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Hair Growth and Antibacterial Effects of Nanoparticle-Based Cosmeceutical Tonic from *Pogostemon cablin* and *Morus alba* Leaves

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Abstract: Hair loss affects a substantial portion of the global population, regardless of age or gender. One effective strategy to address this problem is the use of hair tonics that stimulate hair follicle activity and promote hair growth. This study developed an innovative cosmeceutical nanoparticle-based hair tonic combining Nilam and Mulberry leaf extracts, termed nanoNilamBerry. The formulation was prepared using the ionic gelation method and evaluated on rabbits over 28 days by assessing hair length, growth rate, and hair weight. The nanoNilamBerry formulation fulfilled the physicochemical requirements of a nanoparticle system and demonstrated good stability at 4 °C, 25 °C, and 40 °C for up to 28 days. In antibacterial assays against *Staphylococcus epidermidis*, the Nilam–Mulberry extract produced an inhibition zone of 16.36 ± 0.35 mm, while nanoNilamBerry exhibited a larger zone of 18.4 ± 0.30 mm. The hair tonic formulation showed no signs of skin irritation. The nanoNilamBerry treatment resulted in a hair growth rate of 0.13 ± 0.03 cm/day and a final hair weight of 32.36 mg, compared to 0.09 ± 0.01 cm/day and 27.3 ± 0.31 mg for the simple Nilam–Mulberry extract. Overall, the nanoNilamBerry hair tonic demonstrated superior efficacy, promoting hair growth 3.04 times faster than the untreated control and 1.45 times faster than the non-nano extract combination. These findings indicate that nanoNilamBerry not only provides effective antibacterial protection but also significantly enhances hair follicle stimulation and growth performance.

Introduction

Alopecia or baldness is a common side effect that occurs in cancer patients undergoing chemotherapy. When docetaxel is given as monotherapy at 75 mg/m^2, the prevalence of *alopecia* is seen in 34.3-42.9% of patients. Hair loss makes patients feel seriously ill, and it is difficult to keep their cancer status a secret. Patients lost confidence and feared chemotherapy-induced *alopecia*, resulting in up to 14% of patients considering refusing chemotherapy in their recommended cancer treatment (1, 2). Hair loss is also caused by stress, taking medications, hormonal imbalances, menopause, the presence of bacteria and fungi that cause dandruff, sun exposure, use of chemicals such as hair straighteners, curling irons, and hair coloring (3, 4). Synthetic treatments for hair loss, such as minoxidil and finasteride, are commonly used but can cause side effects like scalp irritation, dizziness, and sexual dysfunction. Studies show that up to 5–15% of users may experience these adverse effects (5).

Patchouli or Nilam leaf (*Pogostemon cablin*) is a plant that has many benefits. Previous review papers have highlighted the occurrence of various phytocompounds and their potential therapeutic properties, including antimicrobial, analgesic, antiinflammatory, antioxidant, antiplatelet, aphrodisiac, antithrombotic, antidepressant, antimutagenic, fibrinolytic, antiemetic, and cytotoxic activities (6). Fresh Nilam leaves are widely used as a hair wash (7) and used in the cosmetic and hygiene industries (8). Nilam leaves contain a wide variety of phytochemicals with significant biological activity. Compounds found in Nilam include flavonoids, alkaloids, polyphenols, and glycosides (9).

Mulberry leaves (*Morus alba* L.) are widely used in medicine. Mulberry leaves contain bioactive substances that have the potential to act as anti-bacterial agents. Mulberry leaves contain active substances, alkaloids, flavonoids, and polyphenols, which show that mulberry leaf extract can inhibit bacterial growth (10-12). The content of flavonoids, alkaloids, and steroids in Nilam leaves and Mulberry leaves can be the capital of cosmeceutical development to stimulate hair growth and as an antibacterial to reduce hair loss. (13). Antibacterial activity in a hair tonic promotes hair growth by maintaining scalp hygiene, reducing infection and inflammation, preventing follicle blockage, and creating

optimal conditions for follicle cycling (14). Flavonoids are compounds that function to accelerate hair growth, and saponins function to clean dirt from the skin because they form foam and can increase peripheral blood circulation to increase hair growth. Alkaloids act as vasodilators because they increase the blood vessels, which can trigger hair growth (15).

Research on the combination of Nilam leaf and Mulberry leaf extracts as hair growers has yet to be conducted. The combination of the two plants was intended to enhance their efficacy and provide maximum therapeutic effect (16). Another major innovation is to make hair tonic in the form of nanoparticles, which are expected to provide more optimal benefits (17). Nanoparticles can increase solubility, permeability, and facilitate penetration into the intercellular space, and increase compound affinity due to the wider surface area. Nanoparticles maximize the penetration of active substances to the inner scalp (18).

Based on this rationale, a study was conducted on the cosmeceutical hair tonic nanoparticle formulation (nanoNilamBerry), which combines extracts from Nilam leaves and Mulberry leaves. The study evaluated its effectiveness in rabbits by assessing hair length, hair growth rate, and hair weight over 28 days. Additionally, antibacterial tests against *Staphylococcus epidermidis*, a common scalp bacterium, were performed to develop a high-quality, preclinically tested cosmeceutical hair tonic.

Experimental Section

Materials

Patchouli or Nilam (*Pogostemon cablin*) Leaf Extract; Mulberry (*Morus alba* L.) Leaf Extract; Chitosan (Sigma); STPP (Brataco); ethanol 96%; propylene glycol (Dow Chemical); PEG 400; methyl paraben (Amresco); Menthol (Macron); acetic acid (Brataco); and distilled water. All other chemicals were of analytical grade, purchased from the material warehouse at the Faculty of Pharmacy, Pancasila University.

Extract Preparation

Each simplisia of Nilam leaves and Mulberry leaves was prepared and weighed at 500 g, then placed into a jar. Seventy percent ethanol (3.5 L) was added to achieve a 1:7 ratio. The jar was covered with aluminum foil and sealed with a lid, then allowed to stand for 24 h with stirring every 24 h. This process was repeated three times. The mixture was then filtered using filter paper to obtain the filtrate, which was subsequently evaporated with a rotary evaporator at 55 °C and 70 rpm until a thick extract was obtained.

Formula Design and Preparation of Hair Tonic

The hair tonic base was prepared by mixing in 10 (%w/v) propylenglycol, 5 (%w/v), 10 (%w/v) ethanol 96%, menthol 0.3 (%w/v), methyl paraben 0.2 (%w/v), and aquadest. In the simple combination extract hair tonic formula, patchouli or nilam leaf extract 0.75 (%w/v) and mulberry leaf 0.25 (%w/v) were suspended in the hair tonic base with a magnetic stirrer until homogeneous.

Nanoparticles of nilam leaf and mulberry leaf extract combination were made by the ionic gelation method using 0.2% chitosan, then crosslinking with the addition of STPP 0.1% drop by drop on a magnetic stirrer with a stirring speed of 500 RPM for 1 hour until homogeneous and stable nanoparticles are formed.

Evaluation of Hair Tonic Preparation

Particle size and polydispersity index (PDI) were measured using a Delsa™ Nanoparticle Size Analyzer (Beckman Coulter, Brea, USA), while zeta potential was determined using a Zetasizer Delsa™ Nano (Beckman Coulter, Brea, USA). Physical evaluations of the nanoNilamBerry hair tonic included organoleptic assessments (odor and color), pH, and viscosity measurements. The stability test was conducted to evaluate the physical stability of the formulation under different storage temperatures. An initial cycling test was performed, followed by storage at three temperature conditions: low (4 ± 2°C), room (25 ± 2°C), and high (40 ± 2°C) for one month, with observations recorded every seven days.

Antibacterial Activity by Agar Diffusion Method

The antibacterial activity of the nanoNilamBerry hair tonic was evaluated against the Gram-positive bacterium *Staphylococcus epidermidis* using the agar diffusion method. Antibacterial efficacy was determined by measuring the diameter of the inhibition zone, characterized by a clear area surrounding the paper disc, indicating bacterial growth suppression.

In Vivo Test

Animal Preparation

The research was conducted at the Department of Pharmacology and Toxicology, Faculty of Pharmacy, Pancasila University, Indonesia, after obtaining ethical approval from the Health Research Ethics Committee of the Faculty of Pharmacy, Pancasila University, with number 044/KEPK-FFUP/XII/2024. The experimental animals used were male rabbits with the New Zealand White strain (*Oryctolagus cuniculus*), 2-3 kg weight, 2-3 months old, obtained from Bogor Agricultural University, as many as 4 rabbits. Before the experiment, all rabbits were kept for approximately one week for environmental adjustment.

Acute Dermal Irritation Test

The test procedure followed the guidelines outlined in BPOM Regulation No. 7 of 2014 concerning *Guidelines for In Vivo Nonclinical Toxicity Tests*, using duplicate testing on one rabbit (19). Rabbits with healthy skin were shaved from the shoulder blade area (scapula) to the groin region on both sides of the body. A 0.5 mL dose of the liquid test preparation was applied onto sterile gauze and affixed to the designated skin area. The application site was then covered with gauze and secured using a non-irritant adhesive plaster. The exposure period lasted for 4 h. The test animals were subsequently observed for signs of erythema and edema at 1, 24, 48, and 72 hour after patch removal. Observations were recorded in a table summarizing irritation scores for erythema and edema at each observation time point.

Assessment of skin irritation reactions, including erythema and edema, was performed using the following scoring criteria: Score 0 – No erythema and no edema formation; Score 1 – Very slight erythema and edema (barely perceptible); Score 2 – Clearly visible erythema and slight edema (well-defined area boundaries); Score 3 – Moderate to severe erythema and moderate edema (swelling extending approximately 1 mm beyond the exposure area); Score 4 – Severe erythema (deep red coloration) up to eschar formation that interferes with erythema assessment, and severe edema (swelling extending more than 1 mm and

spreading beyond the area of exposure to the test preparation).

The Primary Irritation Index (PII) was calculated using **Equation 1**, according to OECD Guideline 404 (Acute Dermal Irritation/Corrosion) and BPOM Regulation No. 7 of 2014 (*In Vivo* Nonclinical Toxicity Tests).

$$\text{PII} = \frac{\text{Mean}_{\text{test group}} - \text{Mean}_{\text{control group}}}{\text{Number of animals}}$$

Equation 1 | Description: PII = Primary Irritation Index.

Hair Growth Activity Test of Hair Tonic

The hair growth activity test was conducted on rabbits. The fur on the rabbit's back was shaved clean and divided into six rectangular sections measuring 2 × 2.5 cm, with a 1 cm distance between each section. After shaving and before applying the hair tonic, the divided areas were swabbed with 70% ethanol to ensure antisepsis. The treatment groups were as follows:

Group I: Untreated (normal control); Group II: Hair tonic base only (negative control); Group III: Commercial herbal hair tonic (Natur®); Group IV: Hair tonic containing 2% minoxidil (positive control); Group V: Simple combination of Nilam leaf and Mulberry leaf extracts; Group VI: NanoNilamBerry hair tonic.

Before treatment, all rabbits were acclimatized for one week to minimize stress. Hair tonic treatments were applied twice daily, in the morning and evening. The first day of application was considered Day 0, and observations were carried out for 28 days.

Hair samples were collected from each section every seven days (on Days 7, 14, 21, and 28). From each treatment area, three strands of hair were plucked, straightened, affixed to adhesive tape, and measured for length using a caliper.

Data Analysis

The study results were presented based on the measurement data of rabbit hair growth, which were also visualized through hair growth graphs. Statistical analysis was performed using the Analysis of Variance (ANOVA), Kruskal–Wallis, and Mann–Whitney tests to determine the differences in hair growth among the following groups: untreated (control), hair tonic base, commercial herbal hair tonic (Natur®), minoxidil 2% tonic, the combination of Nilam and Mulberry leaf extracts, and the nanoNilamBerry hair tonic.

Results and Discussion

Characteristics and Stability of nanoNilamBerry

The Nilam leaf extract yield was 19,62% and the mulberry leaf extract was 16,38%. Based on the particle size examination, the particle size in this formula was 196,8 nm, PI 0,58 with a zeta potential of +24.80. In this study, the measurement results of nanoparticles of a combination of Nilam leaf extract and Mulberry leaf were 196.80 nm. These results indicate that the nanoNilamBerry hair tonic nanoparticles fall within the acceptable nanometer range of 10–1000 nm. For topical delivery targeting hair follicles in baldness treatment, particle sizes between 50–300 nm are generally considered optimal, as they can penetrate through the stratum corneum and localize in follicular openings. The hydrogel-based, hydrophilic formulation of this tonic further supports effective follicular delivery, since it maintains hydration of the scalp and enhances nanoparticle permeation. Thus, the particle size and formulation properties are suitable for delivery via the intended topical route (20).

The combination of Nilam leaf and Mulberry leaf extracts was prepared in the form of nanoparticles using the ionic gelation method. This method is quite easy to do and simple. Nanoparticles with this method will form polymer complexes that are dispersed in the form of similarly charged ions and are reconstituted with compounds that are opposite to the polymer charge. Previously, the extract of Nilam leaves and Mulberry leaves must be dissolved properly first. In this study, the mixed solvents used to dissolve the combination of extracts consisted of 96% ethanol, propylene glycol, and PEG. The extract can dissolve well in 96% ethanol. The addition of propylene glycol and PEG surfactants is used to reduce the surface tension of the combination of Nilam leaf and Mulberry leaf extracts, so that the solubility of the extracts increases and can be used at the same time as a stabilizing agent. After that, chitosan and its linker in the form of sodium tripolyphosphate (STPP) were added with a dripping speed of 1 drop / 3 s until nanoparticles of Nilam leaf extract and Mulberry leaf extract were formed with marked turbidity and color change to matcha green.

Nanoparticle formation by ionic gelation is influenced by the selection of polymers and their cross-linkers. Chitosan is a polymer that is commonly used and has many beneficial properties as a polymer material, namely inert, biocompatible, and has good mucoadhesive properties (21). The cross-linking agent sodium tripolyphosphate is the best crosslinker in the preparation of nanoparticles compared to other materials because it has many anions. Proper addition of sodium tripolyphosphate can form crosslinks that create ionic intermolecular interactions of chitosan ions with the addition of anions from STTP. After depositing, the nanoparticles were stirred with a magnetic stirrer for 15 min to allow the reaction between the cross-linked chitosan polymer and STTP to proceed completely.

Particle size and particle distribution index are important factors in nanoparticles. Nanoparticles with broad particle size distribution show significant variations in drug loading and release, bioavailability, as well as efficacy. Particle size distribution can be seen from the polydispersity index (PI) value. The polydispersity index is a parameter that indicates the homogeneity of a nanoparticle droplet size and the uniformity of the particle size distribution. A good polydispersity index value is 0 - 1, where the closer the value is to 0, the better or more uniform the distribution will be (22). From the measurement results, the polydispersity index of nanoNilamBerry is 0.58 and meets the requirements of particle size distribution, thus indicating that the nanoparticles of the combination of Nilam leaf extract and Mulberry leaf are homogeneously distributed.

In addition to particle size and polydispersity index, zeta potential value is an important parameter in nanoparticle characterization. From the nanoparticle measurements of nanoNilamBerry, a zeta potential of +24.80 was obtained. These results are quite far from the value of 0 and indicate the presence of uniformity of positively charged particles in the nanoparticle suspension, made so that the possibility of aggregation between particles is reduced, and nanoNilamBerry hair tonic nanoparticles become stable (23).

Table 1. Organoleptic (color and odor), homogeneity, sediment, pH, and viscosity of the formula..

Formula	Characteristics of hair tonic preparations					
	Color	Odor	Homogeneity	Sediment	pH	Viscosity (cP)
Hair tonic base	Clear	--	Homogeneous	--	5.4 ± 0.8	0.6250 ± 0.10
Nilam-mulberry extract hair tonic	Blackish green	Typical aromatics	Less Homogeneous	+	5.8 ± 0.6	0.6678 ± 0.12
Hair tonic nanoNilamBerry	Green	Typical aromatics	Homogeneous	--	5.2 ± 0.5	0.6770 ± 0.18

Table 2. Antibacterial activity of the formula.

Bacteria	Sample	Inhibition zone (mm) ± SD (n=3)
Staphylococcus epidermidis ATCC 14990	Nilam-mulberry extract hair tonic	16.36 ± 0.35
	The nanoNilamBerry hair tonic	18.4 ± 0.30
	Hair tonic (Minoxil)	13.56 ± 0.30
	Hair tonic base	0

Three hair tonic formulas were prepared, namely hair tonic base formula without active ingredient extracts; hair tonic formula combined with Nilam-mulberry leaf extracts; and nanoNilamBerry hair tonic formula. As positive controls, hair tonic products containing minoxil and natural ingredient-based hair tonic products on the market were used (NaturÒ). The hair tonic formula was evaluated physically, including organoleptic (odor and color), sediment, pH, and viscosity, which are shown in **Table 1**.

This hair tonic formula of Nilam-mulberry leaf is blackish green because Nilam leaf extract is blackish green with a distinctive odor of Nilam and menthol. There is a difference in color in the nanoNilamBerry hair tonic preparation. Hair tonic nanoNilamBerry is matcha green and more homogeneous. The pH of each formula ranges in the skin pH range (4.5-6.5) so that it does not cause irritation or itching on the scalp. The hair tonic solution preparation is in the form of an aqueous solution. The viscosity of the hair tonic base formula, hair tonic combination of Nilam leaf and Mulberry leaf extracts, and nanoNilamBerry hair tonic formula is 0.6250, 0.6678, and 0.6770 cP against distilled water. The greater the viscosity, the thicker the hair nourishing solution preparation. In this study, the viscosity of the three preparations was in accordance with the characteristics of hair tonic. For the stability of hair tonic combination of Nilam leaf and Mulberry leaf extracts and nanoNilamBerry hair tonic formula at temperatures of 4 °C, 25 °C, and 40 °C, which was observed on days 0, 7,14, 21, 28, appeared stable; however, on day 14, a precipitate occurred in the hair tonic combination of Nilam-mulberry leaf extracts.

Antibacterial Activity by Agar Diffusion

The antibacterial activity of the Nilam–Mulberry leaf extract tonic and the nanoNilamBerry formula was evaluated using the agar diffusion method. The inhibition zone diameter against *Staphylococcus epidermidis*, a bacterium commonly found on the scalp, was measured at a test concentration of 0.75% Nilam leaf extract and 0.25% Mulberry leaf extract. The results are presented in **Table 2**.

Both formulations demonstrated strong antibacterial activity against *Staphylococcus epidermidis*. The Nilam-Mulberry leaf extract tonic produced a clear inhibition zone, confirming its intrinsic antibacterial properties, while the nanoNilamBerry formula generated a slightly larger zone. This increase suggests that nanoparticle formulation enhances the antibacterial effect, likely by improving extract solubility and diffusion through the agar medium.

The scalp microbiome is an array of microorganisms that are important in maintaining scalp homeostasis and mediating inflammation. Disruption of scalp microbial homeostasis is associated with dermatologic conditions, including alopecia, dandruff/ seborrheic dermatitis, and scalp psoriasis. Microbial populations on the scalp and hair are strongly associated with hair growth. The most prevalent bacteria found on individual scalps are *Cutibacterium spp*. (predominantly *Cutibacterium acnes*) and *Staphylococcus spp*. (with a predominance of *S. epidermidis*), which account for approximately 90% of the total scalp microbiome. *S. epidermidis* may contribute to the pathogenesis of common scalp diseases. Inhibition of the growth of *S. epidermidis* bacteria can help prevent hair loss and treat alopecia (24-26). This antibacterial activity is because Nilam leaf and Mulberry leaf extracts contain flavonoids, alkaloids, polyphenols, and glycosides that have antibacterial activity. Polyphenols have antibacterial power by precipitating proteins. The antibacterial effects of polyphenols include reactions with cell membranes, enzyme inactivation, and destruction or inactivation of genetic material functions. Flavonoids function as antibacterials by forming complex compounds against extracellular proteins that disrupt the integrity of bacterial cell membranes. In addition to flavonoids, alkaloids also have the ability as antibacterials. The suspected mechanism is by interfering with the constituent components of peptidoglycan in bacterial cells, so that the cell layer is not formed intact, and causes cell death. Saponins can function as cleaners and antiseptics that function to kill germs or prevent the growth of microorganisms (9-11, 27).

Hair Tonic Formula Irritation Test

In this study, the hair tonic formulations were evaluated for potential skin irritation. The procedure followed the BPOM Regulation No. 7 of 2014 concerning guidelines for *in vivo* non-clinical toxicity testing. In addition, the hair tonic base was also tested separately to assess whether the formulation

vehicle itself could cause irritation. The Nilam–Mulberry leaf extract hair tonic and the nanoNilamBerry hair tonic were applied to the shaved dorsal skin of rabbits and observed at 24, 48, and 72 hours post-application for signs of erythema or edema, indicated by redness or swelling of the skin. Both the negative control (hair tonic base) and the two hair tonic formulations showed no visible erythema or edema, with an irritation index value of zero. These results indicate that both formulations are non-irritating and safe for topical application.

Hair Tonic Effectiveness

Hair tonic formula in the form of nanoparticles (nanoNilamBerry) provides maximum effectiveness compared to the simple extract form (hair tonic combination of Nilam leaf extract and Mulberry leaf). The hair growth activity was observed, and the test parameters were average hair length, hair growth rate, and final hair weight (on day 28).

Hair Length

The average hair length for each treatment group is shown in **Figure 1**. Normal hair growth (without hair tonic) reached 1.54 ± 0.10 cm on day 28, while the negative control group (treated with the hair tonic base) exhibited a comparable result of 1.50 ± 0.12 cm. In contrast, rabbits treated with hair tonics containing minoxidil, commercially available natural-based formulations (Natur®), the combination of Nilam and Mulberry leaf extracts, and the nanoNilamBerry formulation showed markedly faster hair growth than both the untreated and negative control groups. Among all treatments, the nanoNilamBerry hair tonic demonstrated the most significant effect, producing hair growth 3.04 times longer and a growth rate 3.05 times faster over the 28-day period compared to normal hair growth. Furthermore, when compared to the simple Nilam–Mulberry extract formulation, the nanoNilamBerry hair tonic resulted in 1.45 times greater and faster hair growth as seen in **Figure 2**, highlighting the superior efficacy of the nanoparticle-based system.

Hair Weight

Observations were also made of hair weight on the 28th day, namely, shaving the hair in each test area and then weighing the weight. This hair weight parameter is used to see the effect of each treatment on rabbit hair. The results of weight measurements can be seen in **Figure 3**.

Based on the results of the Kruskal–Wallis test for hair length, hair growth rate, and hair weight across all treatment groups—namely the normal control (without hair tonic), hair tonic base group, Natur® group, minoxidil group, Nilam–Mulberry leaf extract combination group, and nanoNilamBerry hair tonic group—on day 28, the Asymp. Sig value was < 0.05. This indicates a statistically significant difference ($p < 0.05$) among the treatment groups, meaning that each formulation exhibited distinct hair growth activity. Consistently, the three measured parameters—hair length, growth rate, and hair weight—showed similar statistical outcomes, confirming the significant variation in efficacy between the tested formulations.

The Mann–Whitney test was conducted to identify specific treatment groups that differed significantly in hair growth parameters—hair length, hair growth rate, and hair weight—on day 28. The results showed no significant difference ($p > 0.05$) between the normal control group (without hair tonic) and the group treated with the hair tonic

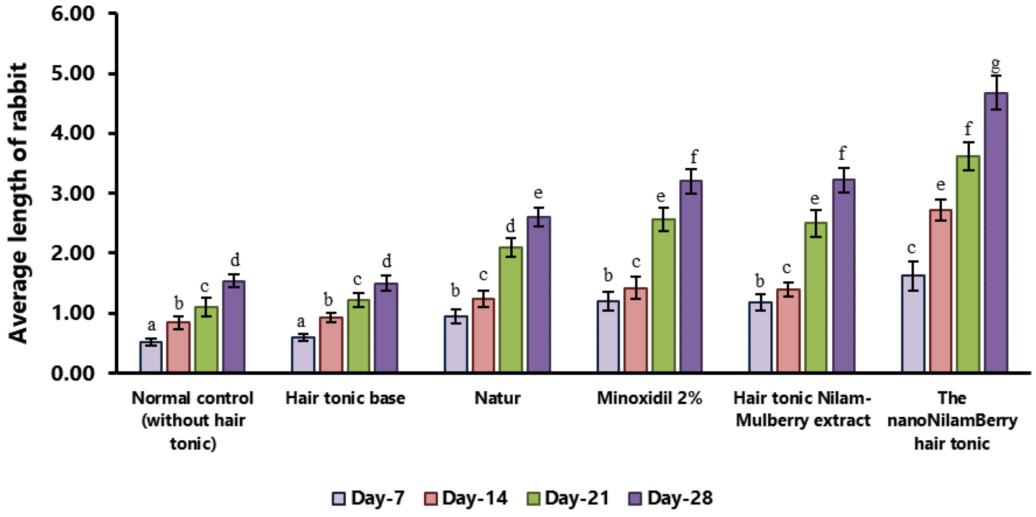

Figure 1. Average rabbit hair length over time. Weekly hair length values are presented as mean ± standard deviation (SD). Different letters within the same week indicate significant differences between groups ($p < 0.05$).

Figure 2. Hair growth in rabbits treated with different formulations. Representative images of rabbits in the following groups: (I) Normal control (no hair tonic), (II) Base tonic, (III) Natur®, (IV) Minoxidil 2% hair tonic, (V) Nilam–mulberry leaf extract hair tonic, and (VI) NanoNilamBerry hair tonic.

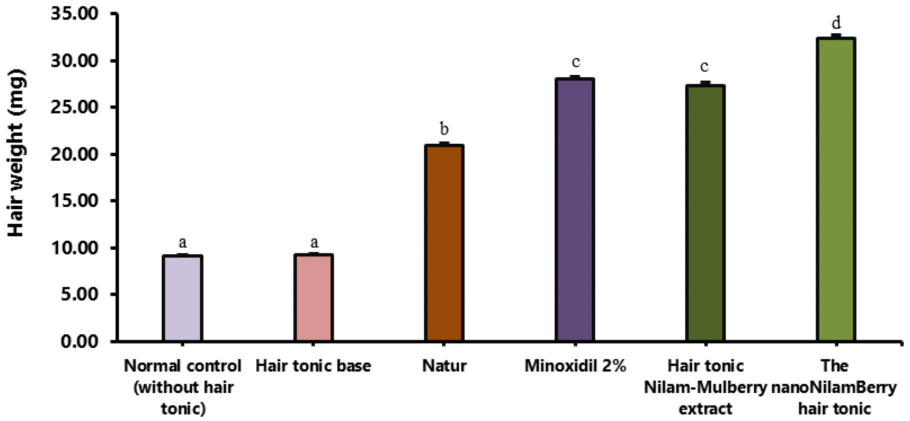

Figure 3. Average rabbit hair weight on day 28. Values are presented as mean ± standard deviation (SD). Different letters indicate statistically significant differences between groups ($p < 0.05$).

base, indicating that the base formulation alone did not affect hair growth. In contrast, significant differences were observed between these control groups (normal and base) and all groups treated with active formulations ($p < 0.05$) (Natur®, Minoxidil 2%, Nilam–Mulberry extract, and nanoNilamBerry).

There was no significant difference ($p > 0.05$) between the Minoxidil 2% and Nilam–Mulberry extract groups, suggesting comparable performance between these treatments. However, nanoNilamBerry demonstrated a significant difference ($p < 0.05$) compared to all other groups, confirming its superior effectiveness in promoting hair growth. Overall, the results indicate that nanoNilamBerry hair tonic markedly accelerates hair growth compared to all other tested formulations.

The nanoNilamBerry contains flavonoids, alkaloids, and steroids. These compounds become capital that can stimulate hair growth and act as an antibacterial to reduce hair loss. Flavonoids are compounds that function to accelerate hair growth and prevent hair loss, and have activity as bactericides. Saponins function to clean dirt from the skin because they form foam and can increase peripheral blood circulation to increase hair growth, and Alkaloids act as vasodilators because they increase blood vessels so that they can trigger hair growth.

Conclusion

In conclusion, this study showed that the nanoNilamBerry hair tonic meets the characteristics of nanoparticles, is safe, and does not cause irritation to the skin of healthy rabbits. The Nilam–Mulberry leaf extract tonic and the nanoNilamBerry formula remained stable at 4 °C, 25 °C, and 40 °C up to 28 days, although precipitation was observed in the extract tonic on day 14. The nanoNilamBerry stimulated hair growth most effectively, working 3.05 times faster than no treatment and 1.45 times faster than the extract-based tonic. Our findings provide a new insight into nanotechnology on natural materials for cosmetic applications.

Abbreviations

The nanoNilamberry= combination of Patchouli or Nilam leaf extract and Mulberry leaf; E= Erythema; U = Udema; STPP = Sodium tripolyphosphate; PSA = Particle Size Analyzer; PI = Polydispersity Index; BPOM = Badan Pengawas Obat Makanan or Food and Drug Administration; ANOVA = Analysis of Variance; ATCC = The American Type Culture Collection.

Declarations

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Conflict of Interest

The authors declare no conflicting interest.

Data Availability

The unpublished data is available upon request to the corresponding author

Ethics Statement

This study was approved by the Health Research Ethics Committee, Faculty of Pharmacy, Universitas Pancasila (Protocol No. 044/KEPK-FFUP/XII/024), dated 6 December 2024. All experimental procedures were conducted in full compliance with ethical standards and were declared ethically appropriate in accordance with the seven WHO (2011) standards.

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Additional Information

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