

Potential utilization of spent *Agaricus bisporus* mushroom substrate for seedling production and organic fertilizer in tomato cultivation

Potencial de utilização do substrato de cogumelo *Agaricus bisporus* usado para produção de mudas e adubação orgânica no cultivo de tomate

Carolina Figueiredo Collela¹, Lívia Martinez Abreu Soares Costa¹, Tatiana Silveira Junqueira de Moraes¹, Diego Cunha Zied², Danny Lee Rinker³, Eustáquio Souza Dias^{1*}

¹Universidade Federal de Lavras/UFLA, Departamento de Biologia/DBI, Lavras, MG, Brasil ²Universidade Estadual Paulista/UNESP, Faculdade de Ciências Agrárias e Tecnológicas, Dracena, SP, Brasil ³University of Guelph, Department of Plant Agriculture, Guelph, Ontario, Canada *Corresponding author: esdias@ufla.br

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ABSTRACT

The production of *Agaricus bisporus* results in significant residual material after cultivation. Due to its physical properties and nutrient content Spent Mushroom Substrate (SMS) has great potential for use in agriculture. Our study evaluated the feasibility of using SMS as an alternative substrate for the production of Paronset[®] hybrid tomato seedlings and as an organic fertilizer in its production. To conduct the fruit production experiment, the plots consisted of three types of fertilizers (SMS of *A. bisporus*, NPK and bovine manure) with four replications. The seedlings produced in the SMS presented higher rates of germination time and speeds, and stood out from the other substrates in the evaluation of seedling quality. In the tomato fruit production phase, the average production of the plants originating from the SMS was 20% higher than the seedlings from the commercial substrate. The plants cultivated with the SMS presented higher production than the plants cultivated with bovine manure incorporated the soil. According to our results, The SMS was as good as the commercial substrates tested. Therefore, the SMS can be recommended for the growth and nutrition of seedling production and denotes potential viability for use in the tomato production cycle in its different phases.

Index terms: Champignon; Lycopersicon esculentum; Spent Mushroom Substrate.

RESUMO

A produção de *Agaricus bisporus* resulta em material residual significativo após seu cultivo. O substrato pós-cultivo do cogumelo (SMS), devido às suas propriedades físicas e conteúdo de nutrientes, tem um grande potencial para uso na agricultura. Nosso estudo avaliou a viabilidade do uso do SMS como substrato alternativo para a produção de mudas de tomate híbrido Paronset® e como fertilizante orgânico em sua produção. Para realizar o experimento de produção de frutos, foram utilizados três tipos de fertilizantes (SMS de *A. bisporus*, NPK e esterco bovino) com quatro repetições. As mudas produzidas no SMS apresentaram maiores índices de tempo e velocidade de germinação e se destacaram dos demais substratos na avaliação de qualidade de mudas. Na fase de produção de frutos de tomate, a média de produção das plantas originárias do SMS foi 20% maior que as mudas advindas do substrato comercial. As plantas cultivadas com o SMS apresentaram maior produção que as plantas cultivadas com esterco bovino incorporado ao solo. De acordo com nossos resultados, o SMS foi tão bom quanto o substrato comercial testado. Portanto, o SMS pode ser recomendado para o crescimento e nutrição da produção de mudas e denota potencial viabilidade para uso no ciclo de produção do tomate em suas diferentes fases.

Termos para indexação: Champignon; Lycopersicon esculentum; Spent Mushroom Substrate.

INTRODUCTION

The commercially produced mushroom *Agaricus bisporus* (Lange) Imbach, is one of five species responsible for the majority of world mushroom production. Although worldwide it represent 15% of total production (Royse; Baars; Tan, 2017), in Brasil *A. bisporus* representes 33% of its mushroom production (Sánchez; Zied; Alberto,

2018). The substrate used for its cultivation is a compost of lignocellulosic carbon sources (eg., wheat or rice straw, sugar cane baggase), additional nutrients (eg., animal manures and chemicals) and gypsum. After spawning and spawn run, the colonized compost is covered with a layer of material with good watering holding capability and porosity (casing layer), often peat moss or soil, is used to cover the substrate that has been colonized by 2

the *Agaricus* fungus (Figueiredo; Dias, 2014). After mushroom production decreases (predetermined by a routine schedule), the SMS is removed, and new crop cycle begins in the same chamber.

SMS is often discarded as waste after mushroom harvest in many countries (Chiu et al., 1998; Hanafi et al., 2018). For every 1 kg of mushrooms produced about 5 kg of SMS remains (Zisopoulos et al., 2016). Therefore, SMS management has become a growing challenge for the mushroom production industry. Consequently, finding environmentally and economically sustainable solutions for this organic waste is highly desirable.

Due to its physical properties and nutrient content, SMS has great potential to be employed in the agricultural and horticultural sectors, as well as contribute to reduce the use of non-renewable resources (García-Delgado et al., 2016). SMS also contains a rich microbiota that can ensure the balance of phytosanitary requirements of crop plants, and the possibility that some species of microorganisms may induce resistance in pre-cultivated seedlings in this substrate (Siqueira et al., 2011).

Agricultural land crops are naturally suitable destinations for SMS. Large volumes of SMS can be apllied to land for large scale agricultural crops (Rinker, 2017). These plant crops include tomato, asparagus, beet, cauliflower, cabbage, pepper, celery, cucumber, lettuce (Kim; Kwak, 2012; Kim et al., 2016).

The production of seedlings for successful field transplant is one of the principle steps in the cultivation of tomatoes. The substrate used in their production plays a fundamental role in the initial development of the plant and must present physical and chemical characteristics adequate to the development of the seedling, such as moisture retention, drainage of excesso water and oxygen and nutrients supply (Gomes et al., 2017).

The commercialization of vegetable seedlings is specialized and the development of the production activity is based on combinations of substrates. Organic, mineral and synthetic materials are used that attempt to meet the requirements of each species. Efforts are made to utilize organically local materials, where-by reducing costs and providing local outlets for agricultural by-products (Costa et al., 2015).

Brasil has an abundance of SMS that could be used in vegetable seedling production. In this context, the use of SMS as a substrate for the production of tomato seedlings of the Paronset[®] hybrid and as fertilizer for tomato production was evaluated.

MATERIAL AND METHODS

Tomato seedling experimental design

The experiment was conducted during 35 days at Hidroceres Saplings and Agricultural Productions Ltda, situated in the municipality of Santa Cruz do Rio Pardo/ SP. A Santa Clara class tomato seedlings (Lycopersicon esculentum) hybrid [Paronset® (Syngenta®)] was used in the evaluation of SMS. The trays (162 cells rigid polypropylene in the trapezoidal format with a volume of 31 cm³ per cell and dimensions of 35 mm in length x 35 mm in width per cell) were filled with three types of substrates (SMS of Agaricus bisporus, Carolina II® - Carolina Soil from Brasil and Pindstrup® - Pindstrup Mosebrug A/S). Seeds (1/cell) were planted at a depth of 2-3 mm. Seedlings were grown in a greenhouse on metal benches and irrigated three times a day. Each substrate was replicated eight times with the trays arranged in a randomized complete block design on the greenhouse bench.

The composition of the SMS of *A. bisporus* consisted of bagasse, horse manure, rice straw, soybean meal, poultry litter, urea, potassium chloride, single super phosphate fertilizer, gypsum and peat moss. The SMS was homogenized. The commercial substrates, Carolina II[®] and Pindstrup[®], are composed basically of *Sphagnum* peat, charred rice straw and perlite, and *Sphagnum* peat and perlite, respectively.

Germination evaluation

The trays were monitored daily over 25 days for the first appearance of the tomato cotyledons. The following variables were calculated based on the first day of emergence: Percent Germination (G), Germination speed index (GSI), Mean germination time (MGT), Mean germination speed (VMG) (De Carvalho; De Carvalho, 2009).

Physical parameters and quality index

Thirty days after sowing, ten seedlings were randomly removed per treatment. The seedlings were evaluated for: root length (RL) (mm), shoot height (SH) (cm), plant height (H) (cm), stem diameter (SD) (mm), root dry mass (RDM), dry shoot mass (SDM), total dry mass (TDM) (sum of RDM and SDM), number of leaves (NL) and Dickson quality index (DQI).

The Dickson quality index (DQI) followed, the methodology of Dickson et al. (1960), using the following equation: DQI = [TDM/(SH/SD + SDM/RDM)].

Statistical analysis for seedling germination and growth

The data were submitted to regression analysis and the means separated at the 5% level according to the Skott-Knott test, using the program SISVAR (Ferreira, 2008).

Evaluation of tomato production from three fertilizers

Tomato production from seedlings produced in SMS and Carolina II[®] substrates were evaluated using three fertilizer sources: SMS, a chemical fertilizer (NPK: 4-12-8) and bovine manure. The plants and fertilizers were arranged in a split-plot randomized complete block design with four replications. The whole plots consisted of the three fertilizers types and the two subplots were the seedlings (produced on the SMS and on Carolina II[®] substrates).

The tomato seedlings were planted into soil. The beds were 1.20 m wide by 20.4 m long with plants spaced 60 cm apart. Soil analyses were taken prior to the application of the fertilizers. Fertilizers were incorporated 30 days prior to transplanting. The bovine manure and the SMS of *A. bisporus* were applied manually at 15t ha⁻¹. The mineral fertilizer NPK (4-12-8) was applied at a rate of 10 kg ha⁻¹ according to the crop recommendations Trani et al. (2015). Plants were watered by drip irrigation. The fruits were harvested until the third bunch. Commercially acceptable tomatoes were weighted; all others were discarded. The control of invasive plants and pruning of the tomatoes were done manually. Pests and diseases were controlled with registered products.

Statistical analysis for tomato production

Tomato production data were analysed through regression analysis and the means separated at 5% level according to the Tukey test using the program SISVAR (Ferreira, 2011).

RESULTS AND DISCUSSION

Parameters of germination of tomato seedlings

No significant differences were observed between the substrates for percent germination and GSI (Table 1). However, MGT and, consequently VMG, showed significant differences between treatments.

The MGT of the seedlings was higher (13.09 days) for the SMS, compared to the other substrates (Table 1). Consequently, the VMG values were lower for the SMS. Although two days longer were required for germination in SMS, compared to the commercial substrates, percent germination was not negatively affected.

Table 1: Germination of tomato seeds [Paronset®(Syngenta®)] on different substrates.

Substrate	Germination (%)	GSI	MGT (days)	VMG (days ⁻¹)
Pindstrup®	88.25a	1.25a	10.68a	0.07a
Carolina II®	90.75a	1.20a	11.05a	0.08a
SMS	89.75a	1.28a	13.09b	0.05b
CV (%)	11.63	15.75	8.03	9.07

*Means followed by different letters in the same columm differ significantly at 5% level according to Tukey test.GSI: Index of speed of germination; MGT: Mean germination time; VMG: Average speed of germination.

Demontiêzo et al. (2016) reported that salinity values higher than 2.5dSm⁻¹ resulted in significantly lower values in germination parameters. The electrical conductivity in our experiment was 2.54 dSm⁻¹, a value within the limit of that presented by Demontiêzo et al. (2016). The commercial substrates values were 0.80 and 1.50 dSm⁻¹ (Pindstrup and CarolinaII, respectively). According to Paula et al. (2017) the SMS of Agaricus mushrooms is known to have a high salinity, due to the different supplements added, especially limestone, gypsum and chemical fertilizers. Therefore, for this work, the SMS was composted for 15 days, in order to make it more stable and to decrease its salinity. Despite this, the electrical conductivity value still fell near the value presented by Demontiêzo et al. (2016), which may explain the longer time to germination. In addition, a possible need to adjust the structure and/or fertility of this substrate should be considered in order to further improve its properties as a substrate to produce tomato seedlings.

Physical parameters and quality index of seedlings

The SMS had a DQI of 0.012 while the indices obtained in the commercial substrates were lower, with values of 0.008 and 0.007, Pindstrup[®] and Carolina II[®], respectively (Figure 1).

Dos Santos et al. (2016) evaluated the production of tomato seedlings (cv. Drica) on alternative substrates (eg., carbonized coffee husks) and observed DQI values similar to those obtained in commercial substrates. However, Silva et al. (2012) observed a reduction in DQI in the tomato cultivar (Santa Clara) when carbonized rice hulls were added, which did not serve as a nutrient source but only a physical conditioner of the substrate. In this context, the better DQI value of the tomato seedlings grown in the SMS can be based on the greater supply and availability of nutrientes.

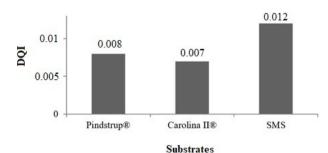


Figure 1: Dickson quality index (DQI) of Paronset[®] hybrid tomato seedlings grown on diferente substrates at 30 days after sowing.

The longer time to germination did not restrict the seedlings in the SMS from having a higher quality index. Despite the possible salinity problem, the SMS had other characteristics that allowed it to be superior to the other substrates. The physical measurements showed taht SMS was superior or equal to the other substrates (Table 2).

The seedlings produced in the SMS of *A. bisporus* were of higher quality because they produced a significantly greater number of leaves which affects all the future development of these plants. Silva et al. (2012) observed that the seedlings produced in organic substrates had greater diameter of the colon, indicating a higher quality of the seedlings after transplanting.

The dry mass of the root of the seedlings produced did not differ significantly between substrates (Table 2). However, the aerial dry mass was greater for SMS, directly reflecting the better results obtained for NL and SH. Lopes et al. (2015) also reported greater SH for tomato seedlings produced on SMS of *A. subrufescens*.

The total porosity of the commercial substrates was higher than the porosity in the SMS (Table 3). This can be explained by the presence of soil in the SMS composition of *A. bisporus*, which can affect its porosity. Clay and silt were detected in the texture in the SMS, corroborating the higher density of this substrate. The water retention capacity was also higher in the SMS, in relation to the commercial substrates tested, indicating high microporosity.

To increase the porosity, many other products, such as vermiculite, could be used to improve the qualities of the physical properties of the SMS. The pH values (6.9) of the SMS, was slightly above the ideal value (6 - 6.5), still within the acceptable range for the culture.

Tomato production

No significant difference in tomato production was observed for the differentt fertilizers (Figure 2). Plants grown in NPK beds showed higher production (4.7 kg/ plant), followed by plants cultivated with SMS (3.7 kg/ plant) and cattle manure (3.4 kg/plant). However, plants grown using SMS produced about 8% more than plants grown with bovine manure.

Based on these results, it can be estimated that the productivity of the Paronset[®] hybrid, using SMS of *A. bisporus* as a biofertilizer, for the traditional and superadded crop system, is 82.5 t/ha and 128.5 t / ha, respectively. For the traditional cultivation system a 12% increase in production was observed. Wamser et al. (2012), observed a production of 133.4 t / ha, which approximates the results estimated in this work, in a superadded crop system.

Physical parameters of tomato seedlings								
Substrate	NL	SD	SH	RL	SDM	RDM	SH/SD	
	Unid	mm	cm		mg		cm/mm	
Pindstrup®	4.5a	2.3a	6.03a	5.32a	8.30a	5.88a	2.62a	
Carolina II®	4.0a	1.9a	5.70a	4.78a	8.05a	6.02a	3.00a	
SMS	5.5b	3.4b	8.76b	5.04a	11.01b	5.83a	2.57a	
Average	4.67	2.53	6.83	5.05	9.12	5.91	2.73	
CV(%)	20.34	13.4	18.74	22.30	17.32	25.13	16.98	

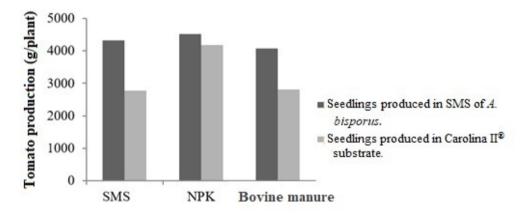
Table 2: Physical parameters of tomato seedlings produced from different substrates, 30 days after sowing.

*Means followed by different letters in the same column differ significantly at 5% level according to Tukey test. NL: Number of leaves; SD: stem diameter; SH: shoot height; RL: root length; SDM: shoot dry mass; RDM: root dry mass.

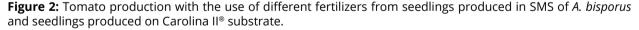
					Parar	neters				
- - Substrate	EC	рН	SOM	CEC	WRC	TP	Density	Clay	Silte	Sand
	dS/m		dag/kg	cmol/dm³	%		kg/m³	dag/kg		
Pindstrup	0.80	5.5	16.87	21.89	57.0	78	217.5	-	-	-
Carolina	1.50	6.3	17.16	12.00	51.0	76	220.0	-	-	-
SMS	2.54	6.9	15.04	58.73	62.3	67	325.0	53.0	30.0	17.0
	К	Р	Ca	Mg	H+Al	P-rem	N-total			
	mg/dm ³		cmol/dm ³		mg/L	g/kg				
Pindstrup	305.0	133.76	10.7	5.9	3.13	38.40	6.6			
Carolina	332.0	126.98	11.3	5.7	4.04	40.52	5.7			
SMS	600.0	542.96	52.7	3.0	1.49	36.13	8.7			

Table 3: Physical-chemical characteristics of substrates Pindstrup[®], Carolina II[®] and SMS of *A. bisporus* used to produce tomato seedlings.

EC: Electric conductivity; pH: Hydrogenionic potential; SOM: Substrate organic matter; CEC: Cation exchange capacity; WRC: Water retention capacity; TP: Total porosity; K: Potassium; P: Phosphorus; Ca: Calcium; Mg: Magnesium; H+AI: Total Acidity; P-rem: Remaining phosphorus.



Fertilizers



The seedlings produced in the SMS of *A. bisporus* presented productivity 20% higher than the seedlings produced in the commercial substratum Carolina II[®] (Figure 2). Lopes et al. (2015), also achieved better production results with tomato seedlings produced in SMS of *A. subrufescens* when compared to the seedlings produced in the commercial substrate Bioplant.

In this context, it can be affirmed that the seedlings produced in the SMS of *A. bisporus* presented greater vigor in relation to the others, thus improving the tomato productivity of these plants in the field. There was a tendency of higher production of the seedlings obtained in substrate based on SMS in the two crops with organic fertilization (SMS and manure). However, when chemical fertilization (NPK) was used, the seedlings produced in SMS practically did not differ in relation to the seedlings produced in the Carolina substrate. These results show that the chemical fertilization with NPK attenuated the differences between the seedlings obtained in the two substrates. On the other hand, for crops with organic fertilization, the seedlings produced in substrates based on SMS may be the most indicated. SMS has relevant concentrations of macronutrients (8.7 g kg⁻¹ N, 542.96 mg/dm³ de P and 600.00 mg/dm³ de K). Therefore, the SMS of *A. bisporus* can be used as an important source of N, P and K for the tomato crop. The SMS of *A. bisporus* presented 62.3% water retention capacity. Thus, its use may contribute to the reduction of soil leaching and consequent reduction of nutrient losses. Thus, in addition to organic fertilizer, SMS can also act as a soil conditioner. Whereas SMS is a byproduct of the *A. bisporus* production chain, its cost is relatively cheap and may be an alternative to cattle or poultry manure.

CONCLUSIONS

In general, the SMS of *Agaricus bisporus* is an excellent substrate for the production of tomato seedlings, as it provides the formation of vigorous and quality seedlings. According to our results, the SMS was as good as the commercial substrates tested. Therefore, the SMS can be recommended for the growth and nutrition of seedling production. In addition, this SMS can also be used as organic fertilizer, mainly as a source of nitrogen, phosphorus, and potassium, for tomato cultivation.

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REFERENCES

- CHIU, S. W. et al. Spent oyster mushroom substrate performs better than many mushroom mycelia in removing the biocide pentachlorophenol. **Mycological Research**, 102(12):1553-1562, 1998.
- COSTA, E. et al. Ambientes e substratos na formação de mudas e produção de frutos de cultivares de tomate cereja. **Horticultura Brasileira**, 33(1):110-118, 2015.
- DE CARVALHO, D. B.; DE CARVALHO, R. I. N. Qualidade fisiológica de sementes de guanxuma em influência do envelhecimento acelerado e da luz. Acta Scientiarum. Agronomy, 31(3):489-494, 2009.
- DEMONTIÊZO, F. L. L. et al. Emergência e crescimento inicial de tomate 'Santa Clara' em função da salinidade e condições de preparo das sementes. Irriga & Inovagri, 1(1):81-92, 2016.

- DICKSON, A. et al. Quality appraisal of whit spruce and whith pine seedling stock in nurseries. **Forestry Chronicle**, 36(1):10-13, 1960.
- DOS SANTOS, A. C. M. et al. Produção de mudas de tomateiro cv. Drica sob substratos alternativos. **Agropecuária Científica no Semiárido**, 11(4):1-12, 2016.
- FERREIRA, D. F. Sisvar: A computer statistical analysis system. Ciência e Agrotecnologia, 35(6):1039-1042, 2011.
- FIGUEIREDO, V. R.; DIAS, E. S. Cultivo do champignon em função da temperatura. Ciência Rural, 44(2):241-246, 2014.
- GARCÍA-DELGADO, C. et al. Purification of polluted water with spent mushroom (*Agaricus bisporus*) substrate: From agricultural waste to biosorbent of phenanthrene, Cd and Pb. **Environmental Technology**, 38(13-14):1792-1799, 2016.
- GOMES, R. B. et al. Production of "Roquesso" tomato seedlings with organic substrates. **Cadernos de Agroecologia**, 13(1):1-6, 2017.
- HANAFI, F. H. M. et al. Environmentally sustainable applications of agro-based spent mushroom substrate (SMS): An overview. Journal of Material Cycles and Waste Management, 20(3):1383-1396, 2018.
- LOPES, R. X. et al. Application of spent *Agaricus subrufescens* compost in integrated production of seedlings and plants of tomato. **International Journal of Recycling of Organic Waste in Agriculture**, 4(3):211-218, 2015.
- KIM, J. S. et al. Effect of microbial inoculant or molasses on fermentative quality and aerobic stability of sawdust-based spent mushroom substrate. **Bioresource Technology**, 216:188-195, 2016.
- KIM, Y. I.; KWAK, W. S. Isolation and identification of lactic acid bacteria from spent mushroom substrate for silage making and determination of optimal medium conditions for growth. Journal of Animal Science and Technology, 54(6):435-442, 2012.
- PAULA, F. S. et al. Stabilisation of spent mushroom substrate for application as a plant growth-promoting organic amendment. Journal of Environment Management, 196(1):476-486, 2017.
- RINKER, D. L. Spent Mushroom Substrate Uses: Technology and Applications. Edible and medicinal mushrooms: Technology and applications (First edit), John Wiley & Sons Ltda. p.427-454, 2017.

- ROYSE, D. J.; BAARS, J.; TAN, Q. Current overview of mushroom production in the world. Edible and medicinal mushrooms: Technology and applications (First edit), John Wiley & Sons Ltda., 5-13, 2017.
- SÁNCHEZ, J. E.; ZIED, D. C.; ALBERTO, E. Edible Mushroom production in the Americas. In: SÁNCHEZ, J. E.; ZIED, D. C.; ALBERTO, E. 9th International Conference on Mushroom Biology and Mushroom Products, Shangai: WSMBMP, 2-11, 2018.
- SILVA, R. R. et al. Influência de casca de arroz carbonizada em diferentes substratos na qualidade de mudas de tomateiro. Revista Brasileira de Ciências Agrárias, 7:803-809, 2012.

- SIQUEIRA, F. G. et al. Biological efficiency of *Agaricus brasiliensis* cultivated in compost with nitrogen concentrations. **Horticultura Brasileira**, 29(2):157-161, 2011.
- TRANI, P. E. et al. **Calagem e adubação do tomate de mesa.** Série tecnologia APTA. Boletim técnico, IAC. 2015. 35p.
- WAMSER, A. F. et al. Produtividade de híbridos de tomate submetidos ao cultivo superadensado. Horticultura Brasileira, 30:168-174, 2012.
- ZISOPOULOS, F. K. et al. A resource efficiency assessment of the industrial mushroom production chain: The influence of data variability. **Journal of Cleaner Production**, 126:394-408, 2016.