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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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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.

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

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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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