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Effect of Different Mixtures with Waste Paper as Casing Soil on the Growth and Production of Mushroom (*Agaricus Bisporus*)

[1]Y.N. Sassine, [2]A.M.R. Abdel-Mawgoud, [1]Y. Ghora and [3]M. Bohme

[1]Department of Horticulture, Faculty of Agriculture, Saint Joseph University, Beirut, Lebanon.
[2]Department of Vegetable Research, National Research Center, Dokki, Cairo, Egypt.
[3]Department of Horticulture, Faculty of Agriculture, Humboldt University in Berlin, Germany.

Abstract: Four experiments were conducted in order to investigate the possibility of replacing the standard peat casing in mushroom production by other alternatives such as waste paper. Waste paper was composted with or without nitrogen addition. After composting, paper was used either alone or mixed with sand and/or peat in different ratios. Mixing ratios ranged from 0 to 100% of both materials. The three materials were also tested in different mixing ratios. In one experiment, the physical shape of composted paper were changed in the shape of balls and used either alone or mixed with sand. The results showed that adding nitrogen to paper during composting not only accelerated the composting process but also improved somehow the properties of paper and enhanced mushroom production but not to the standard level. Mixing composted paper with sand had non consistent effect. Changing the physical shape of composted paper resulted in many dead primordial. The addition of peat to composted paper had a positive effect on mushroom production and the effect can be described as linear. The best mixing ratio of the three materials was 33% paper+33%sand+33%peat which gave a comparable result to the standard peat casing. It could be concluded that waste paper mixed with other materials such as sand and peat can partially replace the standard peat casing soil.

Key words: mushroom, *Agaricus bisborus*, peat casing, waste paper, mixing casing.

INTRODUCTION

Mushrooms are named as the meat of the forest because the high content of proteins. Therefore the consumption of mushrooms was more especially in times of a lack of meat, e.g. during the wars. *Agaricus* mushrooms contain many essential amino acids and can be a valuable food source. Button *Agaricus* mushrooms contain more protein than kidney beans even the same amount as milk. *Agaricus* mushrooms have a high content of proteins such as Tryptophan, Hystin, Histidin and Arginin (MORGREIDE, 1968; SEELKOPF and SCHUSTER, 1957). In the Asian traditions it is expressed that some of the important effects of *Agaricus* mushrooms are the health benefits.

Despite this increasing importance, mushroom cultivation is not introduced yet in Lebanon although the local demands of mushroom are very high and keep claiming. The reason for that is the lack of the necessary materials for cultivation such as peat. Not only the import cost of peat but also the depletion of its available resources worldwide dis-encouraged the investors to start such projects. Therefore other 'local' alternatives have to be considered in order to encourage mushroom production locally. Lebanese paper industry, as a corner stone in the economy, leaves behind a huge amount of side products and residuals which is considered to be a big environmental problem. Therefore, it was suggested to test the wastes of paper as an alternative for peat casing in mushroom production. The possibility of using composted or uncompsted waste paper as casing soil was investigated by Dergham (1994, 1996) and in different thickness by Sassine *et.al.,* (2005). With the latter, however, the results of mushroom production were not up to the commercial level. In this study, other suggestions for using waste paper as casing soil are being introduced.

It is the aim of this study to investigate the possibility of using different mixtures of waste paper with other materials as a casing soil in mushroom production.

Corresponding Author: Y.N. Sassine, Department of Horticulture, Faculty of Agriculture, Saint Joseph University, Beirut, Lebanon.

MATERIALS AND METHODS

Four experiments were carried out to test different mixtures of composted or un-composted paper mixed with different materials in different ratios as casing soil for mushroom production. The materials used for mixing with waste paper were either loamy sand or peat. The ratio of mixing of these materials ranged from 0.0 to 100 percent as shown in Table 1.

Table 1: Some basic information about experiment number, time of composting, rate of nitrogen addition and percentage of components in the casing soil.

Exp. No.	Composting time (days)	Nitrogen addition during composting (g/100L)	Mixture with other materials (in percentage)		
			Paper	Peat	Sand
I	60	0.0, 0.7	100	--	0.0
			90		10
			70		30
			50		50
			30		70
			100	0.0	--
			90	10	
			70	30	
			50	50	
			30	70	
II	60	0.0, 0.7	30	70	--
			100	0.0	
			30	--	70
			0.0		100
III	60	0.0, 0.7	0.0	0.0	100
			33	33	33
			50	25	25
			70	15	15
IV	60	0.0, 0.7	50	50	--
			100	0.0	
			50	--	50
			100		0.0

Paper composting:

Waste paper was collected and shredded in 0.5 cm width with different lengths. Eight plastic pots were filled each with five kilograms of shredded paper for composting. Each pot had five side-holes in order to keep enough aeration and prevent temperature from rising to critical levels. The electrical heater was used to regulate room temperature in a range of 18°C. Temperature of the composted paper was monitored by 50 cm-long mercury thermometer, one in each pot. Temperature of the compost did not fall below 18°C and not higher than 38°C.

In the beginning of composting, water was added to each pot abundantly until drain was enough and paper was completely wet. Water then was added when necessary to keep the substrate moisture. Water was added equally to each pot in every watering-time. The moisture content in this compost was around 60%. Pots were covered with plastic sheets in order to reduce surface evaporation. Compost was mixed thoroughly every while to have homogenous temperature and composting process.

Some experiments were conducted in the Research Centre of Rotterdam, The Netherlands. The composting process took place by digging a big whole in the ground large enough to take up about five tones of paper wastes. Before putting the paper wastes in, the whole was lined with a 15 cm of cement and drainage system was installed. Drained water was collected and passed through a small laboratory for analysis and purification, in particularly, from the large amount of heavy metals in the printing ink usually used in newspaper. For three years, the whole was filled with enough amount of water every six months in order to leach the printing ink with its heavy metals.

Nitrogen Addition During Composting:

In order to accelerate the composting process, C/N ratio had to be narrowed by adding nitrogen. Nitrogen was added in the form of potassium nitrate with 98% purity and in a rate of 0.7% as a result from previous

experience (Sassine *et. al.*, 2005).

Mixtures with Other Materials:

Following the information's from the literature in some experiments, waste paper was mixed as well with sand as with standard casing soil (peat).

Fine clean loamy sand, in average 0.2 mm in size, was used as a casing soil or as supplement to waste paper or standard casing. The aim was to compare the different mixtures. Sand was mixed in different quantities with these materials (Table 1). This loamy sand had a pH of 7. In some cases it was difficult to get a homogenous mix especially with sand and composted paper. Casing with loamy sand was applied in the standard thickness of 5.0 cm.

Physical Analysis:

The physical properties were analysed of paper and peat. In the experimental station of the Institute for horticultural sciences the pore volume, water capacity (WC) and air capacity (AC) using the Air-pycnometer (BÖHME and VORWERK, 1991) was measured. Also was measured the water holding capacity. Water holding capacity is the ability of a material to keep water against gravity. It depends on the physical structure and characteristic of the material.

Water holding capacity was calculated by adding plenty of water to a known weight of shredded paper until drainage was obvious. After drain was stopped, the amount of wet paper was weighted again and difference between dry and wet paper was calculated.

The experiments were conducted in an underground room. Air temperature of the room as well as relative humidity were monitored and recorded by a thermo hygrograph. Air temperature of the room did not fall below 18°C and relative humidity was around 80%. An electrical heater was used when necessary in order to maintain air temperature 18°C.

Temperatures of both air and compost were monitored so that they would not limit the process of composting or the growth of mushroom. The electrical heater was used for that purpose. Minimum temperature was recorded to be 18°C and the maximum was 37° during composting process. The concentration of carbon dioxide was measured till 2300 ppm. During the growth of the mycelium the CO_2 content was often above the scale of the measurement. Normally the CO_2 content decreased to 1600 ppm, but sometimes the level was all the time about 2000 ppm. Relative humidity in the air was above 90% most of the time of the experiments.

Harvesting and Mushroom Head Quality:

Harvest was carried out once a week and a total of six harvests were collected. Total weight and number of heads were recorded in each harvest. Average weight of individual heads was calculated.

Dry matter content in some harvests were measured by measuring the fresh weight of some *Agaricus* mushroom heads and their dry weight after drying in the oven for three days on 108°C.

Statistical analysis:

The statistical analysis of the experimental data was carried out using ANOVA one way analysis. Significant differences were calculated at $P < 0.05$. The LSD method was used in all statistical analyses. Also regression analyses were calculated for different parameters. SPSS package version 11.0 was used for these processes.

RESULTS AND DISCUSSIONS

Results:

The compost ready for use was mixed with loamy sand and/or peat. The pore-volume of the mixture paper/loamy sand was 72.8 %, the WC= 46.6 %, AC= 25.2 % and density= 828 kg m^{-3}

In one experiment in this study where different quantities of paper were mixed with loamy sand, the differences between the variants were not very high (Figure 1). The highest number of mushrooms was in the mix with 70 % with nitrogen composted paper in the casing layer. This difference was significant to all other mixtures. It is obvious that the effect of loamy sand without other components as composted paper is lower. In previous experiments (Sassine *et al.*, 2005), it was found a big difference between the number of mushroom heads and the fresh weight. In this experiment the trend was similar compares the number and the fresh weight (Figure 2). From the variants with the mixture of 70% composted paper to loamy sand were harvested the highest yield with significant difference to all other variants.

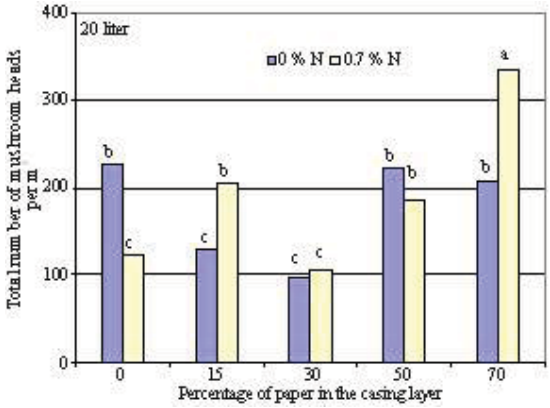

Fig. 1: Total number of *Agaricus* mushroom heads harvested from treatments with different mixtures of paper and sand as a casing soil. Different letters indicate significant differences at $p<0.05$.

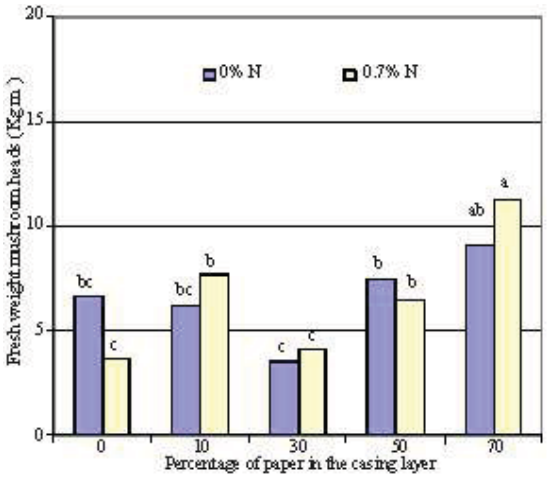

Fig. 2: Total weight of *Agaricus* mushroom heads harvested from different mixtures of paper and loamy sand as a casing soil. Different letters indicate significant differences at $p<0.05$.

Effect of Mixtures of Composted Paper with the Standard Peat:

The situation is clearer in the case of mixture of paper and peat. There was a tendency for lower number of *Agaricus* mushroom heads with increasing the percentage of paper in the mixture of the casing soil (Figure 3). The lowest number was recorded in the 70% paper treatment. The same trends were observed in the total weight. It can not be explained also why composted paper without nitrogen had a similar or even higher mushroom growth.

The parameter fresh weight of mushrooms was similar as with the parameter number of mushrooms (Figure 4).

The variants with adding of nitrogen showed in most of cases a higher fresh weight.

In some cases was the difference significant.

In both experiments, mixture of composted paper with sand and mixture with peat, the average weight of the individual *Agaricus* mushroom heads gave different results. The average weight in the different mixtures of paper and sand seemed to be very variable (Figure 5). The highest weight of individual mushroom head was found in average of all mixtures and substrate volumes by using of compost with 0.7 % nitrogen.

The weight of individual *Agaricus* mushroom heads seemed to be not affected in the experiments with mixture of composted paper and peat. A small but not significant difference in the average weights of single mushrooms is visible for the treatment of 0.7% nitrogen before composting of paper (Figure 6).

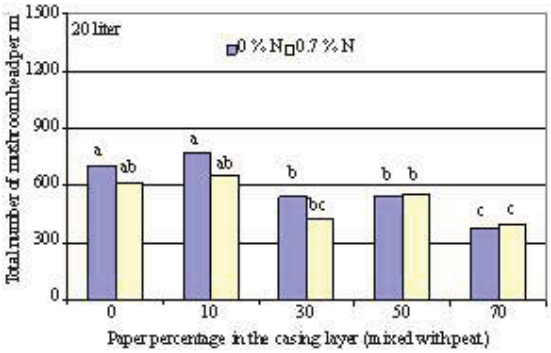

Fig. 3: Total number of *Agaricus* mushroom heads harvested from treatments with different mixtures of paper and peat as a casing soil. Different letters indicate significant differences at $p<0.05$.

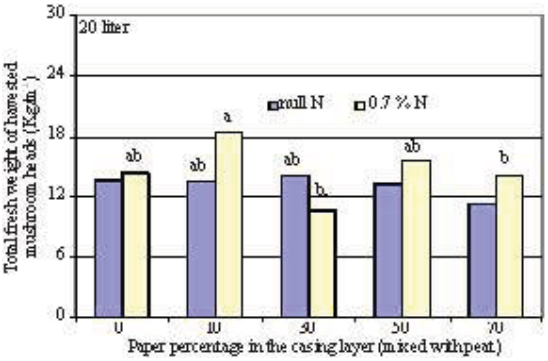

Fig. 4: Total weight of *Agaricus* mushroom heads harvested from treatments with different mixtures of paper and peat as a casing soil. Different letters indicate significant differences at $p<0.05$.

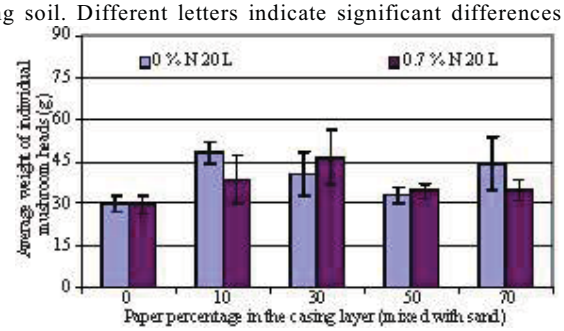

Fig. 5: Average weight of individual *Agaricus* mushroom heads harvested from treatments with different mixtures of paper and sand as a casing soil. Error bars are twice standard error.

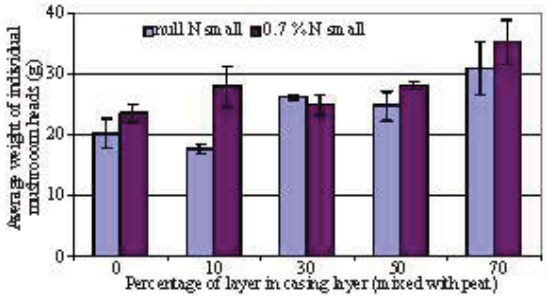

Fig. 6: Average weight of individual *Agaricus* mushroom heads grown in different mixtures of paper and peat as a casing soil. Error bars are twice standard error.

Effect of Mixtures of Three Components:

Because the results getting with different mixtures of loamy sand and paper as well as of peat and paper were not convincing, those treatments were repeated. In this experiment paper slides was formed in globules using a mixer. These globules were used as homogenous casing material and also mixed with sand or peat. In the beginning of this experiment it was observed changing of the size of these globules after wetting. The diameter raised from 10 to 25 mm. Additional were investigated also a mixture of all three components (paper, sand and peat) in different ratios.

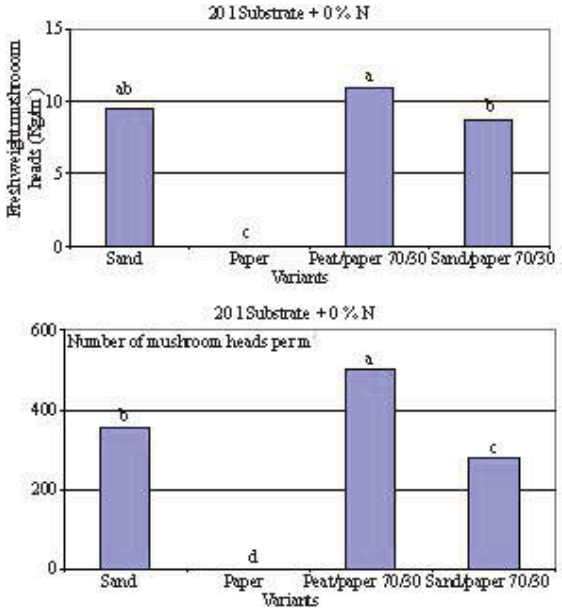

Fig. 7: Total weight and number of *Agaricus* mushroom heads harvested from 20 litres of substrate and treatments of different mixtures of paper, sand and peat as a casing soil. Different letters indicate significant differences at $p<0.05$.

From the variants with paper globules as casing soil no mushrooms were harvested. The mycelium started to grow in the globule, but no primordia were developed.

The total mushroom production in the variant with paper as casing material was zero, because the lack of nitrogen. The yield in the mixture of peat and paper was significantly higher than in all other variants. The same conclusion can be done looking the total number of mushrooms (Figure 7).

Adding nitrogen to the paper during composting had a noticeable positive effect on total yield (Figure 8). The total fresh weight and number were not significantly different between sand casing and casing with the mixture of loamy sand and paper. The number of harvested mushroom heads was significantly higher in the variant peat (70%) and paper (30%). Sand alone and sand (70%) with paper (30%) were not significantly different. As in the previous experiments in the casing soil with content of peat the number of mushrooms is higher.

Other ratios of the three materials were tested as follows:-

Paper 33%, peat 33%, sand 33%

Paper 50%, peat 25%, sand 25%

Paper 70%, peat 15%, sand 15%

The total fresh weight (Figure 9) showed no significant difference between peat casing and casing with the mixture of paper, peat and sand. Increasing the percentage of paper in the mixture to 50% or 70% significantly reduced the total fresh weight and number of mushroom heads (Figure 10). The number of

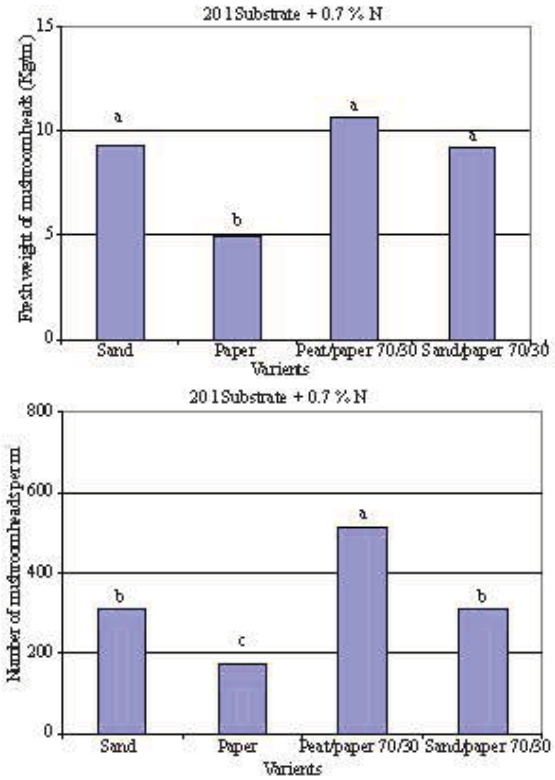

Fig. 8: Total weight and number of *Agaricus* mushroom heads harvested from 20 litres of substrate and treatments of different mixtures of paper, sand and peat as a casing soil. Different letters indicate significant differences at $p<0.05$.

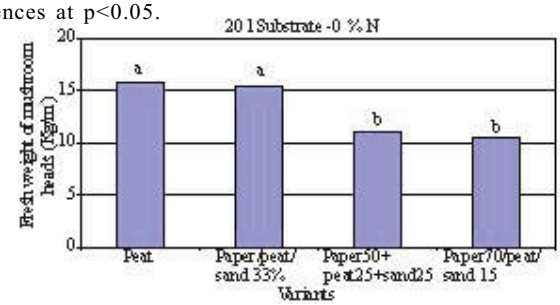

Fig. 9: Total weight of *Agaricus* mushroom heads harvested from 20 litres substrate and different mixtures of paper, sand and peat as a casing soil. Different letters indicate significant differences at $p<0.05$.

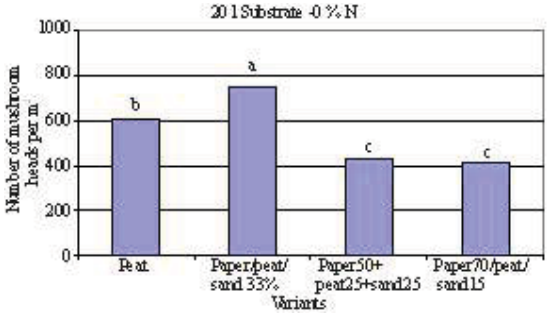

Fig. 10: Total number of *Agaricus* mushroom heads under treatments of different mixtures of paper, sand and peat as a casing soil. Different letters indicate significant differences at $p<0.05$.

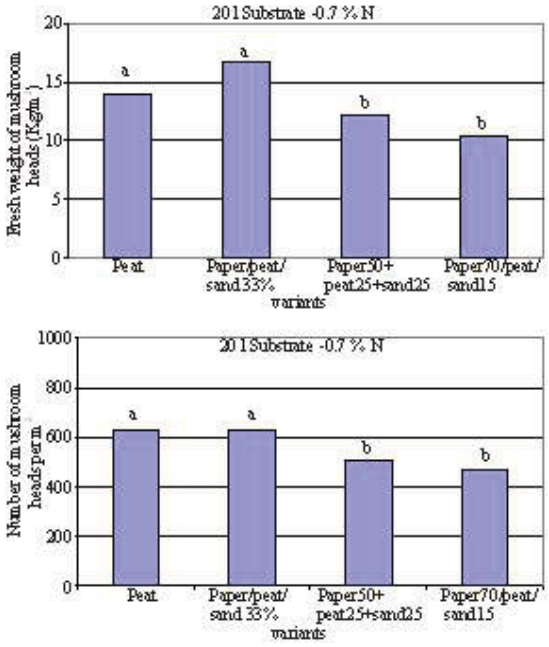

Fig. 11: Total weight and number of *Agaricus* mushroom heads harvested under treatments of different mixtures of paper, sand and peat as a casing soil. Different letters indicate significant differences at $p<0.05$.

harvested mushroom heads using a casing with mixture of paper, sand and peat (33%, 33%, 33%) was the highest compared to all other casing variants. The differences were significant.

The addition of nitrogen had a good effect on the quality of the composted paper.

Figure (11) shows the total yield (fresh weight) and number of mushroom heads under the different mixture of peat and/or sand with composted paper with the addition of nitrogen. Adding nitrogen to paper during composting can improve the fresh weight of mushroom heads. The mixture paper 33%, peat 33%, sand 33% had a significantly higher fresh weight compared to all other treatments including the control treatment. A significantly lower yield can observed of the variant with 70% paper in the mixture.

Discussion:

The results of our previous study (Sassine *et al.*, 2005) indicated to the necessity of the composting process for waste paper before using it as a casing soil. It also indicated that the quality of the composted paper was not in all cases satisfying. In comparison to the standard peat casing soil the mushroom production was much lower for several reasons. The aim of experiments discussed in this paper was to improve the effect of composted waste paper mixed with other materials such as sand and/or peat. The main purpose was to increase the yield of mushrooms through modification of the physical structure of paper used as casing soil. Mixtures of composted paper with loamy sand in different ratios had inconsistent results. This may come from the properties of the mixture in particular the low porosity and inadequate water and air capacity.

Also the variants with mixture where nitrogen was added to the paper had not the expected results.

The best mixtures were consisting from paper, peat and sand (33%, 33%, 33%). This may be due to the high water holding capacity of peat which improved the water content of the casing soil mixture.

In one of the experiments paper globules were investigated. For a technical reason there was no nitrogen addition. The result with these globules was quite lower as expected. The size of this globules seemed to be too large, the water capacity too low and the density of this globules too high. However such globules could be the solution for using of waste paper as casing soil if the physical properties are improved. Because of the inconsistent results of the mixture of paper and sand, changing the physical shape of composted paper was suggested. Adding peat in 70% improved the water holding capacity of the mixture and resulted in a better

growth of mushroom fruit bodies. The combination of sand with paper for casing was not so successful. In further experiments, the structure of this mixture must be improved. This can be reached only if the paper particles are smaller and have a more crumbly consistence.

The positive effect of sand can be due to the good water and air capacity and high pH value.

When adding peat to the mixture (paper and sand) the water capacity of the mixture increases which resulted in a similar or better production compared to the control. Increasing the percentage of paper in the mixture on the expense of sand and peat can lead to a declining of the physical structure and together with this to lower yield.

Crumbled paper resulted in many dead primordia because of dehydration. Adding peat in 70% improved the water holding capacity of the mixture and resulted in a good production. On the other hand, mixing sand with paper for casing resulted in a negative effect compared to using sand only. This can be explained on the basis that sand had smaller interspaces which could hold more water for the growing mushroom compared to the mixture of paper and sand. This was more evident with increasing the percentage of sand in the mixture. When adding peat to the mixture this increased the water holding capacity of the mixture which resulted in a similar production compared to the control. Increasing the percentage of paper in the mixture on the expense of sand and peat resulted in lowering this capacity and reflected negatively on mushroom production.

Also an observed result was the weekly fluctuation. The fluctuation in weekly production was a common phenomenon in all experiments regardless of the type of casing soil. This means that there are other factors affecting the occurrence of this phenomenon. A strong possible factor is the water holding capacity of the casing soil as indicated earlier by (KALBERER, 1985). Another possible factor is bacterial composition of the casing soil (HAYES 1974; HAYES and NAIR, 1976). Changes in these two properties result in fluctuation in weekly production.

Conclusion:

It can be concluded that although waste paper could not completely replace peat material as a casing soil for mushroom production, this investigation proved that it can replace at least one third of this material and a total of two thirds of peat can be replaced in mushroom production.

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