Assessment of the skin photoprotective capacities from coastal plant of *Melaleuca cajuputi* essential oil as a potential UV filters

M Z H Rozaini1,3*, M U Osman2, M H Razali2, M F A Aziz3 and M S Azhar 1,3

1Institute of Marine and Biotechnology, Universiti Malaysia Terengganu, Kuala Nerus 21030, Terengganu, Malaysia
2Faculty of Science and Marine Environment, Kuala Nerus 21030, Terengganu, Malaysia
3Faculty of Fishery and Food Sciences, Kuala Nerus 21030, Terengganu, Malaysia

*Corresponding author: zulhelmi@umt.edu.my

Abstract. UV irradiation can cause cutaneous damage that may be specific according to the wavelength of UV rays. For example, damage from UVB irradiation manifests itself in the form of sunburn cells and enhancement of the expression of p53, while damage from UVA exposure results in an increase in the expression of vimentin. These reactions to UV irradiation were used in this work to evaluate the photoprotective capacities of two sunblock preparations that were applied to the surface of the skin. One sunblock preparation is a UVB absorber containing the coastal plant of *Melaleuca cajuputi* essential oil and titanium oxide (TiO$_2$) exclusively. The other sunblock preparation is a conventional sunblock containing oxybenzone as comparison. Result obtained for UV-Vis test was observed that the new formulation derived showed good absorption and exhibit high potential to be further develop as sunscreen in cosmeceutical applications

Keywords: sunscreen; coastal plant; miroemulsion; *Melaleuca cajuputi* essential oil

1. Introduction

*Melaleuca cajuputi* is a type of Malaysian plant known as gelam or kayu putih in Malaysian vernacular. *Melaleuca leucadendron* is a synonym name for *Melaleuca cajuputi*, which belongs to the Myrtaceae family. When the leaves of *Melaleuca cajuputi* are scraped, they generate a "tea-tree scent," which is the source of cajuputi oil, a type of essential oil. It contains antiseptic, anti-inflammatory, and antibacterial properties that could be used in skincare products [1]. According to the research, it is also effective as an antioxidant [2], has an active cytotoxic impact [3], and is antibacterial [4, 5]. However, the sunscreen effect of the microemulsion made from *Melaleuca cajuputi* essential oil has yet to be established when mixed with a non-ionic surfactant, which is a major worry in this study. Because Malaysia's climate is predominantly tropical, it is hot and humid all year, and sunshine is unavoidable. As a result, *Melaleuca cajuputi*, which has the potential to be used as a sunscreen, is being investigated in this study using a combination of a co-surfactant (olive oil) and a nonionic surfactant (SPAN 80).

2. Materials dan methodology

TWEEN 20, TWEEN 80, SPAN 20, SPAN 80, and olive oil (Sigma-Aldrich); Vaseline Petroleum Jelly; *Melaleuca cajuputi* essential oil (Vaseline); TWEEN 20, TWEEN 80, SPAN 20, SPAN 80, and olive oil (Sigma-Aldrich); *Melaleuca cajuputi* essential oil (Vaseline); *Melaleuca cajuputi* essential oil (Vaseline) (original). All of the substances were used exactly as they were given to us [6].

2.1. Extraction of the *Melaleuca cajuputi* essential oil

MARDI (Malaysia Agriculture Research and Development Institute) at Telong, Bachok, Kelantan Darul Naim, were undergone the extraction process on producing the essential oils. The steam distillation method was adopted at MARDI in this study since it was straightforward to manage and the purification procedures [7]. We were using the fresh *Melaleuca cajuputi* leaves during extraction. Plant material is
placed in a stiller during steam distillation. Water was pumped into the tank and heated in the vessel, while steam was pumped into the stiller through a coil. The essential oil was vaporised and released from the leaves as vapour passes through the plant materials. The saturated vapour of essential oils rises via refrigerated tubing, condenses, and is drained. After the steam procedure, calcium chloride anhydride was added to the essential oil to absorb the air in the solution.

2.2. Preparation of emulsification process

In the emulsification process, the emulsion were mixing and shearing until the particles were suitable size to be stable. The surfactant used was TWEEN 20, TWEEN 80, SPAN 20 and SPAN 80 and co-surfactant is olive oil. Water was used as water phase because it is universal. The formulation of the microemulsion is according to the intersection point of phase diagram constructed. The thickener used was Petroleum Jelly. Melaleuca cajuputi essential oil was used as additives substances in the microemulsion to give pleasant odour and acts as potential sunscreen agent [8].

The emulsion was mixed and sheared until the particles were homogenous and stable during the emulsification process. TWEEN 20, TWEEN 80, SPAN 20 and SPAN 80 were utilized as surfactants, with olive oil as a co-surfactant. Because water is universal, it was chosen as the water phase. The microemulsion is made according to the intersection point of the phase diagram that has been produced. Petroleum Jelly was used as a thickening. Melaleuca cajuputi essential oil was added to the microemulsion as an addition to give it a pleasant odor and to act as a potential sunscreen agent [8].

The microemulsion was made, and the formulation was chosen based on the ternary phase diagram's 3 phases area, which is the most stable. The water bath was used to heat both the oil and water phases to temperatures exceeding 80°C. To avoid overheating, a water bath was employed, and thermometers were placed within the two-phase beaker to get reliable temperature readings [13].

The oil phase was steadily poured into the water phase to build the oil-in-water system. The microemulsion was then heated to above 80°C to ensure that it was homogenized as a colloid system. The microemulsion was then withdrawn from the heat sources and homogenized. Homogenization is the process of breaking down the particles in the microemulsion into smaller sizes and distributing them uniformly throughout the system. As a result, the longer it takes to homogenize the microemulsion, the smaller and more stable it becomes [9].

When homogenization was completed, three drops of Melaleuca cajuputi essential oil were added. At the end of the process, a nice milky microemulsion was generated. Finally, all of the emulsifying samples were stored in a dark closet to prevent contamination and reactions to light, heat, or shaking. The samples were examined to see how long they could maintain their stability.

2.3. The measurement of SPF value

The SPF value, which is defined as a comparative ratio between the lowest UV energy needed to cause a minimal erythema dose (MED) on protected skin against unprotected skin (equation 1), is widely used to determine the effectiveness of a sunscreen:

$$\text{SPF} = \frac{\text{minimal erytherma dose in sunscreen-protected skin}}{\text{minimal erytherma dose in nonsunscreen-protected skin}}$$

(1)

The MED is the minimum exposure duration or UV radiation dosage needed to produce a minimal, perceptible erythema [10-11]. The increase effectiveness of a product in preventing sunburn is based on the increase in its SPF value. Therefore, methods have to be standardized to determine products’ SPF. The in vivo method is tested on volunteers to determine the effective protection sustained by topical sunscreens against exposure. This kind of perseverance are commonly used and even though beneficial and accurate, it is expensive, complex and time consuming [12]. As a result, researchers are more interested in finding in vitro methods to evaluate the photoprotection abilities of different sunscreen compounds. There are two common in vitro methods: (a) UV radiation is transmitted through sunscreen
product films in quartz plates or biomembranes or (b) the measurement of absorption characteristics using spectrophotometric analysis [13].

The minimum exposure length or UV radiation dose required to cause a minimal, detectable erythema is known as the MED [10-11]. An increase in a product's SPF rating indicates that it is more efficient at avoiding sunburn. As a result, procedures for determining the SPF of goods must be standardized. The in vivo approach is used to investigate how efficient topical sunscreens are at protecting people from sun exposure. This type of persistence is widely used, and while it is advantageous and accurate, it is also costly, difficult, and time consuming [12]. As a result, scientists are focusing their efforts on developing in vitro techniques to test the photoprotective qualities of various sunscreen chemicals. In vitro, two typical approaches are used: (a) UV radiation is transmitted via sunscreen product films in quartz plates or biomembranes, or (b) spectrophotometric analysis is used to determine absorption properties [13]. In addition, using UV spectrophotometry, the in vitro approach may be replaced by the following equation below. Additionally, the in vitro method can be substituted by the following equation with the aid of UV spectrophotometry [14]:

$$SPF_{x_{\text{spectrophotometric}}} = CF \times \sum_{290}^{320} \left[ \text{EE}(\tau) \times I(\tau) \times \text{Abs}(\tau) \right] \tag{2}$$

Where: EE (l) – erythemal effect spectrum; I (l) – solar intensity spectrum; Abs (l)- absorbance of sunscreen product; CF – correction factor (= 10). The values of EE x I are constants.

### 3. Results and discussions

#### 3.1. Emulsification process

The emulsification procedure was carried out using the conventional approach and the ternary phase diagrams that had been generated. Four 20 gram samples were made and stored for a month in a cold, dark room. This is the most important division, since it directs the colloidal medium's consistency. The microemulsion created in this study is an oil-in-water (o/w) type, as the oil phase, which consisted of four distinct nonionic surfactants, olive oil, and petroleum jelly (thickener), was poured into the water phase after both phases had been heated to 80°C. This sort of microemulsion is referred to as a simple microemulsion since it only has one degree of emulsification.

In the oil-in-water system, the oil phase was progressively poured into the water phase, and the microemulsion was heated above 80°C to ensure homogeneity as a colloid system. The microemulsion

---

Table 1. Formulation of microemulsion samples.

| Materials          | Formulation 1 | Formulation 2 | Formulation 3 | Formulation 4 |
|--------------------|---------------|---------------|---------------|---------------|
| Surfactants        | TWEEN 20      | TWEEN 80      | SPAN 20       | SPAN 80       |
| Olive oil          | 24%           | 24%           | 24%           | 24%           |
| Water              | 50%           | 50%           | 50%           | 50%           |
| Petroleum Jelly$^a$| 2g            | 2 g           | 2 g           | 2 g           |
| Essential oil$^b$  | 3 drops       | 6 drops       | 12 drops      | 18 drops      |

$^a\text{Means with the same superscripts within a row are not significantly different (P<0.05)}$

Table 2. SPF value of the sunscreen formulation.

| Formulation   | Active Ingredients | Amount (g) | Sunscreen Protection Factor (SPF) |
|---------------|--------------------|------------|----------------------------------|
| Formulation 1 | Melaleuca cajuputi | 3          | 9,630 ± 1.5                      |
| Formulation 2 | Melaleuca cajuputi | 6          | 23,533 ± 1.3                     |
| Formulation 3 | Melaleuca cajuputi | 12         | 37,940 ± 1.2                     |
| Formulation 4 | Melaleuca cajuputi | 18         | 45,469 ± 1.6                     |

All values are presented as Mean ± SD (n =3)
was then withdrawn from the heat sources and homogenized to break down the particles inside the microemulsion into smaller sizes and ensure that they were uniformly distributed throughout the system. As a result, the longer the time required for homogenization, the smaller and more stable the microemulsion becomes [15-16].

The findings revealed that the freshly prepared microemulsion were milky white in colour for Formulation 1, 2 and 4 and yellowish white in colour for Formulation 3. Table 1 showed the formulation used to make the microemulsion samples.

3.2. Sun protection factor (SPF) measurements
The primary goal of the sunscreen test is to assess the absorption of ultraviolet (UV) radiation. This test was conducted with all formulations exposed to sunshine. The absorption peak value was then determined before and after exposure to sunlight using UV-Visible spectroscopy. Water was utilized as a solvent because it absorbs less sunlight and will record the correct value absorption. Table IV showed the findings sorted by increment value.

All formulations exhibited positive results in UV radiation absorption based on the data obtained. UV-C wavelengths ranging from 200 to 280 nm were absorbed by formulations 1, 2, and 3. As a result, the microemulsion generated can only absorb dangerous UV-C but not UV-A or UV-B, yielding 9.630, 23.533, and 37.945 SPF values, respectively. UV-C, on the other hand, is unable to reach the earth's surface because it is absorbed by the ozone layer. As a result, UV-C does not yet affect human skin [17,18]. However, formulation 4 was shown to absorb the maximum UV-B light at 286.2 nm, with a wavelength range of 280-315 nm and an increment value ranging from 0.0516 to 45.469 SPF. UV-A and UV-B radiations are both hazardous to human health since they can damage DNA and cause major illnesses such as skin cancer [19]. Chemical constituents that include a carbonyl group, such as phenanthrene, have this UV radiation inhibitor characteristic [20]. As a result, it is thought that Tween surfactant can be employed as a UV-absorber.

4. Conclusion
The most stable ternary phase diagram which consists of surfactant, co-surfactant which is olive oil and water by comparing four phase diagram which using from different non-ionic surfactant, TWEEN 20, TWEEN 80, SPAN 20, and SPAN 80 were constructed. The region of the liquid crystal and micelle formation also had been discovered and plotted under the microscopic test. All samples of microemulsion had been fabricated from the phase diagram due to its potent liquid crystal whereas had been subjected to stability test. The formulation 4 of microemulsion had tendency to absorb the ultraviolet radiation especially in UV-B that prevent from lead to skin cancer compared to other formulations only absorb UV-C radiation. This is because of chemical properties from *Melaleuca cajuputi* essential oil that gives the main role act as sunscreen due to the carbonyl group such as phenanthrene. So, within nano size range of particle would be advantages for the system with extremely small droplet size which can increase penetration effect, highly interfacial area and capable to produce UV absorber to give sun protection.

**Acknowledgments**
This project consumed huge amount of work, research and dedication. Still, implementation would not have been possible if we did not have a support of many individuals and organizations. Therefore, we would like to extend our gratitude towards Ministry of Higher Education (MOHE) via FRGS 59427 vote number.

**References**
[1] Norudin M, Desnika M A E and Rafi Y M 2010 Cosmetic usage in Malaysia: understanding of the major determinants affecting the users faculty of business management *Int. J. Bussiness Soc. Sci.* 1 273–281.
[2] Chaudhri S K and Jain N K 2009 History of cosmetics Asian Journal of Pharmaceutics 3 164-167.
[3] Epstein H 2009 Cosmeceutical vehicles Clinics in Dermatology 27 453-460.
[4] Mehta S K, Kaur G and Bhasin K K 2017 Analysis of tween based microemulsion in the presence of TB drug rifampicin Colloids Surfaces B. Biointerfaces 60 95–104.
[5] Giri T K, Goswami N G and Jha V K 2013 Prospective and challenges of micro-emulsions as a novel carrier for drug delivery J. Pharma. Sci. Tech. 2 56-61.
[6] Kassim M, Yusoff K M, Ong G, Sekaran S, Yusof M Y M and Mansor M 2012 Gelam honey inhibits lipopolysaccharide-induces endotoxemia in rats through the induction of heme oxygenase-1 and the inhibition of cytokines, nitric oxide, and high-mobility group protein B1 Fitoterapia 83 1054-1059.
[7] Rahim A A 2010 Chemical composition, total phenolic content and radical scavenging activity of Melaleuca cajuputi powell essential oil
[8] Faujan N H, Alitheen N B, Yeap S K, Ali A M, Muhajir A H and Ahmad F B H 2010 Cytotoxic effect of betulinic acid and betulinic acid acetate isolated from Melaleuca cajuputi on human myeloid leukemia (HL-60) cell line African Journal of Biotechnology 9 6387-6396.
[9] Hammer K A, Carson C F and Riley T V 1999 Antimicrobial activity of essential oils and other plant extracts J. Appl. Microbiol 86 985–990.
[10] Jedlickova Z, Ery V, Mott O and Nguyen D C 1993 Antibacterial Properties of Cajuput Oil,” Medicaments Et Aliments: L’approche Ethnopharmacologique
[11] Mohamed Z 2010 Chemical composition and antioxidant potential of Melaleuca cajuputi powell (gelam) essential oil using ftc and tba method”, Final Year Project Report, Universiti Teknologi MARA, 2010.
[12] Rozaini M H Z, Ali R C and Rose L C 2012 Normal micellar value determination in single and mixed surfactant system employing fluorescence technique International Journal of Technology 2 93-99.
[13] Ng D S H, Rose L C, Suhaimi H, Mohamad H, Rozaini M Z H and Taib M 2011 Preliminary evaluation on the antibacterial activities of citrus hystrix oil emulsions stabilized by tween 80 and span 80 Int. J. Pharm. Pharm. Sci. 3 209–211.
[14] Watansirichaikul S, Davies N M, Rades T and Tucker G. 2000 Preparation of biodegradable insulin nanocapsules from biocompatible microemulsions Pharmaceutical Research 17.
[15] Ismail Z 2013 Development of high internal phase emulsions (HIPEs) as colloidal media Final Year Project Thesis, Universiti Malaysia Terengganu
[16] Sonneville-Aubrun O, Simonnet J T, Alloret F L 2014 Nanoemulsions: A new vehicle for skin care products Advances in Colloid and Interface Science 108 145-149.
[17] Epstein J H and Wang S Q 2013 “Understanding UVA and UVB. Skin Cancer Foundation,” [http://www.skincancer.org/prevention/uva-and-uvb/understanding-uva-and-uvb]
[18] Lau S L 2010 Ingredientism Beijing (China): Phoenix Books. Ltd. pp 8-11, 2010
[19] Chisvert A and Salvador A 2007 UV Filters in Sunscreens and other Cosmetics. Regulatory Aspects and Analytical Methods pp 83-120
[20] Kamel S N M 2003 Essential Oil of Gelam (Melaleuca lecadendron) as Additive in Emulsions: Its Potential for Antibacterial and Sunscreen,” Bachelor Degree Thesis, Universiti Malaysia Terengganu (UMT)
[21] Morais G G, Santos O D H, Oliveira W P and Filho P A R 2018 Attainment of O/W emulsions containing liquid crystal from annatto oil (Bixa orellana), coffee oil, and tea tree oil (Melaleuca alternifolia) as oily phase using HLB system and ternary phase diagram 29 297-306.