THE INFLUENCE OF CULTURE TECHNOLOGY ON PRODUCTION AND CHEMICAL CONTENT IN *AGARICUS BLAZEI* MURRILL MUSHROOMS

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Abstract

Until 40-50 years ago, the medicinal properties of this fungus were known only to inhabitants of villages around the Amazonian forest in Brazil, but after scientific recognition of this mushroom, *Agaricus blazei* Murrill has become one of the most important immunomodulatory and mushrooms defense against tumors.

Attempts to cultivate *Agaricus blazei* Murrill, using biotechnologies have not been satisfactorily until around 2000s. The tropical native environment of *Agaricus blazei* Murrill mushrooms is very difficult to reproduce in the locations of culture. A few years ago, when demand for *Agaricus blazei* Murrill greatly increased, and the price has increased almost exponentially mushrooms in Piedade region in Brazil, mushrooms have disappeared almost completely.

All these considerations have led us to approach this work in biotech crop research on the species *Agaricus blazei* Murrill.

The study was conducted on four types of compost (classical, synthetic, mixed and original) with two different protein supplements, applying semi-intensive and semi-mechanized technology of culture. For each experimental variant were made the following chemical determinations: dry matter; total protein; total fat; ash; carbohydrates; energy value; β-glucan 1-3 D and lovastatin. The results were interpreted in terms of statistics.

Key words: *Agaricus blazei* Murrill, β 1-3 D-glucan, lovastatin

Human relationships with mushrooms are ancient and fascinating. The Egyptian believed that they were a gift of god Osiris, while the ancient Romans called them a ‘divine food’ because they thought that mushrooms resulted from the lightning thrown to early by Jupiter during storms. Mushrooms have been valued as delicious and nutritional foods in many countries. (Atkins F., 2006).

Worldwide specialists looking for new products from various plants and fungi containing vitamins, minerals, enzymes that improve human health. Viewed from this perspective, the mushrooms is a food with a high nutritional value, containing essential amino acids in the protein complex structure, and some species have real therapeutic and medicinal virtues. (Ellertsen L.K., 2005)

Traditionally, the fungi were granted with antitumor and immunological therapeutic virtues, which began to be tested scientifically, especially in the last three decades. The most important medical effects of mushrooms are: immunomodulatory genoprotector, antioxidant, anti-allergic, anti-tumor, liver, diabetes, anti-atherogenic, hipocolesterolic.

Currently, supplements and herbal medicines are provided and many products based on mushroom cultivation and medicinal basidiomycetes. (Halpern G.M., 2007)

Many researchers have scientifically proven that beta-glucan content of these fungi is more beneficial to glucans of other medicinal mushrooms. Beta-glucan of *Agaricus blazei* Murrill main form is 1-6, with a spiral which closely replicate the size and shape of normal DNA. (Kaul N.T., 2002)

Some recently isolated and identified compounds, originating from mushrooms, that block the formation of carcinogens, alter membrane structure suppress DNA synthesis, enhance cell differentiation and compete with estrogen receptors (Stamets P., 2010).

MATERIAL AND METHOD

Compost is the substrate on which mushrooms grows. The microbial degradation of organic wastes by several microorganism makes the substrate selective for the growth of *Agaricus blazei* Murrill. During composting, distinct changes occur in the physical, chemical and biological

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characteristics of the straw, all of which influence the productivity. The recipes of the compost for each experimental variant of the experience are shown in the *table 1*.

Table 1

Recipes of the compost used in experience

Type of compost	Components	Cantity for 1 tone of compost
Classical	Horse manure (composed by horse manure and wheat straw bedding 70-75%) Gypsum (calcium sulphate) Superphosphate Ammonium sulfate	500 kg 25 kg 7 kg 7 kg
Synthetic	Wheat straw Poultry litter Gypsum (calcium sulphate) Urea	350 kg 150 kg 20 kg 7 kg
Mixt	Horse manure (composed by horse manure and wheat straw bedding 70-75%) Gunoi de păsări Poultry litter Gypsum (calcium sulphate) Urea Wheat bran 3%	250 kg 100 kg 150 kg 24 kg 2 kg 30 kg
Original	Shredded cane Horse manure (composed by horse manure and wheat straw bedding 70-75%) Poultry litter Gypsum (calcium sulphate) Urea	100 kg 200 kg 150 kg 24 kg 2 kg

In order to achieve a directed composting, were made four identical tanks to control, perform and record optimal environmental conditions necessary for composting (*figure 1*)

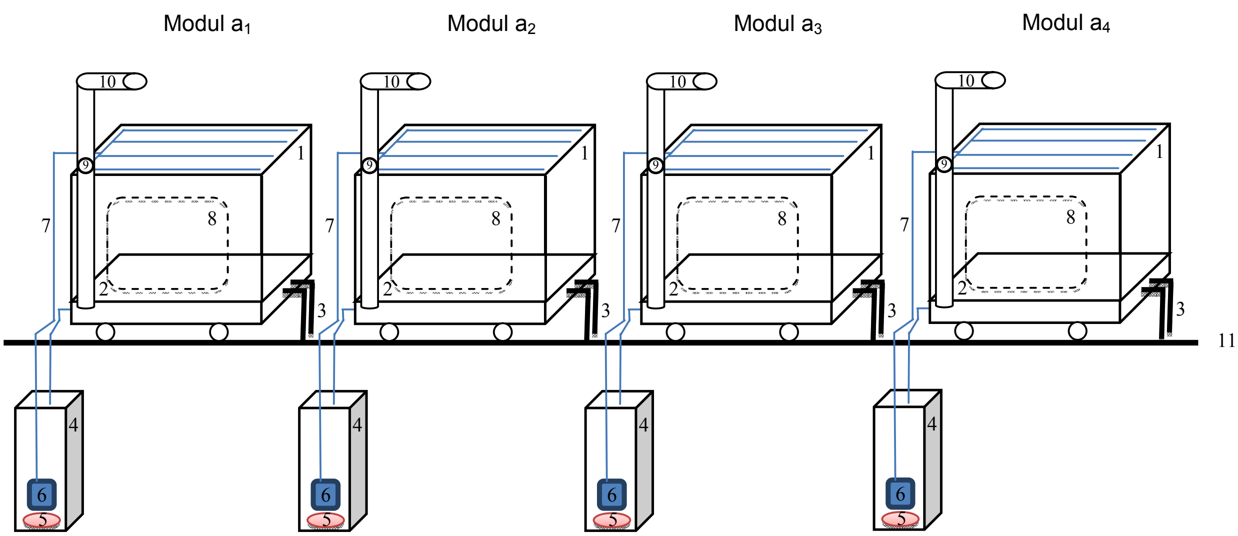

Figure 1 **Sketch for the composting facility - 4 modules**

1 - tank with capacity of 1 m^3 for compost components; 2 – rack for compost; 3 - tank heating system for composting; 4 - tank for the collection and recirculation the water excess; 5 - heating elements for wetting water (purine); 6 – water/purine recirculation pump; 7 - recirculation pipes for wetting water/ purine; 8 - compost discharge door; 9 - air flow control valve for aerobic composting; 10 - air inlet pipe to aerobic composting from the compressor; 11 - ground level.

RESULTS AND DISCUSSIONS

The amounts collected for each experimental variant in part, to the waves 1, 2 and 3 are shown in *table 2*. The results are expressed in kg and are for 1 square meter of cultivated area.

Table 2

Harvest amounts at 1, 2 and 3 flush

Experimental variant	Flush 1 kg/1m^2	Flush 2 kg/1m^2	Flush 3 kg/1m^2	Total kg/1m^2
V1(a_1b_1)*	15.98	12.43	7.09	35.50
V2 (a_1b_2)**	16.74	13.02	7.44	37.20
V3 (a_1b_3)***	16.43	12.78	7.29	36.50
V4(a_2b_1)*	18.23	14.18	8.09	40.50
V5 (a_2b_2)**	18.90	14.70	8.40	42.00
V6 (a_2b_3)***	18.63	14.49	8.28	41.40
V7(a_3b_1)*	17.55	13.65	7.80	39.00
V8 (a_3b_2)**	18.36	14.28	8.16	40.80
V9 (a_3b_3)***	17.96	13.97	7.97	39.90
V10 (a_4b_1)*	13.19	10.26	5.85	29.30
V11 (a_4b_2)**	14.09	10.96	6.25	31.30
V12 (a_4b_3)***	13.82	10.75	6.13	30.70

V1, 2, 3 – classical compost, V4, 5, 6 – synthetic compost, V7, 8, 9 – mixt compost, V10, 11, 12 – original compost
* without supplements, **with wheat bran 3%, *** with corn flour 3%

It was analysed the unilaterally influence of compost on *Agaricus blazei* Murrill mushroom production, table 3 show us the obtained values. The mixed and synthetic compost registered very significantly positive differences (4.35 and 3.04) against average and classic and original compost registered very significant negative differences from the average.

Table 3

Unilateral influence of compost over the production

Compost	Obtained harvest		Difference ±D	Signification of difference
	Obtained values	%		
	36.86	100.0	0.00	Mt.
Classical	36.22	98.3	-0.64	000
Synthetic	41.21	111.8	4.35	***
Mixt	39.90	108.2	3.04	***
Original	30.11	81.7	-6.75	000
DL (p 5%)			0.26	
DL (p 1%)			0.40	
DL (p 0.1%)			0.63	

It was analysed the unilateral influence of protein addition in compost on *Agaricus blazei* Murrill mushroom production, *Table 4* shows us the obtained values. The addition of wheat bran 3% registered very significant positive differences (0.87) from the mean, and the addition of corn flour 3% have not experienced a statistically difference from the mean.

Table 4

Unilateral influence of protein addition over the production

Protein addition	Obtained harvest		Difference ±D	Signification of difference
	Obtained values	%		
	36.86	100.0	0.00	Mt.
Without added protein	35.91	97.4	-0.95	000
Wheat bran 3%	37.73	102.4	0.87	***
Corn flour 3%	36.94	100.2	0.08	-
DL (p 5%)			0.23	
DL (p 1%)			0.32	
DL (p 0.1%)			0.44	

It was analysed the unilateral influence of compost on the dry matter content, expressed in grams per 100g fresh substance, at the *Agaricus blazei* Murrill mushroom. *Table 5* presents the values obtained. The mixed and synthetic compost registered very significantly positive differences (0.33 and 0.14) against average and the original compost recorded very significant negative differences from the average.

It was analysed the unilateral influence of compost on total protein content, expressed in grams per 100g dry weight, at the *Agaricus blazei* Murrill mushroom. *Table 6* presents the values obtained. The classic, synthetic and mixed composts registered very significant positive differences from the average and the original compost recorded very significant negative differences from the average.

Table 5

Unilateral influence of compost over the dry matter of *Agaricus blazei* Murrill mushrooms

Compost	Dry matter g/100g sp (fm)		Difference ±D	Signification of difference
	Obtained values	%		
	8.44	100.0	0.00	Mt.
Classical	8.41	99.7	-0.03	-
Synthetic	8.77	103.9	0.33	***
Mixt	8.58	101.6	0.14	***
Original	8.00	94.8	-0.44	000
DL (p 5%)			0.06	
DL (p 1%)			0.08	
DL (p 0.1%)			0.14	

Table 6

Unilateral influence of compost over the total protein of *Agaricus blazei* Murrill mushrooms

Compost	Total protein g/100g su (dm)		Difference ±D	Signification of difference
	Obtained values	%		
	25.16	100.0	0.00	Mt.
Classical	26.77	106.4	1.60	***
Synthetic	31.61	125.6	6.45	***
Mixt	28.11	111.7	2.95	***
Original	14.17	56.3	-11.00	000
DL (p 5%)			0.34	
DL (p 1%)			0.52	
DL (p 0.1%)			0.83	

It was analysed the unilateral influence of compost on total fat content in grams per 100g dry weight, at the *Agaricus blazei* Murrill mushrooms. *Table 7* presents the values obtained. The mixed and classic composts registered very significant positive differences against average and original and synthetic composts registered significant negative differences from the average.

Table 7

Unilateral influence of compost over the total fat of *Agaricus blazei* Murrill mushrooms

Compost	Total fat g/100g dm		Difference ±D	Signification of difference
	Obtained values	%		
	2.20	100.0	0.00	Mt.
Classical	2.69	122.2	0.49	***
Synthetic	1.80	81.8	-0.40	000
Mixt	2.76	125.3	0.56	***
Original	1.56	70.7	-0.64	000
DL (p 5%)			0.11	
DL (p 1%)			0.17	
DL (p 0.1%)			0.27	

It was analysed the unilateral influence of compost on ash content in grams per 100g dry weight, at the *Agaricus blazei* Murrill mushroom. *Table 8* presents the obtained values. The classical and mixt composts registered very significant positive differences against average and the original and synthetic compost registered very significant negative differences from the average.

Table 8

Unilateral influence of compost over the ash of *Agaricus blazei* Murrill mushrooms

Compost	Ash g/100g dm		Difference ±D	Signification of difference
	Obtained values	%		
	8.78	100.0	0.00	Mt.
Classical	9.60	109.3	0.82	***
Synthetic	8.44	96.2	-0.34	000
Mixt	9.50	108.2	0.72	***
Original	7.58	86.3	-1.20	000
DL (p 5%)			0.11	
DL (p 1%)			0.16	
DL (p 0.1%)			0.26	

It was analysed the unilateral influence of compost on carbohydrate content, in grams per 100g dry weight, at the *Agaricus blazei* Murrill mushroom. *Table 9* presents us the values obtained. The original compost registered very significant positive differences from the average and the classic, mixed and synthetic composts registered significant negative differences from the average.

Table 9

Unilateral influence of compost over the carbohydrates of *Agaricus blazei* Murrill mushrooms

Compost	Carbohydrates g/100g dm		Difference ±D	Signification of difference
	Obtained values	%		
	65.49	100.0	0.00	Mt.
Classical	64.72	98.8	-0.77	000
Synthetic	60.39	92.2	-5.11	000
Mixt	60.53	92.4	-4.96	000
Original	76.33	116.5	10.84	***
DL (p 5%)			0.24	
DL (p 1%)			0.37	
DL (p 0.1%)			0.59	

It was analysed the unilateral influence of compost on energy value, expressed in kcal/kg fresh matter, at the *Agaricus blazei* Murrill mushroom. *Table 10* presents the values obtained. Synthetic compost registered very significant positive differences from the average and the classic, original and mixed composts registered very significant negative differences from the average.

Table 10

Unilateral influence of compost over the energetic value of *Agaricus blazei* Murrill mushrooms

Compost	Energetic value Kcal/kg fm		Difference ±D	Signification of difference
	Obtained values	%		
	336.11	100.0	0.00	Mt.
Classical	330.56	98.3	-5.56	000
Synthetic	390.11	116.1	54.00	***
Mixt	298.89	88.9	-37.22	000
Original	324.89	96.7	-11.22	000
DL (p 5%)			2.22	
DL (p 1%)			3.36	
DL (p 0.1%)			5.40	

It was analysed the unilateral influence of compost on the content of β-glucan 1-3 D, expressed in mg per 100 g dry matter, at the *Agaricus blazei* Murrill mushroom. *Table 11* presents the obtained values. The classical and mixed compost registered very significant positive differences against average and the original compost registered very significant negative differences from the average.

Table 11

Unilateral influence of compost over the β 1-3 D-glucan of *Agaricus blazei* Murrill mushrooms

Compost	β 1-3 D-glucan mg/100g dm		Difference ±D	Signification of difference
	Obtained values	%		
	3292.58	100.0	0.00	Mt.
Classical	3559.22	108.1	266.64	***
Synthetic	3279.44	99.6	-13.14	-
Mixt	3360.00	102.0	67.42	***
Original	2971.67	90.3	-320.92	000
DL (p 5%)			24.76	
DL (p 1%)			37.50	
DL (p 0.1%)			60.24	

It was analysed the influence of compost on unilateral lovastatin content expressed in mg per 100 g dry matter, the *Agaricus blazei* Murrill. *Table 12* presents the obtained values. The classical and mixed composts registered very significant positive differences against average and the synthetic and original compost registered very significant negative differences from the average.

Table 12

Unilateral influence of compost over the lovastatin of *Agaricus blazei* Murrill mushrooms

Compost	Lovastatin mg/100g dm		Difference ±D	Signification of difference
	Obtained values	%		
	15.18	100.0	0.00	Mt.
Classical	18.17	119.7	2.99	***
Synthetic	14.28	94.1	-0.90	000
Mixt	16.14	106.3	0.96	***
Original	12.13	79.9	-3.05	000
DL (p 5%)			0.11	
DL (p 1%)			0.16	
DL (p 0,1%)			0.26	

CONCLUSIONS

Synthetic compost had the highest yield (41.21 kg/m^2), followed by mixed compost (39.90 kg/m^2). The lowest production was carried out on the original compost (30.11 kg/m^2).

The addition of protein in compost, which ensures higher productivity are wheat bran 3% (37.73 kg/ m^2), followed by corn flour 3% (36.94 kg/m^2).

Analysing the dry matter content of the mushrooms, it can be concluded that mushrooms harvested from synthetic compost presents the greatest amount of dry matter (8.77g/100g fresh matter).

Analysing the total protein content of mushrooms, it can be concluded that mushrooms harvested from synthetic compost presents the greatest amount of total protein (31.61g /100 g dry matter), followed by mushrooms harvested on mixed compost (28.11 g / dry matter 100g).

Analysing the compost influence on the content of β-glucan 1-3 D, it can be concluded that harvested mushrooms on classical compost presents the highest value of β-glucan 1-3 D (3559.22 mg / 100g dry matter) followed by mushrooms harvested on mixed compost (3360.00 mg / 100g dry matter).

Analysing the compost influence on the content of lovastatin, it can be concluded that harvested mushrooms on classical compost presents the highest value lovastatin (18.17 mg / 100g dry matter), followed by mushrooms harvested on mixed compost (16.14 mg / 100g dry matter).

REFERENCES

Atkins, F., 2006 - *Mushroom growing today*, Macmillan Publishing Co., New York

Ellertsen, L.K., 2005 - *Effect of a medicinal extract from Agaricus blazei Murrill on gene expression in human monocytes*, Poster 1454, World Allergy Congress, Munich, Germany, June 26-30

Halpern, G.M., 2007 - *Healing mushrooms efective treatments for todays illnesses*, Square One Publishers, New York

Kaul, N.T., 2002 - *Biology and conservation of mushrooms*, Basic Health, Canada

Stamets, P., 2010 - *Growing gourmet and medicinal mushrooms*, Ten Speed Press, Berkele.