

Improvement of productivity and polysaccharide-protein complex in *Agaricus blazei*

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Abstract – The objective of this work was to assess the productivity and polysaccharide-protein complex content of *Agaricus blazei* on rice straw medium, in comparison to conventional sawdust, using four casing soils. The *A. blazei* strain used was BCRC36814^T, purchased from the Food Industry Research and Development Institute, Hsin-Chu, Taiwan. The two media were evaluated as to *A. blazei* productivity, harvesting time, and production costs. The experimental design used was a randomized complete block, with four replicates. Three local casing soils – Typic Paleudult (CCe), Typic Udorthent (Tq) and Oxyaquic Paleudult (TSp) – were compared to imported peat soil (PS, Sapristis, Histosols), used as the control. The productivity of *A. blazei* using Tq and TSp soil was significantly higher. The TSp casing treatment resulted in earlier harvest by at least 14 to 27 days, when compared to the other treatments. The polysaccharide content in CCe (13.2%) and Tq soils (13.2%) did not differ significantly from the PS (13.4%) and TSp (10.6%) treatments. Local casing soils decreased the production costs of *A. blazei* cultivation. Composted rice straw can substitute sawdust as the culture medium for *A. blazei* production with increased yield.

Index terms: *Agaricus blazei*, casing soil, loam soil, rice straw, silty clay soil.

Ganho de produtividade e complexo de proteína-polissacarídeos em *Agaricus blazei*

Resumo – O objetivo deste trabalho foi avaliar a produtividade e o conteúdo do complexo proteína-polissacarídeos de *Agaricus blazei* em meio de palha de arroz, em comparação ao de serragem, tendo-se utilizado quatro tipos de solo como camadas de cobertura. Utilizou-se a linhagem BCRC36814^T de *A. blazei*, procedente do Food Industry Research and Development Institute, Hsin-Chu, Taiwan. Os dois meios foram avaliados quanto à produtividade, ao tempo de colheita e aos custos de produção de *A. blazei*. Utilizou-se o delineamento de blocos ao acaso, com quatro repetições. As camadas de cobertura foram compostas por três solos locais – Argissolo Vermelho-Amarelo distrófico (CCe), Neossolo Litólico distrófico (Tq) e Oxyaquic Paleudult (TSp) –, comparados a solo turfoso importado (PS, Sapristis, Organossolos Háplicos), utilizado como controle. A produtividade de *A. blazei* com Tq e TSp foi significativamente maior. O tratamento com TSp resultou em colheita antecipada de 14 a 27 dias, em comparação aos outros tratamentos. O conteúdo de polissacarídeos em CCe (13,2%) e Tq (13,2%) não diferiu significativamente do em PS (13,4%) e TSp (10,6%). Os custos de produção foram reduzidos com uso das camadas de cobertura locais. O composto de palha de arroz pode substituir a serragem como meio para o cultivo de *A. blazei*, com aumento de produtividade.

Termos para indexação: *Agaricus blazei*, camadas de cobertura, solo franco-arenoso, palha de arroz, solo argilo-siltoso.

Introduction

It is widely known that *Agaricus blazei* Murrill contains active organic ingredients that are associated with the maintenance of human health and the healing of diseases (Lee et al., 2008). Pharmacological studies have shown that bioactive substances, such as polysaccharides and polysaccharide-protein complexes, in *A. blazei*, function as antioxidants, antimutagenics, antitumorigenics, and anticancer

agents (Izawa & Inoue, 2004; Kimura et al., 2004; Firenzuoli et al., 2008). Therefore, it is not surprising that *A. blazei* has drawn the attention of food scientists and biotechnologists.

Culturing of *A. blazei* requires the spawning growth of the mycelium on a solid culture medium, and subsequent casing of the mycelium with suitable soil to stimulate fruiting-body formation. The composition of the culture medium, i.e., compost, provides nutrients for *A. blazei* mycelium growth and also affects the

later production of the fruiting body. *Agaricus blazei* cultivation is commonly done using sawdust as the culture medium (Pokhrel & Ohga, 2007). In Taiwan, cultivation is usually done using sawdust bag-logs as the major medium. Minor ingredients, such as rice bran, ground corn and calcium carbonate, are used for the spawning stage. However, several researchers have modified the composition of the culture medium by applying agricultural-waste compost to enhance the spawning of *A. blazei* mycelium (Donini et al., 2006) and to increase its overall biological efficiency, which is defined as the ratio of the productivity of an organism to that of its energy supply. Mendonça et al. (2005) used compost from corncobs, wheat straw, rice straw, and several types of grass to produce a medium for *A. blazei*. Pokhrel & Ohga (2007) mixed cattle-bedding waste and sawdust, as major ingredients, to wheat, rice and barley bran, as supplements, for the culturing of *A. blazei*. These authors reported a biological efficiency between 28.6–70.9%. In order to cultivate *A. blazei*, Andrade et al. (2007) used crushed sugarcane and coastcross grass trash as major ingredients, supplemented with ground soybean meal, gypsum and calcitic limestone, as minor ingredients, which resulted in a biological efficiency of 33.6%. Therefore, there is no standard composition for *A. blazei* medium, as it is mixed with various major and minor ingredients, which depend on the available natural resources.

After spawning, *A. blazei* requires soil casing to allow the production of the fruiting body (sporophores). Growers worldwide commonly use peat soil (PS) for casing of mushrooms (Peyvast et al., 2007). Several researchers have used PS as a fundamental ingredient to mix with various ratios of agricultural or industrial wastes in attempt to replace PS and manage waste issues. Paper waste, tea residue, vine shoot, sugar beet, bark, coir, coal tailings, and spent mushroom substrate have been used by Gülser & Pekşen, (2003); Noble & Dobrovin-Pennington, (2005); Peyvast et al., (2007); Pardo-Giménez et al., (2010) whereas Mendonça et al., (2005) and Colauto et al., (2010) have been used as casings for *A. bisporus*, whereas lime schist, sand and loam soil have been used for *A. blazei* (Mendonça et al., 2005; Colauto et al., 2010).

Although the submerged fermentation approach has been developed specifically to produce polysaccharides from *A. blazei* (Lin & Yang, 2006), the majority of farmers worldwide cultivate the mushroom using

traditional methods, i.e., a combination of culturing media and of casing soil.

According to Lee et al. (2008), the market demand for *A. blazei* is growing rapidly in Brazil, Canada, China, Japan, Korea, and the USA. However, the highest biological efficiency of *A. blazei* cultivation reported so far is only 70.9% (Pokhrel & Ohga, 2007), which is difficult to sustain when there is a high demand for the product. Moreover, the use of sawdust and PS, which involves tree farming and mining of the natural resource, poses serious threats to the environment and, at the same time, increases production costs. In addition, the accumulation of heavy metals to toxic levels, which are often associated with *A. blazei*, is speculated to be related to the applied culture media and casing soil (Firenzuoli et al., 2008; Huang et al., 2008).

The objective of this work was to assess the productivity and polysaccharide-protein content of *A. blazei* on rice straw medium, in comparison to conventional sawdust, using four casing soils.

Materials and Methods

The *A. blazei* strain used was BCRC36814^T, purchased from The Food Industry Research and Development Institute, Hsin-Chu, Taiwan. The *A. blazei* stock was maintained on potato dextrose agar (PDA) medium (Difco Laboratories Inc., Detroit, MI, USA) and was activated, subcultured (Pokhrel & Ohga, 2007), and cultured on sterilized wheat (moisture content of approximately 60%) to produce the mother spawn.

Rice straw was collected from the National Chung-Hsing University farm, and matured commercial sawdust medium was purchased from the Q-YO Biotechnology Farm, Ta-Chun, Chang-Hua, Taiwan. Two different culture media were prepared: one with rice straw and the other with sawdust as the major ingredient (70 g each). Matured rice straw compost was obtained by mincing rice straw into small pieces, which were placed in a plastic container, with moisture adjusted to 60%. Then, rice straw was turned manually every 4–6 days for one month. Minor ingredients for both media were identical: rice bran (20 g), ground soybean (3 g), yeast powder (3 g), sucrose (2 g), and gypsum (2 g). Rice straw and sawdust media were separately packaged in bag-logs in a 250-mL pot with a total weight of 100 g each. Both culture media

for mycelium spawning were sterilized (121°C per 1.033 kg cm⁻² for 1 hour) before use.

Two grams of grain spawn, prepared from the mother spawn, were inoculated on sterilized medium and placed in an incubator at 28°C for spawning. The light intensity in the growth chamber was measured at 45±5 cm, 20±2 lux, using an TES-1335 Digital Light Meter, (TES Electrical Electronic Corp, Taipei, Taiwan), and the relative humidity was determined at 90±5%.

The physicochemical properties of each culture media are described in Table 1. The C/N ratio of the matured rice straw compost and of sawdust was 16.8 and 17.8, respectively.

The two media were evaluated for *A. blazei* productivity using a peat soil (PS) from Indonesia as the casing soil, at a thickness of 2.0–2.5 cm (Pokhrel & Ohga, 2007). The experimental design used was a randomized complete block, with four replicates. Fruiting-body formation on different media was conducted in an incubator at 25°C (12 hours, daytime), 22°C (12 hours, nighttime), and at 90±5% relative humidity.

Table 1. Physicochemical properties of rice straw and sawdust culture media for cultivation of *Agaricus blazei*.

Parameter ⁽¹⁾	Culture media	
	Rice straw	Sawdust
pH	8.47	7.47
EC (mS cm ⁻¹)	4.14	2.72
WC (%)	52.00	56.70
Ash (%)	26.50	15.00
OM (%)	70.50	79.20
OC (%)	40.80	46.00
N (%)	2.43	2.58
C/N	16.80	17.80

⁽¹⁾EC, electric conductivity; WC, water content; OM, organic matter; OC, organic carbon; N, total nitrogen; C/N, total nitrogen carbon ratio.

In a second experiment, three local casing soils were also evaluated: TSp, coarse-silty, mixed, nonacid, hyperthermic, Oxyaquic Paleudult (FAO-Unesco, 1988); CCe, loamy, mixed, nonacid, hyperthermic, Typic Paleudult (FAO-Unesco, 1988); and Tq, loamy, mixed, nonacid, hyperthermic, Typic Udorthent (FAO-Unesco, 1988). The peat soil (Saprists, Histosols) (FAO-Unesco, 1988) was used as the control. The physicochemical properties of each casing soil are described in Table 2. Non-casing treatment was performed as a negative control, and the casing soils were not sterilized before use. A total of five treatments were conducted with four replicates, in a randomized complete block design. During the cultivation period, 5 mL of water were added weekly to each treatment. In all casing treatments, matured rice straw was used as the medium to spawn *A. blazei* mycelium.

Harvesting for each casing treatment occurs when the mushroom reaches its highest biomass, i.e., during the immature stage with the veil membrane enclosed and the gills still intact (Mendonça et al., 2005; Pokhrel & Ohga, 2007). After the initial emergence of the first harvestable mushroom, mushrooms were harvested every two weeks until the eighth week. Total yield (grams per pot) was measured from the cumulative fresh weight of mushrooms (FWM). Biological efficiency (BE, %) was calculated after eight weeks of harvesting using $BE = (FWM/DWC) \times 100\%$, in which: DWC, is the dry weight of the culture medium (Andrade et al., 2007).

Polysaccharide-protein complex (PSPC) contents were determined according to Nakajima et al. (2002) and presented as the dry weight of PSPC per dry weight of fruiting body (%). Lyophilized PSPC was used to determine the polysaccharide content, using the phenol-sulfuric acid method (Chaplin & Kennedy,

Table 2. Physicochemical properties of casing soils for cultivation of *Agaricus blazei*.

Casing soil ⁽¹⁾	Physicochemical properties ⁽²⁾										
	pH	EC (μS cm ⁻¹)	BD ----- (g cm ⁻³) -----	PD -----	<i>f</i> -----	WHC	Ash	OM (%)	OC	N	C/N
CCe	6.01	244	1.43	2.69	46.8	142	95.5	3.34	1.76	0.06	29.3
Tq	5.82	177	1.32	2.55	48.2	147	94.5	4.35	2.29	0.10	22.9
TSp	8.02	425	1.69	2.57	34.3	126	98.1	1.14	0.60	0.04	15.0
PS	4.09	3,340	0.55	1.44	61.8	263	10.8	85.5	45.0	2.01	22.4

⁽¹⁾CCe, loamy, mixed, nonacid, hyperthermic, Typic Paleudult; Tq, loamy, mixed, nonacid, hyperthermic, Typic Udorthent; TSp, coarse silty, mixed, nonacid, hyperthermic, Oxyaquic Paleudult; PS, Saprists, Histosols. ⁽²⁾EC, electric conductivity; BD, bulk density; PD, particle density; *f*, porosity; WHC, water holding capacity; OM, organic matter; OC, organic carbon; N, total nitrogen; C/N, total nitrogen carbon ratio.

1994). All standards were purchased from Sigma Chemical Co., St. Louis, MO, USA.

The physicochemical properties of each culture media and casing soil were determined. To do that, ash content was analyzed according to Davies (1974); organic matter and organic carbon were determined according to Nelson & Sommers (1982); and total N was evaluated using the Kjeldahl method (Bremner & Mulvaney, 1982). The culture media and casing soils were soaked separately in water with a ratio of 1:5 (w v⁻¹), and pH and electric conductivity (EC) were measured using a pH-electroconductivity meter. Bulk density (BD) was determined using the code technique (Blake & Hartge, 1986). The particle density (PD) of the soil samples was averaged from two measurements (Blake & Hartge, 1986). Porosity (*f*) was calculated as: [$f = (1 - BD/PD) \times 100\%$]. Water-holding capacity (WHC) was calculated by dividing the weight of the casing soil saturated with water by the casing soil weight after oven-dried. The texture of the soil indicates the particle-size distribution and the relative proportions of sand (2.00–0.05 mm), silt (0.05–0.002 mm), and clay (<0.002 mm), according to Gee & Bauder (1986) (Table 3). The minerals P, K, Ca, Mg, Fe, and Mn were measured using double acid (HClO₄:HNO₃ = 1:4, v v⁻¹) analysis (Jones Junior, 2001). Heavy metals (Zn, Cr, Cu, Ni, Cd, and Pb) were extracted according to Jones Junior (2001) and analyzed using an inductively coupled plasma spectrophotometer JY 138 Ultrace

ICP-AES, (AST Instruments Corporation, Taipei, Taiwan). The heavy metals and other inorganic elements in the culture media and in the casing soils are described in Table 4.

Statistical analyses were performed using the CoStat Statistical Procedures, and means were compared by Duncan's multiple range test, at 5% probability. The Pearson product-moment correlation coefficients (*r*) were computed between total yield and all other evaluated parameters in order to measure the strength of the linear relationship. Then, the *r* values were assessed for statistical significance using two-tailed Student's *t* test.

Results and Discussion

The biological efficiency of *A. blazei* cultivated on both media was compared using PS as the casing soil. The rice straw medium showed a biological efficiency of 76.1%, and the sawdust medium of 40.8%. This indicates that composted rice straw can substitute sawdust as the culture medium, providing a higher biological efficiency. Pokhrel & Ohga (2007) compared the biological efficiency of *A. blazei* cultured on different ratios of cattle-bedding waste (matured compost) and sawdust medium. These authors observed a biological efficiency of 70.9% when using 100% cattle-bedding waste, and of 28.6% when 25% cattle-bedding waste was mixed with 75% sawdust.

Table 3. Texture of casing soils for cultivation of *Agaricus blazei*.

Casing soil	Particle size (g kg ⁻¹)			Soil classification
	Sand	Silt	Clay	
CCe	124	424	452	Loamy, mixed, nonacid, hyperthermic, Typic Paleudult
Tq	362	492	145	Loamy, mixed, nonacid, hyperthermic, Typic Udorthent
TSp	887	40	73	Coarse silty, mixed, nonacid, hyperthermic, Oxyaquic Paleudult
PS	600	289	112	Saprists, Histosols

Table 4. Heavy metals and other inorganic elements in the culture media and in the casing soils.

Treatment ⁽¹⁾	(µg g ⁻¹)						(mg g ⁻¹)					
	Cd	Pb	Cr	Cu	Ni	Zn	P	K	Ca	Mg	Fe	Mn
Rice straw	0.27	3.76	9.30	9.39	5.65	93.92	21.47	58.31	25.70	4.73	0.37	0.73
Sawdust	0.20	2.64	2.26	10.20	3.50	69.63	21.65	25.79	36.13	3.72	0.21	0.12
CCe	10.40	10.3	70.2	35.6	56.8	187	3.83	31.40	1.57	2.31	73.8	1.18
Tq	6.77	54.4	74.6	21.8	51.6	145	3.43	20.50	3.03	6.57	51.5	0.67
TSp	8.44	59.2	53.5	28.8	47.8	148	2.49	19.80	12.50	8.54	63.5	0.64
PS	2.11	106.0	64.0	58.4	29.5	221	4.13	3.17	26.00	3.04	14.6	0.21

⁽¹⁾CCe, loamy, mixed, nonacid, hyperthermic, Typic Paleudult; Tq, loamy, mixed, nonacid, hyperthermic, Typic Udorthent; TSp, coarse silty, mixed, nonacid, hyperthermic, Oxyaquic Paleudult; PS, Saprists, Histosols.

Agaricus blazei is a secondary saprophyte and, unlike most wood-decomposing primary saprophytes, such as *Lentinula edodes*, *Ganoderma lucidum* and *Pleurotus ostreatus* (Pokhrel & Ohga, 2007; Gregori et al., 2008), the growth media must be first degraded by microbes or composted in order to allow nutrients to be absorbed by *A. blazei*. These differences may be attributed to the type of culture medium used or to the age of the compost.

Casing of *A. blazei* mycelium with soil is necessary for fruiting-body formation (Andrade et al., 2007; Pokhrel & Ohga, 2007). In order to compare the effects of different local casing soils with the imported PS on the production of *A. blazei* fruiting body, three representative soils were selected in attempt to reduce production costs, using matured rice straw as the medium to spawn *A. blazei* mycelium. All casing treatments resulted in the fruiting-body formation of *A. blazei*, except for the non-casing treatment control, in which no mushrooms were produced. Casing with TSp soil resulted in a harvesting time as early as 47 days after casing, while the harvesting time for the Tq, CCe and PS treatments took longer: 61, 74, and 70 days, respectively (Table 5). Therefore, the TSp soil treatment resulted in a 14 to 27 days earlier harvest, in comparison to the other three treatments. The average number of fruiting bodies per pot was highest in Tq soil (5.5) and lowest in the CCe, TSp and PS casing treatments (4.5, 3.8 and 3.3, respectively). The average fresh weight (fw) of the fruiting body was highest in the TSp treatment (14.1 g), and lowest in the PS, CCe and Tq soil casings (11.2, 10.5 and 9.3 g, respectively). The total fresh yield per pot was higher in Tq and TSp soil (55.1 and 52.5 g per pot), and lower in the CCe and PS treatments (44.2 and 35.9 g per pot, respectively).

The biological efficiency was higher in the Tq and TSp soil treatments (114 and 109%, respectively), and lower in the CCe and PS casings (91.9 and 74.6%, respectively). Overall, the productivity of *A. blazei* using Tq and TSp as casing soils was significantly higher than with the other two treatments. PS appears to be the poorest one.

TSp and Tq casing soils, with sandy and sandy loam soil texture, respectively, yielded higher productivity, and the PS casing, of sandy loam texture, lower (Table 5). Therefore, the soil texture is not the sole determinant of the productivity of *A. blazei*. Other physicochemical properties of the casing soil may contribute as well. Gülser & Pekşen (2003) reported that high amounts of organic matter and salt in the casing soil reduced the productivity of *A. bisporus*. Organic matter and EC values in the PS used in the present study were both significantly higher than that of all three local casing soils evaluated (Table 2). This indicates that the high concentration of organic matter and salt in the casing soil may cause reduction in *A. blazei* yield, similarly as in *A. bisporus*. The correlation analysis on the heavy metals and other inorganic elements in the casing soils indicates that only Cu and Zn showed significant negative correlations with total yield ($p < 0.05$ and $p < 0.01$, respectively). Therefore, high concentrations of Cu or Zn in the casing soil most likely reduce total yield. This type of relationship has not been previously reported for *A. blazei*. The better productivity of *A. blazei* is probably associated to the lower Cu and Zn concentrations in Tq and TSp casing soils. In general, the cultivation of *A. blazei* in different casing treatments using rice straw as a medium had a productivity order of Tq = TSp > CCe > PS, which

Table 5. Comparison of different casing soils as to the number of days to the emergence of the first harvestable mushroom, number and fresh weight of fruiting bodies (FB), total fresh yield, and biological efficiency (BE) and to polysaccharide and polysaccharide protein complex (PSPC) contents after eight weeks of harvest of *Agaricus blazei* with rice straw as the culture medium⁽¹⁾.

Casing soil ⁽²⁾	Emergence (days)	FB number (N° per pot)	FB fresh weight (g per FB)	Total fresh yield (g per pot)	BE	Polysaccharide ----- (%) -----	PSPC
CCe	74a	4.5b	10.5bc	44.2b	91.9b	13.10ab	48.04a
Tq	61b	5.5a	9.3c	55.1a	114.0a	13.22ab	45.58ab
TSp	47c	3.8c	14.1a	52.5a	109.0a	10.61b	45.18ab
PS	70a	3.3c	11.2b	35.9c	74.6c	13.41a	43.30b

⁽¹⁾Means followed by equal letters, in the rows, do not differ by Duncan's multiple range test, at 5% probability. ⁽²⁾CCe, loamy, mixed, nonacid, hyperthermic, Typic Paleudult; Tq, loamy, mixed, nonacid, hyperthermic, Typic Udorthent; TSp, coarse silty, mixed, nonacid, hyperthermic, Oxyaquic Paleudult; PS, Sapristis, Histosols.

is a good indicative that local soils can substitute imported PS and reduce production costs.

Significantly higher PSPC content was observed in the CCE soil treatment (48%), when compared to PS (43.4%); the TSp and Tq soil treatments showed intermediary values and did not differ significantly from the CCE treatment (Table 5). The polysaccharide content in the CCE (13.2%) and Tq (13.2%) soil treatments did not differ significantly from the PS (13.4%) and TSp (10.6%) casing treatments. However, the polysaccharide content was significantly higher in the PS treatment, when compared to TSp soil (10.6%). The obtained results are indicators that casing with Tq soil is more suitable, in comparison to the other three treatments, when polysaccharide/PSPC levels and the total yield of *A. blazei* mushrooms are the focus of production.

Heavy-metal contents were analyzed in dried *A. blazei* mushrooms for food safety concerns (Table 6). Elements such as Cd and Pb affect human health and, therefore, are strictly regulated. Based on the European Union (2006) regulation on the maximum levels for certain contaminants in foodstuffs, the regulated concentrations of Cd should not exceed 0.2 $\mu\text{g g}^{-1}$ fw in *A. bisporus*, *P. ostreatus* and *L. edodes*, and 1.0 $\mu\text{g g}^{-1}$ fw in other mushrooms, such as *A. blazei*. With regard to Pb, there is no regulation on *A. blazei*, but the limit for *A. bisporus*, *P. ostreatus* and *L. edodes* is 0.3 $\mu\text{g g}^{-1}$ fw (European Union, 2008). Therefore, the European Union regulation was considered as a guideline for *A. blazei* mushrooms in the present study. The Cd content was highest in the mushrooms from CCE soil treatment [1.08 $\mu\text{g g}^{-1}$ of dry weight (dw)] and lowest in the TSp treatment (0.92 $\mu\text{g g}^{-1}$ dw). Pb was highest in the CCE casing treatment (1.61 $\mu\text{g g}^{-1}$ dw),

Table 6. Heavy metal contents in dried fruiting bodies of *Agaricus blazei* under different casing treatments⁽¹⁾.

Casing soil ⁽²⁾	Cd	Pb	Cr	Cu	Ni	Zn
CCE	1.08a	1.61a	2.69a	23.3b	0.71a	170a
Tq	0.98bc	1.44ab	0.44c	23.0b	0.55b	155b
TSp	0.92c	1.18b	0.72b	20.5c	0.43c	152b
PS	1.04ab	0.35c	0.75b	26.5a	0.68a	151b

⁽¹⁾Means followed by equal letters, in the rows, do not differ by Duncan's multiple range test, at 5% probability. All concentrations were pooled from four successive harvests. ⁽²⁾CCE, loamy, mixed, nonacid, hyperthermic, Typic Paleudult; Tq, loamy, mixed, nonacid, hyperthermic, Typic Udorthent; TSp, coarse silty, mixed, nonacid, hyperthermic, Oxyaquic Paleudult; PS, Sapristis, Histosols.

and lowest in the PS treatment (0.35 $\mu\text{g g}^{-1}$ dw). The levels of Cd and Pb from the pooled mushroom were 0.10 and 0.16 $\mu\text{g g}^{-1}$ fw, respectively (average water content of harvested mushroom is 90%), both below the European Union regulation. These results are lower than those reported by Xu (1999), in which the Cd content of *A. blazei* fresh fruiting body ranged from 0.3 to 0.35 $\mu\text{g g}^{-1}$. These differences may be due to the contributions of heavy metals from different media or of casing materials that resulted in the levels accumulated within the fruiting body (Kalac & Svoboda, 2000). Therefore, the selection of culturing materials for food safety purposes is an important consideration for *A. blazei* cultivation. Overall, the relatively low content of Cd and Pb in *A. blazei* is in accordance with the European Union regulation for safe mushrooms.

Conclusions

1. Recycling of rice straw agricultural waste can substitute sawdust medium for *Agaricus blazei* production with increased yield.
2. Casing soils from local soil resources can substitute imported peat soil and decrease the production costs of *A. blazei* cultivation.
3. Cu and Zn content in the casing soils show significant negative correlations with *A. blazei* total yield.
4. The polysaccharide-protein complex content in *A. blazei* fruiting bodies is significantly increased using local Typic Paleudult soil, in comparison to peat soil.

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