**ENVIRONMENTAL MICROBIOLOGY - RESEARCH PAPER** 





# Use of peanut waste for oyster mushroom substrate supplementation—oyster mushroom and peanut waste

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#### Abstract

The aim of the research was to verify the influence of macro and micronutrients present in the peanut waste (hulls and nuts) for supplementation of *Pleurotus ostreatus* substrate. The raw materials for base substrate preparation were *Brachiaria dictyoneura*, sugarcane bagasse (bulk material), rice and wheat bran, calcitic limestone, and gypsum. The following supplement formulations were used as treatments: (1) 100% peanut hulls, (2) 80% peanut hulls + 20% nuts, (3) 60% peanut hulls + 40% nuts, (4) 40% peanut hulls + 60% nuts, (5) 20% peanut hulls + 80% nuts, and (6) 100% nuts. A commercial supplement was also used as an additional treatment. The supplementation was done at spawning using the rates of 1% and 2% wet weight of the substrate. Positive correlations between yield and N content, and weight of mushroom and P and K content were verified with 1% supplement. A positive correlation between yield and Cu content, and a negative correlation between yield and Mn content were observed with 2% supplement. The use of peanut waste can be used as supplement for the production of *P ostreatus* increasing biological efficiency up to 61%. A better combination can be reached with 20% peanut hulls + 80% nuts or 100% nuts. The addition of 2% supplement in the substrate provided greater yield than 1%.

Keywords Pleurotus ostreatus · Macronutrient · Micronutrient · Bioconversion · Food quality

#### Introduction

Globally, developed regions are concerned about the quality of the ecosystem and sustainability seeking alternatives to close the food production cycle, avoiding the emission of  $CO_2$ , producing high quantities, with maximum quality of food and avoiding the generation of waste.

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Sugarcane (*Saccharum officinarum*) is an important crop for the production of biofuels. In Brazil between 2015 and 2016, 658 million tons were harvested [1]. The planting of *Brachiaria dictyoneura* has been growing significantly and has been the best option for the development of tropical livestock. In the last 3 years, residues from these two plant crops are the main bulk materials used for the production of *Pleurotus ostreatus* mushroom substrate [2], due to their great abundance and low price.

Peanut (*Arachis hypogaea*, Leguminosae) is the fourth most planted and consumed oilseed worldwide. It also provides a good option for crop rotation [3, 4]. Currently, several sugar cane reform areas are cultivated with peanut with the aim of improving the physical, chemical, and biological conditions of the soil.

In addition to peanuts planted for the improvement of soil quality, they are widely used for the production of oil, milk, peanut butter, confectionery, roasted peanuts, snack products, extenders in meat product formulation, soups, and desserts [5]. Peanuts are consumed all over the world in a wide variety of forms, being used as the complete dietary source for people on expeditions to diverse areas like Antarctica, space, and trekking [5]. It has notably been the source of elimination of

malnutrition amongst the population in many African countries in recent years [6].

With all these applications, the domestic consumption of peanuts in Brazil is high. Even though the export scenario of this product has been growing fast, exports are accompanied by a series of quality standards that the peanut must possess, to ensure the product is exported with the highest quality. Because of these standards and depending on the exporting country, a series of products and by-products are generated after the processing and selection. This may include those grades of poor quality nuts (grain) with and without skin (attacked by pests and diseases) and peanut hulls. The literature reports the applications given to this bulky waste generated in peanut-producing countries, which are often burned, dumped, or left to deteriorate naturally [7].

In the recent past, environmental concerns have led to an interest in using peanut waste for a variety of purposes: fuel, mulch, carrier for chemicals and fertilizers, bedding for live-stock and poultry, pet litter, soil conditioners, etc. [8]. Despite several applications of the use of peanut waste, no literature reported the influence of macro and micronutrient of this sub-product to supplement mushroom substrates.

Mushroom production in Brazil and South America is increasing very fast due to the nutritional benefits of mushroom as a food and as a source of economic income. In spite of this rapid growth in the last few decades, the international companies of supplements and spawns of mushrooms have not made any representation in order to make their products available in Brazil and South America. Supplements have been used successfully to improve mushroom yield [9, 10] and the use of alternative supplements in developing countries could be a good strategy.

Research has already shown an increase of 34.4% in yield, in addition to increasing precocity, thus reducing the length of the crop cycle [11, 12]. The range of supplement composition is characterized by the amount of protein (24 to 62%) and fat (0.8 to 19%) of the material used as base supplement [13]. The peanut wastes have about 25.8% protein and 49.2% fat in the nuts, while the peanut hulls have 7.3% protein and 1.4% fat [14, 15].

In addition to the amount of protein and fat, several authors have also detailed the amount of carbohydrates, fibers, and ashes in the supplements and their possible influence on mushroom yields [16, 17]. Studies have been performed highlighting the importance of the proximate analysis (ash, fibers, fats, proteins, and carbohydrates) of the supplements, but no research was carried out referring to which macro and micronutrients were necessary and essential for the selection of a supplement of high quality. So the aim of this research is to verify the influence of macro and micronutrients present in the waste of the peanut industry (peanut hulls and nuts) and the agronomic viability to use different formulations to supplement the substrate for the production of *Pleurotus ostreatus* (oyster mushroom).

#### **Material and methods**

#### Substrate

*P. ostreatus* substrate was prepared using a short method of composting, totaling 8 days of substrate preparation, 6 days for phase I and 2 days for phase II (semi-composted system). During phase I process, *Brachiaria dictyoneura* and sugarcane bagasse (bulk material) were moistened for 2 days. On the 3rd day, the pile was assembled, and on the 4th day, the pile was turned and, then, the additional materials (rice and wheat bran, calcitic limestone, and gypsum) were added. Afterwards, two more turns were performed and at the 7th day the substrate was transferred to a pasteurization chamber (phase II). The substrate was pasteurized between 65 and 72 °C during 20 h and subsequently conditioned between 55 and 48 °C for 1 day. Table 1 presents the chemical characteristics of the substrate samples were analyzed.

#### Supplement

Peanut hulls and nuts were used as a supplement in the present study (Fig. 1). The runner cultivar was used because it is widely planted in Brazil. The hulls and nuts were dried at 68 °C for 24 h (which serve as heat treatment), until they reached 4-6% moisture, then they were crushed with a sieve to < 0.5 mm. The formulations used as treatments were (1) 100% hulls of peanut, (2) 80% hulls of peanut + 20% nuts, (3) 60% hulls of peanut + 40% nuts, (4) 40% hulls of peanut + 60% nuts, (5) 20% hulls of peanut + 80% nuts, and (6) 100% nuts. As a reference, two more treatments were used, the first one was an international commercial supplement (Spawn Mate II SE®-recommended for the production of *P. ostreatus*) and the second was the substrate control (without supplement), used to compare the viability of the formulations and the efficiency of the technique of substrate supplementation. The supplements with peanut waste and Spawn Mate were added to the substrate in two doses (1 and 2% wet weight of the substrate). The supplement rates used followed the recommendation of the commercial product (treatment 7) and the values reported in the literature [11, 12, 18]. Table 1 presents the chemical characteristics of the supplements. The contents of macro and micronutrients of the supplements and substrate were evaluated, following the methodology presented by Bell and Ward [19] and Sonneveld and van Elderen [20]. For each nutrient, three repetitions were performed.

#### Inoculation

POS 16/01 strain of *Pleurotus ostreatus* var. Florida was used. This strain was obtained from a commercial grower in the city of Mogi-das-Cruzes in the São Paulo State (Brazil). This strain

Table 1 Mac	ro and micronutr.	ients (g kg <sup>-1</sup> ) of th	he supplements w.	ithout being mixed	d and mixed at a r	Macro and micronutrients (g kg <sup>-1</sup> ) of the supplements without being mixed and mixed at a ratio of 1% (w/w) to substrate (1st experiment) and at a ratio of 2% (w/w) to substrate (2nd experiment)	substrate (1st exp	eriment) and at a	ratio of 2% (w/w	<li>v) to substrate (2n)</li>	d experiment)
Treatments	N g kg <sup>-1</sup>	Ρ	K	Ca	Mg	S	B mg kg <sup>-1</sup>	Cu	Fe	Mn	Zn
1	$13 \pm 0.04$	$0.5\pm0.02$	$6 \pm 0.01$	$3 \pm 0.2$	$1.3 \pm 0.08$	$0.7\pm0.002$	$25 \pm 0.6$	$11 \pm 0.1$	781 ± 3	$63 \pm 4.2$	$27 \pm 0.5$
2	$19.8\pm0.1$	$0.8\pm0.02$	$6.4\pm0.04$	$3 \pm 0.2$	$1.42\pm0.02$	$0.76\pm0.008$	$25 \pm 0.2$	$11.8\pm0.03$	$818.8\pm5$	$65.4\pm3.6$	$31.8\pm0.7$
3	$26.6\pm0.1$	$1.26\pm0.04$	$6.8\pm0.03$	$3 \pm 0.1$	$1.54\pm0.04$	$0.82\pm0.004$	$25 \pm 0.5$	$12.6\pm0.04$	$856.6\pm6$	$67.8 \pm 4$	$36.6\pm0.2$
4	$33.4\pm0.3$	$1.64\pm0.03$	$7.2 \pm 0.07$	$3 \pm 0.3$	$1.66\pm0.03$	$0.88\pm0.007$	$25 \pm 0.2$	$13.4\pm0.09$	$894.4 \pm 5$	$70.2 \pm 3$	$41.4\pm0.3$
5	$40.2\pm0.2$	$2.02\pm0.08$	$7.6\pm0.02$	$3 \pm 0.2$	$1.78\pm0.06$	$0.94\pm0.002$	$25 \pm 0.2$	$14.2\pm0.1$	$932.2 \pm 2$	$72.6 \pm 5$	$46.2\pm0.5$
6	$47 \pm 0.3$	$2.4\pm0.05$	$8 \pm 0.1$	$3 \pm 0.1$	$1.9\pm0.04$	$1 \pm 0.003$	$25 \pm 0.3$	$15 \pm 0.08$	$970 \pm 8$	$75 \pm 6.8$	$51 \pm 0.4$
7	$44\pm0.14$	$3.4\pm0.07$	$17 \pm 0.06$	$7 \pm 0.2$	$2.4\pm0.05$	$1 \pm 0.005$	$26\pm0.2$	$11 \pm 0.04$	$334 \pm 5$	$22 \pm 0.8$	$48\pm0.6$
Substrate + 1%	Substrate + 1% of supplements										
1	$10.08\pm0.1$	$1.18\pm0.05$	$6.0 \pm 0.02$	$19.53 \pm 0.4$	$2.47\pm0.02$	$2.06\pm0.05$	$16.2 \pm 0.4$	$6.14\pm0.04$	$358 \pm 2$	$338.1\pm10$	$42.5 \pm 1$
2	$10.27\pm0.3$	$1.19\pm0.03$	$6.01\pm0.04$	$19.53 \pm 0.1$	$2.47\pm0.05$	$2.06\pm0.03$	$16.25\pm0.3$	$6.16\pm0.07$	$359 \pm 5$	$338.2 \pm 2$	$42.6\pm0.3$
3	$10.46\pm0.2$	$1.20\pm0.03$	$6.02\pm0.03$	$19.53 \pm 0.2$	$2.47\pm0.04$	$2.06\pm0.06$	$15.25\pm0.5$	$6.18\pm0.05$	$360 \pm 8$	$338.2\pm 5$	$42.8\pm0.8$
4	$10.65\pm0.4$	$1.21\pm0.08$	$6.03 \pm 0.07$	$19.53\pm0.3$	$2.48\pm0.08$	$2.07\pm0.02$	$16.25\pm0.8$	$6.21\pm0.04$	$361 \pm 4$	$338.3 \pm 7$	$42.9\pm0.6$
5	$10.84\pm0.2$	$1.22\pm0.04$	$6.04\pm0.06$	$19.53 \pm 0.1$	$2.48\pm0.04$	$2.07\pm0.09$	$16.25\pm0.4$	$6.23 \pm 0.02$	$362 \pm 9$	$338.3\pm14$	$43.09\pm0.2$
6	$11.03\pm0.4$	$1.23\pm0.03$	$6.06\pm0.04$	$19.52\pm0.4$	$2.48\pm0.06$	$2.07\pm0.02$	$16.25\pm0.1$	$6.25\pm0.06$	$363 \pm 7$	$338.4\pm9$	$43.2 \pm 2$
7	$10.94\pm0.5$	$1.26\pm0.08$	$6.3 \pm 0.07$	$19.64\pm0.3$	$2.50\pm0.08$	$2.07\pm0.05$	$16.28\pm0.8$	$6.14\pm0.01$	$345 \pm 5$	$337.0 \pm 27$	$43.1\pm1.2$
Substrate	$10 \pm 0.3$	$1.2\pm0.02$	$6 \pm 0.03$	$20 \pm 0.2$	$2.5\pm0.07$	$2.1\pm0.04$	$16\pm0.5$	$6\pm0.02$	$346 \pm 2$	$346 \pm 23$	$43\pm0.5$
Substrate + 2%	Substrate + 2% of supplements										
1	$10.16\pm0.4$	$1.16\pm0.05$	$6.0\pm0.08$	$19.09\pm0.7$	$2.44\pm0.02$	$2.02\pm0.05$	$16.48\pm0.7$	$6.27\pm0.06$	$369 \pm 6$	$330 \pm 7$	$42.1\pm2.8$
2	$10.53\pm0.7$	$1.18\pm0.09$	$6.02\pm0.02$	$19.08\pm0.2$	$2.44\pm0.07$	$2.03\pm0.07$	$16.49\pm0.6$	$6.31\pm0.07$	$371 \pm 7$	$330.8\pm3$	$42.4\pm1.4$
3	$10.90\pm0.3$	$1.20\pm0.02$	$6.04\pm0.05$	$19.08\pm0.4$	$2.45\pm0.04$	$2.03\pm0.1$	$16.49\pm0.3$	$6.36\pm0.2$	$373 \pm 9$	$330.9\pm11$	$42.6\pm1.1$
4	$11.27\pm0.2$	$1.22\pm0.07$	$6.07\pm0.04$	$19.08\pm0.5$	$2.45\pm0.05$	$2.03\pm0.06$	$16.49\pm0.7$	$6.40\pm0.09$	$375 \pm 5$	$331.0\pm 6$	$42.9\pm2.8$
5	$11.64\pm0.5$	$1.24\pm0.05$	$6.09\pm0.08$	$19.07\pm0.8$	$2.46\pm0.02$	$2.04\pm0.02$	$16.49\pm0.5$	$6.45\pm0.2$	$377 \pm 1$	$331.1\pm18$	$43.18\pm1.6$
6	$12.01\pm0.3$	$1.27\pm0.03$	$6.11\pm0.06$	$19.07\pm0.1$	$2.47\pm0.09$	$2.04\pm0.08$	$16.49\pm0.8$	$6.49\pm0.03$	$379 \pm 4$	$331.2\pm28$	$43.4\pm3.1$
7	$11.83\pm0.6$	$1.32\pm0.06$	$6.59\pm0.07$	$19.30\pm0.6$	$2.49\pm0.2$	$2.04\pm0.08$	$16.54\pm0.1$	$6.27\pm0.04$	$345 \pm 8$	$328.5 \pm 9$	$43.27\pm0.7$
Substrate	$10.3\pm0.09$	$1.9\pm0.04$	$7 \pm 0.09$	$20 \pm 0.3$	$2.8\pm0.06$	$1.6\pm0.06$	$15\pm0.2$	$6\pm0.05$	$362 \pm 12$	$321 \pm 11$	$42 \pm 1$

*Treatment 1*, 100% hulls of peanut; 2, 80% hulls of peanut + 20% nuts of peanut; 3, 60% hulls of peanut + 40% nuts of peanut; 4, 40% hulls of peanut; 5, 20% hulls of peanut + 80% nuts of peanut; 6, 100% nuts of peanut; 7, Spawn Mate II SE ( $\otimes$ ; *substrate*, control (without supplement). Each value is expressed as mean  $\pm$  standard deviation (*n* = 3)

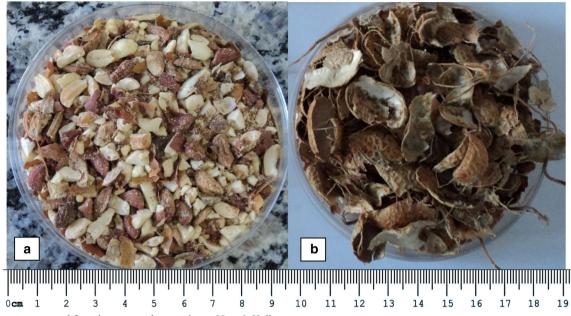


Fig. 1 Peanut waste used for substrate supplementation. a Nuts. b Hulls

was selected in function of the scale of the mushroom production by the grower/company (amount superior to 5 tons of fresh mushrooms harvested per month). The procedures adopted by Zied et al. [21] and Zied et al. [22] were followed for spawn production. The strain was deposited in the public culture collection of São Paulo State University, Campus de Dracena, with open access to other researchers who are interested.

After phase II process or "at spawning", the substrate cooled down and then was mixed with supplements (rate of 1 and 2% of the wet substrate) and the spawn (2% of the wet substrate) homogeneously. The mixture was packed into plastic bags (4 kg wet substrate) and subsequently incubated for 13 days in the greenhouse used specifically for *P. ostreatus* growth, at  $75 \pm 5\%$  relative humidity, and without ventilation. Under these conditions, the substrate temperature was kept at  $28 \pm 1$  °C.

#### Growing cycle

The first flush started after 16 days of spawn run, the second after 29 days, and the third after 37 days, with the growing cycle lasting a total of 45 days. The temperature and relative humidity during the harvest phases were  $24 \pm 3$  °C and  $80 \pm 10\%$ , respectively. The mushrooms were collected twice a day during each flush, weighed and counted for the analysis of the production parameters.

#### Parameters evaluated

The following production parameters were evaluated: (i) the yield calculated as 100 times the fresh weight (f.w.) of mush-rooms divided by the f.w. of substrate, expressed as a

percentage (1st, 2nd, and 3rd flush and total yield); (ii) the number of mushrooms harvested; (iii) the weight per mushroom, expressed in grams (total fresh weight harvested during the cycle divided by the number of mushrooms); (iv) the number of clusters harvested; and (v) biological efficiency calculated as 100 times the f.w. of mushrooms divided by the dry weight of substrate, expressed as a percentage, as previously described by Royse [23] and Pardo-Giménez et al. [12].

#### **Statistical analyses**

Two experiments were performed in a completely randomized blocks design (8 treatments: 6 supplements based with peanut waste + commercial supplement + control), with 6 replicates per treatment (a plastic bag with 4 kg wet weight). The only difference between 1st and 2nd experiment was the amount of supplement added to the substrate (1 and 2% wet weight or 40 and 80 g of the supplement per bag). The means of each production parameter were compared by the least significant difference (LSD) test at p < 0.05. Sigma Stat 3.5 software was used to calculate linear correlations amongst the values for yield, number and weight of mushroom, and the macro- and micronutrients content of the treatments.

#### Results

Comparing the amount of macro and micronutrients in the supplements and in the substrate "control," we verified that the supplements had superior amounts of N, B, Cu, and Fe (except the commercial supplement that had a Fe value of  $334 \text{ mg kg}^{-1}$ , close to that observed in the substrate, 1st

experiment: 346 mg kg<sup>-1</sup> and 2nd experiment: 362 mg kg<sup>-1</sup>). On the other hand, the substrate has a superior amount of Ca, Mg, S, and Mn (except the commercial supplement that had a Mg value of 2.4 g kg<sup>-1</sup>, close to that observed in the substrate, 1st experiment: 2.5 g kg<sup>-1</sup> and 2nd experiment: 2.8 g kg<sup>-1</sup>). In relation to values between hulls of peanut and substrate, it was verified that the hulls of peanut had a lower amount of P and Zn and similar content of K (Table 1).

The application of supplements in the 1st experiment (supplement's 1% w.w.) provided different results compared with the production parameters (Table 2). In the 1st flush, treatment 3 provided the superior yield; in 2nd flush, treatments 1, 5, and 6 provided superior yields; and finally in 3rd flush, treatments 2, 3, 4, 5, and 6 provided superior yield. Neither the commercial supplement nor the substrate control provided superior yield when comparing the flushes separately (1st, 2nd, and 3rd).

Total yield of the supplemented substrate with peanut waste varied from 20.28 to 25.39% while the commercial supplement provided a yield of 22.01%. Both of these results were superior when compared with the substrate without supplement (treatment 8). The only treatment that differed significantly was treatment 5 (20% hulls of peanut + 80% nuts of peanut) when compared with treatment 8 (substrate control), due to the high yield in the 1st, 2nd, and 3rd flush. Separating the obtained results of statistical analysis amongst the treatments, two groups varied in yield values of 20.07 to 25.39% (group a) and of 19.18 to 23.75% (group b); in this sense, it was verified that all treatments that were supplemented are in group a, providing positive physiological responses.

The number of mushrooms and clusters of mushroom did not present a significant difference between treatments. However, the mushroom weight differed, so treatments 4 and 7 presented higher mushroom size than treatments 1, 3, and 8. The substrate non-supplemented provided a high number of clusters, though inferior in mushroom size. In the 2nd experiment (supplement's 2% w.w.), treatment 3 provided the superior yield in the 1st flush; treatments 4, 6, and 7 provided superior yield in the second flush; and finally treatment 5 provided superior yield in the 3rd flush. In both experiments, treatment 3 presented a superior yield at 1st flush and treatment 5 at 3rd flush (Table 3).

Statistical significant differences were verified in treatments 1, 3, 4, 5, 6, and 7 with a high total yield comparable to substrate control (without supplement). The inferior total yield of the substrate without supplement was due to the low yield in 2nd and 3rd flush, which indicates the 2% supplement improved yields in the 2nd and 3rd flush. The treatments that obtained superior yield resulted in a higher number of mushrooms harvested, with a positive correlation (r = 0.829 and p = 0.0108). In both experiments, treatment 3 showed inferior sizes of mushroom.

The supplements based with peanut waste combinations provided superior total yields than a commercial supplement, although not statistically significant. Treatments with higher percentages of nuts (5 and 6) provided superior total yields when compared with treatments with a higher percentage of hulls (1 and 2). When results referring to total yield and biological efficiency for various treatments were compared, they were congruent and presented similar (statistically significant) differences (Table 4).

Linear correlations of the production parameters and macro and micronutrients content of the treatments in the 1st experiment showed a positive correlation between yield and N content (r = 0.744 and p = 0.034), and amongst weight of mushrooms and P (r = 0.867 and p = 0.0005) and K contents (r = 0.960 and p = 0.0002). In the 2nd experiment, a positive correlation existed between the mushroom yield and Cu content (r = 0.875 and p = 0.004), while a negative correlation between yield and Mn content (r = 0.7171 and p = 0.045) were also observed (Table 4).

Treatments	1st flush (%)	2nd flush (%)	3rd flush (%)	Yield (%)	Number of mushrooms ( <i>u</i> )	Weight of mushrooms (g)	Cluster of mushrooms ( <i>u</i> )
1	8.65 <sup>abc</sup>	8.83 <sup>a</sup>	2.67 <sup>b</sup>	20.28 <sup>ab</sup>	147	2.87 <sup>b</sup>	11.33
2	6.55 <sup>c</sup>	7.97 <sup>ab</sup>	6.85 <sup>a</sup>	21.37 <sup>ab</sup>	153	3.02 <sup>ab</sup>	13.66
3	12.51 <sup>a</sup>	2.21 <sup>c</sup>	6.00 <sup>a</sup>	20.78 <sup>ab</sup>	151	2.86 <sup>b</sup>	12.60
4	9.75 <sup>abc</sup>	3.73 <sup>bc</sup>	6.58 <sup>a</sup>	20.07 <sup>ab</sup>	142	3.92 <sup>a</sup>	11.75
5	11.09 <sup>ab</sup>	8.43 <sup>a</sup>	5.81 <sup>a</sup>	25.39 <sup>a</sup>	161	3.15 <sup>ab</sup>	14.66
6	8.01 <sup>bc</sup>	8.72 <sup>a</sup>	7.00 <sup>a</sup>	23.75 <sup>ab</sup>	159	3.18 <sup>ab</sup>	12.0
7	10.39 <sup>abc</sup>	6.76 <sup>ab</sup>	4.86 <sup>ab</sup>	22.01 <sup>ab</sup>	136	3.97 <sup>a</sup>	11.33
8	7.95 <sup>bc</sup>	6.03 <sup>ab</sup>	5.2 <sup>ab</sup>	19.18 <sup>b</sup>	151	2.69 <sup>b</sup>	15
Mean	9.43	5.93	5.57	21.41	149	3.10	12.37

 Table 2
 Production parameters of the treatments supplemented with 1% of wet weight of the substrate

*Treatment 1*, 100% hulls of peanut; 2, 80% hulls of peanut + 20% nuts of peanut; 3, 60% hulls of peanut + 40% nuts of peanut; 4, 40% hulls of peanut + 60% nuts of peanut; 5, 20% hulls of peanut + 80% nuts of peanut; 6, 100% nuts of peanut; 7, Spawn Mate II SE (); 8, substrate control (without supplement). Values followed by different lowercase letters within a column are significantly different at p < 0.05

Treatments	1st flush (%)	2nd flush (%)	3rd flush (%)	Yield (%)	Number of mushrooms ( <i>u</i> )	Weight of mushrooms (g)	Cluster of mushrooms ( <i>u</i> )
1	10.07 <sup>ab</sup>	7.35 <sup>ab</sup>	4.76 <sup>c</sup>	22.18 <sup>ab</sup>	167 <sup>ab</sup>	2.70 <sup>b</sup>	15.75
2	8.27 <sup>b</sup>	5.20 <sup>ab</sup>	5.81 <sup>abc</sup>	19.29 <sup>b</sup>	116 <sup>b</sup>	3.70 <sup>a</sup>	11.66
3	13.06 <sup>a</sup>	4.38 <sup>ab</sup>	6.60 <sup>abc</sup>	24.05 <sup>a</sup>	186 <sup>a</sup>	2.67 <sup>b</sup>	15.83
4	7.01 <sup>b</sup>	7.91 <sup>a</sup>	6.95 <sup>abc</sup>	21.88 <sup>ab</sup>	136 <sup>ab</sup>	3.40 <sup>ab</sup>	12.66
5	8.80 <sup>ab</sup>	5.67 <sup>ab</sup>	8.53 <sup>a</sup>	23.00 <sup>ab</sup>	150 <sup>ab</sup>	3.15 <sup>ab</sup>	15.40
6	10.90 <sup>ab</sup>	7.85 <sup>a</sup>	7.27 <sup>abc</sup>	26.04 <sup>a</sup>	171 <sup>a</sup>	3.06 <sup>ab</sup>	14.66
7	8.55 <sup>ab</sup>	7.83 <sup>a</sup>	5.21 <sup>bc</sup>	21.60 <sup>ab</sup>	151 <sup>ab</sup>	2.89 <sup>ab</sup>	11.25
8	9.11 <sup>ab</sup>	2.68 <sup>b</sup>	4.33 <sup>c</sup>	16.12 <sup>c</sup>	120 <sup>ab</sup>	2.79 <sup>ab</sup>	11.66
Mean	9.47	5.89	6.60	21.96	152	3.05	13.61

 Table 3
 Production parameters of the treatments supplemented with 2% of wet weight of the substrate

*Treatments 1*, 100% hulls of peanut; 2, 80% hulls of peanut + 20% nuts of peanut; 3, 60% hulls of peanut + 40% nuts of peanut; 4, 40% hulls of peanut + 60% nuts of peanut; 5, 20% hulls of peanut + 80% nuts of peanut; 6, 100% nuts of peanut; 7, Spawn Mate II SE B; 8, substrate control (without supplement). Values followed by different lowercase letters within a column are significantly different at p < 0.05

### Discussion

Currently the amount of peanut waste (hulls and nuts) has been increasing due to the strict classification made to meet the maximum quality in the export standard of the peanut. Often this classification discards hulls and broken and defective grains (nuts with and without skin). These residues are currently being used for animal feeding [24], and human feeding is not indicated due to the presence of impurities and even aflatoxin [25, 26]. In this sense, it is fundamental to search for practices that convert these wastes into food quality (closing the cycle of production) for the population, e.g., the bioconversion in fungi protein, known popularly as mushroom.

*P. ostreatus* is the most commonly produced mushroom in Brazil and the 3rd most-produced mushroom in the world, representing approximately 19% of the total world production [27]. In this sense, the application of peanut waste for the supplementation of the substrate at spawning can be an important alternative of food generation, due to the efficiency in the production parameters of this mushroom, as verified in this manuscript, especially in countries where commercial supplements are not available.

Philippoussis et al. [28] studying the use of residues of economic importance in subtropical and temperate countries evaluated the possibility of adding peanut hulls in the substrate formulation of *Pleurotus* spp. production. The authors verified a low biological efficiency, with values of 13.57% for *P. ostreatus*, 18.46% for *Pleurotus pulmonarius*, and 15.31%

Table 4Biological efficiency andincrease in biological efficiencydue to supplementation with 1and 2% of wet weight of thesubstrate

Experiment	Biological efficiency (%)	Increase in biological efficiency* (%)	Biological efficiency (%)	Increase in biological efficiency* (%)
Treatments	1% supplement (	1st experiment)	2% supplement (2	2nd experiment)
1	131.9 <sup>ab</sup>	5	144.2 <sup>ab</sup>	38
2	139.1 <sup>ab</sup>	11	125.4 <sup>b</sup>	20
3	135.2 <sup>ab</sup>	8	156.6 <sup>a</sup>	49
4	130.6 <sup>ab</sup>	4	142.3 <sup>ab</sup>	36
5	165.1 <sup>a</sup>	32	149.8 <sup>ab</sup>	42
6	154.7 <sup>ab</sup>	24	169.3 <sup>a</sup>	61
7	143.3 <sup>ab</sup>	15	140.1 <sup>ab</sup>	34
8	124.7 <sup>b</sup>	0 <sup>(substrate control)</sup>	104.8 <sup>c</sup>	0 <sup>(substrate control)</sup>
Mean	139.4	_	142.9	-

*Treatments 1*, 100% hulls of peanut; 2, 80% hulls of peanut + 20% nuts of peanut; 3, 60% hulls of peanut + 40% nuts of peanut; 4, 40% hulls of peanut + 60% nuts of peanut; 5, 20% hulls of peanut + 80% nuts of peanut; 6, 100% nuts of peanut; 7, Spawn Mate II SE (); 8, substrate control (without supplement). Values followed by different lowercase letters within a column are significantly different at p < 0.05. \*Percentage of increase in the biological efficiency of the supplemented treatments in relation to the substrate without supplement

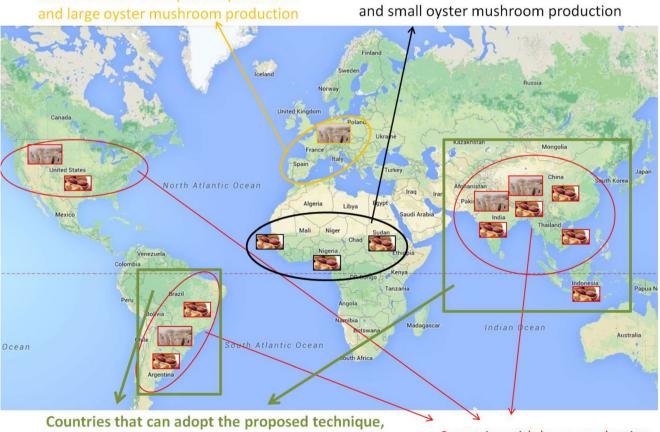
for Pleurotus eryngii. Therefore, in our work, we proposed the use of peanut residues as a supplement, not in the substrate formulation, and we have observed a positive effect.

Increased biological efficiency by 5, 11, 8, 4, 32, and 24% in the 1st experiment and by 38, 20, 49, 36, 42, and 61% in the experiment 2 were verified, for treatments 1, 2, 3, 4, 5, and 6 respectively, when compared with biological efficiency obtained in the substrate control (Table 4). Commercial supplements also provided yield increases, for example, in the 1st experiment (15%) and 2nd experiment (33%). The rate of 2% supplement showed better results in biological efficiency than 1% supplement. Royse et al. [29] studying the effect of the commercial supplement Campbell's S41 at various levels verified increasing values of biological efficiency from 90.5% (substrate control) to 102.6% using the supplement at a rate of 3%. As supplement level increased up to 3%, the biological efficiency decreased.

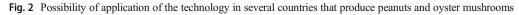
Depending on the origin of the waste material and on the supplement rate, authors have published different results. Narh Mensah et al. [30] obtained biological efficiency of 78.8% using powdered pineapple rind supplement and 65.2% with substrate control. Pardo-Gimenez et al. [31] reported biological efficiency of 100.3, 109.8, 126.9, and 127.6%, respectively, for the control substrate (without supplement) and supplemented with a defatted almond meal at a rate of 0.5%, 1.0%, and 1.5%.

It is important to note that the peanut residue is a material found in some countries (China, India, Nigeria, USA, Indonesia, Brazil, Argentina, and others) with low commercial value, or even with no commercial value. The only treatment that must be performed for its utilization as a supplement is drying (at a temperature of 68 °C for 24 h) and crushing. The drying process, besides being used to remove moisture excess from hulls and nuts, also serves as pasteurization (or heat treatment), reducing the possible presence of contaminants and pests that may influence the mushroom cultivation. Thus, we suggest the use of peanut residue in countries that do not have companies with commercial representation of substrate supplements for mushroom production. Countries with commercial representation may also use peanut residue for substrate supplementation (Fig. 2).

There is a small change in the amount of micro and macronutrient in the final substrate mixed to the supplements (1 and



#### Countries with large production with large production of peanut and oyster mushroom of peanut and oyster mushroom and without companies of mushroom supplements



## Countries with small peanut production

Countries with large peanut production

2%). The N content in the supplement is very high compared with the substrate, while the commercial supplement has 44 g kg<sup>-1</sup> of N, the substrate has only 10 g kg<sup>-1</sup> of N; so in the 1st experiment, the N content of treatment 7 increased by 0.94 g kg<sup>-1</sup> with the addition of commercial supplement. A positive correlation was found in the 1st experiment, which showed much greater N availability improved yields, but in the 2nd experiment we doubled the amount of supplement and not found a correlation between yield and N content, although the yield gains were greater than those obtained in 1st experiment.

The supplementation of compost with soybean base products is common in the cultivation of *A. bisporus*. The majority of modern supplements are based on protein-rich vegetablebased raw materials with contents rich in N [18]. As a result of this, we understand that N content that is important for mushroom growth, but there are also other nutrients involved during the cultivation.

The positive correlation was found between the weight of mushrooms and the amount of P and K of the treatments (1st experiment), showing the importance of the macronutrients in the nutrition of fungi. Micronutrients can also improve or reduce yield which was verified with the positive and negative correlations found in the 2nd experiment. The values of Cu and Mn ranged from 6 to 6.49 mg kg<sup>-1</sup> and 127–331.2 mg kg<sup>-1</sup>, respectively between the treatments. Consequently supplements rich in Cu and poor in Mn should be used, but the same correlations with micronutrients were not found in the 1st experiment.

Rodriguez Estrada and Royse [32] verified the positive effect of the Cu; however, different from the present study, the Mn also presented a positive correlation when applied to the substrate on the yield of *Pleurotus eryngii*. Zied et al. [33] studying the effect of micronutrients in the supplementation of the *Agaricus subrufescens* production verified that Cu and Mn also showed a positive correlation with yield; nevertheless, the Mn values of compost were lower  $(141 \pm 4.9 \text{ mg kg}^{-1})$  than the present study.

No correlation was repeated between the eight treatments in the 1st and 2nd experiment regarding the production parameters and the macro and micronutrients specifically. It is difficult to understand the dynamics that occurred with the addition of the supplements in the substrate and hence further studies are recommended.

#### Conclusion

The use of peanut waste can be used as a supplement in the substrate during the production of *P. ostreatus*, allowing the closing cycle of quality food production. A better combination can be reached with a mix of 20% hulls and 80% or 100% nuts. The addition of 2% supplement in the substrate provided greater yield than 1% supplement.

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