Biodegradation of Viticulture Wastes by *Pleurotus*: A Source of Microbial and Human Food and Its Potential Use in Animal Feeding

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The bioconversion of vineyard pruning and grape pomace by *Pleurotus* spp. using a solid state fermentation (SSF) was evaluated. Fruiting body production and chemical changes in the substrates after harvesting were measured. Biological efficiency and bioconversion ranged from 37.2 to 78.7% and from 16.7 to 38.8%, respectively. The best substrates for mycelial growth and mushroom yield were the mixtures with higher vineyard pruning content. Inclusion of pruning content had higher phenolic components and total sugars, better C/N ratio, and lower crude fat and total nitrogen than pomace. On the contrary, mycelium grew more slowly and scarcely in all treatments with 100% grape pomace. Moisture, protein, fat, and lignin contents were generally higher in mixtures with higher pomace proportion, whereas neutral detergent fiber, hemicellulose, and cellulose contents were higher with pruning content. *Pleurotus* strains may act depending on the availability of fiber fractions of substrate, and dynamic changes in digestion might occur as these fractions change during fungal growth. The recycling of viticulture residues through SSF by *Pleurotus* has great potential to produce human food and yields an available high-fiber feed for limited use in ruminants.

KEYWORDS: Biotransformation; *Pleurotus*; recycling; solid-state fermentation; viticulture byproducts

INTRODUCTION

Viticulture is an important activity in northwestern Mexico, where 33500 ha are cultivated with several vine cultivars (*1*). As a result of this activity, ~270000 tons of agroindustrial wastes are produced every year, of which 93% are vineyard prunings. This waste is usually burned in the field to prevent proliferation of phytopathogens, causing environmental and ecological problems and a risk to human health. Lignin, a main contributor of the total carbon of agroindustrial wastes, produces polycyclic aromatic hydrocarbon components such as benzopyrene, catechol, hydroquinone phenanthrene, and naphthalene when degraded by heat (*2*). All of these compounds can inhibit DNA synthesis and induce cancerous tumors in liver, lung, larynx, and cervix in animals and humans (*3*).

Another waste produced by viticulture is grape pomace. This byproduct is generally disposed of in open areas. Pomace can be used as animal feed, especially in the dry season when pastures are scarce. Its use is limited only up to 30% of the feed for ruminants due to its very low nutritional value and its antinutritional factors such as phenolic components that inhibit

the ruminal symbionts (*4*, *5*). High costs of handling and transportation usually limit the direct benefit of feeding animals with many agricultural byproducts.

On the other hand, *Pleurotus ostreatus-complex* is the third most important edible mushroom cultivated worldwide (*6*). It can efficiently decompose lignocellulose without chemical or biological pretreatment because it possesses an enzymatic complex system that includes phenol oxidases and peroxidases (*7*). Therefore, a broad variety of lignocellulosic wastes can be utilized and recycled by solid state fermentation (SSF) with this mushroom (*8*). Vail et al. (*9*) observed that *P. ostreatus-complex* was often predominant in a wood decay complex on grapevines in northern California.

In an attempt to find alternatives for the recycling of winery agroindustrial wastes, the bioconversion of vineyard prunings and grape pomace by *Pleurotus* spp. with SSF was evaluated by measuring the fruiting body production for human consumption. The chemical changes of spent substrate were determined after mushroom harvesting for potential use in ruminant feeding.

MATERIALS AND METHODS

Strains. Two strains of *Pleurotus ostreatus* (Jacq.:Fr.) Kumm. (CCMC H-041 and IE-8) and one of *Pleurotus pulmonarius* (Fr.:Fr.) Quél. (IE-115) were studied. The CCMC H-041 is from the Strains Collection of CIATEJ (Jalisco, Mexico), whereas strains IE-8 and IE-

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115 are from the INECOL (Xalapa, Mexico). The microorganisms were kept at 4 °C for preservation. Strain propagation was carried out at 28 ± 1 °C on a medium of malt extract agar.

Substrate Preparation and Mushroom Cultivation. Viticulture byproducts were obtained from vineyards of the coast of Hermosillo and a winery located in Hermosillo, Sonora, Mexico. Vineyard prunings were dried and cut into pieces 8−10 mm long. Grape pomace was used as disposed of from the winery. Samples of 500 g (dry weight) were adjusted to a moisture content of 70%, pasteurized at 85 °C for 1 h, and then cooled to room temperature. Five mixtures (1:0, 2:1, 1:1, 1:2, and 0:1) of vineyard prunings and grape pomace were evaluated for each strain (in three replicates). The inoculum was prepared according to the method of Guzmán et al. (*10*) on wheat grains. Substrate mixtures were inoculated at a 5% rate of spawn in a 50 × 30 cm plastic bag and incubated in darkness at 28 ± 1 °C. A 12 h photoperiod with a temperature of 25 ± 1 °C and a relative humidity of 80−90% constituted the conditions for fructification. Comparison between strains was based on biological efficiency (percentage of yield of fresh mushroom in relation to dry weight of substrate). Fruiting body production was classified according to pileus diameter in three groups: < 5 cm, 5−10 cm, and >10 cm. Bioconversion was determined by the loss of dry weight of substrate after mushroom harvesting.

Chemical Analyses. Prior to inoculation and after harvesting, the following analyses were performed to determine changes in substrate composition due to the SSF: moisture by oven dehydration at 100 °C for 8 h (*11*); ash by weighing the incinerated residue obtained at 525 °C after 4 h (*12*); crude fat by Goldfisch extraction with petroleum ether (*12*); crude protein by a micro-Kjeldahl method (*13*); and neutral detergent fiber (NDF), hemicellulose, cellulose, lignin, and silica by a modified Van Soest and Wine technique (*14*) using a Tecator Fibertec apparatus. Total phenols were estimated according to the procedure of Singleton and Rossi (*15*) and total sugars according to the method of Dubois et al. (*16*). Fatty acids were determined by using a modified method of the AOAC (*12*) by GC-MS. Lipids were separated in a Varian 3800 gas chromatograph equipped with an ion trap mass detector (Saturn 2000) and AT Wax glass capillary column (25 m × 0.25 mm i.d. × 0.25 μm film, Supelco). The oven temperature program was of an initial temperature of 50 °C followed by a temperature rise of 10 °C/min to 160 °C and then 1 °C/min to 216 °C. Helium was the carrier gas (1 mL/min). The mass spectra were recorded at 70 eV and an ion source temperature of 200 °C (mass range of 40−500 amu) and compared with a spectral database (NIST 98). All analyses were done by three replicates.

Statistical Analysis. Data of yield, biological efficiency, and chemical analysis were subject to random two-way (strains vs mixtures) analysis of variance and Tukey test ($p < 0.05$) to determine the significance of differences between the mean values using the SAS (*17*) computer program.

RESULTS AND DISCUSSION

Fruiting Body Production. The best substrates for mycelial growth for all studied strains were the mixtures with higher vineyard pruning content. A compact mass was formed because of total covering of mycelium on these substrates. On the contrary, in all treatments with 100% pomace, the mycelium grew more slowly and scarcely, being conspicuously thin. The mycelium of the fungus colonized the substrate within a period of 12 days of spawn run on 2:1 pruning/pomace mixture. The first pinheads were observed on this mixture with CCMC H-041 strain. The rest of the substrates and strains showed pinheads after 15−17 days. Flushes and biological efficiency (BE) of all strains and mixtures are shown in **Table 1**. The highest BE was obtained in 100% pruning substrate inoculated with strain IE-8 (78.7%), whereas the lowest in 100% pomace substrate was with strain CCMC H-041 (37.3%). Two flushes were obtained for all treatments, and mushroom harvesting was finished the 40th day after the spawn was inoculated. During the first flush, 55−89% of the yield was collected, and 11−45% was collected in the second.

Table 1. Biological Efficiency and Bioconversion Percentage of *Pleurotus* Cultivated on Mixtures of Viticulture Byproducts

Pleurotus strain	mixture pruning/ pomace	BE[a] (%)	bioconversion after fructification[a] (%)
CCMC H-041	1:0	64.80 ± 6.1[a−c]	34.93 ± 0.82[ab]
	2:1	55.53 ± 10.0[a−c]	29.76 ± 1.71[bc]
	1:1	57.80 ± 1.10[a−c]	30.76 ± 0.69[bc]
	1:2	54.53 ± 2.30[a−c]	27.88 ± 1.55[cd]
	0:1	37.27 ± 6.90[ac]	20.11 ± 0.15[e−g]
IE-115	1:0	58.13 ± 3.13[a−c]	34.96 ± 1.81[ab]
	2:1	56.93 ± 19.43[a−c]	26.17 ± 1.21[cd]
	1:1	62.67 ± 7.60[a−c]	30.84 ± 1.87[bc]
	1:2	44.60 ± 12.08[bc]	23.06 ± 1.70[d−f]
	0:1	38.27 ± 11.95[a−c]	16.74 ± 0.44[g]
IE-8	1:0	78.73 ± 15.48[bc]	38.89 ± 0.27[a]
	2:1	70.73 ± 20.58[ab]	30.27 ± 0.69[bc]
	1:1	53.67 ± 3.45[a−c]	29.31 ± 5.81[bc]
	1:2	50.53 ± 6.93[ab]	25.74 ± 0.31[c−e]
	0:1	40.93 ± 5.74[bc]	19.42 ± 2.19[fg]

[a] All values are means ± SD of triplicate measurements. Means in a column with different superscripts are significantly different ($p < 0.05$, Tukey).

The best growth, defined by complete colonization of substrates within 12 days of spawn run and the greatest quantity of edible biomass harvested, was obtained from the mixtures with higher vineyard pruning contents. When *Pleurotus* spp. were cultivated on other agroindustrial wastes, fruiting body production began to appear 17−42 days after inoculation (*10, 18, 19*). Strain IE-8 cultivated on pepper leaves, lemon grass leaves, coffee pulp, and cinnamon leaves began to fructify at 15, 18, 20, and 21 days, respectively (*20*). The superiority of the 100% pruning was also evident by the BE in all tested strains.

The most homogeneous pileus size with an 18% coefficient of variation and best commercial size (5−10 cm) were observed for strain IE-115 cultivated on a 2:1 pruning/pomace mixture. From the first flush, 18−23% of fruiting bodies had a diameter between 5.0 and 10 cm, and 21−75% of mushrooms had the same size from the second flush. The smallest pileus size (<5 cm) was present in strain IE-8 cultivated on 100% pruning substrate. This size was present in 56.5−98.2% of basidiomata production in the first flush and in 88.4−100% in the second. Overall, pileus size >10 cm was the least frequently found. The highest percentage (9.9%) of harvesting with this size was obtained in strain CCMC H-041 inoculated in a 1:1 pruning/ pomace mixture. The chemical composition of fruiting bodies of *Pleurotus* harvested on viticulture byproducts, as evaluated in a separate work, showed good nutritional chemical composition (*21*).

Variable ranges of BE have been reported when different lignocellulosic wastes were used as substrate for production of oyster mushroom. The BE results shown in **Table 1** are similar to those obtained by cultivation of *Pleurotus* on sugar cane leaves, where BE ranged from 40.9 to 89.4% (*22*). On the other hand, the efficiency of strain IE-8 cultivated on viticulture wastes was lower than that reported for coffee pulp and dry corn leaves (*10*). The variation on BE agrees with previous reports of *Pleurotus* spp. cultivation on other agroindustrial wastes (*20*).

Bioconversion as a result of mycelial growth and basidiomata production of the three strains of *Pleurotus* developed in five mixtures of winery byproducts is shown in **Table 1**. The highest catabolic and enzymatic activity of macromycetes occurs during fructification; therefore, the percentage of bioconversion by

Table 2. Composition of Intact Viticulture Byproducts (Control) and after Solid State Fermentation of *Pleurotus* Species

Pleurotus strain	mixture pruning/pomace	moisture[a]	crude protein[a]	fat[a]	ash[a]	NDF[a]
control	1:0	6.57 ± 0.52[h]	3.91 ± 1.06[bc]	0.51 ± 0.28[g]	4.30 ± 0.62[a]	76.64 ± 0.29[a−d]
	2:1	7.03 ± 0.21[h]	6.24 ± 0.57[a−c]	2.90 ± 0.19[ef]	4.79 ± 0.26[a]	74.81 ± 1.29[ef]
	1:1	6.94 ± 0.14[h]	6.16 ± 0.22[a−c]	2.06 ± 0.15[f]	4.75 ± 0.35[a]	74.50 ± 0.91[ef]
	1:2	7.24 ± 0.29[h]	6.49 ± 0.57[a−c]	3.70 ± 0.19[de]	5.12 ± 0.05[a]	72.89 ± 1.79[e−g]
	0:1	7.30 ± 0.09[h]	7.94 ± 1.21[a−c]	5.72 ± 0.44[a]	5.73 ± 0.24[a]	72.45 ± 1.65[e−g]
CCMC H-041	1:0	9.96 ± 0.12[e−g]	2.73 ± 0.22[c]	0.43 ± 0.13[g]	4.27 ± 0.12[a]	79.32 ± 1.05[ab]
	2:1	11.53 ± 0.12[a−d]	1.27 ± 0.17[c]	4.59 ± 0.49[a−d]	5.37 ± 0.12[a]	73.66 ± 0.97[ef]
	1:1	11.00 ± 0.62[a−e]	5.61 ± 0.80[a−c]	2.80 ± 0.20[ef]	5.28 ± 0.22[a]	73.36 ± 0.95[e−g]
	1:2	11.76 ± 0.39[a−c]	6.85 ± 1.01[a−c]	4.61 ± 0.20[a−d]	5.52 ± 0.36[a]	71.01 ± 1.78[e−h]
	0:1	12.14 ± 0.45[a]	10.66 ± 0.54[a]	5.75 ± 0.57[a]	5.83 ± 0.29[a]	71.57 ± 1.47[e−h]
IE-115	1:0	10.00 ± 0.35[e−g]	4.11 ± 1.77[a−c]	0.43 ± 0.13[g]	4.41 ± 0.46[a]	80.06 ± 2.74[a]
	2:1	10.80 ± 0.50[b−f]	3.96 ± 1.25[a−c]	3.77 ± 0.74[de]	5.47 ± 0.17[a]	73.74 ± 2.13[ef]
	1:1	10.57 ± 0.19[c−f]	5.67 ± 1.88[a−c]	3.55 ± 0.36[de]	5.30 ± 0.32[a]	75.22 ± 0.61[e]
	1:2	11.50 ± 0.39[a−d]	1.76 ± 0.39[c]	4.53 ± 0.85[a−d]	5.50 ± 0.50[a]	75.17 ± 1.57[e]
	0:1	11.80 ± 0.46[ab]	4.26 ± 0.67[a−c]	5.45 ± 1.00[a−c]	5.41 ± 0.31[a]	74.89 ± 0.49[ef]
IE-8	1:0	9.07 ± 0.40[g]	3.11 ± 0.50[c]	0.29 ± 0.04[g]	5.47 ± 0.19[a]	77.97 ± 0.64[a−c]
	2:1	10.37 ± 0.57[d−f]	6.50 ± 0.43[a−c]	4.19 ± 0.47[b−e]	4.72 ± 0.20[a]	74.46 ± 0.34[ef]
	1:1	9.70 ± 0.66[fg]	5.84 ± 0.51[a−c]	3.19 ± 0.38[d−f]	4.42 ± 0.18[a]	72.93 ± 2.02[e−g]
	1:2	10.67 ± 0.38[b−f]	7.38 ± 0.35[a−c]	4.07 ± 0.34[c−e]	4.61 ± 0.18[a]	73.17 ± 1.08[e−g]
	0:1	11.25 ± 0.31[a−d]	8.50 ± 0.84[ab]	5.55 ± 0.60[ab]	5.17 ± 0.28[a]	73.18 ± 0.80[e−g]

[a] All values are means ± SD of triplicate measurements. Means in a column with different superscripts are significantly different ($p < 0.05$, Tukey).

metabolism of each strain was determined after cropping (*23*). The phenology and the bioconversion of strains changed according to variation in the substrate's content. The lowest bioconversion was found in 100% pomace substrate (16.74%) with strain IE-115, whereas the highest in 100% pruning substrate (38.89%) was obtained with strain IE-8; other values for bioconversion are shown in **Table 1**. BE and bioconversion varied significantly among treatments ($p < 0.05$).

The lowest bioconversion and BE were observed on the 100% pomace substrate with all strains. This behavior could have resulted from the high levels of fatty compounds and perhaps the differences in total nitrogen content (**Table 2**). The fat content of pomace is 11 times higher than in pruning (5.72 vs 0.51%). Rajarathnam and Bano (*23*) mentioned that low levels of fat stimulated the growth of *Pleurotus sapidus;* maybe this is an additional event for growth enhancement in pruning. In a separate experiment, fatty substances were extracted using toluene solvent from a 1:1 pruning/pomace mixture. The radial growth rate was measured and indicated a poor growth (70% of inhibition) compared with control culture, and a thin mycelial mat formation was observed just as in pomace straw (data not shown). The effect of fatty substances has been studied in detail from the early 1970s. Several authors have shown a negative effect on growth by saponins, sterols, glycerol, short-chain fatty acid esters, and unsaponifiables in cultures of *Agaricus bisporus*, *Pleurotus* spp., and yeast (*24−27*). The toxic effect of lipids is closely associated with the final products of lipid peroxidation, mainly aldehydes and acrolein (*28, 29*). Composition analysis of extracted fatty acids showed a prevalence of C18:2, C16:0, and C18:1 in pomace, whereas C16:0, C18:2, and C18:1 were prevalent in pruning (**Table 3**). Although the palmitic acid percentages are similar in pomace and pruning (21.98 vs 28.35), considering the total lipids content, C16:0 is 11 times higher in pomace than pruning. Palmitic acid seems to reduce *Pleurotus* growth (*23*).

The mixtures higher in pruning content had higher total phenols and sugars components than substrates richer in grape pomace. Total sugar of control comprised between 231.42 and 36.28 mg/g, being significantly higher in 100% pruning. Sugar content diminished significantly after SSF in the substrates with

Table 3. Percentage of Fatty Acids in Pomace and Pruning

fatty acid	100% pomace[a]	100% pruning[a]
C12:0	0.46 ± 0.05	0.74 ± 0.10
C14:0	0.84 ± 0.04	1.38 ± 0.02
C16:0	21.98 ± 1.30	28.35 ± 0.60
C16:1	0.95 ± 0.23	0.29 ± 0.02
C18:0	7.15 ± 0.82	4.33 ± 0.08
C18:1	17.53 ± 0.28	6.97 ± 0.22
C18:2	27.52 ± 0.42	17.78 ± 0.25
C18:3	0.84 ± 0.02	1.30 ± 0.01
C20:0	1.41 ± 0.10	1.79 ± 0.01
C20:1	0.12 ± 0.01	ND[b]
C22:0	0.63 ± 0.08	1.83 ± 0.05
C24:0	0.55 ± 0.04	0.75 ± 0.02

[a] All values are means ± SD of triplicate measurements. [b] ND, not detected.

higher pruning content (**Table 4**). On the contrary, the total sugar increased in the mixtures with higher pomace content after SSF.

Because opposite effects on growth and bioconversion of *Pleurotus* were observed on pruning and pomace, changes of phenol and sugars content were determined at the beginning of fermentation and during strain fructification. All strains diminished significantly in phenol content ($p < 0.05$). Strain CCMC H-041 reduced 57% in phenol content in pruning (**Table 5**). There were no observable differences among strains. Although grape pomace had a lower phenol content (21.71%) than pruning, its reduction after fermentation was not significant. The results could be related with fat and nitrogen contents in this residue.

The nitrogen content during growth of mycelia and lignin degradation is still a matter of discussion. Rajarathnam and Bano (*23*) showed that an increased concentration of nitrogen on culture media resulted in a progressive decrease of the yield of *Pleurotus flabellatus* mycelium, although the crude protein content of the mycelium increased. *Agaricus* has also been found to require a substrate of low nitrogen content during spawning in order to raise good crops. Contradictory reports on the role of nitrogen concentration in lignin degradation by white rot fungi have been published. Mostly, high nitrogen levels suppress the lignin removal from lignocellulosic materials (*30*). The present

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Table 4. Total Sugars of Viticulture Byproducts before and after Solid State Fermentation by *Pleurotus* Species

Pleurotus strain	mixture pruning/pomace	total sugars (mg/g)
control	1:0	231.4 ± 30.0^a
	2:1	124.9 ± 21.7^b
	1:1	$167.2 \pm 12.3^{a-c}$
	1:2	$121.2 \pm 6.8^{b-d}$
	0:1	36.3 ± 3.6^g
CCMC H-041	1:0	$107.2 \pm 22.4^{c-e}$
	2:1	$69.1 \pm 7.6^{e-g}$
	1:1	$110.4 \pm 4.9^{c-e}$
	1:2	$107.2 \pm 22.4^{c-e}$
	0:1	$75.71 \pm 8.8^{c-g}$
IE-115	1:0	125.7 ± 24.9^{bc}
	2:1	$73.2 \pm 11.2^{d-g}$
	1:1	$76.4 \pm 10.8^{c-g}$
	1:2	48.7 ± 12.1^g
	0:1	54.2 ± 5.3^{fg}
IE-8	1:0	$111.5 \pm 22.3^{c-e}$
	2:1	$80.9 \pm 1.9^{c-g}$
	1:1	$101.2 \pm 7.7^{c-f}$
	1:2	$100.9 \pm 6.4^{c-f}$
	0:1	$84.1 \pm 18.9^{c-g}$

[a] All values are means $\pm$ SD of triplicate measurements. Means in a column with different superscripts are significantly different ($p < 0.05$, Tukey).

Table 5. Total Phenols of Viticulture Byproducts before and after Solid State Fermentation by *Pleurotus* Species

Pleurotus strain	mixture pruning/pomace	total phenols[a] (μg/g)
control	1:0	47.0 ± 3.6^a
	0:1	21.7 ± 1.1^{bc}
CCMC H-041	1:0	20.3 ± 4.8^{bc}
	0:1	18.0 ± 1.1^{bc}
IE-115	1:0	22.0 ± 3.9^{bc}
	0:1	17.7 ± 1.5^c
IE-8	1:0	26.1 ± 4.8^b
	0:1	19.7 ± 1.9^{bc}

[a] All values are means $\pm$ SD of triplicate measurements. Means in a column with different superscripts are significantly different ($p < 0.05$, Tukey).

study shows both nitrogen and lignin content higher in pomace residues; however, changes in lignin evidently followed the pomace content of substrate mixtures but were not statistically different. The substrates with 100% pruning and 2:1 pruning/pomace mixture presented a C/N ratio (**Table 6**) near the optimum value reported for *Pleurotus* by Rajarathnam and Bano (*23*). Incubations on >50% prunings with CCMC H-041 resulted in numerical decreases in lignin content that cannot be accounted for by differences in N content.

Recently, Thomas et al. (*31*) showed a low BE in leaflets of coconut palm (38.2%) with nitrogen content 3 times higher than that in leaf stalk with a BE of 58.9. Several publications address the high nitrogen content in the medium and substrate related to low manganese-dependent peroxidase activity (*32, 33*). Manganese peroxidase (MnP) is essential for lignin degradation and occurs mostly in white rot fungi. MnP requires hydrogen peroxide (*34*) to depolymerize synthetic lignin in vitro and to peroxidate unsaturated lipids, causing the formation of lipoxy radical intermediates capable of oxidation of nonphenolic lignin compounds (*35*). In a separate unpublished study (data not shown), a low radial growth was found when nitrogen was added

Table 6. C/N Ratio of Viticulture Byproducts before and after Solid State Fermentation by *Pleurotus* Species

Pleurotus strain	mixture pruning/pomace	C/N ratio[a]
control	1:0	$88.52 \pm 5.99^{c-f}$
	2:1	62.84 ± 2.55^{ef}
	1:1	$264.38 \pm 88.56^{c-f}$
	1:2	46.77 ± 3.39^{ef}
	0:1	48.81 ± 7.98^{ef}
CCMC H-041	1:0	$138.17 \pm 10.66^{c-f}$
	2:1	292.61 ± 42.72^a
	1:1	46.96 ± 0.77^{ef}
	1:2	42.14 ± 0.71^f
	0:1	36.76 ± 1.52^f
IE-115	1:0	$157.55 \pm 64.20^{d-f}$
	2:1	$68.85 \pm 7.06^{d-f}$
	1:1	51.60 ± 4.42^{ef}
	1:2	213.67 ± 41.63^{ab}
	0:1	168.65 ± 42.15^{bc}
IE-8	1:0	$122.65 \pm 21.84^{b-f}$
	2:1	$64.70 \pm 5.09^{d-f}$
	1:1	57.28 ± 3.56^{ef}
	1:2	54.80 ± 21.62^{ef}
	0:1	43.37 ± 4.46^f

[a] All values are means $\pm$ SD of triplicate measurements. Means in a column with different superscripts are significantly different ($p < 0.05$, Tukey).

to reach 20 mM to malt extract agar in a culture of *P. ostreatus* strain, as was observed by Fu et al. (*32*).

Compositional Changes of Substrates. Results of composition of the different mixtures of pruning and pomace are shown in **Tables 2** and **7**. Moisture, protein, fat, and lignin contents were generally higher in mixtures with higher pomace proportion, whereas NDF, hemicellulose, and cellulose contents were higher with pruning content. Incubations of strain CCMC H-041 were drier than the control in all mixtures. Strains CCMC H-041 and IE-115 produced a pomace residue with a higher protein content, whereas strain IE-8 reduced it. Mixtures higher in prunings were not changed in protein content. Ash and fat contents were not affected by the incubations, including minerals in the cell wall (silica). Cell wall constituents increased slightly in residues with strain IE-115, especially due to lignin; on the other hand, strain CCMC H-041 showed an increase in cellulose content.

Incubations of 30 days or more of *P. ostreatus* on wheat straw (*36*) and on paddy straw (*37*) showed a decrease in moisture, fat, and all fiber fractions and an increase in protein and ash. Similar findings in moisture and protein contents were reported for fermentations with *Pleurotus sajor-caju* on corn stalks for up to 28 days (*38*). The fiber fractions obtained in this work are similar to those previously reported (*36, 39, 40*). Incubations with strain CCMC H-041 resulted in higher cellulose for all substrates and lower lignin, especially in mixtures containing pruning, showing a preference for different fiber fractions, even though cellulose and hemicellulose are scarce in pomace. Thus, strains of *Pleurotus* may act differently depending on the availability of fiber fractions of the original substrate and on dynamic changes in digestion that might occur as these fractions change during fungal growth.

Adamovic et al. (*36*) studied the biodegradation of wheat straw by *P. ostreatus* and its use in cattle feeding. After SSF, NDF decreased from 824 to 485 g kg^{-1} and ADF decreased from 561 to 412 g kg^{-1}. A similar tendency was found for hemicellulose and cellulose, whereas it was not pronounced for lignin. It was found that animals would not consume mixed

Table 7. Fiber Fractions of Intact Viticulture Byproducts (Control) and after Solid State Fermentation of *Pleurotus* Species

Pleurotus strain	mixture pruning/pomace	hemicellulose[a,b]	cellulose[a]	lignin[a]	silica[a]
control	1:0	17.88 ± 0.04[a]	8.93 ± 0.50[d−g]	49.22 ± 0.20[ef]	0.62 ± 0.03[a]
	2:1	12.11 ± 7.45[ab]	5.96 ± 0.41[f−j]	55.25 ± 0.91[de]	0.93 ± 0.13[a]
	1:1	12.44 ± 0.75[ab]	6.80 ± 0.68[d−i]	53.84 ± 0.38[de]	0.46 ± 0.35[a]
	1:2	4.29 ± 0.00[c−f]	3.72 ± 0.34[g−j]	61.75 ± 0.15[b−d]	1.33 ± 0.19[a]
	0:1	2.34 ± 0.34[d−f]	1.23 ± 0.14[j]	66.20 ± 0.00[ab]	1.74 ± 0.42[a]
CCMC H-041	1:0	11.19 ± 0.26[a−c]	37.77 ± 1.28[a]	29.68 ± 1.85[g]	1.18 ± 1.63[a]
	2:1	3.90 ± 1.29[d−f]	36.33 ± 0.16[a]	32.00 ± 0.84[g]	1.39 ± 0.60[a]
	1:1	−1.19 ± 0.91[f]	25.38 ± 3.97[b]	46.37 ± 4.91[f]	1.32 ± 0.07[a]
	1:2	0.68 ± 1.87[d−f]	18.86 ± 2.16[c]	49.68 ± 1.33[ef]	0.90 ± 0.21[a]
	0:1	−0.99 ± 0.90[ef]	10.84 ± 1.52[d−f]	60.58 ± 1.10[b−d]	1.31 ± 0.29[a]
IE-115	1:0	15.66 ± 1.76[a]	11.85 ± 0.88[d]	50.50 ± 0.59[ef]	0.48 ± 0.12[a]
	2:1	7.07 ± 4.12[b−d]	6.30 ± 1.41[f−j]	59.17 ± 0.66[cd]	0.79 ± 0.40[a]
	1:1	6.43 ± 0.57[b−e]	4.12 ± 0.00[g−j]	62.73 ± 0.56[bc]	1.65 ± 0.50[a]
	1:2	4.43 ± 0.76[c−f]	2.67 ± 0.70[h−j]	66.81 ± 2.29[ab]	1.29 ± 0.05[a]
	0:1	0.54 ± 0.63[d−f]	1.79 ± 0.28[ij]	70.43 ± 0.58[a]	1.91 ± 0.11[a]
IE-8	1:0	15.24 ± 1.76[a]	11.40 ± 0.31[de]	51.00 ± 0.72[ef]	0.61 ± 0.18[a]
	2:1	7.03 ± 0.05[b−d]	7.53 ± 0.55[d−h]	58.92 ± 2.58[cd]	1.06 ± 0.20[a]
	1:1	3.51 ± 3.21[d−f]	5.94 ± 0.33[f−j]	62.34 ± 0.17[bc]	0.94 ± 0.00[a]
	1:2	0.10 ± 1.42[ef]	4.55 ± 0.16[f−j]	66.24 ± 0.95[ab]	1.62 ± 0.63[a]
	0:1	−2.18 ± 1.74[f]	2.03 ± 0.01[ij]	71.97 ± 0.27[a]	1.56 ± 0.32[a]

[a] All values are means ± SD of triplicate measurements. Means in a column with different superscripts are significantly different ($p < 0.05$, Tukey). [b] Negative numbers indicate absence or undetectable contents of hemicellulose in incubated residues.

ration with >17% DM from compost. The utilization of dry matter for body weight gain had a tendency to decrease with the increased amount of compost in the diet.

Bisaria et al. (*39*) analyzed the bioconversion of agroresidues, rice straw, and wheat straw by *P. sajor-caju* in an attempt to increase its nutritional value for animal feed. Bioconversion of unsupplemented rice straw after 20 days of incubation showed a decrease of cellulose from 35.8 to 17.9% and a decrease of lignin from 17.2 to 9.5%. The in vitro dry matter digestibility (IVDMD) increased from 19.7 to 29.8%. A similar tendency was observed for wheat straw after SSF, and the IVDMD augmented from 27.2 to 36.8%. In the case of rice straw, supplementation with ammonium nitrate resulted in the maximum loss of organic matter (34.7%). For wheat straw, urea supplementation caused the highest loss of organic matter. As with rice straw, the nitrogen supplementation increased the degradation of cellulose and hemicellulose but decreased the degradation of lignin. The highest IVDMD values in supplemented rice and wheat straw after 20 days of SSF were 31.8 and 34.8%, respectively.

Karunanandaa and Varga (*40*) found that hemicellulose in rice leaf was selectively and extensively consumed when evaluated in SSF of rice straw by *P. sajor-caju*. Fungus improved the IVDMD of rice leaf, primarily because of increased digestion of cellulose. The improvement of rice straw quality was dependent on fungal species, botanical fractions, and preparation of substrate prior to fungal decay.

The present study establishes the potential use of biotransformation of winery byproducts through solid state fermentation by *Pleurotus* species to produce food of good quality for human consumption. Moreover, after mushrooms are harvested, it is possible to use the fermented viticulture wastes to feed ruminants. The nutritional value of these agroresidues might be increased by some strains of *Pleurotus*, whereas some antinutritional factors such as phenolic components are diminished.

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