Journal of King Saud University – Science (2016) **28**, 143–151

King Saud University
Journal of King Saud University – Science
www.ksu.edu.sa
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ORIGINAL ARTICLE

Utilization of fruit peels as carbon source for white rot fungi biomass production under submerged state bioconversion

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Received 13 February 2015; accepted 3 August 2015
Available online 11 August 2015

KEYWORDS

Substrates;
Biomass;
Fermentable;
Bioconversion;
Phanerochaete chrysosporium;
Panus tigrinus

Abstract The present generation of nutrient rich waste streams within the food and hospitality industry is inevitable and remained a matter of concern to stakeholders. Three white rot fungal strains were cultivated under submerged state bioconversion (SmB). Fermentable sugar conversion efficiency, biomass production and substrate utilization constant were indicators used to measure the success of the process. The substrates – banana peel (Bp), pineapple peel (PAp) and papaya peel (Pp) were prepared in wet and dried forms as substrates. *Phanerochaete chrysosporium* (*P. chrysosporium*), *Panus tigrinus* M609RQY, and RO209RQY were cultivated on sole fruit wastes and their composites. All fungal strains produced profound biomass on dry sole wet substrates, but wet composite substrates gave improved results. *P. tigrinus* RO209RQY was the most efficient in sugar conversion (99.6%) on sole substrates while *P. tigrinus* M609RQY was efficient on composite substrates. Elevated substrate utilization constant (K_u) and biomass production heralded wet composite substrates. *P. chrysosporium* was the most performing fungal strain for biomass production, while PApBp was the best composite substrate.

Abbreviations: Bp, banana peel; PAp, pineapple peel; Pp, papaya peel; SmB, submerged state bioconversion; WRF, white rot fungi; TOS, total soluble sugar; TRS, total reducing sugar

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Peer review under responsibility of King Saud University.

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1. Introduction

Improved fruit and vegetable production through efficient agricultural practices mobilizes huge investments in fruit and vegetable processing across the world. Banana, pineapple and papaya are among the most widely acceptable fruits planted on commercial level worldwide (Jamal et al., 2012). Waste generation through these fruits is on the increase due to sustained surge in world population, improved economic

http://dx.doi.org/10.1016/j.jksus.2015.08.002

growth in developing nations and improved access to nutrition education in high fruit producing countries.

Wastes emanating from aforementioned fruits include peels, pulp and seeds that constitute about 40% of the total mass of each fruit. The majority of these waste materials is often improperly disposed, hence constitute huge environmental disorders (Essien et al., 2005; Lim et al., 2010). Fruit waste dumping sites provide necessary impetus for vectors, pathogenic bacteria and yeast to thrive. A popular approach to mitigating fruit waste poor handling is landfill and incineration; these methods orchestrate an acute air pollution problem by generating massive leachates that contaminate ground water and destroy aquatic lives (Ali et al., 2014; Taskin et al., 2010).

Banana peel (Bp), pineapple peel (PAp) and papaya peel (Pp) are major wastes generated by fruit processing and agro-allied industries (Rasu Jayabalan et al., 2010). These wastes contain simple and complex sugars that are metabolizable by microorganisms through secretion of extracellular products (Saheed et al., 2013). Fruit peels, which constitute a huge part of the waste streams, provide anchorage for filamentous fungi during bioconversion process (Essien et al., 2005). Bioconversion of single fruit waste is a common practice in valorization of fruit peels. Pineapple waste, palm tree waste and cassava waste have received attention for their conversion to bio-ethanol, biogas and animal feed (Alam et al., 2005; Dhanasekaran et al., 2011; Tijani et al., 2012). Designing treatment schemes for specific agricultural residue limits efficiency of waste collection and prolong treatment period. Therefore, adoption of a method that accommodates several fruit wastes is highly robust, cheap and realistic in ameliorating impediments associated with fruit waste disposal (Aggelopoulos et al., 2014). The cultivation of microbial cells (bacteria, yeast, and fungi) that converts fruit wastes into value added products such as biomass that can serve as animal feed supplement is a unique approach.

White rot fungi (WRF) – a class of filamentous fungi - are efficacious in valorizing cellulosic fruit wastes through degradation of complex carbohydrates in recalcitrant agro-residues (Ruqayyah et al., 2013). Several WRF used as edibles, contain essential micronutrients and amino acids at concentrations required for animal health and growth. Their biochemical mechanism of augmenting organic residues involves secretion of lignolytic, amylolytic and other hydrolytic enzymes (Cellulases, Amylases, Lipases etc.) into the fermentation broth during growth to facilitate breakdown of cellulose, starch and lignin in the fruit residues (Sanjay Kumar and Sarkar, 2011). A direct consequence of enzyme secretion is the development of fungal biomass that contains protein, fat and essential amino acids useful for supplementing ruminant and monogastric animal feed (Dhanasekaran et al., 2011; Rasu Jayabalan et al., 2010).

The profile of soluble and reduced carbohydrate content of fruit wastes metabolized by WRF during the bioconversion process is imperative to measure the efficiency of the biochemical process but rarely investigated. Determination of carbon source consumption pattern of fungal cells prior to products synthesis is imperative for measuring opportunities offered by the method (Qureshi et al., 2014). Therefore, this investigation elucidates, the performance of WRF on wet and dried forms of Bp, PAp and Pp. The study also covered the performance of composite substrates developed from the three peels. Parameters compared between individual peel substrate and composites include WRF biomass production, substrate (sugar) conversion efficiency and substrate utilization constant.

2. Materials and methods

2.1. Fungal strains and cultivation

Three white rot fungi comprising two locally isolated *Panus tigrinus* strains RO209RQY and M609RQY (IMI 398363, CABI Europe-UK) (*Polyporales polyporaceae*) and laboratory stock of *Phanerochaete chrysosporium* Burdsall, teleomorph (ATCC 20696) (*P. chrysosporium*) were selected to carry out bioconversion process. RO209RQY (RO2); and M609RQY (M6) were cultivated on malt extract agar (MEA, Merck, Germany) for 7 days at 30 °C while *P. chrysosporium* was cultivated on potato dextrose agar (PDA, Merck, Germany) for 7 days at 30 °C. Each strain was sub-cultured every fortnight.

2.2. Substrate collection and preparation

Fresh banana (*Musa sapientum*) peels, pineapple (*Ananas cosmos*) peels and papaya (*Carica papaya*) peels were collected from fruit processors within the Gombak, Selangor, Malaysia area (Selangor, West Malaysia). The peels were thoroughly washed with tap water to remove attached foreign materials. Wet substrate contained a mixture of one-part peels and one-part distilled water (1:1) and blended for 5 min. 2 mm screen was used to sieve the resulting slurry before being stored at −20 °C for subsequent use. Fruit peels needed in dried form were dehydrated at 60 °C for two days immediately after cleaning to stop destructive microorganism. The peels were ground, sieved to 2 mm particle size and stored in an airtight container for subsequent use, while ungrounded ones were kept at room temperature in airtight plastic bags. Composite forms of dry and wet substrates were prepared by mixing respective peel combination in ratio 1:1:1.

2.3. Determination of total soluble sugar (TOS) and reducing sugar

Total soluble sugar concentration of fruit peel samples before and after bioconversion was determined by using phenol sulfuric acid (Dubois et al., 1956). For reducing sugar of fruit peel samples before and after bioconversion, aqueous extractions of reducing sugar from banana peel, pineapple peel and papaya peel were done in a 50 ml stoppered conical flask containing air-dried peels for dry sample and slurry for wet sample. 10 ml of 0.2 (mol/L) of disodium hydrogen phosphate/0.1 (mol/L) of citrate buffer (pH 4.8) was added before centrifugation was performed. Reducing sugar of the supernatant was determined by the Miller method using dinotrosylsalicylic acid reagent (DNS) (Miller, 1959).

2.4. Fungal biomass determination, substrate utilization constant and microbial efficiency determination

In order to determine the amount of white rot fungi biomass produced, after bioconversion process, all the contents of the

Erlenmeyer flasks were first sieved with screens in such a way that unconverted fibrous fruit strands and residual soluble and reducing sugars were removed. The residue (fungal biomass) was gently washed with distilled water and transferred to pre-weighed whatman No. 1 filter paper (Sigma–Aldrich) (Omar and Sabry, 1991). The filter paper content was dried and total biomass produced was determined by calculating the weight difference before and after drying (Eq. (1))

$$\text{Biomass} = W_2 - W_1 \quad (1)$$

W_1: weight of pre-dried filter paper; W_2: weight of dried biomass and filter paper.

Substrate utilization constant of proportionality (K_u) was obtained with the assumption that edible fungal biomass production is inversely proportional to substrate sugar metabolism in a batch processing; the mathematical expression for determining the constant was given (Eq. (2)):

$$K_u = (B_2 - B_1) \times (S_1 - S_2) \quad (2)$$

K_u: substrate sugar utilization constant; B_1: initial fungal biomass; B_2: final fungal biomass; S_1: initial substrate sugar content; S_2: final substrate sugar content.

The efficiency of each fungus in converting metabolizable sugar in the substrate to biomass over the 7-day bioconversion period was calculated from Eq. (3) below:

$$\text{Conversion efficiency} = \frac{I_o - I_f}{I_o} \times 100 \quad (3)$$

I_o: initial amount metabolizable sugar; I_f: final amount metabolizable sugar.

2.5. Inoculum preparation and submerged state bioconversion (SmB)

Inoculums were prepared by using 25 ml of sterilized distilled water to wash each petri dish of 7 day old fungal mycelium by gently scratching the agar plate surface with L-shaped rod and stored at 4 °C. Submerged state bioconversion was carried out in 250 ml Erlenmeyer flasks comprising 2% (1 g) substrate (solid particles of wet and dry substrates were equalized by determining their moisture content prior to bioconversion) and 2% (1 ml) fungal inoculum. The conversion media contained 0.8 g/L KH_2PO_4, 1.5 g/L $(NH_4)_2SO_4$, 0.45 g/L $MgSO_4$ and 0.05 g/L $MnSO_4$ and distilled water was added to make 50 ml working volume. The flasks were previously autoclaved at 121 °C for 15 min and cooled before inoculation. Samples were transferred to an incubator shaker (Lab companion model SK-300) at 150 rpm and 30 °C cultivation temperature. Fungal biomass was separated and measured after 7 day incubation. All experiments were undertaken in triplicate to minimize experimental error.

2.6. Statistical analysis of data

Analysis of variance (ANOVA) covering single and multi-factors involved in the treatments; Post-hoc *t*-test (to identify the significance level where ANOVA was previously significant at $p < 0.05$) was performed. Statistical analysis was implemented in Microsoft excel 2010 version using data analysis add-on.

3. Result and discussion

3.1. Fungal biomass production on individual fruit peels

In the process of biomass production on wet substrates, *P. chrysosporium* produced the highest biomass when cultivated on wet Bp (Table 1). On wet Bp, biomass production by *P. chrysosporium* was significantly different ($p < 0.05$) compared with M6; similar statistical difference existed between M6 and RO2. The maximum fungal biomass for M6 and RO2 were on Pp at no significant difference between the two microbes; same trend was recorded on PAp. Significant difference was recorded between *P. chrysosporium* and RO2 on PAp and Pp. However, *P. chrysosporium* produced the least biomass at significant levels compared to other two microbes on Pp.

All selected microbes significantly ($p < 0.05$) produced more biomass on dry substrate compared with wet substrate. No significant difference was recorded between *P. chrysosporium* biomass on dry and wet forms of Bp and Pp. However, a significant difference was observed between dry and wet substrates for M6 and RO2; only M6 recorded an insignificant difference. However, on Pp, *P. chrysosporium* and RO2 showed a significant difference in biomass production while M6 was insignificant. Investigations involving protein enrichment of supplemented PAp showed that fungal imperfecti cells recorded profound biomass growth as prelude to high protein synthesis. However the biomass production of all selected strains on either wet or dry substrate forms showed that intense biomass was produced in this report compared with other works (Correia et al., 2007; Dhanasekaran et al., 2011; Nitayavardhana and Khanal, 2010).

Table 1 Fungal biomass production on sole substrate.

Substrate	PC (g/L)		M6 (g/L)		RO2 (g/L)	
	Wet sub	Dry sub	Wet sub	Dry sub	Wet sub	Dry sub
Bp	15.60 ± 0.07^{ax}	17.40 ± 0.05^{b1x}	12.00 ± 0.13^{c}	23.60 ± 0.13^{d1}	15.40 ± 0.25^{e}	24.40 ± 0.25^{f1}
PAp	9.40 ± 0.02^{a}	15.00 ± 0.03^{b2}	15.80 ± 0.05^{c}	15.40 ± 0.06^{d2}	15.00 ± 0.03^{e}	19.60 ± 0.07^{f2}
Pp	15.40 ± 0.03^{ax}	15.00 ± 0.03^{b2x}	17.80 ± 0.08^{c}	13.20 ± 0.06^{d2}	17.80 ± 0.07^{e}	16.80 ± 0.04^{f3}

^{a,b,c,d,e,f}: values with different superscripts in row are significantly different at $p < 0.05$.
^{1,2,3}: values with different superscripts in column are significantly different at $p < 0.05$.
Wet sub: wet substrate, dry sub: dry substrate.
PC: *P. chrysosporium*; M6: *Panus tigrinus* (M609RQY); RO2: *Panus tigrinus* (RO209RQY).
BP: banana peel, PAp: pineapple peel, Pp: papaya peel.

3.2. Effects of fungal growth on metabolizable sugar content of fruit peels

Initial concentration of total soluble sugar (TOS) by wet Bp was 36.71 mg/g; 75.45 mg/g for PAp while Pp had 52.35 mg/g. The final concentration of TOS after bioconversion showed that *P. chrysosporium* utilized more fermentable sugar than M6 and RO2 on Bp and PAp while it consumed least of Pp sugar (Table 2). Raw Bp, PAp, and Pp had 1.30 mg/g, 1.80 mg/g and 4.54 mg/g total reducing sugar (TRS); after 7 day bioconversion *P. chrysosporium* consumed less TRS than M6 and RO2. This shows that *P. chrysosporium* required less reducing sugar for growth and development. RO2 on the other hand, showed preferred TRS compared with TOS in other fruit wastes, but M6 showed an unchanged consumption pattern for TOS and TRS respectively. Results of other workers showed that increased fungal biomass corresponds with increased metabolism of reducing sugar content in fermentation media (Essien et al., 2005; Jamal et al., 2009). A linear biomass production over the fermentation period was documented with a corresponding exponential fall in reducing sugars (Correia et al., 2007).

Initial TOS by dry Bp was 32.84 mg/g; 40.74 mg/g for PAp while Pp had 24.94 mg/g. On dry sample of Bp, final concentration of TOS after bioconversion showed that *P. chrysosporium* consumed more TOS compared with M6 and RO2. The initial values of TRS for each substrate (Bp 1.29 mg/g; Pp 1.70 mg/g; Pw 0.86 mg/g) showed that *P. chrysosporium* performed best only on PAp when comparing its TRS values with others. M6 maintained a middle course on all substrates except on Bp where it consumed the least amount of TOS and TRS. Performance of RO2 on all substrates was moderate with the best result on PAp and Bp; it least performed on Pp among other microbes. In a fermentation process involving *Aspergillus fumigatus* cultivation on optimized media, 1.8 mg of biomass was recorded over 7 days, compared with the present study where an average of 10 g/l was recorded (Essien et al., 2005).

3.3. Substrate utilization constant (K_u) of fungi on sole substrates

Increased biomass production by filamentous fungi often inversely relates to substrate nutrients' concentration – more biomass, less nutrients (Dhanasekaran et al., 2011; Ezekiel et al., 2010). All selected fungal cells demonstrated profound utilization of simple sugars (TRS and TOS) within the wet substrate matrix by recording high values of K_u (Fig. 1a). M6 and RO2 recorded the highest value on Pp and PAp while PC and ROS utilized Bp better than M6. Although no investigator has taken cognizance of this mathematical relationship in batch bioconversion, data concerning decrease in carbon source as a direct response to microbe growth and product formation abound. An inference drawn from other reports showed that K_u values recorded in this research compared favourably with other results' outcome (Ahmed et al., 2010; Munawar et al., 2010). Higher values of K_u were recorded for M6 and RO2 only on dry Bp with wet samples (Fig. 1b); slight variation was evident from other substrates, but, they all showed elevated values when compared with wet samples. This observation was consistent with reports of other workers, where WRF was

Table 2 Residual fermentable sugar of sole substrate.

WRF	Banana peel				Pineapple peel				Papaya peel			
	TOS (g/L)		TRS (g/L)		TOS (g/L)		TRS (g/L)		TOS (g/L)		TRS (g/L)	
	Wet sub	Dry sub	Wet sub	Dry sub	Wet sub	Dry sub	Wet sub	Dry sub	Wet sub	Dry sub	Wet sub	Dry sub
PC	4.01 ± 0.43	4.25 ± 0.10	0.57 ± 0.19	0.75 ± 0.03	4.28 ± 0.10	4.31 ± 0.05	0.79 ± 0.03	0.69 ± 0.04	4.28 ± 0.06	4.21 ± 0.03	0.25 ± 0.10	0.73 ± 0.03
M6	4.21 ± 0.06	4.31 ± 0.01	0.60 ± 0.01	0.82 ± 0.03	4.34 ± 0.05	4.26 ± 0.04	0.78 ± 0.01	0.75 ± 0.03	4.26 ± 0.06	4.23 ± 0.01	0.66 ± 0.16	0.64 ± 0.03
RO2	4.18 ± 0.13	4.30 ± 0.10	0.07 ± 0.03	0.74 ± 0.05	4.33 ± 0.11	1.47 ± 0.49	0.66 ± 0.07	0.74 ± 0.04	4.15 ± 0.09	4.28 ± 0.07	0.02 ± 0.03	0.74 ± 0.03

PC: *P. chrysosporium*; M6: *Panus tigrinus* (M609RQY); RO2: *Panus tigrinus* (RO209RQY).
Wet sub: wet substrate, dry sub: dry substrate.
WRF: white rot fungi.
TOS: total soluble sugar, TRS: total reducing sugar.

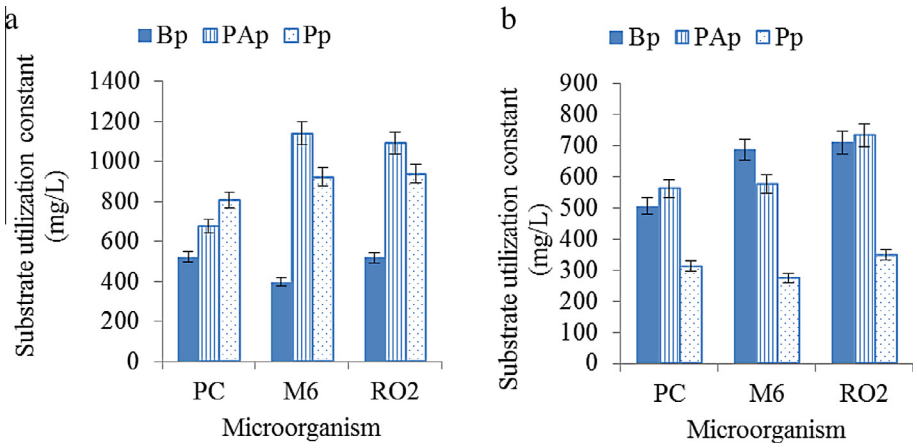

Figure 1 Substrate nutrient utilization constant of WRF strains on dry and wet sole fruit peels (a) on wet Bp, PAp and Pp; (b) on dry Bp, PAp and Pp.

Table 3 Fungal biomass production on composite substrates.

Substrate	PC (g/L)		M6 (g/L)		RO2 (g/L)	
	Dry sub	Wet sub	Dry sub	Wet sub	Dry sub	Wet sub
BpPApPp	18.58 ± 0.08^{a1}	13.83 ± 0.03^{ac2}	17.01 ± 0.02^{a1}	14.23 ± 0.03^{ab2}	20.05 ± 0.13^{a1}	15.44 ± 0.01^{bd1}
BpPp	17.83 ± 0.19^{a3}	15.33 ± 0.02^{ae3}	19.28 ± 0.12^{a3}	17.55 ± 0.04^{bc3}	20.27 ± 0.22^{a3}	20.06 ± 0.02^{df3}
PApBp	21.15 ± 0.17^{a4}	16.38 ± 0.09^{a4}	18.26 ± 0.02^{a4}	15.77 ± 0.06^{a5}	20.15 ± 0.28^{a4}	16.26 ± 0.04^{a4}
PApPp	19.41 ± 0.15^{a5}	10.72 ± 0.07^{a6}	19.93 ± 0.10^{a6}	12.64 ± 0.02^{a7}	23.37 ± 0.27^{a5}	12.47 ± 0.10^{a6}

^{a,b,c,d}: values with different superscripts are significantly different at $p < 0.05$.
^{1,2,3}: values with different superscripts are significantly different at $p < 0.05$.
Wet sub: wet substrate, dry sub: dry substrate.
PC: *P. chrysosporium*; M6: *Panus tigrinus* (M609RQY); RO2: *Panus tigrinus* (RO209RQY).
BpPApPp: banana, pineapple and papaya peel, BpPp: banana and papaya peel, PApBp: pineapple and banana peel, PApPp: pineapple and papaya peel.

recorded to metabolize more sugars locked in solid matrix together with those released into fermentation broth, higher biomass was reported (Gad et al., 2010; Jamal et al., 2009).

3.4. Fungal biomass production through composite substrates

Fungal biomass began to manifest after 72 h in all the selected microorganisms. *P. chrysosporium* produced the highest biomass, followed by RO2 (Table 3). Dry composite substrate significantly produced more biomass compared with wet substrates. RO2 growth on dry matrix of PApPp was most profound albeit, not significantly different from *P. chrysosporium* and M6. A similar trend occurred by *P. chrysosporium* on dry PApBp substrate while M6 made its highest impact on PApPp. A significant difference existed between *P. chrysosporium* biomass on dry and wet substrates of BpPApPp and PApPp, but none occurred between biomass production by BpPp and PApBp respectively. M6 recorded a significant difference in biomass production between dry and wet forms of BpPApPp, PApBp, and PApPp while no significant difference was evident between dry and wet BpPAp. RO2 biomass production differed significantly between dry PApPp and wet type; other substrate combinations (dry and wet) are not profoundly different. Although, there are no reports comparing performance of fungal cells on sole and composite fruit peels, available report showed that WRF biomass and extracellular synthesis increased under combined waste streams than single waste sources (Arumugam and Manikandan, 2011; Essien et al., 2005).

3.5. Effects of fungal growth on sugar content of composite substrates

Initial TOS of wet composite substrate was 164.0 mg/g for BpPApPw, 112.16 mg/g for BpPAp, 89.06 mg/g for BpPp while PApPp had 127.8 mg/g. Similarly, initial TRS for wet composite substrates was 98.52 mg/g for BpPApPw, 73.58 mg/g for BpPAp, 57.78 mg/g for BpPp while PApPp had 127.80 mg/g of TRS. On wet media, all selected strains left an average of 4.0 mg/g TOS while less than 2.0 mg/g was the highest residual TRS on the average (Table 4). *P. chrysosporium* and M6 consumed more TOS of dry substrate compared with the wet form while RO2 left a higher amount of TOS in the dry substrate. All selected fungal strains demonstrated huge metabolic preference for TRS by leaving a paltry 0.5 mg/g in the media after 7 day bioconversion. The tendency of WRF to metabolize more TRS was earlier reported (Gad et al., 2010) however, other fermentative microbes exhibited similar growth requirement for higher synthesis of bioproducts from agro-residues (Dhanasekaran et al., 2011).

Table 4 Residual fermentable sugar of composite substrates after bioconversion.

Substrate	PC				M6				RO2			
	TOS (g/L)		TRS (g/L)		TOS (g/L)		TRS (g/L)		TOS (g/L)		TRS (g/L)	
	Wet sub	Dry sub	Wet sub	Dry sub	Wet sub	Dry sub	Wet sub	Dry sub	Wet sub	Dry sub	Wet sub	Dry sub
BpPApPp	4.04 ± 0.09	2.72 ± 0.30	0.07 ± 0.01	0.49 ± 0.20	3.88 ± 0.39	2.51 ± 0.33	0.03 ± 0.02	0.31 ± 0.03	4.05 ± 0.36	16.87 ± 0.33	0.11 ± 0.01	0.59 ± 0.03
BpPp	4.03 ± 0.08	3.38 ± 0.40	0.09 ± 0.00	0.53 ± 0.10	4.20 ± 0.02	2.35 ± 0.08	1.97 ± 0.03	0.35 ± 0.19	4.02 ± 0.42	16.37 ± 0.08	0.06 ± 0.00	0.49 ± 0.19
PApBp	4.32 ± 0.08	2.65 ± 1.01	0.18 ± 0.11	0.43 ± 0.06	4.25 ± 0.02	2.35 ± 0.08	1.52 ± 0.04	0.29 ± 0.09	4.21 ± 0.07	14.63 ± 0.08	0.51 ± 0.01	0.41 ± 0.09
PApPp	4.17 ± 0.09	3.04 ± 0.76	0.72 ± 0.00	0.58 ± 0.35	4.24 ± 0.04	2.88 ± 0.11	1.94 ± 0.05	0.46 ± 0.65	4.02 ± 0.02	15.92 ± 0.11	0.05 ± 0.00	0.51 ± 0.06

Wet sub: wet substrate, dry sub: dry substrate.
PC: *P. chrysosporium*; M6: *Panus tigrinus* (M609RQY); RO2: *Panus tigrinus* (RO209RQY).
BpPApPp: banana, pineapple and papaya peel, BpPp: banana and papaya peel, PApBp: pineapple and banana peel, PApPp: pineapple and papaya peel.
TOS: total soluble sugar, TRS: total reducing sugar.

Therefore, results presented were consistent with others where improved protein, enzyme and organic acid synthesis were end products of sugar metabolism of agro-wastes (Ezekiel et al., 2010).

3.6. Fungal substrate utilization constant (K_u) on composite substrates

Wet composite substrates supported improved microbial metabolism with high values of K_u recorded for all selected WRF (Fig. 2a). RO2 recorded the highest values on BpPApPp, BpPAp, and PApPp while *P. chrysosporium* was best on BpPp. Although M6 was least performing on composite wet samples, it exhibited improved K_u value compared with sole samples. Generally, all the strains demonstrated improved metabolism on composite wet samples; suggesting synergy among the substrates (Saheed et al., 2013). Similarly, results showed that all fungal strains performed well on dry composite substrates (Fig. 2b); *P. chrysosporium* showed higher K_u on BpPp while M6 outclassed others on BpPAp and PApPp. However, the values of K_u recorded for dry composite substrates were lower when compared with wet samples. This could be caused by high sugar release from the wet compared with the dry sample where osmotic effects may hinder sugar release (Enwefa, 1991). Although there are no reports concerning K_u of fermentation processes, deductions from other fungal investigation showed that K_u of fungal strains are higher for wet media albeit, may not result in higher products. In a report concerning bio-protein production, higher protein was produced in slurry substrates and fruit waste hydrolyzates though their K_u differ greatly (Dhanasekaran et al., 2011; Dimova et al., 2010).

3.7. Substrate conversion efficiency of microbes on each fruit waste

On Bp, selected fungi demonstrated comparable efficiency on TOS (Fig. 3a); this observed similarity between selected fungal strains showed congruence in their metabolism regardless of substrate type. The efficiency of each microbe differed greatly on TRS; RO2 performed better than other fungal strains on wet Bp followed by *P. chrysosporium* and M6. A similar trend was visible on dry Bp where RO2 had a better performance. This result demonstrated RO2 preference for TRS compared with TOS on either dry or wet forms. This trend was previously recorded for WRF for their selective metabolism of fermentable sugar under different fermentation broth conditions (Rosma et al., 2007). However, information on *P. chrysosporium* suggested consistency between the present study and other reports (Gad et al., 2010). Information concerning proficiency of RO2 and M6 showed that they perform optimally on complex substrates. Therefore, this study provided more insight into their biochemical performance (Ruqayyah et al., 2011).

On PAp, all strains demonstrated profound efficiency for TOS metabolism when compared with TRS (Fig. 3b). Fungal cells exhibited closer efficiency on wet PAp for TOS while RO2 performed insignificantly better than *P. chrysosporium* and M6 on dry substrates. However, selected fungal strains were less efficient on TRS except RO2 that slightly metabolize more TRS compared with others; *P. chrysosporium*

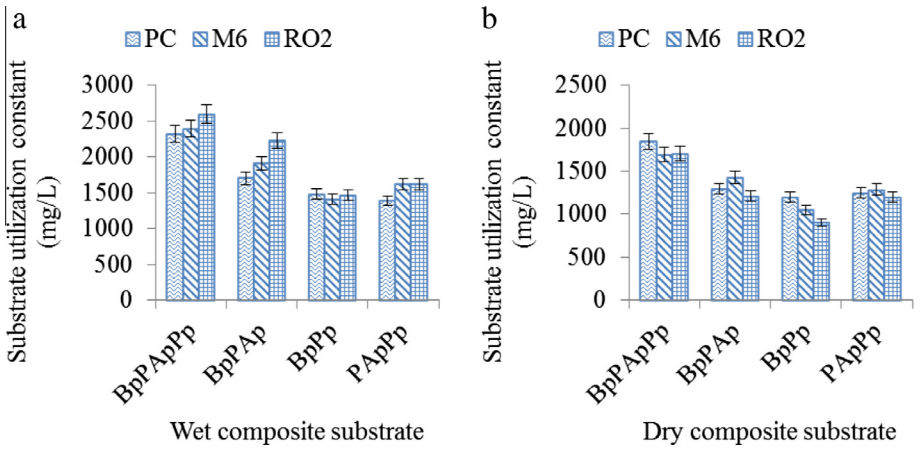

Figure 2 Composite substrate fermentable sugar (TOS and TRS) utilization constant of WRF strains (a) on wet composite substrates; (b) on dry composite substrates.

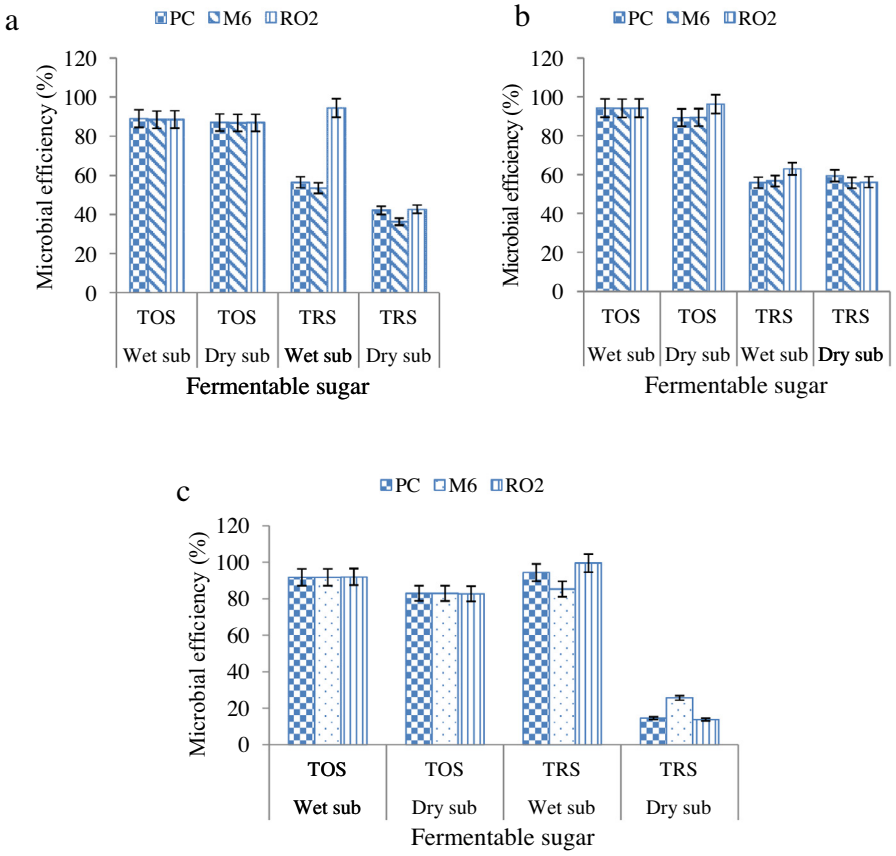

Figure 3 Substrate fermentable sugar (TOS and TRS) utilization efficiency of WRF strains on sole fruit peels (a) banana peels; (b) pineapple peels; (c) papaya peels.

exhibited intense metabolism on dry TRS. The reduction in efficiencies of the strains on TRS could be attributed to high content of the sugar since PAp generally harbors high reducing sugar (Sanjay Kumar and Sarkar, 2011).

The efficiency of the selected strain concerning TOS of Pp showed that all strains demonstrated profound efficiency on wet substrate than dry (Fig. 3c). Same trend was obvious for TRS with a noticeable difference between dry and wet Pp forms. This metabolic performance by fungal strains on Pp revealed that it could support microbial growth for production of value added products. This observation was raised by other workers where high protein synthesis was recorded due to the metabolism of sugar contents of agro-residues (Akin-Osanaiye et al., 2008).

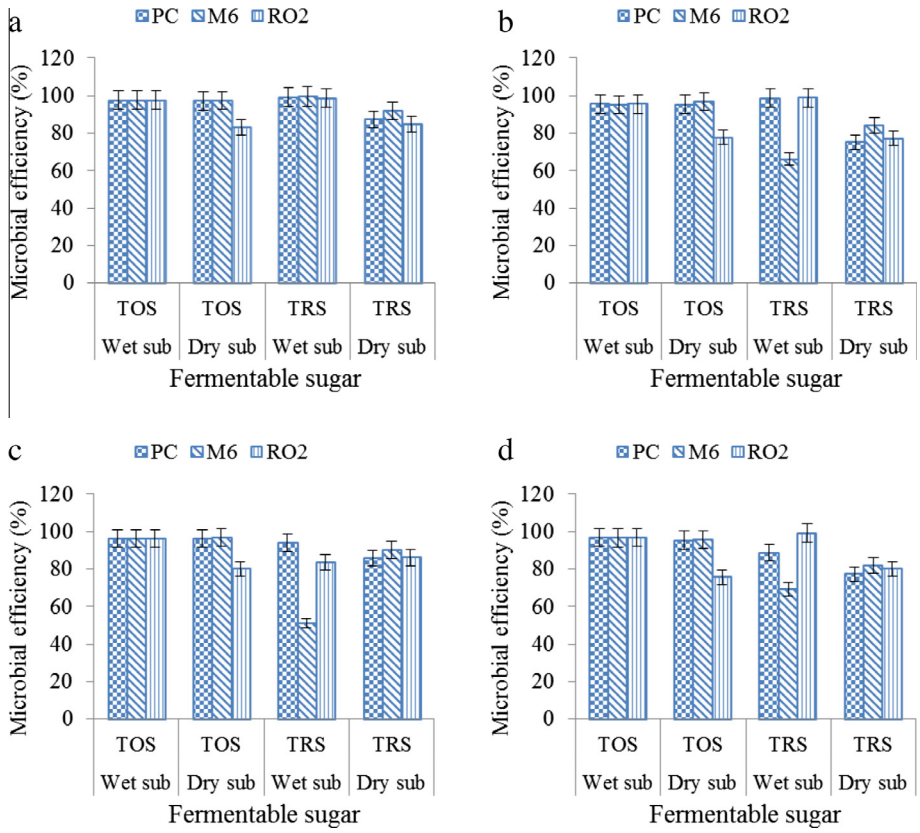

Figure 4 Substrate fermentable sugar (TOS and TRS) utilization efficiency of WRF on composite substrates (a) BpPApPp, (b) BpPAp, (c) PApPp and (d) BpPp.

3.8. Substrate component conversion efficiency of selected microbes on composite substrates

The performance of each selected fungal strain on TOS and TRS of composite substrates (dry and wet forms) showed that high substrate utilization efficiency heralded their growth and development (Fig. 4a–d). The trend was true for TOS and TRS (wet and dry) for all selected strains except M6; combination of all three substrates increased efficiency compared with dual membered substrates (Fig. 4a). However, such differences were not significant owing to comparable compositions of TRS and TOS. An exception to this was M6 that exhibited a significant difference when compared with other microorganisms. Low efficiency was obvious on TRS dry form by all fungal strains when compared with their wet equivalent; though, such effect may not directly influence biomass production (Narasimha et al., 2006).

4. Conclusion

All the sole and composite substrates supported fungal growth and development through the availability of fermentable sugar. Fungal biomass was high in the three fruit wastes, and WRF performed efficiently by consuming TRS and TOS for improved biomass production. Wet and dry sole and composite substrates provided an adequate carbon source for fungal growth, development and product synthesis. Fungal strains proved to be able to metabolize simple sugar components of the substrate by converting them into biomass. Substrate utilization constant was high in all microbial treatments, as fungal strains metabolized sugars contained in the substrates (wet and dry).

Acknowledgements

The research was financially supported by a research Grant EDW B13-004-0889 approved by the Research management Center (RMC), International Islamic University Malaysia. The authors are grateful to the RMC and Department of Biotechnology Engineering, IIUM for supporting and providing the laboratory facilities.

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