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Development of Filtration System for the Collection of Soaking Water during Bedak Sejuk Preparation

(Pembangunan Sistem Penurasan bagi Pengumpulan Air Rendaman Beras sewaktu Penyediaan Bedak Sejuk)

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ABSTRACT

"Bedak Sejuk" or fermented rice powder is a traditional cosmetic product used by locals that is made from fermented rice. To start the process, the rice grains are soaked in water at 1:1 (w:v) ratio and continuously soaked until the rice gains fully dissolved into flour paste slurry. The soaking water was discarded at end of the fermentation process although it is believed valuable active components still exist in the water. Thus, the aim of the present study is to develop filtration system to collect the soaking water and to analyze active components presence. Local rice grains were soaked in the water in a container and within 14 days interval, the soaking water was replaced. The soaking water collected was then filtered using muslin cloth, followed by membrane filter size of 0.45 and 0.20µm. The filtered soaking water was analyzed for amino acid analysis using HPLC. Sample (0.1 ml) was poured onto the nutrient agar plate for microbial growth to determine the effectiveness of the filtration system. Based on these observations, it can be concluded that the rice soaking water during bedak sejuk preparation content were rich in amino acids (15 types of amino acids detected) with the highest concentration were lycine, glutamic acid and arginine – 0.11, 0.12 and 0.16% respectively. After the filtration using 0.20µm, no microbial growth detected suggested that it can be applied directly onto the skin as organic cosmetic product that benefits the skin.

Keywords: Rice grains; organic cosmetic; Bedak sejuk; rice soaking water; amino acids

ABSTRAK

'Bedak sejuk' adalaha produk kosmetik tradisional tempatan yang dihasilkan dari fermentasi beras. Untuk memulakan proses, beras direndam dengan air pada ratio 1:1 (b:i) sehingga ianya terlarut menghasilkan kaldu tepung. Air rendaman dibuaang pada hujung proses fermentasi adalah dipercayai mengandungi bahan aktif bernilai. Jadi, tujuan kajian ini untuk membangunkan proses penurasan yang dapat mengumpul air ini dan melihat kehadiran bahan aktif tersebut. Beras tempatan direndam di dalam bekas tertutup dan setiap 14 hari, air rendaman akan ditukar. Air rendaman ini dituras menggunakan kain muslin, diikuti penuras membran pada saia 0.45 dan 0.20 µm. Air terturas akan diuji dengan kehadiran asid amino menggunakan HPLC. Sampel air juga akan disebarkan pada piring petri agar nutrien untuk melihat pertumbuhan mikroorganisma. Daripada permerhation, air rendaman adalah kaya dengan asid amino (15 jenis asid amino dikesan) dengan kepekatan tertinggi bagi lysin, asid glutamic dan arginine – 0.11, 0.12 dan 0.16 % masing-masing. Selepas penurasan 0.20µm tiada pertumbuhan mikroorganisma dikesan membolehkan ianya digunakan secara terus sebagai bahan kosmetik organik baik untuk kulit.

Kata kunci: Beras; kosmetik organik; Bedak sejuk; air rendaman beras; asid amino

INTRODUCTION

Bedak sejuk is a Malaysia traditional cosmetic product that is made from rice using natural fermentation. During the preparation of bedak sejuk, the rice are soaked in water for certain period of time that could lasting in months. Traditionally, the soaking water normally replaced after few weeks or months and simply thrown away. This soaking water is believed to contain active ingredients that are beneficial to skin and can be used as cosmetic applications. However, the use of rice soaking water directly onto the wearer's skin is not established to be safe due to the presence of microorganism

through the natural fermentation process or any toxic ingredients. The filtered soaking water has the potential to enable it to be applied directly onto the skin as a skin care product. But the usage of rice water as beauty products has been long history amongst Asian women. In Japan, a study stated that women in the Heian court brushed their hair every day using water soaked rice called Yu-Su-Ru (Satoshi et al. 2010) and found out the rice water reducing surface friction, make hair smoother, shinier, stronger and increasing hair length. It is thought to be rich in vitamins and minerals such as amino acids, B vitamin, vitamin E and antioxidants. High antioxidant contents of rice bran oil such as ferrulic acids, gamma-oryzanol and phytic acid are common in cosmetic industry (Coppini et al. 2001) and for treatment of skin diseases (Ha et al. 2018). Apart from that, amino acids such as glutamine, arginine and lysine are also believed to exist in the rice soaking water during the cold powder production (Kalin et al. 2013; Sahelian 2017).

Bioactive ingredients in rice bran are also claimed to poses anti-aging activity (Kanlayavattakul et al. 2016; Chaikul 2016) and it can be developed topical gel formulation using rice water (Marto et al. 2018). Since the main component in rice is starch, it is also being used to treated dermatitis or skin diseases (De Paepe 2002) and could reduce inflammatory activity in vivo due to its polysaccharides content (Garbacki 2018). Starch also is a biodegradable which is safe to be applied in the pharmaceutical industry (Anwunobi & Emeje 2011).

But there is no previous studies have been reported on the rice soaking water components during the preparation of bedak sejuk in Malaysia. Thus, the objective of this study is to develop a filtration system to collect the soaking water and to analyze the amino acids present in the soaking water at the beginning and ends of fermentation process of bedak sejuk production.

METHODOLOGY

SAMPLE PREPARATION

Local rice brand (250g), were soaked in 250 ml of tap water and allowed to ferment naturally at ambient temperature (23-25°C) for 14 days. After 14 days, rice grains were filtered using a muslin cloth. The remaining rice granules on the cloth were weighed and soaked again in tap water with ratio of 1:1 (w:v) (Johar et al. 2018). These procedures were repeated for four more times which resulted in 70 days soaking period. Samples of soaking water were collected at 14 days interval while the supernatants are collected every 24 hours to determine the growth profile of fermentation microorganisms.

GROWTH PROFILE

Soaking water supernatant taken daily during the 14 days intended to measure the dry weight of the sample. As a result of the weights of the sample showed the presence of microorganisms that are present during the fermentation process. 30ml water soaked rice is taken using a 10 ml syringe, inserted into three test tubes and filtered using Whatmann filter paper. Filter paper was weighed using electronic scales and weight readings before and after the filter paper filtered also taken. Subsequently, the filter paper is inserted into the oven and dried at 105°C. The data are tabulated and the growth profile was plotted.

FILTRATION SYSTEM

Filtration systems developed using 3 steps: first, coarse filter cloth (Muslin cloth, 0.2mm) followed by 0.45μm and 0.20μm using syringe filter. The soaking water will be filtered first using muslin cloth to separate large impurities so filter pores are not clogged. Water samples on each filtrations were innoculated onto agar plate to detect any microbes presence.

PLATE TEST

Nutrient agar (Oxoid Ltd) plates are used to detect the growth of microorganisms for each filtrations samples. After inoculation, the plate were incubated at 30°C and observed for 3 days.

ANALYSIS OF AMINO ACIDS

Samples which filtered using 0.45 and 0.2 μm syringe filter are analyzed by using HPLC (Water Corporation 2014). Amino acid analysis is performed to Waters Accq Tag/ HPLC carried out by Unipeq Laboratory, Universiti Kebangsaan Malaysia following all standards procedures of measurement of amino acids.

RESULTS AND DISCUSSION

To ensure the fermentation process occurs, microorganism growth data have been recorded for 14 days (Figure 1). The microbial growth profile indicated no lag phase suggesting the microbes in the fermented soaking water do not require a long time to adapt to a new environment. Cell density continues to increase up to 8 days before starting the stationary phase. This growth profile shows a stationary phase until day 14.

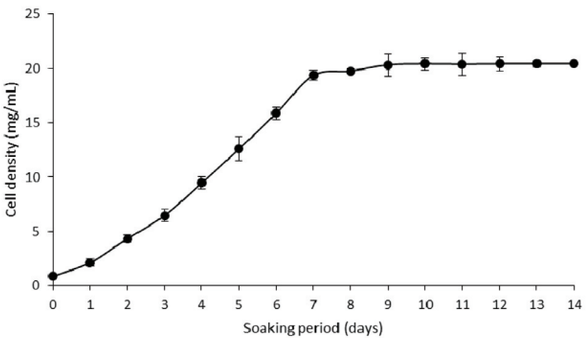


FIGURE 1. Growth profile of microorganisms in the fermentation of local rice for 14 days

The growth profile result supported with the eyes observation on the water surface conditions. Based on the observation, a whitish layer was formed on the surface of the soaking water due to fermentation when the microorganism consume the rice (organic substances as part of their metabolic processes (Chinees 2003). The layer was becoming thicker until the third soaking period before started to become thinner until the end of the soaking process. The figures of the layer on the surface of soaking water for each soaking process are shown in Table 1. It is suggested that natural microorganisms played a role in the presence of this layer (Dzulfakar et al. 2015a); Dzulfakar et al. 2016b). These included lactic acid bacteria family, mold and fungi (Eifert et al. 2009; Perez-Diaz et al. 2013; Breidt et al. 2013).

DEVELOPMENT OF FILTRATION SYSTEM

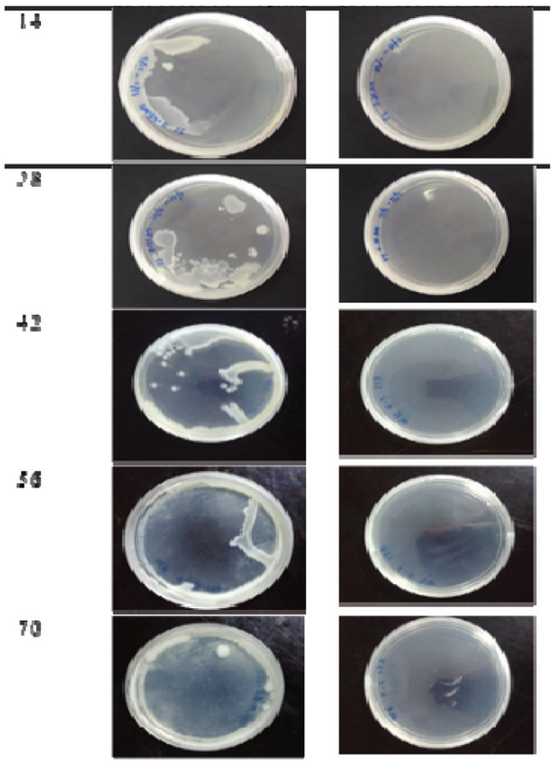
Table 2 shows the growth of microorganisms on the agar plate after the soaking water were filtered using a 0.45 µm and 0.2 µm size filter papers. Based on the results in Table 2, filter papers at size 0.45 µm is unable to get rid of the microorganisms as growth of various colony sizes were observed. Since the fermentation process occurs naturally, at 0.45µm only managed to filter part of the bigger microbes such as yeast, molds and fungi. But no growth of microorganisms were observed after rice soaking water filtered using a syringe filter size 0.20 µm. It is suggested all microorganisms were fully filtered using this size, thus suggesting the filtrate is safe to be applied directly to the skin. This is true since filter at size 0.20 is normally used for sterile preparation (Millipore 2000).

TABLE 1. Rice soaking water surface conditions based on days of soaking

Day	Surface conditions	Observation
0		None
5		Thin layer formed and small bubbles presence on the water surface
10		Thick layer is formed
14		Thicker layer is formed

Table 2 shows the growth of microorganisms on the agar plate after the soaking water were filtered using a 0.45 µm and 0.2 µm size filter papers. Based on the results in Table 2, filter papers at size 0.45 is unable to get rid of the microorganisms as growth of various colony sizes were observed. But no growth of microorganisms were observed after rice soaking water filtered using a syringe filter size 0.20 µm. It is suggested all microorganisms were fully filtered using this size, thus suggesting the filtrate is safe to be applied directly to the skin although there is reported *Pseudomonas aeruginosa* could passed through a 0.22 µm pore size filter (Hesegawa et al. 2003). For safest used, membrane with smaller sizes (nano) might be useful to be used (Teoh et al. 2017).

TABLE 2. The growth of microorganisms on the agar plate after filtration

Day	Type and Filter Pore Size	
	Syringe Filter (Sartorius 0.45 µm)	Syringe Filter (Sartorius 0.20 µm)
14		
28		
42		
56		
70		

AMINO ACID ANALYSIS

Table 3 shows the results of amino acids in the soaking water after 14 days and 70 days. Out of 17 amino acids examined, 15 amino acids were detected in the filtered soaking water. In rice itself, sixteen free amino acids can be detected in rice (Thomas et al. 2015). Amino acids that cannot be detected in the sample are histidine and hydroxyproline because its concentration is less than 0.01% in a 100 gram sample of soaking water. The highest amino acid content on day 14 is lysine with concentration of 0.11% (w/w) and the percentage remained until the 70th day. While on day 70, the highest amino acid content is glutamine with concentration of 0.17% (w / w). The 15 amino acids detected shows the average percentage increase of 0.06% on day 70 compared to day 14. It is suggested that the content of amino acids increases with the length of time soaking in the production of bedak sejuk as can be seen in Figure 2. Four amino acids where found the highest amongst 15 detected are glutamic acid, arginine, alanine and lysine.

TABLE 3 Type of amino acid and its concentration in rice soaking water

Type of amino acids	Amino acid concentration (g/g)		Amino acid concentration w/w (%)
	Day 14th	Day 70th	
Hydroxyproline	ND (< 0.001)	ND (< 0.001)	ND (< 0.001)
Aspartic acid	ND (< 0.001)	0.094	0.094
Serine	ND (< 0.001)	0.0795	0.0795
Glutamic acid	0.0495	0.170	0.1205
Glycine	0.0295	0.074	0.0445
Histidine	ND (< 0.001)	ND (< 0.001)	ND (< 0.001)
Arginine	ND (< 0.001)	0.159	0.159
Threonine	0.0245	0.048	0.0235
Alanine	0.0405	0.110	0.0695
Proline	0.0175	0.072	0.0545
Tyrosine	ND (< 0.001)	0.0835	0.0835
Valine	0.0235	0.074	0.0505
Methionine	0.0265	0.070	0.0435
Lysine	0.1115	0.113	0.0015
Isoleucine	ND (< 0.001)	0.058	0.058
Leucine	0.026	0.0905	0.0645
Phenylalanine	0.014	0.0635	0.0495

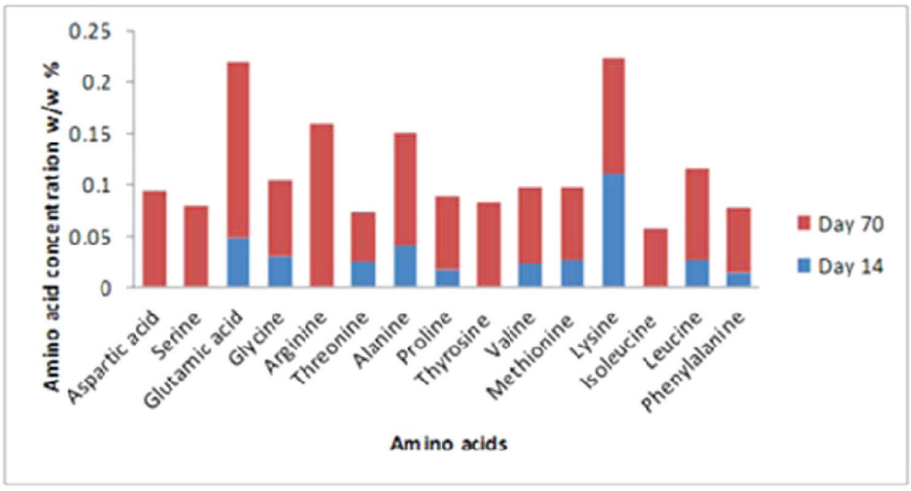


FIGURE 2. The profile of amino acids and its concentration in the rice soaking water

Amino acids that are popular in the cosmetic (Lubkowska et al. 2010) and pharmaceutical industries are arginine, glycine, methionine and glutamine and according to the results of the study, the four amino acids were detected in the soaking water. Arginine, tyrosine, glutamine and lysine are also reported as the main amino acids used in cosmetic industries (Ha et al. 2018). The functions of the amino acids in cosmetics are as an antioxidant and as an ingredient to maintain moisture in the skin by providing a protective layer (Kalin et al. 2013). This support the theory that soaking water is good for the skin. Due to the high content of glutamine detected in the soaking water, it could be used as cosmetic application.

CONCLUSION

From all the results shown and discussed, it can be conclude that the three filtration system is effective to remove the growth of microorganism suggesting it is safe and enable it to apply directly onto the skins. The presence of amino acids normally used in cosmetic industry after the filtration indicated that it has a potential to be used in in preparation of cosmetic application. But more test on toxicity and skin irritation need to be explored in further research.

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