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Development of a New Substrate for the Cultivation of the *Pleurotus sajor–caju* Mushroom through Controlled Composting

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ABSTRACT

The in-vessel composting of a substrate, around 60 kg, composed of 18 kg of green banana leaves (*Musa paradisiaca*) (B), 18 kg green grass (*Stenotaphrum dimidiatum*) (G), 15 kg mixed cardboard waste (C) and 4 kg untreated pinewood shavings (W), was carried out. Oxygen was provided by active aeration with the airflow kept at 540 ± 24 L/h. The average temperature monitored over the 23 days, was 46.2°C with a maximum of 56.5°C on day 8. The final composted BGCW mix had a moisture content (MC) of 42.3%, wet bulk density (BD_w) 186.0 kg/m^3 and particle size ranged from 26.67 mm to 4.699 mm, water holding capacity (WHC) 572.77%, porosity 53.18%, pH 7.94, electrical conductivity (EC) 1.18 dS/m and volatile solids content (VS) 78.7%. The nutrient content for the BGCW compost was 1.72% for nitrogen, 0.11% for phosphorus and 1.36% for potassium, totalizing to a sum of 3.19% as compared to bagasse, 0.001% for nitrogen, 0.003% for phosphorus and 0.006% for potassium. The BGCW compost along with bagasse were packed, in heat resistant bags, 750 g each, and pasteurised at 70°C for 4 h. The total yield of fructifications harvested from the three pickings was 950.30 g for bagasse and 810.60 g for the BGCW compost. The average biological efficiency on bagasse was 42.23 ± 4.4% as compared to the BGCW compost (36.03 ± 5.3%). The difference in yield could be primarily explained by higher fibre content in bagasse (47.3 ± 2.4%) as compared to the BGCW compost (40.5 ± 2.3%). It is envisaged that the BGCW compost was supplemented to bagasse for the cultivation of the *Pleurotus sajor−caju* mushroom.

Keywords: in−vessel composting, oyster mushroom, bagasse, respiration rate, sustainable agriculture

INTRODUCTION

Mushrooms are the reproductive organs of the largest and most complex fungi. Despite the fact that the mushroom neither belongs to the animal nor to the plant monarchy, it is more closely related to Kingdom Animalia than to Kingdom Plantae. This could explain as to why it contains all the nine essential amino acids required by the human body. Furthermore, like animals, mushrooms need to obtain nutrients from organic sources as they do not photosynthesise, since they lack chlorophyll. But, unlike animals, mushrooms are absorptive heterotrophs. This could well explain their classification into their own kingdom that is Kingdom Eumycota. According to Shalchian−Tabrizi *et al.* (2008), fungi are placed with the animals in the monophyletic group of opisthokonts. Furthermore, the 2007 classification of Kingdom Fungi recognises seven phyla (*Basidiomycota*, *Ascomycota*, *Glomeromycota*, *Neocallimastigomycota*, *Blastocladiomycota*, *Chytridiomycota* and *Microsporidia*) on basis of characteristics of their sexual reproductive structures.

Global interest in mushroom production

The global mushroom industry is growing in size and gaining popularity (USDA 2009; Okhuoya 2010). Consequently, higher returns from mushroom cultivation are noted for all world markets. For instance, the production of the three important mushrooms, *Agaricus*, *Lentinula* and *Pleurotus* mushrooms together make up nearly 100% of the mushroom industry in U.S and Spain. Interest is also based, nowadays, on the fact that these saprotrophs can offer an alternative for recycling lignocellulosic wastes into highly alimentative protein−rich biomass. More to the point, potential applications of the mushroom's lignocellulolytic enzymes are being considered in chemical, fuel, food, textile, laundry, pulp and paper industries as well as in animal feed production (Chen *et al.* 2010; Upadhyay and Singh 2010; Diaz *et al.* 2011). Because mushrooms exhibit a broad spectrum of characteristics, this is giving rise to a novel field of study called Mycotechnology. Its predominant domains include mycoremediation, mycofiltration and mycoforestry (Robles−Hernández 2008).

Pleurotus sajor−caju mushroom – Scientific classification and description

The *Pleurotus sajor−caju* mushroom belongs to Phylum Basidiomycota. Being saprophytic, it grows primarily on hardwoods and numerous composted and non−composted lignocellulosic wastes. The *Pleurotus sajor−caju* mushroom belongs to Phylum *Basidiomycota.* Being saprophytic, it grows primarily on hardwoods and numerous composted and non−composted lignocellulosic wastes. **Table 1** summarises the scientific classification and description of the *Pleurotus sajor−caju* mushroom. Compared to *Agaricus* and *Lentinula*, *Pleurotus* mushrooms are the easiest, fastest and cheapest to grow, require less preparation time and production technology (Mandeel *et al.* 2005). Hence, they are being increasingly popularised because they have a wide range of temperature adaptability and most of their substrates are readily available.

Furthermore, Chiu *et al*. (1998a), Martínez−Carrera (1998) and Juárez *et al.* (2010) state that after the mushrooms have been harvested, the fates of the 'spent compost' can be considered as:

- Animal feed as the mushroom mycelium boosts its protein content.
- Substrate for the cultivation of other mushrooms.
- Soil conditioner, since it is still rich in nutrients and

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Table 1 Scientific classification and description of the *Pleurotus sajor-caju* mushroom.

Scientific classification	Description
Kingdom: Fungi	**Cap**
Phylum: Basidiomycota	○ Smooth, oblong becoming convex with time.
Class: Homobasidiomycetes	○ 50 to 200 mm in diameter.
Order: Agaricales	○ Moist, hairless and fragrant.
Family: Tricholomataceae	○ Margin is smooth with a slight wave.
Genus: *Pleurotus*	○ Colour ranges from white to brown to blue-gray.
Species: *P. sajor caju*	**Flesh**
	○ Depending on the variety, the flesh can be thin or thick.
	○ The colour of the most common variety is white.
	○ Different colours can be identified in the wild such as, yellow, pink, blue, and gray, which can be grown.
	○ Chewy texture tasting and like an oyster.
	Gills and Stem
	○ Gills are hairless, decurrent and attached to the stem.
	○ The stem is short, often horizontal and emerging from the substrate and descends the short, stub-like, lateral stalk, when it exists.
	○ The colour of the stem and gills range from white to light yellow with time.
	Spores
	○ Spores form a white to lilac-gray print on dark media.
	Mycelia
	○ White colour.

polymeric components that enhance soil structure.
- Permaculture component.
- Paper manufacture and cardboard production.
- Agents that digest pollutants, like polychlorinated phenols on landfill waste sites as it contains populations of microorganisms able to digest the natural phenolic components of lignin.
- Enzyme production.
- Biogas production.

History of substrate development and cultivation

Oyster mushrooms have been cultivated in most tropical, sub−tropical and temperate zones of the world. Logs, tree stumps and dead wood are the natural habitats of the oyster mushroom. It normally appears in shelf−like layers. Its cultivation traditionally started on tree stumps and wood logs to conform to its substrate. However, this practice became less popular due to long incubation periods, basically 6 to 9 months, and low yields. By the late 1950s, sawdust was used as the substrate material in polypropylene bags. Fruit body formation was reduced to approximately two months (Block *et al.* 1958). This revolutionised the cultivation of oyster mushrooms towards growing them in containers. Most substrates used for the cultivation of the oyster mushroom consist of a high carbon source, basically almost all the available lignocellulosic material (Dundar *et al.* 2009). This is because, the mushroom produces a broad spectrum of lignocellulolytic enzymes which are capable of digesting cellulose, hemicellulose and lignin, which form the bulk of most plant cell walls (Dashtban 2009). Various agricultural wastes have been used as substrates for the cultivation of the *Pleurotus sajor−caju* mushroom. Some of these include rice straw and wheat straw, mostly used in Asia and Europe, respectively.

In South East Asian countries sawdust is more commonly used as substrate. Gregori *et al.* (2007) stated that *P. sajor−caju* could be cultivated on wheat straw and bran substrates containing olive mill waste and wastewaters (OMWW). Other substrates like banana leaves, grass (*Saccharum munja*), coconut coir, coffee pulp, corn cobs, corn leaves, crushed bagasse, molasses waste, mango fruits and seeds, peanut hull, sugarcane leaves, water hyacinth, wheat straw and brassica strawwood chips have also been used (Cangy and Peerally 1995; Buah *et al.* 2010). During substrate preparation, addition of supplements like urea, ammonium sulphate, ammonium nitrate as well as calcium carbonate became quite common to maintain a pH favourable to fungus growth. These additives avoid the increase in acidity caused by fungal metabolism. Additionally, increased substrate degradation and mushroom yield were also reported. Other supplements include bran from rice, wheat grain, oil seed cakes cornmeal and cottonseed meal added to a ratio of approximately 1 to 4 (Mane *et al.* 2007).

Substrate formulation for *Pleurotus sajor−caju* mushroom cultivation in Mauritius

The present local substrate for growing *P. sajor−caju* mushroom is fundamentally, bagasse as substrate, maize seeds as nutrient source and calcium carbonate as supplement. At the start, bagasse is allowed to ferment for two to three days. After this period, it is moistened until the 70% water content is achieved. The maize seeds and calcium carbonate supplement are then mixed with the substrate until a homogeneous mixture is obtained. Polypropylene bags are filled with the resultant mixture to make a net weight of 750 or 3000 g, respectively. The opening of each bag is fitted with a polyvinylchloride (PVC) ring (50 mm in diameter) and plugged with a cubed sponge (50 mm in length). The bags are then heat treated in an iron vessel and allowed to cool to room temperature. They are then inoculated with the mycelium spawn in a sterile area, and then disposed horizontally on wooden or metal shelves to allow substrate colonization. Incubation in total darkness is recommended with a relative humidity of 90 to 100%. Spawn run takes approximately two to three weeks at temperatures varying from 20 to 29°C. After complete colonisation, the sponge is removed and the PVC ring is replaced with another one but with a diameter of 10 mm. The bags are left open to moisten the substrate on a regular basis. Misting is normally carried out twice or thrice a day, depending on ambient temperature. The relative humidity of the incubation room should be kept at 80 to 90% (Futtee, pers. comm., Agricultural Research & Extension Unit (AREU)). Primordia formation will occur in three to five days. The optimal water content of the *Pleurotus* substrate is, as a rule, adjusted to 70% (wet weight) because this water level favours loss of organic matter, degradation of lignin and release of water−soluble substances and therefore easily made available for the mushroom mycelium. According to Choudhary *et al.* (2009), water holding capacity (WHC) appears to be directly related to porosity and bulk density. For instance, low substrate porosity results in degradation occurring only at the substrate outer surface (Prabhakar *et al.* 2005). This is because compaction affects porosity (in terms of FAS) and bulk density is a reflection of sample compaction (Annan and White 1998). Hence, increasing substrate compaction will decrease water holding capacity thus affecting the heat and mass transfer rates in the substrate. Basidiomycetes group of fungi require almost

neutral pH medium for their growth. In terms of microbial activity, the substrate should be properly pasteurised at 60 to 70°C (by immersion in hot water or steam injection) for a minimum of 30 min to 8 h. On the other hand, the substrates can be sterilised in an autoclave at 1 atm, 121°C, for 60 to 90 min to prevent the thriving of competing organisms, generally green moulds (*Trichodema* spp.), which can hinder mycelium development and fruit body formation. However, Moda *et al.* (2005) reported higher contamination rates in pasteurised substrate compared to the washed substrate.

Alternative substrates for oyster mushroom cultivation

Various studies demonstrate the use of composted lignocellulosic wastes, such as cassava peels, corn cobs, fish waste, chopped office papers, cardboard, pulp mill sludge and sawdust as well as plant fibre, such as cotton waste and coconut residue, for growing oyster mushrooms (Buah *et al.* 2010). However, the composting cycle is quite short, from 1-28 days. Normally, sterilisation, prior to inoculation, and the incubation method and period, are near to analogous as to when using the non-composted substrate. Hernández *et al.* (2003) found, by composting a mixture composed of coffee pulp and *Digitaria decumbens*, this improved the Biological Efficiency (BE) of *Pleurotus ostreatus* cultivation. Nevertheless, since heat is generated in compost systems through microbial cleavage of the carbon bonds and loss of organic matter, this means less food available for oyster mushrooms. A substrate's physical properties play an important role in every stage of composting from dictating initial aeration and temperature regimes to determining the degree of handling and utilisation of the end product. Several laccases and lignolytic enzymes are secreted by *P. sajor-caju* which helps in bioremediation processes and substrate degradation. For this reason, oyster mushrooms have been used for heavy metal removal and therefore mycoremediation (Chiu *et al.* 2009; Ferreira *et al.* 2011). Pozdnyakova *et al.* (2011) believe that mushrooms could actually remediate to the aftermath of the British Petroleum (BP) oil spill. This property of mushrooms should be carefully evaluated since they have the ability of growing on toxic components. Therefore, any waste selected as substrate should be monitored for heavy metal contents and other contaminants. An approach towards substrate quality, particularly if compost is to be used for mushroom production intended for human consumption, should be well scrutinised.

Composting process and its product, compost

The essence of composting fundamentally lies in the biodegradation of solid organic substrates, by the microbial community, under aerobic conditions. In the process, energy is released exothermically in the form of heat. Composting begins when the available nutrients found in the organic substrate are gradually consumed by the microorganisms. Since they respire aerobically, the organisms release a large amount of heat, resulting in rapid temperature rise under ideal conditions. In parallel, carbon dioxide and water are released as waste products. The enzymatic activity rates generally double with each 10°C rise in temperature.

The composting process is brought about by successive microbial communities. They can be further classified according to the temperature ranges of their activity (Ben *et al.* 2007). Accordingly, under optimal conditions, composting proceeds from the psychrophilic state through three phases, the mesophilic phase, the thermophilic phase and the cooling and maturation phases (Insam and de Bertoldi 2007; TEG Group 2007). However, the shape of the curve varies with the feedstock being composted and the composting method.

In addition to carbon dioxide, ammonia is generally the main volatile compound produced during the process. The ammonia released can further be oxidised to nitrate compounds by nitrifying bacteria. Though it is assumed that compost temperature should not go beyond 65°C during this stage, Beffa *et al.* (1998) found that thermohygienization can be achieved at temperatures between 65 to 75°C and not exceeding 80°C. Depending essentially on substrate availability, the time−span of the high−rate phase ranges from a few days to several months whereafter thermophilic activity as well as temperature decrease due to a reduction in the availability of nutrients.

Irrespective of the composting system, composts are made from a changing assemblage of organic feedstocks, variations in physical, chemical and biological properties are inevitable. The general physical and chemical characteristics of most composts obtained from several compost standard bodies and literatures (Hassen *et al.* 2001; Basnayake 2003; USCC 2010) are summarised in **Table 2**.

Even so, the specific characteristics of a compost determine how, and in which applications it can be optimally utilised. For that reason, compost quality must be set in accordance to its end use to achieve specific analytical and product performance goals.

Table 2 General physicochemical characteristics of most composts.

Parameter	Range
C/N ratio	10:1 – 35:1
Moisture Content (%)	40 – 60
Wet Bulk Density (kg/m^3)	475 – 950
Particle Size (mm)	9.5 – 25
Water Holding Capacity (%)	75 – 200
Porosity (%)	40 – 60
pH	6.0 – 8.0
EC (dS/m)	2.5 – 4.0
Volatile Solids Content (%)	54 – 75

MATERIALS AND METHODS

Reactor design

The reactor design was obtained from Mohee and Mudhoo (2005) and Mudhoo and Mohee (2008) and was modified by connecting the reactor to a centrifugal fan by means of two PVC shoulders and a PVC T-connection, both having a diameter of 5 cm (Mihilall *et al.* 2010). This pilot-scale batch composter (200L) was designed of PVC plastic with a thickness of 4 mm, an internal diameter of 550 mm and a length of 880 mm. Two adjacent holes were made in both sides of the drum through which two PVC pipes of 1 mm thickness and 50 mm internal diameter were passed. The pipes were perforated at about 20 mm intervals along the upper circumference and 40 mm intervals along the length of the pipes with holes 5 mm in diameter, and allowed the diffusion of air through the compost mixture thus ensuring aerobic conditions. The upper side of the drum was perforated with 3 holes of 50 mm diameter and spaced at 220 mm along the length of the drum. They allowed temperature measurements to be effected and allowed free exchange of air between the compost pile and the atmosphere.

Selection of feedstock

Based on their potential availability, crude fibre content and quality, four potential waste feedstocks were selected to produce the compost substrate. **Table 3** shows their physicochemical characteristics.

Aeration requirements for composting

The stoichiometric oxygen requirement can be determined from the chemical composition of the organic solids and the extent of degradation during composting. The chemical composition of banana leaves, mixed cardboard wastes, green grass (*Stenotaphrum dimidiatum*) and chemically untreated pinewood shavings have been determined using the Perkin-Elmer 2400 Series Carbon-Hydrogen-Nitrogen-Sulphur (CHNS) analyser. **Table 4** shows the average chemical composition of the potential composting feed-

Table 3 Physicochemical characteristics of the potential composting feedstocks.

	Banana leaves	Mixed cardboard waste	Green grass (*Stenotaphrum dimidiatum*)	Untreated pinewood shavings
Wet bulk density (kg/m^3)	812.3 ± 2.5	219.3 ± 18.6	142.6 ± 5.7	54.4 ± 1.4
Moisture content (%)	84.3 ± 0.2	7.1 ± 0.6	83.8 ± 0.1	7.4 ± 0.5
Total dry solids (%)	15.7 ± 0.2	92.9 ± 0.6	16.2 ± 0.1	92.6 ± 0.5
Total volatile solids (%)	89.5 ± 0.2	92.4 ± 0.3	87.7 ± 0.4	98.6 ± 0.1
Ash content (%)	10.5 ± 0.2	7.6 ± 0.3	12.3 ± 0.4	1.4 ± 0.1
Total carbon content (%)	49.7 ± 0.1	51.3 ± 0.2	48.7 ± 0.2	54.8 ± 0.1
Crude fibre content (%)	28.2 ± 1.8	35.7 ± 0.5	29.7 ± 1.8	39.3 ± 0.1

Table 4 Average chemical composition of the potential composting feedstocks.

Substrate	Element (% dry mass)					
	Carbon	Hydrogen	Oxygen	Nitrogen	Sulphur	Ash
Banana leaves	42.05	2.33	53.17	1.33	0.04	1.08
Mixed cardboard wastes	41.42	1.56	55.93	0.15	0.01	0.93
Green grass	39.94	2.37	54.96	1.84	0.02	0.88
Untreated pinewood shavings	46.40	5.10	46.14	0.29	0.08	1.99

Table 5 Relative quantities of the different feedstocks to be composted.

Substrate	Relative quantities (kg)
Green banana leaves	18.0
Mixed cardboard waste	15.0
Green grass	18.0
Untreated pinewood shavings	4.0
Water added	6.0

stocks. The centrifugal fan was set to blow air at 540 L/h in the reactor each time the temperature decreased during the composting process. **Table 5** shows the substrates and their relative amounts used for composting.

Substrate preparation

The preparation of the feedstock was carried out in the composting facility of the University campus. Green banana leaves, mixed cardboard waste and green grass were shredded to an average size of 5 cm in length. After weighing each feedstock, they were all spread and blended on the floor using a rake and a shovel. The resulting mixture was then filled in the reactor.

Temperature monitoring

The temperature of the compost matrix was recorded using a 1m−thermocouple (*CheckTemp1*, ± 0.1°C) at three different openings namely, T_1, T_2 and T_3 along the surface of the pile. At each point, the probe was slowly inserted across different depths of the pile and the maximum temperature was recorded when the reading stabilised on the screen.

Determination of moisture content

70 g to 100 g of the fresh unscreened sample was preheated in a forced-convection oven at 70 ± 5°C for 24 h, or until constant weight. Masses were recorded to the nearest 0.1 g on a Mettler PM3000 top balance. The moisture content was determined by the difference in net dry weight and initial as-received sample weight. The resultant was then divided by the wet mass of the subsequent sample.

Determination of wet bulk density

A 1000 mL beaker was weighed (W_a) and the sample material was gradually added up to the edge of the vessel. The final weight was then taken down (W_b). A Mettler PM3000 top balance was used to record masses to the nearest 0.1 g. The bulk density was calculated by dividing the mass of the compost sample to the volume of the vessel.

Determination of particle size distribution

A Ro-tap testing sieve shaker was used to determine the particle sizes of the mix. 25 g of substrate was weighed and placed on the sieve having the largest mesh size. A total of 7 sieves having mesh sizes (in mm) 26.67, 18.85, 13.33, 6.680, 3.327, 4.699, 1.651 were used. At the bottom of the last sieve, a final metal pan was placed to collect any particle less than 1 mm. The Ro-Tap testing sieve shaker was timed for 10 minutes, after which, the sieves and pan were re-weighed on a Mettler PM3000 top balance.

Determination of porosity

A 2000-mL graduated plastic beaker was modified by drilling four 3-mm holes at the base. Each hole was then covered temporarily with masking tape. 600 cm^3 of sample (of known dry weight) was filled by free-falling in the beaker. After three free-fall drops, more sample was added until the level reached the 1800 mL mark. Known volumes of deionised water were carefully added to the sample up to saturation, over defined time periods. The beaker was then held over a receiving vessel and the masking tape was removed to allow excess water to drain in the recipient. The drain holes were taped again and the collected water was added once more to the sample. This operation was repeated three times. The total volume of the water used to saturate the beaker was also recorded. The Pore Space Volume (PSV) was calculated using the empirically calculated Equation 1:

PSV = (W1800WS, nodrain) – O1800 + (1800−V1800WS, nodrain)

where W1800WS, nodrain (g) = Mass of (beaker + water saturated 1800 cm^3-sample) – Initial mass of beaker, O1800 (g) = (Mass of as-received 1800 cm^3-sample) × (Mass of dried 50 cm^3-sample ÷ Mass of as-received 50 cm^3-sample), and V1800WS, nodrain (g) = Total volume of water used to saturate sample.

Porosity was then determined using the PSV value and Equation 2:

$$Porosity(\%) = \frac{PSV}{1800} \times 100$$

Determination of water holding capacity

Water holding capacity was determined using a modified glass cylinder (height: 120 mm; internal diameter: 35.7 mm) having a closed-meshed plastic net bottom. The sieve base was covered up with moistened filter paper and held in place using a rubber band. The assembly was weighed (M_o) on a Mettler PM3000 top balance. A fresh substrate sample was filled into the cylinder with light shaking and the new mass (M_c) was recorded. The cylinder containing the substrate was disposed into a 2000-mL plastic beaker which was slowly filled with tap water until the sample could soak itself full from the bottom. The substrate was over damped by approximately 1cm. The resultant coating was then allowed to stand

for 24 h after which it was removed out of the water, dried from the outside and placed on a saturated cellulose base covered with a watch glass. After 2 h, the glass cylinder was re−weighed (M_{moist}) until a constant mass was recorded. In parallel with this experiment, the water content (W_c) of the naturally moist sample was determined. The maximum water holding capacity (WK_{max}) was determined using Equation 3:

$$WK_{max} = \frac{E_{moist} - E_{dry}}{E_{dry}}$$

where E_{moist} = mass of wet sample (g) = $M_{moist} - M_o$, E_{dry} = mass of dry sample (g) = $(M_c - M_o)(1 - 0.01W_c)$, M_c = mass of cylinder (g) + mass of wet filter paper (g) + mass of weighed-in sample (g), M_{moist} = mass of cylinder (g) + mass of wet filter paper (g) + mass of wet sample (g), M_o = mass of cylinder (g) + mass of wet filter paper (g), and W_c = water content of fresh sample (%) (Mohee and Mudhoo 2005).

Volatile solids content

5.0 g of oven-dried ground sample material was weighed in a pre-weighed dry porcelain crucible and burnt up at 550°C in a muffle furnace for about 2 h (BS1377 method). The weights were recorded to the nearest 0.001 g on a Mettler PM400 top balance. The volatile solid of each sample was determined by Equation 4 (Mohee and Mudhoo 2005):

$$VolatileSolids, VS(\%) = \frac{M_{dry} - M_{burnt}}{M_{dry} - M_{crucible}} \times 100$$

where M_{dry} = mass of sample and crucible before burning (g), M_{burnt} = mass of sample and crucible after burning (g) and $M_{crucible}$ = mass of empty crucible (g).

Determination of the pH value

200 mL of 0.01 mol/L of calcium chloride solution was added to 20 g of the consistently ground oven-dried sample in a clean dry glass beaker. The mixture was gently stirred with a clean glass rod. After 1 h, the electrode of the calibrated *EcoScan* pH meter (buffer 4, 7 and 10) was immersed into the filtered extract of the mixture. The sample cup was carefully swirled without lifting. pH was recorded when there was no further change in the meter reading.

Determination of the electrical conductivity

200 mL of deionized water was added to 20 g of the consistently ground oven-dried sample in a clean dry glass beaker. The mixture was continuously stirred on a magnetic stirrer for about 2 h. The electrical conductivity was determined over the collected filtrate using a calibrated electrical conductivity meter (Model Lutron pH−201 Hand Held Digital pH meter).

Determination of heavy metal content

The heavy metals content for lead, cadmium and zinc were determined using the Atomic Absorption Spectrophotometer (AAS). The experiment was carried out at the Heavy Metal Analysis Section of the Chemco Laboratory, Chemco Company Limited Mauritius. Reference for the experimental procedures was made in The *Standard Operating Procedure (SOP) for Metal Analysis by AAS SOP MLD 005, California Air Resources Board, Inorganics Laboratory Section – October 2003, Revision 6.0.*

Determination of compost stability

The respiration rate was measured as per the experimental method described by Trautmann and Krasny (1997) and Mohee *et al.* (2008) with a working temperature of 25°C. The experiment consists of measuring the level of carbon dioxide in a compost sample. Three 500-mL jars were filled with 25.0 g of compost sample, with known moisture content. 20.0 ml of 1.0 mol/L potassium hydro-xide was pipetted and transferred into a 25-mL beaker which was carefully placed inside each jar using a malleable metallic plastic-coated wire. The opening of each jar was tightly sealed with a rubber bung. A fourth jar, of the same volume, was set up as control without the compost sample. After 24 h, each beaker was removed and titrated with 1M standardised hydrochloric acid using phenolphthalein indicator. A fresh sample of potassium hydroxide was replaced in each jar on a daily basis. The same exercise was repeated four consecutive days per week.

The mass of carbon dioxide produced was calculated using the Equation 5 (Mohee *et al.* 2008; Mihilall *et al.* 2010):

$$CO2.C(mg) = HCl_b - HCl_s \times [HCl(mol/L)] \times 12gC/mol$$

where HCl_b = volume of HCl used in titration for blank, mL, and HCl_s = volume of HCl used in titration for jars containing sample, mL.

Determination of total nitrogen (Kjeldahl method)

Ground oven-dried sample was weighed in a Mettler PM400 top balance. The sample was then transferred into a boiling tube by washing it with 15 ml concentrated sulphuric acid. A Kjeldahl tablet was then added to the mixture which was heat treated at 360°C. The resultant bright clear sample was allowed to cool at room temperature and then made up to the mark with distilled water. 2.0 mL of the mixture was pipetted and transferred into a distillation vessel followed by 5.0 mL soda lye. Ammonia was collected in an acid receiver containing 10.0 mL boric acid. After the distillation process, a few drops of indicator is added to the acid and then titrated against 0.01 mol/L hydrochloric acid.

Determination of available phosphorus by modified Truog method

Using the method of Puchooa *et al.* (1999), 5.0 g of consistently ground oven-dried sample was weighed in a clean and dry pre-weighed plastic bottle to which 50.0 ml of 0.1 mol/L sulphuric acid was added. The container was safely secured with its lid and agitated on a mechanical shaker for 1 h. The resultant mixture was then filtered and the filtrate was collected for phosphorus determination. To a 100-mL volumetric flask, 80.0 ml of distilled water was added followed by 8.0 ml vanadium ammonium molybdate as the chromogenic reagent, 8.0 ml ascorbic acid solution and 5.0 mL of the sample filtrate made up to the mark. Similarly, a series of standard phosphorus solutions were prepared from 0 to 20 mg/L in 100-mL volumetric flasks. Each flask was allowed to stand for 30 min for full colour development. The absorbance of each solution was then measured in a 10 mm optical cell using a spectrophotometer at 660 nm.

Determination of exchangeable potassium

5.0 g of homogeneously ground oven-dried sample was weighed in a clean and dry pre-weighed plastic bottle to which 50.0 mL of ammonium acetate was added (Dewis and Freitas 1970; Mohee *et al.* 2008). The container was safely secured with its lid and agitated on a mechanical shaker for 1 h. The resultant mixture was then filtered with the filtrate collected for potassium determination. To a 500-mL volumetric flask, 0.954 g of dry potassium chloride was added followed by 1 mL concentrated hydrochloric acid and made up to the mark with distilled water. The flask was capped and gently inverted two to three times to homogenise the solution. A series of five potassium standards were prepared into 100-mL volumetric flasks. The absorbance of the sample filtrate was then measured in a 10 mm optical cell using a calibrated flame photometer.

Determination of crude fibre

2.0 g of consistently ground oven-dried sample was weighed into a dry crucible (adapted from Madsen 1973). The sample was then heated under reflux with 150.0 mL of 0.128 mol/L sulphuric acid in a 600-mL beaker for 30 min. The sample was rinsed three times with 50.0 ml aliquots of hot water and then filtered. The residue was retained and transferred to the 600-mL beaker to be heated under reflux with 150.0 mL of 0.128 mol/L potassium hydroxide for 30 min. The sample was then washed with 50.0 ml aliquots hot water and then filtered once again. The residue thus obtained was retained and washed with 10 mL acetone. The resultant blend was

then transferred into a pre-weighed crucible and dried at 70°C overnight in a forced-convection oven. The following day, the dried sample was allowed to equilibrate in a dessicator and afterwards ashed in a muffle furnace at 550°C for 2 h. The crucible was cooled in a dessicator and then weighed. Crude fibre content was expressed as percentage loss in weight on ignition using the following empirically derived Equation 6:

$$\%Fibre = \frac{b-c}{\alpha} \times 100$$

where α = mass of the sample (g), b = mass of the sample before ashing (g), and c = mass of the sample after ashing (g).

Inoculum preparation

Pure mycelial culture of *Pleurotus sajor–caju* was maintained on potato dextrose agar (PDA). For stock culture production, millet grains were used (Futtee, pers. comm., AREU).

Spawn preparation

Spawn was prepared by inoculating polypropylene bags containing boiled maize seeds mixed with 4% (w/w) calcium carbonate with the stock culture.

Cultivation method

The potential 23-day composted substrate and bagasse were both weighed to 750 g and blended with 10% calcium carbonate and 10% crushed maize on a dry weight basis (adapted from Akavia *et al.* 2009; Hassan *et al.* 2010). The moisture content was adjusted to 70%. Each substrate was then packed in heat resistant polypropylene bags. The opening of each bag was fitted with a polyvinyl (PVC) ring (50 mm length and 53 mm diameter) and plugged with a piece of cubed sponge (50 mm in length). Five bags were prepared for each material. These were pasteurised at 70°C for 4 h in a designed steel drum with a gas burner as heating device and were afterwards allowed to cool gradually to ambient temperature. Each bag was inoculated with the *CC 114* strain spawn at the rate of 5% dry weight of the substrate. Immediately after, the sacks were transported to the growing room and randomly deposited horizontally, 20 cm apart, on galvanised metal shelves (disinfected by 5% domestic bleach). Incubation was carried out in complete darkness where bags were covered with black plastic sheets. The incubation temperature ranged between 23 and 27°C. The growing room was equipped with a table fan, which was switched on when the temperature of the room exceeded 30°C, and a fluorescent ceiling light (40 W, cool white). However, on complete colonisation of the bags, by the mushroom mycelium, the black plastic sheet, the PVC rings and sponge plugs were removed. Subsequently, only a PVC ring (20 mm length and 53 mm diameter) was replaced at the opening of each bag. The exposed surface was hand sprayed with tap water once or twice a day to maintain moisture in the bags at 75-80%. The biological efficiency (B.E.) (yield of mushroom per kg substrate on dry weight basis) was determined by the following formula:

$$B.E.(\%) = \frac{FibreWeight_{mushroom}}{DryWeight_{substrate}} \times 100.$$

RESULTS AND DISCUSSION

Temperature

The average temperature monitored over the 23 days, was 46.2°C with a maximum of 56.5°C on day 8 (**Fig. 1**). Daily temperature monitoring from the start of the experiment until stabilisation near ambient values, which averaged 25.8°C, showed a very gradual rise. This condition is attributed to the initial high carbon-to-nitrogen ratio (72.19:1) of the feedstock. The airflow was kept at 0.009 ± 0.0004 m^3 air min^{-1} (540 ± 24 L/h). The temperature remained above 55°C for 6 days (Finstein *et al.* 1986) hence reaching thermophilic conditions and meeting the "Process to Further Reduce Pathogens" under the USEPA regulations whose minimal temperature requirement is 55°C or higher for three consecutive days (USEPA, 40 CFR Part 503, 1994).

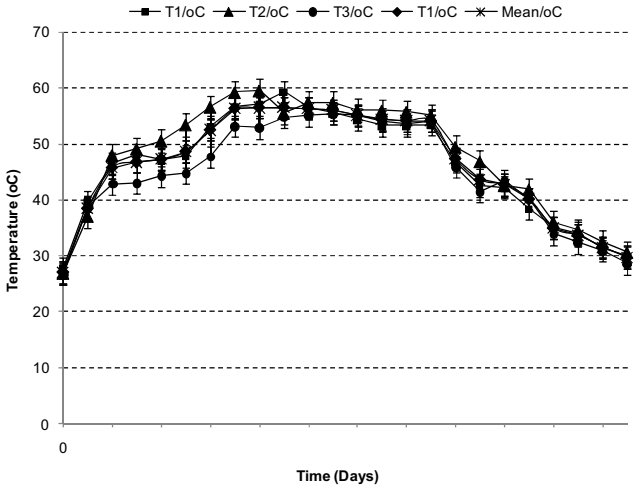

Fig. 1 Average temperature variations for the BGCW mix.

Moisture content

The initial average moisture content of the feedstock was 71.4%. As the composting process continued, moisture content declined to 66.1, 62.4 and 57.7% on days 7, 14 and 21 respectively, principally due to evaporative cooling (Ghaly *et al.* 2006). Most literatures cite a starting moisture content of 40 to 60%. High moisture contents displace air in the micro pores of the feedstock material thus preventing oxygen transfer in the compost mass ensuing anaerobic conditions and excess leachate. However, the moisture percentage range can be extended to 70%, having maximal microbial activity, with respect to the structural strength of the substrates to be composted, basically for those having a low bulk density and high moisture absorbing capacity (Monson and Murugappan 2010). The cardboard and pinewood shavings were dry during compost preparation and hence exhibited the above-mentioned characteristics while also acting as bulking agent, favouring adequate free air space at the initial stage of the composting process.

Total dry solids

The amount of solids remaining after a known volume of compost sample has been oven-dried at steady temperatures, to constant mass is known as total dry solids. This fraction consists of fixed solids, biodegradable volatile solids, and volatile solids not readily biodegradable (USCC 2001). At the start of the process, the average percentage of total dry solids of the BGCW mix was 28.6%. The value then increased gradually to 33.9, 37.6, and 42.3% on day 7, 14 and 21, respectively with a comparative decrease in moisture content over the same period of time. Most literatures mention total solids content for finished composts within the range of 40 to 60% (PennState 2009).

Wet bulk density

The average initial wet bulk density of the starting material was 87.4 kg/m^3. However, on day 7, 14 and 21, bulk density increased to 125.8, 165.6 and 186.0 kg/m^3, respectively. According to Breitenbeck and Schellinger (2004), Iyengar and Bhave (2005) and Larney *et al.* (2007), bulk density should normally increase with composting time, based on a decrease in feedstock physical structure and hence a mass diminution usually accompanied by more pronounced volume reductions. As a result, the wet bulk density influences the mechanical properties such as strength, porosity, and ease of compaction (Agnew and Leonard 2003).

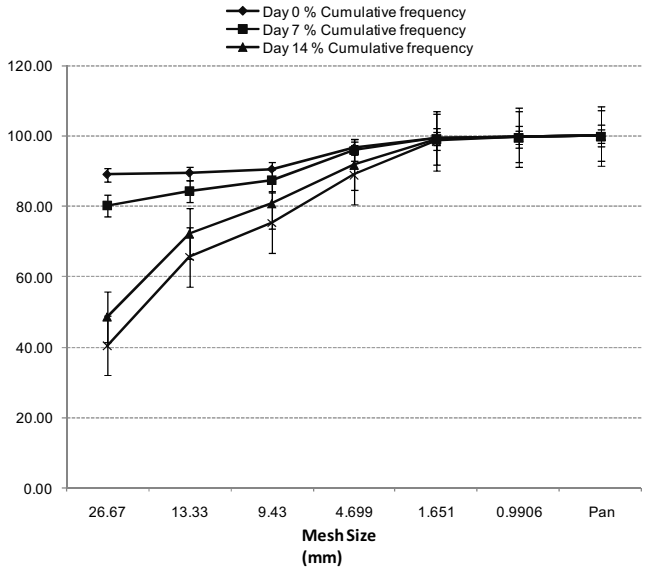

Fig. 2 Cumulative particle size distribution for the BGCW mix.

Particle size distribution

Particle size influences porosity, resistance to compaction and overall mass, in addition to defining microbial surface attachment and carbon availability (Agnew and Leonard 2003). The feedstock particle size range varied between 26.67 and 4.699 mm from day 1 to day 21 with gradual shifting in the percentage cumulative frequency of the mass retained (**Fig. 2**). Gray and Biddlestone (1973) propose a particle size of 13 mm to 76 mm for forced aeration coinciding with the initial feedstock particle size, 50 mm. On day 0, the total percentage of BGCW compost which could pass the sieves having a 26.67 and 13.33 mm mesh sizes was 21.6% of the total mass. However, this percentage increased to 35.3, 79.1 and 94% on days 7, 14 and 21, respectively. The same increasing trend in percentage could be observed for the materials which could pass the 9.43 mm and 4.699 mm sieves. The percentages were 12.9, 16.6, 27.4 and 35.7% on days 0, 7, 14 and 21, correspondingly. Final composts normally have particle sizes ranging from 3.2 to 50.8 mm (Baldwin and Greenfield 2009; USCC 2010).

Porosity

Porosity of the composting mass is a critical factor in forced aeration. Porosity determines airflow resistance and is related to overall particle size and variation in particle sizes of the materials. The larger the particles, the slower the degradation due to the smaller surface area to mass ratio. Hence, feedstock material is usually shredded to a minimum length of 5 cm to ensure the correct porosity. The average porosity on day 0 was 68.35%. This value de-creased to 61.66 and 60.38% on days 7 and 14 to finally reach 53.18% on day 21. Compost guidance recommends 30 to 60% air volume (Annan and White 1998; Ruggieri *et al.* 2008). Furthermore, bulking agents like wood shavings have been added as substrate and also to increase the porosity and integrity of the organic solid matrix (Monson and Murugappan 2010). Additionally, moisture also affects porosity and gas diffusivity and is removed via vaporization (evaporative cooling), as driven by microbial heat generation.

For this reason, it is important that the moisture content be such that the voids are free of water. Since the porosity of the composting matrix was within the required range, despite the initial moisture content of 71.4%, it strongly supports that the in-vessel composting process followed the same standard decomposition trend. This condition may be difficult to maintain in a passive pile and will depend on a number of factors like residence time, pile height and material moisture content.

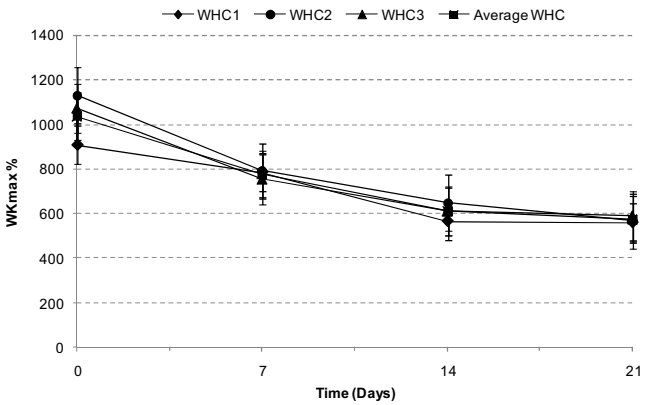

Fig. 3 Water holding capacity of the BGCW mix.

Water holding capacity

Water holding capacity is the ability of the compost to hold water under varying tensions and thus translates to potential reductions in irrigation amount and frequency. The initial water holding capacity was 1035.80% but decreased to 777.02, 608.58 and 572.77% on day 7, 14 and 21, respectively (**Fig. 3**). Water holding capacity diminishes during biodegradation, due to loss of organic content, and thus the ideal level of moisture will likewise diminish (Brinton 2000). Most composts produced from municipal feedstocks possess a water holding capacity of 75 to 200% of their dry weight (Hargreaves *et al.* 2008).

Volatile solids content

Organic matter represents the combustible content or "volatile solids" (VS) and is typically reported in terms of total weight loss (on dry basis) on ignition as VS or OM (Brinton 2000). Specifically, through composting, 20 to 30% of VS in organic waste are biologically degraded to carbon dioxide and small concentrations of other gases, water and heat causing a gradual decrease in percentage. On day 0, the average total volatile solids content was 81.6% (dry matter basis). With time, a decrease in the average percentage was noted, 81.0, 79.6 and 78.7% on days 7, 14 and 21, correspondingly. According to USCC (2010), there is no ideal organic matter content for compost, and it may vary widely, ranging from 30 to 70%.

Ash content

The ash content measures the inorganic residual material left after burning the oven−dried compost sample. The initial average ash content was 18.41%. This value increased to 19.04, 20.45 and 21.33% on days 7, 14 and 21, respectively. As the decomposition progresses, the weight loss and increase in ash content will cause the bulk density to increase and porosity to decrease (Jolanun and Towprayoon 2010). Most composts contain approximately 50% ash (dry weight basis) (USCC 2010).

pH

pH is a measure of the concentration of hydrogen ions in a solution. The pH level of the composting mass normally varies with time. From day 0 to day 7, pH changed from 7.01 to 7.83 (**Fig. 4**). After this period, a decrease in pH, 7.72, was noted, most probably due to the synthesis of organic acids and phenolic compounds in anaerobic zones. Though acidic pH is generally detrimental to aerobic microorganisms, the composting process continued. Consequently, after day 14, a subsequent rise in pH, 7.94, was noted. This coincided with the appearance of the fungi of the Genus *Coprinus* on the composted substrate, strongly implying the utilisation of the acids, in the substrate, by the mushrooms

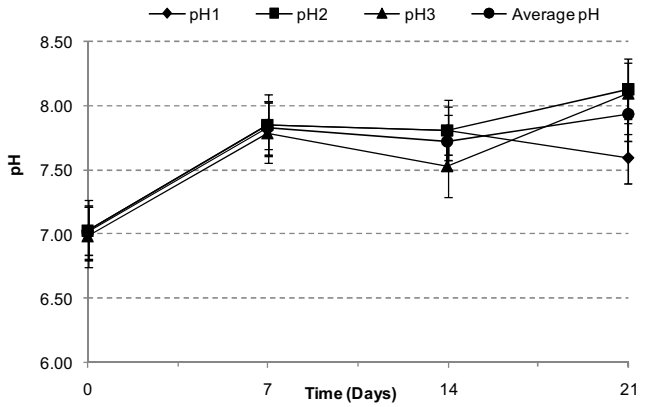

Fig. 4 pH of the BGCW mix.

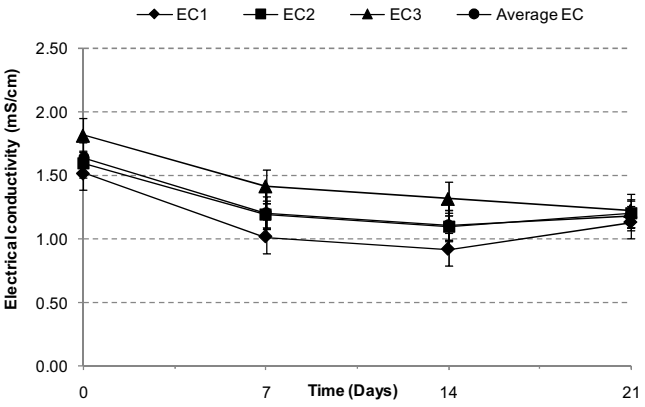

Fig. 5 Electrical conductivity of the BGCW mix.

(Mihilall *et al.* 2010). Final composts normally have a pH ranging between 6.0 and 8.0 (USCC 2010). Yet, the ideal pH depends on compost use (PennState 2009).

Electrical conductivity

The average EC of the substrate was 1.64 dS/m on day 1. As the composting process continued, the EC decreased (**Fig. 5**). Accordingly, on days 7 and 14, the values were 1.21 and 1.11 dS/m. Conversely, on day 21, the EC increased to 1.18 dS/m. The preliminary decrease in EC may be attributed to the utilisation of the salts (mainly sodium, potassium, chloride, nitrate, sulphate and ammonia) during the biodegradation process. The increase in EC on day 21 can be explained by the organic matter loss which suggests an increase of mineral cation concentration not attenuated by salts leaching or by binding to stable organic complex (Francou *et al.* 2005). According to USCC (2010), most composts have a soluble salt conductivity of 1.0 to 10.0 dS/m. Even so, the preferred range for composts depends on their end-use (Soumaré *et al.* 2002).

Heavy metal content

The heavy metal contents for lead, cadmium and zinc were below the detection limits, 0.01 and 0.002 mg/L, respectively. These values were well below the levels specified by the Mauritius Standards (Brinton 2000; Commission of the European Communities 2008; USCC 2010).

Compost stability

The stability of composts can be defined as the degree to which the organic fractions in composts have been stabilised with low microbial activity. Hence, the metabolic quotient of the microbial biomass can be used as a measure of microbial efficiency. On day 1, the quantity of carbon dioxide generated was 11.66 mg CO_2.C/day/gVS (**Fig. 6**).

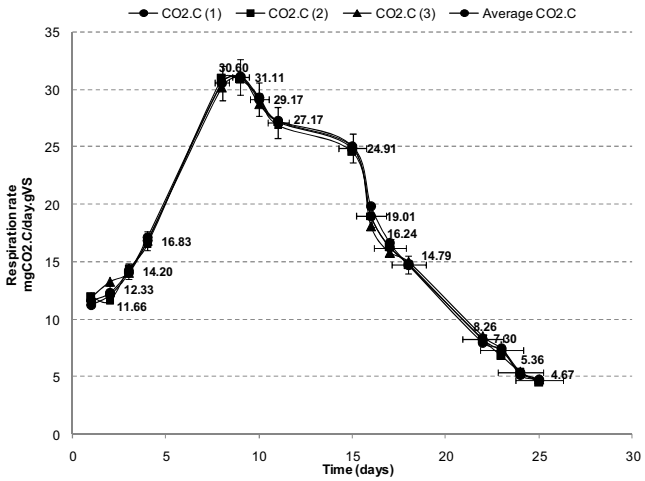

Fig. 6 Evolution of respiration rate with time for the BGCW mix.

This value gradually rose until day 9 where the maximum gas evolution (31.11 mg CO_2.C/day/gVS) was recorded, thus coinciding with the maximum temperature (56.5°C) exhibited on the same day.

This strongly affirms that the process has switched from the mesophilic to the thermophilic phase. As the composting process continued on days 10 to 15, carbon dioxide evolution progressively decreased. Nevertheless, on day 16, there was a sharp decline of 19.01 mg CO_2.C/day/gVS. This swift drop may be explained by a diminution of the thermophilic activity due to the depletion of substrates in the feedstock. This could also suggest the start of the cooling and consequently maturation phases confirmed by the appearance of the fungi of the Genus *Coprinus* coupled with subsequent decline in temperature 47.3°C on the same day. On day 23, the carbon dioxide evolution reached 7.30 mg CO_2.C/day/gVS. According to the Brinton (2000) and California Compost Quality Council Standards (CCQC) (2005), 2 to 8 mg CO_2.C/day/gVS correspond to 'stable compost' and is usually suitable for seeds germination growth and development.

Nutrient content of BGCW as compared to bagasse

The nutrient content for the BGCW compost was 1.72% for nitrogen, 0.11% for phosphorus and 1.36% for potassium, totalising to a sum of 3.19 %. According to USCC (2010), these values are within the range acceptable for a standardised compost and therefore indicate an average nutrient content. However, it is interesting to note that bagasse was very poor for all three nutrients, 0.001% for nitrogen, 0.003% for phosphorus and 0.006% for potassium, suggesting that the oyster mushroom taps its nutrients from the fibre content of the substrate.

Physicochemical characteristics of BGCW compost and bagasse substrates

Table 6 compares the physicochemical characteristics of the BGCW compost substrate to that of the bagasse substrate.

The moisture contents for both BGCW compost and bagasse were nearly the same, 57.7 and 58.1%, respectively. In most of cases, bagasse has a moisture content between 45 and 55% by weight (EFIG 1997). Physical structure decreases and bulk density increases during the decomposition process. This could explain as to why the bulk density of the BGCW compost (186.0 kg/m^3) exceeded that of bagasse (115.2 kg/m^3). The wet bulk density of bagasse ranges from 120 to 150 kg/m^3. Substrates with high bulk densities are more compact due to a greater difference between mass and volume reduction. This results in low air capacity, low water infiltration and water holding capacity. However, it is

Table 6 Physicochemical characteristics of BGCW compost and bagasse substrates.

Parameter	BGCW compost	Bagasse
Moisture content (%)	57.7 ± 3.1	58.1 ± 0.1
Wet bulk density (kg/m^3)	186.0 ± 1.9	115.2 ± 1.7
Particle size (mm)	4.7 - 9.4	0.50 - 50.0
Water holding capacity (%)	572.7 ± 16.5	188.0 ± 3.8
Porosity (%)	53.2 ± 0.4	92.3 ± 0.6
pH	7.94 ± 0.3	5.6 ± 0.05
EC (dS/m)	1.18 ± 0.05	0.31 ± 0.01
Volatile solids content (%)	78.7 ± 0.51	92.5 ± 1.14
Ash content (%)	21.3 ± 0.51	7.49 ± 1.14
Total carbon content (%)	44.0 ± 0.6	54.2 ± 0.07
Crude fibre content (%)	40.5 ± 2.3	47.3 ± 2.4

interesting to note that the dry fibre of bagasse can absorb 5 to 6 times its own mass of water without significant change in the bulk volume as opposed to the BGCW compost. This could be explained by the fact that particle size changes follow a logarithmic normal distribution during composting which resulted in a decrease of particle size in the BGCW compost (4.7 to 9.4 mm) as compared to that of bagasse (0.50 to 50.0 mm) which only underwent a 2−day fermentation, prior to substrate preparation. Kok-Kheng (1989) and Iwase *et al.* (2000), recommend substrate particle size within a minimum range of 1 mm to 3 mm for mycelium development and subsequent colonisation of the *P. sajor−caju* mushroom. Yet, Tabi *et al.* (2008), notifies on substrates with finer particle size which tend to become more compact when water is added, thus reducing voidage available. This renders the substrate nutrients less accessible impairing spreading and permeation of mushroom mycelia through the substrate. Water holding capacity diminishes during biodegradation, due to loss of organic content, with a parallel diminution in the ideal moisture content. Conversely, the BGCW still had a higher water holding capacity (572.7%) as compared to bagasse (188.0%). According to Arora *et al.* (2008), bagasse has a water holding capacity of 260% for a moisture content of 6.83%. However, excessive moisture above the water holding capacity of the substrate will cause local accumulation of fluid containing organic acids and other metabolites which might ferment and hinder mushroom yield. Bagasse had a higher porosity, 92.3% as compared to that of the BGCW compost (53.2%). Low porosity leads to low water holding capacity in most composts. According to Tomme *et al.* (1995) and Grethlein (1985), decreased porosity affects the accessibility of the mushroom's lignocellulosic enzymes to surface area adsorption of the substrate fibers. The lowest pH was recorded in bagasse, 5.6 while the BGCW compost had a pH of 7.94. *P. sajor−caju* normally thrives on substrates with pH ranges of 6.0 to 8.0 (Fanadzo *et al.* 2010). According to Choudhary *et al.* (2009) increase in electrical conductivity (normally greater than 6 dS/m) is almost proportional to the decrease in the number of pinheads. How-ever, both EC for BGCW compost and bagasse were found in the required range, 1.18 and 0.31 dS/m, respectively, to favour the growth and development of the oyster mushroom. The volatile solids fraction of a substrate comprises the percentage of organic matter that can be used directly by the mycelia (Magingo *et al.* 2004). It refers to the biodegradeble content of the substrate consisting mainly of cellulose with the organic matter content lying within a range of 68 to 95% (Silveira *et al.* 2008). Bagasse had the highest volatile solids content (92.5%) as compared to the BGCW compost (78.7%) suggesting a lower nutrient content available for the *P. sajor−caju* mushroom mycelium in the novel substrate. This statement is further confirmed by the crude fibre content and the total carbon content of substrates, 47.3 and 54.2% for bagasse and for the BGCW compost, 40.5 and 44.0%, correspondingly. Crude fibre measures the cellulose, hemicellulose and lignin content of food (Zakpaa *et al.* 2010). Most substrates used for oyster mushroom cultivation have a crude fibre

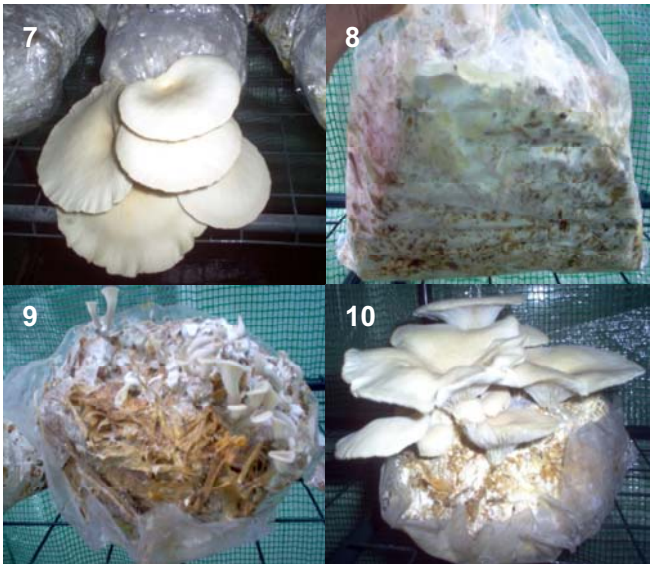

Fig. 7 Growth of *Pleurotus sajor–caju* on bagasse.
Fig. 8 Colonisation of *Pleurotus sajor–caju* on BGCW compost.
Fig. 9 *Pleurotus sajor–caju* pinheads on BGCW compost.
Fig. 10 Growth of *Pleurotus sajor–caju* on BGCW compost.

content ranging from 28 to 65% as demonstrated by Mshandete and Cuff (2007). For instance, *Triplochiton scleroxylon* which had the highest crude fibre content (63.8%), also showed the highest biological efficiency in oyster mushroom yield compared to other substrates namely, rice straw, banana leaves, maize stover, corn husk, rice husk, fresh sawdust, and elephant grass. For that reason, the BGCW compost was not allowed to mature otherwise this would have caused extensive degradation of its crude fibre content which contains the prime nutrients for the nourishment of the oyster mushroom. For instance, Obodai *et al.* (2010) confirmed a decrease in crude fibre content while composting *Triplochiton scleroxylon* K. (Schum) sawdust for 28 days. Similar results were obtained by Zhang *et al.* (1996) and Elamin and Hamed (2010) during traditional and ventilation fermentation of cotton seed hulls and after four months composting of farmyard manure with chicken manure, correspondingly. Furthermore, Huang *et al.* (2009) found the highest lignin degradation ratio and good cellulose degradation at the cooling stage of the composting process, by quinone profiling.

Biological efficiency

The spawn run on the substrate could be observed from the fifth day of incubation on bagasse and BGCW compost. Primordia formation appeared on the bagasse substrate only after 14 to 18 days of incubation while on the BGCW compost, 7 to 14 days later. Mushrooms were harvested three to five days later after primordia formation. **Fig. 7** shows the *P. sajor−caju* mushrooms growing on bagasse and **Figs. 8-10** show the same mushroom growing on the BGCW compost. The second and third flushes occurred after 15 and 25 days, respectively. The total yield of fructifications harvested from the three pickings was 950.30 g for bagasse and 810.60 g for the BGCW compost. The average biological efficiency on bagasse was 42.23 ± 4.4% as compared to the BGCW compost which was 36.03 ± 5.3%.

CONCLUSION

The general findings of this study indicate that compost from biowaste and lignocellulosic wastes using a controlled composting reactor may be produced. The feasibility of using this compost for the cultivation of the *Pleurotus sajor-caju* mushroom has also been verfiied. The composting time, with an airflow of 540 ± 24 L/h, was relatively

short, 23 days, to prevent the degration of fibre in the BGCW substrate. Nevertheless, the process met with the "Process to Further Reduce Pathogens" under the USEPA regulations consequently, achieving sanitisation. Additionally, the final carbon dioxide evolution of the BGCW compost indicated 'stable compost' according to Brinton (2000) and the California Compost Quality Council Standards (2005). The heavy metal contents for lead, cadmium and zinc of the composted BGCW substrate were well below the detection limits as specified by the current Mauritius Standards. The total nutrient content, nitrogen, potassium and phosphorus, for the BGCW compost (3.19%) was well above that of bagasse (0.01%). Despite this fact, slightly higher mushroom yields were observed on bagasse denoting that the mushroom mycelium taps its nutrients from the fibre content of the substrate itself. Hence, the average biological efficiency on the BGCW compost was 6.2% lower than bagasse.

REFERENCES

Agnew JM, Leonard JJ (2003) The physical properties of compost. *Compost Science and Utilization* **11 (3)**, 238-264

Akavia E, Beharav A, Wasser SP, Nevo E (2009) Disposal of agro-industrial by-products by organic cultivation of the culinary and medicinal mushroom *Hypsizygus marmoreus*. *Waste Management* **29 (5)**, 1622-1627

Annan JS, White RK (1998) Evaluation of techniques for measuring air filled porosity in composts of municipal biosolids and wood chips. In: Das KC, Graves EF (Eds) *Composting in the Southeast*, Proceedings of the 1998 Conference, SC, USA, pp 88-96

Arora NK, Khare E, Naraian R, Maheshwari DK (2008) Sawdust as a superior carrier for production of multipurpose bioinoculant using plant growth promoting rhizobial and pseudomonad strains and their impact on productivity of *Trifoliumrepense. Current Science* **95 (1)**, 90-94

Beffa T, Staib F, Lott Fischer J, Lyon P-F, Gumowski P, Marfenina OE, Dunoyer-Geindre S, Georgen F, Roch-Susuki R, Gallaz L, Latgé JP (1998) Mycological control and surveillance of biological waste and compost. *Medical Mycology* **36 (1)**, 137-145

Ben AL, Hassen A, Jedidi N, Saidi N, Bouzaiane O, Murano F (2007) Microbial C and N dynamics during composting process of urban solid waste. *Waste Management and Research* **25 (1)**, 24-29

Block SS, Tsao G, Han L (1958) Production of mushrooms from sawdust. *Journal of Agriculture and Food Chemistry* **6 (12)**, 923-927

Breitenbeck GA, Schellinger D (2004) Calculating the reduction in material mass and volume during composting. *Compost Science and Utilization* **12 (4)**, 365-371

Brinton WF (2000) Compost quality standard and guidelines: An international view. New York State Association of Recycler, pp 1-44

Buah JN, Van Der Puije GC, Bediako EA, Abole EA, Showemimo F (2010) The growth and yield performance of oyster mushroom (*Pleurotus ostreatus*) on different substrates. *Biotechnology* **9 (3)**, 338-342

California Compost Quality Council (CCQC) (2005) Compost Maturity Index. Available online: http://www.anr.state.vt.us/dec/wastediv/compost/documents/CompMaturity.pdf

Cangy C, Peerally A (1995) Studies of *Pleurotus* production on sugarcane bagasse. *African Journal of Mycology and Biotechnology* **3 (2)**, 67-79

Chen S, Zhang X, Singh D, Yu H, Yang X (2010) Biological pretreatment of lignocellulosics: potential, progress and challenges. *Biofuels* **1 (1)**, 177-199

Chiu SW, Ching ML, Fong KL, Moore D (1998) Spent oyster mushroom substrate performs better than many mushroom mycelia in removing the biocide pentachlorophenol. *Mycological Research* **102 (12)**, 1553-1562

Chiu SW, Gao T, Chan CSS, Karman Ho C (2009) Removal of spilled petroleum in industrial soils by spent compost of mushroom *Pleurotus pulmonarius*. *Chemosphere* **75 (6)**, 837-842

Choudhary DK, Agarwal PK, Johri BN (2009) Characterization of functional activity in composted casing amendments used in cultivation of *Agaricus bisporus* (Lange) Imbach. *Indian Journal of Biotechnology* **8 (1)**, 97-109

Dashtban M, Schraft H, Qin W (2009) Fungal bioconversion of lignocellulosic residues; Opportunities and perspectives. *International Journal of Biological Science* **5**, 578-595

Dewis J, Freitas F (1970) Physical and chemical methods of soil and water analysis. *FAO Soils Bulletin* **10**, 162-164

Diaz R, Alonso S, Sanchez C, Tomasini A, Bibbins-Martinez M, Diaz-Godinez G (2011) Characterization of the Growth and Laccase Activity of strains of *Pleurotus ostreatus* in submerged fermentation. *BioResources* **6 (1)**, 282-290

Dundar A, Acay H, Yildiz A (2009) Effect of using different lignocellulosic wastes for cultivation of *Pleurotus ostreatus* (Jacq.) P. Kumm. on mushroom yield, chemical composition and nutritional value. *African Journal of Biotechnology* **8 (4)**, 662-666

Elamin EA, Hamed NE (2010) Evaluation of maturing and nutrient content of farmyard manure composted with chicken manure. In: *International Conference on Organic Farming (ICOF), College of Agricultural Studies (CAS), Sudan University of science and Technology (SUST)*. 6–7 April 2010, Friendship Hall, Khartoum, Sudan, 7 pp

Fanadzo M, Zireva DT, Dube E, Mashingaidze AB (2010) Evaluation of various substrates and supplements for biological efficiency of *Pleurotus sajor-caju* and *Pleurotus ostreatus*. *African Journal of Biotechnology* **9 (19)**, 2756-2761

Ferreira LFR, Aguiar MM, Messias TG, Pompeu GB, Lopez AMQ, Silva DP, Monteiro RT (2011) Evaluation of sugar-cane vinasse treated with *Pleurotus sajor-caju* utilizing aquatic organisms as toxicological indicators. *Ecotoxicology and Environmental Safety* **74 (1)**, 132-137

Francou C, Pointrenaud M, Houot S (2005) Stabilization of organic matter during composting: Influence of process and feedstocks. *Compost Science and Utilization* **13 (1)**, 72-83

Futtee PH (2010) pers.comm., AREU. Senior Research Assistant. Agricultural Research and Extension Unit (AREU), Mauritius

Ghaly AE, Alkoaik F, Snow A (2006) Thermal balance of invessel composting of tomato plant residues. *Canadian Biosystems Engineering* **48**, 6.1-6.11

Gray KR, Biddlestone AJ (1973) Composting – Process parameters. *The Chemical Engineer* **2**, 71-76

Gregori A, Mirjan V, Pohleven J (2007) Cultivation techniques and medicinal properties of *Pleurotus* spp. *Food Technology and Biotechnology* **45 (3)**, 238-249

Hargreaves JC, Adl MS, Warman PR (2008) A review of the use of composted municipal solid waste in agriculture. *Agriculture Environment Ecosystems* **123**, 1-14

Hassan FRH, Ghada M, Medany SD, Hussein A (2010) Cultivation of the king oyster mushroom (*Plerrotus eryngii*) in Egypt. *Australian Journal of Basic and Applied Sciences* **4 (1)**, 99-105

Hassen A, Belguith K, Jedidi N, Cherif A, Cherif M, Boudabous A (2001) Microbial characterization during composting of municipal solid waste. *Bioresource Technology* **80**, 217-225

Hernández D, Sánchez JE, Yamasaki K (2003) A simple procedure for preparing substrate for *Pleurotus ostreatus* cultivation, *Bioresource Technology* **90 (2)**, 145-150

Huang DL, Zeng GM, Feng CL, Hu S, Lai C, Zhao MH, Su FF, Tang L, Liu HL (2010) Changes of microbial population structure related to lignin degradation during lignocellulosic waste composting. *Bioresource Technology* **101 (11)**, 4062-4067

Iwase K, Umezawa Y, Masuda K (2000) Cultivation *of Pleurotus ostreatus* with beer spent grains and utilization of carbonized waste substrate as a soil ameliorant. *Mushroom Science* **15 (2)**, 819-826

Iyengar SR, Bhave PP (2005) In-vessel composting of household wastes. *Waste Management* **26 (10)**, 1070-1080

Jolanun B, Towprayoon S (2010) Novel bulking agent from clay residue for food waste composting. *Bioresource Technology* **101 (12)**, 4484-4490

Juárez RAC, Dorry LLG, Bello-Mendoza R, Sánchez JE (2011) Use of spent substrate after *Pleurotus pulmonarius* cultivation for the treatment of chlorothalonil containing wastewater. *Journal of Environmental Management* **92 (3)**, 948-952

Kok-Kheng T (1989) Mushroom cultivation US Patent 4833821. Available online: http://www.freepatentsonline.com/4833821.html

Larney FJ, Olson AF, Miller JJ, Demaere PR, Zvomuya F, Mcallister TA (2007) Physical and chemical changes during composting of wood chip–bedded and straw-bedded beef cattle feedlot manure. *Journal of Environmental Quality* **37 (2)**, 725-735

Madsen E (1973) Effect of CO_2-concentration on the morphological, histological and cytological changes in tomato plants. *Acta Agriculturae Scandinavica* **23 (4)**, 241-246

Magingo FS, Oriyo NM, Kivaisi AK, Danell E (2004) Cultivation of *Oudemansiella tanzanica* nom. prov. on agricultural solid wastes in Tanzania. *Mycologia* **96 (2)**, 197-204

Mandeel QA, Al-Laith AA, Mohamed SA (2005) Cultivation of oyster mushrooms (*Pleurotus* spp.) on various lignocellulosic wastes. *World Journal of Microbiology and Biotechnology* **21 (4)**, 601-607

Mane VP, Patil SS, Syed AA, Baig MMV (2007) Bioconversion of low quality lignocellulosic agricultural waste into edible protein by *Pleurotus sajor-caju* (Fr.) Singer. *Journal of Zhejiang University Science* **8 (10)**, 745-751

Martínez-Carrera D (1998) Oyster mushrooms. In: Licker MD (Ed) *McGraw-Hill Yearbook of Science and Technology 1999*, McGraw-Hill, Inc., New York, pp 242-245

Mihilall Y, Mudhoo A, Mohee R (2010) Preliminary study on compost production as substrate for *Pleurotus sajor-caju* (Fr.) Singer mushroom cultivation in Mauritius. *Dynamic Biochemistry, Process Biotechnology and Molecular Biology* **4 (1)**, 32-43

Moda EM, Horii J, Spoto FHM (2005) Edible mushroom *Pleurotus sajor-caju* production on washed and supplemented sugarcane bagasse. *Scientia Agricola* **62 (2)**, 127-132

Mohee R, Mudhoo A, Unmar GD (2008) Windrow co-composting of shredded office paper and Broiler Litter, *International Journal of Environment and Waste Management* **2**, 3-23

Mohee R, Mudhoo A (2005) Analysis of the physical properties of an in-vessel composting matrix. *Powder Technology* **155**, 92-99

Mshandete AM, Cuff J (2007) Proximate and Nutrient composition of three types of indigenous edible wild mushrooms grown in Tanzania and their utilization prospects. *African Journal of Food Agriculture Nutrition and Development* **7 (6)**, 1-7

Mudhoo A, Mohee R (2008) Modelling heat loss during self-heating composting based on combined fluid film theory and boundary layer concepts. *Journal of Environmental Informatics* **11**, 74-89

Obodai M, Amoa-Awua W, Odamtten GT (2010) Physical, chemical and fungal phenology associated with the composting of 'wawa' sawdust (*Triplochiton scleroxylon*) used in the cultivation of oyster mushrooms in Ghana. *International Food Research Journal* **17**, 229-237

Okhuoya JA, Akpaja EO, Osemwegie OO, Oghenekaro AO, Ihayere CA (2010) Nigerian mushrooms: underutilized non-wood forest resources. *Journal of Applied Sciences Environmental Management* **14 (1)**, 43-54

PennState (2009) Agricultural Analytical Services Laboratory, College of Agricultural Sciences. The Pennsylvania State University. Available online: http://www.frederickcountymd.gov/documents/Utilities%20&%20Solid%20Waste%20Management/Recycling/2009-07-17.PDF

Pozdnyakova NN, Dubrovskaya EV, Makarov OE, Nikitina VE, Turkovskaya OV (2011) Production of ligninolytic enzymes by white-rot fungi during bioremediation of oil-contaminated soil. *Soil Enzymology* **22**, 363-377

Prabhakar A, Krishnaiah K, Janaun J, Bono A (2005) An overview of engineering aspects of solid state fermentation. *Malaysian Journal of Microbiology* **1 (2)**, 10-16

Puchooa D, Ponnuswamy B, Wan Chow Wah YF (1999) An investigation on low cost, locally-available substrates potentially suitable for use in the hardening of tissue-cultured plantlets. *Science and Technology – Research Journal* **4**, 127-143

Robles-Hernández L, González-Franco AC, Crawford DL, Chun WWC (2008) Review of environmental organopollutants degradation by whiterot basidiomycete mushrooms. *Tecnociência* **2 (1)**, 32-40

Ruggieri L, Gea T, Artola A, Sánchez A (2008) Influence of different co-substrates biochemical composition on raw sludge co-composting. *Biodegradation* **19 (3)**, 403-415

Shalchian-Tabrizi K, Minge MA, Espelund M, Orr R, Ruden T, Jakobsen KS, Cavalier-Smith T (2008) Multigene phylogeny of choanozoa and the origin of animals. *PLoS ONE* **3 (5)**, e2098

Silveira MLL, Furlan SA, Ninow JL (2008) Development of an alternative technology for the oyster mushroom production using liquid inoculums. *Ciência e Tecnologia de Alimentos Campinas* **28 (4)**, 858-862

Soumaré M, Demeyer A, Tack FM, Verloo MG (2002) Chemical characteristics of Malian and Belgian solid waste composts. *Bioresource Technology* **81 (2)**, 97-101

Tabi M, Nafissa A, Zakil A, Fauzai F, Fauzai M, Fauzan WN, Onn A, Onn N, Onn H (2008) The usage of empty fruit bunch (efb) and palm pressed fibre (ppf) as substrates for the cultivation of *Pleurotus ostreatus*. *Jurnal Teknologi* **49**, 189-196

TEG Group PLC (2007) The way forward with organic waste. Available online: http://www.theteggroup.plc.uk

Tomme P, Warren RA, Gilkes NR (1995) Cellulose hydrolysis by bacteria and fungi. *Advanced Microbiology Physiology* **37**, 1-81

Upadhyay RC, Singh M (2010) Production of edible mushrooms. *The Mycota* **10 (1)**, 79-97

USCC (2001) US Composting Council. *Compost maturity index*. Available online: http://www.anr.state.vt.us/dec/wastediv/compost/documents/CompMaturity.pdf

USCC (2010) US Composting Council, *Test Methods and Parameters*. Available online: http://www.compostingcouncil.org/programs/sta/test_methods.php

USDA Economics, Statistics and Market Information System (2009) *Mushroom Industry Report*. United States Department of Agriculture: Albert R. Mann Library Cornell University, (94003). Available online: http://usda.mannlib.cornell.edu/MannUsda/viewDocumentInfo.do?documentID=1395

USEPA (1994) United States Environmental Protection Agency. Pathogen Equivalency Committee (PEC). Available online: http://www.epa.gov/nrmrl/pec/basic.html#whatare

Zakpaa HD, Mak-Mensah EE, Adubofour J (2010) Production and characterization of flour produced from ripe "apem" plantain (*Musa sapientum* L. var. *paradisiacal;* French horn) grown in Ghana. *Journal of Agricultural Biotechnology and Sustainable Development* **2 (6)**, 92-99

Zhang CK, Gong F, Sheng XF, Li DS, Feng YJ (1996) Ventilation fermentation of cotton-seed hulls: population dynamics of microflora and biochemical changes during fermentation. In: Royse DJ (Ed) *Proceedings of the Second International Conference*, June 9-12, 1996, World Society for Mushroom Biology and Mushroom Products, University Park, Pennsylvania, pp 147-152