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Sequential fractionation of value-added coconut products using membrane processes

Ching Yin Ng [a], Abdul Wahab Mohammad [a,b,*], Law Yong Ng [a], Jamaliah Md Jahim [a,b]

[a] Department of Chemical and Process Engineering, Faculty of Engineering and Built Environment, Universiti Kebangsaan Malaysia, 43600 Bangi, Selangor, Malaysia

[b] Research Centre for Sustainable Process Technology (CESPRO), Faculty of Engineering and Built Environment, Universiti Kebangsaan Malaysia, 43600 Bangi, Selangor, Malaysia

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ABSTRACT

The coconut waste-skimmed coconut milk was employed for sequential fractionation using UF and NF membranes to produce value-added products (coconut proteins, plant hormones – kinetin and zeatin). The retention factors achieved by UF membrane (PS10): albumin (0.9822 ± 0.0799) and globulin (0.9975 ± 0.0783); NF membrane (NF1): kinetin (0.9238 ± 0.0345) and zeatin (0.9511 ± 0.0355). Coconut protein powder was obtained after spray-drying process using concentrated coconut protein (UF retentate). SDS-PAGE showed that molecular weights of the coconut proteins were 17, 34, 55 and 150 kDa. Proximate and HPLC analyses revealed that the obtained samples were enriched with basic nutrients and well-balanced amino acids composition, respectively.

Introduction

Coconut (*Cocos nucifera L.*) is an important palm species around the region of Asia Pacific and South East Asia. It is commonly used as the cooking ingredient in these countries. Besides, coconut is also famous with its versatile applications [1]. One of the renowned coconut products is virgin coconut oil (VCO). VCO has been recognized as the healthiest crop oil and can be extensively employed in various fields such as food, beverage, medicinal, pharmaceutical, nutraceutical, cosmetic, etc. [2]. The high-valued fatty acids and anti-viral properties of the VCO have increased the demand of VCO worldwide and thus its production [3]. Large amount of valuable by-products have been discarded during the production of VCO. This has been considered as a wastefulness which also creates environmental issue in some areas near to the coconut processing plant. These discarded by-products still contain some high-valued nutritional components such as coconut protein, plant hormones, vitamins, minerals, amino acids and so forth, which are highly beneficial to the consumers [4]. These valuable components can be potentially extracted by suitable separation techniques or other appropriate processes to fully utilize the raw materials employed during the VCO production. Full utilization of these by-products would definitely increase the profit of the coconut industry.

The skimmed coconut milk is the main by-product after the coconut oil extraction process. The skimmed coconut milk consists of approximately 70% of the total proteins, carbohydrates, sugars, vitamins and minerals [5]. Through fractionation and concentration processes, the by-products from skimmed coconut milk can be employed in functional food, food supplement and food formulation. In addition to the compounds mentioned above, there are some other smaller-size minor components exist in the skimmed coconut milk. For example, there is a group of plant hormone called cytokinins which consists of kinetin, zeatin and traces of other compounds [6,7]. The kinetin and zeatin have been recognized as valuable compounds which possess anti-viral, anti-bacteria and anti-aging properties. They can be employed to stimulate the cell division of human body [1,8]. If the kinetin and zeatin from skimmed coconut milk can be separated and concentrated using appropriate processes, they will have huge potential to be applied in many applications such as medicinal, pharmaceutical and nutraceutical sectors [9,10]. The great versatility of these valuable compounds from the

* Corresponding author at: Department of Chemical and Process Engineering, Faculty of Engineering and Built Environment, Universiti Kebangsaan Malaysia, 43600 Bangi, Selangor, Malaysia. Tel.: +603 89216102; fax: +603 89252546.

E-mail addresses: echingyin@gmail.com (C.Y. Ng), wahabm@eng.ukm.my, awm.ukm@gmail.com (A.W. Mohammad), nglawyong@yahoo.com (L.Y. Ng), jamal@eng.ukm.my (J.M. Jahim).

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coconut has attracted the attentions of many researchers to produce more profitable value-added coconut products.

Many attempts have been tested to separate the valuable compounds in agricultural or food products such as distillation, evaporation, chemical extraction and so forth [11,12]. However, the conventional separation methods which involve heat treatment will deteriorate some thermal sensitive compounds like proteins [13,14]. Membrane filtration is an environmental friendly process compared to conventional methods [15]. The advantages of membrane filtration technique including low labor cost, low maintenance cost, less space consuming, energy saving, feasible to scale up, no chemicals used, no heat treatment, able to prolong the shelf-life of product, can retain the natural flavor and aroma of raw materials [16,17]. Due to the numerous advantages possessed by membrane filtration, it is used widely from resource recovery to wastewater treatment [18–21]. Therefore, the membrane separation technique is proposed in this study as one of the most probable ways to fractionate the skimmed coconut milk to produce those value-added products.

In food technology field, the fractionation method using membrane technology has been well developed since past decades. For example, the fractions of fat and volatile compounds from goats' milk cheese has been successfully obtained using the fractionation method by sequential ultrafiltration and nanofiltration processes [16]. Compounds with varying molecular sizes managed to be retained by different membrane categories such as microfiltration, ultrafiltration and nanofiltration. In the fractionation of dairy products, ultrafiltration [22,23] and nanofiltration [24–26] are always employed in order to obtain the valuable milk components. The protein hydrolysates can be efficiently separated by nanofiltration process due to the size exclusion mechanism and Donnan effect. In beverage industry, the membrane filtration technique is a common method to clarify and concentrate the fruit juice. Combination of various membrane types have been employed to enhance the quality of the produced juice [27–29]. However, there is not much study about the fractionation of coconut milk using membrane technology to obtain the valuable compounds. If the membrane processes can be employed to fractionate and concentrate the valuable nutrients from coconut waste effectively, this will contribute to a great breakthrough in the coconut industries. Besides that, implementation of spray-drying process could further enhance the quality of value-added products produced (such as coconut protein). Spray-drying is the most widely used commercial dehydration method owing to its short duration of heat-contact and high rate of evaporation, which can produce high quality products. Powders with precise specifications can be generated using spray-drying process in continuous operation [30].

The objective of present study is to determine the capability of sequential ultrafiltration and nanofiltration processes to fractionate the value-added products from skimmed coconut milk. In addition, the membrane performance will be evaluated and discussed in terms of normalized flux decline and solute retentions for protein, kinetin and zeatin compounds.

Materials and methods

Materials

Coconut of *Malayan Tall* variety has been selected for this research work. To determine the concentration of albumin and globulins proteins in UF streams, standard bovine serum albumin (BSA), standard immunoglobulin (IgG) and protein assay dye reagent concentrate were supplied by Bio-Rad (USA). During the UF process, polysulfone (PS) membrane with 10 kDa (supplied by Koch, USA) was employed. The employed NF membrane was manufactured by Amfor Inc., China. High performance liquid chromatography (HPLC) analysis was used to determine the concentration of kinetin and zeatin in NF streams. The standards of kinetin and zeatin were supplied by Sigma-Aldrich (Steinheim, Germany). Kinetin and zeatin solutions with the concentration ranging from 10 to 500 μM were prepared by dissolving the kinetin and zeatin standards in HPLC-grade methanol. These varying concentrations of kinetin and zeatin standards were stored at or below temperature of 4 °C. The chemicals used for HPLC analysis including HPLC-grade methanol (Tedia, USA), formic acid (Tedia, USA) and triethylamine (TEA) (Merck, Germany). Buffer solutions employed in the HPLC analysis has been prepared using methanol and 0.1% formic acid with pH 3.2.

Preparation of skimmed coconut milk

The milky white color fresh coconut milk was produced from solid grated coconut endosperm using a coconut extraction machine. Then, the extracted fresh coconut milk was filtered through micro-size sieving cloth to remove the bigger-size particles. Simple pasteurization process was conducted prior to the membrane filtration process in order to prolong the shelf-life of extracted coconut milk. The fresh coconut milk was heated at 60 °C for 15 min. This pasteurization method was conducted in order to reduce the microbial loading up to 10% according to the Hagenmaier method [31]. After that, the pasteurized coconut milk was poured into 125 L capacity cream separator machine (Elecrem, France) to separate the coconut cream/fat from the fresh coconut milk. There are two outputs from the cream separator: concentrated coconut cream and skimmed coconut milk. The produced skimmed coconut milk needs to be stored at a cold condition (0–4 °C) before it is used as a feed solution for the membrane filtration process. Membrane filtration processes (UF and NF membranes) were then employed to fractionate the skimmed coconut milk to produce the desired value-added coconut products.

Membrane filtration process

The polysulfone (PS) membrane was employed in the UF process. Polysulfone membrane was employed owing to its several superior properties such as high material toughness, good stability at high temperature, high resistant to various solvent, good resistant to wide pH range (pH 2–13) and low protein binding tendency [32]. The molecular weight cut-off (MWCO) value of this PS membrane is 10 kDa. The abbreviation of the UF membrane used in this experiment is PS10. NF1 membrane was employed in the NF filtration process. The membrane material of NF 1 is polyamide. Polyamide provides the desired properties such as high rejection of undesired materials (like salts), good mechanical strength and high filtration rate at low pressure. Prior to the filtration process, the membranes were soaked overnight to remove any preservative layer and dirt particle. Membrane compaction step was carried out at a higher pressure value (greater than operating pressure) for each employed membrane in order to enhance the permeate flux and membrane permeability [33]. Ultra-pure water was used during the membrane permeability test.

In this study, a cross-flow system was used in the UF process. The cross-flow system was equipped with a 4 L stainless steel jacketed feed tank (embedded with a mixer) and a variable feed pump (Hydra-cell Pump, Mn, USA). The temperature of feed solution was controlled by the circulating water within the jacketed feed tank. The cross-flow velocity (CFV) of feed and the trans-membrane pressure (TMP) was controlled using a feed flowmeter (F-400, Blue-White, USA) and a permeate needle valve