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Biological efficiency and nutritional value of *Pleurotus ostreatus* cultivated in agroindustrial wastes of palm oil fruits and cocoa almonds

Eficiência biológica e valor nutricional de **Pleurotus ostreatus** *cultivados em resíduos agroindustriais de dendê e amêndoa de cacau*

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ABSTRACT: The cocoa and palm oil agro-industries active in the state of Bahia, Brazil, generate high quantities of lignocellulosic wastes that could be recycled through their use in the formulation of substrates to cultivate edible mushrooms. *Pleurotus ostreatus,* also known as oyster mushroom, is the second most cultivated mushroom in the world due to its highly appreciated gastronomic, nutritional, and medicinal characteristics. This work evaluated the vertical mycelium growth, biological efficiency, mushroom yield, and nutritional composition of *P. ostreatus* produced in substrates formulated with a combination of palm oil fruit mesocarp (POFM) and cocoa almond peels (CAP) processing wastes. The substrates were formulated with the following POFM/CAP proportions (%/%): S1 – 86.4/9.6; S2 – 76.8/19.2; S3 – 67.2/28.8; S4 – 57.6/38.4, and S5 – 48.0/48.0. Substrates also received 3% powdered charcoal and 1% calcium carbonate. Substrates S1, S2, S3, and S4 were superior for vertical mycelium growth. S2 promoted the best biological efficiency (148.8%) and yield (560.5 g·kg^{-1}). The mushrooms produced in all substrates presented good nutritional values, although mushrooms produced using the S2 presented the highest crude protein content. Overall, S1 is the recommended substrate as it results in higher yields of nutrient rich mushrooms. Production of *P. ostreatus* in substrates composed of POFM and CAP represents a good alternative for recycling these wastes with potential economic and ecological benefits to regions where palm oil and cocoa are grown.

KEYWORDS: oyster mushroom; fungal protein; bioconversion.

RESUMO: As indústrias de cacau e óleo de dendê no estado da Bahia, Brasil, geram grandes quantidades de resíduos lignocelulósicos que podem ser reciclados na formulação de substratos para o cultivo de cogumelos comestíveis. *Pleurotus ostreatus* ou cogumelo ostra é o segundo cogumelo mais cultivado no mundo por apresentar características gastronômicas, nutricionais e medicinais muito apreciadas. Este estudo avaliou o crescimento micelial vertical, a eficiência biológica, a produção e a composição nutricional de *P. ostreatus* produzido em substratos formulados com a combinação de resíduos do processamento de frutos de dendê (mesocarpo do fruto de dendê – MFD) e de amêndoas de cacau (tegumento de amêndoas de cacau – TAC). Os substratos foram formulados com as seguintes proporções de MFD e TAC (%/%): S1: 86,4/9,6; S2: 76,8/19,2; S3: 67,2/28,8; S4: 57,6/38,4 e S5: 48,0/48,0. Os substratos também receberam 3% de carvão e 1% de carbonato de cálcio. Os substratos S1, S2, S3 e S4 foram superiores quanto ao crescimento micelial vertical. S2 promoveu os melhores resultados para eficiência biológica (148,8%) e produção (560.5 g·kg^{-1}). Os cogumelos produzidos em todos os substratos apresentaram valores nutricionais promissores. Entretanto, os cogumelos produzidos com o substrato S2 apresentaram o maior conteúdo de proteína bruta. De modo geral, S1 é o substrato recomendado por resultar na maior produção de cogumelos ricos em nutrientes. A produção de *P. ostreatus* em substratos compostos por MFD e TAC representa uma boa alternativa para a reciclagem desses resíduos com potenciais benefícios econômicos e ecológicos para as regiões produtoras de dendê e cacau.

PALAVRAS-CHAVE: cogumelo ostra; proteína fúngica; bioconversão.

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INTRODUCTION

Agricultural activities generate high quantities of lignocellulosic wastes represented by leaves, stems, roots, straws, and husks of cultivated plants. The increase in wastes from agriculture directly relates to the world's population growth and the consequent rising demand for food production. A good alternative to recycle these wastes is through their use in the formulation of substrates for the cultivation of edible mushrooms (KUMLA et al., 2020).

Pleurotus ostreatus (Jacq.) P. Kumm. 1871, commonly known as oyster mushroom or white-rot fungi, is the world's second most cultivated mushroom. This edible mushroom is appreciated worldwide for its flavor, as well as its nutritional and medicinal properties (PÉREZ-MARTÍNEZ et al., 2015). The commercial production of *P. ostreatus* using lignocellulosic wastes as substrates has an important role in preserving natural resources and ecosystems by recycling agricultural wastes and removing water micropollutants (HULTBERG et al., 2020). *Pleurotus ostreatus* participates in the decomposition of lignocellulose, which is composed of lignin, cellulose, and hemicellulose, through the synthesis of enzymes that act on the cell wall components of woody materials (ALFARO et al., 2020).

Palm oil (*Elaeis guineensis* Jacq.) and cocoa (*Theobroma cacao* L.) are two socioeconomically important crops cultivated in the southern region of the state of Bahia, Brazil. In 2018, Brazil produced 1.565.197 tons of palm oil fruit bunches out of the 272.055.131 tons produced worldwide in the same year (FAO, 2020). The processing of palm oil fruit bunches generates several wastes such as palm kernel, stems, fruit fibers, and peels. The fibers resulting after palm oil fruit pressing (palm oil fruit mesocarp – POFM) represent 12% of the wastes generated from the palm oil processing industry (ROSA et al., 2010).

In 2018, the worldwide production of cocoa was 5.252.377 tons, with Brazil occupying the sixth position in the global ranking, with 239.387 tons produced (FAO, 2020), of which almost 50% were from the state of Bahia (IBGE, 2019). In 2014, it was estimated that the state of Bahia, alone, generated approximately 16.2 thousand tons of cocoa almond peels (CAP). Indeed, 80-120 kg of the waste from CAP are produced from every 1,000 kg of cocoa almonds with 7% humidity (SILVA et al., 2015). Fruit peels are the main waste produced by the industrial processing of cocoa almonds to obtain the liquor (raw material to make chocolate).

Various agricultural wastes have been used in formulations of substrates to grow *P. ostreatus*, straw from alfalfa, pecan nuts, wheat, rice, corn, coffee, coconut, eucalyptus, sugarcane, cocoa, cotton, sawdust from bamboo, and several woods (CONDÉ et al., 2017; RODRÍGUEZ, 2018; YAMAUCHI et al., 2019). However, the combination of the agro-industrial wastes POFM and CAP as substrates for the production of high-protein foods such as *P. ostreatus* has not been investigated so far.

Therefore, the present study aimed to evaluate the mycelium growth and production of *P. ostreatus* in substrates formulated with different proportions of POFM and CAP. Besides the obvious environmental impact, studies involving the recycling of these wastes are warranted because mushroom production has the potential to increment food production, jobs, and income generation in regions that produce cocoa and palm oil in Brazil and other countries, and, consequently, contribute to food security and poverty reduction. Importantly, these are amongst the objectives for sustainable development of the 2030 Agenda of the United Nations (UN, 2020).

MATERIAL AND METHODS

Wastes from the agro-industries of palm oil and cocoa

Palm oil fruit mesocarp waste was collected from the Kidendê palm oil production plant, located in the municipality of Taperoá, Bahia, Brazil (13°19'40.7"S; 39°02'47.0"W). Cocoa almond peel waste was donated by the cocoa almond processing plant Barry Callebaut Company, located in the municipality of Ilhéus, Bahia, Brazil (14°47'35.6"S; 39°03'07.2"W).

Growth of *P. ostreatus*

Pleurotus ostreatus isolate Plo 02 was donated from the culture collection of the Laboratory of Mycorrhizal Associations, Department of Microbiology, Universidade Federal de Viçosa, Minas Gerais, Brazil. The culture was preserved in test tubes with potato dextrose agar (PDA, Acumedia) at 4 °C before being reactivated by mycelium inoculation onto PDA medium and incubation for 7 days at 25 °C.

Preparation of *P. ostreatus* spawn

Sorghum grains were soaked in tap water for 2 h, and the excess of water was drained with a common household plastic sieve. Then, 100 g of the soaked grains were transferred to 200 mL-glass flasks, closed with a metallic cover, and sterilized in an autoclave at 121 °C for 55 min. After cooling, each flask, containing the sterilized grains, was inoculated with three agar plugs (0.5 mm diameter) of the *P. ostreatus* culture (grown as mentioned above) and incubated at 25 °C for 20 days, without photoperiod.

Substrates formulation

For substrate formulation, POFM and CAP were combined as described in Table 1. The substrate mixture received 3% of triturated charcoal and 1% $CaCO_3$, to maintain the humidity

and pH, as recommended by some mushroom producers in Brazil. Activated carbon or charcoal is also recommended for grain-based substrate casing in *Agaricus bisporus* production (BECHARA et al., 2009).

Vertical mycelium growth of *P. ostreatus*

To evaluate the growth of *P. ostreatus*, the formulated substrates (Table 1) were transferred to test tubes 20 cm long and 3 cm wide. Each tube was filled with 28.5 g of fresh substrate, which allowed forming a substrate column of 12 cm length within the tubes. The tubes were than covered with a cotton plug and wax paper and sterilized in an autoclave at 121 °C for 55 min. After sterilization, inoculation was conducted by transferring 1 g of the prepared spawn (inoculum) to the substrate surface within the tubes. The tubes were closed with the cotton plug and wax paper and incubated at 25 ± 1 °C, in the dark, in an incubation room. Mycelium vertical growth was measured along the substrate by using a ruler placed over the tubes, at every two days. The measurements of mycelium growth for all treatments were stopped when growth reached the total substrate length (12 cm), for the first time in one of the tubes. Mycelium vigor was analyzed through the non-microscopic observation of its growth characteristics following the scale described by PEDRA (2006): 1 – weekly dense, 2 – medium dense, and 3 – highly dense.

Production of *P. ostreatus*

For mushroom production, the substrates (Table 1) were transferred to 1 kg polypropylene bags fitted with a filter for gas exchange. The bags with the substrates were sterilized in an autoclave at 121 °C for 55 min. After cooling, the surface of the substrates was inoculated with 20 g of *P. ostreatus* spawn (seed inoculum produced as described above). The bags were sealed and transferred to a mushroom colonization chamber maintained at 25 ± 2 °C in the dark, until complete colonization of the substrate.

After substrate colonization, the bags were transferred to a mushroom growth chamber with automated control of humidity and temperature through a system using nebulization and fans. Initially, the colonized bags received a shock treatment with the reduction of the temperature in the chamber to 20 °C by turning on the nebulization system, three times during the night, for 3 min each. Following this temperature shock, 12 equidistant holes were made in the bags, using a stainless steel scalpel, to allow the emission of mushroom primordia. The air humidity of the growth chamber was maintained at 85–90% and the temperature was kept at 25 ± 2 °C during the entire production period.

Fruiting body (mushroom) harvest was done twice a day, when the fruiting bodies presented the pileus with an almost flat format, which is considered the harvest point for *P. ostreatus.* The fruiting bodies were weighted and measurements such as the number of bunches, basidiomes, average size of the stipe, and average diameter of the pileus were taken. The fruiting bodies were dried in an oven with ventilation at 45 °C to obtain the dry weight.

Biological efficiency (BE) and yield (Y) were calculated as follows (Eq. 1):

$$BE = \frac{MFW}{SDW} \times 100 \quad (1)$$

where BE = biological efficiency (%), MFW = mushroom fresh weight (g) and SDW = substrate dry weight (g).

Y was calculated with Eq. 2:

$$Y = \frac{MFW}{SFW} \quad (2)$$

where Y = yield ($g \cdot kg^{-1}$), MFW = mushroom fresh weight (g) and SFW = substrate fresh weight (kg).

Table 1. Proportions of palm oil fruit mesocarp (POFM) and cocoa almond peels (CAP) processing wastes used to formulate substrates for the cultivation of *P. ostreatus*. All substrate formulations received 3% of powdered charcoal and 1% of $CaCO_3$.

Substrates	POFM (%)	CAP (%)
S1	86.4	9.4
S2	76.8	19.2
S3	67.2	28.8
S4	57.6	38.4
S5	8.0	48.0

Bromatological analysis of substrates and mushrooms

Substrate samples were analyzed for dry matter (DM), mineral matter (MM), crude protein (CP), ether extract (EE), fiber in neutral detergent (FND), fiber in acid detergent (FAD), and lignin. The analytical methods used are described by SILVA et al. (2017).

Other measurements included the content of carbon (C) to calculate the C/N ratio, as described by CARMO; SILVA (2012). Mushroom crude fiber content was determined by the gravimetric method, as defined by the Association of Official Analytical Chemists (AOAC). Nitrogen (N) content was determined by the micro-Kjeldahl method described by AOAC (1996). Total carbohydrates (C) were calculated with Eq. 3:

$$C = [100\ g - (\text{total fibers} + \text{proteins} + \text{fat} + \text{ash})] \quad (3)$$

Energy was calculated with Eq. 4:

$$\text{Energy(kcal)} = (4 \times \text{crude protein}) + (9 \times \text{fat}) + (4 \times \text{carbohydrate}) \quad (4)$$

using the Atwater general factor system (FAO, 2003a). The conversion factor used for N to protein was 6.25 (FAO, 2003b).

Statistical analysis

The experimental design was completely randomized with eight replications for vertical mycelium growth and 15 replications for mushroom production. Data were submitted to analysis of variance (ANOVA) and the grouping of means was done using the Scott–Knott test ($p \leq 0.05$). The correlations between the variables were studied using multivariate analysis, the principal component analysis (PCA) and the correlation matrix (correlogram), using the statistical software packages: R v.3.6.3 (R CORE TEAM, 2020); RStudio v.1.2.5042 (RSTUDIO TEAM, 2020); the ggplot2 package v3.3.2 (WICKHAM, 2016); the dplyr package v1.0.2 (WICKHAM et al., 2020); the FactoMineR package (LÊ et al., 2008) and the corrgram package v1.13 (WRIGHT, 2018).

RESULTS AND DISCUSSION

The contents of mineral matter, protein, hemicellulose, cellulose, lignin, and C/N ratio of the wastes used in the formulation of the substrates are shown in Table 2. These values are within the range reported for lignocellulosic wastes commonly used for mushroom production, as described by FIGUEIRÓ; GRACIOLLI (2011), using substrates composed of rice and wheat straw.

Mycelium growth is an important initial phase for mushroom production, since it leads to substrate colonization and allows mushroom growers to determine if growth conditions are adequate (POKHREL et al., 2013). Moreover, rapid mycelium growth lowers the possibility of contamination and growth of competing fungi and bacteria (JONATHAN et al., 2008). Substrates S1, S2, S3 and S4 promoted similar mycelium growth rates (0.8 $cm \cdot d^{-1}$) and reached total colonization of the substrate mixtures in the tubes after 15 days of inoculation. The substrate S5 supported a lower daily growth rate of 0.73 $cm \cdot d^{-1}$ and 15 days after inoculation, colonization by *P. ostreatus* reached 10.95 cm of the substrate (Fig. 1). In banana wastes, the growth rate of *P. ostreatus* mycelium was 1.08 $cm \cdot d^{-1}$ (CARVALHO et al., 2013). In sawdust and coconut shells, the growth rates of different isolates of *P. ostreatus* were 0.63 and 0.67 $cm \cdot d^{-1}$, respectively (MARINO et al., 2008).

Table 2. Contents of mineral matter (MM), crude protein (CP), hemicellulose, cellulose, lignin, and C/N ratios of palm oil fruit mesocarp (POFM), cocoa almond peels (CAP) and charcoal used to formulate the substrates for the cultivation of *P. ostreatus*.

Bromatology	POFM	CAP	Charcoal
(%)			
MM	6.3	11.4	10.7
CP	7.6	12.8	6.9
Hemicellulose	15.8	8.9	2.2
Cellulose	30.1	15.4	7.6
Lignin	11.2	21.7	66.1
C/N ratio	42.5	13.5	45.2

After the 15th day of inoculation, mycelium vigor was evaluated and classified (PEDRA, 2006). Substrates S1, S2, and S3 supported a highly dense mycelium, whereas substrates S4 and S5 presented medium dense mycelium (Fig. 1B). Medium dense mycelium was reported when *P. ostreatus* was grown in sawdust from coconut shells (MARINO et al., 2008). These authors pointed out the importance of fast substrate colonization by a vigorous mycelium, which was observed in this study with the substrates S1, S2, and S3 (Fig 1B).

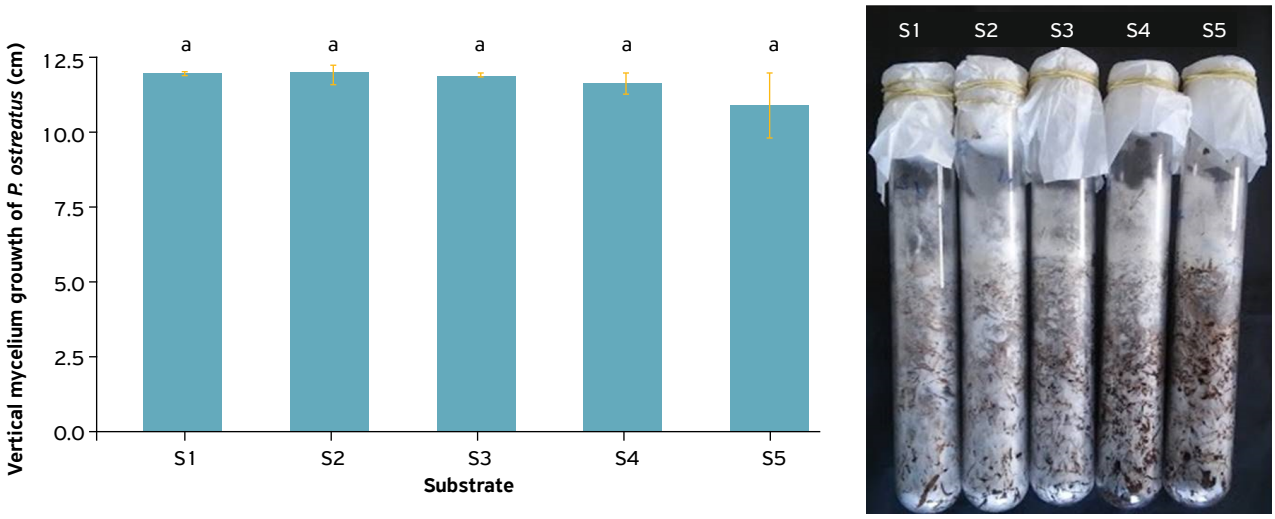
Mycelium growth rate and substrate colonization are directly related to the nutritional composition and granulometry of the substrate, as well as O_2 concentration, temperature, and fungal genome (SOFI et al., 2014; HOA; WANG, 2015). The porosity of the POFM waste allows the translocation of nutrients, water, and rapid fungal colonization (ZHU et al., 2017). The substrates formulated with POFM and CAP were not tested for granulometry and aeration, but allowed for fast and dense mycelium growth of *P. ostreatus.* Wastes with smaller particles can affect gas exchange and the colonization rate of the substrate (PEDRA; MARINO, 2006).

The substrates tested herein promoted rapid mycelium growth of *P. ostreatus*, reaching complete colonization on the 15th day after inoculation. MARLINA et al. (2015) reported that complete colonization of *P. ostreatus* in bags with 1 kg of substrate formulated with palm oil fruit waste, rice bran, and mineral fertilization took 28-35 days. CONDÉ et al. (2017) observed complete substrate colonization 20 days after inoculation with *P. ostreatus* by using substrates formulated with sugar cane (*Saccharum officinarum*) processing waste enriched with different concentrations of Fe. *Pleurotus ostreatoroseus* and *Pleurotus florida* grown in substrates formulated with cotton residues or sawdust and supplemented with rice bran showed total substrate colonization within 24 to 30 days after inoculation, and started the emission of primordia within 5 to 10 days after colonization (REIS et al., 2010).

In the present study, primordia emission occurred 17 days after inoculation, which is a shorter period than that described previously for *P. ostreatus* in other substrates. Formation of fruiting bodies of *P. florida* and *Pleurotus flabellatus* cultivated in substrate composed of anaerobically digested biomass waste, mostly banana leaf biomass from a biogas plant, took 20-30 days after inoculation (CHANAKYA et al., 2015).

Biological efficiency (BE) and mushroom yield

The BE and mushroom yield for *P. ostreatus* cultivated with all the formulated substrates described herein were higher



S1: 86.4% POFM + 9.6% CAP; S2: 76.8% POFM + 19.2% CAP; S3: 67.2% POFM + 28.8% CAP; S4: 57.6% POFM + 48.0% CAP; S5: 48.0% POFM + 48.0% CAP, after 15 days of incubation at 25 °C.

Figure 1. Vertical mycelium growth of *P. ostreatus* in the substrates formulated with wastes from palm oil fruit mesocarp (POFM) and cocoa almond peels (CAP) processing and in vitro growth.

than the values reported in the literature. Indeed, BE ranged from 92.7 to 148.8% and yield from 349.3 to 560.5 $g \cdot kg^{-1}$ (Fig. 2). For BE and mushroom yield, S1 was the most efficient substrate, followed by S2, and S3, while S4 and S5 were the least efficient substrates (Fig. 2). The greater the proportion of palm oil fruit mesocarp processing waste in the substrates (Table 1), the more efficient they were in terms of BE and yield. Lower values were reported for *P. ostreatus* produced in substrates composed of corn cobs and wheat straw, with BEs of 52.82 and 66.93% and yields of 119 and 165.7 $g \cdot kg^{-1}$, respectively (KOUTROTSIOS et al., 2014). In addition, *P. ostreatus* cultivated in substrates containing wastes of sisal (*Agave sisalana*) leaves and stems, enriched with caw manure presented values for BE and yield of 62.9% and 188.64 $g \cdot kg^{-1}$, respectively (RAYMOND et al., 2013).

The higher BE and mushroom yield presented here in relation to those previously reported could be due to the medium to high mycelium vigor supported by our substrate mixtures (Fig. 2). Accordingly, colonization and production of fruiting bodies in lignocellulosic substrates are directly related to the fungus mycelium vigor (FIGUEIRÊDO; DIAS, 2014). Another reason is the nutritional characteristics of the substrate and the environmental conditions of the cultivation process, as reviewed by KUMLA et al. (2020). Indeed, the C/N ratio of the wastes used to formulate the substrates to cultivate *Pleurotus* spp. is particularly important since it interferes directly with substrate colonization and the production of fruiting bodies (EIRA, 2004). The C/N ratios of the POFM (42.5/1) and CAP (12.5/1) used in the substrates formulated herein were adequate for the successful production of *P. ostreatus*, as suggested by EIRA (2004) who indicated that the ideal C/N ratio for early colonization should be between 20/1 and 50/1.

Taken together, the good results for BE and yield presented herein indicate that the tested substrate formulations using POFM and CAP are adequate to cultivate *P. ostreatus*. Substrate S1, composed of 86.4% POFM and 9.6% CAP, presented the best BE and yield results and is, therefore, recommended for the production of *P. ostreatus*.

Mushroom nutritional composition

Several factors affect the nutritional composition of mushrooms, such as fungal genetics, substrate origin and nutritional composition, growth conditions, and the developmental stage of the fruiting bodies (WANG et al., 2015). In this study, the nutritional composition of *P. ostreatus* fruiting bodies did not significantly vary according to the different substrate formulations for the parameters: humidity (92.36%), ashes (8.19%), fat (1.24%), fiber (13.4%), and carbohydrates (51.64%). The energy value of the fruiting bodies varied from 311.37 to 333.17 $kcal \cdot g^{-1}$ (data not shown). However, the protein content was affected by the substrate composition. Mushrooms produced in the S2 substrate were the richest, followed by those cultivated in the substrates S1 and S3, while mushrooms cultured in the substrates S4 and S5 had lower crude protein content (Table 3). Overall, the nutritional composition of *P. ostreatus* fruiting bodies produced in this study is similar to that reported for *P. ostreatus* produced in substrates derived from several agro-industrial wastes (OYETAYO; ARIYO, 2013; KOUTROTSIOS et al., 2014; FERNANDES et al., 2015).

In the present study, the composition of the substrates was the only varying factor, since only one isolate of *P. ostreatus* was tested and all other growth conditions were kept the same. Substrate S1 promoted the highest yield and BE, which was 22.7% higher than the BE obtained for

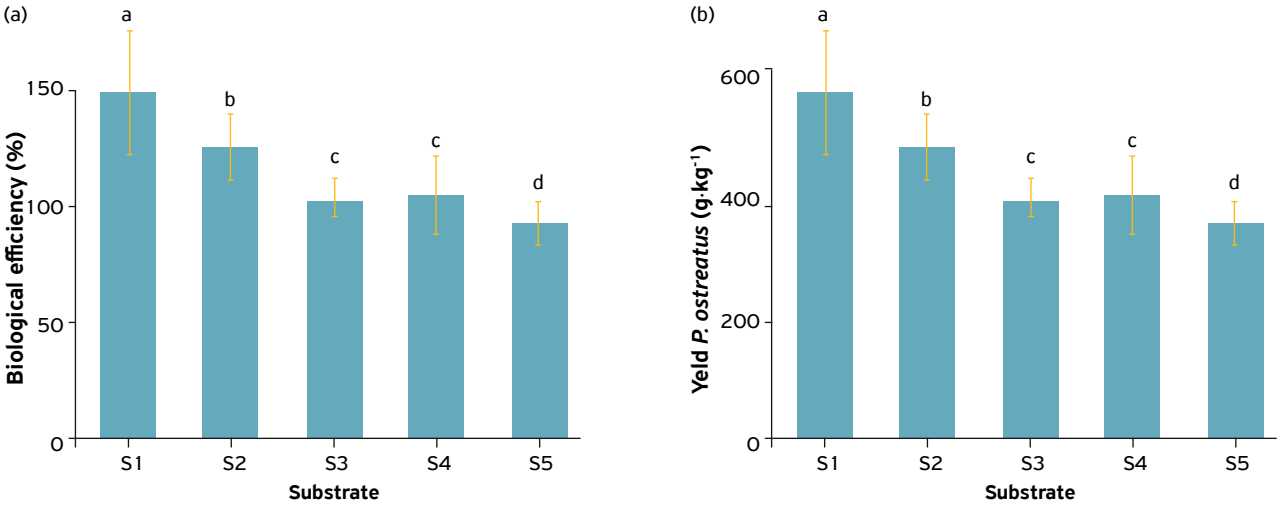


Figure 2. Biological efficiency (a) and yield (b) of *P. ostreatus* produced in substrates formulated with different concentrations of wastes derived from the processing of palm oil fruit mesocarp (POFM) and cocoa almond peels (CAP). Harvesting period lasted 31 days.

the S2. Considering a large-scale production system, this difference in BE can represent an increase of approximately 220 kg in mushroom production for each ton of dried substrate. Therefore, although S2 allowed the production of mushrooms with higher protein content, it is not the most viable or profitable formulation when compared to the yield that can be obtained with S1.

Mycelium vigor is another factor that must be considered for mushroom production. Besides promoting less incidence of competitive microorganisms, rapid substrate colonization, which in this study occurred 15 days after inoculation, allows the mushroom producer to run more production cycles. In turn, profit margins can potentially increase since more mushrooms will be produced within a fixed period and allocated space and resources.

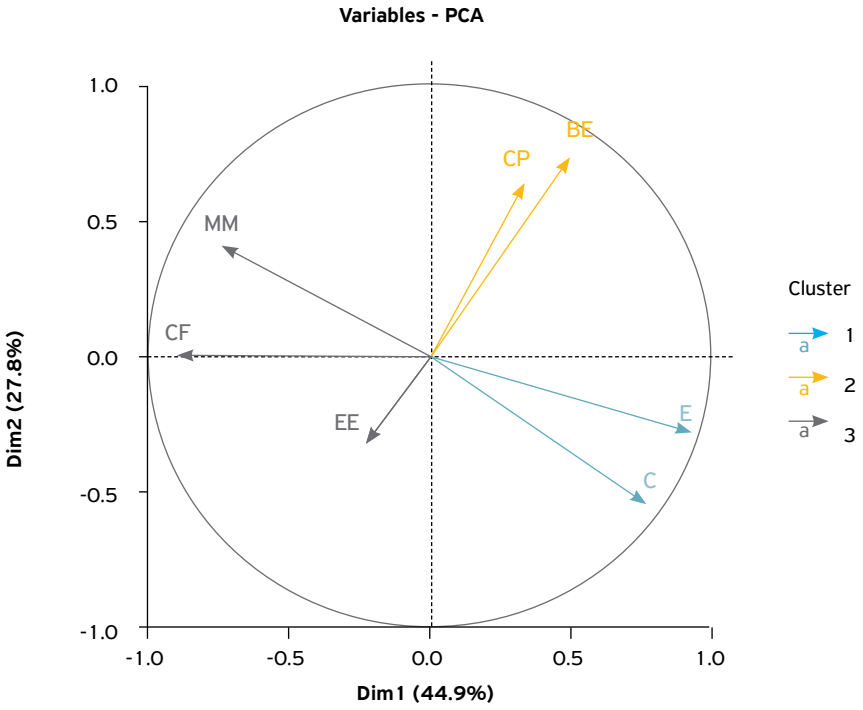
Table 3. Content of humidity (H), mineral matter (MM), fat, crude protein (CP), fiber, and carbohydrates (C) (g in 100 g of dry matter, means ± SD, n = 5) of *P. ostreatus* fruiting bodies grown in substrates with different concentrations of the palm oil fruit mesocarp (POFM) and cocoa almond peel (CAP) processing wastes.

Substrates	H	MM	Fat	CP	Fiber	C
S1	92.6±0.45a	7.58±1.61a	1.23±0.10a	25.2±0.11c	12.4±1.02a	53.5±1.57a
S2	92.5±0.66a	8.55±0.02a	1.30±0.09a	27.5±0.56a	13.3±0.74a	49.2±2.37a
S3	92.4±0.99a	8.36±0.28a	1.01±0.01a	26.6±0.10b	12.9±0.45a	51.0±1.42a
S4	92.6±0.39a	8.36±0.18a	1.21±0.00a	25.0±0.02c	13.1±0.62a	52.1±0.83a
S5	91.7±0.80a	8.12±0.20a	1.47±0.01a	22.4±0.24d	15.4±0.65a	52.4±0.75a
CV (%)	1.32	4.41	9.57	2.03	9.22	2.66

Means with the same letter in the columns do not differ statistically by the Scott–Knott test at 5% probability.

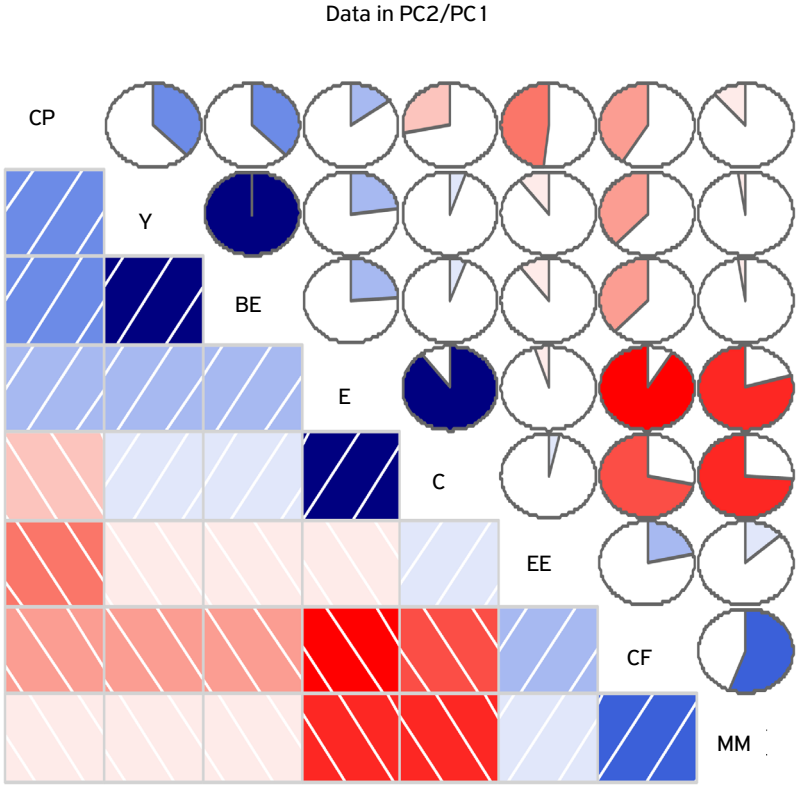
The principal component analysis displayed in Figure 3 indicates that BE, yield, and crude protein were directly and positively correlated, and formed one cluster. These parameters are of special interest for mushroom producers to evaluate the quality of the mushrooms, and they are affected by the quality of the growth substrate. Thus, S1, which had the highest content of the palm oil processing waste, stimulated a high production of mushrooms with high protein content. On the other hand, BE, yield, and crude protein were inversely correlated with fat content, which formed another cluster with crude fiber and mineral matter. Furthermore, carbohydrate content and energy were directly and positively correlated and formed a third cluster.

Accordingly, the correlogram (Fig. 4) indicates a strong positive correlation between BE and yield, and between carbohydrate and energy. It also shows the strong positive correlation between BE, yield, and crude protein content as well as between crude fiber and mineral matter. A weaker, but positive correlation is observed between energy, BE, yield, and crude protein, as well as between fat and crude fiber. On the other hand, energy and carbohydrate have a strong negative correlation with crude fiber and mineral matter. Negative weaker correlations are observed between fat and crude protein, and between crude protein, crude fiber, and mineral matter (Fig. 4). In fact, BELLETTINI et al. (2019) discussed that fruiting bodies with high protein contents tend to have low fat content.



CP: crude protein; BE: biological efficiency; Y: yield; E: energy; C: carbon; EE: ether extract (fat); CF: crude fiber; MM: mineral matter (ashes).

Figure 3. Principal Component Analysis (PCA), with distribution of the variables studied in the dimensions 1 and 2 (Dim 1 and Dim 2)



CP: crude protein; BE: biological efficiency; Y: yield; E: energy; C: carbon; EE: ether extract (fat); CF: crude fiber; MM: mineral matter (ashes). The red color indicates a negative correlation between the variables while the blue color indicates a positive correlation. The size and the color intensity of the pizza slice indicate how strong the association is between the variables.

Figure 4. Correlogram of biological efficiency, yield, and the nutritional values of the fruiting bodies of *P. ostreatus*, grown in substrates formulated with palm oil fruit mesocarp (POFM) and cocoa almond peels (CAP) processing wastes.

CONCLUSIONS

Substrates S1, S2, and S3 supported good mycelium growth and vigor, with highly dense substrate colonization by *P. ostreatus*. Since complete substrate colonization was reduced to 15 days, which is shorter than most of the reported periods in the literature, substrates composed of palm oil fruit mesocarp and cocoa almond peel processing wastes have the potential to allow more production cycles of *P. ostreatus*. The recommended substrate is formulated with 86.4% of POFM and 9.4% of CAP (substrate S1), as it results in higher yields of nutrient rich mushrooms. The bioconversion of these waste products into high protein and nutritious mushrooms is suggested as an excellent recycling alternative with potentially significant ecological and economic benefits to the regions where palm oil and cocoa are produced.

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