG.g ES⁻¹), a basidiomycete used in traditional Chinese medicine and *Flammulina velutipes* (21 mg EAG.g ES⁻¹) an edible mushroom (Kim *et al.*, 2008).

Table 1. Biological efficiency and productivity rate of wild strains of *Hericium erinaceus* grown on different substrates.

CEPA	Treatments									
	A		C		D		E		F	
	EB	TP	EB	TP	EB	TP	EB	TP	EB	TP
CCH001	2.76 ± 0.15 ^f	0.05 ± 0.01 ^a	5.63 ± 0.11 ^f	0.09 ± 0.01 ^a	14.10 ± 0.20 ^d	0.23 ± 0.01 ^f	1.37 ± 0.07 ^d	0.02 ± 0.01 ^b	16.89 ± 0.11 ^d	0.28 ± 0.01 ^c
CCH002	7.10 ± 0.20 ^e	0.08 ± 0.01 ^a	9.13 ± 0.15 ^e	0.40 ± 0.05 ^a	27.13 ± 0.25 ^e	0.30 ± 0.01 ^d	2.26 ± 0.15 ^{bc}	0.02 ± 0.01 ^b	18.96 ± 0.25 ^c	0.14 ± 0.01 ^d
CCH003	12.46 ± 0.25 ^a	0.13 ± 0.01 ^a	13.36 ± 0.15 ^a	0.14 ± 0.01 ^a	42.33 ± 0.73 ^a	0.47 ± 0.01 ^a	4.07 ± 0.20 ^a	0.04 ± 0.01 ^a	36.13 ± 0.25 ^a	0.40 ± 0.01 ^a
CCH005	5.06 ± 0.15 ^d	0.33 ± 0.10 ^a	7.63 ± 0.11 ^d	0.12 ± 0.01 ^a	26.13 ± 0.35 ^e	0.44 ± 0.01 ^b	1.96 ± 0.11 ^c	0.03 ± 0.01 ^b	19.10 ± 0.65 ^c	0.32 ± 0.01 ^{ab}
CCH006	1.38 ± 0.07 ^g	0.02 ± 0.01 ^a	3.26 ± 0.20 ^e	0.05 ± 0.01 ^a	11.76 ± 0.25 ^e	0.20 ± 0.01 ^e	0.46 ± 0.05 ^e	0.01 ± 0.00 ^c	11.43 ± 0.40 ^e	0.18 ± 0.01 ^d
CCH007	8.00 ± 0.17 ^b	0.09 ± 0.01 ^a	11.26 ± 0.15 ^b	0.12 ± 0.05 ^a	33.06 ± 0.35 ^b	0.37 ± 0.01 ^c	2.56 ± 0.15 ^b	0.03 ± 0.01 ^b	22.13 ± 0.29 ^b	0.23 ± 0.01 ^d
CCH008	4.26 ± 0.15 ^c	0.07 ± 0.01 ^a	6.80 ± 0.10 ^e	0.11 ± 0.05 ^a	14.96 ± 0.21 ^d	0.25 ± 0.01 ^c	1.24 ± 0.05 ^d	0.02 ± 0.00 ^c	17.33 ± 0.30 ^d	0.29 ± 0.01 ^{ab}

a, b, c, d, e, f, g Different letters in the same column indicate significant differences ($P < 0.05$). EB= Biological efficiency; TP= Total productivity.

Table 2. Antioxidant content evaluated for wild strains of *Hericium erinaceus*.

No. CBLH	Place	Técnicas				
		Phenols	Flavonoids	CAT	ABTS EC50	DPPH
CHDIR-DGO		mg EAG.g ⁻¹ ES	mg EQ.g ⁻¹ ES	mg EAA.g ⁻¹ ES	µg EAG.g ⁻¹ ES	µg EAG.g ⁻¹ ES
CCH001	Los túneles	21.33 ± 0.0186 ^a	1.61 ± 0.02157 ^b	40.31 ± 0.0005 ^b	396 ± 0.4727 ^f	382 ± 0.4799 ^b
CCH002	Puentecillas	51.03 ± 0.0324 ^d	1.84 ± 0.01851 ^c	48.96 ± 0.0096 ^d	364 ± 1.5271 ^e	192 ± 1.7143 ^c
CCH003	Espinazo del diablo	60.00 ± 0.0182 ^e	4.21 ± 0.0131 ^f	71.16 ± 0.0028 ^e	157 ± 0.0898 ^b	121 ± 0.1071 ^b
CCH008	Coscomate	36.66 ± 0.0079 ^c	1.79 ± 0.01567 ^c	40.64 ± 0.0099 ^b	371 ± 0.4870 ^e	299 ± 0.6477 ^e
CCH005	La peña	36.78 ± 0.0152 ^c	2.11 ± 0.01517 ^d	46.11 ± 0.0068 ^c	222 ± 0.3463 ^d	254 ± 0.6595 ^f
CCH006	Puentecillas M	20.31 ± 0.0132 ^a	0.69 ± 0.01957 ^a	24.52 ± 0.0004 ^a	522 ± 0.5204 ^e	455 ± 0.4683 ⁱ
CCH007	La gallina	53.20 ± 0.0126 ^d	4.01 ± 0.01211 ^f	51.48 ± 0.0015 ^e	164 ± 1401 ^c	175 ± 0.2026 ^d

a, b, c, d, f, g, h, i Different letters in the same column indicate significant differences ($P < 0.05$).

Total flavonoid content

The strains from the PuenteCillas and Coscomate areas did not show significant statistical differences ($P < 0.01$) between them (they were found in shady places). The highest flavonoid content was obtained for the wild basidiomes belonging to Espinazo del Diablo and La gallina (4.2 and 4.0 mg EQ.g ES⁻¹). The highest flavonoid concentration reported in the literature for *H. erinaceus* is 1.46 mg EQ.g ES⁻¹ by Li *et al.* (2012) extracted with chloroform, which is lower than that obtained in this work. Wilkinson & Kasparbauer (1972) mentioned that plants use flavonoids as a defense mechanism against oxidations promoted by UV light. This could explain the different concentrations of flavonoids present in wild mushrooms, since the specimens with high concentrations of flavonoids were exposed to light, compared to those with lower concentrations growing inside trunks or in shady places.

Total antioxidant capacity

The results obtained agree with the determination of total phenolic content, suggesting a close relationship between these compounds. Higher concentrations (table 2) were obtained than those reported by Charumathy *et al.* (2016) in India for *H. erinaceus* (11.93 mg EAA.g ES⁻¹) using hot water as solvent, but lower than those found by Kosanić *et al.* (2012) in other mushrooms such as *Agaricus campestris*, *Boletus edulis* and *Hydum repadum* (187.73 mg EAA.g ES⁻¹).

Effect of ABTS radical scavenging capacity

The generation of the ABTS⁺ radical cation involves the direct production of the blue-green chromophore ABTS by the reaction between ABTS and potassium persulfate. Addition of antioxidant extracts (EHES) to the preformed radical reduces it to ABTS (Wootton-Beard *et al.*, 2011). Specimen CCH003 showed significant differences compared to the other samples, as well as the most prominent concentration (145 µg EAG.g ES⁻¹) similar to that found by Smolskaitė *et al.* (2015) in the fungus *Inonotus hispidus* (165 µg EAG.g ES⁻¹). Lower values have been reported by Rani *et al.* (2015) in major commercial cultivated mushrooms such as the mushroom *Ganoderma lucidum* (580 µg EAG.g ES⁻¹).

Effect of DPPH radical scavenging capacity

Significant differences were obtained between all the wild-type and cultivated strains that were analyzed. The highest DPPH radical uptake was observed in strain CCH003 (121 µg EAG.g ES⁻¹), lower than that found in wild strains and *Ganoderma lucidum* (290 µg EAG.g ES⁻¹) by Rani *et al.*, (2015); and higher than those reported by Smolskaitė *et al.* (2015) for *Phaeolus schweinitzii* (8.89 µg EAG.g ES⁻¹).

Conclusions

The addition of agave bagasse, oak sawdust and corn tazole allowed optimal growth of wild *H. erinaceus* strains with higher biological efficiency. These easily accessible substrates in the state of Durango are considered agroindustrial wastes, mainly agave bagasse, which results from mezcal production. So far, no specific use has been reported. However, an alternative for its use is presented here, which could promote the cultivation of *H. erinaceus* in the mezcal zone of the state, thus diversifying rural activities in the region. With respect to the bioactive compounds of wild basidioma evaluated, they presented outstanding antioxidant activity in comparison with other studies carried out for this species, and even higher than many of the most consumed medicinal mushrooms in the world. The variability of

the results in the assays performed with respect to the concentrations of antioxidant capacity provides information on how climatic, geographical and maturity conditions influence the production of secondary metabolites, even for the same species. It is hoped that this information will be useful to generate new materials on the effect of conditions on the production of bioactive compounds and how variations in these compounds can be adjusted through cultivation, so that they can be used in the diet or as health promoters, with proper and responsible management.

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