



Food waste compost as an organic nutrient source for the cultivation of *Chlorella vulgaris*

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ABSTRACT

The present study investigates the prospective of substituting inorganic medium with organic food waste compost medium as a nutrient supplement for the cultivation of *Chlorella vulgaris* FSP-E. Various percentages of compost mixtures were replaced in the inorganic medium to compare the algal growth and biochemical composition. The use of 25% compost mixture combination was found to yield higher biomass concentration (11.1%) and better lipid (10.1%) and protein (2.0%) content compared with microalgae cultivation in fully inorganic medium. These results exhibited the potential of combining the inorganic medium with organic food waste compost medium as an effective way to reduce the cultivation cost of microalgae and to increase the biochemical content in the cultivated microalgae.

1. Introduction

Over one billion tonnes of food wastes are produced every year and these food wastes either require treatments or are disposed in landfills. The transformation of food waste into value-added products through biological means such as composting is an environmental friendly method for effective organic waste management (Cerdeira et al., 2018). The end product of composting is a stabilized organic matter rich in humic substances which contains essential plant nutrients to be used as organic fertilizer (Zhou et al., 2016). Compared with inorganic fertilizer, the production of organic fertilizer can achieve a saved emissions of 4–81 kg CO₂ per tonne of food waste and 4–67 kg CO₂ per tonne of garden waste (Boldrin et al., 2009). The nitrogen and phosphorus content in compost produced from food waste can be used in agriculture for food production. Moreover, food waste contains high content of organic components such as carbohydrates, proteins, lipids and organic acids which makes it a reasonable feedstock for cultivating microorganisms (Zeng et al., 2018). The recycling of nutrients from food waste could potentially replace the non-renewable feedstock in various microorganisms' cultivation, which will lead to the alleviation

of the global energy crisis. This will contribute to better nutrient use efficiencies and a reduction on the pollution that would otherwise be caused by inorganic fertilization.

Microalgae has been cultivated and applied extensively due to its promising properties as a biomass feedstock. Microalgae has high growth rates, less competition with terrestrial crops and a wide distribution. The production of biofuel from microalgae was found to be much greater than food crops and non-food crops feedstock, in addition to the shorter cultivation time need for microalgae and its renewability (Chia et al., 2017). Apart from that, its untapped potential to produce multiple valuable products in addition to biofuels has aroused the interest of various industries (Chew et al., 2017; Yen et al., 2013). The expanding interest and commercial development of products with functional compounds puts focus on microalgae metabolites which can be used in food, nutraceutical and pharmaceutical industries (Cuellar-Bermudez et al., 2015). Microalgae can produce a wide range of functional ingredients which include proteins, polysaccharide, natural pigments, vitamins, essential amino acids and other bioactive molecules. The functional protein biomolecules from microalgae are especially of interest for their health-promoting effect and biological properties like

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antioxidants, anticancer, anticoagulant, antitumor, antihypertensive and immune-modulatory activities (Samarakoon and Jeon, 2012).

The commercialization and scaling up of microalgae production for biomolecules extraction requires a large quantity of freshwater and high-cost nutrients. This creates the need to find a sustainable low-cost alternative medium which can provide similar nutrients needed for microalgae cultivation (Chia et al., 2018). Wastewaters have been used to cultivate microalgae since their growth can purify the wastewater before discharging them back into water bodies (Wang et al., 2016). Nevertheless, the utilization of wastewater poses a few uncertainties such as the possibility of contamination, presence of hazardous chemicals and varying nutrient contents which will affect the growth of microalgae. Another potential source of nutrient medium is the use of food waste medium for microalgae cultivation. Food waste hydrolysates have successfully been used for the cultivation of microalgae and showed good growth of microalgae (Lau et al., 2014; Pleissner et al., 2013). A coupled system of dark fermentation and microalgae cultivation was also performed using substrates of food waste and provided a sustainable route for efficient energy recovery (Ren et al., 2018). The major pollutant in food waste is mainly ammonium and phosphate, and the concentration of these pollutants can be reduced and utilized effectively to grow microalgae, bringing about a minimization of the contamination level in water (Cerdeira et al., 2018; Zhang et al., 2018). The integration of food waste compost medium and inorganic medium could potentially substitute the major nutrients such as nitrate and phosphate needed for microalgae cultivation. This will lead to a more sustainable industrial microalgae growth process for subsequent bio-refinery.

The aim of this study was to investigate the potential of using food waste compost as a nutrient source for the cultivation of *Chlorella vulgaris* FSP-E. The feasibility of replacing inorganic fertilizers with organic fertilizers for microalgae cultivation were explored by using different concentrations of food waste compost mixture and to determine the algal biomass accumulation from each combination. The optimum concentration mixture was identified and the biochemical compositions in microalgae, such as lipid, protein and carbohydrates contents were extracted from the microalgae biomass to evaluate the productivity of the biomass for fuel, food and feed productions.

2. Materials and methods

2.1. Microalgae strain and medium compositions

A high protein-yielding microalgae strain *C. vulgaris* FSP-E used in this study was isolated from a freshwater area located in southern Taiwan. The microalgae was cultivated in a modified version of BG-11 medium containing 1.5 g NaNO₃, 0.03 g K₂HPO₄, 0.075 g MgSO₄·7H₂O, 0.006 g citric acid anhydrous, 0.02 g Na₂CO₃, 0.036 g CaCl₂·2H₂O, 0.006 g Ammonium iron (III) citrate, 0.001 g EDTA·2Na, 0.00286 g H₃BO₃, 0.00181 g MnCl₂·4H₂O, 0.000222 g ZnSO₄·7H₂O, 0.00039 g Na₂MoO₄·2H₂O, 0.000079 g CuSO₄·5H₂O, and 0.000049 g Co (NO₃)₂·6H₂O in 1 L of deionized water. The pre-culturing was done in an indoor photobioreactor (PBR) which consist of a 250 ml glass vessel containing 200 ml of medium, aerated with compressed air and equipped with external LED light sources on both sides of the vessel. The PBR was operated at room temperature with agitation. The light intensity on the vessel wall of the PBR was adjusted to 200 μmol/m²/s using a LI-250 light meter with a LI-190SA pyranometer sensor (LI-COR, Inc., Lincoln, USA). Air was filtered (0.45 μm) and mixed with CO₂ to give a CO₂ concentration of 2.0%. The culture broth was aerated continuously at a rate of 200 ml/min (0.2 vvm, volume gas per volume media per minute). Similar PBR operating conditions were used for the subsequent batch cultures but in 1 L of medium. All cultures were performed in duplicate and average values were reported.

2.2. Microalgae cultivation with food waste compost

Food waste co-composted with green waste compost was provided by the Campus Services of University of Nottingham Malaysia Campus. The compost was generated using the aerated windrow composting method and produced using food waste obtained from the cafeteria in the campus. The compost solutions were prepared by immersing 10 g of compost in 200 ml of water into individual 250 ml Erlenmeyer flasks. The flasks were placed in a shaker incubator and rotated at 150 rpm in room temperature for 24 h. The compost mixture was filtered through glass microfiber filters (934H, Whatman, USA) to remove large particles and indigenous bacteria. The organic compost mixture rates applied for microalgae cultivation was 15% organic (85C), 25% organic (75C), 35% organic (65C), 50% organic (50C), 75% organic (25C), 100% organic (100F), 100% organic with 75:25 compost solution to water dilution (75F), using 100% inorganic (100C) as control. The elemental composition of the food waste compost are 30.84 ± 0.45% C, 4.10 ± 0.04% H, 2.68 ± 0.01% N and 0.84 ± 0.15% S. The resulting organic compost medium contains a total NO₃ of 360 ± 4 mg/L, total P of 208 ± 12 mg/L, pH range of 7.2–7.4 and a dark brown colour of maximum absorbance (λ_{max}) of around 284–293. Sufficient compost mixture volumes were generated to fulfill the respective rates.

2.3. Determination of microalgae cell concentration

The microalgae biomass concentration was determined by measuring the optical density at 680 nm using a UV–Vis spectrophotometer (UV-1800, Shimadzu). The dry cell weight of the microalgae biomass was obtained by filtering 5 ml aliquots of culture using cellulose acetate membrane filter (0.45 μm pore size, 25 mm in diameter). Each loaded filter was dried at 105 °C until the weight was constant. The dry weight of the blank filter was subtracted from that of the loaded filter to obtain the microalgae dry cell weight (DCW). The OD₆₈₀ values were converted to biomass concentration via calibration between OD₆₈₀ and the DCW (1 OD₆₈₀ = 0.23 g/L DCW). To eliminate the influence of the medium colour, a blank control of each medium combinations was prepared and quantified. The microalgae culture broth for each mixture combination was centrifuged at 6000 rpm for 15 min and the clear supernatant was removed. The pellet was lyophilized for 24 h. The resulting dry microalgae biomass was used for further characterization studies.

The biomass productivity (P_b) and specific growth rate (μ) of the microalgae in the exponential growth were calculated using the following equations:

$$P_b (\text{mg/L/d}) = (N_2 - N_1) / (t_2 - t_1) \quad (1)$$

$$\mu (\text{d}^{-1}) = \ln(N_2/N_1) / (t_2 - t_1) \quad (2)$$

where N₁ and N₂ are dry cell weight (g/L) at time t₁ and t₂, respectively.

2.4. Quantification of nitrate and phosphorus content in medium

Nitrate content was determined using the spectrophotometric method (Collos et al., 1999). The microalgae samples were collected from the bioreactor and centrifuged at 6000 rpm for 5 min. After centrifugation, the samples were diluted with deionized water and measured at the optical density of 220 nm using a UV–Vis spectrophotometer. Pure sodium nitrate (NaNO₃) at different concentrations was used for generating the standard curve. Phosphorus content in the medium was determined using atomic absorption spectroscopy (AAS, PerkinElmer AAnalyst 400) (Hoft et al., 1979). 30 ml of each mixture combination mediums were heated with occasional stirring to ensure the liberation of phosphorus. The spectrophotometer was set to isolate light at the phosphorus resonance line of 213.6 nm. Dibasic ammonium phosphate of known concentrations were used to establish the standard

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