

# Formulation and Physicochemical Evaluation of Nanoemulgel Star Anise Essential Oil (*Illicium verum* Hook. f.) and Butterfly Pea Extract (*Clitoria ternatea* L.) as Antioxidant Cosmetics

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## Abstract

Oxidative stress from reactive oxygen species (ROS) is a major factor in various skin issues, including inflammation, dullness, uneven tone, and impaired barrier function. *Illicium verum* essential oil and *Clitoria ternatea* extract are natural antioxidants with complementary properties—one is oil-based, effective in skip lipid layers, while the other is water-based, active in hydrophilic environments. Their combination offers broader antioxidant protection, but different solubility properties—one is oil-based and the other is water-based—which makes it challenging to combine them in one stable product. This study developed a nanoemulgel combining these actives using varying emulsifier ratios of Tween 80 and Span 80 to optimize the hydrophilic-lipophilic balance (HLB). High-shear homogenization was used to produce nanoemulsions, which were then gelled with Carbopol 940. Four formulations (F1 - F4) were characterized for organoleptic properties, pH, viscosity, spreadability, droplet size, polydispersity index (PDI), zeta potential, and antioxidant activity via DPPH assay. Among all formulations, F3 (HLB 11.79) exhibited the smallest droplet size ( $134.7 \pm 2.1$  nm), lowest PDI (0.183), and zeta potential ( $-32.1 \pm 1.3$  mV), indicating superior nanoemulsion stability. F3 also showed optimal pH (5.79), moderate viscosity, and good spreadability. Antioxidant activity analysis revealed that F3 had the highest scavenging capacity, with an  $IC_{50}$  of  $98.53 \mu\text{g/mL}$ . Statistical analysis confirmed significant differences between formulations ( $p < 0.05$ ), with F3 being significantly superior. F3 was identified as the optimal nanoemulgel formulation, combining physical stability and strong antioxidant activity. These findings support the potential of nanoemulgel systems as effective carriers for delivering plant-based actives in cosmetic formulations. Future research should focus on evaluating their *in vivo* efficacy and assessing formulation stability under real-time storage conditions.

**Keywords:** *Illicium verum*, *Clitoria ternatea*, Nanoemulgel, Physicochemical, Antioxidant

## Introduction

Cosmeceuticals, which combine cosmetic and pharmaceutical properties, have garnered increasing attention for their ability to deliver bioactive compounds

that enhance both the appearance and physiological function of the skin [1]. One major focus in modern skincare is antioxidant, particularly targeting oxidative

stress-induced damage caused by intrinsic and environmental factors such as ultraviolet (UV) radiation, pollution, and lifestyle factors [2]. These factors increase the production of reactive oxygen species (ROS), trigger biological responses in the skin, including the activation of key signaling pathways such as MAPK and NF- $\kappa$ B. These pathways then increase the production of enzymes called matrix metalloproteinases (MMPs), which are responsible for breaking down important structural proteins like collagen and elastin [3].

Natural antioxidants—particularly those derived from plants—have attracted growing interest in cosmeceutical research and development due to their ability to target multiple skin-related pathways, their favorable safety profile, and their alignment with consumer demand for natural, clean-label ingredients [4]. In recent years, the global cosmeceutical market has experienced remarkable growth, driven by increasing awareness of skin health and a clear preference for natural, multifunctional alternatives over synthetic compounds [5]. In line with this trend, plant-based antioxidants rich in flavonoids, phenolic acids, and terpenes have emerged as promising candidates due to their strong antioxidant and anti-inflammatory activities, making them highly effective for protecting the skin from oxidative damage while supporting regeneration and repair [6].

*Illicium verum* (star anise) is traditionally used as a spice in the food industry; however, its essential oil and fruit have gained increasing attention in pharmaceutical and cosmetic applications due to their diverse biological activities, including antibacterial, antifungal, anti-inflammatory, and antioxidant effects. *I. verum* contains phenylpropanoids, flavonoids, neolignans, monoterpenoids, and sesquiterpenoids. The dominant component in its essential oil is trans-anethole, phenylpropanoids, which makes up 72% - 92% of the oil [7-9]. In cosmetic applications, these bioactives help neutralize free radicals and reduce oxidative stress, thereby preserving skin elasticity and minimizing signs of aging such as wrinkles and pigmentation [9,10]. The antioxidant activity of *I. verum* essential oil has been demonstrated in DPPH assays, with an IC<sub>50</sub> value of 9.88 mg/mL [9]. Additionally, according to the European Commission's CosIng database, *I. verum* is approved for cosmetic use as a

skin-conditioning agent and fragrance ingredient, supporting its multifunctional role in skincare formulations [10].

*Clitoria ternatea* (butterfly pea) is a well-known Indonesian medicinal plant rich in bioactive compounds such as anthocyanins, flavonoids, phenolic acids, and saponins, which contribute to its strong antioxidant and skin-protective properties [11]. Ethanolic extracts of *C. ternatea* have demonstrated significant antioxidant activity, with reported IC<sub>50</sub> values  $41.36 \pm 1.19$   $\mu$ g/mL in DPPH assays [12]. Topically, its efficacy has been supported by studies showing that formulations containing *C. ternatea* extract (100 mg/mL) enhanced fibroblast proliferation and collagen thickness *in vivo* Nursyafillah *et al.* [13], while emulgel preparations with 20 mg/mL demonstrated improved skin hydration, elasticity, and formulation stability [14]. These findings highlight its potential as a multifunctional antioxidant ingredient for cosmetic applications.

Furthermore, the combination of *I. verum* and *C. ternatea* offers a complementary therapeutic profile: *I. verum* contributes anti-inflammatory and antimicrobial properties Cosmetic Ingredient Database [10], while *C. ternatea* enhances antioxidant protection and promotes collagen synthesis [15,16]. The synergistic combination of *Illicium verum* and *Clitoria ternatea* extracts may provide enhanced skin-regenerative effects compared to using either extract alone. However, their integration into a single formulation poses challenges due to their differing solubility profiles—trans-anethole from *I. verum* is lipophilic, while anthocyanins from *C. ternatea* are hydrophilic [17]. This incompatibility can affect formulation stability and reduce overall efficacy. Formulating plant-based extracts into emulsions can help address these issues by improving solubility, uniform distribution, and delivery of both hydrophilic and lipophilic compounds. Therefore, conventional emulsions are often insufficient due to risks of phase separation, low dermal absorption, and limited bioavailability when attempting to integrate such diverse extracts [18]. Developing plant-based extracts in emulsion form enhances the delivery and bioavailability of plant-derived actives by promoting better skin penetration. These systems are commonly formulated into cosmetic preparations such as serums, creams, and lotions [19,20].

The nanoscale with droplet sizes between 20 - 200 nm, offer even greater benefits by enhancing surface area and skin permeability, leading to better absorption and improved bioavailability of the active compounds. To further improve the stability and applicability of nanoemulsions in topical applications, they can be incorporated into a gel base to create a nanoemulgel. Nanoemulgels combine the benefits of both nanoemulsions and hydrogels—offering excellent stability, ease of application, improved skin adherence, and prolonged retention time on the skin surface. While nanoemulsions alone provide increased solubility, penetration, and bioavailability of active ingredients, their low viscosity can limit practical use in topical products. The gel matrix not only resolves this issue but also provides a soothing, moisturizing and non-greasy texture preferred in skincare applications. Moreover, nanoemulgels help control the release of active compounds, which may enhance their antioxidant efficacy over time.

The nanoemulgel formulation will be developed using a combination of nonionic surfactants—Tween 80 and Span 80—which are widely used in topical products due to their safety, skin compatibility, and ability to reduce interfacial tension. The hydrophilic-lipophilic balance (HLB) system of these emulsifiers allows for precise adjustment of the surfactant ratio to match the required HLB of the oil phase, ensuring stable emulsion formation. A comprehensive evaluation of the nanoemulgel's physical and efficacy properties—including organoleptic, pH, viscosity, spreadability, droplet size, polydispersity index (PDI), zeta potential, and antioxidant activity—is crucial to ensure formulation quality, stability, and efficacy.

## Materials and methods

### Materials

Star anise and butterfly peas were taken from local farmers Gunung Arjuno area, Surabaya East Java, and determined at UPT Herbal Materia Medika Laboratory Batu, East Java, Indonesia. 96% ethanol (PT. Brataco, Surabaya), methanol pa, DPPH (Sigma), ascorbic acid (Merck), olive oil, Span 80 (CV. Chemical Indonesia Multi Sentosa, Surabaya), Tween 80 (PT. Brataco, Surabaya), phenoxyethanol, PEG 400, Carbomer 940, triethanolamine (CV. Chemical Indonesia Multi Sentosa, Surabaya), aquadest.

### Preparation of star anise essential oil

The essential oil was extracted through the steam distillation method, adapted from Qin *et al.* [21] with some modifications to enhance yield and purity. The ratio of plant material to water was adjusted to 500 g of dried star anise in 7.0 L of distilled water to improve yield. The distillation process was conducted at an operating temperature of 100 °C and atmospheric pressure of 1 atm. The distillation process was carried out for 6 h to ensure optimal extraction of essential oil. The distillation duration was extended from 4 to 6 h to ensure optimal extraction of volatile components. Once the water boiled, steam extracted volatile compounds from the plant material and condensed into a liquid distillate. The resulting distillate was separated into 2 distinct layers: The upper layer consisting of essential oil and the lower layer composed of hydrosol. After distillation, the collected essential oil was dried using anhydrous sodium sulfate (Na<sub>2</sub>SO<sub>4</sub>) to remove residual moisture. The purified essential oil was then weighed and stored in an amber-colored, well-sealed glass bottle to prevent oxidation and degradation.

### Preparation of butterfly pea extract

The sample extraction process according to Widyowati *et al.* [22] was extracted using 70% ethanol by maceration. A total of 500.0 g flowers were macerated in 5.0 L of 70% ethanol for 24 h, ensuring complete submersion of the powder, with periodic stirring to enhance extraction. The mixture had filtration through a Buchner funnel, followed by maceration until a consistent % brix was achieved, repeated 3 times. The filtrates from both maceration processes were then concentrated with a rotary evaporator at temperatures ranging from 45 - 55 °C to remove the solvent. The concentrated extract was oven-dried to a thick, constant-weight extract. Finally, extraction efficiency was determined based on the extract yield, which was calculated using Eq. (1).

$$\% \text{ yield} = \frac{\text{Weight of the dried extract}}{\text{Initial weight of the crude material}} \times 100 \% \quad (1)$$

### Preparation of nanoemulgel

The active ingredients were formulated using a fixed concentration of star anise essential oil (1%) and butterfly pea extract (2.5%), both of which exceed their

reported IC<sub>50</sub> values from previous studies. Star anise essential oil demonstrated antioxidant activity with an IC<sub>50</sub> of approximately 9.88 mg/mL (~0.98 %w/v) while butterfly pea extract showed potent activity with IC<sub>50</sub> values ranging from 4 mg/mL (~0.4 %w/v) in DPPH assays. Surfactant ratios of Tween 80 and Span 80 were varied to achieve different HLB values: F1 (6:4), F2 (7:3), F3 (8:2), F4 (9:1) as shown in **Table 1**. The formulation process followed the method described by

[24]. First, the oil phase (containing star anise essential oil, PEG 400, and Span 80) and the aqueous phase (containing butterfly pea extract, Tween 80, ethanol 96%, and distilled water) were prepared separately and heated to 40 ± 2 °C. The oil phase was slowly added dropwise to the aqueous phase under continuous stirring. Both phases were then homogenized using a magnetic stirrer at 600 rpm for 5 min.

**Table 1** formula of nanoemulgel star anise essential oil and butterfly pea extract.

Materials	Used	F1 (6:4)	F2 (7:3)	F3 (8:2)	F4 (9:1)
Star anise essential oil	Active (API)	2.00	2.00	2.00	2.00
Butterfly pea extract	Active (API)	1.00	1.00	1.00	1.00
Olive oil	Emollient	0.50	0.50	0.50	0.50
PEG 400	Humectant	5.00	5.00	5.00	5.00
Tween 80	Emulsifier	9.00	10.5	12.0	13.5
Span 80	Emulsifier	6.00	4.50	3.00	1.50
Ethanol 96 %	Co-solvent	3.00	3.00	3.00	3.00
Carbopol 940	Gelling agent	1.00	1.00	1.00	1.00
TEA	Alkalizer	0.75	0.75	0.75	0.75
Phenoxyethanol	Preservative	0.25	0.25	0.25	0.25
Distilled Water	Solvent	ad 100	ad 100	ad 100	ad 100

### Characterization of nanoemulgel

#### *Organoleptic evaluation*

The organoleptic characteristics of the nanoemulgel were evaluated by observing its color, odor, and phase separation. The formulation was evaluated for homogeneity, consistency, and ease of application on the skin [23].

#### *Droplet size and polydispersity Index (PDI) examination*

The size of the droplets and the polydispersity index (PDI) were measured simultaneously by diluting 1 mL of nanoemulsion in 25 mL of distilled water, followed by analysis in a cuvette using a Particle Analyzer (Delsa™ Nano C, US) [24].

#### *Zeta potential examination*

The zeta potential was determined using a Particle Analyzer (Litesizer 500, Anton Paar, Austria). A 50 µL sample was diluted in 2 mL of distilled water and

subsequently injected into a disposable zeta cell (DT1060C) [25].

#### *pH assessment*

A 2 g sample of the nanoemulgel was diluted in 20 mL of distilled water. The pH of various formulations was determined using a pH meter (Eutech pH 700, US). This measurement was conducted in triplicate, following the method described in reference [24].

#### *Measurement of viscosity*

A 100 g sample was carefully positioned on the lower plate of the rheometer, with the cone spindle (CP-52) set to rotate at 100 rpm to attain an appropriate torque range of 10 to 100. The measurements were conducted under controlled conditions, maintaining a temperature of 25 ± 1 °C and relative humidity of 48%, utilizing a Brookfield Digital Rheometer (DV-I<sup>+</sup>, US) [26].

### Spreadability test

The spreadability of the nanoemulgel was assessed using the glass plate method. A 0.5 g sample was placed at the center of a circle with a 1 cm diameter inscribed on a glass plate. A second glass plate was carefully positioned on top of the first. Different weights (2 g, 5 g, and increments of 5 g) were then applied for 1 min. The diameter of the spread area was measured after each weight application [27].

### Evaluation of antioxidant capacity

The antioxidant activity of the nanoemulgel formulations was evaluated using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay. A stock solution was prepared by dissolving 10 mg of the nanoemulgel in 10 mL of methanol to obtain a concentration of 1000 µg/mL. Serial dilutions were then performed with methanol to prepare working solutions in the range of 25, 50, 75, 100, 200, 500 µg/mL. For each concentration, 100 µL of the sample was transferred into a 96-well microplate, followed by the addition of 100 µL of freshly prepared DPPH solution (0.1 mM in methanol). A blank solution containing only methanol and DPPH served as the negative control. The microplate was covered with aluminum foil to protect it from light, gently shaken for 2 min, and incubated in the dark at room temperature (25 °C) for 30 min. After incubation, the absorbance of each well was measured at 517 nm using a microplate reader [28]. The radical scavenging activity was expressed as the percentage of inhibition using the following Eq. (2):

$$\% \text{ DPPH scavenging} = \frac{\text{Abs control} - \text{Abs sample}}{\text{Abs control}} \times 100 \% \quad (2)$$

where Abs control is the absorbance of the DPPH solution without sample, and Abs is the absorbance in the presence of the sample.

### Statistical analysis

The results are presented as mean values  $\pm$  standard deviation (SD) derived from 3 replicates ( $n = 3$ ). The data were analyzed using the statistical program SPSS software (SPSS for Windows, Version 27, IBM, SPSS Inc) used for statistical analysis. One-way ANOVA followed by Tukey's Multiple Comparison test

were used to compare groups with a significance level of 0.5 ( $p$ -value  $< 0.05$ ).

### Results and discussion

The selection of surfactant blends in this study was guided by the required hydrophilic-lipophilic balance (HLB) of the oil phase, primarily composed of star anise essential oil. Although the exact required HLB value for star anise oil has not been extensively documented, its chemical composition—rich in anethole—shares similarities with other essential oils such as clove and fennel, which typically require an HLB in the range of 11 to 14 for stable oil-in-water emulsions. Therefore, the formulations were designed using varying ratios of Tween 80 (HLB = 15.0) and Span 80 (HLB = 4.3) to achieve this expected HLB range. The calculated HLB values for the formulations were: 10.72 (F1, 6:4), 11.79 (F2, 7:3), 12.86 (F3, 8:2), and 13.93 (F4, 9:1), allowing for the evaluation of emulsifier composition on nanoemulsion characteristics. Ethanol was used as a co-surfactant to reduce interfacial tension, enhance solubility of poorly water-soluble compounds, and improve emulsion formation. Its presence facilitates better dispersion of star anise oil in the aqueous phase, contributing to overall nanoemulsion stability.

The organoleptic assessment of the 4 nanoemulgel formulations containing *I. verum* essential oil and ethanolic extract of *C. ternatea* (Table 2, Figure 1), F1 and F2 exhibited a milky blue appearance with smooth gel-like consistency; however, both showed signs of phase separation, indicating insufficient emulsion stability and lower homogeneity. This instability may be attributed to larger droplet sizes resulting from less optimal HLB values, which hindered effective emulsification and uniform dispersion. In contrast, formulation F3 (HLB 12.86) presented clear bluish-purple, smooth, semi-solid gel texture without any visible signs of phase separation, demonstrating homogeneity. Formula F4 showed a bluish-purple but slightly opaque appearance, no observed phase separation, but thicker consistency and reduced spreadability compared to F3. The odor of the formulations was dominated by the characteristic aroma of *I. verum* essential oil, which was strong yet pleasant aromatic. The bluish-purple color, attributed to the presence of anthocyanins in the *C. ternatea* extract.

**Table 2** Organoleptic test results for Nanoemulgel Star Anise Essential Oil and Butterfly Pea Extract.

Formula	Color	Odor	Consistency	Homogeneity
F1	Milky blue	Strong star anise aromatic	Smooth gel-like	Signs of phase separation
F2	Milky blue	Strong star anise aromatic	Smooth gel-like	Signs of phase separation
F3	Clear bluish-purple	Strong star anise aromatic	Smooth gel-like	No visible separation
F4	Bluish-purple, slightly opaque	Strong star anise aromatic	Slightly thicker, less spreadable	Slightly denser than F3

The characterization of droplet size, polydispersity index (PDI), and zeta potential revealed significant differences among the 4 nanoemulgel formulations (**Table 3**), which were influenced by the emulsifier ratio and resulting HLB values. Formula F3, with an HLB value of 12.86, demonstrated the smallest droplet size ( $134.7 \pm 2.1$  nm) and the lowest PDI (0.183), indicating a uniform and homogeneous nanoemulsion system. This can be attributed to the emulsifier blend in F3 closely matching the required HLB of the oil phase, resulting in optimal reduction of interfacial tension and effective dispersion of the oil droplets. In contrast, F1 (HLB 10.72) exhibited the largest droplet size ( $254.6 \pm 3.2$  nm) and a higher PDI (0.394), suggesting poor

emulsification efficiency and less uniform droplet distribution due to excessive hydrophilic surfactant content. Similarly, F2 (HLB 11.79) and F4 (HLB 13.93) showed increased droplet size  $181.2 \pm 2.8$  nm and  $196.4 \pm 4.0$  nm, respectively. Previous studies have established that transdermal permeation can be achieved with 200 nm nanoemulsions, indicating significant therapeutic possibilities [30]. Moreover, smaller droplet sizes, specifically those less than 80 nm, have the capability to diffuse into the viable epidermis without penetrating the skin, while larger droplets, exceeding 500 nm, tend to migrate alongside hair follicle canals [30,31].



**Figure 1** organoleptic appearance of nanoemulgel formulations.

The polydispersity index (PDI) analysis revealed notable differences in particle size uniformity across all formulations, lower PDI values suggest a more uniform distribution and minimal particle size variation Bayat *et al.* [33], Li *et al.* [34], whereas higher values indicate increased variability. F3 exhibited the most optimal result with a PDI of 0.183, indicating a highly homogeneous and stable system. This low value suggests efficient emulsification and minimal particle size variation, which is crucial for maintaining long-term stability and bioavailability in topical applications.

F2 also demonstrated acceptable uniformity with a PDI of 0.276, falling within the ideal range ( $< 0.3$ ), is indicative of a homogeneous system. In contrast, F1 (0.394) and F4 (0.321) showed significantly higher PDI values ( $> 0.3$ ), suggesting broader size distribution and reduced emulsification efficiency. According to literature, PDI values below 0.3 indicate monodispersity and formulation stability, while values between 0.3 - 0.5 are still considered acceptable for nanoparticle systems but may show increased risk of instability due to droplet coalescence [19,33].

**Table 3** Characteristics of Nanoemulgel Star Anise Essential Oil and Butterfly Pea Extract.

Characteristics	F1	F2	F3	F4
pH	5.42 ± 0.05	5.61 ± 0.04	5.79 ± 0.03	5.53 ± 0.04
Viscosity (cP)	12800 ± 120	17050 ± 150	16100 ± 110	18500 ± 125
Droplet size (nm)	254.6 ± 3.2	181.2 ± 2.8	134.7 ± 2.1	196.4 ± 4.0
PDI	0.394 ± 0.012	0.276 ± 0.009	0.183 ± 0.007	0.321 ± 0.011
Zeta potential (mV)	-23.4 ± 1.2	-27.6 ± 1.5	-32.1 ± 1.3	-25.0 ± 1.4
Spreadability (cm)	5.87 ± 0.17	4.72 ± 0.12	5.12 ± 0.14	4.38 ± 0.11

Note: Data are presented as mean ± standard deviation (n = 3).

Zeta potential measurements further supported the superior stability of F3, which showed the most negative value at  $-32.1 \pm 1.3$  mV, indicating strong electrostatic repulsion between droplets and reduced risk of aggregation. Formulations F1 and F4 exhibited less negative zeta potentials ( $-23.4$  and  $-25.0$  mV, respectively), reflecting lower colloidal stability. The potential of zeta, which measures the surface electrostatic charge of the droplet, plays a crucial role in determining the physicochemical properties of nanoemulsions, including aggregation and flocculation [36]. According to Wilson *et al.* [37], nanoemulsions with droplet sizes below 200 nm and PDI values under 0.3 are considered physically stable and well-dispersed systems, while zeta potentials beyond  $\pm 30$  mV contribute significantly to colloidal stability by creating sufficient charge repulsion to inhibit coalescence and flocculation [37].

The pH values of all nanoemulgel formulations (F1 - F4) were within the acceptable range for topical application, typically between 4.5 and 6.5 [38]. Among the formulations, F3 showed a pH of  $5.79 \pm 0.03$ , which closely matches the natural pH of the skin and is therefore unlikely to cause irritation or barrier disruption. F1 and F4 showed slightly lower pH values ( $5.42 \pm 0.05$  and  $5.53 \pm 0.04$ , respectively), possibly due to differences in surfactant concentrations affecting the microenvironment. Maintaining pH within this physiologically compatible range is crucial for product safety and user comfort.

Viscosity plays a vital role in the stability and application characteristics of a nanoemulgel. Viscosity is an important factor in drug delivery through the skin because the viscosity of topical formulations directly affects drug release, distribution, stability, and ease of

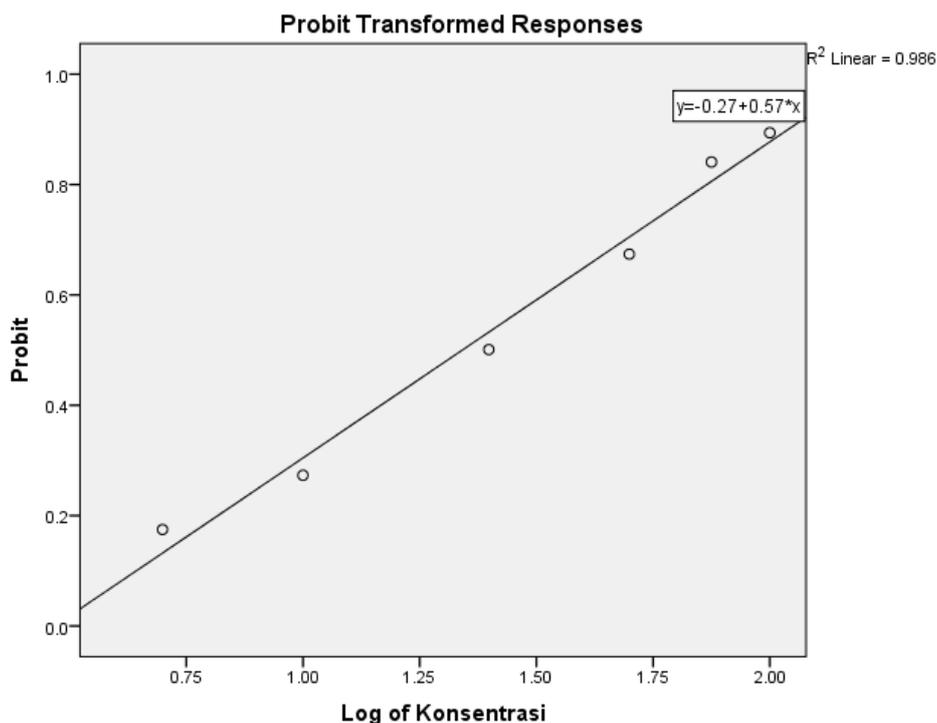
use throughout the body [40]. The viscosity of the formulations ranged from 12,800 to 18,500 cP, with F3 showing a moderate viscosity of  $16,100 \pm 110$  cP. This viscosity level allowed for sufficient structural integrity while maintaining ease of application. F1 exhibited the highest viscosity ( $18,500 \pm 125$  cP), likely due to a higher concentration of hydrophilic surfactants and increased gel matrix density. In contrast, F4 demonstrated the lowest viscosity ( $12,800 \pm 120$  cP), which may have compromised the formulation's physical stability and led to a more runny texture. Optimal viscosity ensures both user satisfaction and controlled release of active ingredients.

Spreadability reflects the ease with which the product can be evenly distributed on the skin. It was inversely related to viscosity across all formulations. F3 displayed a spreadability of  $5.12 \pm 0.14$  cm, which is considered ideal for topical products, enabling smooth application without excessive thinning or dripping. F1, with higher viscosity, had the lowest spreadability ( $4.38 \pm 0.11$  cm), while F4, being the least viscous, had the highest spreadability ( $5.87 \pm 0.17$  cm). These results align with the understanding that formulations with moderate viscosity exhibit balanced spreadability, offering both ease of use and sufficient contact time on the skin surface. Nanoemulgel products should disperse under light pressure for easy application to the skin, defined as 5 - 7 cm [5].

Statistical analysis using one-way ANOVA revealed significant differences ( $p < 0.05$ ) among the formulations in terms of droplet size, polydispersity index (PDI), and zeta potential. Tukey's post-hoc test indicated that F3 was significantly superior, with the smallest droplet size (134.7 nm), lowest PDI (0.183), and most negative zeta potential ( $-32.1$  mV) compared

to the other formulations. Statistical analysis using one-way ANOVA confirmed significant differences ( $p < 0.05$ ) among the formulations for all 3 parameters—droplet size, PDI, and zeta potential. Tukey's post-hoc test indicated that F3 was significantly superior, with the smallest droplet size, lowest PDI, and most negative zeta

potential compared to the other formulations. These findings align with previous studies indicating that nanoemulsions with droplet sizes under 200 nm and PDI values below 0.3 are optimal for cosmetic and pharmaceutical applications due to their improved stability and delivery potential.



**Figure 3** linear correlation of probit response vs. log concentration for ascorbic acid.

The antioxidant potential of the nanoemulgel formulations (F1 - F4) was evaluated using the DPPH radical scavenging assay, and results were expressed as  $IC_{50}$  values (the concentration required to inhibit 50% of DPPH radicals). The results (**Table 4**) showed significant variation among the different formulations. All formulations exhibited moderate antioxidant activity, with  $IC_{50}$  values ranging from 98.4 to 123.7  $\mu\text{g/mL}$ , indicating the presence of active radical-scavenging compounds derived from both star anise essential oil and butterfly pea extract. Among the 4, Formula F3 demonstrated the highest antioxidant activity, with the lowest  $IC_{50}$  value ( $98.53 \pm 0.23$

$\mu\text{g/mL}$ ), while Formula F2 followed with an  $IC_{50}$  of  $148.67 \pm 0.29 \mu\text{g/mL}$ , suggesting moderate antioxidant potential, likely influenced by a slightly lower concentration or less effective interaction between the active compounds. Formula F4 recorded an  $IC_{50}$  of  $123.7 \pm 0.31 \mu\text{g/mL}$ , which was better than F2 but did not surpass F3, possibly due to differences in droplet size or emulsifier distribution affecting bioavailability. Formula F1, with the highest  $IC_{50}$  among the active formulations ( $192.84 \pm 0.24 \mu\text{g/mL}$ ), showed the weakest antioxidant performance, indicating suboptimal formulation ratios or less effective dispersion of the active ingredients.

**Table 4** antioxidant activity of nanoemulgel formulations based on IC<sub>50</sub> values (µg/mL).

Formula	IC <sub>50</sub> (µg/mL)	p-value (< 0.05)
F1	192.84 ± 0.24	
F2	148.67 ± 0.29	
F3	98.53 ± 0.23	
F4	123.7 ± 0.31	0.00
Active ingredients combination	2.47 ± 0.48	
Placebo (negative control)	421.38 ± 0.57	
Ascorbic acid (positive control)	2.59 ± 0.28	

Note: Data are presented as mean ± standard deviation (n = 3).

The placebo formulation (without active extracts) showed negligible antioxidant activity, with an IC<sub>50</sub> value of 421.38 ± 0.57 µg/mL, validating that the observed radical scavenging effects in the active formulations were due to the presence of *C. ternatea* and *I. verum* extracts. In contrast, the positive control (ascorbic acid) displayed potent antioxidant activity (IC<sub>50</sub> = 2.59 ± 0.28 µg/mL), serving as a benchmark for comparison. Notably, a separate evaluation of the pure combination of 1.0% star anise essential oil and 0.25% butterfly pea extract (prior to nanoemulgel incorporation) exhibited a synergistic antioxidant effect, with an IC<sub>50</sub> of 2.47 ± 0.48 µg/mL—closely approximating the performance of ascorbic acid. This result underscores the strong radical-scavenging potential of the 2 plant-derived actives when combined. The enhanced performance of F3 can be attributed to its smaller droplet size and more uniform particle distribution, which likely facilitated better release and surface interaction of antioxidant molecules despite the presence of a gel matrix that can potentially hinder diffusion. It is also important to consider that dilution factors may influence the outcome of DPPH assays. Higher dilution levels may reduce the frequency of interaction between antioxidant molecules and free radicals, potentially increasing IC<sub>50</sub> values. Therefore, using standardized concentration ranges is essential to enable accurate comparisons across formulations.

Although the IC<sub>50</sub> values were higher compared to those reported for the raw essential oil or extract (e.g., IC<sub>50</sub> of star anise oil at 9.88 mg/mL in DPPH solutions) Cosmetic Ingredient Database [10], the incorporation into a nanoemulgel system introduces matrix diffusion barriers that reduce the immediate availability of the

active compounds. However, this also reflects a controlled-release advantage, which is desirable in topical applications where prolonged antioxidant protection is beneficial. These findings are consistent with previous studies that reported a moderate increase in IC<sub>50</sub> when antioxidants are delivered through gel-based nanocarriers, due to restricted molecular mobility and delayed interaction with DPPH radicals [31,40].

The antioxidant efficacy of the nanoemulgel formulations can be attributed to the rich phytochemical content present in *Clitoria ternatea* and *Illicium verum*. *Clitoria ternatea* contains a high concentration of anthocyanins, primarily ternatins, along with flavonoids such as quercetin, kaempferol, and myricetin. These polyphenolic compounds are well-known for their strong radical scavenging activity, primarily through hydrogen donation and metal ion chelation mechanisms [4]. On the other hand, *Illicium verum* essential oil is dominated by trans-anethole, a phenylpropanoid with proven antioxidant and anti-inflammatory properties [7]. It also contains minor components like limonene, linalool, and eugenol, which synergistically enhance the free radical-neutralizing capacity of the oil. In the nanoemulgel system, the encapsulation of both hydrophilic (e.g., flavonoids) and lipophilic (e.g., trans-anethole) compounds within nano-sized droplets improves their dispersion, stability, and skin permeability. While the current study focused on a nanoemulgel based on an oil-in-water (O/W) nanoemulsion, future work may explore dual emulsion systems—such as water-in-oil-in-water (W/O/W)—to further optimize the simultaneous delivery of hydrophilic and lipophilic antioxidants. Such systems have the potential to provide more controlled release

profiles and enhanced skin retention, especially in cosmeceutical applications.

This optimized delivery likely contributes to the observed antioxidant performance across the formulations, particularly in F3 (HLB 12.86), where a balanced ratio of both extracts and favorable droplet characteristics may have maximized bioactive availability [32,39]. The result supports the use of natural flavonoid- and terpenoid-rich extracts in nanocarrier-based topical antioxidants for cosmetic applications. This study did not include particle morphology analyses using Scanning Electron Microscopy (SEM) or Transmission Electron Microscopy (TEM), which could have provided valuable information on the surface characteristics and internal structures of the nanoemulgel system. Additionally, *in vivo* evaluations, skin permeation studies, and long-term storage stability under various environmental conditions were not conducted. These limitations will be addressed in future work to comprehensively assess the performance and applicability of the formulation.

### Conclusions

The physicochemical characteristics of the nanoemulgel were influenced by variations in emulsifier ratios, affecting the HLB values. Formula F3 (Tween 80: Span 80 = 8:2) with HLB value 12.86 showed optimal results with a smallest droplet size (134.7 nm), narrow PDI (0.183), and zeta potential (-32.1 mV), indicating stability and uniform dispersion. It also maintained a skin-compatible pH (5.79), appropriate viscosity (16,100 cP), and good spreadability (5.12 cm), ensuring suitability for topical application. It exhibited strong antioxidant activity with an  $IC_{50}$  value of 98.53  $\mu\text{g/mL}$ . These findings highlight the significance of emulsifier ratio optimization in achieving multifunctional, stable, and effective nanoemulgel systems. Future research should explore particle morphology analyses using Scanning Electron Microscopy (SEM) or Transmission Electron Microscopy (TEM), study *in vivo* efficacy, long-term storage stability, and advanced delivery enhancements to further develop this formulation for commercial cosmeceutical applications.

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### Declaration of Generative AI in Scientific Writing

The authors acknowledge the use of generative AI tools (e.g., QuillBot, Grammarly, and ChatGPT by OpenAI) in the preparation of this manuscript, specifically for language editing and grammar correction. No content generation or data interpretation was performed by AI. The authors take full responsibility for the content and conclusions of this work.

### CRedit Author Statement

**Trisiyana Sholika Sari:** Conceptualization, Data analysis, Experimental design, Investigation, Writing – original draft preparation.

**Tristiana Erawati Munandar:** Methodology, Validation, Data analysis, Writing – review & editing.

**Hsin-I Chang:** Provided methodological input, Visualization, Validation, and Critical review.

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