

## **Polyherbal gel with Azadirachta indica, Adhatodavasica, Piper betle, Ocimumtenuiflorum, and Pongamia pinnata extracts**

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### **Abstract**

The present study involves the development and evaluation of a polyherbal gel containing extracts of Azadirachta indica, Adhatodavasica, Piper betle, Ocimumtenuiflorum, and Pongamia pinnata. The gel was formulated using a suitable gelling agent and evaluated for its physicochemical properties, stability, and in vitro release. The results indicate that the polyherbal gel is stable and possesses good physicochemical properties. The in vitro release studies showed sustained release of the active compounds, indicating the potential use of the gel in various dermatological and wound healing applications. Furthermore, the individual extracts used in the formulation of the polyherbal gel have been extensively studied for their medicinal properties, such as anti-inflammatory, antibacterial, antifungal, and antioxidant activities. The combination of these extracts in the gel is expected to enhance their therapeutic efficacy and provide a synergistic effect. The polyherbal gel can be a promising alternative to conventional topical formulations, which often have limitations such as poor stability, low efficacy, and adverse effects. Additionally, the use of natural plant extracts in the formulation of the gel may reduce the risk of adverse effects associated with synthetic compounds. Overall, the results of this study suggest that the polyherbal gel containing extracts of Azadirachta indica, Adhatodavasica, Piper betle, Ocimumtenuiflorum, and Pongamia pinnata has great potential for use in various dermatological and wound healing applications. However, further studies are needed to determine its safety and efficacy in humans.

### **INTRODUCTION**

Polyherbal formulations have gained increasing attention in recent years due to their potential synergistic effects and therapeutic benefits. Several studies have reported the use of polyherbal preparations in traditional medicine for various ailments, including skin disorders and wound healing.

Azadirachta indica, Adhatodavasica, Piper betle, Ocimumtenuiflorum, and Pongamia pinnata are widely used in traditional medicine for their medicinal properties. Azadirachta indica, commonly known as neem, is known for its antibacterial, antifungal, and anti-inflammatory activities. Adhatodavasica, also known as Malabar nut, is used

for its expectorant and bronchodilator properties. Piper betle, also known as betel leaf, has been reported to possess antioxidant and anti-inflammatory activities. Ocimumtenuiflorum, also known as holy basil, has been shown to possess antimicrobial, anti-inflammatory, and antioxidant activities. Pongamia pinnata, also known as karanja, has been used for its wound healing and anti-inflammatory properties.

In this study, a polyherbal gel containing extracts of these five plants was formulated and evaluated for its physicochemical properties, stability, and in vitro release. The aim was to develop a novel topical formulation that can potentially be used in the treatment of various skin disorders and wounds. The use of natural plant extracts in the formulation of the gel may provide a safer and more effective alternative to conventional topical formulations.

## RESULTS

### Evaluation of polyherbal gel

The polyherbal gel that has been produced for a physical analysis

It was noticed that the freshly made formulations ranged in hue from slightly off-white to yellow (Table 1). Regarding the base as well as formulations A, B, and C, there was no change in colour, odour, or appearance up to the observation period of 30 days at 80°C and 40°C using different storage conditions; additionally, formulations A, B, and C were stable [6]. Additionally, there was no change in colour, odour, or appearance up to the observation period of 30 days at different temperatures.

the pH levels of the finished formulations

After being exposed to a variety of storage conditions for a period of thirty days, it was determined to be in the range of 6.62 to 7.08. The pH of the formulations and the base that were stored at 8 degrees Celsius for one month did not exhibit considerable change, and the results showed that control (the base) was significantly better than experimental ( $p < 0.05$ ). It is interesting to note that during the one-month trial, formulation A showed an increased shift in pH (7.08), whilst the other formulations maintained a minor degree of stability. At a temperature of 40 degrees Celsius, it was discovered that the data of formulations A, B, and C were significant. Table 2.

### Examination of Viscosity

It was discovered that the viscosity and rheological characteristics of the formulations ranged from 78693.54 to 79683.92. Table 2 demonstrates that there was a statistically significant difference between the formulations A, B, C, and Control when measured at both 8°C and 40°C.

### Centrifugation

After thirty days, a centrifugation test was carried out on the formulation and the base after they had been held in a variety of storage settings. During the one-month research, formulations A, B, C, and base at temperatures of 8 and 40 degrees Celsius showed no evidence of phase separation following centrifugation. Table 1.

Spreadability

After doing research, it was discovered that the base and the three formulations (A, B, and C) had a spreadability that fell somewhere in the range of 8.30.09 to 10.00.01. It was discovered that all of the formulations and the base have high spreadability.

Washability

After applying several formulations to the skin, a manual evaluation of the ease and thoroughness of washing with water was performed. All of the formulations demonstrated excellent washability and, as a result of their non-greasy qualities, removed all residues from the skin after being washed off with water.

Acid value, peroxide value and total fatty matter determination

The acid value, peroxide value, and total fatty matter of the base as well as the formulations were monitored for a period of thirty days while being kept in a variety of storage conditions; the values for the base as well as formulations A, B, and C were found to fall within the range shown in Table 2. For the formulations and the base that were kept at various storage conditions for a period of 30 days, the acid value was found to be in the range of 2.30 to 2.91, the peroxide value was found to be in the range of 1.64 to 1.88, and the total fatty matters were found to be in the range of 15.01 to 15.5. During the stability research that lasted for one month, it was discovered that the data of acid values of the formulations and base were significant (p 0.05). It was determined that the peroxide value data for formulations A, B, and C as well as the control at 8oC and 40oC were significant (p 0.05). The findings on total fatty matter were found to be significant in every case, with the exception of formulations A, B, C, and the control at both 8 and 40 degrees Celsius. Table 2.

Table1.Physicalstudyofthepreparedformulationsduringonemonth.

Duration	Storage Conditions					
	7 days		15 days		30 days	
Parameter	8° C	40° C	8° C	40° C	8° C	40° C
Appearance						
Formulation A	Semisolid	Semisolid	Semisolid	Semisolid	Semisolid	Slightly Liquid
Formulation B	Semisolid	Semisolid	Semisolid	Semisolid	Semisolid	Semisolid
Formulation C	Semisolid	Semisolid	Semisolid	Semisolid	Semisolid	Slightly Liquid
Control or Base	Semisolid	Semisolid	Semisolid	Semisolid	Semisolid	Semisolid
Color						
Formulation A	Yellow	Yellow	Yellow	Yellow	Yellow	Dark Yellow
Formulation B	Yellow	Yellow	Yellow	Yellow	Yellow	Yellow
Formulation C	Yellow	Yellow	Yellow	Yellow	Yellow	Dark Yellow
Control or Base	Colorless	Colorless	Colorless	Colorless	Colorless	Colorless
Odour						
Formulation A	Characteristic	Characteristic	Characteristic	Characteristic	Characteristic	Bad Smell
Formulation B	Characteristic	Characteristic	Characteristic	Characteristic	Characteristic	Characteristic
Formulation C	Characteristic	Characteristic	Characteristic	Characteristic	Characteristic	Characteristic
Control or Base	Characteristic	Characteristic	Characteristic	Characteristic	Characteristic	Characteristic
Centrifugation test						
Formulation A	NSL	NSL	NSL	NSL	NSL	NSL
Formulation B	NSL	NSL	NSL	NSL	NSL	NSL
Formulation C	NSL	NSL	NSL	NSL	NSL	NSL
Control or Base	NSL	NSL	NSL	NSL	NSL	NSL

NSL: No separation of layer; SL: Separation of layer

Patchtestevaluationofvolunteers

A patch test was carried out to determine whether or not the formulation and base were safe to use on human skin. Volunteers had their forearms covered with the prepared formulations and base for a period of forty-eight hours [6]. The results that were obtained are presented in Table 3. According to the findings, the characteristics, namely the ease of application, were found to fall somewhere in the range of 2.40.2341 to 3.90.3214. The results of the spreadability tests on the formulations and the base were found to fall somewhere in the range of 2.5 0.2537 to 3.7 0.2321. It was discovered that the sense shortly after application of the formulations and base fell anywhere in the range of 2.5 minus 0.2642 to 4.0 minus 0.2775. Irritation as well as a sensation of softness on application were noted in the range of 2.560.2212 to 4.40.3431 for formulations A, B, and C across the forearms of volunteers. In contrast to the base, the findings of the patch test were uniformly favourable in every respect. The results of the paired sample t-test made it abundantly clear that the effects of formulations and base were extremely significant ( $p < 0.001$ ) in relation to all of the parameters of the patch test. Following the application of the produced formulations A, B, and C, the volunteers did not report experiencing any discomfort or redness. Figure 1 displays the findings obtained from the patch test.

The consistency of the polyherbal gel

In this experiment, the stability of the base and three distinct formulations (A, B, and C) was investigated under a variety of storage circumstances and evaluated for their colour, appearance, and odour (for a period of thirty days) [6]. Table 1 presents the findings in their entirety. After a period of 30 days, Formulation A exhibited no discernible changes in terms of its look, odour, or colour.

Antimicrobial activity

The diameter of the zone of inhibition was measured, and that was how it was calculated. Table 4 and Figures 2A, 2B, 2C, and 2D display the findings that were obtained from an analysis of the antibacterial activity of Formulation A, Formulation B, Formulation C, and the control (base) against the microorganisms that were chosen for study. When tested against *S. aureus*, *E. coli*, *B. subtilis*, and *A. niger*, base exhibited a zone of inhibition that fell somewhere between 8.98 and 0.7943 and 9.76 and 0.8798. As compared to Formulation A, Formulation B, and the base, Formulation C exhibited a zone of inhibition that was superior in the range of 15.870.7804 to 19.010.6542. As a result, formulation C shown the highest level of effectiveness against certain strains since it included a greater quantity of herbal extracts than the other formulations. It was determined that there was a statistically significant link between the two variables ( $p < 0.05$ ).

Table2. Chemical investigation of the finished formulations throughout the course of one month.

Duration	7 days		15 days		30 days	
	Storage Conditions					
Parameter	8° C	40° C	8° C	40° C	8° C	40° C
pH						
Formulation A	6.63±0.22	6.91±0.17	6.87±0.19	6.93±0.23	6.80±0.38	7.08±0.14
Formulation B	6.90±0.17	6.91±0.19	6.80±0.35	6.82±0.54	6.91±0.42	6.81±0.98
Formulation C	6.98±0.20	6.95±0.25	6.90±0.35	7.01±0.44	6.98±0.24	6.71±0.88
Control or Base	6.71±0.25	6.62±0.30	6.78±0.19	6.85±0.54	6.80±0.25	6.85±0.35
Viscosity and Rheological studies (Cps)						
Formulation A	7968±3.92	7962±2.33	7958±3.50	7952±4.01	7945±2.90	7939±3.09
Formulation B	7939±2.67	7934±3.65	7925±4.01	7918±3.65	7910±4.06	7903±3.56
Formulation C	7918±2.65	7910±3.90	7895±2.65	7894±4.11	7885±4.03	7865±3.96
Control or Base	7911±2.12	7898±2.76	7891±3.76	7882±4.04	7876±3.65	7869±3.54
Spreadability (gm-cm <sup>2</sup> )						
Formulation A	8.4±0.01	8.4±0.04	8.3±0.09	8.4±0.01	8.3±0.06	8.3±0.10
Formulation B	9.8±0.02	9.8±0.03	9.8±0.08	9.7±0.04	9.6±0.08	9.7±0.09
Formulation C	9.9±0.03	9.9±0.02	9.9±0.06	9.8±0.06	9.8±0.07	9.7±0.09
Control or Base	10.0±0.01	10.0±0.03	10.0±0.04	10.0±0.05	9.5±0.10	9.5±0.07
Acid Value						
Formulation A	2.65±0.12	2.59±0.20	2.63±0.22	2.74±0.16	2.85±0.23	2.91±0.23
Formulation B	2.55±0.15	2.57±0.19	2.57±0.21	2.75±0.21	2.66±0.23	2.79±0.15
Formulation C	2.56±0.10	2.76±0.16	2.54±0.12	2.76±0.21	2.45±0.16	2.76±0.17
Control or Base	2.30±0.06	2.37±0.09	2.45±0.11	2.43±0.05	2.46±0.15	2.50±0.17
Peroxide Value						
Formulation A	1.49±0.23	1.53±0.25	1.55±0.30	1.62±0.25	1.64±0.28	1.67±0.30
Formulation B	1.61±0.11	1.64±0.09	1.70±0.15	1.76±0.22	1.78±0.25	1.85±0.32
Formulation C	1.65±0.10	1.70±0.07	1.72±0.09	1.79±0.20	1.80±0.21	1.88±0.33
Control or Base	1.64±0.09	1.67±0.12	1.70±0.16	1.73±0.12	1.74±0.15	1.79±0.21
Total fatty matters						
Formulation A	15.55±0.23	15.42±0.32	15.34±0.26	15.30±0.15	15.24±0.32	15.18±0.35
Formulation B	15.40±0.12	15.32±0.15	15.25±0.21	15.28±0.18	15.20±0.23	15.10±0.30
Formulation C	15.33±0.19	15.32±0.30	15.20±0.23	15.13±0.28	15.07±0.29	15.00±0.32
Control or Base	15.35±0.10	15.49±0.11	15.31±0.23	15.25±0.21	15.20±0.26	15.01±0.24

Table3.Patch testing on human subjects to evaluate the effects of various prepared formulations on the skin

Variable	Average point for Control ± SEM	Average points for Formulation A ± SEM	Average points for Formulation B ± SEM	Average points for Formulation C ± SEM
Ease of application	2.4±0.2341	3.9±0.3214	3.8±0.1765	3.8±0.2343
Spread ability	2.5±0.2537	3.4±0.2142	3.6±0.2621	3.7±0.2321
Sense just after application	2.5±0.2642	4.0 ±0.2775	3.5±0.2227	3.8±0.1243
Sense on Long term	2.4±0.2361	3.8±0.2794	3.7±0.2632	3.9±0.1986
Irritation	3.7±0.2735	4.4±0.3431	4.0±0.2741	4.2±0.2012
Sense of softness	2.56±0.2212	3.7±0.2341	3.8±0.2161	3.9±0.2103

Table4.Antimicrobial sensitivity as a result of the A, B, C, and Control formulations that were developed.

Test organism	Zone of inhibition (mm)			
	Control	Formulation A	Formulation B	Formulation C
<i>S. aureus</i>	9.76±0.8798	15.56±0.6012	17.70±0.6609	19.01±0.6542
<i>E.coli</i>	9.65±0.7809	11.45±0.8648	13.76±0.6098	15.87±0.7804
<i>B. subtilis</i>	8.98±1.020	13.33±0.6523	15.65±1.0090	17.68±1.0245
<i>A. niger</i>	8.98±0.7943	15.64±0.8090	17.79±1.1565	19.22±0.9807



Figure 1. A: Formulation A; B: Formulation B; C: Formulation C and D: Control (Base) (i) Formulation and control (base) application at time = 0 hr (ii) Effect of all formulation and control at time = 48 hr after application on forearms of volunteers.

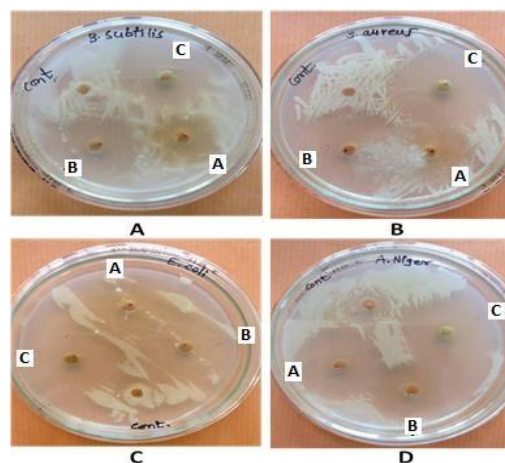


Figure 2. (A) Zone of inhibition of control and herbal formulations A, B, C and control against *B. subtilis* (B) of inhibition of control and herbal formulations v against *S. aureus* (C) Zone of inhibition of control and herbal formulations C and control against *E. Coli* (D) Zone of inhibition of control and herbal formulations C and control against *A. niger*.

## DISCUSSION

Plants are regarded to be an essential source of potentially beneficial components for the development of novel therapeutic medicines since the vast majority of plant-based constituents are risk-free and have few or no adverse effects (s). As compared to topical applications of creams and ointments, topical applications of gels at pathological locations offer significant benefits in terms of a quicker release of a medication straight to the site of action [7, 8]. These days, gels are frequently utilised as a vehicle for the topical administration of several medications. It is possible to include, as active components in this dosage form, extracts of plants and herbs that have special medical qualities in order to get additional advantages. [6, 37] *S. aureus*, *E. coli*, *Bacillus subtilis*, and *Aspergillus niger* are some of the most prevalent infectious agents that can lead to skin infections. [48], [49], and [50] Some plant and human pathogens have been used in the past to test the antimicrobial properties of plants such as *Azadirachta indica* [9, 10, 14, 17, 50], *Ocimum sanctum* [25], *Adhatodavasica* [28, 29], *Piper betle* [30-32], and *Pongamia pinnata* [35, 36]. [9, 10, 14, 17, 50]; *Ocimum sanctum* [25]; *Piper betle* [ Nevertheless, because it is difficult to apply and use these plants in their raw form on the skin's surface, the extracts of these plants have been created in the form of a gel formulation.

When it comes to the cosmetic industry, the antibacterial and natural preservation properties of the chemical components of *Azadirachta indica*, *Adhatodavasica*, *Piper betle*, *Ocimumtenuiflorum*, and *Pongamia pinnata* are highly valued.

It is generally accepted that herbal cosmetics may be used safely for extended periods of time. However, quality control for the efficacy and safety of herbal cosmetic products is of the utmost importance; and quality control tests must therefore be carried out for these preparations. Stability studies and the patch test are well-known methods that will demonstrate the efficacy and efficiency of the cosmetic herbal formulations [6, 37]. According to the criteria provided by the ICH, short-term stability testing found that the pH of all of the formulations and the base indicated variations when they were stored in different settings. Viscosity, Rheological tests, Spreadability, Acid Value, Peroxide Value, and Total Fatty Matters all revealed minor differences in their results, which demonstrated that all of the developed formulations are stable for both 8 and 400 C. The findings of the herbal preparation's viscosity and spreadability tests demonstrated that the formulation's applicability was adequate. Throughout the course of our research, we found that the created formulations rapidly spread when they were applied to the skin or the afflicted portion, and the homogeneity tests proved that there were no lumps present. Also, the physicochemical characteristics that were used in the testing of the stability of cosmetics formulations made it evident that formulations C is much superior than formulations A, B, and the base due to the comparatively larger concentration of active ingredients that it contains.

According to a search of the relevant published material, each particular extract may have a history of being recognised for its antimicrobial activity. The development of a polyherbal formulation that includes the extracts of *Azadirachta indica*, *Adhatodavasica*, *Piper betle*, *Ocimumtenuiflorum*, and *Pongamia pinnata* has not been the subject of any published research.

The findings of the patch test are depicted in Figure 1, which shows that there was no irritation or redness on the underarm following the application of formulation A, B, or C. This was based on the reports of the volunteers. Also, the results of the washability test demonstrated that none of the created formulations were greasy in any

way.

According to the findings of this research, the formulations A, B, and C that contained plant extracts were significantly more effective than the base. The existence of active elements of plants that demonstrate antibacterial action is one probable explanation for this phenomenon. Yet the efficacy of Formulation C was found to be higher than that of the other formulations when compared with each other.

## CONCLUSION

After conducting a patch test on the underarms of human volunteers for a period of 24 hours, the results of the studies showed that the prepared polyherbal formulations A, B, and C did not cause any skin irritation. These formulations contained ethanolic extracts of *Azadirachta indica*, *Adhatodavasica*, *Piper betle*, *Ocimumtenuiflorum*, and *Pongamia pinnata* at concentrations of 0.1, 0.3, and 0. The synthesised polyherbal gel was subjected to both a physical investigation and stability test, both of which demonstrated its potency and effectiveness. Thus, it is safe to apply these compositions directly to human skin. The synergistic action of the plant ingredients in the formulation may be responsible for the effective activity demonstrated by polyherbal formulations; this action may be linked to the formulation's effectiveness. The antibacterial activity of the formulation enhanced as a result of the large amount of plant extracts that were included (0.5%).

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