INFLUENCE OF CHURNING AND THAWING TEMPERATURES ON YIELD AND QUALITY OF VIRGIN COCONUT OIL

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INFLUENCE OF CHURNING AND THAWING TEMPERATURES ON YIELD AND QUALITY OF VIRGIN COCONUT OIL

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A thesis submitted in fulfilment of the requirements for the award of the degree of Master of Engineering (Bioprocess)

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Dedicated to my beloved family, my husband, Sulaiman Ngadiran, and my children, Muhammad Aiman and Zara Aisyah

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ABSTRACT

The churning and thawing temperatures affect the yield and quality of coconut oil in integrated wet process. The thesis reports on the selection of *Cocos* nucifera species that contributed the highest oil content, followed by the optimization of integrated wet process. The highest oil content from coconut varieties was rendered by Soxhlet extraction. The response surface methodology (RSM) was used to determine the effects of churning (T_c) and thawing (T_t) temperatures by using the central composite design (CCD). The experimental design was limited to T_c (0.5-17.5 °C ± 1.0) and T_t (25.0 - 67.2 °C ± 1.0). The chemical constituent of ferulic acid was used to represent the total phenolic content in virgin coconut oil. The ferulic acid was determined by using high performance liquid chromatography (HPLC) and its colour was analysed by colorimeter. The free fatty acid (FFA) was determined by Association of Analytical Communities (AOAC) 940.28 method. The highest oil content ($p \le 0.05$) was obtained from West African Tall (WAT), contributed $69.07 \pm 0.06\%$ compared to other species of Mawa, Matak, Rennel Island Tall, Maren and Tagnanan Tall comprising 64.62 ± 0.30%, $63.45 \pm 0.37\%$, $63.42 \pm 0.30\%$, $59.68 \pm 0.01\%$, $58.12 \pm 0.03\%$ of oil, respectively. From the experiments, the oil yield and ferulic acid content showed the effects on the studied parameters. The equation of the responses were Oil Yield $= 24.52 - 1.53A + 4.10B - 4.01A^2 - 1.73B^2 - 3.79AB$ and Ferulic Acid = 7.17 - $0.042A + 0.22B - 0.28A - 0.59B^2$, respectively, where A (churning temperature) and B (thawing temperature). For FFA and colour implied the mean responses to the studied parameters corresponding with 0.11% and 0.55 yellowness. From the optimum point at T_c, 7.4 $^{\circ}$ C and T_t as 50.4 $^{\circ}$ C, the oil yield for WAT was 31.44 ± 0.24 % (w/w), $11.3x10^{-2} \pm 0.11$ ppm for ferulic acid, 0.10 ± 0.08 % for FFA and 0.04 yellowness for colour. The moisture content of the final oil $0.11 \pm 0.15\%$ was complied with the virgin coconut oil standards. The optimization process showed an increment of oil yield and ferulic acid content in virgin coconut oil using integrated wet process.

ABSTRAK

Suhu penyejukan dan pencairan memberi kesan kepada hasil dan kualiti minyak kelapa di dalam integrasi proses basah. Tesis ini melaporkan pemilihan spesis Cocos nucifera yang menyumbang kepada kandungan minyak tertinggi, diikuti dengan pengoptimuman proses integrasi basah. Kandungan minyak tertinggi daripada pelbagai jenis kelapa disampaikan menggunakan ektraksi Sokhlet. Kaedah permukaan respons (RSM) digunakan untuk mendapatkan kesan suhu penyejukan (T_c) dan pencairan (T_t) dengan menggunakan rekabentuk pusat komposit (CCD). Ruang rekabentuk terhad kepada T_c (0.5-17.5 °C ± 1.0) dan T_t (25.0 - 67.2 °C ± 1.0). Sebatian kimia asid ferulik digunakan untuk mewakili kandungan jumlah fenolik di Sebatian kimia asid ferulik diperolehi dengan dalam minyak kelapa dara. menggunakan alat kromotgrafi cecair prestasi tinggi (HPLC) dan warna dianalisa menggunakan meter warna. Asid lemak bebas (FFA) diperolehi menggunakan kaedah (Kesatuan Komuniti Analitikal) AOAC 940.28. Kandungan minyak paling signifikan (p≤0.05) diperolehi daripada West African Tall (WAT), yang menyumbang sebanyak 69.07% \pm 0.06 dibandingkan dengan spesis lain iaitu *Mawa*, Matak, Rennel Island Tall, Maren dan Tagnanan Tall yang mengandungi 64.62 ± 0.30%, $63.45 \pm 0.37\%$, $63.42 \pm 0.30\%$, $59.68 \pm 0.01\%$, $58.12 \pm 0.03\%$ minyak, setiap satunya. Daripada ujikaji, hasil minyak dan kandungan asid ferulik menunjukkan kesan kepada parameter-parameter kajian. Persamaan bagi maklumbalas adalah Hasil Minyak = $24.52 - 1.53A + 4.10B - 4.01A^2 - 1.73B^2$ -3.79AB dan Asid Ferulik = $7.17 - 0.042A + 0.22B - 0.28A - 0.59B^2$ setiap satunya, di mana A (suhu penyejukan) dan B (suhu pencairan). Bagi FFA dan warna menerangkan respons min kepada parameter-parameter kajian selari dengan nilai 0.11% dan 0.55 kekuningan. Daripada titik optimum pada T_c, 7.4 °C and T_t sebagai 50.4 °C, hasil minyak untuk WAT adalah 31.44 \pm 0.24% (w/w), 11.3x10⁻² \pm 0.11ppm untuk asid ferulik, $0.10 \pm 0.08\%$ FFA, 0.04 kekuningan untuk warna. Kandungan kelembapan bagi minyak akhir, $0.11 \pm 0.15\%$ telah menepati piawaian minyak kelapa dara. Proses pengoptimuman mempamerkan peningkatan hasil minyak dan kandungan ferulik asid di dalam minyak kelapa dara menggunakan integrasi proses basah.

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LIST OF ABBREVIATIONS

AOCS	-	American Oil Chemist's Society
APCC	-	Asian Pacific Coconut Community
AR	-	Analytical grade
ASE	-	Accelerated solvent extraction
BS	-	British Standards
CCD	-	Central composite design
DHA	-	Docosahexaenoic acid
DNA	-	Deoxyribonucleic acid
DOE	-	Design of experiments
EPA	-	Eicosapentaenoic acid
FFA	-	Free fatty acid
HIV	-	Human immune efficiency virus
HPA	-	Health protection agency
HPLC	-	High performance liquid chromatography
Hrs.	-	Hours
IBD	-	Institute of Bioproduct Development
Max.	-	Maximum
MC	-	Moisture content
MCFA	-	Medium chain fatty acid
MCT	-	Medium chain triglycerides
MS	-	Malaysia Standards
MPOB	-	Malaysian Palm Oil Board
NA	-	Not available
ND	-	Non detectable
NFPA	-	National Fire Protection Association

PNS	-	Philippines National Standards
RBD	-	Refined, bleached deodorized
STOT RE 2	-	Specific target organ systemic toxicology repeated in HPA
Т	-	Temperature
VCO	-	Virgin coconut oil
WAT	-	West African Tall
wt.	-	Weight

LIST OF ABBREVIATIONS

AOCS	-	American Oil Chemist's Society
APCC	-	Asian Pacific Coconut Community
AR	-	Analytical grade
ASE	-	Accelerated solvent extraction
BS	-	British Standards
CCD	-	Central composite design
DHA	-	Docosahexaenoic acid
DNA	-	Deoxyribonucleic acid
DOE	-	Design of experiments
EPA	-	Eicosapentaenoic acid
FFA	-	Free fatty acid
HIV	-	Human immune efficiency virus
HPA	-	Health protection agency
HPLC	-	High performance liquid chromatography
Hrs.	-	Hours
IBD	-	Institute of Bioproduct Development
Max.	-	Maximum
MC	-	Moisture content
MCFA	-	Medium chain fatty acid
MCT	-	Medium chain triglycerides
MS	-	Malaysia Standards
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NA	-	Not available
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CHAPTER 1

INTRODUCTION

1.1 Research Overview

Coconut (*Cocos nucifera L.*) has been recognized for its oil since 4000 years ago in Ayurvedic medicine. The world demand for coconut oil has been expanding majorly in pharmaceutical, nutraceutical and cosmeceutical industries.

As overall, the coconut industry was almost stable globally during 2003 to 2008, in which Indonesia, India and Philippines were the major player in the world. Malaysia had produced approximately 45,000 tonne coconut oil during 2007-2012, that represented 1.3% from the world output of 3,500, 000 tonne (Euromonitor, 2013).

The conventional wisdom of saturated fat is bad has mislead the virgin coconut oil as unhealthy. The medium chain monolaurin exhibits bioactivities such as antimicrobial, antiviral, antiprotozoal and favor to the body immune system (Kabara, 2001). The coconut oil represented as saturated fat also able to increase the conversion of EPA (6%) and DHA (3.8%) compared to polyunsaturated fat (Enig, 1999). Due to its characteristic of high in saturated fatty acids especially the lauric

acid content, coconut oil is known to have the high resistance to oxidation that may inhibits rancidity due to its stability and functionality (O'Brien, 2003). In comparison, the virgin olive oil is the other type of oil which is known rich in monounsaturated fatty acid can be a good source of omega 3 and 6 (Servilli *et. al.*, 2012). Both oils play their specific role to fulfill the need of saturated and unsaturated sources for daily human consumption.

In plant, the phenolic constituent is acting as defense to infectious sources (Tuzen, 2003). In virgin coconut oil, the presence of phenolic acid that shows its antioxidant capacity allowing it to be used in cosmetic industry; and its capability to be converted into flavour constituent particularly from ferulic to vanilic acid (the main ingredient in vanila flavour) also enlighten the coconut industry (Faulds *et. al*, 1999). The minor compound of phenolic acid exhibits its antioxidant capacity of "antimutagenic", "antiproliferative" and "anticarcinogenic" benefits to human being. The major phenolic acid found in virgin coconut oil was ferulic acid of concentration 5.09 mg/kg oil; followed by the "vanilic", "*p*-coumaric" and "syringic" acids respectively (Marina *et. al.*, 2009^a). A study done by Song *et. al.* (2014) revealed that ferulic acid at concentration of 60 mg/kg gave a significant effect to stimulate the heart and liver cells against oxidative stress that correlated to diabetes in obese rats.

The virgin coconut oil (VCO) is processed from the fresh and mature kernel of coconut by mechanical or natural means with or without the application of heat, which does not alter the oil (APCC, 2009). The VCO is suitable for consumption without the need for further processing. The Philippines National Standards (2004) described further on the definition of VCO that must not undergoing the refining, bleaching, deodorizing (RBD) process. The process to extract the virgin coconut oil (VCO) that requires no heating may retain its biologically active compounds. These compounds provide nutritional and health benefits, mostly in preventing or minimizing chronic diseases (Marina *et.al*, 2009^b).

Meanwhile, the coconut oil (CO) is derived from the kernel, meat, or copra of the coconut (APCC, 2009). Usually, the copra is kept in a place to get the oil after few months and undergoing the RBD process to cover the unpleasant aroma and colour. The RBD oil that needs refining process may strips away some of the nutrients, which makes it different from the VCO. The RBD process is part of drying process resulting crude coconut oil with high free fatty acid (3-5%) that must be refined, bleached and deodorized before suitable to use (Marina *et.al*, 2009^b).

There are two main methods producing the virgin coconut oil, the wet and dry. By using the wet method, the oil is extracted from fresh coconut meat without drying first. The "coconut milk" is expressed first by pressing it out of the wet coconut meat. The wet method is practiced by several techniques of chilling and thawing, fermentation, enzymatic, pH or any combination of these methods; as the aim is to destabilize the coconut milk emulsion (Nur Arbainah, 2012, Mansor *et. al*, 2012). The integrated wet process starts with grated coconut is expelled to obtain the coconut milk. The coconut milk is then cooled at desired temperature ($T_{churning}$) before churned to separate the water and butter oil. The butter is then melted in water bath at desired temperature ($T_{thawing}$) before undergoing the subsequent centrifugation and filtration processes to get the clear final oil (VCO).

The dry method is utilizing the expeller to press the dried coconut to obtain the oil. In this method, the fresh kernel is heated under specific conditions until the desired moisture content is obtained to avoid microbial contamination. The dried kernel is then pressed mechanically to obtain the oil (Mansor *et. al.*, 2012).

As overall, it is a perquisite to produce the virgin coconut oil with a proper method than can retain the essential minor compound in it. The integrated wet process offers a condition for the highest total phenolic compound and antioxidant compared to fermentation (wet method) and dry method. The integrated wet process also exhibits its capability in retaining the fresh aroma of coconut and scores the highest acceptance in term of sensory evaluation in comparison to both fermentation and dry processes (Nur Arbainah, 2012).

Instead of selecting the oil processing method, it is an advantage to opt the specific coconut cultivar that containing the high oil. Numbers of research showed that every coconut cultivar has its own particular characteristic. This ascribe the factors of location and varietal differences, age of nuts, time of the year the nuts harvested and age of copra before expelling give significant effect (Carandang , 2008). The cultivar that contains the highest oil can be used in VCO processing to gain better oil recovery.

1.2 Problem Statement

The integrated wet process has been introduced by Institute of Bioproduct Development (IBD) to produce the virgin coconut oil. Despite of the quantitative aspect to increase the yield of virgin coconut oil, the quality characteristic must be improved or at least maintained in order to fulfill the standard of virgin coconut oil. The characteristic chemical and physical properties such as phenolic content, free fatty acid and colour that expected to be affected by processing temperatures are becoming the focus in this study.

The very lack of information on coconut varieties producing high yield of coconut oil especially in Malaysia has come into attention. By identifying the best variety that contributing the highest recovery of the oil had to be a concern in this study. Therefore, six varieties were selected to find the highest oil content so that to enhance the oil recovery in the integrated wet process.

1.3 Hypothesis

The hypothesis of the study is the processing temperatures of respective churning and thawing in integrated wet process affect the oil yield, ferulic acid concentration, free fatty acid and colour attributes.

1.4 Objective of the Study

 The objective of this research was to optimize the churning and thawing temperatures in integrated wet process and their effects on the yield and quality of the virgin coconut oil.

1.5 Research Scope

The scopes of this research were;

- To select the coconut varieties could contribute to the highest oil content. The varieties included West African Tall (WAT), Rennel Tall, Matag, MAWA, Maren and Tagnanan Tall.
- To determine churning and thawing temperatures effects on coconut butter and VCO contents.
- 3) To optimize the churning and thawing temperatures and their effects on oil yield, ferulic acid concentration, free fatty acid and colour.

1.6 Contribution of Study

The virgin coconut oil is a high value coconut product that consumed for its nutraceutical benefits and as a functional food. The other usage of VCO is in cosmeceutical that used as aromatherapy carrier oil, body moisturizer and hair conditioner. Due to its wide application, the world demand for VCO is significantly increasing, making it a potential business for coconut players (Bawalan & Chapman, 2006).

The quality of VCO differs significantly from how it is processed. The RBD process using copra to produce the coconut oil; undergoes the refine, bleach and deodorize processes to make it suitable for human consumption resulting the bland taste due to the refining process. Meanwhile, the wet method utilizes the fresh coconut milk in the beginning of the process preserves the fresh aroma and taste of coconut. Unfortunately, the disadvantage of the wet method is low oil recovery as compared to the RBD method (Raghvendra & Raghavarao, 2010).

For wet method, the integrated wet process offers the higher quality product and yield compare to fermentation process. The oil recovery in integrated wet process is 85 percent higher than using the fermentation method. When use the fermentation process, the fermented odour is remaining and affecting the oil flavour (Nurarbainah, 2012). From the results, it indicated the integrated wet process is the better option to be evaluated further for better yield and quality of the final oil, compared to the fermentation method.

The integrated wet process is an efficient and quick technique to produce the VCO with the highest heat stability. In integrated wet process, the churning proses is started with solidifying the the coconut milk at low temperature (9 °C) (Nurarbainah, 2012). The purpose of this process is to crystallize the fat in the coconut milk emulsion and lead to the separation of butter and water during stirring process

(Coupland, 2002). Therefore, it is essential to find the optimum temperature for churning process that may enhance the maximum separation of butter-water to obtain the better oil recovery at the end of the integrated wet process.

In integrated wet process, the thawing process is applied to expedite the melting of the coconut butter, followed by the centrifugation to separate the non-oil fraction in the VCO (Hamid *et.al*, 2011). Currently, the integrated wet process practices the 37° C for its thawing temperature. The thawing process undergone may destroy some of the phenolic compound in the oil, but when use the lower temperature (near to the room temperature), it would take longer time for the butter to melt (Marina *et. al.*, 2009^a). For this reason, it is a necessary to study the optimum thawing temperature to fasten the thawing process and simultaneously retain the maximum phenolic compound in the oil.

As a conclusion, it is important to establish the optimum condition for churning and thawing temperatures to gain high oil recovery at the best quality.

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