VOLUME 13 NOMOR 1 JUNE 2026 ISSN 2548 – 611X

JURNAL
BIOTEKNOLOGI & BIOSAINS INDONESIA

Homepage Jurnal: http://ejournal.brin.go.id/JBBI/index

EVALUATION OF CREAM FORMULATION FROM NANOEMULSION *Euphorbia tirucalli* L. WITH STEARIC ACID VARIATIONS

Evaluasi Formulasi Krim dari Nanoemulsi *Euphorbia tirucalli* L. dengan Variasi Asam Stearat

Meylani Nur Intan[1], Aldesra Fitri[1]*, Iqlima Ayu Prestisya[1]
[1]Department of Pharmacy, Faculty of Science and Technology, dr. Soepraoen Kesdam V/ Brawijaya Institute of Technology, Science and Health, Malang, East Java 65147, Indonesia
*Email: aldesra@itsk-soepraoen.ac.id

ABSTRACT
The macerate of *Euphorbia tirucalli L.* contains alkaloids, flavonoids, saponins, steroids, terpenoids, and tannins with antibacterial potential for acne management. This study formulated a nanoemulsion of *E. tirucalli* extract via the high-energy method, which was incorporated into cream formulations with varying stearic acid concentrations (F1;7.5%, F2;9%, F3;12%, F4;14.5%) and physical quality evaluation was carried out. The physical quality test of the cream preparations found that concentrations of stearic acid significantly affected the organoleptic properties (color and skin feels after application), spreadability 4.86-6.55 cm, water washability 8.5-19.1 mL, and viscosity 15557-31433 cPs. Meanwhile, homogeneity, phase separation, and pH of the preparation were not found the significantly different. An increase in stearic acid was directly proportional to washability and viscosity, but inversely proportional to spreadability. The formulations met the physical criteria for an acceptable cream preparation, except for pH, which was excessively alkaline. F2 and F3 establishing them as the most suitable formulations for further development. The antibacterial test against *Propionibacterium acnes* using the well diffusion agar assay, resulted; positive control (Mediklin; clindamycin 0.01%) in strong activity, 5% extract was in the weak activity, 10% and 20% extract was in moderate activity, 30% extract showed strong activity which is comparable to the positive control, and 20% nanoemulsion had no activity. The Spearman's Rho test confirmed a strong correlation between extract concentration and antibacterial potency (r = 0.927).

Keywords: *Antibacterial, Euphorbia tirucalli L., Nanoemulsion, Propionibacterium acnes, Stearic acid*

ABSTRAK
Maserat *Euphorbia tirucalli* L. mengandung alkaloid, flavonoid, saponin, steroid, terpenoid, dan tanin dengan potensi antibakteri untuk manajemen jerawat. Penelitian ini memformulasikan nanoemulsi ekstrak *E. tirucalli* melalui metode energi tinggi, yang dimasukkan ke dalam formulasi krim dengan berbagai konsentrasi asam stearat (F1; 7,5%, F2; 9%, F3; 12%, F4; 14,5%) dan evaluasi kualitas fisik dilakukan. Uji kualitas fisik sediaan krim menemukan bahwa konsentrasi asam stearat secara signifikan mempengaruhi sifat organoleptik (warna dan rasa kulit setelah aplikasi), daya sebar 4,86-6,55 cm, daya cuci air 8,5-19,1 mL, dan viskositas 15557-31433 cPs. Sementara itu, homogenitas, pemisahan fase, dan pH sediaan tidak ditemukan berbeda secara signifikan. Peningkatan asam stearat berbanding lurus dengan daya cuci dan viskositas, tetapi berbanding terbalik dengan daya sebar. Formulasi tersebut memenuhi kriteria fisik untuk sediaan krim yang dapat diterima, kecuali untuk pH, yang terlalu basa. F2 dan F3 menjadikannya sebagai formulasi yang paling sesuai untuk pengembangan lebih lanjut. Uji antibakteri terhadap *Propionibacterium acnes* menggunakan uji agar difusi sumur,

***Received**: 24 November 2025* ***Accepted**: 09 April 2026* ***Published**: 09 June 2026*

menghasilkan; kontrol positif (Mediklin; klindamisin 0,01%) dalam aktivitas kuat, ekstrak 5% dalam aktivitas lemah, ekstrak 10% dan 20% dalam aktivitas sedang, ekstrak 30% menunjukkan aktivitas kuat yang sebanding dengan kontrol positif, dan nanoemulsi 20% tidak memiliki aktivitas. Uji Spearman's Rho mengonfirmasi korelasi kuat antara konsentrasi ekstrak dan potensi antibakteri (r = 0,927).

Kata kunci: *Antibakteri, Euphorbia tirucalli L., Nanoemulsi, Propionibacterium acnes, Asam stearat*

INTRODUCTION

Acne is a chronic inflammatory disorder of the pilosebaceous unit that often leads to scarring, irritation, and psychological distress (Vasam et al. 2023). It results from excessive sebum secretion, follicular obstruction, and bacterial proliferation, triggering inflammation. Environmental factors, particularly pollution, exacerbate acne severity. A retrospective study of 59,530 dermatology patients in Beijing revealed a strong association between exposure to traffic- and industry-related pollutants and acne prevalence. Similarly, a La Roche-Posay study in Beijing found that elevated $PM_{2.5}$, PM_{10}, and NO_2 levels increased sebum production and lesion formation (Krutmann et al. 2017).

Indonesia, one of the world's most biodiverse countries, hosts approximately 31,750 plant species, of which 15,000 possess medicinal potential, yet only about 7,000 are pharmaceutically utilized (Kinho et al. 2011; Retnowati and Susan 2019). *E. tirucalli* (pencil cactus or milk bush), widely distributed in tropical regions (Abd Wahab et al. 2024), contains such as alkaloids, saponins, terpenoids, and tannins. Its methanolic extract exhibits antibacterial and anti-inflammatory properties, showing inhibition zones of 7–22 mm against Gram-positive and 7–13 mm against Gram-negative bacteria (Afifah et al. 2023). Furthermore, a 10% *E. tirucalli* cream demonstrated wound-healing activity in rats, supporting its potential as a natural topical anti-acne agent (Fox et al. 2016).

Nanoemulsions (20–500 nm) offer high stability, superior dermal penetration, and enhanced bioavailability, making them effective drug delivery systems (Singh and Kumar 2015; Geetanjali et al. 2021). Creams serve as ideal topical vehicles due to their favorable texture, comfort, and washability (Kumalasari et al. 2020). Stearic acid, a key emulsifier, is crucial in maintaining cream consistency and stability (Rowe et al. 2009). Therefore, this study aimed to develop and evaluate a nanoemulsion-based *E. tirucalli* cream with varying stearic acid concentrations to optimize its physicochemical characteristics and antibacterial efficacy against *P. acnes*.

MATERIALS AND METHODS

This experimental laboratory study, conducted from at the Pharmacy Laboratory, Institute of Science and Health Technology, dr. Soepraoen Hospital Malang, evaluated the antibacterial activity of *E. tirucalli* extract against *P. acnes*. The conventional extract and nanoemulsion compared to assess their potential as alternative anti-acne therapies addressing bacterial resistance. The study further examined the effect of varying stearic acid concentrations (7.5%, 9%, 12%, and 14.5%) on the physical properties and stability of the nanoemulsion cream. Antibacterial assays were performed using extract concentrations of 5%, 10%, 20%, and 30%. Evaluated parameters included pH, spreadability, viscosity, washability, and inhibition zone diameter. All tests conducted in triplicate to ensure reproducibility. The findings elucidate the relationship between formulation composition, physical stability, and antibacterial efficacy, supporting the development of *E. tirucalli* based nanoemulsion creams for pharmaceutical application.

The equipment used in this study consist of ultra turrax, brookfield viscometers, pH meter, laminar air flow, centrifugator, petri dish, autoclave, desiccator, burette set, incubator, vortexmixer, and UV-Vis spectrophotometry.

The materials needed in this study are *E. tirucalli* plants *tirucalli* from Malang, East Java, Indonesia and authenticated at Materia Medica Batu City, 70% ethanol, isolates of gram-positive bacteria *P.acnes*, magnesium powder, dragendorf reagent, HCl, H_2SO_4, $FeCl_3$, chloroform, physiological NaCl, castor oil, tween 80, aloe mucilago, hydroxypropyl methylcellulose (HPMC), paraffin liquidum, stearic acid, lanolin, triethanolamine (TEA), methylparaben, propylparaben, Mediklin gel 1%, NA (Nutrient Agar), and aquadest.

Preparation of plant extract with maceration method

400 g of dried *E. tirucalli* powder macerated in 2,000 mL of 70% ethanol for 24 hours at room temperature with intermittent stirring. The macerate filtered and concentrated using a rotary evaporator at 60°C to obtain a viscous ethanolic extract.

Phytochemical screening

Alkaloids were identified by reacting the diluted extract (1 mL HCl and 3 mL distilled water) with 2 drops Dragendorff's reagent, producing an orange-red precipitate (Masniawati et al. 2021; Qomaliyah et al. 2023). Flavonoids were confirmed by adding magnesium powder and 10 drops concentrated HCl to the extract, yielding an orange-red coloration (Qomaliyah et al. 2023; Iskandar et al. 2024). Steroids and triterpenoids were detected by treating the diluted extract with anhydrous acetic acid and concentrated H_2SO_4 (2:1 drop), producing a purple to blue-green color. Although structurally related, triterpenoids and steroids are differentiated by distinct colorimetric reactions (Nola et al. 2021). The purple or red then turns blue green indicating the presence of them both. The water layer of distilled extract with chloroform (1:1) were added 0.1 ml HCl and several grains of magnesium metal, resulting in a pink-red color. Tannins were confirmed by adding reacted the distilled extract to 2-3 drops of $FeCl_3$. The positive results are indicated by blackish green or blackish blue color (Laoli 2018).

Nanoemulsion preparation of *E. tirucalli* extract

The primary mixture consisted of 20% concentration of *E. tirucalli* extract (Table 1) dissolved in castor oil and homogenized using an ultra turrax for 30 minutes at 13000 rpm. Then it homogenized again for 5 minutes with 3% tween 80. HPMC 0.2% which has been developed in 17% of aloe vera mucilago was gradually added to the mixture to be ultra turraxed again at the same speed for 25 minutes.

Table 1. Nanoemulsion formulation (Geetanjali et al. 2021)

Composition	Formulation	Function
E. tirucalli extract	20%	Active ingredient
Aloe mucilage	17%	Surfactant
Tween 80	3%	Surfactant
Hydroxypropyl methylcellulose (HPMC)	0,2%	Hydrophilic polymer
Castor oil	Ad 100 g	Solvent

Characterization of nanoemulsion from *E. tirucalli* extract

Clarity was determined using a UV–Vis spectrophotometer by dissolving 1 mL of nanoemulsion in 100 mL of distilled water and measuring transmittance at 650 nm, with distilled water as the blank (Zulfa et al. 2019). Higher transmittance values indicated greater clarity, suggesting droplet sizes within the nanometer range (Lestario et al. 2024). The pH of the formulation was measured using a calibrated digital pH meter.

Preparation cream from nanoemulsion of *E. tirucalli* extract

The oil phase components (liquid paraffin, stearic acid, and lanolin) and the aqueous phase (nipagin, nipasol, TEA, and distilled water) were separately heated to 60–70°C until fully melted, indicated by reduced viscosity and intensified color (Setiorini et al.

2014). The hot aqueous phase was then combined with the hot oil phase and stirred rapidly in a warm mortar until a uniform cream base formed upon cooling. The *E. tirucalli* nanoemulsion was gradually incorporated into the base with gentle stirring until homogeneous (Table 2).

Table 2. Anti-acne cream preparation formula from nanoemulsion of *E. tirucalli* extract (Setiorini et al. 2014)

Composition	F1	F2	F3	F4	Function
Nanoemulsion of *E. tirucalli* extract	5%	5%	5%	5%	Active ingredient
Paraffin liquidum	25%	25%	25%	25%	Base
Stearic acid	7,5%	9%	12%	14,5%	Oil phase emusifier
Lanolin	3%	3%	3%	3%	Emolient
Triethanolamine (TEA)	1,5%	1,5%	1,5%	1,5%	Water phase emulsifier
Methylparaben	0,1%	0,1%	0,1%	0,1%	Preservative
Propylparaben	0,05%	0,05%	0,05%	0,05%	Preservative
Aquadest	Ad 100%	Ad 100%	Ad 100%	Ad 100%	Solvent

Physical quality evaluation test
Organoleptic test

Tests the shape, color, odor, and taste when the cream applied on the skin (Saryanti et al. 2019).

Homogeneity test

A thin layer of cream is placed between two glass slides and pressed, observe the surface, whether it is evenly distributed and smooth, were not lumpy are positives sign (Saryanti et al. 2019).

pH test

One gram of cream was diluted in 10 mL of aquadest, and the pH was measured using a calibrated digital pH meter (Iskandar et al. 2024).

Spreadability test

0.5 g sample of cream was placed on a glass plate, covered with another plate and left for 1 minute to obtain several diameters of the spread formed by adding 50, 100, and 150 g loads then recorded the average (Saryanti et al. 2019).

Phase separation test

Five g of cream placed in a centrifuge tube and spun at 3000 rpm for 30 minutes.

Water washability test

One gram of cream applied to the back of the hand and running water through a burette over it.

Viscosity test

The cream were put at 25 mL beaker glass is tested with rotor 6^{th} mounted at Brookfield viscometer with speed setting at 30 rpm.

Antibacterial test using the agar well diffusion assay

Nutrient agar (Oxoid) was prepared by dissolving 14 g of medium in 500 mL of distilled water, heated to boiling, and sterilized at 121°C for 15 minutes. The sterile medium was poured into petri dishes (20 mL as the base layer and 15 mL as the top layer) and into test tubes for slant cultures (10 mL) (Setiorini et al. 2014). Pure *P. acnes* colonies were inoculated onto slant media and incubated anaerobically in a desiccator at 37°C for 48 h (Septiyana et al. 2022).

Test samples consisted of *E. tirucalli* extract (5%, 10%, 20%, and 30%), its nanoemulsion formulation (20%), and Mediklin 0.01% as the positive control. The bacterial suspension was prepared by transferring colonies into 6 mL of 0.9% physiological NaCl, followed by vortexing for 30s. Turbidity was adjusted to match the McFarland 0.5 standard (absorbance 0.08–0.13) using a UV–Vis spectrophotometer (McFarland 1907).

Seven wells (5 mm diameter) were made in each nutrient agar plate. After inoculation with 8 µL of *P. acnes*, 40 µL of each test sample or control was introduced into the respective wells. Plates incubated anaerobically at 32°C for 24 h, and inhibition zones were measured to evaluate antibacterial activity.

Statistical data analysis

Qualitative data encompassed the antibacterial activity of both conventional and nanoemulsion forms of *E. tirucalli* extract, as well as the physicochemical evaluations of the cream formulations (pH, spreadability, viscosity, and washability), presented in tabular form for clarity. Quantitative analyses were performed using (SPSS) version 25 to evaluate the influence of stearic acid concentration and the antibacterial activity against *P. acnes*. Data normality assessed using the Shapiro–Wilk test. For normally distributed data (Sig. > 0.05), homogeneity of variance was confirmed prior to One-Way ANOVA and Pearson correlation analyses. Non-normally distributed data were analyzed using the Kruskal–Wallis test and Spearman's rho correlation, following the interpretation criteria (Nurhalijah et al. 2024).

RESULTS AND DISCUSSION

Preparation of plant extract with maceration method

The concentrated extract yield in this study was 8.60%, lower than that reported by Adawiyah et al. (2024) (18.10%), despite all using 70% ethanol. The reduced yield was attributed to the shorter maceration period (1:5 days), which limited diffusion efficiency. The maceration was selected for its simplicity, low cost, and ability to preserve heat-sensitive metabolites by avoiding thermal degradation. The obtained extract displayed a caramel-like, brownish-black appearance and mild sweet odor, differing from the dark green viscous extract. The higher water bath temperature during solvent evaporation, leading to partial degradation and moisture loss (Sukma et al. 2017; Yuliantari et al. 2017).

Phytochemical screening

The phytochemical screening confirmed six major secondary metabolite classes (Table 3) exhibited antibacterial activity. Flavonoids disrupt bacterial membranes and denature proteins, impairing respiration (Arifin et al. 2025). Alkaloids inhibit peptidoglycan synthesis, weakening cell walls, while steroids increase membrane permeability, leading to lysis (Sadiah et al. 2022). Triterpenoids destabilize porin proteins, saponins promote cytoplasmic leakage, and tannins form enzyme–protein complexes that reduce membrane permeability (Rini et al. 2017; Arifin et al. 2025). This differs from Adawiyah et al. (2024), who identified only flavonoids, saponins, and tannins despite using 70% of ethanol. Beyond maceration duration, such variation reflect the geographical differences, as earlier samples were collected in Surabaya, while the present study used specimens from Malang Regency. Environmental factors such as soil composition, fertilizer use, temperature, and humidity are known to influence secondary metabolite biosynthesis. The results indicating that the shorter extraction did not diminish phytochemical diversity and more complex than that (Adawiyah et al. 2024). Similarly, prolonged maceration increased yield but reduced flavonoid content due to oxidative degradation of thermolabile compounds.

Collectively, these findings indicate a multifactorial antibacterial mechanism, suggesting that *E. tirucalli* extract possesses broad-spectrum activity through combined membrane and metabolic disruption.

Table 3. Phytochemical content of *E. tirucalli* extract

Phytochemical tests					
Alkaloids	Flavonoids	Steroids	Terpenoids	Saponins	Tannins
++	+++	++	+	++	+++

Indicator:
+ = The presence was detected, but in a small amount.
++ = The presence was detected in a sufficient amount.
+++ = The presence was detected in a large amount.

Preparation and characterization of nanoemulsion from *E. tirucalli* extract

The nanoemulsion formulation by Geetanjali et al. (2021) required modification due to phase separation between oil and aqueous phases. Stability was improved by increasing tween 80 concentration and incorporating HPMC as a hydrophilic polymeric thickener. Ultrasonic-assisted extraction (UAE) was applied as an efficient technique to enhance mass transfer and reduce extraction time, as cavitation disrupts cell walls, increases permeability, and promotes compound release. This method improves polyphenol yield by 6–35% compared with maceration (Febriyanto et al. 2021). During transmittance testing, prolonged sonication produced a lighter extract and clearer solution (Figure 1), confirming nanoemulsion formation with 90–100% transmittance, comparable to distilled water (Widyastuti and Saryanti 2023).

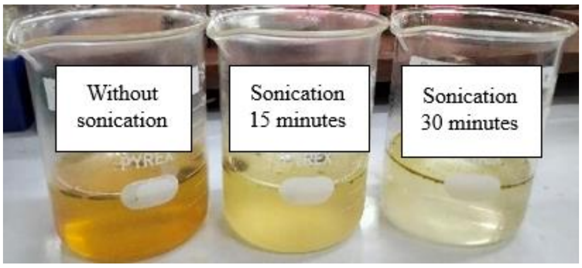

Figure 1. Preparation for transmittance testing

Physical evaluation tests of cream from nanoemulsion of *E. tirucalli* extract with the statistic analyst

Organoleptic evaluation of formulations F1–F4 showed distinct differences in texture, color, and spreadability corresponding to increasing stearic acid concentrations (Table 4). In the *E. tirucalli* nanoemulsion cream, higher stearic acid levels gradually lightened the characteristic brown color to yellowish tones and produced a sweet, fatty caramel-like odor. The active nanoemulsion was reformulated by reducing extract concentration from 15% to 5%, as excessive oil content caused physical instability and phase separation.

Table 4. Physical quality evaluation test of cream preparation results

	Physical Quality Evaluation			
Preparation cream	pH	Speadability (cm)	Water washability (mL)	Viscosity (cPs)
F1	7,98 ± 0,031	6,55 ± 0,21	8,5 ± 1,5	15556,667 ± 310,86
F2	8,25 ± 0,28	6,18 ± 0,39	11 ± 2	23366,667 ± 1366,47
F3	8,41 ± 0,02	5,57 ± 0,26	13,7 ± 2,51	27353,333 ± 365,56
F4	8,38 ± 0,012	4,86 ± 0,23	19,1 ± 2,15	31433,333 ± 208,17

*F1 = 7.5% stearic acid concentration, F2 = 9% stearic acid concentration,
F3 = 12% stearic acid concentration, F4 = 14.5% stearic acid concentration.

Homogeneity testing confirmed all formulations had smooth, uniform textures without coarse particles or agglomerates. Such irregularities, though minor, may impair active compound distribution and reduce formulation efficacy (Swastini et al. 2015). Macroscopic analysis further revealed that higher stearic acid concentrations yielded more widely dispersed oil globules across the slide surface.

pH analysis showed that all formulations exhibited slightly alkaline values, posing minor risks of dryness and desquamation. The nanoemulsion alone had a pH of 6.72, and its low concentration (5% in 50 g) minimally influenced the overall pH. The Kruskal–Wallis test indicated no significant difference among formulations, suggesting stearic acid variation had limited impact. Similar findings were reported by Saepudin et al. (2024) in kesum leaf anti-acne creams ($p = 1.00 > 0.05$). Although higher stearic acid levels slightly increased pH, the effect was moderated by TEA (pH 10.5). Previous studies using 17% stearic acid and 2% TEA achieved pH 6, while this study (7.5–14.5% stearic acid, 1.5% TEA) yielded an average pH ≈ 8, still acceptable for topical use (Saryanti et al. 2019).

Spreadability tests showed that higher applied weight increased spread diameter, whereas higher stearic acid levels reduced spreadability and produced stiffer textures. Statistical analysis confirmed significant differences (Table 5), indicating an inverse correlation between stearic acid content and spreadability. These results align with previous research, who observed increased viscosity and reduced spreadability in papaya leaf cream with higher stearic acid levels.

Table 5. Statistical analysis

	Statistic Tests				
Analysis data	pH	Speadability (cm)	Water washibility (mL)	Viscosity (cPs)	Antibacterial activity (cm)
Differences	Sig. 0,147 > 0,05	Sig. 0,000 < 0,05	Sig. 0,001 < 0,05	Sig. 0,016 < 0,05	Sig. 0,012 < 0,05
Correlation	Sig. 0,147 > 0,05 n = 0,462	Sig. 0,000 < 0,05 n = -0,934	Sig. 0,000 < 0,05 n = 0,9	Sig. 0,000 < 0,05 n = 0.972	Sig. 0,000 < 0,05 n = 0.972

Centrifugation-based phase separation tests revealed that F1 showed the highest oil separation, followed by F3 (≈50% less), while F4 and F2 exhibited only slight turbidity and minimal foaming. This contrasts with reports suggesting that triethanolamine–stearic acid emulsions typically form stable o/w systems (Saryanti et al. 2019). The instability may result from CO_2 incorporation during preparation, which weakens emulsifying capacity by disrupting polar headgroup interactions and hydrogen bonding (Xu et al. 2015).

Washability tests indicated that higher stearic acid concentrations required more water for removal, reflecting increased oil content. Statistical analysis confirmed a significant positive correlation between stearic acid level and water volume used. Thus, higher concentrations reduced washability, consistent with (Husni et al. 2019), who found that w/o creams require more water for rinsing than o/w types. Reduced washability likely stems from higher interfacial tension, as nonpolar stearic acid resists interaction with polar water, necessitating greater rinsing effort.

Viscosity measurements showed a direct proportional relationship with stearic acid concentration, with F4 exhibiting the highest value. Statistical analysis confirmed significant differences, consistent with (Thomas et al. 2024), who reported that increased solid content in seaweed-based creams elevated viscosity proportionally to total formulation volume.

Variations in stearic acid concentration markedly influenced the physical characteristics and stability of the *E. tirucalli* nanoemulsion cream. Higher stearic acid levels increased viscosity and firmness but decreased spreadability and washability, potentially reducing user comfort. All formulations exhibited acceptable pH values and uniform appearances, indicating good emulsification and stability. Excessive stearic acid produced rigid, less washable textures, whereas lower concentrations reduced structural stability. The formulation with a moderate stearic acid level showed the most balanced viscosity, spreadability, and consistency, making it optimal for topical application. These findings underscore the importance of optimizing emulsifier concentration to achieve desirable rheological behavior, user acceptability, and formulation stability in nanoemulsion-based creams.

Antibacterial test using the agar well diffusion assay with the statistical analysis

The antibacterial assay showed that Mediklin® (Clindamycin 0.01%) produced a strong inhibition zone, while the 5% extract exhibited weak activity and the 10–20% extracts demonstrated moderate effects (Table 6). The 30% extract showed the strongest inhibition, comparable to the reference drug. The Kruskal–Wallis test indicated significant differences among treatments ($p < 0.05$), and Spearman's Rho ($r = 0.927$) revealed a strong positive correlation between concentration and antibacterial activity. Clindamycin acts by binding to the 50S ribosomal subunit at the peptidyl transferase center and nascent peptide exit tunnel, blocking peptide bond formation (Armillei et al. 2024). The antibacterial effect of *E. tirucalli* is attributed to synergistic interactions of flavonoids, alkaloids, steroids, triterpenoids, saponins, and tannins acting on multiple bacterial targets.

Table 6. Antibacterial activity against *P. acnes*

Antibacterial Activity Against *P. acnes* (mm)						
Extract 5%	Extract 10%	Extract 20%	Extract 30%	Aquadest	Mediklin 0,01%	Nanoemulsion 20%
2,13 $\pm$ 0,56	6,52 $\pm$ 0,69	9,67 $\pm$ 0,37	14,72 $\pm$ 5,02	0	13,7 $\pm$ 3,49	0

*Data are means of three replicates (n = 3)

In contrast, the nanoemulsion showed no inhibition zone, similar to the negative control. Although the Kruskal–Wallis test showed significance, the correlation ($r = –0.111$; $p < 0.05$) was weak. Reduced activity likely resulted from castor oil's low antibacterial potential, thermal degradation of bioactives during homogenization (>50°C), lack of co-surfactant, and excess tween 80 (20%) limiting compound availability. The lipophilic oil phase may also hinder diffusion through the agar medium. Further optimization of carrier system, temperature control, and active fraction enrichment is recommended to enhance antibacterial efficacy.

CONCLUSION

Variations in stearic acid concentration in the anti-acne cream formulated from the 70% ethanolic nanoemulsion of *E. tirucalli* extract showed distinct effects; organoleptic revealed differences in color and comfort, while homogeneity and pH showed no significant variation. However, increasing stearic acid markedly enhanced viscosity while reducing spreadability and washability. Formulations F2 and F3 demonstrated the most favorable balance for further development.

The 70% ethanolic extract of *E. tirucalli* exhibited concentration-dependent antibacterial activity against *P. acnes*, with the 30% extract showing inhibition comparable to Mediklin® (clindamycin 0.01%). Spearman's Rho analysis indicated a strong positive correlation between extract concentration and antibacterial potency, confirming that higher phytochemical content enhances antimicrobial efficacy.

Further optimization of surfactant and co-surfactant ratios, coupled with low-energy emulsification and cold extraction methods, is recommended to preserve formulation stability and bioactivity. Despite limitations in laboratory access and experimental replication, this study provides a foundational basis for subsequent formulation optimization and pharmacological evaluation.

ACKNOWLEDGEMENT

The author would like to thank to dr. Soepraoen Kesdam V/ Brawijaya Institute of Technology, Science and Health Malang, for their facilities and support.

REFERENCES

Abd Wahab NZ, Malza NMNM, Rukayadi Y (2024) Phytochemical screening and evaluation of antibacterial, anticandidal, and sporicidal properties of *Euphorbia tirucalli* extract in Terengganu, Malaysia. HAYATI J Biosci 31:903–915. doi: 10.4308/hjb.31.5.903-915

Adawiyah R, Mukti AW, Sanulingga AS (2024) Perbandingan aktivitas ekstrak ranting patah tulan (*Euporbia tirucalli*) dan daun yodium (*Jatropha muitifida*) terhadap *Staphylococcus Aureus*. J Kesehat Tambusai 5:10071–10077

Afifah N, Riyanta AB, Amananti W (2023) Pengaruh waktu maserasi terhadap hasil skrining fitokimia pada ekstrak daun mangga harum manis (*Mangifera Indica* L.). J Cryst Publ Penelit Kim Ter 5:54–61. doi: 10.36526/jc.v5i1.2634

Arifin SHAG, Firdhausi NF, Hidayati I, Faizah H (2025) Phytochemical screening and antimicrobial activity of combination of *Piper betle* and *Moringa oleifera* extracts. Indones J Pharm Sci Technol 12:184–191. doi: 10.24198/ijpst.v12i2.45288

Armillei MK, Lomakin IB, Del Rosso JQ, Grada A, Bunick CG (2024) Scientific rationale and clinical basis for clindamycin use in the treatment of dermatologic disease. Antibiotics 13:270. doi: 10.3390/antibiotics13030270

Febriyanto F, Hanifa NI, Muliasari H (2021) Penetapan kadar fenolik total ekstrak kulit buah kopi robusta (*Coffea canephora* L.) di pulau Lombok. Lumbung Farm J Ilmu Kefarmasian 2:89–95

Fox L, Csongradi C, Aucamp M, Du Plessis J, Gerber M (2016) Treatment modalities for acne. Molecules 21:1063. doi: 10.3390/molecules21081063

Geetanjali R, Sreejit V, Sandip P, Preetha R (2021) Preparation of *Aloe vera* mucilage- ethyl vanillin nano-emulsion and its characterization. Mater Today Proc 43:3766–3773. doi: 10.1016/j.matpr.2020.10.990

Husni P, Pratiwi AN, Baitariza A (2019) Formulasi krim ekstrak etanol daun kelor (*Moringa oleifera* Lamk). J Ilm Farm Farmasyifa 2:101–110. doi: 10.29313/jiff.v2i2.4796

Iskandar D, Putri DAM, Hidayani R (2024) Uji kualitatif senyawa metabolit sekunder ekstrak daun malapari (*Pongamia pinnata* L. Pierre) pada pelarut etanol dan n-heksana sebagai kandidat sunscreen. BADAA J Ilm Pendidik Dasar 6:107–114. doi: 10.37216/badaa.v6i1.1400

Kinho J, Arini DID, Tabba S, Kama H, Kafiar Y, Shabri S, Karundeng MC (2011) Tumbuhan obat tradisional di Sulawasi Utara Jilid I. Balai Penelitian Kehutanan Manado, Manado

Krutmann J, Moyal D, Liu W, Kandahari S, Lee G-S, Nopadon N, Xiang LF, Seité S (2017) Pollution and acne: is there a link? Clin Cosmet Investig Dermatol 10:199–204. doi: 10.2147/CCID.S131323

Kumalasari E, Mardiah A, Sari AK (2020) Formulasi sediaan krim ekstrak daun bawang Dayak (*Eleutherine palmifolia* (L) Merr) dengan basis krim tipe A/M dan Basis Krim tipe M/A. J Farm Indones AFAMEDIS 1:23–33. doi: 10.61609/afamedis.v1i1.2

Laoli NS (2018) Uji aktivitas antibakteri ekstrak etanol daun bandotan (*Ageratum conyzoides* L.) terhadap bakteri *Bacillus substilis* dan *Proteus vulgaris*. Bachelor’s thesis, Universitas Sumatera Utara

Lestario JR, Sari IP, Andareza A, Fadillah S, Rachmaniar R (2024) Karakteristik nanoemulsi isolat brazilin dari tanaman kayu secang (*Caesalpinia sappan* L.) asli Indonesia. Maj Farmasetika 9:205–215

Masniawati A, Johannes E, Winarti W (2021) Analisis fitokimia umbi talas Jepang *Colocasia esculentai* L.(Schott) var. antiquorum dan talas kimpul *Xanthosoma sagittifolium* L.(Schott) dari dataran rendah. J Ilmu Alam Lingkung 12:7–14

McFarland J (1907) The nephelometer:an Instrument for estimating the number of bacteria in suspensions used for calculating the opsonic index and for vaccines. J Am Med Assoc XLIX:1176–1178. doi: 10.1001/jama.1907.25320140022001f

Nola F, Putri GK, Malik LH, Andriani N (2021) Isolasi senyawa metabolit sekunder steroid dan terpenoid dari 5 tanaman. Syntax Idea 3:1612–1619

Nurhalijah SD, Nina C, Romadhona A, Maulani N, Rahayu MS (2024) Analisis korelasi Spearman untuk mengetahui hubungan antara penggunaan media sosial dan tingkat produktivitas akademis mahasiswa agribisnis (Studi kasus : Universitas Sultan Ageng Tirtayasa). J Ilm Wahana Pendidik 10:800–809. doi: 10.5281/zenodo.13766003

Qomaliyah EN, Indriani N, Rohma A, Islamiyati R (2023) Skrining fitokimia, kadar total flavonoid dan antioksidan daun cocor bebek. Curr Biochem 10:1–10

Retnowati A, Susan D (2019) Kekayaan jenis jamur. In: Retnowati A, Rugayah, Rahajoe JS, Arifiani D (eds) Status keanekaragaman hayati Indonesia: Kekayaan jenis tumbuhan dan jamur Indonesia. LIPI, Jakarta

Rini AA, Supriatno S, Rahmatan H (2017) Phytochemical screening and antibacterial test of ethanolic extract of kawista (*Limonia acidissima* L.) from Aceh Besar against *Escherichia coli*. J Ilm Mhs Pendidik Biol 2

Rowe RC, Sheskey PJ, Weller PJ (2009) Handbook of pharmaceutical excipients. Pharmaceutical Press, London

Sadiah HH, Cahyadi AI, Windria S (2022) Kajian daun sirih hijau (*Piper betle* L) sebagai antibakteri. J Sain Vet 40:128–138. doi: 10.22146/jsv.58745

Saepudin S, Hartono K, Wasih EA (2024) Formulasi krim anti-jerawat ekstrak daun kesum (*Polygonum minus* Huds) dengan variasi konsentrasi asam stearat. Farmasains J Ilm Ilmu Kefarmasian 11:66–75. doi: 10.22236/farmasains.v11i2.14707

Saryanti D, Setiawan I, Safitri RA (2019) Optimasi asam stearat dan TEA pada formula sediaan krim ekstrak kulit pisang kepok (*Musa paradisiaca* L.). J Ris Kefarmasian Indones 1:225–237

Septiyana E, Dewi ERS, Sumarno (2022) Uji daya hambat ekstrak kulit buah ketapang muda (*Terminalia catappa* L.) terhadap pertumbuhan bakteri *Propionibacterium acnes*. SNSE VIII 1:80–90

Setiorini MS, Soegihardjo C. J, Atmodjo K (2014) Potensi antimikrobia krim ekstrak ranting patah tulang (*Euphorbia tirucalli* Linn.) terhadap *Propionibacterium acnes* ATCC 11827 dan *Candida albicans* ATCC 24433. J Farm Sains Komunitas 11:64–71

Singh D, Kumar A (2015) Effects of nano silver oxide and silver ions on growth of *Vigna radiata*. Bull Environ Contam Toxicol 95:379–384. doi: 10.1007/s00128-015-1595-4

Sukma IWA, Harsojuwono BA, Arnata IW (2017) Pengaruh suhu dan lama pemanasan ekstraksi terhadap rendemen dan mutu alginat dari rumput laut hijau *Sargassum* sp. J Rekayasa Manaj Agroindustri 5:71–80

Swastini DA, Yanti NLGT, Udayana NK, Arisanti CIS, Wirasuta IMAG (2015) Uji sifat fisik cold cream kombinasi ekstrak kulit buah manggis (*Garcinia mangostana* L.), daun binahong (*Anredera cordifolia*), herba pegagan (*Centella asiatica*) sebagai antiluka bakar. J Farm Udayana 4:279721

Thomas NA, Suryadi AMA, Latif MS, Hutuba AH, Susanti S (2024) Formulasi dan uji stabilitas fisik krim pelembab ekstrak rumput laut (*Eucheuma cottonii*). Indones J Pharm Educ 4:1–9. doi: 10.37311/ijpe.v4i1.20522

Vasam M, Korutla S, Bohara RA (2023) *Acne vulgaris*: A review of the pathophysiology, treatment, and recent nanotechnology based advances. Biochem Biophys Rep 36:101578. doi: 10.1016/j.bbrep.2023.101578

Widyastuti AI, Saryanti D (2023) Formulasi dan evaluasi sediaan nanoemulsi ekstrak umbi bawang putih (*Allium sativum* L.): Formulation and evaluation of nanoemulsion of garlic (*Allium sativum* L.) extract. J Sains Kesehat 5:178–185

Xu W, Gu H, Zhu X, Zhong Y, Jiang L, Xu M, Song A, Hao J (2015) CO_2-controllable foaming and emulsification properties of the stearic acid soap systems. Langmuir 31:5758–5766. doi: 10.1021/acs.langmuir.5b01295

Yuliantari NWA, Widarta IWR, Permana IDGM (2017) Pengaruh suhu dan waktu ekstraksi terhadap kandungan flavonoid dan aktivitas antioksidan daun sirsak (*Annona muricata* L.) menggunakan ultrasonik. Media Ilm Teknol Pangan Sci J Food Technol 4:35–42

Zulfa E, Novianto D, Setiawan D (2019) Formulasi nanoemulsi natrium diklofenak dengan variasi kombinasi Tween 80 dan Span 80: Kajian karakteristik fisik sediaan. Media Farm Indones 14:1471–1477