The 7th Proceeding Strada International Conference on Health
Kediri – East Java, Indonesia, September 24-25, 2025
https://proceeding.thesich.org | Page 61-66

Antibacterial Activity of Facial Wash Formulated with Pegagang (*Centella asiatica*) and Sweet Orange (*Citrus sinensis*) Extracts against Staphylococcus aureus and Propionibacterium Acnes

Devita Riafinola Andararit
Department of Pharmacy, Universitas Strada Indonesia
**Corresponding author: devita.riafinola@gmail.com*

ABSTRACT

Acne is a common skin disorder caused by *Staphylococcus aureus* and *Propionibacterium acnes*, and is influenced by hormonal, sebum, and lifestyle factors. One of the preventive measures can be the use of facial washes made from natural extracts. This study aimed to determine the most effective concentration of a facial wash combining Pegagang (*Centella asiatica*) and Sweet Orange (*Citrus sinensis*) extracts in inhibiting the growth of acne-causing bacteria. The disc diffusion method was used in this studyThe facial wash formulations were prepared in three variations of Pegagang extract concentrations (6%, 12%, and 15%) with a fixed Sweet Orange extract concentration of 10%. The F3 formula showed the highest antibacterial activity, with an average inhibition diameter of 22.74 mm, and was not significantly different from the positive control. These findings suggest that Formula F3 is the most effective and has strong potential to be developed as a natural-based facial wash for acne prevention**.**

Keywords: acne, antibacterial, centella asiatica, citrus sinensis, facial wash

BACKGROUND

Acne (Acne vulgaris) is a common skin condition among adolescents and adults. This condition often causes discomfort and even lowers self-confidence (Aryani & Riyaningrum, 2022). The main cause of acne is bacterial infection, particularly *Staphylococcus aureus*, which triggers inflammation of the skin follicles (Husnani et al., 2020) and *Propionibacterium acnes*, a Gram-positive bacterium that lives in sebaceous follicles and plays a role in exacerbating skin inflammation.

Conventional acne treatments generally use topical antibiotics such as clindamycin and erythromycin, or chemical agents like benzoyl peroxide. However, these therapies have drawbacks, including skin irritation, dryness, and the risk of bacterial resistance with long-term use (Sasebohe et al., 2023). This has prompted research into safer, more affordable, natural-based alternatives. Indonesia, as a megabiodiverse country, has many medicinal plants with antibacterial potential. Sweet orange (*Citrus sinensis*) contains alkaloids, saponins, and tannins, which have both soothing and antibacterial properties (Endarini et al., 2022). Meanwhile, Pegagang extract (*Centella asiatica*) is rich in bioactive compounds such as flavonoids, triterpenoids, tannins, and saponins, which have been shown to have antibacterial activity (Utari, 2023).

One form of pharmaceutical preparation that is relevant in acne prevention efforts is *facial wash* , because it not only functions as a facial cleanser, but can also be formulated with natural active ingredients that have the potential to inhibit the growth of acne-causing bacteria. The use of medicinal plant extracts in facial wash is safer, more practical, and economical (Herawati, 2020). Based on this potential, this study focused on the formulation of a facial wash combining pegagang *(Centella asiatica*) and sweet orange (*Citrus sinensis)* extracts, with its antibacterial activity tested against *Staphylococcus aureus* and *Propionibacterium acnes* as the main bacteria that cause acne.

METHODS

This study is an experimental laboratory research. Pegagan (*Centella asiatica*) leaves and Sweet Orange (*Citrus sinensis*) fruits were extracted using the maceration method with 70% ethanol as the solvent for 3 × 24 hours. The maceration filtrate was then filtered and concentrated using a rotary evaporator to obtain a thick extract. Subsequently, phytochemical screening was conducted to identify the active compounds present in each extract.

Tools

The tools used in this study included a maceration vessel, analytical balance, rotary evaporator, glass beakers, measuring cups, test tubes, stirring rods, knives, blender, porcelain cups, separating funnels, pipettes, filter paper, mortar, pH meter, stamfer, water bath, weighing bottles, glass slides, oven, Brookfield viscometer, Bunsen burner, loop needle, and autoclave.

Material

The materials used in this study included a combination of Pegagang (*Centella asiatica*) extract at concentrations of 6%, 12%, and 15% and Sweet Orange (*Citrus sinensis*) extract at a fixed concentration of 10%, distilled water, 70% ethanol, Sodium Lauryl Sulfate (SLS), Glycerin, Triethanolamine, Methyl Paraben, NaCl, Adeps Lane, Citric Acid, and Stearic Acid.

Table 1. Facial Wash Formulation (Senja et al., 2024)
Antibacterial Test

No.Name Material	FI	F2	F3	K (-)	K(+)	Function
1.Extract Pegagang Leaf	6%	12%	15%	-	Acne	Active substance
2.Extract orange sweet	10%	10%	10%	-		Active substance
3.Stearic acid	0.75%	0.75%	0.75%	0.75%		Stabilizer
4.Adeps lana	0.5%	0.5%	0.5%	0.5%		Saponification
5.Glycerin	1 %	1 %	1 %	1 %		Humectant
6.Methyl paraben	0.2%	0.2%	0.2%	0.2%		Preservative
7.Citric acid	0.1%	0.1%	0.1%	0.1%		pH regulator
8.NaCl	1.67%	1.67%	1.67%	1.67%		Thickening agent
9.(SLS)	0.1%	0.1%	0.1%	0.1%		Surfactant
10.Triethanolamine	0.15%	0.15%	0.15%	0.15%		Emulsifier
11.Aquadest	Ad 100 ml	Ad 100 ml	Ad 100 ml	Ad 100 ml		Solvent

Antibacterial activity was determined based on the diameter of the inhibition zone formed, then the results were analyzed using the One-Way ANOVA statistical test, followed by the Tukey HSD *post-hoc test* to determine significant differences between formulas.

RESULTS

Moisture content (%)

This section presents the results of the water content analysis of sweet oranges and mandarins. The test results are presented in Table 2.

Table 2. Moisture Content of Pegagang Leaves and Sweet Orange

Powder	Weight (kg)	Level Water (%)
Pegagang leaves	2	6.83
Sweet Orange	2	5.52

Centella asiatica and *Citrus sinensis* were dried and powdered using a blender. The powders were then sieved through a No. 60 mesh to obtain 2 kg of fine powder. Subsequently, the moisture content was determined using a moisture balance, resulting in 6.83% for *Centella asiatica* and 5.52% for *Citrus sinensis*, which met the moisture content requirement of less than 10%.

Ethanol Residue Test Results

The ethanol residue test was conducted to ensure that no residual ethanol remained in the concentrated extracts obtained (Sandy et al., 2021). The test results are presented in Table 3.

Table 3. Free Ethanol Pegagang Leaves and Orange Sweet

Procedure	Observation results	Conclusion
Extract Pegagang leaves added with concentrated H2SO4 and 1% CH3COOH	No characteristic ester odor detected	Negative (-) / Free Ethanol
Extract Sweet Orange plus concentrated H2SO4 and 1% CH3COOH	No characteristic ester odor detected	Negative (-) / Free Ethanol

Phytochemical Screening of Pegagang (*Centella asiatica*) Leaf Extract Tables 4 and 5 present the results of the phytochemical screening of *Centella asiatica* and *Citrus sinensis* extracts using the test tube method.

Table 4. Phytochemical Screening of *Centella asiatica* Extract by the Test Tube Method

Sample	Compound	Color Which Formed	Results
Extract *Centella asiatica*	Flavonoid	Brownish yellow	+
	Saponin	Foam stable	+
	~~Tannin~~	~~Blackish green~~	+
	Alkaloid	No There is sediment yellow	-

Table 5. Phytochemical Screening of *Citrus sinensis* Extract by the Test Tube Method

Sample	**Compound**	**Color Which Formed**	**Results**
Extract *Centella asiatica*	Flavonoid	Brownish yellow	+
	Saponin	Foam stable	+
	~~Tannin~~	~~Blackish green~~	+
	Alkaloid	No There is sediment yellow	-

Phytochemical screening of *Centella asiatica* extract showed positive results for flavonoids, indicated by the formation of a yellowish-brown color, positive for saponins, indicated by the formation of stable foam, and positive for tannins, indicated by the formation of a greenish-black color. Meanwhile, the alkaloid test was negative, as no yellow precipitate was formed. The *Citrus sinensis* extract showed consistent results, being positive for flavonoids, saponins, and tannins, and negative for alkaloids. These secondary metabolites contribute to the potential antibacterial activity, with flavonoids and tannins known to disrupt bacterial cell walls, while saponins can increase the permeability of microbial cell membranes.

Results Evaluation Preparation *Facial Wash*

Test Organoleptic

Organoleptic testing was conducted visually to observe the shape, odor, and color of the three formulations. The observation results are presented in Table 6

Table 6. Organoleptic Test of Facial Wash Formulations

Formula	**Form**	**Color**	**Aroma**
F1	Fluid thick	Dark brown	Typical extract
F2	Fluid thick	Dark brown	Typical extract
F3	Fluid thick	Dark brown	Typical extract

Test Homogeneity

The homogeneity test was conducted to ensure that all components in the formulation were evenly mixed. The observation results are presented in Table 7.

Table 7. Homogeneity Test of Facial Wash Formulations

Formula	Replication 1	Replication 2	Replication 3	Information
F1	Homogeneous	Homogeneous	Homogeneous	Qualify
F2	Homogeneous	Homogeneous	Homogeneous	Qualify
F3	Homogeneous	Homogeneous	Homogeneous	Qualify

Based on Table 7, the observations showed that all three facial wash formulations (F1, F2, and F3) had a homogeneous composition. This indicates that all components were well dispersed and that the formulations met the homogeneity requirements.

Antibacterial Activity Test Results

The antibacterial activity test was conducted to determine the ability of the facial wash formulations to inhibit the growth of *Staphylococcus aureus* and *Propionibacterium acnes*. The disc diffusion method (Kirby-Bauer) was used, where the effectiveness of the formulations was measured based on the diameter of the clear zone (inhibition zone) formed around the paper discs. The inhibition zone diameters from three replicates are presented in Table 8.

Table 8. Activities Zone Resistor Bacteria

Treatment	**F1**	**F2**	**F3**	**K+**	**K-**
Replication 1	15.45	20.46	22.23	14.79	0

Replication 2	15.25	20.27	22.68	15.78	0
Replication 3	16.5	21.49	23.32	14.08	0
Average ± SD	15.73 ± 1.42	20.74 ± 3.02	22.74 ± 4.30	14.88 ± 0.85	0.00 ± 0.00
Category	Strong	Strong	Strong	Strong	No There is

Information (CLSI, 2023)

Strength	:	Diameter 10 - 19 mm
Medium	:	Diameter 5 - 9 mm
Weak	:	Diameter < 5 mm
No There are	:	Diameter 0 mm

Based on the results in Table 8, all facial wash formulations exhibited inhibitory activity against the growth of *Staphylococcus aureus* and *Propionibacterium acnes*. A dose-response relationship was observed, where an increase in Sweet Orange extract concentration corresponded with an increase in the inhibition zone diameter. Formula F3, containing 15% Pegagang leaf extract and 10% Sweet Orange extract, showed the strongest inhibitory activity among the tested formulations.

The One-Way ANOVA statistical test indicated a significant difference ($p < 0.05$) in antibacterial activity among the treatment groups. Further analysis using the Tukey HSD test confirmed that F3 had the most effective inhibitory effect. The most notable finding was that there was no statistical difference ($p > 0.05$) between the activity of F3 (22.74 mm) and the positive control (14.88 mm). This indicates that the antibacterial potential of the herbal facial wash at 15% concentration is comparable to that of a commercial antiseptic product.

DISCUSSION

This study focused on the formulation and evaluation of a facial wash containing Pegagang (*Centella asiatica*) leaves and Sweet Orange (*Citrus sinensis*) extracts against *Staphylococcus aureus* and *Propionibacterium acnes*. Analyses were conducted to ensure the physical quality of the formulations and their antibacterial effectiveness, providing a basis for the development of natural-based skincare products for acne..

Moisture content analysis showed that *Centella asiatica* had 6.83% and Sweet Orange 5.52%. These values meet the Indonesian Herbal Pharmacopoeia requirements (<10%), indicating that both herbs comply with quality standards. Low moisture content is important to prevent microbial and fungal growth and to maintain the stability of bioactive compounds (Ansel, 2017).

The ethanol-free test showed negative results for both extracts, indicating that the concentration process successfully removed residual solvent. This step is important because residual ethanol can compromise the stability of the preparation, cause skin irritation, and reduce product safety (Rowe et al., 2012). Based on these results, the pegagan and sweet orange extracts are confirmed to meet safety criteria and are therefore suitable for formulation into *facial wash preparations.*

Phytochemical screening showed that the extracts of pegagang and sweet orange contain flavonoids, saponins, and tannins, but are negative for alkaloids. These secondary metabolite compounds are known to play an important role in antibacterial activity. Flavonoids can disrupt the integrity of bacterial cell membranes, tannins work by precipitating proteins thereby inactivating enzymes, while saponins act as natural surfactants that lyse cell membranes (Cowan, 2018). The content of these secondary metabolites plays a synergistic role in supporting the antibacterial activity of the combination of pegagang *(Centella asiatica*) and sweet orange (*Citrus sinensis)* extracts.

The results of the physical quality evaluation showed that all *facial wash formulas containing* pegagang *(Centella asiatica*) and sweet orange (*Citrus sinensis)* extracts met the organoleptic and homogeneity requirements. Antibacterial tests showed that all formulas were able to inhibit the growth of *Staphylococcus aureus* and *Propionibacterium acnes* in the strong category. Formula F3, a combination of 15% *Centella asiatica* leaf extract and 10% sweet orange extract, provided the highest inhibitory power (22.74 mm) and was not significantly different from the positive control. The results of the One-Way ANOVA statistical test ($p < 0.05$) and the Tukey HSD tests confirmed that F3 was the most effective formulation, with antibacterial activity comparable to commercial antiseptic products.

CONCLUSION

This study shows that facial wash formulations containing Pegagang (*Centella asiatica*) and Sweet Orange (*Citrus sinensis*) extracts meet physical quality requirements and contain bioactive compounds, including flavonoids, saponins, and tannins, which contribute to antibacterial activity. Antibacterial tests showed that all formulations were able to inhibit the growth of *Staphylococcus aureus* and *Propionibacterium acnes*, with Formula F3 showing the strongest antibacterial activity, comparable to the positive control. These results highlight the potential of this formulation as a herbal facial wash for acne prevention.

REFERENCES

Ansel, H. C. (2017). *Pengantar Bentuk Sediaan Farmasi*. Jakarta: UI Press.

Aryani, D. T., & Riyaningrum, W. (2022). Hubungan Acne Vulgaris Dengan Kepercayaan Diri Pada Mahasiswa Universitas Muhammadiyah Purwokerto Angkatan 2021. *Jurnal Kesehatan Tambusai, 3*(3), 434–441.

Cowan, M. M. (2018). Plant products as antimicrobial agents. *Clinical Microbiology Reviews, 12*(4), 564–582.

Endarini, L. H., Mutiarawati, D. T., & Nugrahini, A. (2022). Pertumbuhan Bakteri Bacillus Cereus Dan Salmonella Typhi Pada Ekstrak Mentimun (Cucumis Sativus). *Jurnal Penelitian Kesehatan" Suara Forikes"(Journal Of Health Research" Forikes Voice"), 13*(3), 663-666.

Herawati, R.D., Riyanta, B.A., Febriyanti, R. (2020). Formulasi Dan Uji Sifat Fisik Sediaan Gel Facial Wash Dari Ekstrak Lobak (Raphanus Satinus L) Dan Bengkuang (Pachyrizus Erosus). *Jurnal Politeknik Tegal*. 2089-5313.

Husnani, H., Suhaimi, S., Puspasari, H., & Sari, Y. (2020). Uji Daya Hambat Ekstrak Kental Daun Kratom (Mitragyna Speciosa Korth) Terhadap Bakteri *Staphylococcus aureus* Sebagai Penyebab Jerawat. *Medical Sains: Jurnal Ilmiah Kefarmasian, 4*(2), 95-100.

Rowe, R. C., Sheskey, P. J., & Quinn, M. E. (2012). *Handbook of Pharmaceutical Excipients* (6th ed.). London: Pharmaceutical Press.

Sandy, M., Wardani, T. S., & Septiarini, A. D. (2021). Uji Aktivitas Antibakteri Ekstrak, Fraksi N-Heksan, Fraksi Etil Asetat, Fraksi Air Daun Pegagan (*Centella asiatica* (L.) Urb) Terhadap Escherichia coli ATCC 25922. Media Farmasi Indonesia, 16(2), 1683–1692.

Sasebohe, V. Y., Cantya Prakasita, V., & Aditiyarini, D. D. (2023). Aktivitas Antibakteri Ekstrak Etanol Daun Binahong Terhadap *Staphylococcus aureus* Dan *Propionibacterium acnes* Penyebab Jerawat. *Jurnal Farmasi, 4*(1).

Senja, R. Y., Indriaty, S., & Rohadi, D. (2024). Antibacterial Activity of Ethanol Extract of Cucumber Fruit (Cucumis Sativus L.) On Staphylococcus Epidermidis and *Propionibacterium acnes*. *Jurnal Kesehatan Muhammadiyah, 5*(2).

Utari, N. N. P. (2023). Literature Review: Ekstrak Daun Pegegan Sebagai Anti Jerawat. Jurnal Ilmiah Farmasi Akademi Farmasi, 6(1).