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The Influence of Uncaria Gambir Extract and Tween 80/PEG-400 Ratio on the Physicochemical Properties and Physical Stability of Nanoemulsion-Based Moisturizer

[1] Mutia Devi Hidayati, [2] Nuril Kusumaningtyas, [3] Arief Rahmatulloh
[1, 2, 3] Department of Chemical Engineering, Politeknik Negeri Malang, Indonesia

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Corresponding Author: **Mutia Devi Hidayati**

Abstract
Uncaria gambir is a plant-derived material containing bioactive compounds that have potential for use in cosmetic formulations. However, incorporating plant extracts into nanoemulsion-based moisturizers may affect their physical characteristics during storage. This study aimed to evaluate the effects of *Uncaria gambir* extract concentration and Tween 80/PEG-400 ratio on the homogeneity, organoleptic properties, and spreadability of nanoemulsion-based moisturizer formulations. A 4 × 2 factorial experimental design was employed using four concentrations of *U. gambir* extract (0, 0.3, 0.5, and 0.7 g/100 g formulation) and two Tween 80/PEG-400 ratios (25:15 and 30:10), resulting in eight formulations (F1–F8). The formulations were evaluated during four weeks of storage based on visual homogeneity, organoleptic characteristics, and spreadability. All formulations remained homogeneous throughout the four-week storage period, with no visible phase separation, clumping, or coarse particles. Incorporating U. gambir extract primarily affected the formulations' color, which changed from transparent in the extract-free formulations to brown in the extract-containing formulations, while no noticeable changes in odor or texture were observed during storage. Spreadability showed a slight increase over the storage period, while formulations containing the 25:15 Tween 80/PEG-400 ratio generally exhibited greater spreadability than those containing the 30:10 ratio. Variations in extract concentration did not cause visible deterioration in homogeneity or organoleptic characteristics. Overall, *U. gambir* extract can be incorporated into nanoemulsion-based moisturizer formulations while maintaining acceptable physical characteristics in terms of homogeneity, organoleptic properties, and spreadability over four weeks of storage. The Tween 80/PEG-400 ratio of 25:15 provided favorable overall physical performance, particularly with respect to spreadability.

Keywords: *Uncaria Gambir*, Nanoemulsion, Moisturizer, Tween 80, PEG-400, Homogeneity, Spreadability

Introduction

The growing demand for natural, multifunctional cosmetic products has driven the development of topical formulations that incorporate plant-derived bioactive compounds. In topical applications, a formulation's effectiveness depends not only on the biological activity of its active ingredients but also on the physicochemical characteristics of the delivery system [1]. Parameters such as homogeneity, appearance, spreadability, and physical stability matter because they affect a topical product's consistency, ease of application, and overall acceptability [2, 3].

Uncaria gambir Roxb. is a plant material traditionally used in various applications and is recognized as a source of phenolic compounds, particularly catechins. The incorporation of *U. gambir* extract into topical formulations provides an opportunity to develop a plant-based cosmetic preparation with functional properties. However, incorporating a concentrated botanical extract into a semisolid formulation may alter its physicochemical characteristics, including color, viscosity, spreadability, and physical stability. Therefore, appropriate formulation design is required to obtain a preparation with acceptable physical characteristics while maintaining the properties of the incorporated bioactive extract [4, 5].

Nanoemulsion-based delivery systems have attracted considerable interest for topical formulations because their dispersed structures and interfacial characteristics can facilitate the incorporation and distribution of poorly water-soluble compounds. The composition and concentration of surfactants and cosurfactants strongly influences the physicochemical properties of

nanoemulsions. In particular, the surfactant-to-cosurfactant ratio can affect interfacial film formation, droplet formation, viscosity, and the physical behavior of the resulting formulation. Consequently, optimization of the surfactant system is an important step in developing a stable nanoemulsion-based topical preparation [6, 7, 8].

Tween 80 is a nonionic surfactant widely used in pharmaceutical and cosmetic formulations because of its ability to reduce interfacial tension and facilitate the formation of dispersed systems. PEG-400 can function as a co-surfactant and may improve the flexibility and stability of the interfacial film formed during emulsification. The combination of Tween 80 and PEG-400 therefore provides a suitable surfactant system for developing a nanoemulsion formulation. Nevertheless, changes in their relative proportions may produce different physicochemical characteristics in the final formulation. In a moisturizer system, these changes may be reflected in spreadability, as well as in the maintenance of homogeneity and appearance during storage [9, 10, 11].

The development of a *U. gambir* nanoemulsion-based moisturizer consequently requires simultaneous consideration of extract concentration and surfactant composition. Increasing the concentration of plant extract may modify the appearance and consistency of the formulation, whereas changes in the Tween 80/PEG-400 ratio may influence the dispersion characteristics and rheological behavior of the system. These effects are particularly relevant during storage because changes in physicochemical properties can indicate instability or deterioration of formulation performance.

Previous work on the present formulation system demonstrated the feasibility of incorporating *U. gambir* extract into a Tween 80/PEG-400 nanoemulsion-based moisturizer. However, a systematic evaluation of how extract concentration and surfactant ratio affect the physicochemical characteristics and short-term physical stability of the resulting moisturizer remains necessary. In particular, the relationship between formulation composition and spreadability requires further consideration because these parameters directly influence the handling and topical application characteristics of semisolid preparations [12, 13, 14].

Therefore, this study aimed to investigate the effects of *U. gambir* extract concentration and Tween 80/PEG-400 ratio on the physicochemical properties and four-week physical stability of a nanoemulsion-based moisturizer. The formulations were evaluated in terms of homogeneity, organoleptic characteristics, and spreadability. The results are expected to provide a formulation-based understanding of the influence of extract concentration and surfactant composition on the physical performance of *U. gambir* nanoemulsion-based moisturizers.

Material and Methods

Materials

Dried *Uncaria gambir* Roxb. leaves were obtained from West Sumatra, Indonesia. Sodium carboxymethyl cellulose (Na-CMC, food grade), glycerin (pharmaceutical grade), Tween 80 (polysorbate 80, Sigma-Aldrich, USA), polyethylene glycol 400 (PEG-400, Merck, Germany), extra virgin olive oil (food grade), α-tocopherol (Sigma-Aldrich, USA), triethanolamine (TEA, Merck, Germany), and ethylenediaminetetraacetic acid (EDTA, Merck, Germany) were used for the formulation. Ethanol (70%, technical grade) was used for extraction. Deionized water was used throughout the formulation and analytical procedures.

Instrumentation

The instruments used in this study included an analytical balance, a magnetic stirrer with hotplate, rotary evaporator.

Preparation of *Uncaria gambir* Extract

Fresh *U. gambir* leaves were washed thoroughly with running water and dried in an oven at 40 °C for 48 h until a constant weight was obtained. The dried leaves were subsequently ground using a blender and sieved through a 60-mesh sieve to obtain a uniform powder. A total of 100 g of gambir leaf powder was macerated with 2 L of 70% ethanol at a solid-to-solvent ratio of 1:20 (w/v). The extraction was conducted at room temperature (25 ± 2 °C) for 48 h, with intermittent stirring every 8 h. The extract suspension was filtered using Whatman No. 1 filter paper, and the filtrate was concentrated using a rotary evaporator at 40 °C under reduced pressure. The resulting viscous extract was transferred into an amber glass container and stored at 4 °C until further formulation.

Experimental Design and Formulation

A 4 × 2 factorial design was employed to investigate the effects of *U. gambir* extract concentration and Tween 80/PEG-400 ratio on the physicochemical properties of the nanoemulsion-based moisturizer. The first independent variable was the concentration of *U. gambir* extract at four levels: 0, 0.3, 0.5, and 0.7 g per 100 g of formulation. The second independent variable was the Tween 80/PEG-400 weight ratio at two levels: 25:15 and 30:10. The combination of these factors resulted in eight formulations (F1–F8). Olive oil and α-tocopherol were maintained at 3.0 and 0.3 g, respectively, in all formulations. Na-CMC, glycerin, TEA, and EDTA were used at 1.0, 2.0, 0.5, and 0.1 g per 100 g formulation, respectively. Deionized water was added to obtain a final formulation weight of 100 g.

Preparation of Nanoemulsion

The nanoemulsions were prepared using a low-energy self-emulsification method. The aqueous phase was first prepared by dissolving the predetermined amount of *U. gambir* extract in deionized water. Tween 80 and PEG-400 were separately combined according to the predetermined weight ratio and mixed using a magnetic stirrer until a homogeneous surfactant mixture (Smix) was obtained. The Smix was gradually incorporated into the aqueous phase under continuous stirring at 2000 rpm for 30 min.

The oil phase, consisting of olive oil and α-tocopherol, was subsequently added dropwise to the aqueous-Smix mixture while maintaining stirring at 2000 rpm for 60 min. Stirring was continued until a visually homogeneous dispersion was obtained. The resulting nanoemulsions were allowed to stand at room temperature for 24 h to facilitate the formation of a transparent dispersion and the removal of entrapped air bubbles.

Preparation of the Nanoemulsion-Based Moisturizer

The gel base was prepared by dispersing Na-CMC in deionized water under continuous stirring at 500 rpm until a clear and lump-free gel was obtained. Glycerin was then incorporated as a humectant, followed by EDTA as a chelating agent. Triethanolamine was subsequently added to

adjust the formulation pH toward the skin-compatible range. The gel base was stirred until homogeneous and allowed to hydrate at room temperature for 24 h before incorporation of the nanoemulsion. Each nanoemulsion was incorporated into the hydrated gel base and homogenized using a magnetic stirrer at 500 rpm for 15 min. A control moisturizer without nanoemulsion was prepared for comparison.

Physicochemical Characterization

Homogeneity

Homogeneity was evaluated by placing approximately 0.1 g of each moisturizer formulation on a glass slide and covering it with a cover glass. The preparation was visually examined under bright light for the presence of coarse particles, aggregates, or other visible inconsistencies. A formulation was considered homogeneous when no coarse particles or visible non-uniform regions were observed.

Organoleptic Properties

The organoleptic characteristics of the moisturizer formulations were evaluated in terms of color, odor, and texture. The evaluation was performed by 15 semi-trained panelists using a five-point hedonic scale, where 1 represented very poor and 5 represented excellent. The organoleptic evaluation was conducted during the storage period to monitor visible changes in the physical characteristics of the formulations.

Spreadability

Spreadability was evaluated by placing 0.5 g of moisturizer between two glass plates. A 50-g weight was applied to the upper plate for 1 min, after which the diameter of the resulting spread circle was measured.

Spreadability (S) was calculated using Eq. (1):

$$S = \frac{M \times L}{T}$$

Where S is the spreadability, M is the applied load (g), L is the spread diameter (cm), and T is the application time (s).

Physical Stability Study

The physical stability of the formulations was evaluated under three storage conditions: Refrigerated storage at 4 °C, room-temperature storage at 25 °C, and accelerated storage at 40 °C.

The formulations were stored for four weeks and evaluated at predetermined intervals (0, 1, 2, and 4 weeks). Homogeneity, color, odor, and texture were evaluated to identify visible changes in the physical characteristics of the formulations.

Results and Discussion

Homogeneity

The homogeneity of the *Uncaria gambir* nanoemulsion-based moisturizers was evaluated throughout the four-week storage period to determine whether the incorporation of the nanoemulsion into the Na-CMC gel matrix resulted in visible physical instability. As presented in Table 1, all formulations (F1–F8) remained homogeneous during the entire observation period. No visible coarse particles, clumping, aggregation, or phase separation were observed in any formulation from week 1 to week 4. This result indicates that the nanoemulsion could be incorporated into the gel matrix without producing visible instability of the final moisturizer.

The maintained homogeneity can be associated with the formulation design in which the nanoemulsion was stabilized using a combination of Tween 80 and PEG-400. Tween 80 functions as a nonionic surfactant that facilitates the formation and stabilization of the dispersed oil phase, while PEG-400 acts as a cosurfactant that can contribute to the formation of a flexible interfacial layer. The presence of the Na-CMC gel matrix may further help maintain the distribution of the dispersed phase throughout the formulation [15, 16].

Importantly, the increase in *U. gambir* extract concentration from 0 to 0.7 g/100 g did not result in an observable loss of homogeneity. Although increasing the amount of botanical extract introduces additional dissolved and suspended constituents into the formulation, no visible aggregation or phase separation was observed even at the highest extract concentration. This finding suggests that the formulation composition was sufficiently compatible to maintain a visually uniform preparation during the four-week observation period [17].

Similarly, changing the Tween 80/PEG-400 ratio from 25:15 to 30:10 did not produce a detectable difference in homogeneity. Both surfactant systems maintained uniformity throughout storage. Thus, within the formulation range investigated, neither extract concentration nor the Tween 80/PEG-400 ratio caused visible deterioration in the moisturizer's physical uniformity [18, 19].

However, visual homogeneity should be interpreted as a macroscopic physical assessment rather than definitive evidence of colloidal stability. The absence of visible particles or phase separation does not exclude changes occurring at the microscopic or nanoscale level. Therefore, homogeneity in the present study primarily demonstrates the maintenance of acceptable visual uniformity during storage.

Table 1: Homogeneity Test Results for Moisturizer

Analysis	Formula	Storage duration (Week)			
		1	2	3	4
Homogeneity	F1	homogen	homogen	homogen	homogen
	F2	homogen	homogen	homogen	homogen
	F3	homogen	homogen	homogen	homogen
	F4	homogen	homogen	homogen	homogen
	F5	homogen	homogen	homogen	homogen
	F6	homogen	homogen	homogen	homogen
	F7	homogen	homogen	homogen	homogen
	F8	homogen	homogen	homogen	homogen

Organoleptic Properties

The organoleptic characteristics of the formulations were evaluated based on color, odor, and texture during the four-week storage period. The evaluation was used to monitor visible and sensory changes that could indicate physical deterioration of the moisturizer.

The results showed that all formulations maintained their characteristic appearance, odor, and texture throughout storage. No noticeable changes were observed in these parameters during the observation period, indicating that the formulations maintained acceptable organoleptic characteristics under the storage condition employed.

A clear difference in color was observed between formulations without and with *U. gambir* extract. Formulations F1 and F5, which did not contain *U. gambir*

extract, remained transparent. In contrast, formulations F2–F4 and F6–F8 exhibited light to dark brown coloration. The difference in color was associated with the presence of *U. gambir* extract, whose naturally occurring phenolic constituents contribute to its characteristic coloration. The intensity of the brown color tended to increase with increasing extract concentration [20].

The color characteristics are therefore primarily determined by the botanical extract rather than by the Tween 80/PEG-400 ratio. Formulations containing the same extract concentration but different surfactant ratios showed comparable visual characteristics. This observation suggests that changing the surfactant composition within the investigated range did not substantially alter the macroscopic appearance of the final moisturizer [21].

During storage, no obvious progressive darkening or other noticeable color change was observed in any formulation. This result indicates that the formulations maintained their visible color characteristics throughout the four-week observation period. Nevertheless, the absence of visible color change should not be interpreted as direct evidence of chemical stability because organoleptic observation cannot quantitatively determine changes in individual phytochemical constituents [22].

In terms of odor, all formulations exhibited a mild characteristic odor associated with the formulation components. No pronounced unpleasant or rancid odor was detected during the storage period. The presence of *U. gambir* extract did not produce a substantial change in the overall odor characteristics of the moisturizer. The absence of a noticeable odor change indicates that no obvious sensory deterioration occurred during the four-week observation period [20, 22].

The texture of all formulations remained smooth and semisolid throughout storage. No visible liquefaction, coarse aggregation, or structural separation was observed. This consistent texture, together with the maintained homogeneity, indicates that the nanoemulsion could be incorporated into the Na-CMC-based moisturizer without causing an obvious deterioration of the physical appearance of the product.

Overall, the organoleptic observations demonstrate that the formulations retained their characteristic color, odor, and texture throughout four weeks of storage. Both Tween 80/PEG-400 ratios were capable of maintaining acceptable organoleptic characteristics across the investigated extract concentrations. These findings support the physical compatibility of the nanoemulsion formulations with the moisturizer base [23].

Spreadability

Spreadability is an important quality attribute of semisolid topical formulations because it determines the ease with which a product can be distributed over the skin surface. A formulation with adequate spreadability can be applied more uniformly and with less mechanical effort, which is relevant to its practical use as a moisturizer.

Fig 1 presents the spreadability of formulations F1–F8 during the four-week storage period. Overall, the spreadability values of all formulations showed a slight increase during storage. Despite these changes, the formulations retained their ability to spread uniformly under the applied load throughout the observation period.

The gradual increase in spreadability suggests that the internal structure of the semisolid formulations underwent minor changes during storage. Such changes may be associated with gradual rearrangement of the polymeric gel network and redistribution of the formulation components during the storage period. Because no visible phase separation or loss of homogeneity was observed, the increase in spreadability was not accompanied by an obvious failure of the formulation structure [24].

The formulation groups prepared using different Tween 80/PEG-400 ratios also showed differences in spreadability. Formulations F1–F4, containing the 25:15 Tween 80/PEG-400 ratio, generally exhibited greater spreadability than formulations F5–F8 prepared using the 30:10 ratio.

This difference indicates that the relative proportion of Tween 80 and PEG-400 can influence the deformation and flow behavior of the final semisolid formulation. Increasing the Tween 80 proportion while reducing the PEG-400 proportion may alter the internal organization of the dispersed system and the interaction between the nanoemulsion and the Na-CMC matrix. As a consequence, the resistance of the formulation to deformation under the applied load may change, resulting in differences in spreadability [25].

The effect of *U. gambir* extract concentration was less pronounced than the effect associated with the surfactant ratio. Within each surfactant system, increasing the extract concentration did not produce a consistent linear decrease or increase in spreadability. This suggests that the contribution of the extract to the spreading behavior is influenced by the overall formulation composition rather than being determined solely by extract concentration.

During storage, the slight increase in spreadability observed across the formulations may also indicate gradual equilibration of the formulation components after preparation. The nanoemulsion droplets, surfactant system, botanical extract, and Na-CMC matrix may undergo structural rearrangement during storage until a more equilibrated semisolid structure is established. Since the changes were relatively small and were not accompanied by visible phase separation or loss of homogeneity, the observed variation can be regarded as a change in formulation consistency rather than evidence of physical failure [22].

An important observation is that all formulations retained their spreading characteristics throughout the four-week storage period. The combination of maintained homogeneity, stable organoleptic characteristics, and relatively consistent spreadability indicates that the developed nanoemulsion-based moisturizers possessed acceptable physical characteristics during the investigated storage period.

Taken together, the spreadability results demonstrate that both Tween 80/PEG-400 systems were capable of producing a usable semisolid moisturizer containing *U. gambir* extract. The 25:15 ratio generally provided higher spreadability than the 30:10 ratio, while all formulations maintained their spreading characteristics during storage. These results indicate that the surfactant composition is an important

formulation variable when optimizing the application characteristics of *U. gambir* nanoemulsion-based moisturizers [24, 25].

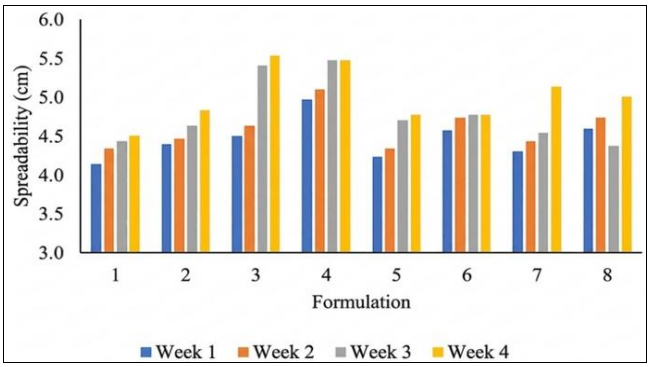


Fig 1: Spreadability Test of Moisturizers F1 – F8

Overall Physical Performance

The combined evaluation of homogeneity, organoleptic characteristics, and spreadability provides an overall assessment of the macroscopic physical performance of the developed moisturizers. All eight formulations remained homogeneous throughout the four-week observation period, with no visible phase separation or coarse particles. Their characteristic color, odor, and semisolid texture were also maintained during storage [26].

The absence of visible deterioration across the formulations indicates that the incorporation of *U. gambir* extract and the two Tween 80/PEG-400 ratios investigated did not compromise the basic physical acceptability of the moisturizer. Meanwhile, the slight changes in spreadability during storage suggest that the formulations underwent minor structural adjustments without apparent loss of physical integrity [27, 28].

Among the investigated formulation variables, the Tween 80/PEG-400 ratio appeared to have a more noticeable influence on spreadability, whereas extract concentration primarily affected the visual color of the formulation. Importantly, neither factor caused observable loss of homogeneity during the four-week observation period [28].

Therefore, based solely on homogeneity, organoleptic characteristics, and spreadability, the developed *U. gambir* nanoemulsion-based moisturizers demonstrated satisfactory macroscopic physical stability during four weeks of storage. Further instrumental characterization would nevertheless be required to establish the underlying microstructural and rheological mechanisms responsible for the observed differences among formulations.

Conclusion

The incorporation of *Uncaria gambir* extract and different Tween 80/PEG-400 ratios produced nanoemulsion-based moisturizers with good physical characteristics in terms of homogeneity, organoleptic properties, and spreadability. All formulations (F1–F8) remained homogeneous during four weeks of storage, with no visible phase separation, clumping, or coarse particles. The incorporation of *U. gambir* extract mainly influenced the color of the formulations, resulting in a characteristic brown appearance, while no noticeable deterioration in color, odor, or texture was observed during the storage period.

Spreadability showed a slight increase during four weeks of storage, indicating that the formulations maintained their ability to spread on the surface without visible physical instability. The formulations containing a Tween 80/PEG-400 ratio of 25:15 generally exhibited higher spreadability than those containing a 30:10 ratio, suggesting that the surfactant combination influenced the physical spreading behavior of the moisturizer formulations. Meanwhile, variation in *U. gambir* extract concentration did not result in a consistent deterioration of homogeneity or organoleptic characteristics.

Overall, the results demonstrate that *U. gambir* extract can be incorporated into nanoemulsion-based moisturizer formulations while maintaining acceptable homogeneity, organoleptic characteristics, and spreadability over four weeks of storage. Among the tested formulations, the 25:15 Tween 80/PEG-400 system provided favorable physical performance, particularly in terms of spreadability. Further studies involving instrumental characterization, chemical stability, and longer-term storage are required to confirm the nanoscale and overall stability of the formulations.

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