Cultivation of *Pleurotus ostreatus* and *Lentinus edodes* on lignocellulose substrates for fruiting bodies and animal feed production

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Abstract

Wheat straw was fermented in the solid state with *Pleurotus ostreatus* and *Lentinus edodes* at 22°, 25° and 30°C for 30 and 60 days. During the fermentation the loss of organic matter, loss of lignin, water soluble substances and *in vitro* digestibility of substrate were increased. The supplementation of wheat bran on wheat straw and sugarcane bagasse increased the yield of fruiting bodies of *P. ostreatus* and *L. edodes.*

Keywords: lignin, in vitro digestibility, wheat straw, sugarcane bagasse, mushroom

Introduction

Lignolytic microorganisms are mainly wood degrading fungi. They are able to colonize different plant residues (Zadrazil 1979) and increase the digestibility of the substrate. In addition, lignocellulosic wastes can also be used for the cultivation of edible mushrooms (Zadrazil and Grabbe 1984). Therefore, the cultivation of wood degrading fungi has beneficial not only for human food but also for animal feed production.

In the present study, the *in vitro* digestibility of wheat straw during fungal colonization and the influence of supplementation of wheat bran on wheat straw and sugarcane bagasse as substrate for fruiting bodies production of *Pleurotus ostreatus* and *Lentinus edodes* were determined.

Materials and Methods

Fungi

The Indonesian strains of *Pleurotus ostreatus* and *Lentinus edodes* were used in the present study. The fungi were grown on malt extract agar medium at 25°C for 7 days.

Substrate Preparation

Twenty five gram of milled wheat straw (particle size ± 1 mm) were placed in 500 ml erlenmeyer flasks and added with 75 ml of deionized water. The flasks were closed with a cotton stopper and sterilized at 121°C for 30 min. After cooling, three replicates were inoculated with two agar plugs (7-mm diam.) per flask, and incubated at 22°, 25° and 30°C for 30 and 60 days. After each incubation period the substrate were dried at 105°C and milled to homogeneity. Loss of organic matter, loss of lignin (Halse 1926), pH, water soluble substances and *in vitro* digestibility (Tilley and Terry,1968) were determined.

Fruiting bodies production

Fifty g of substrate (wheat straw and sugarcane bagasse) was placed in 1500-ml jars. The substrates were supplemented with 5, 10 and 15% of wheat bran. Substrate without supplementation was used as control. Deionized water (150 ml) was added to the jars, which were then sterilized at 121°C for 30 min. Under aseptic condition, four replicates were inoculated with 3 agar plugs per jar, sealed with polypropylene and incubated in the dark at temperature 25°C for 45 days.

After the colonization period, the jars were opened and placed in a light incubator at temperature ± 18°C and relative humidity 75 - 90%. The fruiting bodies were collected until 150 days. The yield of fruiting bodies was determined after drying at 105°C, and was calculated as percent dry matter mass at the original substrate.

Results and Discussion

Loss of organic matter, loss of lignin, pH and water soluble substances

The fermentation process with white rot fungi is completed in two stages. In the first stage the fungus colonizes the substrate and utilizes easily degradable carbohydrates (Zadrazil 1977). In the second stage, lignin is degraded relatively faster than other components. In this experiment *P. ostreatus* completely colonized the substrate in 12 days, while *L. edodes* did so in 14 days under standard aerobic conditions.

Table 1 shows the influence of temperature and incubation time on organic matter decomposition, lignin degradation, pH and water soluble substances. The decomposition of organic matter of wheat straw with *P. ostreatus* was optimal at 30°C, while with *L. edodes* at 25°C. The highest loss of organic matter of wheat straw after incubation with *P. ostreatus* and *L. edodes* for 60 days were 28.9% and 23.7% respectively.

Table 1. Loss of organic matter (LOM), lignin degradation (LD), pH and water soluble substances (WSS) of substrates after fermentation with *P. ostreatus* and *L. edodes* at 22°, 25° and 30°C for 30 and 60 days.

Fungi	Days	Temp. °C	LOM (%)	LD (%)	pH	WSS (%)
P. ostreatus	30	22	11.8	17.8	4.4	14.1
		25	12:8	26.8	4.3	14.2
		30	15:3	33.1	4.5	13.8
	60	22	25.2	39.5	4.4	18.8
		25	28.0	49.5	4.5	17.1
		30	28.9	55.9	4.7	17.5
L. edodes	30	22	10.1	12.3	3.8	17.6
		25	10.5	21.3	4.3	19.0
		30	13.4	29.9	4.5	19.4
	60	22	21.2	39.6	3.9	25.5
		25	23.7	46.4	4.1	26.4
		30	20.4	44.5	4.1	19.7

The trend of lignin degradation is similar to that loss of organic matter. The highest lignin decomposition occurred in substrate fermented with *P. ostreatus* (55.9%) at 30°C for 60 days. At 30°C, lignin decomposition

began earlier than at 22° or 25°C, therefore, at highest temperatures, more lignin was decomposed than at lower temperature.

During fermentation the pH of substrate decreased until 4.3 (*P. ostreatus*) and 4.1 (*L. edodes*). After 30 days incubation with *P. ostreatus*, the concentration of water soluble substances in the substrate had decreased, due to mycelium production, but the concentration increased after 60 days of incubation. With *L. edodes,* the water soluble substances had increased after fermentation for 30 and 60 days.

In vitro *digestibility*

In vitro digestibility is the important parameter to determine the feed quality. The *in vitro* digestibility of wheat straw without fermentation was 45.8%. After fermentation with the both fungi, the *in vitro* digestibility increased (Table 2).

Table 2. Change of *in vitro* digestibility (Δ IVD) and process efficiency of substrate after fermentation with *P. ostreatus* and *L. edodes* at 22°, 25° and 30°C for 30 and 60 days

Fungi	Days	Temperature °C	Δ IVD (%)	Process Efficiency*
P. ostreatus	30	22	+ 4.6	0.39
		25	+ 5.2	0.34
		30	+ 5.6	0.20
	60	22	+ 10.6	0.90
		25	+ 11.1	0.73
		30	+ 3.8	0.14
L. edodes	30	22	+ 13.4	1.32
		25	+ 9.1	0.68
		30	+ 22.3	0.94
	60	22	+ 24.6	2.44
		25	+ 23.6	1.76
		30	+ 24.2	1.02

* Process efficiency: change of *in vitro* digestibility divided by loss of organic matter.

In general, the *in vitro* digestibility of the substrates after fermentation with *L. edodes* was higher than with *P. ostreatus.* The change of *in vitro* digestibility correlated with the increasing of the incubation time. But, there was no correlation between the temperature and the change of *in vitro* digestibility. It was also difficult to make correlation between the

loss of lignin and the change of digestibility. The amount of lignin decomposed does not always correlated with a change of digestibility (Zadrazil, 1980).

The process efficiency for *in vitro* digestibility decreased with the increasing of the incubation time, but decreased with the increasing of the temperature. The highest process efficiency (2.44) occurred in fermentation with *L. edodes* at 22°C for 60 days.

Yield of fruiting bodies

The first fruiting bodies of *P. ostreatus* were formed after 57 days on sugarcane bagasse and 103 days on wheat straw. The first fructification was earlier by the supplementation with wheat bran. The *L. edodes* grew well on wheat straw and produced the fruiting bodies after 86 days of inoculation, however the fungus did not form fruiting bodies on sugarcane bagasse.

Figure 1. Effect of supplementation of wheat bran on fruiting bodies yield of *P. ostreatus* growing on wheat straw and sugarcane bagasse

The supplementation of wheat bran increased the yield of fruiting bodies of *P. ostreatus* and *L. edodes*. The lowest yield of *P. ostreatus* (0.3%) was obtained using sugarcane bagasse and increased to 5.6% after

supplementation with 15% wheat bran. The yield of *P. ostreatus* growing on wheat straw with supplementation of 15% wheat bran was higher (8.3%), this result related with Permana *et al* (2000). The yield of *L. edodes* growing on wheat straw supplemented with 15% wheat bran was 8.7%, as compared with substrates not supplemented (1.5%).

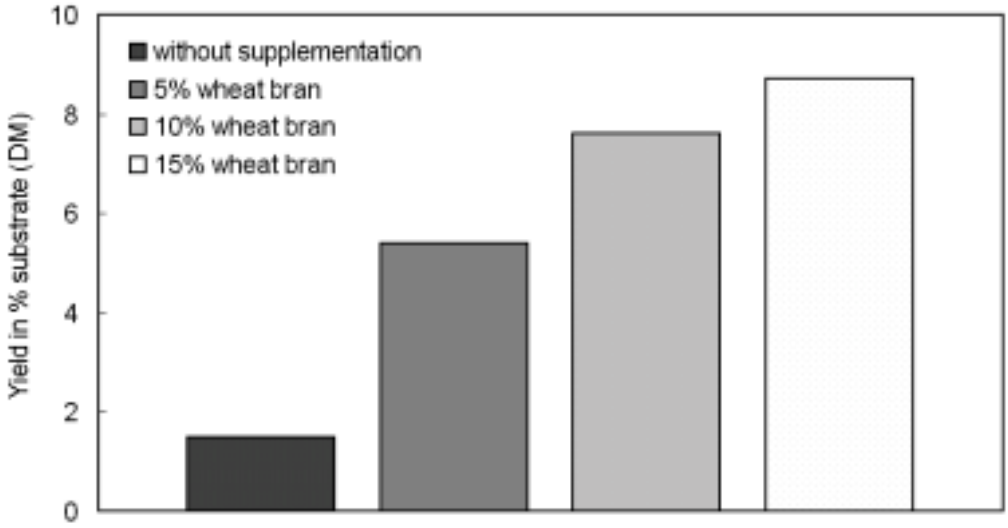

Figure 2. Effect of supplementation of wheat bran on fruiting bodies yield of *L. edodes* growing on wheat straw

Conclusion

The fermentation of wheat straw with *P. ostreatus* and *L. edodes* increased the *in vitro* digestibility. The supplementation of wheat bran on wheat straw and sugarcane bagasse increased the yield of the mushroom.

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