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Formulation and Physical Evaluation of Hydrogel Extract Combination of *Centella asiatica* and *Moringa oleifera* using Simplex Lattice Design Method

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Abstract. This research aims to develop an optimal hydrogel formula using Design Expert 13.0 and the Simplex Lattice Design (SLD) method. The hydrogel combines *Centella asiatica* and *Moringa oleifera* extracts with carbopol 940 as a gelling agent and triethanolamine (TEA) as an alkalizer. Hydrogels were chosen due to their ease of use, cooling effect, and superior biocompatibility compared to other topical formulations like ointments or creams. The study focused on optimizing the formula to meet Indonesian National Standards (SNI). After eight hydrogel formula trials using the SLD method, physical evaluations were conducted, including organoleptic tests, pH measurements, spreadability, and viscosity tests. The optimal formula contained 0.6% Carbopol and 0.4% TEA, validated with three replications. The hydrogel exhibited a deep green color, a distinctive extract aroma, and a thick texture, meeting SNI standards with an average pH of 7.1±0.29, spreadability of 5.7±0.21 cm, and viscosity of 20,080±0.35 cPs. Stability tests confirmed its quality after cycling and centrifugal evaluations. This study promotes sustainable and eco-friendly practices by utilizing natural extracts, reducing reliance on synthetic chemicals, and contributing to environmentally conscious topical drug innovations. The development of such hydrogels aligns with sustainability goals, offering biodegradable and safe formulations that minimize environmental impact.

Keywords: Carbopol 940, Hydrogel, Simplex Lattice Design, TEA

1. Introduction

The increasing demand for wound care and inflammatory products is driven by public awareness of wound healing management [1]. The use of herbal medicine is increasingly in demand by the public as an alternative anti-inflammatory therapy. Alternative therapy has mild side effects because the content in herbal plants is balanced and neutralizes each other [2]. Herbal plants whose properties are widely known by the public and are easy to obtain include gotu kola and moringa [3].

Gotu kola (*Centella asiatica*) is a plant that contains flavonoids and triterpenoid compounds, so this plant is considered to have anti-inflammatory, antioxidant and neuroprotective activities [4]. [5] stated that gotu kola also contains active compounds from the triterpenoid group of saponins, which include asitic acid, madecassoside, and asiaticoside which are able to heal wounds as well as anti-inflammatory agents. *Moringa oleifera* contains bioactive compounds including flavonoids, carotenoids, polyphenols, tannins, and saponins. Flavonoids are compounds that are able to work to accelerate the inflammatory healing process [6]. Thus, to obtain active compounds from gotu kola and moringa plants, it is necessary to carry out an extraction process.

Pulse Electric Field (PEF) is used in the pre-MUAE or Microwave-Ultrasound-Assisted Extraction process to improve the efficiency of polyphenol extraction. Research shows that PEF can increase the extraction of bioactive compounds such as flavonoids [7]. MUAE is an extraction method by combining microwave and ultrasonic. Simultaneous irradiation of both waves accelerates the extraction and release of the target substance from the matrix. This combination of techniques is very effective in producing bioactive chemicals from various parts of the plant such as flowers, fruits, leaves, and seeds [8].

An important factor for a drug to reach the desired concentration is its solubility and release of the active substance [2]. This study aims to find the optimal formula for anti-inflammatory topical drug preparations that are of high quality according to Indonesian national standards (SNI) and environmentally friendly. [9] stated that use of topical preparations faces the challenge of low skin penetration due to the stratum corneum with small pores that are difficult to penetrate. Therefore, the development of topical preparations that are more pharmaceutical and practical is needed [3].

According to [4], hydrogel is a topical or transdermal preparation with a highwater content that can hydrate the stratum corneum, facilitating drug penetration through the skin compared to ointments and creams. Hydrogel also has advantages such as good dispersion on the skin, cooling effect of slow evaporation of water, does not inhibit the physiological function of the skin, and effective drug release [10]. Hydrogel combination of gotu kola and moringa is expected to be able to become a preparation that has good quality. This is one of the important factors for the formulation and evaluation of the physical character and stability of hydrogel preparations with a combination of gotu kola and moringa extracts to find out whether the hydrogel preparations have physical qualities that have met the standards.

One of the most widely used optimization methods is Simplex Lattice Design (SLD). The SLD method is an experimental design technique with an effective, simple, and fast way to create a formulation compared to the 'trial and error' method [11]. Previous research by [12] stated that the results of the optimization of the gel formula of Arabica coffee powder nanoparticle extract (*Coffea arabica* L.) using *the* Design Expert (DE) software version10.0 is 0.500% Carbopol 940, 0.400% triethanolalamin (TEA), and 2.313% extract. This study shows that the optimization of inflammatory inhibition in the formulation of nanoparticle gel of Arabica coffee powder can be done by using a surface response methodology using DE software.

Based on the description above, this study aims to determine the optimal formulation of hydrogel preparations using gotu kola and moringa combination extract as a good active compound and in accordance with Indonesian national standard preparations (SNI). Formula optimization was carried out using the Simplex Lattice Design method in the Design Expert software version 13.0. The physical evaluation of the hydrogel includes organoleptic tests, dispersibility, viscosity, pH, and hydrogel stability tests.

2. Materials and Methods

2.1 Preparation of Centella and Moringa Simplisia

The centella and moringa simplisia were obtained from the Research and Development Center for Medicinal Plants and Traditional Medicine in Tawangmangu, Central Java, in powdered form. A total of 176 grams of simplisia from both plants was used in the study.

2.2 Extraction of Centella and Moringa Combination

The extraction process utilized a continuous combination of PEF (Pulse Electric Field), microwave, and ultrasonic methods. First, PEF extraction was carried out for 3 minutes at a frequency of 1000 Hz and a voltage of 2000 Volts [13]. The second step involved microwave extraction, where 176 grams of centella and moringa extracts were dissolved in 8,000 mL of distilled water and extracted for 4 minutes at 300 Watts [14]. Finally, ultrasonic extraction was performed at 120 Watts and 40 kHz for 9.5 minutes. The resulting extracts were freeze-dried, ground using a blender, and sieved with a 100-mesh sieve.

2.3 Preparation of Centella and Moringa Hydrogel

The hydrogel formulation referred to [15]. Carbopol 940 was weighed and dissolved in 30 mL of distilled water, stirred with a magnetic stirrer at 80°C for 30 minutes, and then placed in a mortar. TEA, propylene glycol (10%), sodium benzoate (0.1%), and 2% of the centella and moringa powder extract were added to the mixture and stirred until homogeneous. Each formulation was prepared to yield 40 grams of hydrogel.

2.4 Verification of the Optimal Formula of Hydrogel

The optimal formula is created by the Design Expert® software version 13.0 using the SLD method as the previous 8-run formula. The optimum formula hydrogel is tested for its physical properties and compared with the predicted physical properties of the Design Expert® software version 13.0. The observed physical properties include pH, dispersion, and viscosity by making three replications of the optimal formula of the hydrogel for the process of verifying the optimal formula of the SLD running results.

2.5 Evaluation and Optimization of Hydrogel Physical Properties

Eight hydrogel formulations were tested for physical properties such as organoleptic observations, pH, spreadability, and viscosity to determine the optimal formulation. The results were analyzed using Design Expert® version 13.0 software. The optimal hydrogel formulation was verified through physical evaluations, including organoleptic, pH, spreadability, and viscosity, using three replications to ensure the accuracy of the predicted results.

2.5.1 Organoleptic

Observation of organoleptis tests was carried out on color, aroma, physical shape in the form of uniformity of consistency of preparations visually to evaluate the hydrogel formulation of the combination of gotu gotu and moringa extracts. Hydrogels usually have a clear preparation with a semi-solid form [16]. The organoleptis test criteria of gel preparations that are good in color, odor and shape meet the specifications of the materials used [17].

2.5.2 pH Measurement

Hydrogel pH testing is determined using a pH meter (Senz, Fisher Scientific) pH testing is carried out on each hydrogel composition using a semi-solid pH meter that has been reset with a solution of pH 4, pH 7, and pH 10 [18]. The pH test was carried out to determine the pH value of the gel preparations that met the requirements according to SNI 16-3499-1996. A good pH value for the skin is 4.5-8 [19].

2.5.3 Spreadebility Test

The diffusion test of bioactive hydrogel containing gotu kola and moringa extracts was carried out using a sterile syringe. 0.5 g of prepared hydrogel is applied to a glass plate. The formulation is covered with a calibrated plate, on the surface of which a load of 150 gr is placed [15]. The result of a good dispersion of gel preparations is 5-7 cm or 5.54-6.08 cm (based on SNI standards) [20].

2.5.4 Viscosity Test

Bioactive hydrogels of gotu kola and moringa were tested for viscosity using the Brookfield DV2-T viscometer on spindles size 63 and 64 with the speed of the viscometer spindle rotated at 12 rpm [21]. According to the Indonesian National Standards Agency (BSNI/BSN/SNI) in [22] stated that in SNI 16-4380-1996, the standard viscosity value of gel preparations for skin ranged from 3,000-50,000 cps.

2.6 Hydrogel Physical Stability Test

The hydrogel temperature stability test was carried out by cycling test, while the mechanical stability test was carried out by centrifugal test. In the cycling test, the hydrogel was placed in a tightly sealed container and stored in the refrigerator for 24 hours at 4°C, then transferred to the oven for 24 hours at 40°C. This process is repeated for 3 cycles or 6 days. After that, the hydrogel sample was visually assessed to assess its physical stability, including pH, dispersion, and viscosity [23]. In the centrifugal test, 1 gram of hydrogel sample is centrifuged at 4,000 rpm and 25°C for 10 minutes to observe its physical stability, such as discoloration, phase change or separation [10].

3. Results and Discussion

3.1 Optimization of Hydrogel Formula from the Results of 8 Formula Runs

The formula optimization was analyzed using the Simplex Lattice Design (SLD) method on Design Expert (DE) software version 13.0. It involved two components (X), namely carbopol 940 and triethanolamine (TEA), and three responses (Y), namely pH, spreadability, and viscosity (Table 1). The analysis of the variance of the design model used in this study is presented in Table 2. A high R^2 determination coefficient indicates a good model fit with the data [24]. The results of the analysis also show that the model used is not overfitting, as evidenced by the difference between Adjusted R^2 and Predicted R^2 which is less than 0.02. In addition, the Adeq Precision values of all three responses (pH, spread, viscosity) already meet the minimum requirements, allowing for further testing. This is in accordance with the statement of [25] about the importance of comparing Adjusted R^2 and Predicted R^2 in the evaluation of response surface models to ensure good prediction capabilities.

Table 1. Results of physical properties testing of 8 hydrogel formula runs.

Run	Carbopol (%)	TEA (%)	pH	Spreadibility (cm)	Viscosity (rpm)
1	0,6	0,4	7,1	5,7	17.100
2	0,5	0,5	6,2	6,5	3.900
3	0,5	0,5	6,2	6,5	3.900
4	0,6	0,4	7,1	5,7	17.100
5	0,575	0,425	5,9	5,3	17.580
6	0,525	0,475	6,1	6,1	4.860
7	0,55	0,45	6,2	6,1	5.646
8	0,55	0,45	6,2	6,1	5.646

[a] Software Design Expert 13.0.

Table 2. Characterization of hydrogel formulation modeling design.

Respons	Source	Std. Dev	R-Square	Adj R-Square	Pred R-Square	Adeq Precision
pH	Linear	0,2491	0,4354	0,3413	0,212	-
	Quadratic	**0,2491**	**0,9896**	**0,7055**	**0,5928**	**6,5291**
	Cubic	0,1750	0,7169	0,8546	0,3999	-
Spreadibility	**Linear**	**0,2277**	**0,8407**	**0,6975**	**0,5827**	**7,8072**
	Quadratic	0,2332	0,7734	0,6828	0,5619	-
	Cubic	0,2240	0,7327	0,7072	0,2086	-
Viscosity	**Linear**	**3062.16**	**0,9091**	**0,7773**	**0,7161**	**9,5096**
	Quadratic	3062.16	0,8626	0,8076	0,7342	-
	Cubic	2442.37	0,8190	0,8583	0,4153	-

[a] Software Design Expert 13.0.

The results of the observation of the 8-run organoleptis formula showed identical characteristics, namely a slightly bubbly, clear, distinctive smell of gotu moringa extract, and homogeneous. The pH measurement results of the 8 run formulas showed that all run formulas met the good pH requirements for the skin, namely 4.5-6, the formula run 1 and 4 produced the highest pH, while run 5 produced the lowest pH (Table 1). The results from running eight formulas showed that the higher the concentration of carbopol 940, the higher the viscosity value (Table 2). This effect was clearly seen in the contour plot analysis results. From the figure, it was proven that carbopol 940 had an impact on both spreadability and viscosity. As stated by [26], this is due to carbopol's ability to increase viscosity by binding more water, which leads to an increase in molecular size. The higher the carbopol concentration, the smaller the gel's spreadability, whereas TEA had little effect on spreadability.

3.2 Verification of the Optimum Hydrogel Formula

The results of the ANOVA (analysis of variance) analysis of the eight running DE formulas showed significant results, namely in accordance with the requirements for modeling the optimal formula determination process. These results prove that the data can be continued towards the optimization process which shows a contourplot graph and a value of 52 desirability. [24] stated that the contourplot graph was analyzed to make it easier for researchers to identify the optimal combination of the components being studied, allowing for the determination of the most effective hydrogel formula. This shows the importance of this analysis in understanding the relationship between component concentrations and physical characteristics of hydrogels. The results from the contourplot graph show that increasing the concentration of carbopol increases the pH and viscosity values, but decreases the dispersion (Figures 1 & 3). In contrast, increased triethanolamine (TEA) concentrations decreased pH and viscosity, but increased dispersion (Figure 2).

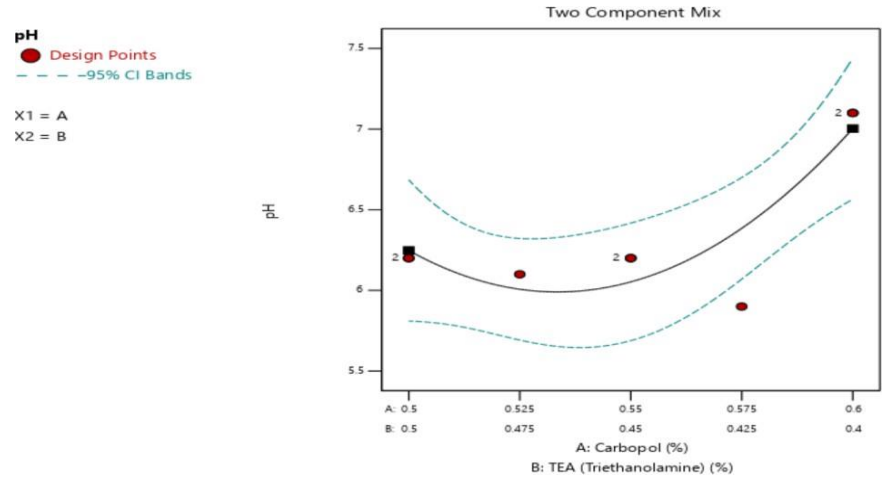

Figure 1. pH response of hydrogel from contourplot graph by Design Expert 13.0.

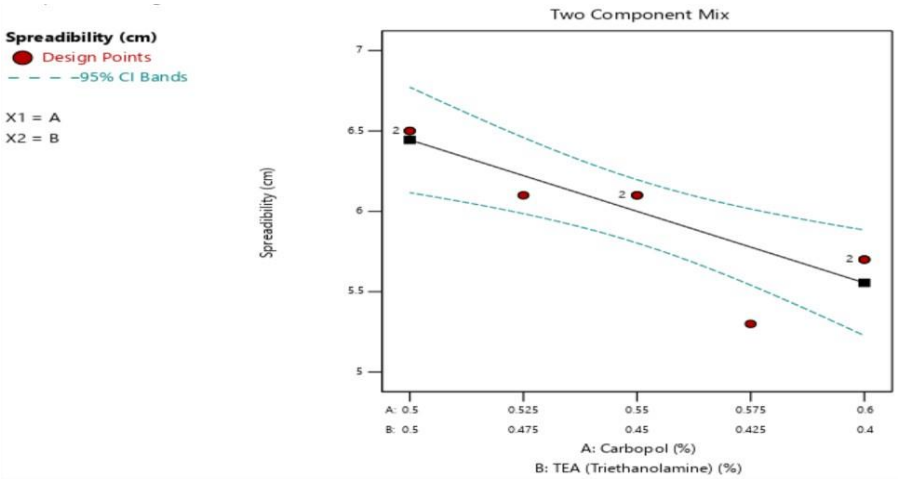

Figure 2. Spreadibility response of hydrogel from contourplot graph by Design Expert 13.0.

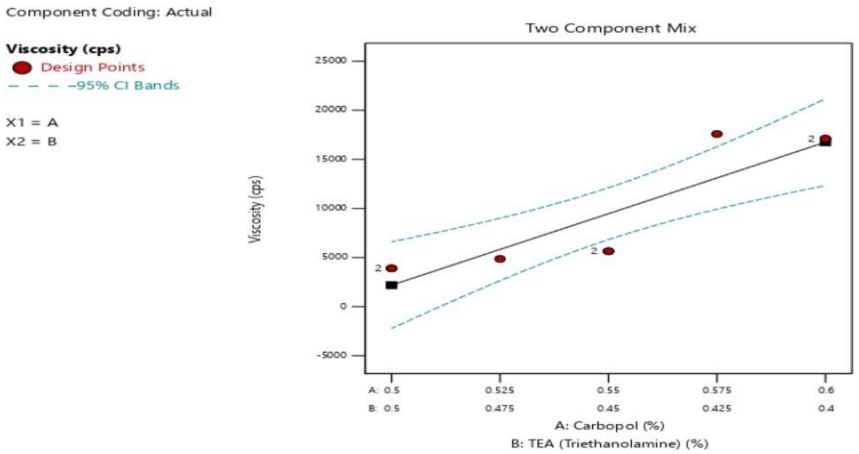

Figure 3. Viscosity response of hydrogel from contourplot graph by Design Expert 13.0.

3.3 Physical Character Testing of the Optimum Hydrogel Formula

3.3.1 Organoleptic Test

Observations of the optimal hydrogel formula showed all three replications had a dark green color, a distinct herbal scent from the centella and moringa extracts, a thick texture, and homogeneity. This aligns with [27], who found that dermato-cosmetic hydrogels with Sanguisorba minor Scop extract showed unique color and scent matching the extract mixture.

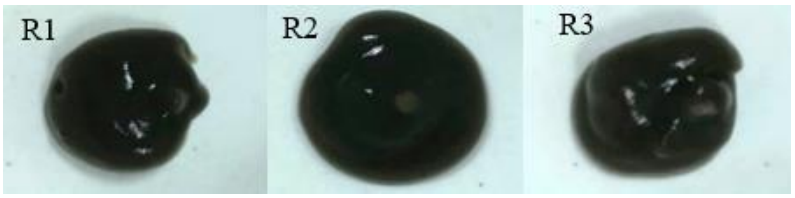

Figure 4. Optimal formula organoleptal test results.

3.3.2 H Test

The results of the optimal formula show a pH value between 6.8-7.5 (Table 3.), in accordance with the Indonesian national standard (SNI) which states that a good pH value for the skin is 4.5-8. pH adjustment is carried out using TEA (triethanolamine), which functions as an alkylation agent to form the pH and gel matrix through the formation of polymer chains. Optimization of the formula by the Simplex Lattice Design (SLD) method showed that carbopol 940 mixed with TEA produced a gel with a pH coefficient that met SNI standards, in accordance with the study that showed that the addition of TEA affected the pH value and stiffness of the gel.

The pH test showed results that met the SNI standard, with a pH value between 6.8-7.5 which is classified as normal-alkaline for the wound healing process. [28] stated that the more alkaline wound microenvironment, such as those found in acute and chronic wounds, can spur bacterial growth, so it is important to maintain acidic pH to prevent microbial growth. [29] also stated that the acidic pH value of the skin is important for skin protective regeneration and antimicrobial response, so that hydrogel formulations made according to the physiological pH of the skin can be applied without causing irritation during application.

Table 3. Physical character of pH test results.

Replication	pH
R1	7.5
R2	7.1
R3	6.8
Mean ± SD	7.1±0.29

[a] Statistical analysis.

3.3.3 Spreadability Test

The dispersion results of the optimal formulation showed a value between 5.54-6.08 cm (Table 4.), in accordance with the SNI standard range of 5-7 cm. [30] stated that a large dispersion indicates the ability of the active substance to spread widely in the skin. TEA in formulations affects the dispersion of hydrogels through the formation of polymer chains, while carbopol 940 acts as a thickener, surfactant, and stabilizer that determines the dispersion and viscosity of gel preparations.

Dispersion in accordance with SNI standards ensures that the hydrogel can cover the entire wound area evenly, preventing infection, and accelerating the inflammatory process. This is as stated by [31] that hydrogel functions in the healing of chronic wounds through adhesives, as well as antimicrobial and anti-inflammatory activities. Good dispersion helps the hydrogel penetrate and spread effectively on the surface of the wounded skin, healing without causing irritation or dryness. [18] also stated that a good hydrogel also absorbs wound exudate and adapts to the shape of the wound, providing a moist environment that supports optimal healing.

Table 4. Physical character of spreadibility test results.

Replication	Spreadibility (cm)
R1	5.6
R2	5.7
R3	6.0
Mean ± SD	5.7±0.208

[a] Statistical analysis.

3.3.4 Viscosity Test

The optimal formula viscosity value of the hydrogel meets the SNI standard for topical preparations, which is 3,000-50,000 cPs (Table 5.). [33] stated that viscosity is inversely proportional to the dispersion power; The greater the viscosity, the smaller the spread. Increased concentrations of Carbopol 940, which is used as a gelling agent, leads to higher viscosity. Carbopol 940, as a synthetic, hygroscopic and highly ionizable polymer, plays an important role in regulating the viscosity of gel preparations through the binding of solvents to their polymer structures, resulting in cross-linking in the polymer tissue.

Proper viscosity in the hydrogel is important to ensure that the active ingredient can be evenly distributed on the wound surface and adhere well, allowing direct contact with the wound tissue and working effectively [34]. Hydrogels with optimal viscosity allow active ingredients to absorb well without drying out or flowing from the application area too quickly, supporting better distribution of active ingredients and faster curing. The viscosity of the hydrogel affects hydration, oxygen permeability, and the ability to absorb wound exudate, all of which contribute to an effective healing process, reduce wound size, and accelerate the formation of new tissue [35].

Table 5. Physical character of viscosity test results.

Replication	Viscosity (cPs)
R1	20.320
R2	19.920
R3	20.000
Mean ± SD	20.080±0,35

[a] Statistical analysis.

3.4 Hydrogel Physical Stability Test

The physical stability test of the hydrogel was evaluated using the cycling test method by checking the viscosity, pH, and dispersion. After 3 cycles of cycling test, the hydrogel remained thick with a

deep green color and a distinctive aroma of the extract. The test results showed that drastic temperature changes (4°C to 40°C) affected pH values, dispersibility, and viscosity, according to [36] stating that polymer hydrogels respond to temperature changes through sol-gel phase transitions. These supramolecular hydrogels exhibit thermogel properties, forming gels at high temperatures and re-melting at low temperatures, making them potentially used as biocompatible injectable thermogels.

The results of the mechanical stability test using a centrifugation device showed that the hydrogel preparation of combination gotu kola moringa after the centrifugal test did not experience phase separation and change in the consistency of the preparation. These results are in line with the research of [37] in their study testing anti-inflammatory, antibacterial and anti-aging properties with hydrogels of *Epilobium angustifolium* L. extract and gelling agent carbopol that all analyzed hydrogels showed good physical properties and no separation of plant extracts was observed after centrifugation tests. No discoloration and odor of the hydrogel was found compared to the hydrogel before the stability test.

4. Conclusion

The study concluded that the optimized hydrogel formulation of gotu kola and moringa extract, using Simplex Lattice Design (SLD), had the best composition: 0.6% carbopol 940, 0.4% TEA, 2% extract, 10% propylene glycol, and 0.1% sodium benzoate. Physical tests confirmed it met Indonesian topical medication standards (SNI), with a herbal scent, thick texture, dark green color, pH of 7.1±0.29, a spreadability of 5.7±0.21 cm, and a viscosity of 20,080±0.35 cPs. Stability tests showed no significant changes in pH, spreadability, or viscosity, with no phase separation after mechanical testing. The study highlights the potential of combining *Centella asiatica* and *Moringa oleifera* extracts in a stable, effective herbal hydrogel for anti-inflammatory use and emphasizes the utility of SLD in formulation development. Future research should explore additional bioactives, long-term stability, and clinical validation to enhance its application.

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