

**FORMULATION AND EVALUATION OF HAIR
SHAMPOO CONTAINING TEA TREE
(*Melaleuca alternifolia*) AND VIRGIN
COCONUT (*Cocos nucifera*) OIL**

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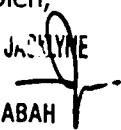
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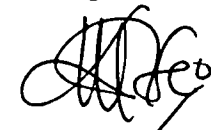
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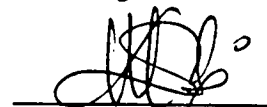
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ABSTRACT

The components of essential oil for *Melaleuca alternifolia* or commonly known as tea tree oil (TTO) are well known for their various beneficial properties. Whereas, the medium chain fatty acids in oil of Virgin Coconut (*Cocos nucifera*) oil (VCO) have the ability to protect hair follicles from heat, restoring hair's moisture and other damage. The objective of the current study is to incorporate TTO and VCO into shampoo formulations and systematically evaluate their properties and effects on hair. This thesis describes the physical properties of seven hair shampoo formulations containing differing amount of TTO and VCO. The essential oils applied in these formulations were extracted from fresh tea tree leaves using steam distillation method and the virgin oil was produced from fermentation of fresh mature kernel coconut. Gas Chromatography-Mass Spectrometry (GC-MS) analysis was conducted to determine the essential oil components of TTO and fatty acid composition of VCO. The shampoo formulations were subjected to evaluation of several parameters. The GC-MS results show that the TTO was composed of terpene hydrocarbons with Terpinene-4-ol as the major component. As for the VCO, its major component is lauric acid. All formulations were acid balanced with pH range between 6.23 – 6.43. Previous study shown that acid balanced shampoos will cause less frizzing for generating less negative static electricity on the hair fiber surface. Total solid contents were between 29.92 – 35.61 % with shampoo formulation of 6% TTO 0% VCO showed the best. All formulations showed the same foam volume for 4 minutes observation and no dirt were observed which indicates their stability in foaming and ability of cleansing are excellent. Rheological evaluations were conducted with only shampoo containing 6% TTO 0% VCO showed pseudo plastic behavior which is a desirable attribute in shampoos formulation. It showed high viscosity at low rotational speed and decrease in viscosity at high rotational speed which indicates a favorable property to spread the shampoos on hair. Antimicrobial effectiveness test was carried out to determine the efficacy of shampoo formulation on elimination of *Staphylococcus aureus* bacteria. From the results, shampoo with 6% TTO 0% VCO exhibits the most satisfying result due to the highest percentage reduction of bacterial load which is 95.8%. The Scanning Electron Microscope (SEM) showed that hair samples washed with shampoo containing 6% VCO 0% TTO indicates overlapping scales with no crack and holes indicating well-conditioned and healthy hair. In summary, TTO can be formulated specifically as antimicrobial shampoo focusing in combating severe itchiness and bacterial infection on human hair and scalp. Whereas, VCO is greater in the preparation of hair conditioning and damage treatment shampoo due to its ability in penetrating hair fiber and high affinity for hair proteins.



ABSTRAK

FORMULASI DAN PENILAIAN SYAMPU RAMBUT YANG MENGANDUNGI MINYAK TEA TREE (*Melaleuca alternifolia*) DAN MINYAK KELAPA DARA (*Cocos nucifera*)

*Komponen minyak pati untuk *Melaleuca alternifolia* atau yang lebih dikenali sebagai minyak tea tree (TTO) terkenal dengan pelbagai ciri-ciri bermanfaat. Manakala, asid lemak rantai sederhana dalam minyak kelapa (*Cocos nucifera*) dara (VCO) mempunyai keupayaan untuk melindungi folikel rambut daripada haba, memulihkan kelembapan rambut dan juga kerosakan yang lain. Tesis ini menerangkan sifat fizikal dari tujuh formula syampu rambut yang mengandungi jumlah TTO dan VCO yang berbeza. Minyak pati yang digunakan dalam formulasi ini diekstrak daripada daun segar pokok tea tree menggunakan kaedah penyulingan wap dan minyak kelapa dara dihasilkan daripada penapaian buah kelapa matang. Analisis Gas Chromatography-Mass Spectrometry (GC-MS) dijalankan untuk menentukan komponen kimia dalam TTO dan komposisi asid lemak VCO. Setiap formulasi syampu yang dihasilkan dinilai mengikut beberapa parameter. Keputusan analisis menunjukkan bahawa TTO terdiri daripada hidrokarbon terpena dengan Terpinene-4-ol sebagai komponen utama. Bagi VCO, komponen utamanya adalah asid laurik. Semua formulasi adalah berkeadaan asid dengan julat pH antara 6.23 - 6.43. Kajian terdahulu menunjukkan bahawa syampu bersifat asidik mengurangkan masalah rambut kusut dan mengurangkan penghasilan cas negatif elektrostatik pada permukaan serat rambut. Jumlah kandungan pepejal adalah antara 29.92 - 35.61%, dan keputusan syampu dengan 6% TTO 0% VCO menunjukkan nilai terbaik di antara formula lain. Semua formulasi menunjukkan isipadu penghasilan buih yang sama selama 4 minit pemerhatian dan tiada kotoran yang dihasilkan yang menunjukkan kestabilan dalam berbuih dan keupayaan pembersihan sangat baik. Penilaian rheologi dilakukan dan hanya syampu yang mengandungi 6% TTO 0% VCO menunjukkan sifat pseudoplastik yang merupakan sifat yang wajar dalam penghasilan syampu. Ia menunjukkan kelikatan yang tinggi pada kelajuan putaran rendah dan penurunan kelikatan pada kelajuan putaran tinggi yang membuktikan kualiti penyebaran syampu yang rapi pada rambut. Ujian keberkesanan antimikrob dijalankan untuk menentukan keberkesanan setiap formula syampu dalam menghapuskan bakteria *Staphylococcus aureus*. Hasil keputusan adalah syampu mengandungi 6% TTO 0% VCO memperlihatkan hasil yang paling memuaskan kerana pengurangan peratusan tertinggi jumlah bakteria iaitu 95.8%. Scanning Electron Microscopy (SEM) menunjukkan sampel rambut dibasuh dengan syampu yang mengandungi 6% VCO 0% TTO menyatakan lapisan rambut bertindih antara satu sama lain dan tiada kerekahan dan lubang menunjukkan sifat rambut yang sihat dan kelembapan yang baik. Secara ringkasnya, TTO boleh dirumuskan secara khusus sebagai syampu antimikrobial yang mana lebih fokus kepada mengawal kegatalan yang teruk dan jangkitan bakteria pada rambut manusia dan kulit kepala. Manakala, fokus utama VCO lebih kepada syampu kelembapan dan mengawal kerosakan rambut kerana keupayaannya menembusi serat rambut dan tarikan tinggi untuk protein rambut.*

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LIST OF ABBREVIATIONS

APCC	Asian and Pacific Coconut Community
CMC	Cell membrane complex
EDTA	Ethylenediaminetetraacetic acid
ISO	International Standard Organization
GC-MS	Gas Chromatography-Mass Spectroscopy
RBD	Refining, Bleaching and Deodorizing
SAIP	Sabah Agro-Industrial Precinct
SEDIA	Sabah Economic Development and Investment Authority
SEM	Scanning Electron Microscopy
SLES	Sodium Lauryl Ether Sulphate
TTO	Tea Tree Oil
VCO	Virgin Coconut Oil



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CHAPTER 1

INTRODUCTION

1.1 Background of Study

Shampoo is one of must have item in our bathroom. It is known as a cosmetic product for cleansing hair and scalp, resulting soft, manageable, and lustrous hair. Shampoos texture can be clear or opaque and also available as creams, lotions, gels, pastes, liquids or even dry-powder aerosols (Johnson, 1997). Varieties of shampoo are marketed nowadays based on their specific functions such as anti-dandruff shampoos, baby shampoos, conditioning shampoos, and dry shampoos. From the ancient time, human being topically uses the natural approach especially in maintaining and treatment of hair. Recently, there has been a huge increase in the use of natural products in cosmetics preparation. Nowadays, herbal shampoos are commercially can be found in the market which contains natural ingredients such as plant extracts and essential oils. There are a numerous number of plants which are claimed to have great effects on hair and are commonly used in haircare formulations (Anusha *et al.*, 2013).

The objective of the current study is to incorporate tea tree oil (TTO) and virgin coconut oil (VCO) into shampoo formulations and systematically evaluate their properties and effects on hair. This thesis describes the physical properties of seven hair shampoo formulations containing differing amount of TTO and VCO. The essential oils applied in these formulations were extracted from fresh tea tree leaves using steam distillation method and the virgin oil was produced from fermentation of fresh mature kernel coconut. Gas Chromatography-Mass Spectrometry (GC-MS) analysis was conducted to



determine the essential oil components of TTO and fatty acid composition of VCO. The shampoo formulations were subjected to evaluation of parameters namely appearance, pH, viscosity, total solid contents, foam stability, and dirt dispersion. Antimicrobial effectiveness test was also carried out to determine the efficacy of shampoo formulation on elimination of *Staphylococcus aureus* bacteria.

Since TTO originated from Australia, it has been used medicinally for more than 80 years, with uses relating primarily to its anti-inflammatory and antimicrobial properties. According to Hammer *et al.* (2004) the essential oil of tea tree is obtained by using steam distillation process, from the Australian native plant tea tree also known as *Melaleuca alternifolia*. The chemical composition contains approximately 100 components, most of it consist of monoterpenes, sesquiterpenes and other related alcohols. Major compound of TTO is Terpinen-4-ol, which has long been considered as the main anti-microbial properties to TTO (Carson *et al.*, 2005). Based on previous study Carson and Riley (1995), they did an evaluation on the antimicrobial effectiveness for eight components of TTO using two methods, which are disc diffusion and broth microdilution. It resulted that terpinen-4-ol was active against all the test organisms (*Candida albicans*, *Staphylococcus aureus* and *Escherichia coli*) while there was no antimicrobial activity reported for *p*-cymene performed. Meanwhile, other terpenes and α -terpineol were actively against all test organisms except for *Pseudomonas aeruginosa*.

The beneficial effect of coconut oil has been studied previously by water retention measurements and protein loss effect. In addition to providing a lubricating film, coconut oil minimized the water penetration into the hair fiber as well. This is due to the hydrophobicity properties of coconut oil, which reduces the water penetration into the fiber (Relle and Mohile, 1999).

As TTO and VCO were well proven to have beneficial properties, incorporating these two oils in a formulation of shampoo will benefit in overcoming hair and scalp problem. Therefore, this research was designed to formulate shampoo containing TTO and VCO and systematically evaluate their properties and effects on hair.



1.2 Problem of Statement

In the past 20 years, cosmetics industry was reported to be increasing by average 4.5% market's growth rates per annum which makes it as one of the most stable industries due to demand that keep increasing all over the world (Lopaciuk & Loboda, 2013). According to Hassali (2015) the extensive of global beauty market is greatly influenced by the increasing demand especially from Europe and Asia Pacific region.

One of the major challenges for the cosmetic manufacturer is to provide and produce a good product which both low cost and environmentally friendly to the consumer. Despite the positive news related to the grown of the cosmetics industry, the fact that the safety of cosmetic used is always at a topmost priority which cannot be taken lightly. Safety of cosmetics production and usage has become the major concern (Draelos, 2012). One of the common reasons a consumer was referred to the hospital was due to an adverse reaction from the application of beauty products. Some of the ingredients or additive chemicals are hazardous and prohibited that can impact in human health in a negative way, it may cause cancer, reproductive toxicity, endocrine disruption and mutation as well (Amasa *et al.*, 2012). To obtain fast result or effectiveness of products, manufacturer tends to incorporate dangerous chemicals to their product for the sake of economic benefit. Heavy metals, for example, are one of the prohibited chemicals to be found in a beauty product. Even at low concentration, these elements will cause serious damage to the internal body organ of animals and humans which lead to concerning health issue. Metal poisoning was reported to cause various respiratory diseases, various cancers, intellectual retardation and organ failure (Ayenimo, 2010).

The major concern and consciousness about the safety of cosmetics trigger the natural cosmetics market to grow. The concept incorporating natural-based ingredients in cosmetic preparation is widely grown in the market which attracts consumers attention. Investigation of natural products has gained popularity among the researchers recently because of its abundantly available, renewable as well as degradable materials (Awang *et al.*, 2012). Active compounds in natural ingredients present tremendous benefits and valuable bioactivities such as anti-aging, antimicrobial, antioxidant,



photoprotection, and anti-inflammatory actions that useful for production of skincare, hair care and dental care (Mohd *et al.*, 2018).

Tea tree oil and VCO among oils which possess a lot of beneficial properties, it has been used largely in various industries of its antimicrobial properties (Southwell and Lowe, 1999), whereas virgin coconut oil has been used since ancient time as multipurpose oils and nowadays it's become a trend in cosmetic application (Rele and Mohile, 2003). This research aimed to prepare and evaluate shampoo formulation containing tea tree oil and virgin coconut oil. Tea tree oil and Virgin coconut oil were analyzed for its major component and results obtained were in accordance with other previous studies. Tea tree oil and virgin coconut oil contains components which prominent for its various benefits. By incorporating the two oils in shampoo formulation, besides its contribution to the treatment of hair and scalp, the usage of synthetic additives will be minimized as well. However further research and development are still required to improve and enhance its properties and quality especially on the safety assessment of the formulation.

1.3 Significance of Study

Significance of this study is the usage of natural oils as ingredients in formulating shampoo which benefit in hair treatment. TTO and VCO were used with various percentages in each formulation of shampoo. TTO is an essential oil that widely known because of its antibacterial, antimicrobial, antiviral and anti-fungal properties and some anti-inflammatory activity. Whereas, VCO which refers to an oil that is obtained from fresh endocarp of the coconut by natural methods, without the use of heat and without undergoing chemical refining. The fats in VCO can penetrate each strand of hair, restoring lost moisture. In particular, the medium chain fatty acids are good at restoring lost moisture to hair and VCO can even protect hair follicles from heat or other damage. Therefore, with incorporating both TTO and VCO will be resulted in producing shampoo which can benefit more to human hair and scalp as well as minimizing usage of synthetic additives to shampoo formulation.



1.4 Research Objectives

The specific objectives of this research are:

1. To prepare shampoo formulations with differing amount of Tea Tree (*Melaleuca alternifolia*) oil and Virgin Coconut (*Cocos nucifera*) oil.
2. To analyses physicochemical and biologicals characteristics of the shampoo formulations.
3. To evaluate the effect of conditioning to human hair after treatment with the shampoo formulations.

1.5 Scope of Study

This research consists of three main parts. First part was the preparation of both oils TTO and VCO from fresh tea tree leaves and mature kernel coconut respectively. Both oils were prepared using steam distillation and fermentation method which afterwards were analyzed by GC-MS in order to provide and maintaining the purity and quality of TTO and VCO. Second part focuses in incorporating TTO and VCO in hair shampoo preparation. Seven hair shampoo formulation was successfully developed with differing amount of TTO and VCO with not more than 6% w/w content of the oils in each shampoo formulation. 6% TTO and 6% VCO was set as the maximum oils content for this research due to safety purposes and stability of the emulsion prepared. The third and last part of the research was on the assessment of shampoo formulation for their properties and effects on hair. The assessment was including the organoleptic characteristics, pH determination, total solid content, the ability and stability of foaming and rheological evaluation as well. The Scanning Electron Microscopy (SEM) was used to study the morphology of human hair after the treatment using seven hair shampoo formulation. Finally, the antimicrobial effectiveness test was also carried out in order to investigate the effectiveness of shampoo against bacteria, which is in this research *Staphylococcus aureus* to be specific.



CHAPTER 2

LITERATURE REVIEW

2.1 Introduction

This chapter includes an extensive review of human's hair structure, hair damaged, scalp disorders and shampoo review. The review on both TTO and VCO are also presented in this chapter.

2.2 The Structure of Human's Hair

Human hair is a bio-synthesized material which consist of a complex internal structure with chemical and physical behavior consisting mostly proteins in particular keratin (Yang and Rheinstädter, 2014). Hair is divided into two distinct parts: the hair follicle which resides in the dermal layer of skin, and the visible hair fiber on scalp. The follicle of hair begins at the surface of the epidermis which extends to deep dermis (Harkey, 1993). The terminal region of the hair follicle existed within the skin or well known as hair bulb, it is the structure formed by actively growing cells that generates long, fine and cylindrically shaped hair fibers (Cruz *et al.*, 2016). Figure 2.1 shows the structure of hair follicle. The hair fibre component is divided into three main parts: Cuticle, cortex and medulla (Dias, 2015). Figure 2.2 shows the schematic cross section of hair fibre.



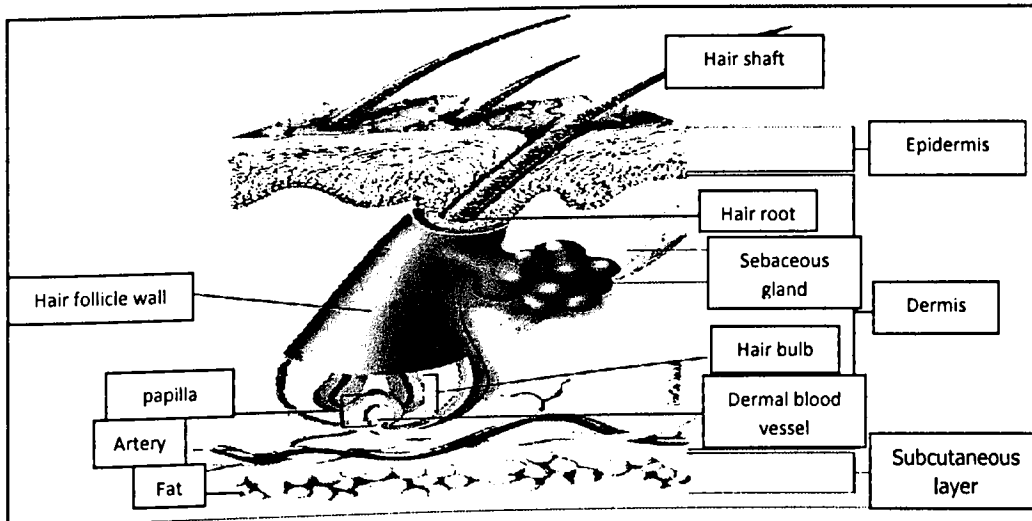


Figure 2.1: Structure of Hair Follicle

Source : Jain & Das (2016)

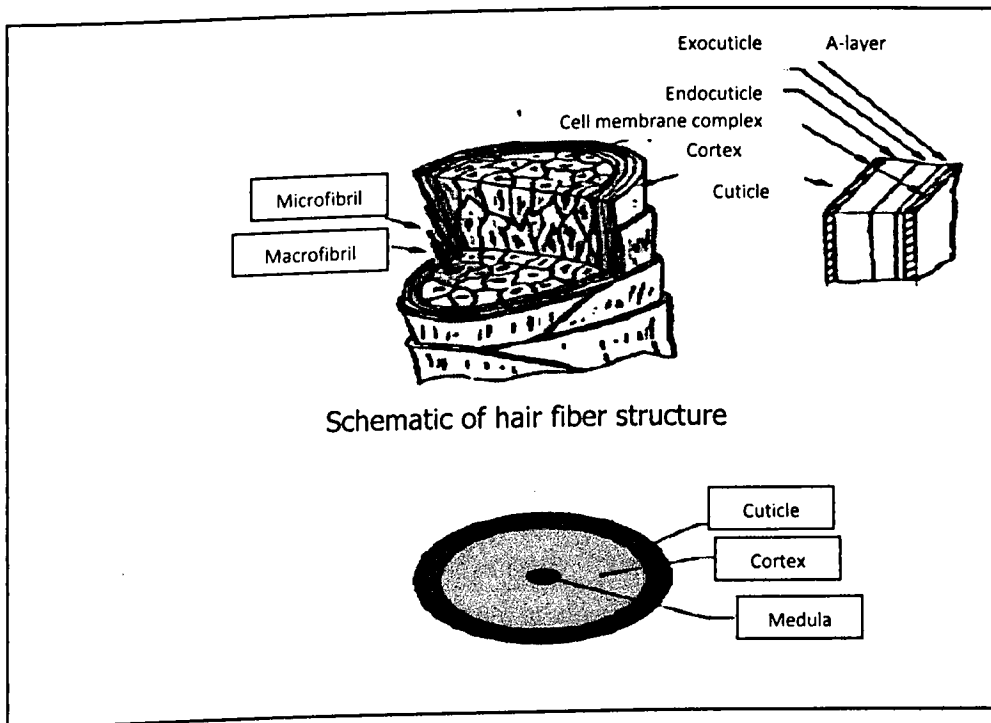


Figure 2.2: Schematic cross section of hair fibre

Source : Wei *et al.* (2005)

2.2.1 Cuticle

The hair thread are covers by the cuticle, it acts as a barrier by protecting the underlying cortex from any external unwanted environmental damage (Cruz *et al.*, 2016). As cuticle part will be more or less affected by treatment whether naturally or synthetically, it is the most important part of the human hair. Cuticle is the most outer layer of human hair, application of cosmetic products is deposited on this layer. Chemical treatments, such as straightening, dyeing and curling products spread themselves via the cuticle to expatiate their effects by the hair fiber.

The cuticle contains approximately 8 to 11 layers of overlapping scales in the lateral direction of the thread, based on the condition, length, and type of the hair. Each of the individual layer is formed by only one cell and each of the cell is a rectangular in shape and overlapping in such way, that only one-sixth of them are exposed (Robbins, 2012). The epicuticle (~ 3 nm) is a thin external membrane, which is a protein coat covered by a strong lipid structure that includes 18-methyl eicosanoic acid (18-MEA) and free lipids (Velasco *et al.*, 2009). It provides lubricity to the hair and consequently act as the first line of defense against any environmental attack. The A-Layer lies immediately beneath the epicuticle cell membrane with approximately 30% cystine content (Cruz *et al.*, 2016). The following layer which is the exocuticle also known as B-Layer, with approximately 15% cysteine content and corresponds to 55% of the cuticle layer (Velasco *et al.*, 2009). The last layer is endocuticle which is mainly composed of remaining cell organelles, with low content of cysteine ($\sim 3\%$). This layer swells more in water than the layer with a high content of cysteine, and it is mechanically softer.

The intercellular matter that holds the cuticle cells together well-known as the cell membrane complex (CMC). CMC mostly consists of cell membranes, an adhesive material in CMC binds the cell membranes which comes between two cuticle cells, two cortical cells and cuticle-cortex cells. It comprises of beta layer which contemplated to be the intercellular cement and it is packed by other layers from each cell (Dias, 2015).

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