





## Oyster mushroom cultivation with rice and wheat straw

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Received 15 January 1999; received in revised form 24 September 2001; accepted 27 September 2001

#### Abstract

Cultivation of the oyster mushroom, *Pleurotus sajor-caju*, on rice and wheat straw without nutrient supplementation was investigated. The effects of straw size reduction method and particle size, spawn inoculation level, and type of substrate (rice straw versus wheat straw) on mushroom yield, biological efficiency, bioconversion efficiency, and substrate degradation were determined. Two size reduction methods, grinding and chopping, were compared. The ground straw yielded higher mushroom growth rate and yield than the chopped straw. The growth cycles of mushrooms with the ground substrate were five days shorter than with the chopped straw for a similar particle size. However, it was found that when the straw was ground into particles that were too small, the mushroom yield decreased. With the three spawn levels tested (12%, 16% and 18%), the 12% level resulted in significantly lower mushroom yield than the other two levels. Comparing rice straw with wheat straw, rice straw yielded about 10% more mushrooms than wheat straw under the same cultivation conditions. The dry matter loss of the substrate after mushroom growth varied from 30.1% to 44.3%. The straw fiber remaining after fungal utilization was not as degradable as the original straw fiber, indicating that the fungal fermentation did not improve the feed value of the straw. © 2002 Elsevier Science Ltd. All rights reserved.

Keywords: Mushroom; Agricultural waste; Rice straw; Wheat straw; Pleurotus sajor-caju

## 1. Introduction

Cultivation of edible mushrooms with agricultural residues, such as rice and wheat straw, is a value-added process to convert these materials, which are otherwise considered to be wastes, into human food. It represents one of most efficient biological ways by which these residues can be recycled (Madan et al., 1987). The mushrooms are protein-rich, palatable food. Their production in the United States has been steadily increasing over the past ten years (Molin, 1996). The oyster mushroom, Pleurotus sajor-caju, is one of the most successfully cultivated specialty mushrooms and is now considered to be a delicacy. Its cultivation on various lignocellulosic materials has been investigated by a number of researchers (Baskaran et al., 1978; Zadrazil, 1980a,b; Chang et al., 1981; Mira and Ragini, 1984; Bisaria et al., 1987; Madan et al., 1987). Zadrazil (1980b) showed that P. sajor-caju has a very high saprophytic colonizing ability and can degrade wheat straw efficiently. Bisaria et al. (1987) studied the growth

of this mushroom on several different agricultural wastes, including paddy straw and wheat straw, and found that the paddy straw supplemented with cotton seeds yielded the highest bioconversion efficiency of 12.82% (defined as the percentage conversion of substrate into fruit bodies of mushrooms on a dry weight basis). The protein content of the mushrooms was found to be 26.3–36.7%. Detailed analyses of straw composition during mushroom growth have not been reported. Based on previous research results, *P. sajor-caju* may be capable of nitrogen fixation (Ginterota and Gallon, 1979)

Large quantities of rice and wheat straw are produced as agricultural by-products in California. In the Sacramento Valley alone, about 1.5 million tons of rice straw are produced each year (CARB-CDFA, 1995). Alternative methods of utilizing these agricultural residues are needed to mitigate the environmental pollution problems associated with current disposal methods, such as open-field burning and soil incorporation. Mushroom cultivation on rice and wheat straw may offer economic incentives for agribusiness to examine these residues as valuable resources and develop new enterprises to use them to produce nutritious mushroom

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products. Based on 10% bioconversion efficiency and 90% moisture content in the fresh mushrooms (Madan et al., 1987; Bisaria et al., 1987), one dry ton of rice straw would yield about 1000 kg of the oyster mushroom, which would be worth \$8800 at the current retail price of \$8.8 per kg. Therefore, the mushroom cultivation may become one of the most profitable agri-businesses that could produce food products from rice and wheat straw as well as help dispose of them in an environmentally friendly manner.

The primary objectives of this study are (1) to investigate the differences between rice and wheat straw in growing *P. sajor-caju* without nutrient supplementation, (2) to determine the effects of different substrate preparation procedures on the mushroom yield, and (3) to determine the biodegradation efficiencies of rice and wheat straw and transformation of mineral elements in the straw during the mushroom growth. A secondary objective is to determine the chemical composition and fermentation characteristics of rice straw at the optimal inoculation level.

## 2. Experimental methods

Four steps were used in the mushroom cultivation procedure: straw preparation, inoculation, growing, and harvesting. Straw preparation includes particle size reduction and sterilization. To determine the effects of different straw preparation procedures on the mushroom growth, two size reduction methods (grinding and chopping) and two particle sizes for each method (0.5 and 2.5 cm for ground straw, and 2.5 and 5.0 cm for chopped straw) were tested. The hypothesis was that grinding would be better than chopping and smaller particles better than larger particles for enhancing the mushroom growth by rupturing the cell walls and increasing the surface areas of straw and making nutrients more accessible for mushroom growth, resulting in higher mushroom yields. Different levels (12%, 16% and 18%, on dry matter basis) of spawn to straw ratio were also tested in order to determine the optimum level of spawn inoculation. A lower level of spawn means lower production cost because the spawn is a relatively expensive item in mushroom production. A total of 14 test runs were performed in this study as listed in Table 1. The cultivation procedure for each test run is described in detail as follows.

## 2.1. Substrate preparation and inoculation

Rice and wheat straw were obtained as bales from a feed company in Winters, California. The straw was approximately two months in storage after harvesting. For each cultivation, the straw was first ground or chopped to a desirable particle size. Grinding was per-

formed with a hammer mill and chopping was performed with a hand cutter. The prepared straw (ground or chopped) was weighed, mixed, and placed into a 60 l plastic container filled with 45 l tap water and 0.3% (on volumetric basis) chlorine bleach. The straw was allowed to soak for 10 min to achieve sufficient moisturization and sterilization. The soaked straw was removed from the water, spread on the surface of a clean bench and air dried for 10 min to allow chlorine to evaporate and excess water to drain. The dried straw with a final moisture content of about 80% was weighed into small portions each weighing 146.4 g dry matter. Each portion was mixed with a predetermined amount of spawn, depending on the test conditions, and then put into a  $40 \times 50 \text{ cm}^2$  plastic bag. Each bag was closed with a plastic ring and a porous sponge plug on the top to prevent possible contamination by airborne organisms while allowing air exchange. The spawn of *P. sajor-caju* was obtained from a commercial supplier (Amycel, Monterey, CA). Eight bags were inoculated for each test condition. All the bags inoculated were successfully cultivated.

## 2.2. Mushroom growth and harvesting

The mushrooms were grown in a  $4 \times 4 \times 3$  m<sup>3</sup> environmental chamber where temperature, ventilation and relative humidity could be precisely controlled. Four stages of mushroom growth were carried out for each test run. The four growth stages were spawn run, primordial formation, first flush or first harvest, and second flush or second harvest. The temperature and relative humidity were controlled at  $24 \pm 1$  °C and 85– 90%, respectively, during the first stage; at  $18 \pm 1$  °C and 90–95% during the second stage; and  $22 \pm 1$  °C and 85–90% during the last two stages. These environmental conditions were determined from preliminary tests to be the best conditions. The inoculated bags were placed in the chamber with the top of each bag closed until the substrate in the bags was fully colonized, which marked the completion of the second stage of growth. The bags were then opened to allow the development of fruit bodies. For all the test runs, two flushes of mushrooms were harvested from each bag. Eight bags were used as replicates for each sampling time. At each harvesting, mushroom fruit bodies in each bag were picked manually and weighed. Very little mushroom was left on the substrate after harvesting. The substrate in the bag was also weighed.

The samples of both mushrooms and substrates were analyzed for the contents of dry matter and ash to determine the mushroom yield, bioconversion efficiency, biological efficiency, and dry matter loss of the substrate. The bioconversion efficiency was defined as the grams of dry mushrooms produced per 100 g of dry straw used. The biological efficiency was defined as the grams of

Table 1 Experimental conditions and results for *P. sajor-caju* cultivation on rice and wheat straw

Test run	Straw type	Mechanical method	Size (cm)	Initial substrate weight (dry weight, g)		Spawn level (%)	Mushroom Yield (g)			Biological efficiency	Substrate dry master loss (%)
				Straw	Spawn		First flush	Second flush	Total		
1	Rice	Ground	0.5	146.4	27.5	18	112.7 <sup>A</sup> (20.0) <sup>B</sup>	45.9 (10.5)	158.6 (33.4) <sup>a</sup>	108.0 (9.6) <sup>a</sup>	32.3 (2.5) <sup>a</sup>
2	straw		2.5				118.3 (21.2)	70.0 (18.4)	188.3 (36.9) <sup>b</sup>	128.8 (13.8) <sup>b</sup>	33.2 (4.1) <sup>b</sup>
3		Chopped	2.5	146.4	27.5	18	114.2 (18.5)	42.0 (11.0)	156.2 (28.8) <sup>c</sup>	106.8 (6.2)°	30.7 (2.0)°
4			5.0				114.0 (19.1)	38.1 (8.5)	152.1 (25.7) <sup>c</sup>	104.0 (4.9) <sup>c</sup>	30.1 (1.9) <sup>c</sup>
5	Rice	Ground	2.5	146.4	17.9	12	105.6 (20.1)	43.5 (10.2)	149.1 (28.5) <sup>d</sup>	101.8 (5.6) <sup>d</sup>	36.2 (2.6) <sup>d</sup>
6	straw				23.6	16	122.1 (32.8)	56.6 (13.4)	178.7 (30.0) <sup>b</sup>	122.1 (6.5) <sup>b</sup>	39.4 (4.1) <sup>b</sup>
7					27.5	18	124.6 (31.7)	67.2 (15.2)	191.8 (33.7) <sup>b</sup>	131.0 (9.7) <sup>b</sup>	36.7 (2.3) <sup>b</sup>
8		Chopped	2.5	146.4	17.9	12	82.0 (19.2)	28.6 (5.3)	110.6 (27.1) <sup>e</sup>	75.5 (5.4) <sup>e</sup>	29.0 (0.5)e
9		• •			23.6	16	92.1 (21.2)	45.1 (12.5)	142.2 (22.0) <sup>c</sup>	97.1 (3.0)°	29.1 (5.2)°
10					27.5	18	94.1 (25.6)	60.9 (18.3)	155.0 (32.0) <sup>c</sup>	105.9 (8.3) <sup>c</sup>	30.4 (0.8) <sup>c</sup>
11	Rice	Chopped	2.5	146.4	27.5	18	114.0 (21.4)	38.1 (10.5)	152.1 (22.0)°	104.0 (2.9)	44.1 (3.6)°
	straw										
12	Wheat						94.7 (20.8)	47.2 (14.6)	141.9 (19.1) <sup>c</sup>	97.0 (2.8)°	34.8 (2.0)°
	straw										
13	Mixture						114.0 (18.5)	34.4 (8.9)	148.4 (26.0) <sup>c</sup>	101.2 (5.0)°	42.5 (4.0)°
14	Mixture						108.0 (19.2)	50.2 (12.8)	158.2 (30.4)°	108.3 (6.7)°	44.3 (3.1) <sup>c</sup>

<sup>(</sup>a–e) Mean values within the same column with no common superscript letter differ (P < 0.1).

fresh mushrooms produced per 100 g of dry straw used. The carbon dioxide (CO<sub>2</sub>) concentration in the center of the substrate and in the headspace of each bag were analyzed periodically by taking the air samples with a gas tight syringe and analyzing them with a gas chromatograph (GC) (HP5890A, Hewlett-Packard, Avondale, PA) equipped with a thermal conductivity detector. The CO<sub>2</sub> concentration was used as an indicator for the biological activity and air exchange in each bag.

## 2.3. Analysis of compositional changes of substrate

To understand the fate of major nutrient elements, including carbon (C), nitrogen (N), phosphorus (P), and potassium (K), in the substrate during the growth of mushrooms, detailed elemental analyses were performed for two test runs (Runs 6 and 9 in Table 1). The contents of these elements in the substrate before and after mushroom growth and in the mushrooms produced were analyzed and mass balance calculations were performed. The initial substrate was the inoculated substrate, i.e. mixture of straw and spawn, and the final substrate was the spent substrate after the second flush of mushrooms was harvested. Analysis of C and N were done using a dynamic flash combustion system coupled with a GC that was equipped with a thermal conductivity detector (TCD). The method for C and N analysis is given by Pella (1990a,b). Phosphorus (P) and potassium (K) were analyzed using an inductively coupled plasma atomic emission spectrometer (ICP-AES).

The substrate contents of ash, silica, and fiber were analyzed as well as the in vitro degradation of the rice straw substrate at different times of the mushroom growth. The purpose of the in vitro degradation analysis was to determine whether P. sajor-caju fermentation would enhance the ruminant feed value or degradation properties of the colonized or spent straw. The substrates used in the test runs 6, 9 and 10 (Table 1) were sampled at different times of mushroom growth. The test runs 6 and 9 were used to study the compositional and digestibility change of the whole substrate, i.e., mixture of rice straw and spawn, during different stages of the mushroom growth, and to determine the effects of two different physical forms (chopped and ground). For test runs 6 and 9, samples were taken at zero time (0 day, just before the bag was closed), after the first harvest (23rd day for test run 6 and 28th day for test run 9), and after the second harvest (35th day for test run 6 and 40th day for test run 9). The test run 10 was used to study the compositional and digestibility change of rice straw during different stages of the mushroom growth. At each sampling, straw was manually separated from spawn. For test run 10, samples were taken at four times: zero time, after the straw was fully colonized and the bag was opened (20th day), after the first harvest (28th day) and after the second harvest (40th day). At each sampling time for runs 6, 9, and 10, sets of three bags were randomly selected, airdried and were then ground through a Wiley mill to pass a 1 mm screen and stored for further analyses.

Analyses of the ground samples were done in duplicate for dry matter, ash, (AOAC, 1990), neutral detergent fiber (NDF) (Van Soest et al., 1991), acid detergent fiber (ADF), lignin, and silica (Goering and Van Soest, 1970). The NDF remaining after 12 and 48 h in vitro

A Mean value.

<sup>&</sup>lt;sup>B</sup> Standard deviation.

fermentation were determined using the procedure presented by Goering and Van Soest (1970). All the fiber components were determined on an ash-free basis (Crocker et al., 1998). All the results are presented on a dry matter basis.

## 2.4. Statistical analysis of experimental data

Microsoft Excel was used for the statistical analysis of the mushroom yield data to determine the significance of the test conditions. A two factorial *T*-test was performed to compare the different straw size reduction methods, types of straw, particle sizes, and spawn levels.

The general linear models procedure of SAS (1987) was used for analysis of rice substrate compositions and degradability. The model for the test runs 6 and 9 included the main effect of time of sampling (zero, first harvest, and second harvest), physical form (chopped and ground), and the interaction of time × physical form. The interaction was not significant at all *P* values greater than 0.5. The model for the test run 10 included the main effect of time of sampling. If the main effects were significant, the means were separated using Duncan's multiple range test (Snedecor and Cochran, 1980).

#### 3. Results and discussion

The mushroom yield, bioconversion efficiency, biological efficiency, and substrate dry matter loss after the mushroom growth for different test conditions are given in Table 1. The test runs 1 to 4, 5 to 10, and 11 to 14, were carried out as three separate cultivation runs to determine the effects of size reduction method and particle size, spawn inoculation level, and type of straw on the mushroom growth and substrate degradation, respectively. Each datum in Table 1 is the average of eight replicate determination. The test conditions for test runs 3, 10, and 11 were the same but the results exhibited a slight variation. This could be due to experimental errors introduced by different cultivation runs.

# 3.1. Effects of straw size reduction method and particle size

Generally speaking, grinding yielded better results than chopping, perhaps because it ruptured the cell walls of the straw to a greater degree, potentially making the nutrients in the straw more accessible for mushroom growth. The mushrooms also grew faster on the ground straw with their growth cycles being five days shorter than on the chopped straw. Among the four types of substrate tested, the ground, 2.5 cm rice straw showed the best results in terms of mushroom yield, bioconversion efficiency, biological efficiency, and substrate dry matter loss. No significant difference was found between

the two sizes (2.5 and 5.0 cm) of the chopped straw (P>0.1). With the same particle size (2.5 cm), the ground substrate resulted in 20.5% higher mushroom yield than the chopped substrate, which was significant (P<0.1). Further reduction of the particle size by grinding the straw to 0.5 cm, however, resulted in a lower mushroom yield. This could be because the particles that were too small resulted in the over-compaction of the substrate in the bags, which may have led to hindered air exchange between the void spaces in the substrate and headspace, especially when the bags were closed. This reasoning is further evidenced by the higher  $CO_2$  concentration profile in the center of ground, 0.5 cm substrate as shown in Fig. 1. The  $CO_2$  was presumably produced from the respiration of the fungi.

The  $CO_2$  concentrations in the center of both ground substrates (0.5 and 2.5 cm) were higher than in the chopped substrates (2.5 and 5.0 cm) as shown in Fig. 1. This might have resulted from faster growth of the mushroom in the ground substrate. The  $CO_2$  concentrations in the headspaces of all four types of substrates were similar, as shown in Fig. 2, indicating similar air

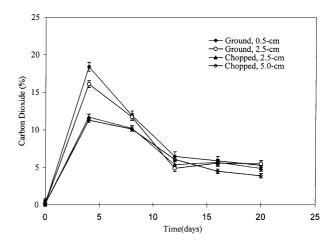


Fig. 1. Carbon dioxide concentration in the center of the substrate.

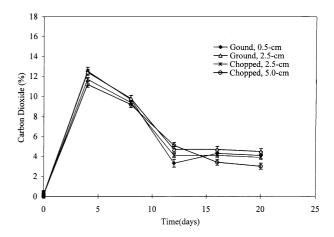


Fig. 2. Carbon dioxide concentration in the headspace of the substrate.

exchange rates between the headspace of each bag and the surrounding environment. It can also be seen that the CO<sub>2</sub> concentrations in all four substrates increased quickly immediately after inoculation and reached the maximum on the fourth day, indicating that the mushroom mycelia colonized the substrate rapidly in the first few days of cultivation.

It has been reported in the literature that the concentrations of CO2 and O2 play important roles in the growth of fungi and the production of mushrooms. The effects of CO<sub>2</sub> vary with the types and strains of the fungi. Jenson (1967) reported that the growth of Agaricus is inhibited by CO<sub>2</sub> concentrations above 2%. Donoghue and Denison (1995) studied the effect of CO<sub>2</sub> and O<sub>2</sub> concentrations inside plastic mushroom growing bags on the colonization and growth of Shiitake mushrooms. They found that the visible, biological indications of fungal development (colonization, lumping and browning and pinning in the bags) appeared to be significantly affected by the concentrations of  $CO_2$  and  $O_2$ . Their general observation was that the cultures in the bags with lower CO<sub>2</sub> concentrations and higher O<sub>2</sub> concentrations developed more rapidly. Detailed documentation of the effects of CO2 on the growth of P. sajor-caju has not been found in the literature. However, inhibitory effects of the high concentrations of CO<sub>2</sub> inside the mushroom growth bags or rooms on the fungal development and mushroom production are well understood.

## 3.2. Effects of spawn inoculation level

Spawn inoculation level is an important factor that relates to the production costs of the mushroom cultivation. Compared to the straw, spawn is a relatively expensive item. Using the lowest inoculation level possible without sacrificing the mushroom yield would give the best economics. The spawn level is defined as the weight ratio of spawn to substrate on a dry weight basis. The optimum spawn level depends on the type of substrate, mushroom species, spawn quality, and the cultivation conditions. Among the three spawn levels (12%, 16%, and 18%) tested, the 12% spawn level resulted in significantly lower mushroom yield and biological efficiency than the other two levels. The mushroom yield at the 12% level was 22.2% lower than at the 16% level. The mushroom growth cycle at the 12% level was longer. No statistically significant difference was found between 16% and 18% levels (P > 0.1), even though the mushroom yield and biological efficiency at 18% level were slightly higher than at 16% level. Therefore, based on these results, it would be recommended that at least 16% spawn level be used to achieve a desirable mushroom yield. In real production, bench-scale cultivation tests as performed in this study are recommended for determining the optimum spawn inoculation levels.

## 3.3. Effects of substrate characteristics

Four types of substrate were compared to determine the differences between rice and wheat straw in mushroom cultivation. They were rice straw, wheat straw, mixture of rice and wheat straw in 1:2 dry weight ratio, and mixture of rice and wheat straw in 2:1 dry weight ratio. Generally speaking, rice straw was a better substrate than wheat straw. The mushroom yield with rice straw was about 10% higher than with wheat straw and the yield with the mixture of 2:1 rice straw to wheat straw was also about 10% higher than the other mixture which contained less rice straw. The findings of this study agree with the findings of Bisaria et al. (1987) who reported that the P. sajor-caju yield with rice straw was 11.7% higher than with wheat straw. But the statistical analysis results of this study show that this yield increase is not statistically significant (P > 0.1).

## 3.4. Substrate degradation and nutrient utilization

The dry matter loss is in agreement with the mush-room yield and biological efficiency. The higher mush-room yield and biological efficiency corresponded to the higher dry matter loss. The dry matter lost was partly assimilated into the mushroom fruit bodies and partly lost into the atmosphere as carbon dioxide due to mushroom respiration. The dry matter loss of the substrate with all the test runs varied from 30.1% to 44.3%. This is substantial mass reduction from the standpoint of straw disposal. The biological efficiency is in the range 75.5–131.0% (Table 1). With the average moisture of mushrooms harvested being 92.5%, the corresponding bioconversion efficiency is in the range 5.7–9.8%.

The elemental analysis results with two types of substrate (ground and chopped) are given in Table 2. To make the data easy to visualize, the initial weight of the substrate has been adjusted to 100 g and all the other data have been adjusted accordingly. The initial contents of nutrients except for K were about the same in both ground and chopped substrates. The K content in the ground straw was lower than in the chopped straw, due to leaching that occurred in the straw moisturization process. It can been seen that the C, N, P, and K assimilated into the mushrooms accounted for 9.3%, 42.8%, 70.3%, and 36.7% of these elements in the original substrate, respectively, for the ground substrate and 7.7%, 35.4%, 58.3%, and 20.1%, respectively, for the chopped substrate. The higher uptake of these elements with the ground substrate is in agreement with the higher yield of mushrooms with this substrate. The protein content of the mushrooms (calculated by multiplying the nitrogen content by 6.25) was about 27.2%, which is within the range of protein content of P. sajorcaju (26.3-36.7%) reported by Bisaria et al. (1987). The C and N losses were 38.2% and 13.2% for the ground

Table 2 Nutrient contents of substrate and mushrooms

Rice straw	Materials	Dry weight (g)	Nutrient content (%)						
			C	N	P	K			
Ground	Initial substrate	100.0	35.1 (0.6)	0.81 (0.02)	0.11 (0.001)	0.77 (0.01)			
	Spent substrate	67.7	31.5 (0.4)	0.54 (0.01)	0.05 (0.001)	0.71 (0.01)			
	Mushroom	8.0	40.9 (0.2)	4.35 (0.10)	0.97 (0.01)	3.55 (0.07)			
Chopped	Initial substrate	100.0	35.1 (0.6)	0.81 (0.02)	0.11 (0.001)	1.17 (0.02)			
**	Spent substrate	69.3	32.5 (0.3)	0.64 (0.01)	0.06 (0.001)	1.23 (0.01)			
	Mushroom	6.6	40.9 (0.2)	4.35 (0.10)	0.97 (0.01)	3.55 (0.07)			

Note: Nutrient content is calculated as the percentage of dry matter.

substrate, respectively; 35.8% and 11.3% for the chopped substrate, respectively. The carbon loss was due to the mushroom respiration. The N loss might be due to volatilization during the N mineralization process. The P and K contents were well balanced among the initial substrate, spent substrate, and mushrooms.

The substrate composition of ash, silica and fiber and the degradation characteristics of fiber are shown in Table 3. The time of sampling was more important than physical size. The physical form of the straw had little effect on the changes of all components measured. However the ash content was lower (P < 0.1) for the chopped straw, which supports the previous observations in this study that the mushrooms grown on ground straw had higher yields than mushrooms grown on chopped straw. The contents of ash and silica increased

with time (P < 0.1). The contents of other components remained the same or decreased. If the content of a component remained the same or decreased, the implication is that P. sajor-caju utilized the component. The fiber plus ash remained similar or decreased slightly over time indicating that the decrease in NDF was relatively consistent over time compared to the increase in ash. Both cellulose and hemicellulose decreased over time while lignin remained fairly constant. The decrease in hemicellulose was relatively greater than cellulose implying that the fungi utilized a greater percentage of hemicellulose than cellulose. The fungi also used some lignin. The amount of NDF or fiber degraded in 48 h was less for the second stage harvest compared to zero sampling time. The amount of NDF degraded in 48 h decreased over time as the amount of initial NDF

Table 3
Composition and digestibility of rice straw and spawn mixture from two physical forms sampled at three times during mushroom growth

Time <sup>A</sup>	Physical <sup>A</sup>	Ash	Silica	$NDF^{B}$	$Hemi^{C}$	$\mathrm{ADF^D}$	Cell <sup>E</sup>	ADF lignin	NDF degraded between hou		een hours
									0-12 <sup>F</sup>	12–48	0-48
ZT	Ch	18.26	13.01	63.33	24.77	38.56	34.80	3.76	10.47	22.28	32.75
1st	Ch	25.47	18.83	53.27	17.98	35.29	31.22	3.96	6.35	19.57	25.92
2nd	Ch	28.13	21.03	48.71	15.48	33.22	29.35	3.76	6.17	15.24	21.41
ZT	Gr	19.56	14.06	62.98	24.10	38.87	35.02	3.85	10.58	21.26	31.83
1st	Gr	26.11	19.41	53.44	18.21	35.23	31.35	3.87	7.63	18.03	25.66
2nd	Gr	29.55	21.08	48.58	15.86	32.73	29.24	3.59	7.09	13.83	20.92
Time											
Ave ZT		18.78 <sup>c</sup>	13.43°	63.19 <sup>a</sup>	24.51a	38.69a	34.89a	3.79	10.51a	21.87a	32. 38 <sup>a</sup>
Ave 1st		$25.79^{b}$	19.12 <sup>b</sup>	53.35 <sup>b</sup>	18.09 <sup>b</sup>	35.26 <sup>b</sup>	31.29 <sup>b</sup>	3.92	$6.99^{b}$	18.80 <sup>b</sup>	25.79 <sup>b</sup>
Ave 2nd		28.84a	21.05a	48.64°	15.67°	$32.97^{\circ}$	29.30°	3.67	6.63 <sup>b</sup>	14.53°	21.16 <sup>c</sup>
S.E.		0.581	0.425	0.687	0.442	0.545	0.569	0.163	0.566	0.766	0.932
Physical											
Ave Ch		23.95a	17.62	55.10	19.41	35.69	31.79	3.83	7.66	19.03	26.69
Ave Gr		25.76 <sup>b</sup>	18.70	54.00	18.80	35.20	31.48	3.76	8.16	17.26	25.42
S.E.		0.712	0.521	0.841	0.541	0.668	0.696	0.199	0.694	0.938	1.141

(a–c) Means within the same column with no common subscript letter differ (P < 0.1).

S.E. - Standard error.

A Times are ZT = zero time, 1st = first harvest, 2nd = second harvest. Physical: Ch = chopped, Gr = ground.

<sup>&</sup>lt;sup>B</sup> NDF = neutral detergent fiber.

<sup>&</sup>lt;sup>C</sup> Hemi = Hemicellulose = NDF – ADF.

 $<sup>^{\</sup>mathrm{D}}\mathrm{ADF}\!=\!\mathrm{acid}$  detergent fiber.

<sup>&</sup>lt;sup>E</sup> Cell = cellulose = NDF - Lignin.

F<sub>0</sub>-12, 12-48, 0-48: NDF degraded between 0 and 12 h, 12 and 48 h, and 0 and 48 h, respectively. Note this is in vitro fermentation time not time of mushroom growth.

Table 4
Composition and digestibility of rice straw sampled four times during mushroom growth

_	_			_	_	_				
Time <sup>A</sup>	Ash	Silica	$NDF^{B}$	Hemi <sup>C</sup>	$\mathrm{ADF}^\mathrm{D}$	Cell <sup>E</sup>	ADF lignin	NDF degraded between hours		
								0-12 <sup>F</sup>	12–48	0-48
ZT	19.53 <sup>d</sup>	14.62 <sup>d</sup>	67.54 <sup>a</sup>	25.67a	41.86a	38.23a	3.75	6.77	27.36 <sup>a</sup>	34.13 <sup>a</sup>
OB	23.21°	$17.00^{\circ}$	58.29 <sup>b</sup>	19.42 <sup>b</sup>	$38.87^{b}$	$35.09^{b}$	3.77	4.50	23.24 <sup>b</sup>	$27.74^{b}$
1st	25.39 <sup>b</sup>	18.92 <sup>b</sup>	55.21°	$19.10^{\rm b}$	36.11°	$32.49^{\circ}$	3.62	3.91	22.63bc	26.55bc
2nd	$28.20^{a}$	21.22a	$52.00^{d}$	18.15 <sup>b</sup>	$33.85^{d}$	29.91 <sup>d</sup>	3.92	5.39	19.39°	24.78°
S.E.	0.204	0.305	0.445	0.402	0.346	0.462	0.214	0.874	0.909	0.552

(a-c) Means within the same column with no common subscript letter differ (P < 0.1).

decreased and a smaller percentage of the initial NDF was digested in the later stages compared to initial NDF. This finding implies that the NDF remaining after fungi utilization was not as degradable as the initial substrate NDF. The dry matter loss of substrate after mushroom growth can be estimated by using the ash content of initial and second harvest. Based on change in ash content, the loss in dry matter substrate was calculated to be about 35%, which supports the direct measurements, made in this study. Similar results are shown in Table 4 even though the spawn was removed. The amount of dry matter lost was about 31%, which is slightly lower. This lower dry matter loss is probably because the spawn was removed.

## 4. Conclusions

The major findings of this study are summarized as follows. The ground straw yielded better mushroom production than the chopped straw. The mushrooms grew faster on the ground substrate with their growth cycles being five days shorter than on the chopped straw for a similar particle size. However, it was found that when the straw was ground into particles that were too small, such as 0.5 cm, the mushroom yield was lowered. The ground, 2.5 cm straw yielded the best results. No significant difference was observed between the two sizes, 2.5 and 5.0 cm, of chopped straw. The results of this study showed that too low a spawn level would result in a lower mushroom yield and a longer growth cycle. With the three levels tested (12%, 16% and 18%), the 12% level resulted in significantly lower mushroom yield than the other two levels. Although a 16% spawn level is recommended for mushroom production, the optimum spawn level should be specifically determined with experiments under actual production situations. Comparing rice straw with wheat straw, rice straw yielded about

10% more mushrooms than wheat straw under the same cultivation conditions. The average protein content of mushrooms was 27.2%.

The dry matter loss of the substrate after mushroom growth varied from 30.1% to 44.3%. Substrate fiber remaining after fungi utilization was not as degradable as substrate fiber at the initial stage of fungi utilization. Therefore, improvement of feed value of the straw through fungal fermentation was not demonstrated in this study. The loss in dry matter is significant in terms of mass reduction of straw for later disposal. Therefore, mushroom cultivation proves to be a highly efficient method for disposing of agricultural residues, such as rice and wheat straw, as well as producing nutritious human food. The alternative uses for the spent straw after mushroom production include animal feed, soil conditioner, and the feedstock for energy generation via combustion or anaerobic digestion.

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S.E. - Standard error.

A Times are ZT = zero time, OB = open bag, 1st = first harvest, 2nd = second harvest.

<sup>&</sup>lt;sup>B</sup> NDF = neutral detergent fiber at zero time, NDF\_12 = NDF remaining after 12 h or fermentation, NDF\_48 = NDF remaining after 48 h of fermentation.

<sup>&</sup>lt;sup>C</sup> Hemi = hemicellulose.

 $<sup>^{\</sup>mathrm{D}}\mathrm{ADF} = \mathrm{acid}$  detergent fiber.

 $<sup>^{</sup>E}$  Cell = cellulose.

F 0-12, 12-48, 0-48: NDF degraded between 0 and 12 h, 12 and 48 h, and 0 and 48 h, respectively. Note this is in vitro fermentation time not time of mushroom growth.

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