UTILIZATION OF SELECTED URBAN WASTES AS SUBSTRATE SOLUTIONS IN THE GROWTH AND YIELD PERFORMANCE OF *PLEUROTUS SAJOR-CAJU* (FR.) SINGER (GRAY OYSTER MUSHROOM)

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Abstract: Mushrooms are being cultivated worldwide for its nutritional and medicinal value. Agricultural wastes rich in lignocellulosic substances are commonly used as substrates for mushroom cultivation. Moreover, selected urban wastes that contain lignocellulosic substances are also potential substrates which when utilized can help lessen wastes in the environment. This study tested the possible utilization of selected urban wastes as solution substrates for the growth and yield performance of *Pleurotus sajor-caju* (Fr.) Singer. Four different urban waste substrates (banana peels, newspaper, cotton textile scraps and mixture of leaf litters) and their combinations were used in the study with sawdust which served as the control. Percentage of mycelial growth, colonization period, primordial formation, harvesting time, number of fruiting bodies, cap diameter, stalk length, total yield and size of the mushroom and biological efficiency of the substrates used were assessed to determine the effects on growth and yield. Results revealed that the substrate with 50% newspaper + 50% sawdust showed rapid colonization, fast primordial formation and earliest harvest day and obtained the highest yield of mushroom and biological efficiency. Mushrooms did not proliferate mostly in the substrates with the combinations of leaf litter, however, 25% leaf litter + 75% sawdust produced less yield of mushrooms. The least yield of mushrooms was from the substrate with 100% cotton textile scraps. Other combinations used in the study are effective substrates in cultivating P. sajor-caju, but have different average number and yield of mushrooms produced. The study demonstrated that faster mycelial growth is consistent with better yield of the mushroom and highest biological efficiency of the substrate.

***Keywords**: Pleurotus sajor-caju, urban wastes, mycelial growth, yield, biological efficiency*

1. INTRODUCTION

Gray oyster mushroom, *Pleurotus sajor-caju* (Fr.) Singer is a basidiomycete, which belongs to the order Agaricales. It is an edible mushroom which is readily available in most supermarkets and is considered as the third largest cultivated mushroom in the world. Cultivation of the oyster mushroom has recently increased tremendously throughout the world because of its ability to grow in a wide range of agro-based residues. It is useful decomposer of various agricultural wastes (Kurt and Buyukalaca, 2010).

The Philippines is an agricultural country and its economy is mainly dependent on agriculture. Even though mushroom cultivation is a profitable business, mushroom farming in the Philippines is not spreading fast. Its present cultivation protocols are limited, perhaps due to the limited local knowledge about its culture. Hence, it is

necessary that people are made aware of the simple methods required for the successful cultivation of edible mushrooms.

The organic materials, on which mushrooms derive their nutrition, are referred to as substrates. Mushrooms are unique group of organisms which assemble their food by secreting and degrading enzymes, decompose the complex food materials present in the biomass where they grow and generate simpler compounds which they then absorb and transform into their own peculiar tissues. The substrate materials used in mushroom cultivation are usually by-products from industry, households and agriculture and are usually considered as wastes (Chang and Miles, 2004). These wastes, when carelessly disposed in the surrounding environment by dumping or burning, will consequently lead to environmental pollution which may cause health hazards.

It is a common knowledge that lignocellulosic wastes are readily available in both rural and urban areas. The most apparent environmental issue we literally see every day is the problem on solid waste management. Solid waste disposal continues to be a growing crisis in urban centers in the Philippines. Some of these types of solid wastes can be harmful to the environment when improperly disposed. Indeed, the issue on solid waste management impinges development, safety and security of the human race (Payongayong, 2011). Cities are now grappling with the problems of high volume of wastes, the costs involved, the disposal technologies and methodologies, and the impact of wastes on the local and global environment. Moreover, these situations have also provided a window of opportunity for cities to find solutions - the exploitation of such wastes by the mushroom industry. The use of lignocellulosic materials, which provide sustainable biomass resource for the growth of edible and medicinal mushrooms, is of great environmental importance by recycling organic wastes; thereby help in controlling problems on pollution (Chang and Miles, 2004). Mushroom farming is a highly remunerative enterprise with quick return in a short period of time (Dhal, 2012). Because of the tropical and humid climate, the Philippines is an ideal country which favor the growth of many types of edible mushrooms. This means that mushroom farming has a big potential and can even be considered as revenue-generating industry. The present study was conducted in order to evaluate the use of different urban wastes such as banana peels, old newspaper, cotton textile scraps, and a mixture of leaf litters from the trees of *Mangifera indica, Terminalia catappa, Swietenia mahogani,* and *Bambusa vulgaris* as substrates for the cultivation of *P. sajor-caju* (Fr.) Singer.

2. METHODOLOGY

2.1 Study Site

Mushroom cultivation and composting of substrates were conducted in the experimental mushroom house located at Gabriela Silang Building area of the Polytechnic University of the Philippines Sta. Mesa, Manila. Bagging and sterilization of the substrates were performed at the Institute for Science and Technology Research Laboratory.

2.2 Propagation of Subculture

Mother culture of *P. sajor-caju* purchased at the Rizal Technological University Mushroom Laboratory was aseptically subcultured on the prepared 10 bottles of potato dextrose agar (PDA). All subcultures were incubated at room temperature for 10-14 days or until the media was fully impregnated with the mycelia.

2.3 Production of the Spawn

Three kilograms of sorghum grains were soaked in 6L of water for one night. It was washed thoroughly in tap water and strained to remove all water. Then, it was placed in a clean casserole with water 1 inch above level of grains and boiled for 30 minutes to soften the grains. It was strained and cooled by spreading in a nylon cloth or fine screen leaving the grains just damp with 65-70% moisture. Three-fourths of the grains were distributed equally in bottles, plugged with cotton and covered with foil supported with rubber band. All bottles containing sorghum grains were sterilized at 15lbs. for 1 hour. Sterilized sorghum grains were inoculated aseptically with young and vigorous subcultures of the mushroom mycelium and were incubated at room temperature until the grains were fully impregnated with the mushroom mycelia.

2.4 Composting of Substrates

Four urban wastes such as banana peels, old newspapers, cotton textile scraps and mixtures of leaf litters from the trees of *Mangifera indica, Terminalia catappa, Swietenia mahogani,* and *Bambusa vulgaris* were used as mushroom substrates. Sawdust served as control treatment for *P. sajor-caju* cultivation.

The fresh banana peels weighing 13.3 kg, obtained from the banana cue vendors along the vicinity of Pureza Street in Sta. Mesa, Manila, were allowed to dehydrate by sun drying for one week. The dried and ground peels were allowed to compost with calcium carbonate and water in the ratio 32:2:66 of substrate, $CaCO_3$ and water respectively for one day. The mixed substrate drained with water was further allowed to compost in a large container for a period of 14 days.

Shredded newspaper wastes weighing 13.3 kg, obtained from the junkshops around Metro Manila, were soaked in fresh water added with 331 g urea, 331 g gypsum ($CaSO_4 \bullet 2H_2O$) and 133 g potassium chloride (KCl). The mixtures were allowed to stand for a week with occasional mixing, allowing all the materials to absorb the water and salts. The composted newspapers were squeezed manually by hand, leaving at least 85% moisture.

Cotton textile leftovers weighing 13.3 kg gathered from the tailoring shop at Cainta, Rizal and Divisoria Market, were composted by soaking in water added with 1662.5 g calcium carbonate ($CaCO_3$) for a week. The cotton textile wastes drained with excess water were spread on a clean 1 m×2 m platform and sprinkled with urea and rice bran. Three layers of cotton textile wastes with urea and rice bran were done in a platform

covering it with black polythene tarp and turning twice in two-day intervals. After seven days, the excess water was removed manually by squeezing (Rajapakse, 2011).

Leaf litters, *Mangifera indica, Terminalia cattapa, Switenia mahogani* and *Bambusa vulgaris*, each weighing 3.5 kg were collected at the Ninoy Aquino Parks and Wildlife Center. These were fragmented into small pieces (1-3cm), mixed and soaked in water for two days, squeezed manually, and were spread in the ground to build up a heap. Urea, grass clippings and wheat bran were sprinkled and mixed into the leaf litters. Three layers of leaf litters with urea, grass clippings and wheat bran were done in a platform covered with black polythene tarp which were allowed to compost for a period of 20 days.

The sawdust medium which served as the control was prepared by mixing the 28 kg sawdust with 7 kg rice bran, 350 g gypsum, 175 g lime and 350 g sugar dissolved in distilled water.

2.5 Bagging and Sterilization of Substrates

The materials were mixed and the moisture was increased by adding clean water until 65% moisture content was reached. Nutrient supplements such as 25 grams of rice bran and 5 grams of sugar were added to each substrate and substrate combinations (Hanko, 2001). Calamansi extract was also added to each 500 grams of substrate and substrate combinations to adjust the pH of the substrate (Custodio, 2004), for at least pH 5.6-6.0 (Gibriel *et al*., 1996). Prepared substrates were packed tightly in polypropylene bags containing 500 g per pack. Each substrate bag was provided with PVC ring which served as neck tightened by rubber band and tightly plugged with cotton ball. The fruiting bag for each substrate and substrate combination was prepared with five replicates which were autoclaved for two hours at 121°C and 15 psi. All sterilized fruiting bags were allowed to cool for 24 hours before the inoculation of the *P. sajor caju* mycelia.

2.6 Seeding Spawn to the Substrates and Incubation of the Fruiting Bags

One teaspoonful of mother grain spawn culture containing mycelia was placed aseptically through the hole of each fruiting bag separately. Fruiting bags were kept in the mushroom house allowing the completion of the whitish mycelial growth (Hanko, 2001). Relative humidity of around 80% to 85% was maintained in the room while temperature was monitored daily assuring that the room was kept cool and moistened.

2.7 Culture Condition for Fructification of Basidiocarp

After mycelia have completely colonized the bags, the bags were then opened to trigger fructification. Water spraying was done thrice a day until the mushrooms were matured enough to be harvested. The temperature was monitored daily. High relative humidity of 80 to 85% and proper ventilation of the mushroom house was maintained for the development of fruiting body with the use of hygrometer (Custodio, 2004).

2.8 Harvesting of Mature Basidiocarp

Only the first mushroom flush of each bag was harvested and used for data analysis.

2.9 Data Collection and Statistical Analysis

The experiment was laid out in Completely Randomized Design (CRD) with sixteen (16) treatments and a control with five (5) replicates each. The parameters that were used for the collection of data were:

a. percentage of mycelium running per week;

b. number of days for colonization of the mycelia, primordial formation and first harvest;

c. expanded caps diameter, stalk length, and size of the mushroom produced; and

d. number and weight of the harvested fruit bodies, and its biological efficiency of the substrates.

Number of days for colonization was measured as long as the mycelium was growing on the substrate or until the substrate was fully covered and the diameter of expanded caps and length of stalks were measured with a digital vernier caliper. Total weight of the fruiting bodies harvested from the first mushroom flush was measured as total yield of mushroom. Size of the mushrooms was calculated by total weight of the mushroom / number of fruiting bodies harvested. The biological efficiency (BE) of the different substrates used was calculated by following the formula of Chang *et al.,* (1981):

$$BE=\frac{\text{Fresh weight (g) of the mushroom harvested}}{\text{Dry weight (g) of the substrate}} \text{ x } 100$$

The data were statistically analyzed by SPSS 17.0 software. Due to the heterogeneity and non-normality of the data, the means were compared by using Kruskal-Wallis ANOVA followed by non-parametric post-hoc test, Mann-Whitney U.

3. RESULTS AND DISCUSSION

Four different urban waste substrates (banana peels, newspaper, cotton textile scraps, and mixture of leaf litters) and their combinations with sawdust (control) for a total of seventeen (17) treatments were utilized to determine their effects on the growth and yield of oyster mushroom (*P. sajor-caju*) and to identify the optimum substrate suited for the mushroom.

3.1 Weekly mycelial growth and colonization period of *P. sajor-caju* on different substrates and substrate combinations

Mycelial examination for the growth and yield performance is the preliminary step in determining whether the substrate creates suitable internal conditions for fruiting of the mushroom. Outstanding growth of mycelia is a vital factor in mushroom cultivation (Pokhrel *et al*., 2009). Mushroom mycelia (vegetative phase) is important in the ecosystem because it is able to biodegrade the substratum. Therefore the use of wastes of urban products as potential substrate for mushroom production is possible.

Percental weekly mycelial growth on different substrates and substrate combinations is shown in Table 1 and the colonization period of the mycelia on each treatment is shown in Table 2. Mycelial growth was observed on different substrates and substrate combinations after inoculation. Upon incubation, percentage of mycelial growth was measured every week. The number of days taken to fully colonize the different substrates and substrate combinations differ from one another spanning a range of 21 to 35 days. Kruskal-Wallis H - test showed that the percentage of mycelial growth per week were significantly different from all the treatments at $p \leq 0.050$, excluding the fifth week. In the first week of incubation, T7 – 75% newspaper + 25% sawdust had the highest percentage of mycelium coverage with 25%, followed by T4 – 100% cotton textile scraps and T11 – 50% newspaper + 50% sawdust which had the same percentage of the mycelium run with 24%. T2 – 100% banana peels had the least percentage of mycelium run. However, the mycelium slightly dropped on T4, which completed its colonization on the fourth week of incubation. On T11, the mycelium continuously grew and completed growth on the third week of incubation. On most of the substrates, mycelial growth was completed at the fourth week. Moreover, T6 – 75% banana peels + 25% sawdust, T9 – 75% leaf litter + 25% sawdust, and T13 – 50% leaf litter + 50% sawdust had the slowest growth of mycelia which had a colonization period of 28 to 35 days. Thus, T11 and T8 - 75% cotton textile scraps + 25% sawdust possessed the fastest growth for mycelia which have both a total colonization period of 21 days (Figure 1).

Table 1. Comparison of weekly mycelial growth of *P. sajor-caju* on different substrates and substrate combinations (Mean ± SE, n = 5).

Substrates	Week				
	1st (%)	2nd (%)	3rd (%)	4th (%)	5th (%)
T1 Sawdust, S (control)	9.00±2.45	55.00±8.80	88.00±5.15	100.00±0.00	
T2 Banana Peels, BP	6.20±2.25	29.00±9.14	15.60±3.30	100.00±0.00	
T3 Newspaper, N	17.00±1.23	57.00±4.47	80.00±12.15	100.00±0.00	
T4 Cotton Textile Scraps, C	24.00±2.55	69.00±4.58	94.00±0.00	100.00±0.00	
T5 Leaf Litters, L	no growth	no growth	no growth	no growth	no growth
T6 BP + S, 3:1	17.60±6.19	48.00±11.04	80.00±8.94	97.00±2.89	100.00±0.00
T7 N + S, 3:1	25.00±2.74	60.00±6.12	90.00±6.12	100.00±0.00	
T8 C + S, 3:1	22.00±1.58	71.00±7.42	100.00±2.00		
T9 L + S, 3:1	8.00±1.23	23.00±1.23	63.00±2.00	94.00±6.04	100.00±0.00
T10 BP + S, 1:1	12.00±1.23	32.00±2.00	83.60±6.55	100.00±0.00	
T11 N + S, 1:1	24.00±2.92	75.00±0.10	100.00±3.32		
T12 C + S, 1:1	20.00±1.87	55.00±2.45	98.00±3.74	100.00±0.00	
T13 L + S, 1:1	12.00±2.00	17.00±7.00	52.00±13.04	78.00±6.00	100.00±0.00
T14 BP + S, 1:3	17.00±4.64	43.00±11.28	76.00±9.80	100.00±0.00	
T15 N + S, 1:3	21.00±3.67	60.00±8.57	91.00±4.00	100.00±0.00	
T16 C + S, 1:3	16.00±2.00	51.00±7.00	93.00±7.18	100.00±6.00	
T17 L + S, 1:3	22.00±4.06	46.00±1.00	73.00±4.48	95.40±2.62	100.00±0.00

Table 2. Comparison of colonization period, primordial formation and first harvest days of *P. sajor-caju* on different substrates and substrate combinations. (Mean±SE, n = 5).

Substrates	Colonization Period (days)	Primordial Formation (days)	First Harvest (days)
T1 Sawdust, S (control)	25.20±1.72	28.40±1.81	32.60±1.91
T2 Banana Peels, BP	28.00±0.00	30.60±0.51	35.20±0.66
T3 Newspaper, N	25.20±1.72	29.40±2.06	33.40±1.72
T4 Cotton Textile Scraps, C	23.80±1.72	37.50±8.50	41.00±9.00
T5 Leaf Litters, L	no growth	no growth	no growth
T6 BP + S, 3:1	28.00±3.13	31.60±3.27	35.00±3.33
T7 N + S, 3:1	23.80±1.72	27.20±1.36	30.00±1.45
T8 C + S, 3:1	21.00±1.72	33.20±2.69	36.40±2.62
T9 L + S, 3:1	35.00±1.40	no growth	no growth
T10 BP + S, 1:1	28.00±0.00	31.80±0.97	35.20±0.97
T11 N + S, 1:1	21.00±1.72	25.00±0.00	28.00±0.00
T12 C + S, 1:1	22.40±0.00	27.60±7.08	30.60±7.80
T13 L + S, 1:1	35.00±1.40	no growth	no growth
T14 BP + S, 1:3	25.20±3.83	33.20±2.11	35.60±2.32
T15 N + S, 1:3	26.60±0.00	32.00±1.58	36.20±1.59
T16 C + S, 1:3	25.20±1.40	no growth	no growth
T17 L + S, 1:3	28.00±0.00	37.60±1.67	40.60±1.47

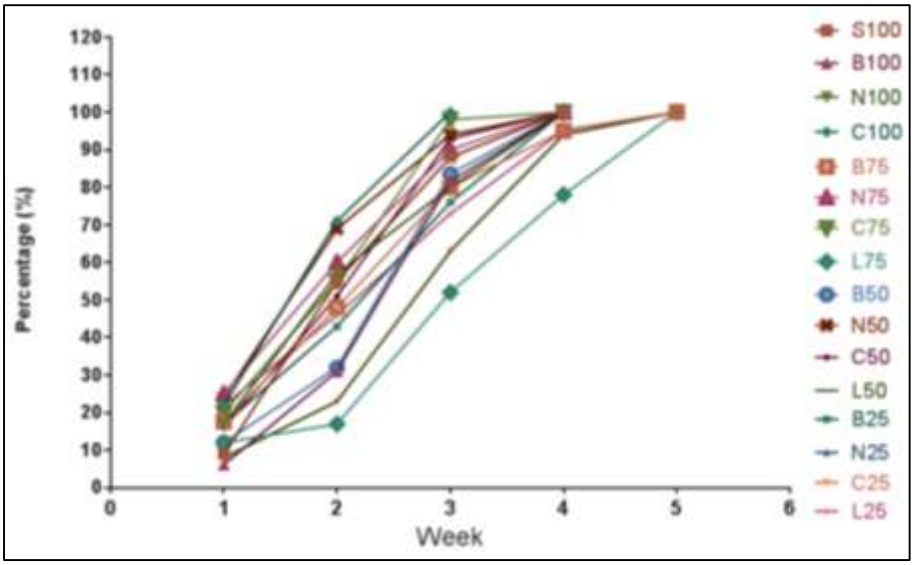

Figure 1. Graphical illustration of the relationship between the percentages of weekly mycelial growth of *P. sajor-caju* on different substrates. Substrate names refer to the abbreviations designated in the text.

Zadrazil (1980) and Bisaria *et al.* (1987) reported the ability of *P. sajor-caju* to degrade major components of lignocellulosic materials. Conversely, there is no growth of mycelia observed from T5 – 100% leaf litters probably because it didn't meet the nutritional requirement for the growth of the mycelia into the substrate. This was also supported by Gerrits (1868) as he recorded the rapid degradation of lignin and slow degradation of cellulose and hemicellulose during spawn-run.

Narain *et al.* (2008) reported that mushroom mycelia growth and primordial development is dependent on the lignocellulosic materials especially the C:N ratio. The physical nature and high C:N ratio of leaf litters may not be suitable for the cultivars of oyster mushroom. On the other hand, the slow spawn running on cotton textile scraps substrate may be due to high nitrogen content of the urea that was added to the substrate. Such may probably inhibit mushroom growth.

3.2 Primordial formation and first harvest period of *P. sajor-caju* on different substrates and substrate combinations

Primordial formation is the second stage of mycelial growth during cultivation of *P. sajor-caju*. Primordia (pinheads) are the tiny fruit bodies of mushroom with a size greater than 0.01mm. They appeared after the completion of mycelial growth of mushroom. Environmental conditions of the substrate affect directly the growth of primordia. Data regarding number of days taken for mycelium colonization, primordial formation and first harvest is shown in Table 2.

Small pinheads were observed at 4 to 9 days immediately after the completion of mycelium running (21 to 35 days). These primordia became first prominent particularly in T11 – 50% newspaper + 50% sawdust on the 25th day. The latest occurrence of primordia was observed in T17 – 25% leaf litters + 75% sawdust at 37th day. As these primordia grow, number of days was recorded until it was fully matured and taken to be harvested. Mature mushrooms were produced 7 to 18 days after the completion of mycelium running and basidiocarps from T11 – 50% newspaper + 50% sawdust were the earliest to be harvested after 28 days. In contrast, mushrooms produced on T4 – 100% cotton textile scraps (41 days) and T17 – 25% leaf litters + 75% sawdust (40.60 days) needed the highest amount of days until harvest. Kruskal-Wallis H - test showed that the number of days of mycelial colonization, primordial formation and first harvest were significantly different from all the treatments at $p \leq 0.050$. Khan *et al*., (2001) described the cultivation of oyster mushroom using different ligno-cellulosic substrates and found that pinhead formations take place after 7-8 days while basidiocarps formation takes place after 10-12 days of mycelium running. Shah *et al*., (2004) investigated oyster mushroom cultivation and observed that pin head like structures are formed after 6-7 days of mycelium running.

Overall, T11 – 50% newspaper + 50% sawdust showed the best results in terms of colonization period, primordial formation days and first harvest days when compared to the control, while T17 – 25% leaf litters + 75% sawdust demonstrated the opposite effect in these respects (see Figure 2). Oei (2005) reported that substrate having high quality lignin and cellulose contents delay the start of the pinning and fruit body formation.

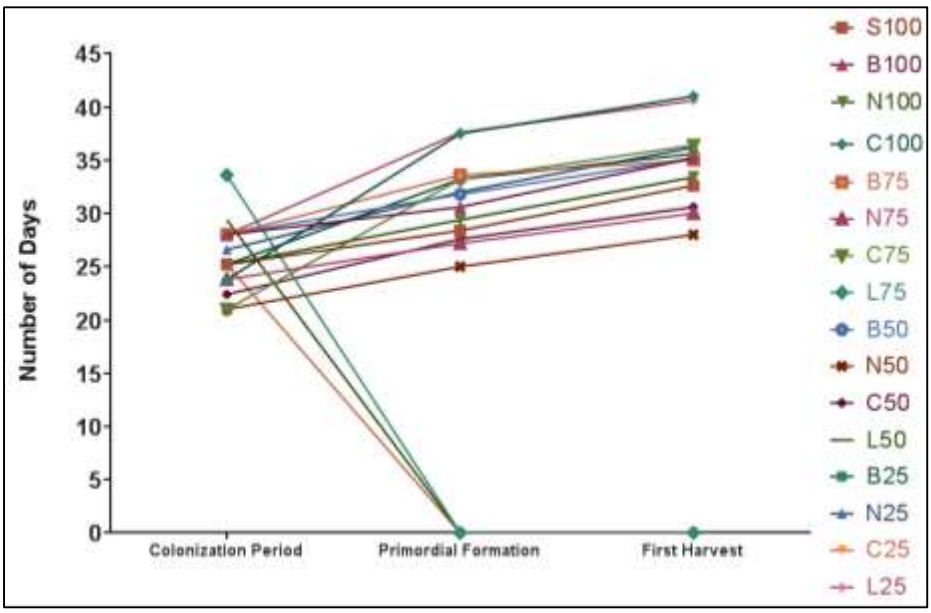

Figure 2. Graphical illustration of the relationship between colonization, primordial formation, and first harvest days of *P. sajor-caju* on different substrates.

In the present study, the fruiting bodies appeared 28-41 days after inoculation of spawn. These findings are in conformity with the results of Pokhrel *et al.* (2013) who reported that formation of fruiting bodies was 32-44 days after inoculation of spawn.

A complete mycelium growth was observed for T9 – 75% leaf litters + 25% sawdust, T13 – 50% leaf litters + 50% sawdust, and T16 – 25% cotton textile scraps + 75% sawdust, however, pinheads and fruiting bodies were not formed. The lack of primordia and fruiting bodies may be due to nutritional requirements of *P. sajor-caju*, which were not fulfilled by the respective substrates or because the substrates were not fully degraded.

3.3 Mushroom yield performance

Determining the yield of mushroom produced on a particular waste is very essential to identify the ability of that mushroom to produce the enzymes essential to degrade the major components of the waste, and thereafter absorb it as food.

The number of well-developed fruiting bodies, cap diameter and stalk length during the first flush of *P. sajor-caju* produced from each substrate and substrate combination is presented on Table 3. Total yield and size of *P. sajor-caju* for each treatment and biological efficiency of each substrates used are shown in Table 4.

3.3.1 Number of fruiting bodies produced on each treatment

The fruiting body is the edible part of the mushroom and they differ on size, shape and coloration which aid in identification of the specific species. Dry and pin headed fruiting bodies were discarded but twisted and tiny fruiting bodies were included during counting. The number of effective fruiting body did not vary significantly ($p \geq 0.05$) in different substrates. The highest number of effective fruiting body was obtained from T15 – 25% newspaper + 75% sawdust with an average number of 25.60, while the lowest amount was obtained from T4 – 100% cotton textile scraps having only one fruiting body.

The formation and growth of fruiting bodies are sensitive to environmental conditions, such as temperature, humidity, carbon dioxide concentration, and moisture content in the mushroom substrate. Improper balance of these factors can induce fruiting body deformations. According to Chang and Miles (2004) nutrient content of substrates affects the growth and formation of fruit bodies of *Pleurotus* species.

3.3.2 Cap diameter

Samples of mushroom caps were taken from each treatment and the diameters were recorded. T2 – 100% banana peels produced bigger caps with an average diameter of 48.83 mm, while T10 – 50% banana peels + 50% sawdust produced smaller caps with

an average diameter of 34.16 mm. It was also observed that cap diameters of other substrates were similar to those of the control (T1 – 100% sawdust). Statistically, there were no significant differences among the treatments in terms of the size of the cap diameter. Therefore it can be concluded that the size of the caps is not influenced by the composition of the substrates. As the cap diameter increases, the yield decreases, and other substrates gave better results than control.

Table 3. Number of individual growth of fruiting bodies, cap diameter and stalk length during the first flush of *P. sajor-caju* in corresponding substrates and substrate combinations (Mean±SE, n = 5).

Substrates	Number of Fruiting Bodies	Cap Diameter (mm)	Stalk Length (mm)
T1 Sawdust, S (control)	16.80±2.65	41.21±3.79	24.04±2.03
T2 Banana Peels, BP	12.00±3.13	48.83±6.09	35.97±1.96
T3 Newspaper, N	19.40±4.75	44.69±3.00	34.45±3.40
T4 Cotton Textile Scraps, C	1.00±0.00	34.41±0.00	32.94±0.00
T5 Leaf Litters, L	no growth	no growth	no growth
T6 BP + S, 3:1	17.60±1.60	36.05±1.58	23.00±2.00
T7 N + S, 3:1	20.40±4.30	47.69±2.60	35.90±5.56
T8 C + S, 3:1	9.80±2.80	36.90±2.75	24.52±2.98
T9 L + S, 3:1	no growth	no growth	no growth
T10, BP + S, 1:1	20.20±5.52	34.16±2.44	25.32±3.20
T11 N + S, 1:1	24.00±4.12	44.15±2.79	34.79±1.84
T12, C + S, 1:1	10.40±3.86	36.64±12.01	26.54±10.48
T13 L + S, 1:1	no growth	no growth	no growth
T14 BP + S, 1:3	16.40±2.77	38.74±2.79	26.09±1.90
T15 N + S, 1:3	25.60±6.30	36.06±2.96	25.43±1.12
T16 C + S, 1:3	no growth	no growth	no growth
T17 L + S, 1:3	16.40±3.76	42.25±5.34	28.75±0.83

Onyango *et al.* (2011) reported that large-sized fruit bodies were considered to be of good quality and rated highly in mushroom production but Shen and Royse (2001) reported this as an inferior quality since such fruit bodies tend to break during packaging thereby reducing their quality. However, major ecological factors that affect stalk height, stalk diameter and cap size in mushrooms are temperature, humidity, fresh air and compact material (AMGA, 2004).

3.3.3 Stalk length

The different substrates used in this study have significant influence on stalk. The longest length of stalk was recorded in T2 – 100% banana peels having an average length of 35.97 mm. The shortest stalk length was recorded on T6 – 75% banana peels + 25% sawdust having 23 mm in average.

A variation in mushroom cap diameter and stalk length was observed in the seventeen substrates used in this study. Relatively small mushroom cap diameter and long mushroom stipe length are undesirable characteristics for marketable quality. Environmental conditions as well as supplementation of substrates with various additives including nitrogen sources had been reported to improve growth, yield and quality of mushrooms (Royes, 2002; Panjabrao *et al*., 2007; Onyango *et al*., 2011). In our study, the supplement of rice bran and urea to the substrates could shorten mushroom stipe length and enlarge mushroom cap diameter, which may result from the supplement changing the physical properties and C/N ratio of the substrates particularly in the newspaper.

3.3.4 Total yield of mushroom produced in first flush

Substrate with 50% newspaper + 25% sawdust (T11) yielded the highest average weight of fruit bodies (123.90 g), followed by T7 75% newspaper + 25% sawdust with an average weight of 106.20 g and the substrate with pure newspaper (T3) which gave an average weight of 84.80 g. The lowest yield was recorded in the 100% cotton textile scraps (T4) having an average weight of 3.54 g. Statistical analysis showed significant differences between the treatments in terms of total yield of mushrooms produced with $p \leq 0.05$.

Another study done by Baysal and Peker (2001) showed that the waste paper substrate combination induced faster mycelial growth and higher yield of oyster mushroom (*P. ostreatus*) when compared to waste paper alone. Yield increases may be due to several factors. Firstly, the increased level of nutrient available for higher production rates would provide more energy for mycelial growth and primordial formation. Secondly, supplement of the substrates increased the water-holding capacity, and decreased the mortality of young fruiting bodies due to water shortage.

3.3.5 Size of mushroom

Among the treatments, the biggest size of mushrooms was observed on the substrate with pure banana peels (T2) having an average size of 8.08 g. The smallest size was recorded in T6 – 75% banana peels + 25% sawdust and T10 – 50% banana peels + 50% sawdust with an average of 2.34 g. This was followed by T15 – 25% newspaper + 75% sawdust with 2.77 g in average. Statistically, there were significant differences among the treatments in terms of the size of the mushroom ($p \leq 0.05$).

3.3.6 Biological efficiency of substrates used

Results showed that the 50% newspaper + 50% sawdust (T11) produced the highest biological efficiency with 68.89%, followed by the substrate with 75% newspaper + 25% sawdust (T7) with an average percentage of 54.82%. The lowest percentage for the biological efficiency was calculated for pure cotton textile scraps (1.22%). The biological efficiency of the urban waste used in the cultivation of *P. sajor-*

caju showed a highly significant difference ($p \leq 0.05$). Superiority of newspaper substrate was reported by Vyas *et al.* (2003). The variation in biological efficiency of different substrates might be due to low lignolytic and cellulonitic activity of the substrates used (Pathak and Goel, 1988).

Table 4. Total yield and size of *P. sajor-caju* in corresponding treatments and biological efficiency of each substrate used (Mean±SE, n = 5).

Substrates	Total Yield (g)	Size of Mushroom (g)	Biological Efficiency (%)
T1 Sawdust, S (control)	48.91±7.37	3.13±0.62	18.73±2.08
T2 Banana Peels, BP	60.22±5.44	8.08±3.25	19.86±1.61
T3 Newspaper, N	84.80±18.23	4.76±0.74	35.26±8.62
T4 Cotton Textile Scraps, C	3.54±0.00	3.54±0.00	1.22±0.00
T5 Leaf Litters, L	no growth	no growth	no growth
T6 BP + S, 3:1	40.05±1.97	2.34±0.19	14.52±1.04
T7 N + S, 3:1	106.20±19.46	5.77±0.78	54.82±12.41
T8 C + S, 3:1	26.06±3.63	3.26±0.67	9.90±1.79
T9 L + S, 3:1	no growth	no growth	no growth
T10, BP + S, 1:1	39.09±4.69	2.34±0.46	14.55±2.19
T11 N + S, 1:1	123.90±5.29	5.73±0.9	68.89±5.12
T12, C + S, 1:1	29.19±7.86	4.38±2.86	12.48±3.19
T13 L + S, 1:1	no growth	no growth	no growth
T14 BP + S, 1:3	43.64±4.27	3.16±0.61	13.73±1.59
T15 N + S, 1:3	47.03±6.14	2.77±0.86	19.16±2.57
T16 C + S, 1:3	no growth	no growth	no growth
T17 L + S, 1:3	51.08±6.02	4.54±1.87	21.32±2.21

Mann-Whitney U Test revealed that T11 – 50% newspaper + 50% sawdust showed significant differences towards T1 (control), T2 (100% banana peels), T6 (BP+ S, 3:1), T8 (C + S, 3:1), T10 (BP + S, 1:1), T12 (C + S, 1:1), T14 (BP + S, 1:3), T15 (N + S, 1:3), and T17 (L + S, 1:3) in terms of the total yield of the mushrooms produced and biological efficiency of the substrates used. It also showed significant differences in terms of colonization period, primordial formation days and first harvest days ($p \leq 0.05$). The higher mushroom yield and biological efficiency correspond to the mycelial growth, colonization period and harvest period (Pokhrel, 2013).

The measured parameters proofed that T11 is the best substrate composition among all tested substrates to grow *Pleurotus sajor-caju*. The probable reason for the best performance of *P. sajor-caju* on newspaper substrates might be that the nutrient required for the mushroom particularly for its mycelium run, primordia development and yield were supplied by these substrates which decomposed quicker than the other substrates.

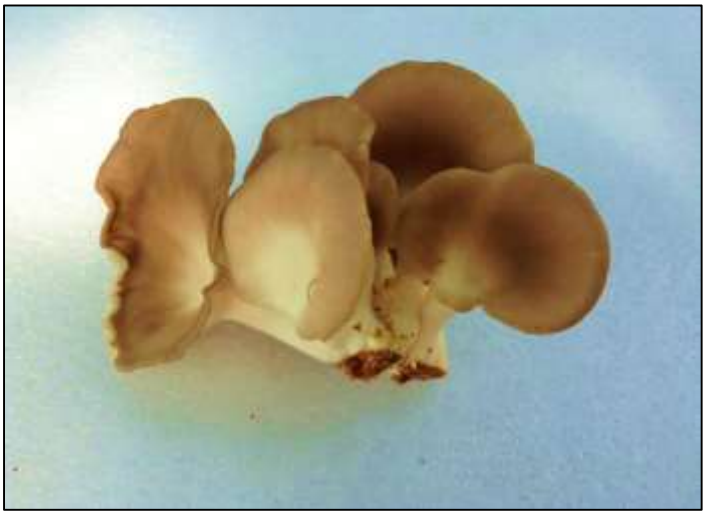

Figure 3. Harvested P. sajor-caju from sawdust (Jambaro and Neri, 2014).

4. CONCLUSIONS AND RECOMMENDATIONS

The study reveals that 50% newspaper+ 50% sawdust showed rapid colonization, primordial formation, and earliest harvest day and obtained the highest yield of mushroom and biological efficiency. Thus, it can be established as the optimum substrate used to grow *P. sajor-caju*. The least yield of mushrooms was from 100% cotton textile scraps. The remaining substrates with their respective concentrations used in this study are also effective in promulgating *P. sajor-caju*, but have different number and yield of mushrooms produced and grown. The study demonstrated that faster mycelial growth is consistent with better yield and highest biological efficiency of the substrate. The present study thus explored the opportunities and possibilities for the cultivation of *P. sajor-caju* using largely available urban wastes. This in turn would help to meet the growing demand of the protein need.

Figure 4. Harvested P. sajor-caju from (A) 100% banana peels; (B) 75% banana peels + 25% sawdust; (C) 50% banana peels + 50% sawdust; (D) 25% banana peels + 75% sawdust; (E) 100% newspaper; (F) 75% newspaper + 25% newspaper; (G) 50% newspaper + 50% sawdust; (H) 25% newspaper + 75% sawdust; (I) 100% cotton textile scraps; (J) 75% cotton textile scraps + 25% sawdust; (K) 50% cotton textile scraps + 50% sawdust; (L) 25% cotton textile scraps + 75% sawdust (Jambaro and Neri, 2014).

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