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AGARICUS BLAZEI MURRILL MUSHROOM COMPOST STUDY ANAEROBIC AND AEROBIC PHASES

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Abstract
Compost for the production of Agaricus blazei Murrill mushrooms, is produced from wheat straw, straw-bedded horse manure, chicken manure and gypsum. The substrate is made in two processes called Phase I (anaerobic) and Phase II (aerobic). Phase I includes mixing and moistening of the ingredients and a period of uncontrolled self-heating where temperatures will rise to 80ºC. Phase II starts with a pasteurization period of 8h at 56-60ºC and continues with a conditioning period at 45ºC for up to 7 days until volatile NH_3 has been cleared from the process by air.
Quality parameters for compost cannot be established directly. Moisture and nitrogen contents and pH can be adjusted at the start of Phase I, but the values will be affected during processing.
In this paperwork, we studied the physical properties (water content, electrical conductivity) and chemical composition (pH, organic matter, nitrogen, calcium, magnesium, ammonia) of four recipes of compost: classical, synthetic, mixt and original. During the experience, we recorded every hour the compost and the air temperature and the air relative humidity. The highest yield was obtained on synthetic compost with 42 kg mushrooms on 100 kg of compost.

Keywords: Agaricus blazei Murrill, compost, Phase I, Phase II.

1. INTRODUCTION

Because cultivated mushrooms are having a saprophyte diet, they are forced to feed with decomposing organic substances, decomposed with the enzymes they possess. Cultivated mushrooms are distinguished as important sources of nutrition, the carbon, the prime representative of organic substances, and mineral nutrition (Colak, 2004). An enrichment of the compost in carbohydrates is done by adding wheat or barley straw, corn cobs, corn cobs, which are materials with a high percentage of carbohydrates in their composition (Chakravarty, 2011). Due to the hydrolytic enzymes emitted by mushroom mycelium, these carbohydrates are converted into directly assimilable sugars (Vedder, 2012). During the composting and pasteurization of the substrate, the protein nitrogen is transformed into peptides and amino acids that are absorbed by mycelium hippocampal cells. As a result of researches, it was found that between the total nitrogen content of the nutrient substrate (up to 2.7%), the production of mushrooms and the protein content of the mushrooms has a direct correlation (Zicari et al., 2012). Research has shown that the osmotic pressure, the degree of diffusion through the cell membrane, depends on the concentration of the mineral salts in the nutrient substrate or the coating mixture. In case of higher concentration of

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mineral salts, fewer nourishing substances will penetrate with the osmotic phenomenon inside the mycelium (Chakravarty, 2011). Materials used to cultivate mushrooms of the genus Agaricus, must be subjected to composting prior to use, under the action of several microorganisms (Chang and Miles, 2004; Sánchez, 2004). During the composting, the temperature increases, the sugars are degraded and thermophilic microorganisms are formed, which transforms the cellulosic materials into substrate (Ross and Harris, 1983; Stölzer and Grabble, 1991).

2. MATERIALS AND METHODS

To study the influence of compost recipes on substrate quality and mushroom production, a bifactorial experience has been organized.

Factor A was the culture substrate with 4 graduations:

a1-classical
a2-synthetic
a3-mixed
a4-reed + horse manure

Factor B was the protein addition with 3 graduations:

b1 - without addition
b2 - wheat bran 3%
b3 – corn flour 3%

The combination of experimental factors resulted in 12 variants presented in Table 1.

Table 1. Experimental variants

Variant	Culture substrate	Protein addition
V1 (a_1b_1)	Classical	without addition
V2 (a_1b_2)	Classical	wheat bran 3%
V3 (a_1b_3)	Classical	corn flour 3%
V4 (a_2b_1)	Synthetic	without addition
V5 (a_2b_2)	Synthetic	wheat bran 3%
V6 (a_2b_3)	Synthetic	corn flour 3%
V7 (a_3b_1)	Mixed	without addition
V8 (a_3b_2)	Mixed	wheat bran 3%
V9 (a_3b_3)	Mixed	corn flour 3%
V10 (a_4b_1)	reed + horse manure	without addition
V11 (a_4b_2)	reed + horse manure	wheat bran 3%
V12 (a_4b_3)	reed + horse manure	corn flour 3%

In order to record the temperatures in the experiments carried out, the composting tanks and the humidifying / purine water tanks, they were equipped with LOGGER S0141 temperature readers, which are designed to record the readings read at different time intervals, set by user. Values are stored in non-volatile electronic memory. In the past, the S0141 has been configured to read the hourly temperature readings.

The compost recipes used in the experiment are presented in Table 2.

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Table 2. Compost recipes used in experience

Type of compost	Compost components	Quantity for 1 tone of compost
Classical	Horse manure (with bedding straw) Gypsum (calcium sulphate) Superphosphate Ammonium sulphate	500 kg 25 kg 7 kg 7 kg
Synthetic	Wheat straw Poultry manure Gypsum (calcium sulphate) Urea	350 kg 150 kg 20 kg 7 kg
Mixed	Horse manure (with bedding straw) Poultry manure Wheat straw Gypsum (calcium sulphate) Urea	250 kg 100 kg 150 kg 24 kg 2 kg
Reed	Crumbled reed Horse manure (with bedding straw) Poultry manure Gypsum (calcium sulphate) Urea	100 kg 200 kg 150 kg 24 kg 2 kg

Phase I - Soaking (anaerobic phase) was carried out for 5-6 days. (figure 1). Considering that the experimental composting plant has a capacity of 1 m^3 for an experimental variant, on day 6 the compost temperature rose to 60 °C to trigger the anaerobic fermentation process of the compost.
At the start of aerobic composting, the aeration system was started, which introduced air into each composting trough, thus on day 7 it began with 25m^3 of air per tonne per hour, then on day 11 it was reduced to 10m^3 air per tonne per hour, and from day 15 only 5m^3 of air per tonne per hour were administered.
The air was introduced into the composting tanks under the grate on which the compost is placed. (figure 2). The introduced air is filtered through a HEPA 13 filter.

Figure 1. Phase I - compost components presoaking

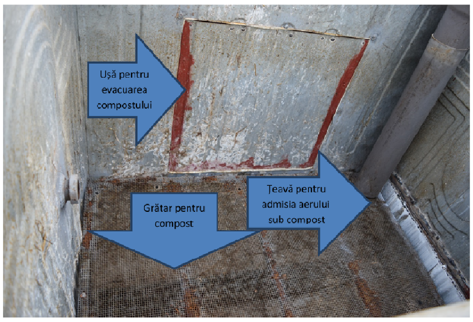

Figure 2. Composting tank, internal view

Source : Foto original – SC CIUPERCĂRIA SRL, Aghireșu-Fabrici, Cluj County

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Phase II of compost preparation, heat treatment, pasteurization, was achieved by raising the compost temperature to 58-60 °C for a period of about 8 hours, then the compost temperature was lowered to 50 °C with fresh air mixing and cooling continued to 45 °C. The temperature of 45 °C was maintained until the ammonia content of the compost fell below 0.05% and the pH stabilized in the range 7.3-7.5.

3. RESULTS AND DISCUSSIONS

The physicochemical characteristics of the mixtures that were introduced into the compost at the beginning of the anaerobic composting phase are presented in the table 3.

Table 3. Chemical characteristics of the mixture at the beginning of phase I

Type of compost	Determined element							
	Water content **%**	Organic matter / d.m. **%**	Nitrogen / d.m. **%**	Calcium **mg/l**	Magnesium **mg/l**	Ammonia (NH_3) **%**	pH	Electrical conductivity (EC) **dS/cm**
Classical	75	65	1.8	95	4	0.5	8.95	3.45
Synthetic	78	63	1.9	85	5	0.6	8.98	3.65
Mixed	74	68	1.7	87	4	0.55	8.87	3.78
Reed	75	68	1.8	82	4.6	0.58	8.92	3.67

At the beginning of the anaerobic composting phase, the water content of the mixtures for each experimental variation ranged from 74-78%, organic matter determined at 63-68%, nitrogen 1.7-1.9%, calcium 82-95 mg/l, magnesia 4-5 mg/l, ammonia (NH_3) 0.5-0.6%, determined pH 8.87-8.98, and electrical conductivity (EC) 3.45-3.78 dS/cm. The data obtained are also found in the scientific literature (Andrade et al., 2007, Chatterjee et al., 2013, Xiang et al., 2014).
The temperature values recorded in the aerobic composting tanks, days 7-22, are shown in figure 3.

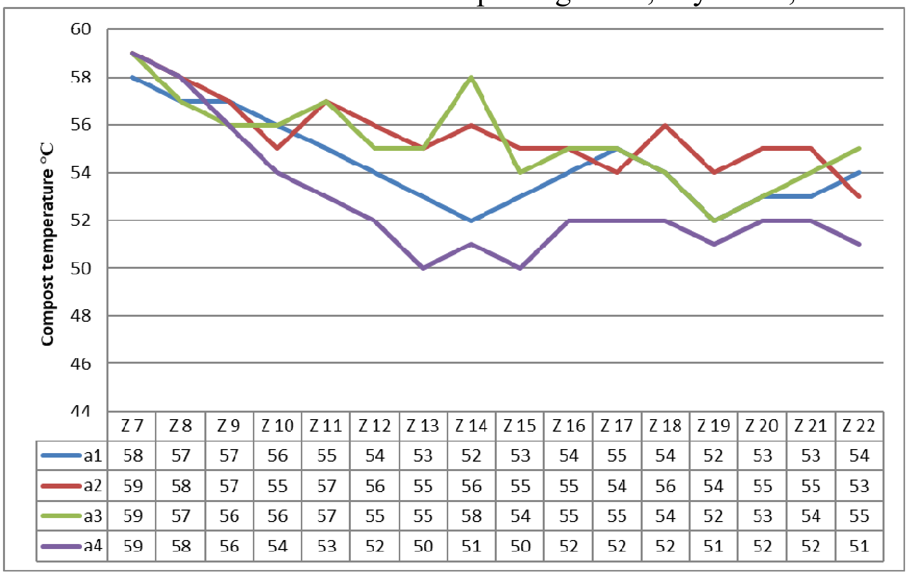

Figure 3. Temperature variation at aerobic composting of mixture, days (Z) 7-22

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The physicochemical characteristics of the mixture at the end of the aerobic composting, ie the beginning of the pasteurization for each experimental variant, are presented in the table 4.

Table 4. Chemical characteristics of the mixture at the end of aerobic composting

Type of compost	Determined element							
	Water content **%**	Organic matter / d.m. **%**	Nitrogen /d.m. **%**	Calcium **mg/l**	Magnesium **mg/l**	Ammonia (NH_3) **%**	pH	Electrical conductivity (EC) **dS/cm**
Classical	69	55	1.7	90	4	0.20	8.11	2.92
Synthetic	71	53	1.8	80	4.85	0.25	8.24	2.98
Mixed	68	56	1.7	82	3.7	0.21	7.98	2.83
Reed	67	56	1.8	79	4.1	0.29	8.12	2.93

By comparing the data in tables 3 and 4, it can be said that during aerobic composting, the water content in the compost decreases from 74-75% to 67-71%, the organic matter content drops from 63-68% to 53-56%, the content of nitrogen decreases by about 0.1%, calcium content decreases by 5mg/l, magnesium decreases by 0.1-0.3mg/l, ammonia (NH_3) decreases by 0.3% and pH decreases from 8.95 to 8.24, the best values being the mixed compost variant.
The physicochemical characteristics of compost after pasteurization are shown in the table 5.

Table 5. Chemical characteristics of mixture after pasteurization

Type of compost	Determined element							
	Water content **%**	Organic matter / d.m. **%**	Nitrogen /d.m. **%**	Calcium **mg/l**	Magnesium **mg/l**	Ammonia (NH_3) **%**	pH	Electrical conductivity (EC) **dS/cm**
Classical	65	51	2.4	87	3.8	0.03	7.21	2.32
Synthetic	63	49	2.6	78	4.7	0.032	7.32	2.48
Mixed	64	52	2.4	78	3.2	0.045	7.35	2.53
Reed	63	51	2.1	75	3.8	0.054	7.5	2.63

At the end of the pasteurization phase, the compost water content for each experimental variation ranged from 63-65%, Horm and Ohga (2008) mentioning 60-65%, Chatterjee et al., (2013) 60-70%.
Organic matter determined from d.m. at this stage, it was in the range of 49-52%, being included in the values studied by other authors Stanek (2010) 45-50% and Siqueira et al., (2011) 50%.
Nitrogen determined from d.m. at this phase, it was in the range of 2.1-2.6% below the value obtained by Gerben et al., (1998) 2.8%.
Calcium was in the range of 75-87 mg/l and magnesium in the range of 3.2-4.7 mg/l, Silva et al., (2009) reporting 80 mg/l for calcium and Colak (2004) reporting 5.3 mg/l for magnesium.
Ammonia (NH_3) determined in this phase was in the range of 0.03-0.054%, Xiang et al., (2014) reporting 0.049% (NH_3).
The pH was in the range of 7.21-7.5, the values being found in several authors. (Garcia et al., 1992, Gerben et al., 1998, Maynard 2000, Horm and Ohga 2008). The determined electrical conductivity (EC) was in the range of 2.32-2.63 dS/cm, Xiang et al., (2014) reporting 2.2-2.9 dS/cm.

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The averages of the quantities harvested for each experimental year in part on production flushes (1, 2 and 3) are presented in Table 6. The results are expressed in kg and refer to a cultivated area of 1m^2. Flush 1 represented 49.95% of total production in 2015, ie 52.59% in 2016, Flush 2 29.79% in 2015, 28.29% in 2016, and Flush 3 of 20.26 % in 2015 and 19.16% respectively in 2016.

Table 6. Harvest amounts at 1, 2 and 3 flushes, expressed in kg/m^2

Variant	Year **2015**				Year **2016**			
	Flush 1	Flush 2	Flush 3	**Total**	Flush 1	Flush 2	Flush 3	**Total**
V1	16,30	9,20	5,70	31,20	18,30	10,50	6,20	35,00
V2	17,10	10,30	6,77	34,17	19,20	9,80	7,93	36,93
V3	14,80	8,80	7,70	31,30	17,50	10,90	7,57	35,97
V4	16,20	10,50	8,10	34,80	21,30	11,20	7,60	40,10
V5	**19,20**	**13,20**	**8,87**	**41,27**	**22,30**	**12,80**	**6,73**	**41,83**
V6	18,50	12,70	7,37	38,57	20,90	11,70	8,40	41,00
V7	17,70	11,60	6,20	35,50	19,50	11,10	8,20	38,80
V8	19,50	11,90	8,67	40,07	21,40	12,40	6,77	40,57
V9	19,60	12,50	6,57	38,67	19,90	10,7	9,03	39,63
V10	15,30	7,20	5,07	27,57	16,50	7,10	5,20	28,80
V11	16,10	8,50	6,83	31,43	17,20	8,9	5,23	31,33
V12	14,80	7,50	5,73	28,03	16,70	7,80	5,67	30,17

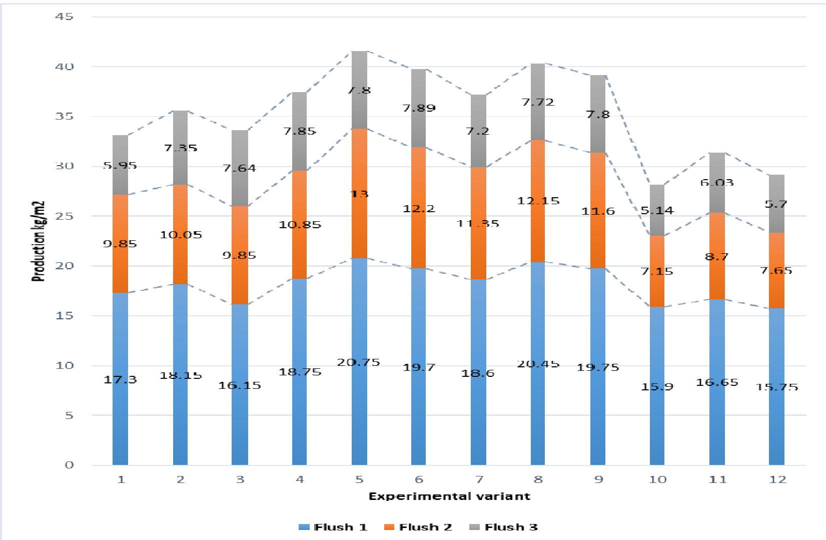

Figure 4. Dynamic of fructification flush in years 2015-2016

4. CONCLUSIONS

Researching the composition and methodology of composting for *Agaricus blazei* Murrill mushroom culture, by analogy with the specificity of the culture of the *Agaricus bisporus* mushroom, it is found that good results are obtained by using grain straws and horse mackerel or poultry manure.

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During aerobic composting, the water content in synthetic compost decreases from 78% to 71%, the organic matter content from 63% to 53%, the nitrogen content drops by about 0.1%, the calcium content by 5mg/l, magnesium with 0.15 mg/l, ammonia decreases by 0.35%, and the pH is reduced from 8.98 to 8.24.

During the pasteurization of the compost there are losses of up to 36.3% of the fresh material and up to 30.1% of the dry matter of the compost, the electrical conductivity of the synthetic compost has the higher values (2.98 dS/cm). General aspect, colour, smell, texture and presence of actinomycetes are more favorable to the culture of mushrooms than to the other compost variants.

In order to prepare limited quantities of compost, for experimental purposes, the complex, own conception plant, consisting of a composting bowl and grids, a purine recirculation and heating basin, a compressor for aeration, can be used.

The highest production of carpophores, determined by the composition and mode of composting, is obtained on the synthetic compost substrate with an average value of 39.5 kg/m^2, exceeding the average production of the other compost variants. Good production results can also be obtained on mixed compost. (38,3kg/m^2).

Compost quality and effects on production can be substantially improved by adding a protein supplement to the composition of the compost. Protein with 3% wheat bran provides a mean production increase of 6.9% for synthetic compost and 10.9% for mixed compost.

The addition of 3% honey flour has significant effects on the level of production compared to the compost variant without additional protein supplementation.

The production gained from the wheat bran supplement can be attributed to nutrient intake with compost protein.

The environmental conditions during the incubation period recommended for the *Agaricus bisposus* species are also suitable for the *Agaricus blazei* Murrill fungus.

5. REFERENCES

Andrad,e M.C.N., Kopytowski, Filho J., Minhoni, M.T.A., Coutinho, L.N., Figueiredo, M.B. (2007). Productivity, biological efficiency and number of Agaricus blazei mushrooms grown in compost in the presence of Trichoderma sp. and Chaetomium olivacearum contaminants. *Braz. J. Microbiol, 38*, 243-247.

Chakravarty, B. (2011). Trends in mushroom cultivation and breeding. *Australian Journal of Agricultural Engineering, 2(4)*, 102-109.

Chang, S.T., Miles, P.G. (2004). Mushrooms: cultivation, nutritional values, medicinal effect and environmental impact, 2nd ed. *Boca Raton: CRC*.

Chatterjee, N., Flury, M., Hinman, C., Cogger, C.G. (2013). Chemical and Physical Characteristics of Compost Leachates, *Washington State University Journal, 4*, 79-85.

Colak, M. (2004). Temperature profiles of Agaricus bisporus in composting stages and effects of different composts formulas and casing materials on yield. *African Journal of Biotechnology, 3 (9)*, 456-462.

Garcia, C., Hernandez, T., Costa, F., Ceccanti, B., Ciardi, C. (1992). Changes in ATP copntent, enzyme activity and inorganic nitrogen species during composting of organic wastes. *Can. J. Soil. Sci, 72*, 243-253. http://dx.doi.org/10.4141/cjss92-023.

Gerben, S., Jan, P.G., Jac, T.N.M., Jos, G.M. (1998). Adjustment of the composting process for mushroom cultivation based on initial substrate composition. *Bioresource Technology, 72,* 67-74.

Horm, V., Ohga, S. (2008). Potential of compost with some added supplementary materials on the development of Agaricus blazei Murill. *J. Fac. Agr. Kyushu Univ, 53(2),* 417-422.

Maynard, A. (2000). Compost: The process, *Connecticut agricultural experiment station*, *New Haven*, 128-145.

Ross, R.C., Harris, P.J. (1983). An investigation into the selective nature of mushroom compost. *Scientia Horticulturae*, 19, 55-64.

Rózsa, S. (2017). Study of some technological aspects of Agaricus blazei Murrill mushroom culture. PhD thesis. University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, Faculty of Horticulture Cluj-Napoca, Romania.

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ISSN-L: 2284-9521

Sánchez, C. (2004). Modern aspects of mushroom culture technology. *Applied Microbiology and Biotechnology 64*, 756-762.

Silva, C.F., Azevedo, R.S., Braga, C., Silva, R., Dias, E.S., Schwal, R.F. (2009). Microbial diversity in a bagasse-based compost prepared for the production of Agaricus brasiliensis. *Braz. J. Microbiol*, 40, 590-600.

Siqueira, F.G., Dias, E.S., Silva, R., Martos, E.T., Rinker, D.L. (2009). Cultivation of Agaricus blazei ss. Heinemann using different soils as source of casing materials, *Sci. Agric, 66(6),* 827-830.

Stanek, M. (2010). Microorganisms inhabiting mushroom compost during fermentation. *Mushroom Sci, 8,* 797-811.

Stölzer, S. Grabble, K. (1991). Mechanisms of substrate selectivity in the cultivation of edible fungi. In: MAHERMJ. (Ed.). *Science and cultivation of edible fungi. Rotterdam: Balkema 1*, 141-146.

Vedder, P.C. (2012). Modern mushroom growing. *Madisonville, Tex.: Pitman Press*.

Xiang, Z., Kenlin, C., Xingdong, W., Meng, L., Yin, W. (2014). Effects of Biochar from Spent Mushroom Substrate on Physic-chemical Properties during Pig Manure Composting. *Frontier of Environmental Science, 3(2)*, 67-74.

Zicari, G., Rivetti, D., Soardo, V., Cerrato, E. (2012). The cultivation of the mushroom Agaricus bisporus (Champignon) and some environmental and health aspects, *Brazilian Sanita Pubbl. May-Jun, 68(3),* 435-446.

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