Trop J Nat Prod Res, February 2023; 7(2):2414-2420 ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

Tropical Journal of Natural Product Research

Available online at https://www.tjnpr.org

Original Research Article

Evaluation of the Physical Properties and Anti-aging Microemulgel Sunscreen Nyamplung Oil (*Calophyllum inophyllum* L.)

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ARTICLE INFO

Article history:
Received 12 January 2023
Revised 20 February 2023
Accepted 23 February 2023
Published online 01 March 2023

ABSTRACT

Exposure to UV rays can cause photoaging, and a photoprotective compound can be used to prevent it, as well as being used as an active ingredient in sunscreen. The potential side effects and contamination of existing sunscreens on the market call for alternatives such as natural ingredients. Nyamplung oil (*Calophyllum inophyllum* L.) has the potential to be used as a sunscreen gel, with a 30.46 SPF value in its formulation. However, Nyamplung oil has the disadvantage of being hydrophobic. Therefore, this study aims to formulate it in the form of a microemulsion gel as a dual-action sunscreen with anti-aging activity. The optimization of the microemulsion formula were performed using Design Expert software, with observations on droplet size, polydispersity index (PDI), and zeta potential. The sunscreen was tested for its SPF value, percent (%) erythema transmission (%Te), percent (%) pigmentation transmission (%Tp), and percent (%) residual elastase enzyme activity (%AE), to demonstrate its anti-aging activity, as well as being tested for its physical properties against the effects of storage, temperature, and photostability, and its safety in potential irritation on rabbits. The optimum microemulsion formula had a droplet size of 172.46 nm, a PDI of 0.196, and a zeta potential of -6.36 mV. The microemulgel sunscreen met the physical requirements of gel preparations, with an SPF value of 25.73, %Te and %Tp values for sunblock protection categories, and providing 62.96% anti-elastase activity at a concentration of 10%. The sunscreen was stable during storage and with the influence of temperature. However, the sunscreen indicated a decrease in SPF value by 18.19% after a photostability test.

***Keywords*:** microemulgel, nyamplung oil, sunscreen, anti-aging .

Introduction

UV exposure can cause damage to the skin, including sunburn, aging, and cancer. Currently, dual-action sunscreen and anti-aging preparations are widely developed. The mechanism of sunscreen and anti-aging is closely related to active substances that are capable of providing photoprotective effects.[1] Photoprotectors play a role in skin protection through two mechanisms, namely direct protection against UV rays to prevent skin damage, as well as protection against radical reactions from reactive oxygen species formed due to exposure to UV rays, which are also factors that cause skin aging.[2]

Sunscreen with organic active ingredients can provide good protection, but there is a risk of adverse effects if used in the long term, including effects on hormones, allergic reactions, and carcinogenicity.[3] In addition, due to the harmful effects of chemicals, special attention should be paid to the use of chemicals on the environment. Natural products are now in high demand as they are considered safer for long-term use and environmentally friendly.

Several types of natural ingredients are currently being developed as active ingredients for natural sunscreens, such as nyamplung oil, peppermint oil, grape seed oil, and carrot seed oil.[4,5]

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Citation: Sari PKN, Zulkarnain AK, Lukitaningsih E. Evaluation of the Physical Properties and Anti-aging Microemulgel Sunscreen Nyamplung Oil (*Calophyllum inophyllum* L.). Trop J Nat Prod Res. 2023; 7(2):2414-2420 http://.www.doi.org/10.26538/tjnpr/v7i2.18

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria.

Nyamplung oil (*Calophyllum inophyllum* L.) contains compound components in the form of neoflavonoids and pyranocoumarin derivatives with the main components being calophyllolide, inophyllums, calanolides, and tamanolides compounds.[6] Topical application of nyamplung oil was able to inhibit the increase in MMP-1 expression and the amount of collagen in the skin of male Wistar rats exposed to UV-B light.[7] In a single nyamplung oil formulation as a sunscreen gel, an SPF value of 30.46 was obtained in the UV-B absorption area.[4]

Topical formulations with active ingredients in the form of oil can be made in the form of microemulsions. A microemulsion is a stable form of emulsion because it has a globule size in the range of 10-200 nm, a clear appearance, and a high level of solubilization.[8] Sunscreen preparations are generally made in the form of lotions or gels. However, the gel form is preferred because of its appearance and ability to increase contact time on the skin.

This study aims to formulate a dual-action sunscreen preparation as an anti-aging treatment with nyamplung oil content in the form of a microemulsion gel that meets the requirements for physical properties and is safe to use on the skin.

Materials and Methods

Materials

The active ingredient used is nyamplung oil (*Calophyllum inophyllum* L.) purchased on 29 July 2022 from PT. Lansida (Indonesia), Tween 80, Tween 20, Span 20, isopropyl alcohol, Carbopol 940, TEA, aquadest (pharmaceutical grade) purchased from Bratachem® (Indonesia), dimethylsulfoxide (DMSO, p.a degree), ethanol (p.a degree), methanol (p.a degree), hydrochloric acid (p.a degree) purchased from Merck® (Indonesia); as well as elastase enzymes (Enzo Life Science, USA).

Determination of the SPF value of nyamplung oil
A preliminary test was conducted to determine the potential SPF value of nyamplung oil as an active ingredient in sunscreen. The test was performed in vitro using UV-Vis spectrophotometry. Nyamplung oil was made in different concentrations, namely 0.3 mg/mL; 0.5mg/mL; 0.7 mg/mL; and 0.9 mg/mL. The absorbance curve of the samples was measured at a wavelength of 290-320 nm with three replications, and ethanol was used as a blank. The average absorbance was then determined at each concentration with a wavelength interval of 5 nm. The SPF value was calculated based on the Mansur method, using the following formula:

$$SPF = CF \times \sum_{290}^{320} EE\ (\lambda) \times I(\lambda) \times Abs(\lambda) \quad (1)$$

Information:
EE : Erythemal effect spectrum
I : Solar intensity spectrum
Abs : Absorbance of the sunscreen sample
CF : Correction factor (= 10)
The value of EE x I is constant and is shown in Table 1.

Table 1: The normalized product function is used in the SPF calculation

Wavelength (nm)	***ee*x I**
290	0.0150
295	0.0817
300	0.2874
305	0.3278
310	0.1864
315	0.0839
320	0.0180
Total	1

Oil and S_{mix} comparison screening
The determination of the type of S_{mix} was carried out by an admixture test between oil and surfactants/cosurfactants. The mixability test was perfomed by mixing oil and several types of surfactants/cosurfactants in a ratio of 1:1, afterward the mixability and stability were observed visually. Several types of surfactants used were Tween 80, Tween 20, Span 80, and several types of surfactants, such as isopropyl alcohol, PEG 400 and glycerin.
The ratio of oil and S_{mix} was determined by mixing oil with surfactants and cosurfactants in several ratios according to the ratios at each point contained in the pseudoternary diagram. The three components were put into vials, made in a total volume of 1 mL, and stirred with a magnetic stirrer for 2 minutes at a speed of 600 rpm and a temperature of 25°C. The mixture was then emulsified in distilled water and visually observed, which were categorized into grade A, which was a nanoemulsion system with a clear appearance, grade B was a microemulsion system with a bluish appearance, and grade C was a microemulsion system with a milky white appearance. The results of the emulsification of each ratio were included in the pseudoternary diagram. The plotting results on the diagram showed the area with the ratio of oil and S_{mix} which could produce microemulsions. This data was used to determine the upper and lower levels of each component in optimization using Design Expert.

Optimization of microemulsion formulas
The upper and lower levels of oil and S_{mix} data were entered into the Design Expert software version 13 and analysed using the D-Optimal Design method. From the results of the analysis, 13 run formulas were obtained according to Table 1 and then tested and observed for the response in the form of droplet size, PDI, and zeta potential of each microemulsion. Testing the droplet size and polydispersity index was carried out by dissolving the microemulsion sample in distilled water and then analysing it using a Particle Size Analyzer. The zeta potential test was carried out by dissolving the sample in distilled water and analysing it using a zetasizer at 25°C.

Determination and verification of optimum microemulsion formula
Determination of the optimum formula was based on the results of the analysis of the response value with the highest desirability value. The optimum formula was verified by comparing the response test results data to the software's prediction data. The selected optimum formula could be accepted if the response value of the test results was included in the predicted value range.

Making the optimum microemulgel sunscreen formula
Microemulsion resulting from the optimum formula was incorporated into Carbopol 940 gel base. Microemulsion was prepared by mixing the oil phase, water phase, and S_{mix} followed by stirring at 300 rpm using a magnetic stirrer. Furthermore, the gel base was prepared by first developing a 0.5% gelling agent (Carbopol 940) in distilled water for 24 hours and then adding 0.3% TEA, stirring for 15 minutes using a homogenizer. The microemulsion formula was then added and stirred until homogeneous into a gel base with a ratio of 3:1.

Evaluation of the physical properties of microemulgel sunscreen
Organoleptic
Organoleptic observations were carried out visually, including color, odor, clarity and consistency of the microemulgel.

Homogeneity
The sample on the object glass was then pressed with another glass object until it was evenly distributed, then the homogeneity of the preparation was observed visually.

pH
Sample pH measurements were carried out using a pH meter at room temperature 25°C for sunscreen preparations.
Spreadability
Spreadability was measured by placing 0.5 g of microemulgel between two plates with an added weight of 500 g and allowing it to stand for 5 minutes.

Adhesiveness
The adhesiveness measurement was carried out by placing 0.5 g of the microemulgel preparation at the midpoint of a glass and covering it with a glass plate with an added weight of 200 g for 2 minutes. The determination of adhesiveness was in the form of the time required until the two slides could be separated.

Viscosity
Viscosity and flow properties were measured using a Brookfield viscometer at 100 rpm, spindle number 7 for 15 seconds. The results of viscosity measurements were presented in units of d.Pas.

Stability test of sunscreen microemulgel
In the storage stability test, the preparations were stored and observed for physical parameters for 4 weeks at room temperature (28±2°C). To see the effect of temperature in extreme conditions, a cycling test was carried out for 3 cycles by storing the preparation at cold temperature 4°C for 24 hours then removed and placed and stored at 40°C for 24 hours (1 cycle counted).

Photostability test of sunscreen microemulgel
Photostability testing was carried out based on Lv (2018) with modifications[9]. UV irradiation at a wavelength of 366 nm is given to sunscreen preparations. A total of 1.5 grams of microemulsion gel was irradiated with a UV lamp with a wavelength of 366 nm with various irradiation durations of 0, 1 and 2 hours in a closed cabinet. Furthermore, absorbance measurements were carried out at a wavelength of 290-320 nm using UV-Vis spectroscopy with an ethanol blank to determine any change in the SPF value of the test sample.

Determination of SPF microemulgel sunscreen
Determination of the SPF value was carried out *in vitro* using a UV-Vis spectrophotometer. The preparation was diluted using ethanol to a concentration of 10% and the absorbance was read at 5 nm interval in the wavelength range λ 290-320 nm. Ethanol was used as the blank. The calculation of the SPF value was based on the Mansur method (Equation 1 and Table 1).
Determination of %Te and %Tp microemulgel sunscreen
Calculation of the value of % erythema transmission was calculated for each wavelength from 292.5 to 317.5 nm based on the following formula:

$$\%\ Transmisi\ Eritema = \frac{\sum(T.Fe)}{\sum Fe} \qquad (2)$$

Information:
T : value of % Erythema Transmission
Fe : erythema flux constant
ΣFe : total amount of sunlight erythema flux
Σ(T.Fe): the amount of erythema flux that is attenuated by the sunscreen agent at the wavelength of the erythema spectrum.
Calculation of the value of % pigmentation transmission was calculated for each wavelength from 322.5 to 372.5 nm based on the following formula:

$$\%\ Transmisi\ Eritema = \frac{\sum(T.Fp)}{\sum Fp} \qquad (3)$$

Information:
T : value of % Erythema Transmission
Fe : erythema flux constant
ΣFe : total amount of sunlight erythema flux
Σ(T.Fe): the amount of erythema flux that is attenuated by the sunscreen agent at the wavelength of the erythema spectrum.

Microemulgel sunscreen anti-elastase activity test
The antiaging activity test was measured based on the activity of inhibiting the elastase enzyme. The elastase enzyme inhibition test was carried out based on the product manual of Enzo Life Science's Drug Discovery Kit (Manual, Neutrophil Elastase Colorimetric). 20 µL of sample solution was diluted with 65 µL of buffer solution (10 mM HEPES, 50 mM NaCl and 0.05% Tween 20 in DMSO) on a 96-well microplate. A buffer solution was used for both the blank and negative control, and Elastatinal (100 µM) was used as a comparison with the inhibitor control. An activity test was carried out by adding 2.2 µU/µL of Neutrophil elastase enzyme to the sample solution, negative control solution and 10 µL of the control inhibitor. Afterward, Incubation has been done for 10 minutes at 37°C and added by 5 µL of substrate (MeOSuc-Ala-Ala-Pro-Val-pNA, 100 µM). The samples were then measured for absorption at a wavelength of 405 nm which were observed for 10 minutes at 1 minute intervals.

Microemulgel sunscreen irritation test
The test was carried out in accordance with Regulation No. 7 of 2014, issued by the Head of BPOM RI. Albino adult rabbits, weighing 1.5-2 kg (3 tails) and in good health, were used for the test. The preparation was topically applied to the rabbits in a dose of 0.5 g and left for 4 hours. The rabbits were then observed at 1, 24, 48, and 72 hours, and up to 14 days. Observations were made on the incidence of erythema and edema in the test animals.

Ethical clearance
The protocol for the use and treatment of test animals has been approved by the Ethics Commission at Gadjah Mada University's LPPT institution with certificate number 00032/04/LPPT/X1/2022.

Statistical analysis
The entire data from the test results was carried out by replication and the mean and standard deviation was calculated. Statistical analysis was performed using SPSS 25 with ANOVA to compare mean differences between before and after testing or between each group. A $p < 0.05$ was considered as significant.

Result and Discussion

The results of measuring the SPF value of nyamplung oil alone at a concentration range of 0.2 mg/mL to 0.9 mg/mL showed a positive linear correlation with an increase in oil concentration, which was statistically significant (Figure 1). This is related to the increased content of active compounds in nyamplung oil, such as phenolic compounds, flavonoids, and coumarins. Several coumarin group compounds, identified in nyamplung oil, showed a structure with a long chromophore group and an aromatic structure followed by a conjugate bond system.[10] Compounds with this structure are able to optimally absorb UV light in the 290-320 nm range, resulting in SPF values ranging from 11 to 34, calculated using the Mansur method. In a study conducted by Rejeki, the SPF value of nyamplung oil was found to be in the range of 10-26 at concentrations between 0.2 mg/mL to 0.3 mg/mL.[4]
The microemulsion formula consists of oil, water and S_{mix}. S_{mix} is a mixture of surfactants and cosurfactants, both of which are responsible for lowering the interfacial tension between oil and water to form a thermodynamically stable colloidal dispersion system.[11] There are several factors that influence the microemulsion formulation, including the type of S_{mix} used, the ratio of oil to S_{mix}, and the amount of water phase present in the system.
The appropriate type of surfactant and cosurfactant is determined based on their ability to form a stable microemulsion system, which is visually shown as a transparent or bluish mixture. The results of the mixing test indicate that Tween 80 is the surfactant that mixes well with nyamplung oil. Tween 80 is a type of non-ionic surfactant with an uncharged hydrophilic group that is widely used in microemulsion formulations with vegetable oils. A research showed that Tween 80 can produce a more stable nyamplung oil nano emulsion system compared to Tween 20 or Tween 40 because of the presence of an unsaturated nonpolar structure that reduces the interfacial tension between oil and water.[12]
For the type of cosurfactant, the result showed that isopropyl alcohol (IPA) provides the best miscibility compared to glycerin and PEG 400. IPA is a group of short-chain alcohols widely used as cosurfactants due to their easy nature of diffusing between two phases, optimizing the performance of surfactants in reducing interface voltage. Based on existing research, IPA can act as a cosurfactant in the nyamplung oil microemulsion formulation, producing a stable microemulsion with a particle size of 34.37 ± 1.06 nm and a zeta potential of 37.48 ± 0.87 mV.[13]
The next aspect to consider is the amount of water phase, which is determined by optimizing the amount of water used to disperse the oil and S_{mix} mixture. The amount of water phase in the microemulsion affects the resulting droplet size. The optimized amount of water phase was determined to be 95%, based on consideration of globule size and desired zeta potential value. The desired values were more than 100 nm for the globule size, and more than ±5 mV at zeta potential value.
To select the microemulsion area in the form of a visual bluish mixture, an area pseudo ternary diagram is used in each category. The selected area is then used as the upper and lower levels in optimization using D Optimal mixture design in Design Expert 13 software.
The results of calculating the linear equation for the response to droplet size can be seen in equation (3).

$$Y = 8655.03\ (A) - 257.69\ (B) + 74.00\ (C) \qquad (3)$$

Information :
Y = Droplet size
A = Nyamplung oil
B = Tweens 80
C = Isopropyl Alcohol

Based on equation (3), it can be observed that the droplet size increases with the addition of the oil component (Table 2). This occurs because an increase in the oil component leads to greater flow resistance, which makes the droplet size reduction process more difficult, thus limiting

the speed of droplet formation. On the other hand, decreasing the surfactant component can result in an increase in particle size. This is because a smaller amount of surfactant reduces its ability to emulsify the oil and water phases. Conversely, a high surfactant concentration increases the flexibility of the interfacial membrane, resulting in a microemulsion with a smaller particle size.[14]
The linear equation for the PDI response is shown in equation (4).

$$Y = -50.45\ (A) - 0.13\ (B) + 0.52\ (C) \quad (4)$$

Information :
Y = PDI
A = Nyamplung oil
B = Tweens 80
C = Isopropyl Alcohol

Based on equation (4), it can be inferred that the PDI value decreases as the oil component increases (Table 2). This result contradicts the analysis of the droplet size response data. An increase in the oil component makes the droplet size reduction process more difficult, resulting in the formation of larger droplets. Larger droplets tend to coalesce more quickly, leading to a wider distribution of particle sizes and a higher polydispersity index value.

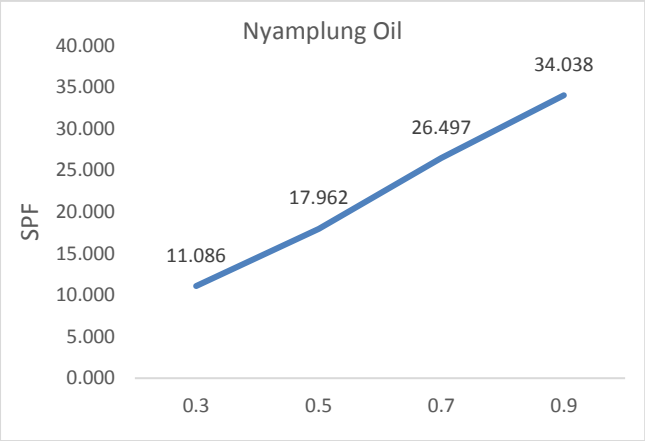

Figure 1: Graph of the relationship between Nyamplung Oil concentration (mg/mL) and SPF value.

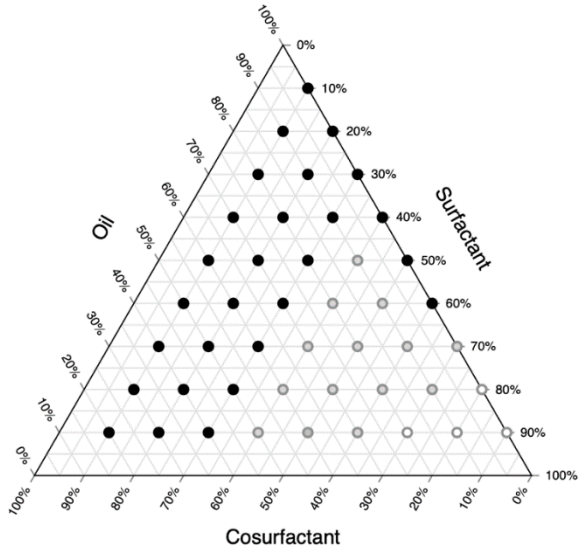

Figure 2: Pseudo ternary Diagram Nyamplung Oil Microemulsion.

The black dots indicate the emulsion area, the grey dots indicate the microemulsion area, the white dots indicate the nanoemulsion area.
The linear equation for the zeta potential response is shown in equation (5).

$$Y = 111.80\ (A) + 12.81\ (B) - 1.08\ (C) \quad (5)$$

Information :
Y = zeta potential
A = Nyamplung oil
B = Tweens 80
C = Isopropyl Alcohol

Based on the equation (5) above, it can be stated that a decrease in the oil component will lead to a higher or negative (away from 0) zeta potential value (Table 2). This can be attributed to the mechanisms of stabilization, which include electrostatic and steric stabilization. A lower oil composition causes a steric stabilizing effect to occur with the presence of surfactants and cosurfactants in the system. Conversely, if the oil composition is increased, the amount of surfactant and cosurfactant becomes insufficient to enhance the electrostatic repulsion in the system, resulting in a decrease in the zeta potential value (approaching 0).[14]
The results of the analysis based on the three responses showed that the optimum formula for nyamplung oil microemulsion consisted of 6,67% nyamplung oil; 48,69% surfactant component; 44,65% cosurfactant component; and 95% water component. This mixture of components provided response data in the form of a droplet size value of 166.47 nm, a PDI value of 0,125, and a zeta potential value of -11,48 mV, which corresponds to the predictive value and tolerance value.
The aspects of viscosity, pH, spreadability, and adhesiveness determine the comfort of application on the skin. Gels with a soft consistency will be more evenly distributed, easily absorbed, and comfortable to use on the skin. The viscosity of the preparation will affect the spreadability and adhesiveness produced.[15] The preparation is expected to have a spreadability of between 5-7 cm and an adhesiveness of >1 second. The amount of spreadability will determine the ability of the active substance to spread and come in contact with the skin, while the adhesiveness determines the ability of the preparation to stick to the skin when applied. The magnitude of the pH value affects the irritating effect that will be caused on the skin. The desired pH of the gel is close to the pH of the skin, which ranges from 4,5 to 6,5; this is because if the pH of the gel is too acidic compared to the pH of the skin, it will potentially irritate the skin. The results showed that the preparation met the criteria for a good gel preparation in terms of viscosity, pH, spreadability, and adhesiveness Table 3.
Visual observation of the preparations during the 4-week shelf life showed that the physical properties of the preparations were homogeneous and did not change in color, smell or texture, and did not show any phase separation. Other physical parameters, namely viscosity, pH, adhesiveness, and spreadability, indicated that there were changes in the overall physical parameters, but these changes were not significant and still met the requirements for acceptance of the physical properties of the preparation (Figure 3). These data indicate that the preparation is stable during a shelf life of 4 weeks at room temperature (25°C).
The results of the cycling test showed that the visual aspects of the preparation, such as homogeneity, color, odor, and texture, did not change with the effect of temperature, and there was no phase separation in the preparation. Other physical parameters, such as viscosity, pH, adhesiveness, and spreadability, have changed due to the influence of the test. Statistically, changes in viscosity and pH values occurred significantly, but not in the values of adhesiveness and spreadability. Overall, the physical parameter values were still within the desired range (Figure 4).
Sunscreen should be applied every 2 hours with the hope that they can maintain their ability to protect against UV rays, which is indicated by a maximum decrease in the SPF value of 10% to be declared as a photostable sunscreen.[16] The test results showed that the preparation decreased in SPF value by 18.19% after irradiating UV light at a wavelength of 366 nm for 2 hours (Figure 5). This value is higher than

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

the expected maximum value, and it may be caused by the inability of the preparation to provide protection against UV rays for a duration of 2 hours, so re-application with a faster duration is required. To increase the duration of use, other active ingredients can be added in combination form, or other excipients can be added to the preparation, such as the use of antioxidants.

The determination of SPF showed that the sunscreen preparation had an SPF value of 25.73 with a final oil concentration of 0.231%. This data shows that sunscreen preparation has a medium SPF strength category (15-20).[2] The ability to protect against UV rays is related to the presence of phenolic compounds, flavonoids, and coumarins in nyamplung oil.

The results of formulating nyamplung oil gel alone with a concentration of 50% showed an SPF value of 30.46.[4] These data support the statement that in the form of microemulsions, preparations are able to provide higher SPF values, thus allowing the use of a minimum amount of active ingredients to avoid potential skin irritation.

The determination of %Te and %Tp value showed that the preparation had %Te value of 0.24% and %Tp value of 1.13%. Based on the rating category of sunscreen strength, this value is included in the high SPF category. This category indicates that the preparation is able to provide full protection against the occurrence of erythema and pigmentation, which indicates that the preparation has a broad spectrum, so that it can work to provide protection in the UV A and UV B areas.

The protective activity against erythema and pigmentation provided by the nyamplung oil microemulgel sunscreen preparations is related to the presence of phenolic, flavonoid, and coumarin compound groups. These compounds play an important role in UV light absorption activity and act as antioxidants that contribute to the prevention of pigmentation resulting from non-enzymatic oxidation.[17]

The sunscreen was tested for their anti-elastase activity at three different concentrations, namely 5%, 7.5%, and 10%, with a comparison to 10% concentration of vitamin C. The inhibitory activity kinetics were observed at 1-minute intervals for 10 minutes based on absorbance data at a wavelength of 405 nm. The observed data was plotted against time until the slope value of each test sample was obtained in the form of a negative control, positive control, sunscreen preparation, and Vitamin C comparison. Enzyme inhibitory activity was expressed as the percentage of remaining enzyme activity compared to the negative control.[18] The test results showed that the inhibitory activity of the elastase enzyme at concentrations of 5% and 7.5% exhibited a significantly different ability compared to the elastatinal positive control and Vitamin C (Figure 6). However, at the highest concentration of the sunscreen preparation (10%), the preparation showed enzyme inhibitory activity that was not significantly different from the inhibitory activity of the positive control and Vitamin C. This may be due to the presence of phenolic and sterol group compounds.[19] These results were also supported by previous research that demonstrated the ability of nyamplung oil to reduce MMP-1 expression and inhibit the reduction of collagen in male Wistar rats exposed to UV-B.[7]

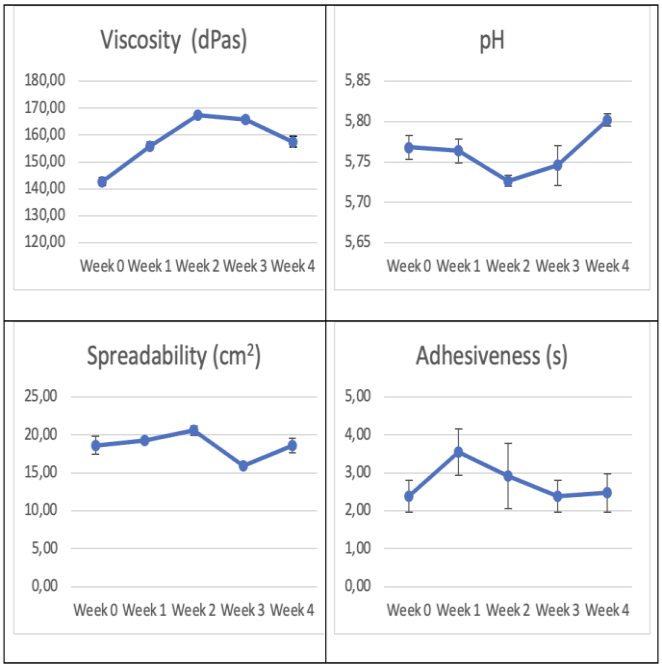

Figure 3: Graph of physical properties of microemulgel sunscreen at 4 weeks shelf life.

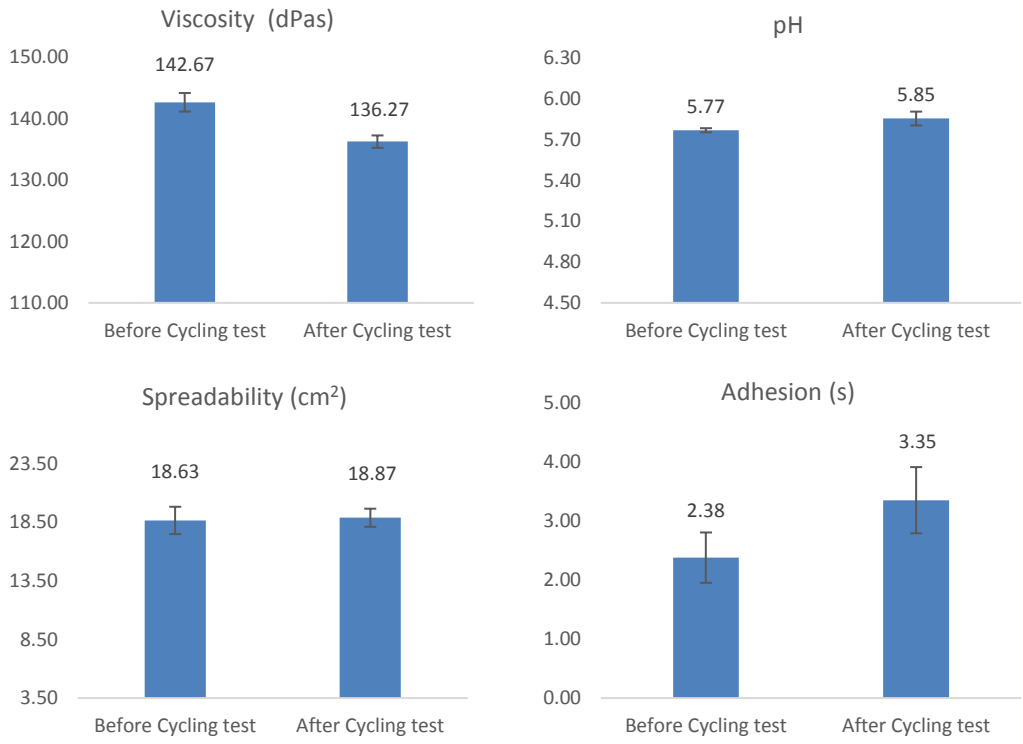

Figure 4: Graph of physical properties of microemulgel sunscreen in cycling test.

Figure 5. Graph of SPF for microemulgel sunscreen on photostability test.

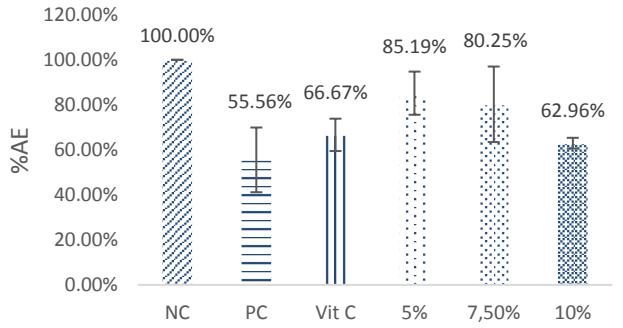

Figure 6: Graph of percent decrease in elastase enzyme activity by inhibitors.
Description: %AE-Percentage of Enzyme Activity Remaining; NC:Negative Control; PC:Positive Control.

This indicates that natural ingredients containing compounds such as triterpenes, polyphenols, and sterols have the potential to provide inhibitory activity against enzymes responsible for the degradation of elastin and collagen in the skin.[17] Based on the percentage of remaining enzyme activity, it can be concluded that the optimal concentration for inhibiting elastase enzyme activity is 10%.

The irritation test results showed that all tested animals did not show any skin irritation response in either the control or the microemulgel sunscreen during observation from 1 hour to 72 hours. Likewise, during follow-up observations for 14 days, there was no response to irritation or other clinical signs of toxicity that could be observed, such as hyperkeratosis, hyperplasia, or scaling. These results indicate that the preparation has a negligible irritation category response so that it can be said that the microemulgel sunscreen is safe for skin.

Conclusion

The optimal microemulsion formulation had a droplet size of 172.46 nm, a PDI of 0.196, and a zeta potential of -6.36 mV. The microemulsion gel meets the physical requirements of a gel formulation and is stable during storage and exposure to temperature. This sunscreen has an SPF value of 25.73 at a 10% dilution, and the sun protection category is based on % Te and % TP values and an anti-elastase activity of 62.96% at a 10% concentration. Results indicate potential for the sunscreen to be further developed to market maturity

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

Table 2: Design and response of nyamplung oil microemulsion formula.

run	Oil (%)	Surfactant (%)	Cosurfactant (%)	Droplet Size* (nm)	PDI*	Zeta Potential* (mV)
1	8.03085	50.7963	41.1729	159.53	0.228	-13,13
2	5	48.9601	46.0399	144,27	0.242	-11,13
3	5	45	50	115,10	0.304	-9.43
4	9.22146	44.5762	46.2024	152.70	0.192	-8.53
5	10.6003	40	49.3997	155.97	0.198	-7.94
6	7,5	55.0195	37.4805	129.57	0.288	-8.63
7	10.6003	40	49.3997	167.30	0.183	-8.25
8	10	60	30	15.99	0.381	-7,11
9	8.03085	50.7963	41.1729	160.47	0.226	-8.00
10	12	54.8948	33.1052	168.33	0.205	-8.63
11	12	50.3815	37.6185	176.33	0.233	-6.44
12	8.03085	50.7963	41.1729	153,13	0.200	-11.87
13	5	59,875	35,125	9,74	0.089	-1.15
14	12	46.5165	41.4835	165,63	0.201	-5.86

*Data are average values(n=3)

Table 3: Physical properties of microemulgel sunscreen.

Base	Viscosity* (dPas)	pH*	Spreadability* (cm^2)	Adhesiveness* (s)
Carbobol 0.5%	142.67 ± 1.51	5.77 ± 0.02	18,63 ± 1,16	2.38 ± 0.43

*Data are average values± SD (n=3)

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