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Application of water to the mushroom colonized compost prior the addition of the casing layer

Aplicação de água no composto de cogumelos colonizado antes da adição da camada de cobertura

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Abstract

The mushroom *Agaricus bisporus* is produced on all the continents. To increase yield and efficiency during the process of mushroom production, and reduce costs and size of the mushroom farm, production of fully colonized compost in specialized chambers (a.k.a. Phase III compost) evolved. One challenge in this technology is achieving appropriate substrate moisture. Average moisture content at the end of pasteurization and conditioning in this kind of chamber is approximately 60%. The aim of this study was evaluate the effect of the addiction of different levels of irrigation before casing onto the surface of A. bisporus PIII compost. Five irrigation rates were applied to the surface of the colonized compost: T1 - control (60% moisture); T2 - 200 ml box^{-1} (62% moisture); T3 - 400 ml box^{-1} (63.5% moisture); T4 - 800 ml box^{-1} (67% moisture); T5 - 1.600 ml box^{-1} (74% moisture); T6 - 3.200 ml box^{-1} (87% moisture). Mushroom yield increased with increasing moisture levels in the compost, obtaining higher values for the treatment 3200 ml box^{-1}. The application of water to the colonized compost can become a low cost practice in a mushroom commercial production.

Additional keywords: *Agaricus bisporus*; button mushroom; compost phase III; high technology; yield

Resumo

O cogumelo Agaricus bisporus é produzido em todos os continentes. Para aumentar a produtividade e eficiência biológica durante a produção e reduzir custos e tamanho da fazenda de cogumelos, a produção de composto totalmente colonizado em câmaras especializadas (também conhecido como composto fase III) evoluiu. Um desafio nessa tecnologia é alcançar a umidade ideal do substrato. O teor médio de umidade ao final da pasteurização e condicionamento nesse tipo de câmara é de aproximadamente 60%. O objetivo desse estudo foi avaliar o efeito da adição de diferentes níveis de irrigação antes da adição da camada de cobertura em composto fase III de A. bisporus. Cinco taxas de irrigação foram aplicadas na superfície do composto colonizado: T1 –controle (60% de umidade); T2 – 200 ml caixa^{-1} (62% de umidade); T3 – 400 ml caixa^{-1} (63,5% de umidade); T4 – 800 ml caixa-1 (67% de umidade); T5 – 1600 ml caixa-1 (74% de umidade); T6 – 3200 ml caixa^{-1} (87% de umidade). A produtividade dos cogumelos aumentou com o incremento de umidade no composto, obtendo maiores valores para o tratamento 3200 ml caixa^{-1}. A adição de água no composto colonizado pode se tornar uma prática de baixo custo no cultivo de cogumelos.

Palavras-chave adicionais: *Agaricus bisporus*; cogumelo botão; composto fase III; alta tecnologia; produtividade.

Introduction

The mushroom *Agaricus bisporus* (Lange) Imbach is cultivated on all continents. As a saprophyte this mushroom requires a balanced substrate for its nutrition (Bhushan and Kulshreshtha 2018; McGee 2018; Suwannarach et al., 2022; Zhang et al., 2019). Water is the second most important constituent for the metabolic development of this fungus. These mushrooms are composed of 85-95% moisture by weight (Harshavardhini & Sharma, 2021; Zhang et al., 2018).

Commercial mushrooms have been produced in a protected environment for centuries. Some of the first records indicate they were produced in limestone caves in France during the reign of Louis 14th (Aydoğdu et al., 2020; Dhar, 2017). Later the composted substrate was prepared outside of any building and then it was transferred inside a constructed facility to complete the composting process. There it remained until the crop was terminated. This single-zone system has been replaced by multi-zone systems whereby the different stages of the process (composting, pasteurization, spawn-run and production) are all carried out in specialized chambers. The multi-zone system together with the advancements in computerized control has increased the efficiency and the annual yield per growing chamber. There are basically four stages (a.k.a. phases): Phase I (composting of the raw ingredients), Phase II, (pasteurization and continuation of composting), Phase III, (inoculation and colonization of the substrate with the spawn) and Phase IV (casing and production).

Although the advances in system technologies have increased overall productivity, these have also created challenges. Since the substrate is physically handled less than in the original single-zone system, achieving appropriate compost density and moisture have become challenges. According to Zied et al. (2011a) an ideal moisture level of spawn-run compost at casing for high productivity is 67%. However, Seaby (1995) showed that an increase in moisture from 63.8% to 73.7% was associated with a reduction in mushroom yield. Mushroom producers as a matter of personal judgment have added water routinely to compost at spawning or casing for decades. The aim of this study was to evaluate the effect of the addition of water to spawn-run compost before addition of the casing in *A. bisporus*.

Material and methods

Spawn

Button mushrooms (ABI 11/21 strain) were collected from a commercial company, Compobras® in the city of Castro, in Parana State (Brazil). This strain is characterized by mushrooms with high texture, white, smooth, and with lamellae that do not easily break (Zied et al., 2015). Grain spawn was produced on sorghum seeds, as described by Zied et al. (2018a). Briefly, the seeds were boiled at 100°C for 30 h and then placed (0.5 kg wet weight) in polyethylene bags and mixed with $CaSO_4$ (1%) and limestone (0.5%). Afterwards, the bags were inoculated and incubated in a dark room at 20°C for 15 days.

Compost

Commercial compost was prepared with sugar cane bagasse, wheat straw, chicken manure, urea, limestone and gypsum. Phase I (PI) was held in bunker for 16 days. Both Phase II (PII) and III (PIII) were completed in a pasteurization tunnel that lasted 7 and 10 days, respectively. At the end of PII, the compost had a pH of 7.6, moisture of 60%, N content of 2.4% and C/N ratio of 17.6/1. The substrate was spawned at the rate of 1% wet weight of compost. After colonization, 4.5 kg (wet weight) of substrate was transferred and pressed into the plastic box (measure – 30 cm wide x 30 cm long x 30 cm high, which totals 0.09 m^2). So, the compost area of production surface was 50 kg of compound m^{-2}. The compost surface was then irrigated with five rates of water with no mixing: T1 - control (60% moisture); T2 - 200 ml box^{-1} (62% moisture); T3 - 400 ml box^{-1} (63.5% moisture); T4 - 800 ml box^{-1} (67% moisture); T5 - 1.600 ml box^{-1} (74% moisture); T6 - 3.200 ml box^{-1} (87% moisture).

Casing layer

A Brazilian commercial peat more $CaCo_3$ (maisterra®, Castro, Parana, Brazil) was used as a casing to cover the spawn-run compost. The material was applied 3 cm deep. The physico-chemical characteristics of the peat were pH 7.8, electrical conductivity 206 µS cm^{-1}, organic matter 712 g kg^{-1}, total pore space 875 ml l^{-1}, and water holding capacity 1.43 kg kg^{-1}.

Growing cycle

Mushrooms were harvested for 52 days, with the 1st flush occurring 22 to 27 days after casing, the 2nd flush 33 to 37 days after casing and the 3rd flush 43 to 52 days after casing. The air temperature and relative humidity during production were maintained at 19±1°C and 85±5%, respectively. The casing layer humidity was kept between 60 and 70%, according to the methodology of Pardo-Gimenez et al. (2017).The cultivation was conducted in a mushroom production chamber used specifically for *A. bisporus* growth where temperature, relative humidity and CO_2 content are controlled. The mushrooms were harvested twice a day during each flush in the optimal commercial development stage corresponding to morphogenetic stages 2, 3 and 4, and in accordance with the classification established by Hammond and Nichols (1976). Them the mushrooms were weighed and counted for the analysis of the production parameters.

Parameters evaluated

The following production parameters were evaluated: i) the yield expressed as a percentage of substrate fresh weight, calculated by dividing the fresh weight (4.5 kg compost) of the mushrooms by the fresh weight of compost, (1st, 2nd, 3rd flush and total yield); ii) the number of mushrooms (NM) harvested; and iii) the individual mushroom weight (WM), expressed in g (total fresh weight harvested during the cycle divided by the number of mushrooms) as previously described by Pardo-Giménez et al. (2016).

Analyzes in the compost and casing

The following measurements were taken according to Zied et al. (2017): - pH (extraction ratios of 1+5 (V/V) were used); - moisture (drying at 103-105 ºC to a constant weight); - total N content (Kjeldahl method); - C/N ratio (calculation from organic matter and nitrogen contents); - electrical conductivity (extraction ratio of 1+5 (V/V) were used); - organic matter (measuring the loss of weight after calcination at 540 ºC); total of pore space (calculation from dry bulk density and real density); - water holding capacity (saturation and drainage method).

Statistical Analyses

The experiment was carried out in a randomized block design containing 6 treatments, each one represented by 5 replicates (plastic boxes with 4.5 kg of wet compost), which provided 42 experiment units. ANOVA was used to analys the data, and the Tukey test was used to separate means (P = 0.05). All calculations were performed using the Statgraphics Plus software, v. 4.1 (Statistical Graphics Corp., Princeton, NJ, USA). Finally, linear regression analysis and Person correlation were performed using SAS statistical software (SAS Institute Inc., Cary, NC, USA).

Results

The volume of water applied to the spawn-run compost significantly affected mushroom yield. In the 1st flush yield increased with increasing rates of water such that the highest rate of 3200 ml box^{-1} nearly doubling the yield (11.13% to 19.01%) (Table 1). Only this high rate was significantly different from the control. In the 2nd flush again, the irrigation treatments were empirically higher than the control but none were significantly different from the unirrigated compost. The 3rd and last flush provided low yield, which is typical for commercial producers. Current production practices harvest two flushes because of the low 3rd flush production.

Total yield clearly demonstrated the efficiency of irrigation doses applied to the colonized compost prior to application of the casing. Increasing irrigation rates of 200, 400, 800, 1600 and 3200 ml box-1 demonstrated an increase in yield of 37, 21, 52, 43, 67% over the control (Figure 1). Only 800 and 3200 ml box^{-1} were significantly greater in productivity than the control. The relationship between yield and water addition to the spawn-run compost is positive with an R^2 value of 0.6363 (Figure 2).

Table 1 Parameters evaluated during mushroom cultivation (1st, 2nd, 3rd, yield total, number and weight of mushroom) when varying rates of water were applied to spawn-run compost

Irrigation rate, ml box $^{-1}$	1st flush	2nd flush	3rd flush	total	NM[1]	WM[2]
			Yield, %		u	g
Control	11.13 b	8.91	1.56	21.61 c	39	25.60
200	13.00 ab	15.29	1.37	29.67 abc	49	27.40
400	12.66 ab	13.20	0.32	26.19 bc	44	23.72
800	14.18 ab	15.62	3.06	32.87 ab	56	27.44
1600	14.82 ab	14.08	2.05	30.96 abc	62	23.28
3200	19.01 a	15.73	1.40	36.14 a	55	30.33
Media	14.13	13.81	1.62	29.57	50.72	26.29
LSD	6.51	9.35	3.57	8.73	27.77	7.25
P value	0.0196	0.2447	0.3435	0.0004	0.1797	0.0553

Values followed by different lowercase letters within a column are significantly different among the parameters evaluated at $p < 0.05$, according to Tukey's test. 1NM = number of mushroom. 2WM = weight of mushroom

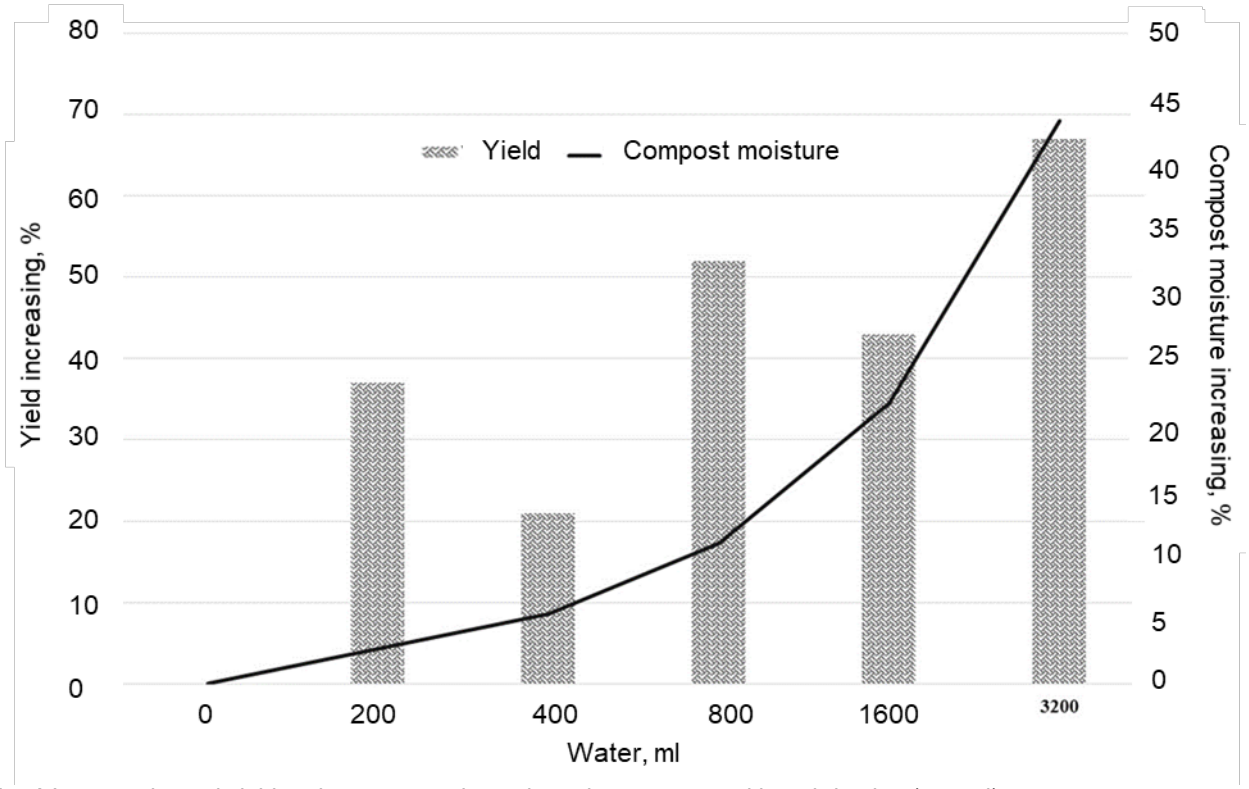

Fig. 1 Increase in total yield and compost moisture based on compost without irrigation (control).

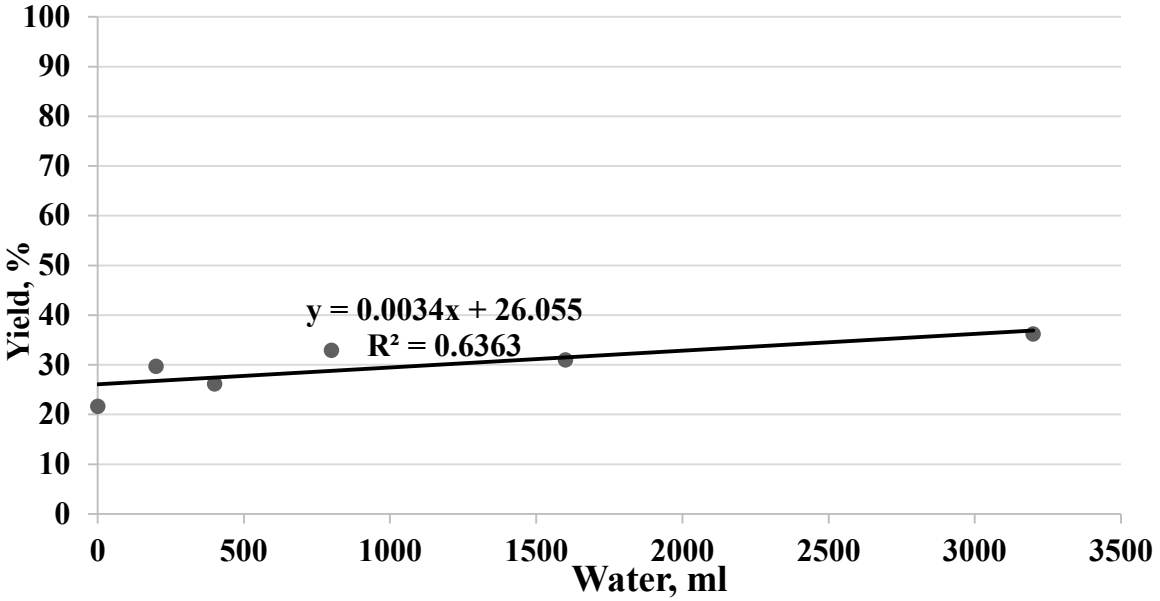

Fig. 2 Regression analysis between total yield and irrigation doses

NM and WM did not differ significantly as a function of the applied irrigation dose. However, there was a positive correlation between total yield and NM (0.8341). A higher yield gave a higher number of mushrooms harvested. Although WM increased empirically with increasing rates, there was no significant correlation between total yield and WM. Thus, an increase in moisture did not adversely affect individual mushroom weight (Table 2).

Table 2 Person correlation coefficients between total yield, number of mushrooms and individual mushroom weight

Pearson correlation	NM[1] (r/ P values)	WM[2] (r/P values)
Yield	**0.8341** **0.0390**	0.6498 0.1625
NM	-	0.1259 0.8122

[1]NM= number of mushrooms. [2]WM = weight of mushroom; N = 7

Discussion

Preparation of fully colonized compost in a specialized chamber is a widely adopted technology throughout Europe (Jurak et al., 2015; Thai et al., 2022) and the Americas. Attributes of quality indicators for compost production of A. bisporus include moisture, C/N ratio, pH, total nitrogen and presence of competitor/indicator organisms such as mites, nematodes and molds (Zied et al., 2011a). Within this context, moisture would be considerate the second important variable in the compost, behind only the C/N ratio. In the past there was great attention given to moisture contents of the compost (Seaby 1995), where levels of approximately 67% before phase I, 65% end phase I, and finally 70% end phase II were recommended (Laborde et al., 1987). However, the concern was whether excess moisture might cause anaerobic zones in the PI compost and/or make it difficult for air to pass through the compost during PII in the pasteurization tunnel.

In this study, we increased the moisture content to 87% with the application of an irrigation dose of 3200 ml box-1, an increase of 43% over the original Phase III compost. Addition of moisture at this time did

not contribute to the problems associated with the augmentation of the moisture content at Phase I or II. In this situation, the irrigation doses had the role of solving the low moisture content of the compost, prior to the addition of the casing layer. The yield improvement is noticeable, especially in the first and second flush. Neither timing to first flush or nor yield was reduced with the higher moisture levels.

Potable water, such as would be used for preparation of the casing and irrigation of the mushroom crop, was used in this study. Use of any other water source could firstly affect consumer health and secondly influence mushroom quality through introduction of competitor or post-crop deterioration organisms.

The simple application of water to the colonized compost which demonstrated an increase in yield even with the lowest rate of 200 ml box-1 (2,2L.m2) can become an important practice in a mushroom commercial operation. Water is low cost item compared to the investments associated with commercial supplements (Schisler & Sinden 1966; Zied et al., 2011b; Pardo et al., 2012; Zied et al., 2018b).

Conclusion

We concluded that mushroom yield increased with increasing moisture levels in the compost. The application of water to the colonized compost can become a low cost practice in a mushroom commercial production.

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