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POTENTİAL OF GOTU KOLA (*CENTELLA ASİATİCA* L.) EXTRACT NANOPHYTOSOME İN TOPİCAL THERAPY: A STUDY ON TEWL AND ANTİOXİDANT ACTİVİTY

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Asiaticoside in Centella asiatica exhibits antioxidant and skin-hydrating properties. Gotu kola (Centella asiatica L.), which is known to have various therapeutic benefits. Centella asiatica extract contains active compounds such as asiaticoside, madecasoside, asiatic acid, and madecasic acid, where asiaticoside has pharmacological activity as an antioxidant that can fight free radicals and stimulate type 1 collagen synthesis in fibroblast cells. This study formulated Centella asiatica extract nanophytosome using soy lecithin and evaluated their effects on TEWL and antioxidant activity. Nanophytosome were prepared by the thin-layer hydration method, resulting in four formulas: F1 (blank), F2 (0.25%), F3 (0.5%), and F4 (0.75%). All formulas showed good organoleptic properties, skin-compatible pH (4.1–5.8), particle sizes <200 nm, PDI <0.5, and zeta potentials above ±30 mV, indicating good stability. Antioxidant activity increased with higher extract concentrations, correlating with enhanced skin protection and hydration potential. One-way ANOVA revealed significant differences among formulas (p <0.05), with Formula 4 showing the greatest reduction in TEWL and the best moisturizing effect. These results indicate that Centella asiatica nanophytosome can be effectively formulated with stable physical characteristics and potential skin protection effects.

Keywords: *Centella asiatica; asiaticoside; nanophytosome; antioxidant; moisturizing*

INTRODUCTION

Skin is the largest and outermost organ of the human body, serving as a vital barrier against various external factors. The skin has an important role in protecting the body from mechanical forces, such as friction and vibration, as well as detecting physical changes in the environment, thus helping to avoid uncomfortable stimuli[1]. Long-term exposure to sunlight causes photoaging. Photoaging refers to accelerated skin aging due to prolonged sun exposure, which can cause the skin to become dry, rough, sagging, wrinkled, and hyperpigmented due to intense ultraviolet[2], that damages collagen and elastin structures. The protective function of the skin involves the stratum corneum and melanin pigment, which help absorb some UV rays, but this mechanism does not completely prevent molecular damage such as the breakdown of dermal support fibers, which accelerates the signs of aging[3]. In this modern era, various skin care products have been developed to address skin problems, one of them is reducing the effects of photoaging by maintaining moisture, strengthening the skin's protective layer, and protecting it from UV rays.

Gotu kola (*Centella asiatica* L.), which is known to have various therapeutic benefits. *Centella asiatica* extract contains active compounds such as asiaticoside, madecasoside, asiatic acid, and madecasic acid, where asiaticoside has pharmacological activity as an

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antioxidant that can fight free radicals and stimulate type 1 collagen synthesis in fibroblast cells, which is important for preventing skin aging[4]. In addition, asiaticosides can also increase the expression of Aquaporin-3 (AQP3) protein in Normal Human Epidermal Keratinocytes (NHEK) cells, which plays a role in maintaining skin hydration[5]. However, the lipophilic nature and poor aqueous solubility of asiaticosides pose challenges in their use[6, 7].

The problem of asiaticoside solubility can be overcome with pharmaceutical technologies such as nanophytosomes (NPs). Nanophytosomes are an innovation in herbal product development that binds active ingredients with phospholipids, thereby increasing absorption rates compared to conventional herbal extracts[8]. In addition, nanophytosome are lipophilic, making them easier to penetrate the lipid layer of the skin and reach the dermal target more effectively than conventional herbal extracts. Using phospholipids, which are similar to cell membrane components, makes nanophytosome preparations more biocompatible and less irritating, making them suitable for cosmetic and topical applications[9]. In the preparation of nanophytosomes, soy lecithin is used as a phospholipid. Soy lecithin contains phosphatidylcholine. Phosphatidylcholine is biocompatible, stable, biodegradable and can increase the bioavailability and absorption of drugs by forming vesicles that are more efficient in penetrating cell membranes[10].

This study aims to evaluate the physical characteristics and stability of *Centella asiatica* extract nanophytosome preparation. Antioxidant activity will be tested using 2,2-diphenyl-1-picrylhydrazyl (DPPH) method. Hopefully, the results of this study can produce nanophytosome preparations that are effective in providing stable moisture and antioxidant activity.

MATERIALS AND METHODS

Materials

Analytical scales (Ohaus PA312), oven (Memmert®), micropipette (Socorex®), hotplate stirrer (Thermo Scientific™ Cimarec+™), magnetic stirrer, sungu spoon, iron spatula, glassfirm, tweezers, sonicator (Elma-Ultrasonic S10H), UV-Vis spectrophotometry (Shimadzu®), PSA cuvette, tewameter TM 300, pH meter (Ohaus ST20), evaporator (Buchi Rotavapor® R-300), Centrifuges (Eppendorf 5430R), Tewameter TM300, aluminum foil, glassware (IWAKI and Pyrex). *Centella asiatica* L. extract, ethanol PA (Emsure®), soy lecithin (Lansida), dichloromethane (Emsure®), 70% ethanol (Onemed), 2,2-diphenyl-1- picrylhydrazyl (Himedia), vitamin C (Merck) and distilled water.

Extraction of *Centella asiatica*

Extraction of *Centella asiatica* herb was carried out by maceration using 70% ethanol (v/v) at a ratio of 1:5 (w/v). Fifty grams of powder was placed in an Erlenmeyer flask containing 250 mL of 70% ethanol and macerated for 24 hours in a macerator at a speed of 150 rpm. The solution was filtered using a Büchner funnel and the remaceration process was repeated three times for 24 hours each with a total solvent volume according to the protocol. The extraction results were evaporated using an evaporator to obtain a thick extract[11].

Thin Layer Chromatography (TLC)

Identification of asiaticoside in *Centella asiatica* extract was performed by thin-layer chromatography using a silica gel GF254 plate (11 × 5 cm) activated at 105–110 °C for 30 minutes; The extract (0.2 g) and asiaticoside standard (0.01 g) were dissolved in 10 mL of 70% ethanol to obtain concentrations of 20,000 ppm and 1,000 ppm, respectively. Separation was performed with a mobile phase of n-hexane:ethyl acetate:diethylamine (80:20:2), first saturated in a chamber containing filter paper to stabilize the vapor pressure, then the sample and standard spots were spotted with a distance of 2 cm between spots and a margin of 1.5 cm so that the effective elution length was 8 cm. After elution to the upper limit, the plate was left to dry and observed under 366 nm UV light to determine the Rf values of the extract and asiaticoside standard.

Nanophytosome Preparation

Nanophytosomes were prepared using the thin-layer hydration method. *Centella asiatica* extract was dissolved in ethanol p.a, soy lecithin was dissolved in dichloromethane. The phytoactive solution and phospholipids were mixed using a magnetic stirrer at 35°C speed 2000 rpm for 10 minutes, the nanophytosome complex was made thin layer on a rotary

evaporator at 55°C speed 50 rpm until the dichloromethane evaporated. The thin layer formed on the wall of the round bottom flask was hydrated with distilled water characterized by the formation of colloidal dispersion. The colloidal dispersion formed was sonicated using a sonicator bath for 45 minutes[12] (**Table 1**).

Physical Characteristics Evaluations
Organoleptics

This physical appearance is related to acceptability to patients. The parameters seen from this nanophytosome preparation are color, odor, shape/texture of the preparation that is physically visible. The physical appearance of nanophytosomes can be seen from the liquid consistency, clarity and slightly yellowish color[13].

pH

Testing pH using an automatic pH meter. Skin has a pH range of 4.1-5.8 or close to 6[14]. Good topical preparations have a pH range between 4.5-8.0[15].

Particle Size, Polydispersity Index and Zeta Potential

The tests were conducted at the Terpadu Laboratory of Islamic University of Indonesia, Yogyakarta. Particle size, PDI and zeta potential of nanophytosome preparations were measured by Dynamic Light Scattering (DLS) method with Particle Size Analyzer (PSA). The particle size should be <200 nm. A good polydispersity index is in the range of 0-0.5, where the lower the polydispersity index value, the more uniform the particle size in the system[16]. The zeta potential value that shows good dispersion stability has an absolute value above ± 30 mV[17].

Nanophytosomes Preparation Stability

The stability of the preparation was carried out on preparations that had been stored for 21 days. Observation of the stability of the preparation was carried out on physical changes (odor, color and shape/texture of the preparation), pH, particle size, polydispersity index and zeta potential observed on days 0, 7, 14, and 21 days. A good preparation showed no significant changes during the period of storage time[18].

Fourier Transform Infra-red (FTIR)

FTIR was performed on the prepared nanophytosomes to determine the chemical groups. Nanophytosomes of *Centella asiatica* extract were examined to determine the interaction of gotu kola extract with phosphatidylcholine compounds. To determine the compound spectrum, the sample was mixed with potassium bromide (KBr) powder (1:1) in a mortar and pestle. A 4000-400 cm transmission spectral scan was used to characterize the nanophytosomes of the *Centella asiatica* extract[8].

Table 1: Formulation of Nanophytosome *Centella asiatica* extract.

Materials	Formula (g)			
	Formula 1	**Formula 2**	**Formula 3**	**Formula 4**
Centella asiatia extract	-	0,25	0,5	0,75
Ethanol 70%	-	0,25 mL	0,5 mL	0,75 mL
Soy Lecithin	1	0,5	1	1,5
Dichloromethane	33,33 mL	16,67 mL	33,33 mL	49,97 mL
Distilled water	Add 100 mL	Add 100 mL	Add 100 mL	Add 100 mL

Note :
Formula 1 : Nanophytosome formula without extract concentration
Formula 2 : Nanophytosome formula containing 0.25% extract
Formula 3 : Nanophytosome formula containing 0.5% extract
Formula 4 : Nanophytosome formula containing 0.75% extract

Antioxidant Activity

DPPH solution was prepared by dissolving DPPH with a concentration of 100 ppm, in ethanol p.a protected from light. DPPH 10 mg was dissolved with ethanol p.a in a 100 mL volumetric flask, then shaken until homogeneous[19].

Vitamin C solution was used as positive control. Vitamin C powder of 10 mg was dissolved in ethanol p.a and then put in a 100 mL volumetric flask. The volume was sufficient with ethanol p.a until the limit mark. Vitamin C mains solution was made in several concentrations, namely 2; 4; 6; 8 ppm in a 10 mL volumetric flask. The volume was sufficient with ethanol p.a until the limit mark[19].

Nanophytosome main solution was made by weighing 100 mg of *Centella asiatica* extract nanophytosomes, dissolved in ethanol p.a and then put in a 50 mL volumetric flask, the volume was sufficient with ethanol p.a until the limit mark. Nanophytosome main solution was made in several concentrations, namely 100; 200; 300; 400 ppm and put into a 10 mL volumetric flask, the volume was sufficient with ethanol p.a until the limit mark[20].

Test and control solutions as much as 1 mL of each concentration was pipetted and put in vials, then added with DPPH solution as much as 1 mL, and added 2 mL ethanol p.a. Then shaken until homogeneous, then incubate for 30 minutes in a place protected from light. The absorbance was read at a wavelength of 517 nm. Determination of antioxidant activity using DPPH is expressed by the DPPH silencing value (IC_{50})[19].

Transepiderma Water Loss (TEWL)

TEWL is an important parameter in evaluating skin barrier function. If the skin barrier is compromised, the skin will lose its ability to retain moisture. The test was conducted on 12 volunteers (6 men and 6 women) aged 21-36 years with healthy skin. The volunteers were placed in a controlled room at a temperature of 18-22°C for 30 minutes prior to measurement. Each sample (0.1 mL) was applied to the designated area of the volunteer's forearm and allowed to dry for 20 minutes. The samples were applied to the left and right arms of the subjects. The left arm was given Formula 1 and Formula 2 samples. Then, the right arm was given Formula 3 and Formula 4 samples. The area of application of the sample on the arm was approximately 3x3 cm. Measurements were taken before sample application and after application at 45, 90, and 135 minutes. Each measurement was taken three times, and the results were averaged. Measurements were taken using a Tewameter TM300 device. The test was conducted as an open-label study[21].

Studies involving human volunteers have been reviewed and approved by the Health Research Ethics Committee of the Faculty of Medicine, Duta Wacana Christian University, Yogyakarta, Indonesia (Approval Number: **1770/C.16/FK/2025**). All procedures involving human participants were conducted in accordance with the ethical standards of the institutional research committee. The TEWL test was supervised by a physician appointed by the ethics committee and a clinical physician to monitor the procedure and observe potential allergic reactions among volunteers during the study. Written consent containing a complete explanation of the study was obtained from all participants before they participated in the study.

Rf Value of *Centella asiatica* Extract and Asiatioside Standard

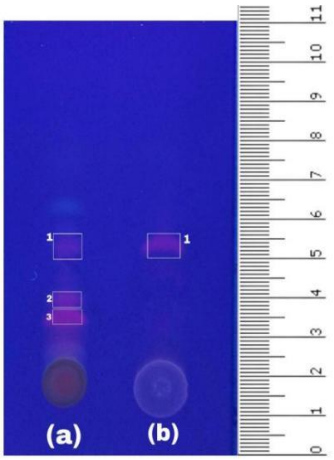


Fig. 1: (a) *Centella asiatica* extract; (b) Asiaticoside standard.

Data Analysis

Data from pH, particle size, polydispersity index (PDI), zeta potential, antioxidant activity, and the raw TEWL measurements were processed and tabulated using Microsoft Excel 2016 (Microsoft Corporation, USA). Descriptive statistics, including mean and standard deviation (SD), were calculated for all parameters. For inferential analysis of TEWL

data, one-way analysis of variance (ANOVA) followed by Tukey's post-hoc multiple comparison test was performed using IBM SPSS Statistics version 25.0 (IBM Corp., Armonk, NY, USA). A *p-value* of less than 0.05 was considered statistically significant.

RESULT AND DISCUSSION

Based on the literature, the comparator Rf value for asiaticoside was obtained as 0.33[22]. The optimal Rf value is in the range of 0.2-0.8, where the higher the Rf value, the further the compound moves on the TLC plate[23]. If two compounds have the same Rf value or the difference is less than 0.05, then they can be considered identical[24, 25]

The Rf value of *Centella asiatica* extract and standard asiaticoside **(Table 2)** is 0.33, so it can be concluded that the extract contains asiaticoside compound. In addition, spots with Rf values of 0.20 and 0.14 also appeared, which are thought to be other compounds in gotu kola extract, namely madecoside and asiatic acid. According to Monton et al. (2019), titrated *Centella asiatica* extract contains asiatic acid (30%), madecascic acid (30%), and asiaticoside (40%)[26].

Results of Evaluation of Nanophytosome Characteristics

Evaluation of the characteristics of nanophytosome preparations was carried out by observing organoleptic, pH, particle size, polydispersity index and zeta potential. The organoleptical test is seen from the color, odor, shape/texture of the preparation that is physically visible[12].

Based on organoleptic test observations, the results obtained for F1 (formula without extract) produced a cloudy white color, odorless and liquid texture. Preparation F2 (0.25% extract) produces a bright greenish brown color, has a distinctive aroma and has a liquid texture. Preparation F3 (0.5% extract) produces a bright greenish brown color, has a distinctive aroma and has a liquid texture. Meanwhile, preparation F4 (0.75% extract) produces a greenish murky brown color, has a distinctive aroma and has a liquid texture. During the 21-day storage period, the preparation was observed on days 0, 7, 14, and 21 days. The four nanophytosome formulas did not experience changes in color, odor and texture. So it can be said that the resulting preparation is stable[18].

The pH test (**Table 3**) is carried out so that the pH of the nanophytosome preparation is in accordance with the pH of the skin, so it is hoped that the nanophytosome preparation of *Centella asiatica* extract will not cause irritation to the skin[12].

Table 2: Rf value of *Centella asiatica* extract and standard asiatioside.

Rf Extract	Rf Asiaticoside
0.33	0.33
0.20	
0.14	

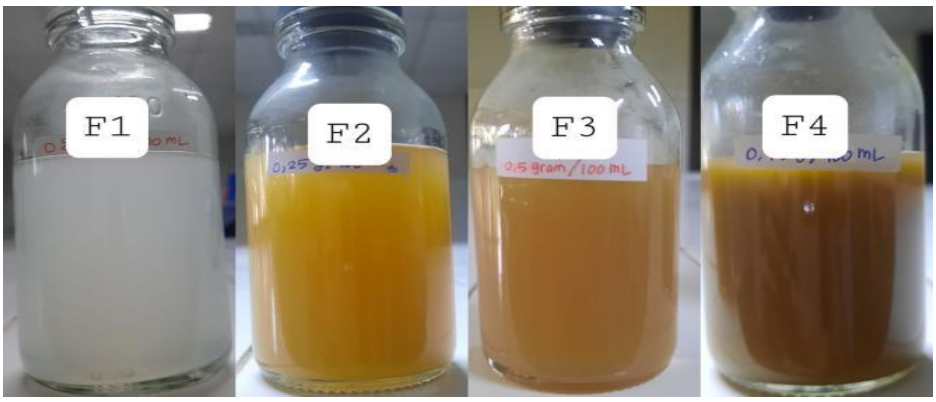


Fig. 2: F1= without extract; F2= 0.25 g extract; F3= 0.5 g extract; F4= 0.75 g extract.

Table 3: pH value of nanophytosome *Centella asiatica* extract.

Formula	Day-			
	0	**7**	**14**	**21**
F1	4.10	4.17	4.15	4.12
F2	4.52	4.83	4.81	4.82
F3	4.66	4.54	4.51	4.49
F4	4.92	4.87	4.92	4.86

Based on the pH test that has been carried out, the formulas made, namely F1, F2, F3 and F4, produce a pH that meets the criteria, where the skin has a pH ranging from 4.1-5.8 or close to 6[11] and good topical preparations have a pH range between 4.5-8.0[15]. During the 21-day storage period, pH measurements were taken on days 0, 7, 14 and 21. F1, F2, F3 and F4 did not show any significant changes in pH, so the preparation can be said to be good and stable because it does not show significant changes in pH during storage.

Measurement of particle size, polydispersity index (PDI) and zeta potential (**Table 4**) was carried out by Dynamic Light Scattering (DLS) method, employing a Particle Size Analyzer (PSA). The particle size distribution provides insight into the homogeneity of the formulation, while the PDI reflects the uniformity of particle dispersion. Zeta potential measurements were used to assess the surface charge and electrostatic stability of the particles.

Table 4: Results of particle size, polydispersity index, and zeta potential of Nanophytosome *Centella asiatica* extract.

Day-0			
Formula	**Particle Size ± SD (nm)**	**PDI ± SD**	**Zeta Potential ± SD (mV)**
F1	195.267 ± 1.328	0.407 ± 0.054	-61.2 ± 0.321
F2	197.300 ± 1.100	0.403 ± 0.034	-52.3 ± 0.557
F3	181.267 ± 0.709	0.340 ± 0.027	-49.0 ± 0.500
F4	195.883 ± 0.551	0.374 ± 0.017	-44.2 ± 0.907
Day-21			
F1	195.300 ± 2.464	0.451 ± 0.041	-31.0 ± 0.400
F2	193.467 ± 1.922	0.443 ± 0.021	-36.3 ± 0.200
F3	183.933 ± 1.401	0.275 ± 0.012	-33.9 ± 1.002
F4	187.200 ± 3.822	0.362 ± 0.037	-36.6 ± 0.802

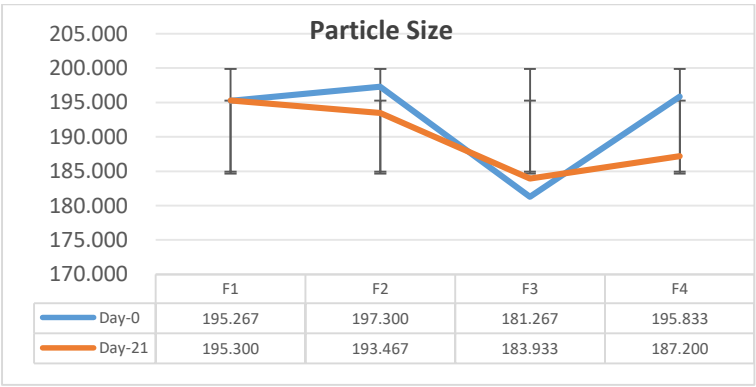


Fig. 3: Graph of particle size of *Centella asiatica* nanophytosome formulations on day 0 and day 21.

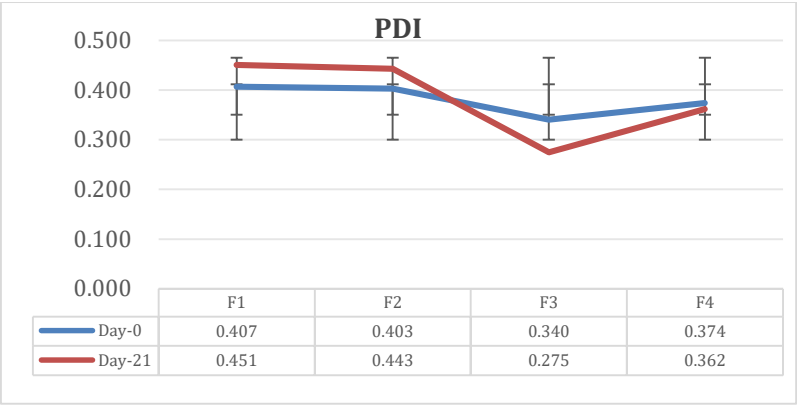


Fig. 4: Graph of polydispersity index (PDI) of *Centella asiatica* nanophytosome formulations on day 0 and day 21.

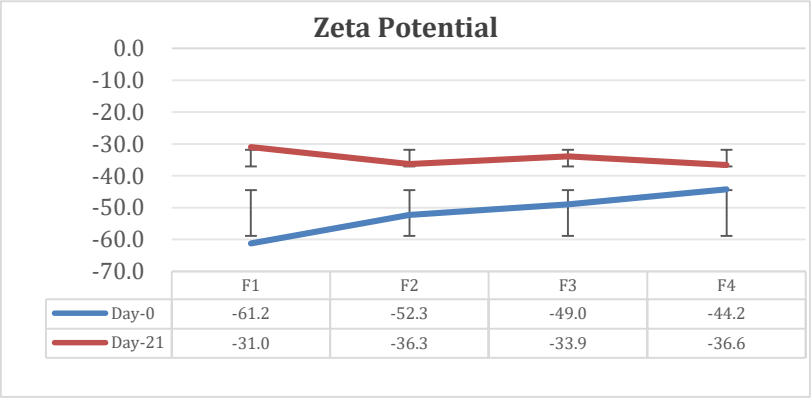


Fig. 5: Graph of zeta potensial of Centella asiatica nanophytosome formulations on day 0 and day 21.

Based on particle size tests on day 0 and day 21, *Centella asiatica* extracts in the form of nanophytosomes in formulas F1, F2, F3, and F4 were all classified as nano because they had particle sizes ≤ 200 nm[16], which falls within the ideal nanoscale range for topical delivery systems, as nanoparticles smaller than 200 nm can penetrate the stratum corneum and enhance drug retention in the skin[9]. The polydispersity index plays a role in determining particle size homogeneity. This parameter is used to measure the level of variation in particle size distribution[12]. The polydispersity index of F1, F2, F3 and F4 on day 0 and day 21 met the criteria, where the four nanophytosome formulas of *Centella asiatica* extract had a polydispersity index value below 0.5. A good polydispersity index is in the range of 0-0.5, where the lower the polydispersity index value, the more uniform the particle size in the system[16]. The potential zeta value indicates a strong repulsive force between particles, which serves to prevent aggregation and maintain the stability of the dispersion system. Based on the results of zeta potential measurements on day 0 and day 21, the nanophytosome preparations of *Centella asiatica* extract F1, F2, F3 and F4 are already in the good category and show an electrostatically stable dispersion system, where the zeta potential value that shows good dispersion stability has an absolute value above ± 30 mV[17] similar to findings by Husni and Puspitaningrum (2017), who reported that zeta potentials exceeding ±30 mV indicate good colloidal stability and prevent particle aggregation.

Based on FTIR spectrum analysis, the formation of nanophytosomes from *Centella asiatica* extract can be confirmed through significant changes in spectrum characteristics. In the spectrum of pure *Centella asiatica* extract (**Fig. 6**), a clear peak is observed in the frequency range of 3200–3600 cm^{-1} with a transmittance intensity of approximately 55–60%, indicating the presence of hydroxyl (-OH) groups from the phenolic compounds contained in the extract. This frequency region is a characteristic feature of the -OH bonds of phenolic compounds, which are the primary bioactive components in *Centella asiatica* extract. After the formation of nanophytosomes (**Fig. 7**), a noticeable change in the spectrum

occurs at the same frequency region. The intensity of the -OH peak decreases significantly to approximately 45-50%, accompanied by a shift in the peak position. This is strong evidence of hydrogen bonding interactions. The shift in the spectrum of phytosomes compared to the extract indicates the interaction between the extract and the phosphate group (P=O) part of phosphatidylcholine, which is characterized by the removal or decrease in intensity of the OH peak[27]. Similar spectral changes were reported in nanophytosome formulations of other herbal extracts such as carvacrol[27] and myricetin[12], indicating successful formation of the phytosome complex.

Antioxidant Activity

Antioxidant activity seen with IC_{50} value is defined as the concentration of substance required to inhibit activity by 50%. The IC_{50} value is obtained by calculating the %inhibition using the following formula:

$$\%Inhibition = \frac{Abs\ blanko\ -\ Abs\ sample}{Abs\ blanko}\ x\ 100\%$$

The calculation of this value is done using linear regression, which shows the relationship between the concentration of the test solution (x) and the percent inhibition (y) in the form of the equation y = bx + a. Based on this analysis, a regression curve was made which was then used to determine the IC_{50} value of each nanophytosome preparation of *Centella asiatica* extract and vitamin C. The smaller the IC_{50} value, the higher the IC_{50} value. The smaller the IC_{50} value, the higher the ability of the preparation to inhibit free radicals[28].

A compound is said to be a very strong antioxidant if the value <50 ppm, strong if the value of 50-100 ppm, moderate if the value of 100-150 ppm, weak if the value of 150-200 ppm and very weak >200 ppm[19]. Based on the antioxidant activity test conducted, the result of vitamin C is 4.326 ppm where the result is categorized as very strong. F1 preparation produced a result of 352.903 ppm which is included in the very weak category. The F1 preparation does not contain *Centella asiatica* extract, so F1 is declared to have no antioxidant activity. F2 preparation has an IC_{50} value of 149.084 ppm and F3 has an IC_{50} value of 112.613 ppm, so F2 and F3 have antioxidant activity that falls into the moderate category. Then for F4 has an IC_{50} value of 90.580 ppm, which means F4 has strong antioxidant activity, similar results were observed by Witkowska et al. (2024), who reported that the incorporation of *Centella asiatica* extract into lipid based nanocarriers enhanced free radical scavenging efficiency due to improved stability and bioavailability of triterpenoid saponins. Antioxidants can fight free radicals and stimulate the synthesis of type 1 collagen in fibroblast cells, which is important for preventing skin aging, where aging causes the skin to become dry, rough, saggy, wrinkled, and hyperpigmented[2, 4].

FTIR Nanophytosome *Centella asiatica* Extract

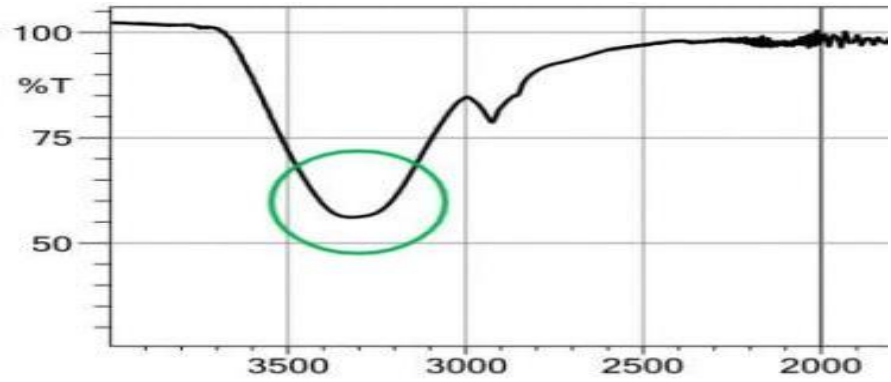


Fig. 6 :FTIR Spectrum *Centella asiatica* Extract.

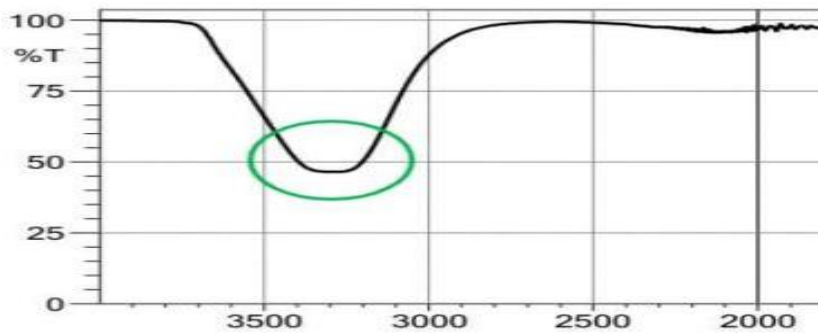


Fig. 7 :FTIR Spectrum F3 Nanophytosome *Centella asiatica* Extract.

Table 5: Result of antioxidant activity of nanophytosome *Centella asiatica* extract.

Sample	IC_{50} (ppm)	Category
Vitamin C	4.326	Very Strong
F1	352.903	Very Weak
F2	149.084	Moderate
F3	112.613	Moderate
F4	90.580	Strong

Transepidermal Water Loss (TEWL)

TEWL is the process of passive diffusion of water through the stratum corneum layer. An increase in TEWL values can inhibit the desquamation process, causing corneocytes to accumulate and adhere, resulting in dry, rough, and scaly skin.

A TEWL value of 0–8 g/m²/h reflects very healthy skin, 9–14 g/m²/h indicates healthy skin, 15–20 g/m²/h suggests normal skin, 21–24 g/m²/h signals a compromised skin barrier, and ≥25 g/m²/h represents a critical barrier condition. Based on **Table 6**, all formulations (F1–F4) showed a consistent and statistically significant decrease in TEWL values from 0 to 135 minutes ($p<0.05$). Initially, TEWL values were relatively high, but they declined steadily over time, indicating an improvement in skin barrier function after application. The one-way ANOVA revealed significant differences between most formula pairs, particularly between Formula 4 and Formula 1, which had the largest mean difference of 35.1556 ($p=0.000$). Increasing the concentration of active ingredients resulted in a measurable reduction in TEWL, reflecting enhanced barrier repair.

Based on the average percentage reduction, Formula 1 achieved a 20.0% decrease, Formula 2 reached 31.4%, Formula 3 achieved 40.5%, and Formula 4 reached 41.6%, confirming that all formulas were effective, with Formula 4 demonstrating the highest improvement in skin moisture retention. These findings are consistent with previous studies reporting the moisturizing and barrier-restoring effects of Centella asiatica through the upregulation of aquaporin-3[5] and collagen synthesis[2]. The results also align with Jaslina et al. (2022), who observed similar TEWL reductions in lipid-based nanoemulsion systems due to their occlusive and hydrating properties. During TEWL testing and subsequent observation, no participants exhibited signs of irritation or allergic reaction, indicating that all formulations were well tolerated and safe for topical application under the study conditions.

Table 6: Result of TEWL measurement**.**

Subyek	0-Minute				45-Minute				90-Minute				135-Minute			
	F1	F2	F3	F4	F1	F2	F3	F4	F1	F2	F3	F4	F1	F2	F3	F4
1	12.5	11.8	11.8	10.4	12.2	9.0	8.5	5.3	12.3	8.5	7.4	5.7	12.6	7.8	6.4	5.2
2	11.9	10.9	10.6	9.6	11.8	9.9	9.1	4.8	11.6	8.7	9.0	4.8	12.3	8.4	7.3	4.7
3	8.9	8.4	8.4	10.0	8.3	7.0	5.7	5.5	8.4	6.7	5.3	4.4	8.2	6.3	4.9	4.0
4	14.7	13.2	14.4	15.4	14.6	11.6	12.1	11.3	14.9	10.2	10.6	10.2	15.0	9.8	10.1	10.2
5	16.9	16.5	16.7	17.5	16.2	14.2	13.4	11.4	17.5	13.8	11.6	10.7	17.3	13.3	10.6	10.8
6	15.5	15.4	14.4	15.1	15.4	13.6	12.3	10.7	16.4	13.4	11.7	10.3	17.0	12.3	10.7	10.2
7	16.4	15.7	16.5	14.7	15.6	13.5	13.3	8.9	16.6	13.1	13.0	8.5	16.8	12.4	11.6	8.6
8	16.1	15.1	14.9	15.7	15.3	13.2	14.1	10.7	14.1	13.3	13.7	9.5	14.0	13.2	10.8	9.6
9	11.4	11.6	15.3	17.2	11.2	10.7	13.3	12.0	11.8	9.5	12.7	11.0	11.5	10.0	11.4	11.0
10	18.5	16.1	15.3	15.7	16.5	15.1	14.6	11.4	17.6	14.8	12.7	10.8	17.8	14.6	12.3	10.8
11	15.0	15.7	15.1	14.6	13.1	14.0	14.5	11.3	12.3	13.3	11.9	10.5	13.2	12.3	11.7	10.2
12	17.7	18.6	18.5	17.9	18.0	15.6	14.3	14.1	18.2	14.5	13.9	11.4	17.6	14.5	12.5	11.0
x̄	**14.6**	**14.1**	**14.3**	**14.5**	**14.0**	**12.3**	**12.1**	**9.8**	**14.3**	**11.7**	**11.1**	**9.0**	**14.4**	**11.2**	**10.0**	**8.9**

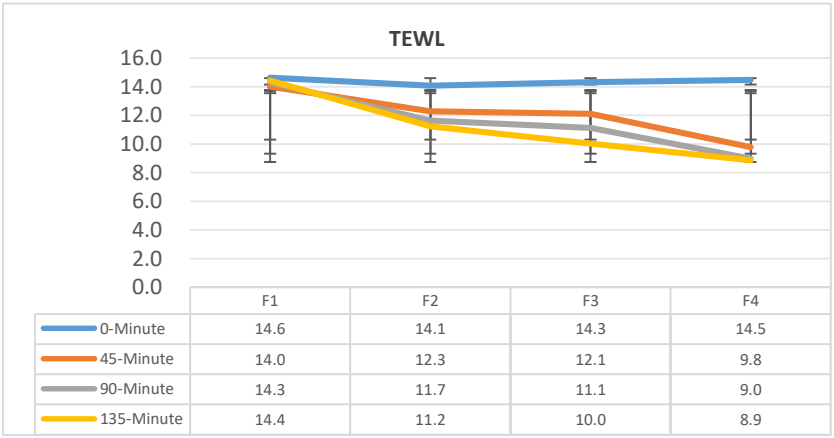


Fig. 8 : Graph of the average TEWL values of all formulations at 0–135 minutes after application.

Conclusions

Centella asiatica extract in the form of nanophytosome can be formulated with good physical characteristics and stable conditions. The higher the amount of extract, the higher antioxidant activity and also affects a significant decrease in TEWL values of the nanophytosome.

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إمكانات النانوفيتوسوم لمستخلص الغوتو كولا (Centella asiatica L.) في العلاج الموضعي: دراسة حول فقدان الماء عبر البشرة والنشاط المضاد للأكسدة

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يُظهر مركب الأسياتيكوسيد الموجود في نبات Centella asiatica خصائص مضادّة للأكسدة ومرطِّبة للبشرة. ويُعرف نبات الغوتو كولا (Centella asiatica L.) بفوائده العلاجية المتعددة، إذ يحتوي مستخلصه على مركبات فعّالة مثل الأسياتيكوسيد، والماديكاسوسيد، وحمض الأسياتيك، وحمض الماديكاسيك، حيث يمتلك الأسياتيكوسيد نشاطًا دوائيًا مضادًا للأكسدة يساهم في مكافحة الجذور الحرة وتحفيز تصنيع الكولاجين من النوع الأول في الخلايا الليفية.

هدفت هذه الدراسة إلى تحضير نانوفيتوسوم لمستخلص Centella asiatica باستخدام ليسيثين الصويا، وتقييم تأثيره في فقدان الماء عبر البشرة (TEWL) والنشاط المضاد للأكسدة. تم تحضير النانوفيتوسومات باستخدام طريقة الترطيب بالغشاء الرقيق، مما أسفر عن أربع صيغ F1 :(فارغة)، و F2(0.25%)، و F3 (0.5%)، و F4٠,٧٥%.

أظهرت جميع الصيغ خصائص حسية جيدة، وقيم pH متوافقة مع البشرة تراوحت بين (٤,١–٥,٨)، وأحجام جسيمات أقل من ٢٠٠ نانومتر، ومؤشر تشتت متعدد (PDI) أقل من ٠,٥، إضافة إلى جهود زيتا تجاوزت ±٣٠ ملي فولت، مما يدل على ثباتية جيدة للمستحضرات.

ازداد النشاط المضاد للأكسدة مع ارتفاع تركيز المستخلص، وهو ما ارتبط بتحسن القدرة على حماية البشرة وترطيبها. كما أظهر تحليل التباين الأحادي الاتجاه (One-way ANOVA) وجود فروق ذات دلالة إحصائية بين الصيغ المختلفة($p < 0.05$) ، حيث حققت الصيغة الرابعة (F4) أكبر انخفاض في فقدان الماء عبر البشرة وأفضل تأثير مرطّب.

تشير هذه النتائج إلى إمكانية تحضير نانوفيتوسوم لمستخلص Centella asiatica بخصائص فيزيائية مستقرة وتأثيرات واعدة في حماية البشرة.