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EFFECT OF *Yucca schiedigera* BAGASSE AS SUBSTRATE FOR OYSTER MUSHROOM ON CULTIVATION PARAMETERS AND FRUIT BODY QUALITY

EFECTO DEL BAGAZO DE *Yucca schiedigera* EN EL SUSTRADO DEL HONGO OSTRA SOBRE LOS PARÁMETROS DE CULTIVO Y LA CALIDAD DE LOS CUERPOS FRUCTIFERO

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Abstract

Yucca bagasse (YB) was evaluated as cultivation substrate for *Pleurotus ostreatus*. The effect of YB on physical and morphological properties and on chemical composition of fruit bodies, was studied with three strains. A factorial design was used to study the effect of strain and different proportions of yucca bagasse in the substrates. All strains consistently showed higher yields on wheat straw (WS) substrates and YB:WS 25:75 (average biological efficiency values of 143% and 115%, respectively). Pileus width and length was found to be influenced by both, strains and substrates ($p < 0.05$) but stipe length was significantly influenced only by strain. Both, whiteness (WI) and yellow indexes (YI) were significantly affected only by strain. Using PCA, an important relationship between textural parameters and chemical composition of fruit bodies was established in this study and that strain and substrate are important for both texture and chemical composition of fruit bodies.

Keywords: *Yucca schidigera*, *Pleurotus*, texture, color.

Resumen

En este trabajo se evaluó el bagazo de yuca (YB) como sustrato para el cultivo del hongo comestible Pleurotus ostreatus. Se utilizaron tres cepas y se determinó el efecto de YB sobre las propiedades físicas, morfológicas, de composición química y productividad de los cuerpos fructíferos. Se utilizó un diseño factorial para estudiar el efecto de la cepa y de diferentes proporciones de bagazo de yuca como sustrato. Todas las cepas mostraron sistemáticamente mayores rendimientos en los sustratos de paja de trigo (WS) y YB:WS 25:75 (valores promedio de eficiencia biológica de 143% y 115%, respectivamente). Se encontró que el ancho y la longitud del píleo están influenciados por las cepas y los sustratos ($p < 0.05$), pero la longitud del estípite solo por la cepa utilizada. Tanto el índice de blancura (WI) como amarillez (YI) se vieron afectados significativamente únicamente por el tipo de cepa. Usando PCA, en este estudio se estableció una relación importante entre los parámetros de textura y la composición química de los cuerpos fructíferos, indicando que estos parámetros son determinados tanto para la cepa como el sustrato utilizado.

Palabras clave: *Yucca schidigera*, *Pleurotus*, textura, color.

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1 Introduction

Yucca schiedigera is a plant whose species are found in north and central Mexico and the southeast of the United States in arid and semi-arid zones (Kawai *et al*., 2000; Vlcková *et al*., 2017). The leaf fibre is used to make rope, sandals and clothing. Its flowers and fruits are edible, the seeds are milled to make flour and its roots are used to make soup. This plant has been used for years to diminish pain and swelling, as well as to fight allergies and to strengthen the immune system (Oleszek, *et al*., 2001, Tenon *et al*., 2017).

The trunk is also edible. It is crushed, dried and milled to produce yucca powder and the crushed material is pressed to produce yucca juice. This is concentrated by evaporation and it is known as yucca extract (Cheeke, 2000). During the extraction process, three principal by-products are generated: bark, bagasse, and fine and coarse powder. Most of the manufacturing of the commercial products from *Y. schidigera* is carried out in Mexico.

Cellulose, hemicelluloses form a complex three-dimensional structure that is difficult to degrade. However, the lignocellulosic materials are an important source of carbohydrates with vast potentials that are currently underutilized (Akmar & Kennedy, 2001; Kunasundari, 2017). The *Y. schidigera* trunk waste produced by different Mexican industries is a lignocellulosic by-product available in large quantities but of no commercial value and it is either confined or donated to locals. Therefore, there is an opportunity to process it to make added value products. The lignocellulose compounds in these agro-industrial residues are important for edible mushroom cultivation.Edible mushrooms of the genus *Pleurotus* are present in a wide geographical distribution in most temperate regions of the world (Zervakis, *et al*., 2004; Gomes *et al*., 2016). Their cultivation is very popular since they can be produced on a large variety of agricultural residues and agro-industrial by-products (Melo de Carvalho, *et al*., 2010). An important requirement for further development of their cultivation is and increased in productivity through exploitation of novel cheap substrates in order to enhance mushroom yields and quality parameters of fruity bodies.

The aim of this work was to evaluate *Yucca schiedigera* as a substrate for production of oyster mushrooms (*Pleurotus* spp.).

2 Materials and methods

2.1 Fungal strains, spawn production and mushroom cultivation process

2.1.1 Biological material

Three Pleurotus ostreatus strains from the culture collection of the Cell Culture Laboratory (UPIBI-IPN) were used: CS (a commercial strain), UAP9 (University of Puebla (BUAP), Puebla, Mexico) and CxU, an hybrid obtained by cross breeding neohaplonts from CS and UAP9 strains. For spawn production, sterilized wheat grain was inoculated with malt extract agar (MAE) plates full grown with fresh mycelium, and then incubated in darkness at 28 ºC until grain spawn was completely invaded by mycelium (approximately 15 to 20 days).

2.2 Cultivation substrates

Wheat straw was obtained from a local feedstock trader, PROPECUA (Tlalnepantla, México) and *Yucca schiedigera* bagasse from a company located in Ciudad Serdán, Puebla, México. Dried yucca bagasse and wheat straw were chopped into pieces of 2 to 5 cm. Materials were soaked in water at room temperature for 24 h in raffia sacks, drained to eliminate excess water and then pasteurized with steam for 2 h. After cooling down, 1 kg of wet substrate (72% moisture) was packed into 40x30 cm polypropylene bags according to following formulations YB (yucca bagasse at 100%), WS (wheat straw at 100%), YB:WS at 75:25%, 50:50% and 25:75%. Substrates were then inoculated with spawn at a 5% w/w rate and mixed uniformly. Ten replicates per substrate and strain were used. Inoculated substrates were incubated in darkness at 20 to 28 ºC. Three days after spawning, bags were perforated (15-20 holes for bag) to allow gaseous exchange and were incubated for 15 days until substrate was completely invaded by mycelium. Once primordia formation was evident, substrates were freed from plastic covers and they were moved to a fruiting chamber. Conditions in the fruiting chamber were 40 to 70% relative humidity (RH), CO_2 levels less than 1200 ppm, 15 to 30 ºC temperature and indirect natural light for 8 to 12 h/day. Fruiting substrates were watered five times a day to maintain humidity.

Fruit bodies were cut at their base, approximately a week after primordia formation, before over

maturing occurred. Data from the first, second and third flushes were registered. Following parameters were determined to assess substrate quality: (a) duration of each stage of the mushroom crop cycle (incubation, primordial formation, and three flushes of mushroom production); (b) earliness, defined as the time elapsed between the day of inoculation and the day of primordia appearance; (c) yield (g fresh mushrooms/1 kg fresh substrates), biological efficiency, i.e. BE (g fresh mushrooms/100 g dry substrate), productivity rate (BE/total cultivation period in days), and average mushroom weight (MW), i.e.: total weight of fresh mushrooms harvested/total number of mushrooms harvested (Royse, 2002).

2.3 *Morphological and physical characteristics*

Morphology of fruit bodies was assessed by measuring color, texture and size, the latter by assessing pileus diameter and stipe length (both parameters with a Vernier calliper). Fruit bodies were classified in relation to pileus diameter according to Salmones *et al*. (1997): G1 (less than 5 cm), G2 (5 to 9.9 cm), G3 (10 to 14.9 cm) and G4 (15 cm upwards) and stipe size was classified according to Cruz (2009), i.e. group "a" (less than 5 cm) and group "b" (5 to 9.9 cm). The color of a sample from the first crop was measured using a MINOLTA colorimeter, model CR-300 with a DP-301 data processor, considering the CIELab parameters L^*, a^* and b^*. These analyses were made by triplicate on the front face of the pileus (smooth side). Later, the difference of ΔE^* color and Chroma were calculated (Eq. 1):

$$\Delta E* = \sqrt{(L_0^* - L^*)^2 + (a_0^* - a^*)^2 + (b_0^* - b^*)^2} \quad (1)$$

Subscript "0" refers to the color reading of straw. Wheat straw was used as the reference and a larger ΔE^* denotes larger color change from the reference material. Whiteness index (WI), Yellowness index (YI) and Browning index (BI) (which represents the purity of brown colour and is considered an important parameter associated with browning) were calculated, see Eq. (2)-(5), (Maskan, 2001; Bozkurt & Bayram, 2006; Rhim *et al*., 1999):

$$WI = 100 - \left[(100 - L^*)^2 + a^{*2} + b^{*2}\right]^{1/2} \quad (2)$$

$$YI = \frac{142.86 * b^*}{L^*} \quad (3)$$

$$BI = 100\left(\frac{x - 0.31}{0.17}\right) \quad (4)$$

Where:

$$YI = \frac{a^* + 1.75L^*}{5.645L^* + a^* - 3.012b^*} \quad (5)$$

Texture Profile Analysis (TPA) was performed with a sample from the first crop with a LLOYD TA 500 model texturometer, with a 9.8067 N load cell and a 1.27 cm acrylic cylinder at a speed of 1.7 mm/s at room temperature. This test was applied on the front face of the pileus (smooth side) to obtain the values for hardness, cohesiveness, springiness, chewiness, adhesiveness and gumminess.

2.4 *Chemical analysis*

Mushrooms from the first crop were shredded and dried at 35 °C for 3 days, then milled to perform proximal analysis using standard methods. Moisture, ash, crude fibre and crude fat were determined according to the Association of Official Analytical Chemists methods (AOAC, 1997). Total nitrogen was evaluated with the Micro-Kjeldahl method, crude protein was calculated from total nitrogen content by employing the converting factor 4.38 (Crisan & Sands, 1978; Barros *et al*., 2008). Total Carbohydrates (Nitrogen-free extracts) were calculated by the formula: 100 - (moisture + protein + fat + ash contents), and gross energy (kcal 100 g^{-1} f.w.) was estimated according to the equation: energy = 4 × (g protein + g carbohydrate) + 9 × (g fat) (Manzi *et al*., 2004).

2.5 *Statistical treatment of experimental data*

Productivity experiments were performed using ten replicas (for each substrate); morphological, textural and chemical analyses were carried out by triplicate. Results were expressed as mean values ± standard error of mean. Levene test was used for determination of homogeneity of variance; in the absence of it, the Welch test was used with Games-Howell post hoc test. With homoscedasticity the Analysis of variance multivariable was performed followed by post hoc Duncan test at 5% level of significance, when differences were significant in interaction, the Bonferroni test was performed. Spearman correlation coefficient and Principal Component Analysis (PCA) were employed to assess relationships (at significance

levels of 0.05) between variables such as substrate composition versus cultivation, morphological, textual and chemical composition parameters. The SPSS software (version 18) was used for all analysis.

3 Results and discussion

3.1 Assessment of cultivation parameters

Table 1 shows mushroom production parameters for *Pleurotus ostreatus* strains (CS, UAP9 and CxU) growing on five different cultivation substrates (100% yucca bagasse (YB), 100% wheat straw (WS), YB:WS 75:25, YB:WS 50:50 and YB:WS 25:75). Analysis of variance for two factors indicated significant differences between strains and substrates for all production parameters although there were not statistical differences for the strain and substrate interaction. Though strain UAP9 produced the highest BE on WS 100% (154.33%), all 3 strains consistently showed higher yields on substrates WS 100% and YB:WS 25:75 (EB values from 107.5 up to 157.3) (Table 1). The values produced in this study are similar to those obtained by other researchers as well, Philippoussis (2009) indicated that wheat straw showed higher biological efficiency (85.4-90.9%) than soft woods waste (48.1-76%) used as substrate for cultivation of *P. ostreatus* and *P. pulmonarius*. Perhaps this is because straw has a C/N ratio 6-8 times lower than soft wood waste (Philippoussis, 2009). Moreover, it has been reported that the nutrient composition of the substrate is one of the factors limiting colonization and mushrooms cultivation yield (Philippoussis, 2009).

Regarding average mushroom weight (MW), significant differences were obtained for strain and substrate, larger MW were produced on substrate YB (7-12 g) and WS (8-10 g) and the strain producing larger fruit bodies was CS (8.8-12.5 g). Royse (2002) reported values of 7.2-19.6 g for MW for fruit bodies of *Pleurotus cornucopia* grown on a mixture of straw with cotton. In relation to earliness (table 1), the lowest weighted average value was obtained on substrate YB 100%, 23.6 days, while on other substrates; they were 25 to 27 days (data not shown). Strain UAP9 showed the lowest values on all substrates (20.3-25.3 days). Curvetto *et al*. (2002) reported primordia initiation for *P. ostreatus* strains between day 24 and 28, while fruit body formation on coir pith substrate, occurred after 24 days for *P. florida* and after 30 days for *P. flabellatus* (Chanakya *et al*., 2015). Commercial production on 100% yucca bagasse has therefore a good potential. Earliness is indicative of the invasion rate of substrate by a strain and it is dependent both on the metabolic capabilities resulting of the genotype of each strain and of the specific characteristics of substrate. Figure 1 shows productivity rates for *P. ostreatus* strains (CS, UAP9 and CxU) growing on the five cultivation substrates (YB 100%, WS 100%, YB:WS 75:25, YB:WS 50:50 and YB:WS 25:75).

Table 1. Mushroom production parameters for *Pleurotus ostreatus* strains (CS, UAP9 and CxU) growing on five cultivation substrates (100% yucca bagasse (YB), 100% wheat straw (WS), YB:WS 75:25, YB:WS 50:50, and YB:WS 25:75). Values are expressed as means ± standard errors of means, n = 10. Different letters indicate statistically significant differences (Duncan's test. $p < 0.05$) for comparisons of treatment means between different strains (capital letters) and between different substrates (lowercase letters).

Substrates	Strain	Earliness	Total cultivation	Yield(g)			BE		Mushroom
		(days)	1st flush	2nd flush	3rd flush (g)	Total	(%)	weight (g)	
YB 100%	CS	23.1 ± 1.4^{Aa}	69.6 ± 1.3^{Aa}	75.4 ± 9.1^{Aa}	55.5 ± 5.2^{Ba}	40.3 ± 3.7^{Ba}	$171.4\pm17.^{Aa}$	54.7 ± 5.6^{Aa}	12.5 ± 0.8^{Bb}
	UAP9	20.3 ± 1.6^{Ba}	66.0 ± 1.7^{Aa}	164.9 ± 15.1^{Ba}	57.1 ± 4.8^{Aa}	29.5 ± 2.6^{Aa}	251.6 ± 16.4^{Ba}	85.7 ± 5.4^{Ba}	11.3 ± 1.6^{Ab}
	CxU	27.4 ± 1.4^{Ca}	72.8 ± 1.2^{Ba}	128.8 ± 8.8^{Aa}	48.7 ± 4.9^{Aa}	25.1 ± 3.3^{Aa}	202.7 ± 12.5^{Aa}	69.3 ± 4.2^{Aa}	7.8 ± 0.4^{Ab}
YB:WS 75:25%	CS	26.6 ± 0.6^{Ab}	75.0 ± 1.5^{Ac}	112.1 ± 3.5^{Aa}	67.2 ± 3.1^{Bb}	31.7 ± 2.5^{Ba}	211.1 ± 6.3^{Aa}	74.8 ± 2.5^{Aa}	8.8 ± 1.0^{Ba}
	UAP9	25.3 ± 0.9^{Bb}	77.1 ± 1.8^{Ac}	133.6 ± 12.8^{Ba}	53.8 ± 3.4^{Ab}	23.7 ± 2.9^{Aa}	211.2 ± 14.3^{Ba}	78.1 ± 5.0^{Ba}	6.7 ± 0.5^{Aa}
	CxU	29.2 ± 0.6^{Cb}	80.5 ± 2.0^{Bc}	125.7 ± 7.9^{Aa}	61.3 ± 5.4^{Ab}	28.8 ± 2.8^{Aa}	215.9 ± 7.9^{Aa}	77.7 ± 2.1^{Aa}	9.4 ± 1.1^{Aa}
YB:WS 50:50%	CS	24.7 ± 0.9^{Ab}	73.2 ± 1.1^{Ab}	131.6 ± 6.1^{Ab}	70.0 ± 3.3^{Bb}	37.5 ± 2.9^{Bb}	239.2 ± 10.4^{Ab}	95.2 ± 4.3^{Ab}	9.2 ± 0.5^{Ba}
	UAP9	22.3 ± 0.3^{Bb}	72.4 ± 1.7^{Ab}	168.6 ± 7.2^{Bb}	63.0 ± 3.8^{Ab}	27.5 ± 2.9^{Ab}	259.2 ± 9.4^{Bb}	102.8 ± 3.4^{Bb}	6.6 ± 0.4^{Aa}
	CxU	28.7 ± 0.6^{Cb}	76.2 ± 1.1^{Bb}	146.5 ± 10.8^{Ab}	58.9 ± 5.9^{Ab}	30.2 ± 3.5^{Ab}	235.7 ± 16.5^{Ab}	92.3 ± 5.7^{Ab}	8.0 ± 0.9^{Aa}
YB:WS 25:75%	CS	24.6 ± 0.5^{Ab}	73.4 ± 0.7^{Ab}	145.5 ± 11.0^{Ab}	53.2 ± 4.0^{Ba}	36.3 ± 3.1^{Bb}	235.2 ± 14.3^{Ab}	107.5 ± 5.9^{Ac}	11.2 ± 0.6^{Ba}
	UAP9	21.5 ± 0.3^{Bb}	71.5 ± 1.7^{Ab}	165.8 ± 7.5^{Bb}	67.4 ± 5.9^{Aa}	34.5 ± 3.0^{Ab}	267.7 ± 8.7^{Bb}	125.6 ± 3.5^{Bc}	6.8 ± 0.2^{Aa}
	CxU	28.9 ± 1.3^{Cb}	74.2 ± 0.9^{Bb}	153.7 ± 6.8^{Ab}	57.5 ± 2.6^{Aa}	29.7 ± 2.5^{Ab}	241.1 ± 6.7^{Ab}	113.7 ± 3.0^{Ac}	7.9 ± 0.7^{Aa}
WS 100%	CS	23.1 ± 0.9^{Ab}	68.4 ± 1.6^{Aa}	130.8 ± 4.8^{Ab}	81.5 ± 3.9^{Bb}	43.7 ± 3.3^{B}	256.0 ± 4.8^{Ab}	137.1 ± 2.5^{Ad}	10.8 ± 1.1^{Bb}
	UAP9	22.8 ± 1.4^{Bb}	67.6 ± 2.4^{Aa}	175.0 ± 11.4^{Bb}	68.2 ± 4.1^{Ab}	30.9 ± 3.9^{Ab}	274.1 ± 12.1^{Bb}	154.3 ± 7.6^{Bd}	8.5 ± 0.7^{Ab}
	CxU	29.5 ± 0.8^{Cb}	74.4 ± 1.3^{Ba}	140.5 ± 5.8^{Ab}	61.0 ± 3.9^{Ab}	32.7 ± 1.6^{Ab}	234.4 ± 7.72^{Ab}	133.8 ± 4.8^{Ad}	8.6 ± 0.8^{Ab}

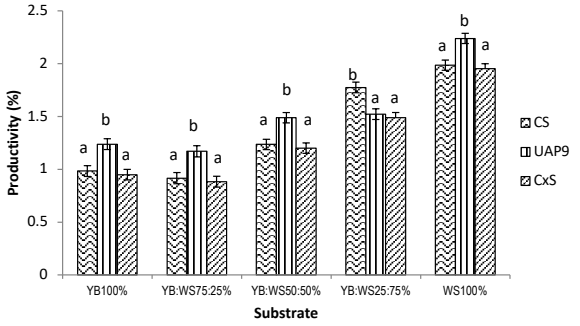

Fig. 1. Productivity (ratio of BE over total cropping period) for *Pleurotus ostreatus* strains (CS, UAP9 and CxU) growing on five cultivation substrates (100% yucca bagasse (YB), 100% wheat straw (WS), YB:WS 75:25, YB:WS 50:50 and YB:WS 25:75). Values are expressed as means ± standard error of means, n = 10. Different letters indicate statistically significant differences (Duncan's t-test, $p < 0.05$) for comparisons of treatment means between different strains (lowercase letters).

This parameter is important because the time spent for mushroom production of fruit bodies is considered. WS substrate clearly presented the best productivity rate (1.8-2.3%) in comparison to a productivity rate of 1.0% reported for *P. ostreatus* cultivated in wheat straw by Koutrotsios *et al*. (2014). The most productive strain was UAP9 (1.3-2.3%) on all substrates used in this study. However, for YB:WS 25:75% substrate was obtained a productivity rate of 1.4 to-1.7% with the BE values of 107.5 to 125.6%. Furthermore, Ruiz-Rodríguez *et al*. (2010) obtained with various *P. ostreatus* strains, a productivity rate of 2.5% and BE of 80% on a substrate of olive mill waste and wheat straw (50:50%). Therefore, commercial production on YB:WS 25:75% substrate has also a good potential.

3.2 Morphological and physical characteristics

Once the ripe fruiting bodies from all the strains were harvested, the phenotypic characteristics and color of the first crop were determined (see table 2). Pileus width and length were found to be influenced by both, strains and substrates ($p < 0.05$) but stipe length was significantly influenced only by strain. The majority of the mushrooms of the three strains were classified within groups G1 (less than 5cm) and G2 (5cm to 9.9cm), in accordance with data reported for various *Pleurotus* species grown on different substrates (Salmones *et al*., 1997; Pérez & Mata, 2005; Gaitán-Hernández & Salmones, 2008). Remarkably, strain CS gave fruit bodies of larger size (6.6 to 8.2 cm length and 5.7 to 7.2 cm width) than the other two strains.

Table 2. Morphologic parameters and color for *Pleurotus osteratus* strains (CS, UAP9 and CxU) growing on five cultivation substrates (100% yucca bagasse (YB), 100% wheat straw (WS), YB:WS 75:25, YB:WS 50:50 and YB:WS 25:75). Values are expressed as means ± standard errors of means, n = 3. Different letters indicate statistically significant differences (Duncan's t-test, $p < 0.05$) for comparisons of treatment means between different strains (capital letters) and between different substrates (lowercase letters).

Substrates	Strain	Pileus		Stipe	ΔE^*	Browning	Whiteness	Yellowness
		Length (cm)	width (cm)	cm		Index (BI)	Index1	Index (YI)
YB100%	CS	8.2 ± 0.9^{Bb}	7.2 ± 0.8^{Bb}	4.5 ± 0.7^{Ba}	4.7 ± 1.7^{Ab}	29.5 ± 1.2^{Aa}	46.3 ± 1.7^{Ba}	29.3 ± 1.2^{Aa}
	UAP9	6.6 ± 0.5^{Ab}	5.7 ± 0.3^{Ab}	3.0 ± 0.1^{Aa}	2.7 ± 0.7^{Ab}	35.1 ± 1.6^{Aa}	47.2 ± 0.2^{Ca}	35.8 ± 2.1^{Aa}
	CxU	5.9 ± 0.4^{Ab}	5.0 ± 0.3^{Ab}	2.6 ± 0.0^{Aa}	6.3 ± 0.7^{Bb}	32.3 ± 2.7^{Aa}	49.7 ± 1.3^{Aa}	33.2 ± 3.0^{Ba}
YB:WS75:25%	CS	7.5 ± 0.8^{Ba}	6.8 ± 0.8^{Ba}	4.2 ± 0.7^{Ba}	1.4 ± 0.5^{Ab}	24.4 ± 0.8^{Aa}	51.9 ± 0.6^{Ba}	26.4 ± 0.7^{Aa}
	UAP9	4.3 ± 0.3^{Aa}	4.1 ± 0.2^{Aa}	2.5 ± 0.1^{Aa}	2.2 ± 0.9^{Ab}	29.1 ± 2.4^{Aa}	46.2 ± 1.7^{Ca}	29.0 ± 1.9^{Aa}
	CxU	6.0 ± 0.6^{Ba}	5.4 ± 0.4^{Aa}	2.9 ± 0.1^{Aa}	5.2 ± 0.4^{Bb}	37.7 ± 0.1^{Aa}	49.1 ± 0.3^{Aa}	39.3 ± 0.1^{Ba}
YB:WS50:50%	CS	6.9 ± 0.7^{Ba}	6.2 ± 0.5^{Ba}	3.8 ± 0.4^{Ba}	4.9 ± 0.4^{Ab}	31.6 ± 1.3^{Aa}	45.9 ± 0.4^{Ba}	31.5 ± 1.6^{Aa}
	UAP9	4.4 ± 0.1^{Aa}	4.2 ± 0.1^{Aa}	2.6 ± 0.1^{Aa}	3.2 ± 0.8^{Ab}	35.5 ± 2.8^{Aa}	44.8 ± 1.7^{Ca}	34.0 ± 1.9^{Aa}
	CxU	5.0 ± 0.3^{Aa}	4.7 ± 0.3^{Aa}	2.5 ± 0.1^{Aa}	5.1 ± 2.0^{Bb}	32.9 ± 4.7^{Aa}	51.2 ± 3.1^{Aa}	35.4 ± 4.5^{Ba}
YB:WS25:75%	CS	6.7 ± 0.31^{Ba}	5.9 ± 0.1^{Ba}	3.5 ± 0.1^{Ba}	3.4 ± 1.0^{Ab}	34.7 ± 2.9^{Aa}	47.5 ± 0.9^{Ba}	35.2 ± 2.5^{Aa}
	UAP9	4.5 ± 0.1^{Aa}	4.1 ± 0.0^{Aa}	2.7 ± 0.1^{Aa}	1.4 ± 0.4^{Ab}	29.4 ± 0.2^{Aa}	45.3 ± 0.6^{Ca}	30.1 ± 0.4^{Aa}
	CxU	5.3 ± 0.4^{Aa}	4.9 ± 0.3^{Aa}	2.7 ± 0.1^{Aa}	6.0 ± 1.7^{Bb}	26.1 ± 1.7^{Aa}	52.0 ± 2.1^{Aa}	29.4 ± 2.0^{Ba}
WS100%	CS	6.5 ± 0.6^{Ba}	5.7 ± 0.5^{Bb}	3.5 ± 0.4^{Ba}	0.0 ± 0.0^{Aa}	26.8 ± 1.2^{Aa}	50.7 ± 0.4^{Ba}	28.4 ± 1.2^{Aa}
	UAP9	5.3 ± 0.2^{Aa}	4.8 ± 0.2^{Ab}	2.8 ± 0.1^{Aa}	0.0 ± 0.0^{Aa}	29.0 ± 1.4^{Aa}	46.7 ± 0.7^{Ca}	29.5 ± 1.4^{Aa}
	CxU	5.8 ± 0.4^{Aa}	5.2 ± 0.3^{Ab}	3.2 ± 0.2^{Aa}	0.0 ± 0.0^{Aa}	35.1 ± 4.8^{Aa}	53.7 ± 1.8^{Aa}	37.3 ± 4.5^{Ba}

This is important because pileus size increases market value. However, hybrid CxU was considered the best strain due to the large pileus/stipe ratio, 1.7-2.2, a larger value than those of the two parental strains, UAP9 and CS. Stipe length is an important parameter for mushrooms producers because too long stipes have to be cut and rejected. Additionally, mushrooms with large pileus and short or no stipe are preferred because of stipe fibrosity and unpleasant flavour (Kamat, et al, 2010; Lechner & Albertó, 2011).

Samples from the first crop were taken in order to measure color. A MINOLTA CR 300 colorimeter was used to evaluate the frontal part of the pileus. Strain and substrate were found to influence the value of ΔE^* ($P < 0.05$), while for browning index (BI) no significant differences were found for strain and substrate. However, both whiteness (WI) and yellowness indexes (YI) were significantly affected only by strain (see Table 2).

Concerning L^* (46.18 to 56.59), a^* (2.53 to 5.49) and b^* (9.55 to 14.88) values (not shown), some differences were observed in regards to values obtained by Ruiz-Rodríguez *et al*. (2010), who reported L^* values from 58 to 77, a^* values from 0.7 to 3.1 and b^* values between 12.5 and 13.5 for seven strains of *Pleurotus* spp. grown on straw. These dissimilarities could be explained because strains were different.

The CIELab color difference (ΔE^*), numerically quantifies the difference in color perception by the human eye between two samples. In this specific case the CIE2000 formula was used, comparing the values of the fruit bodies to that of straw as reference. This parameter was influenced by substrate, and fruit bodies cropped from substrate WS 100% showed significant lower values than fruit bodies from other substrates. An influence of strains on this parameter was also observed, ΔE^* values of fruit bodies from hybrid CxU (ΔE^* = 5.1) were significantly higher than those from strains CS and UAP9 (ΔE^* = 1.4) (Table 2). This parameter (ΔE^*) indicates the influence on the color of fruit bodies as a result of the nutrients present in the substrate and of the genetics of the strain. Apparently, in opposition to it, browning index (BI) was not influenced neither by strain nor by substrate. Nevertheless, browning is related to enzymatic and no enzymatic processes in fruit bodies taking place during ripening or in the post-harvest steps, i.e. storage, drying, freezing (Maskan, 2001; Mohapatra *et al*., 2010). However, in this study, measurements were performed with fresh cut fruit bodies. Whiteness (WI) and yellowness (YI) indexes were different in the hybrid strain compared to the parental strains.

Table 3. Proximate analysis results for *Pleurotus ostreatus* strains (CS, UAP9 and CxU) growing on five cultivation substrates (100% yucca bagasse (YB), 100% wheat straw (WS), YB:WS 75:25, YB:WS 50:50 and YB:WS 25:75). Values (% d.w., except of gross energy: kcal 100 g^{-1} d.w.) are expressed as means ± standard errors of means, n = 3. Different letters indicate statistically significant differences (Duncan's test, $p < 0.05$) for comparisons of treatment means between different strains (capital letters) and between different substrates (lowercase letters).

Substrates	Strain	Humidity	Crude protein	Crude fat	Crude fibre	Total carbohydrates	Ash	Gross energy
YB100%	CS	94.2 ± 0.0^{Bb}	25.2 ± 0.0^{Ac}	1.8 ± 0.1^{Ca}	17.5 ± 0.2^{Bb}	64.8 ± 0.2^{Cc}	8.0 ± 0.0^{Ba}	376.7 ± 0.6^{Ca}
	UAP9	92.1 ± 0.2^{Ab}	25.2 ± 0.0^{Cc}	1.4 ± 0.0^{Aa}	16.5 ± 0.1^{Ab}	64.6 ± 0.0^{Ac}	8.6 ± 0.0^{Ba}	372.6 ± 0.4^{Ab}
	CxU	92.1 ± 0.1^{Bb}	22.6 ± 0.6^{Bc}	2.9 ± 0.1^{Ba}	16.2 ± 0.1^{Ab}	68.0 ± 0.5^{Bc}	6.4 ± 0.1^{Aa}	388.6 ± 0.6^{Bb}
YB:WS75:25%	CS	91.1 ± 0.0^{Ba}	18.5 ± 0.0^{Aa}	1.9 ± 0.1^{Ca}	15.7 ± 0.7^{Ba}	70.9 ± 0.1^{Cd}	8.5 ± 0.0^{Bb}	375.5 ± 0.2^{Ca}
	UAP9	91.9 ± 0.3^{Aa}	25.2 ± 0.0^{Ca}	2.1 ± 0.1^{Aa}	16.4 ± 0.1^{Aa}	64.7 ± 0.0^{Ad}	7.7 ± 0.0^{Bb}	379.7 ± 0.9^{Aa}
	CxU	93.2 ± 0.4^{Ba}	23.6 ± 0.4^{Ba}	2.1 ± 0.1^{Ba}	16.0 ± 0.3^{Aa}	66.1 ± 0.5^{Bd}	8.0 ± 0.0^{Ab}	378.4 ± 0.8^{Ba}
YB:WS50:50%	CS	93.1 ± 0.3^{Bd}	21.9 ± 0.0^{Ac}	3.3 ± 0.1^{Cb}	17.4 ± 0.1^{Bb}	66.4 ± 0.1^{Cb}	8.3 ± 0.1^{Bc}	383.4 ± 1.3^{Ca}
	UAP9	93.4 ± 0.2^{Ad}	28.6 ± 0.0^{Cc}	1.5 ± 0.0^{Ab}	16.5 ± 0.1^{Ab}	60.9 ± 0.0^{Ab}	8.8 ± 0.0^{Bc}	372.0 ± 0.1^{Aa}
	CxU	93.2 ± 0.1^{Bd}	22.3 ± 0.4^{Bc}	2.7 ± 0.2^{Bb}	16.2 ± 0.0^{Ab}	66.1 ± 0.2^{Bb}	8.7 ± 0.0^{Ac}	378.9 ± 1.6^{Ba}
YB:WS25:75%	CS	92.6 ± 0.2^{Bb}	21.9 ± 0.0^{Ab}	3.0 ± 0.4^{Ca}	16.7 ± 0.5^{Ba}	66.9 ± 0.5^{Cd}	8.0 ± 0.0^{Ba}	382.9 ± 2.1^{Cb}
	UAP9	92.3 ± 0.2^{Ab}	25.2 ± 0.0^{Cb}	1.5 ± 0.0^{Aa}	16.3 ± 0.3^{Aa}	66.1 ± 0.0^{Ad}	7.0 ± 0.0^{Ba}	379.3 ± 0.7^{Ab}
	SAUA	93.1 ± 0.1^{Bb}	23.2 ± 0.3^{Bb}	1.7 ± 0.1^{Ba}	16.4 ± 0.2^{Aa}	67.3 ± 0.2^{Bd}	7.7 ± 0.1^{Aa}	377.7 ± 1.3^{Bb}
WS100%	CS CS	93.6 ± 0.2^{Bc}	25.2 ± 0.0^{Ad}	3.3 ± 0.0^{Ca}	17.2 ± 0.1^{Ba}	63.5 ± 0.0^{Ca}	7.8 ± 0.0^{Bb}	385.2 ± 0.2^{Ca}
	UAP9	92.9 ± 0.2^{Ac}	25.2 ± 0.0^{Bd}	1.5 ± 0.0^{Aa}	16.4 ± 0.1^{Aa}	65.0 ± 0.0^{Aa}	8.1 ± 0.0^{Bb}	374.8 ± 0.2^{Aa}
	CxU	92.8 ± 0.1^{Bc}	29.0 ± 0.0^{Cd}	1.5 ± 0.0^{Ba}	15.4 ± 0.0^{Aa}	60.9 ± 0.1^{Ba}	8.4 ± 0.1^{Ab}	373.5 ± 0.4^{Ba}

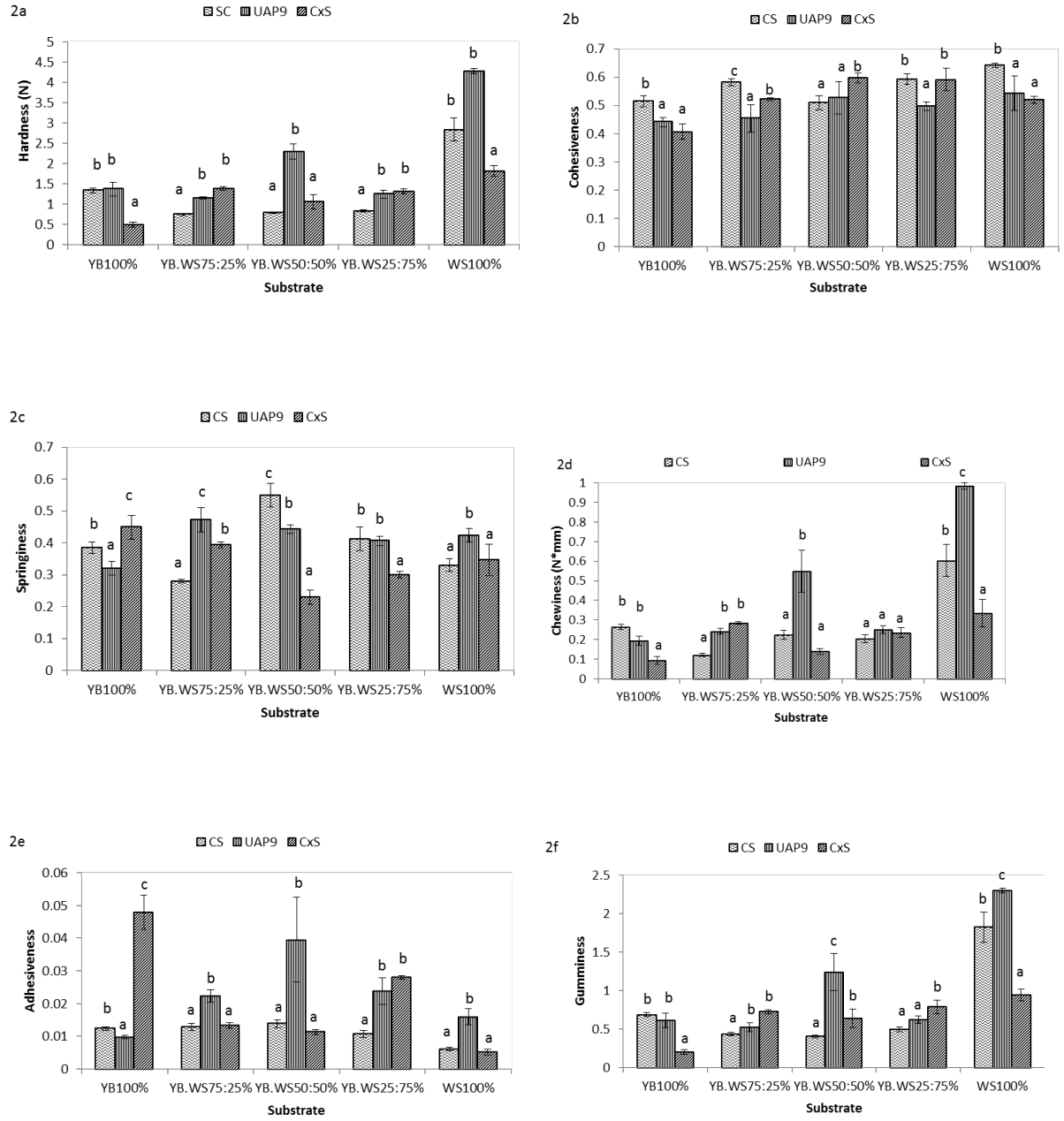

Fig. 2. Texture profile: 2a Hardness, 2b Cohesiveness, 2c Springiness, 2d Chewiness, 2d Adhesiveness and 2e Gumminess for Pleurotus ostreatus strains (CS, UAP9 and CxU) growing on five cultivation substrates (100% yucca bagasse (YB), 100% wheat straw (WS), YB:WS 75:25, YB:WS 50:50, and YB:WS 25:75). Values are expressed as means ± standard error of means, n = 6. Different letters indicate statistically significant differences (Duncan's t-test, $p < 0.05$) among substrates for each strain examined.

Several researchers have proposed that these color indexes allow a direct correlation with the visual appearance of a food and can be used in studies of maturation, preservation or storage (Carreño & Martínez, 1995; Pathare *et al*., 2013). In accordance to a previous report by Liu *et al*. (2005) indicating that color of fruit bodies was not influenced by the type of substrate, in this study both, WI and YI indexes were not affected by substrate. Both indexes are important parameters to detect differences in the color of fruit bodies, an important attribute in mushrooms cultivation.

3.3 Texture characteristics

Six parameters of texture profile analysis (TPA) were measured, i.e. hardness, cohesiveness, springiness, adhesiveness, gumminess and chewiness. No interaction was found between strain and substrate with texture parameters, however significant differences for these texture parameters were observed on the different substrates for the three strains (Figure 2). Hardness is the parameter most commonly used to determine the quality and freshness of edible mushrooms (Zivanovic *et al*., 2000). In this study, hardness values in the range of 0.49 to 4.27 were obtained, in agreement with the value of 0.46 N reported by Kotwaliwale *et al*. (2007) for *Pleurotus* spp. Addition of yucca bagasse to wheat straw resulted with all 3 strains, in fruit bodies with lower hardness, similarly to the negative effect on hardness when olive milling residues were added to the wheat straw as reported by Ruiz-Rodríguez |it et al. (2010).

In addition to higher hardness values, fruit bodies produced on 100% substrate showed also higher values for chewiness and gumminess. Cohesiveness values were in the range of 0.40-1.59, for springiness 0.28 to 0.55 and for chewiness 0.09 to 0.60 N mm, in accordance with values reported by Kotwaliwale *et al*. (2007) for cohesiveness, springiness and chewiness (0.53, 0.63 and 0.16 N mm, respectively) for fruit bodies of *Pleurotus* spp. Regarding adhesiveness, strain CS consistently showed the lower values of all 3 strains.

3.4 Chemical characteristics

The chemical composition of the fruit bodies produced on the various substrates is presented in table 3. Statistical analysis showed significant differences in humidity of fruit bodies for both strains and substrates. Mushrooms from strain UAP9 showed a significantly lower humidity content. Mushroom moisture was affected by composition of substrates, the highest value was found in mushrooms from YB:WS 50:50 substrate, while those from YB:WS 75:25 showed the lowest value. Ahmed *et al*. (2009) found significant differences in the moisture of fruit bodies growing on different agro-wastes like soybean straw, paddy straw, wheat straw and their combination in 1:1 proportion and Khan *et al*. (2013) reported similar findings on sawdust substrates of different woods. The values reported in the literature are close to those obtained in this study, namely 91.1 to 94.27%.

Crude fat values obtained in this study (1.4-3.3%) were similar to those reported by Forero *et al*. (2008), who found low levels of total lipids (ranging from 1.41-2.85%) in *P. ostreatus* grown on chili residues (*Capsicum* spp) with husk and King grass and by Ingale & Ramteke (2010) for *P. sajor-caju*, *P. florida* and *P. eous* mushrooms growing on soy straw (1.2% to 1.9%). Similarly, Akyüz & Kirbağ (2010) found that in *Pleurotus* sp. the fat content varies from 0.9 to 1.3% for wild mushrooms, and from 0.5 to 1.0% for cultivated mushrooms. In this study, fruit bodies from substrate YB:WS 50:50 showed a significantly higher crude fat content than fruit bodies from the other substrates, so lipids content was affected by the type of substrate in accordance to previously stated by Liu *et al*. (2005).

Furthermore, crude fat content was found to be affected also by strain, i.e. parental strain CS showed the highest value while the other parental strain UAP9 showed the lowest value and the hybrid strain, CxU, was in between.

Crude fibre values in the range of 10.9 up to 12.0% have been previously reported for fruiting bodies of *P. sajor-caju*, *P. florida* and *P. eous* grown on soy straw (Ingale & Ramteke, 2010). Data obtained in this study (15.4-17.5%) were higher than the reported values and significant differences were found depending of the strains and substrates.

Fruit bodies from parental strain CS showed significantly higher crude fibre content while all 3 strains produced fruit bodies with higher crude fibre content on two substrates, yucca bagasse 100% and YB:WS 50:50. Such high fibre content in the mushrooms produced in this study are advantageous since it promotes an efficient intestinal regulation helping the body to digest and eliminate undigested food (Silva *et al*., 2002).

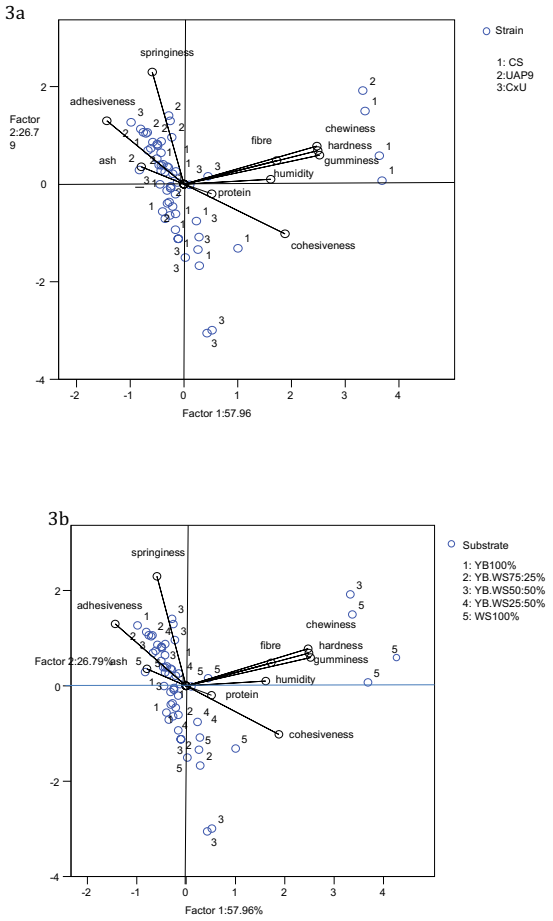

Fig. 3. Dimensional graphics obtained of Principal Component Analysis (PCA) for texture parameters and chemical composition of the fruit bodies of Pleurotus ostreatus strains (3a), grown on five cultivation substrates (100% yucca bagasse (YB), 100% wheat straw (WS), YB:WS 75:25, YB:WS 50:50 and YB:WS 25:75) (3b).

Raw protein in fruit bodies was influenced by substrate composition, the highest protein content was obtained on WS100% substrate, i.e. 26.0%, while YB:WS 50:50 substrate was in a second place with 28.6% protein content.

Strain also influenced protein content, the hybrid strain CxU produced the significantly highest value, 29.0%, followed by strain UAP9 (28.6%). Interestingly, the content of carbohydrates in fruit bodies were inversely related to the values obtained for raw protein content, i.e. fruit bodies with the lowest carbohydrates content were cropped from WS100% substrate and the highest were from 75:25 substrates.

Principal component analysis (PCA) is a dimensional projection method and data reduction, in which there is an association between variables in order to verify the degree of participation of each of them (Heenan *et al*., 2008). The parameters of texture and chemical composition were correlated by PCA for their values of similarities and differences (Kihlberg *et al*., 2006) and the coefficients were obtained from the correlation matrix of transformed variables (Bordes *et al*., 2008). Figure 3 shows the two dimensional graphics with the projection of texture parameters and chemical composition of fruit bodies of the three strains grown on 5 different substrates. The two main components, factors 1 and 2, describe about 57.96% and 26.79% of the variance, respectively, so explaining 84.75% of the variance.

The first and second axes describe virtually all variations in texture and chemical composition. It shows at the positive end of the first axis (factor 1) parameters like hardness, chewiness and gumminess, as well as moisture and crude fibre, which in the case of Figure 3a are correlated with strains CS and UAP9, while in figure 3b, correlation with substrates YB:WS50:50% and WS100% is observed. Cohesiveness and protein content were found at the negative end of the second axis, they are therefore correlated with strains CS and CxU and with all substrates.

Springiness, adhesiveness and ash content are at the positive end of the second axis (factor 2). Figure 3 shows the vectors of parameters strongly correlated, i.e. hardness, gumminess and chewiness with moisture and crude fibre. There were different parameters positively correlated with each other, such as adhesiveness, springiness with ash content, as well as cohesiveness with protein content. The parameters whose vectors form an angle close to 180° are negatively correlated, just like springiness and adhesiveness with cohesiveness while hardness, gumminess and chewiness with springiness and adhesiveness.

The correlation matrix of transformed variables for texture parameters and chemical composition, indicates that significant correlations (≥0.36, data not shown) were obtained between the different parameters of texture and between these and the chemical composition of fruit bodies (Borda *et al*., 2008). Using PCA, this study shows that there is an important relationship between textural parameters and chemical composition of fruit bodies and that the strain and type of substrate is important for both texture and chemical composition.

Conclusions

Yucca bagasse is a suitable material for the formation and development of oyster mushrooms, and therefore it is an option to produce oyster mushrooms commercially. It improves production while maintaining the physical and chemical quality of the fruiting bodies, when added as a supplement to wheat straw in proportions up to 50%. The use of yucca bagasse is a sustainable alternative as it provides an opportunity to make use of an existing agro-industrial residue from the extraction of yucca juice.

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