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PHYSICOCHEMICAL EVALUATION OF A NEEM-LOADED BIOPOLYMER PEEL-OFF HYDROGEL MASK WITH ANTIBACTERIAL ACTIVITY AGAINST CUTIBACTERIUM ACNES

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Abstract

Topical therapies for acne vulgaris must be evaluated both in terms of antimicrobial activity and the feasibility of spreading over the skin. A biopolymer peel-off hydrogel mask loaded with neem extract and polyvinyl alcohol, gelatin, and Aloe vera gel was developed and tested. It was prepared using polymer blending and solvent casting techniques, and twelve formulations, T1–T12, were prepared. The physical appearance, morphology, flexibility, thickness, diameter, pH, freeze–thaw stability against Cutibacterium acnes (C. acnes) activity were evaluated. Optimization was progressive, which alleviated cracking, brittleness, and non-uniformity in structure. The formulation T12 showed the best profile in terms of excellent flexibility, homogeneity, dimensional consistency, and high freeze–thaw stability. Formulation pH values ranged from 5.21 to 5.79, and the pH of T12 was 5.79, which was skin-compatible. Among the three strains, T12 showed the highest inhibition zone for C. acnes, which was 17.2 mm. The results suggest that T12 is a promising formulation with active compounds extracted from neem, which, if standardized and properly compared, can be subjected to long-term stability tests, irritation testing, and clinical testing to translate into the daily care of acne.

INTRODUCTION

Acne vulgaris is a chronic inflammatory condition of the pilosebaceous unit and is a common dermatological condition in adolescents and young people. It appears as comedones, papules, pustules, nodules, and sometimes as permanent scarring and hyperpigmentation. While acne is not a life-threatening condition, the condition itself can significantly impact self-esteem, social functioning, and psychological well-being [1]. Epidemiological evidence indicates that acne is a multifactorial disease that includes follicular hyperkeratinization, increased sebum production, microbial dysbiosis, and inflammation [2, 3]. These interconnected processes make these conditions difficult to treat and require systems that can simultaneously target both the microbes and the formulation.

Cutibacterium acnes is a commensal, anaerobic bacterium that pays to inflammatory acne when changes in the follicular environment favor the proliferation of pathogenic strains. It has lipase activity, which converts sebum triglycerides to free fatty acids that induce irritation and inflammatory signalling in the follicles. Bacterial antigens may also trigger innate and adaptive immune responses, leading to the release of cytokines and development of inflammatory lesions [4, 5]. With antibiotic-resistant C. acnes, long-term antimicrobial treatment is less effective, and there has been a greater need for non-antibiotic management [6]. Furthermore, systemic and topical antibiotics can alter the skin microbiome and increase resistance to an antibiotic that is not the intended pathogen [7].

Acne is effectively treated with conventional topical agents such as retinoids, benzoyl peroxide, azelaic acid, and antibiotics; however, they are often ineffective due to poor adherence, peeling, burning, erythema, and skin irritation. Simple creams and normal gels can also have a brief duration of action, such as being washed off, sweating, or as a result of sebum production. Therefore, this treatment will need repeated use, and the lack of consistency will influence the poor therapeutic results and the possible recurrence of lesions [8, 9]. These constraints have led to the development of systems for patient-centred drug delivery, which are able to offer extended release of the drug to the skin, target chemical actions, greater tolerance, and ease of administration.

Hydrogels are three-dimensional hydrophilic polymer networks that can hold a significant amount of water in their structure. They are also hydrated, which can result in a cooling effect, support skin compatibility, and facilitate the diffusion of incorporated bioactive compounds. These can also help with prolonged drug release using the hydrogel films and peel-off masks, which constitute a continuous layer on the application surface, providing an effective alternative to fast-removable semisolid preparations [10, 11]. The hydrophilicity, mechanical strength, and ability of polyvinyl alcohol to form coherent peelable films give it a wide range of applications as a film-forming polymer. Gelatin may be used to enhance the structural properties (elasticity, flexibility, biocompatibility) of the natural polymer [12]. However, optimisation of polymer concentrations is required, as too little network formation will allow brittle films to form, or an inappropriate polymer balance will limit the ability to dry, achieve dimensional stability, or peel ability.

Neem (*Azadirachta indica*) is a medicinal plant reported to have antimicrobial, anti-inflammatory, and protective anti-skin properties. The rationale for the attention given to neem as a plant-derived anti-acne agent is its phytochemical constituents such as limonoids, flavonoids, tannins, and phenolic compounds. Neem constituents could be incorporated into a peel-off hydrogel that would keep them at the skin surface and provide a slow release in the applied polymer. Aloe vera can be added as a complementary ingredient since it contains polysaccharides responsible for its gel properties, which provide hydration, soothing, and film-forming capabilities [13]. The matrix of PVA-gelatin combined with these botanical materials could thus be a multifunctional formulation with good handling properties and antibacterial activity.

Although several studies have been conducted on the optimization of physical and chemical properties and temperature-stress stability of herbal hydrogel, there is not enough information available to optimize the anti-C. acnes properties of neem-loaded peel-off masks. A facial product in its formulation should have an even color, proper consistency, uniform size, pliability, skin-friendly pH, and should not degrade or lose its shape when in storage. If the film cracks, shrinks, tears, or isn't easily removed, biological activity isn't enough. Furthermore, it is possible to use a medicinal plant as an ingredient embedded in a flexible biopolymer and thus enhance its topical applicability, as well as reduce the need for long-term treatment with conventional antibiotics in acne-prone skin. This study was undertaken to develop and optimize a neem-loaded biopolymer peel-off hydrogel mask and assess its physical parameters, thickness, diameter, pH, freeze-thaw stability, and in vitro antibacterial properties against C. acne.

MATERIALS AND METHODS

Study design

An experimental formulation study based on lab-designed formulation techniques was performed to develop, optimize, and evaluate a neem-loaded biopolymer peel-off hydrogel mask for topical management of acne. The polymer composition was systematically changed, with 12 different formulations (T1-T12) prepared. The physical appearance, morphology, flexibility, homogeneity, thickness, diameter, pH, freeze-

thaw stability, and antibacterial properties against Cutibacterium acnes of the formulations were assessed. The formulation screening was done using consistent procedures for all evaluations.

Materials

Botanicals used were fresh leaves of Aloe vera and neem (Azadirachta indica). The main film-forming polymer used was polyvinyl alcohol (PVA), and gelatin was added as a natural biopolymer to enhance the elasticity, flexibility, and skin compatibility of the film. R2 water was used as a formulation medium and polymer solvent. Appropriate microbiological culture medium and a standard strain of C. acnes were used for antibacterial testing.

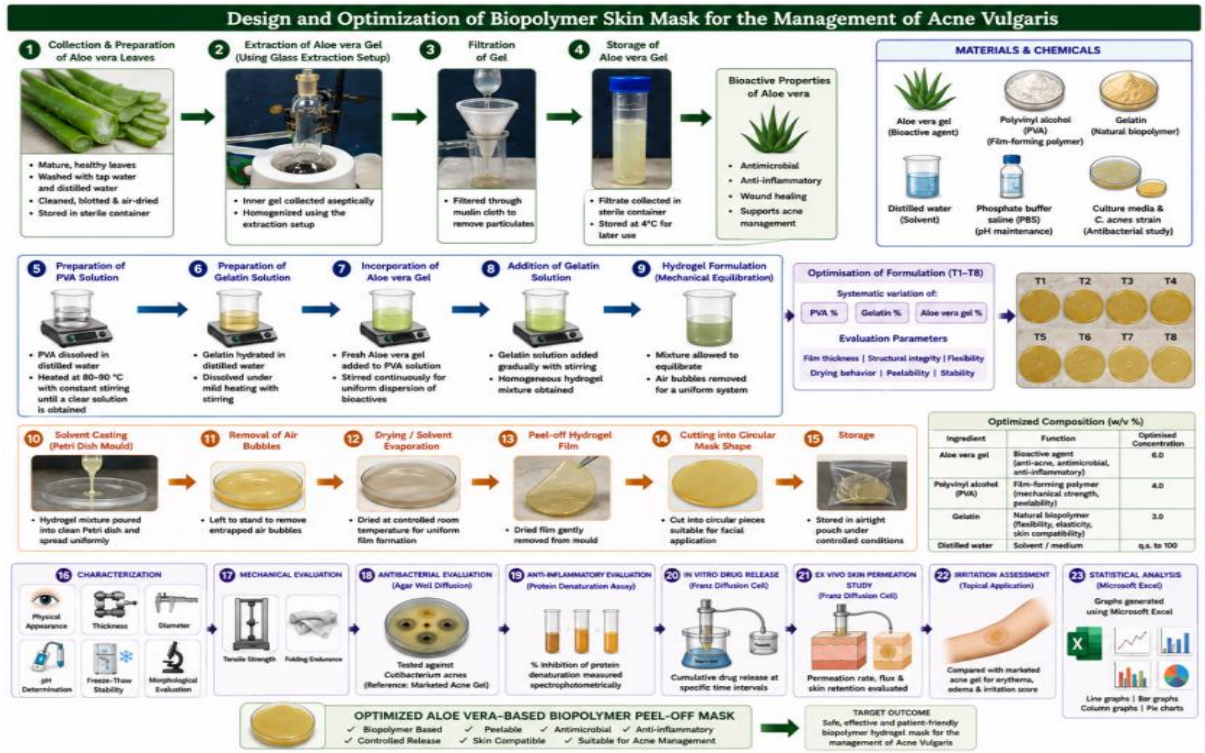


Figure 1. Experimental workflow for the preparation and evaluation of the neem-loaded biopolymer peel-off hydrogel mask

Preparation of neem extract

The leaves of Neem were washed to remove the dust and other impurities and dried. Dried leaves were powdered and macerated in an appropriate solvent. The mixture was shaken periodically to aid in extracting the soluble phytochemical constituents. After maceration, the maceration mixture was filtered to eliminate the plant particles, and the filtrate was concentrated to get an extract suitable for incorporation into the hydrogel.

Preparation of Aloe vera gel

The leaves of Aloe vera were washed with running tap water and then washed with distilled water. Flaws or discolours were cut out, and care was taken with the removal of the marginal spines and outer green rind. The transparent mucilaginous gel was harvested in a clean environment and homogenized with a sterile blender to ensure that it became more viscous and homogeneous. The homogenized gel was strained through the muslin cloth, and the material was settled in the muslin strainer. The filtrate was placed in a sterile airtight container and kept at 4°C for formulation.

Preparation of the hydrogel

predetermined amount of PVA was dispersed in distilled water with continuous magnetic stirring. The dispersion was heated at 80-90℃ until the polymer dissolved and became a clear viscous solution. The

separate hydration and dissolution of the gelatin in distilled water was achieved under mild heating. Both the prepared Aloe vera gel and concentrated neem extract were added to the PVA solution and stirred continuously to achieve uniformity of the botanical constituents. The gelatin solution was slowly added and mixing continued until a smooth and homogeneous hydrogel was made. Before casting, the preparation was left to equilibrate to eliminate bubbles. The optimized formulation consisted of 2.0% neem extract, 6.0% Aloe vera gel, 4.0% PVA, and 3.0% gelatin, with distilled water added to give 100% of the formulation.

Fabrication of peel-off masks

All the prepared formulations were converted into films by the solvent casting technique. The prepared hydrogel solutions were filled into clean Petri dishes/circular moulds and were evenly applied. The cast formulations were left to stand for a short time to drive out the bubbles trapped in the formulation and then were dried at controlled room temperature. Coherent polymeric films were obtained by the process of solvent evaporation. The films were carefully removed from the moulds without causing damage to their structure after complete drying. The produced hydrogels were cut into round samples for testing and application on the face. Masks were made ahead and stored in sealed packages in a controlled environment to reduce absorption of moisture.

Formulation optimization

The formulations have been gradually modified in the composition of the compounds T1—T12. In the optimization, factors such as film formation, structural integrity, flexibility, drying behaviour, shrinkage, peel ability, surface smoothness, and tearing resistance were considered. Formulations that yielded films that were cracked, brittle, not uniform, and/or unstable in dimension were rejected or modified. Those formulations with clearly visible structure, uniform appearance, sufficient flexibility, easy removal properties, and reasonable stability were retained. Not all of the measured properties were used to select the optimized formulation, but rather, all of them were considered.

Physical and morphological evaluation

Dried films were observed with regard to colour, transparency, homogeneity, smoothness on the film surface, flexibility, any cracking, air bubbles, phase separation, and general structural integrity. Handling and peeling behaviour were observed to assess the practicality for use with a facial mask. Surface morphology was observed and analyzed under visual and microscopic observation.

Thickness, diameter, and pH

The film thickness was determined by a digital micrometer at three different spots at a distance measured on each mask, and the meaning of the three was obtained. The uniformity of the cast film was evaluated by measuring it at several points. Each circular sample's diameter was measured with a vernier caliper. Dimensional variation, shrinkage, or deformation after drying was detected by repeated measurements.
When measuring pH, a hydrogel-mask sample was dispersed or dissolved in distilled water, and the pH was then measured with a calibrated digital pH meter. All measurements were taken in the same lab under the same conditions. The recorded values were compared to the mildly acidic range of the pH of the facial skin to determine its topical compatibility.

Freeze–thaw stability

To test the physical stability, repeated freezing and thawing of formulations was performed. After temperature stress, each film was analyzed in comparison to the original. The changes in colour, texture, shape, flexibility, shrinkage, cracking, surface deformation, phase separation and structural integrity were observed. If there were no major changes after cycling, the film was considered stable, while films which showed observable changes were assigned lower stability ratings.

Antibacterial activity

A typical agar-diffusion method was used to determine antibacterial activity against C. acnes. The culture medium was inoculated with a bacterial suspension, which was streaked evenly over the medium, creating a uniform microbial lawn. Inoculated agars were prepared, and a quantity of each formulation was added to one of the prepared wells. Samples of hydrogel discs were also created and tested. As reference material, a commercially available anti-acne gel was used under similar conditions. Inoculated plates were cultured for the growth of C. acnes under the specific growth conditions required for C. acnes. The zones of inhibition around the samples were measured after incubation in millimetres. The bigger the zones, the more antibacterial activity was seen in vitro.

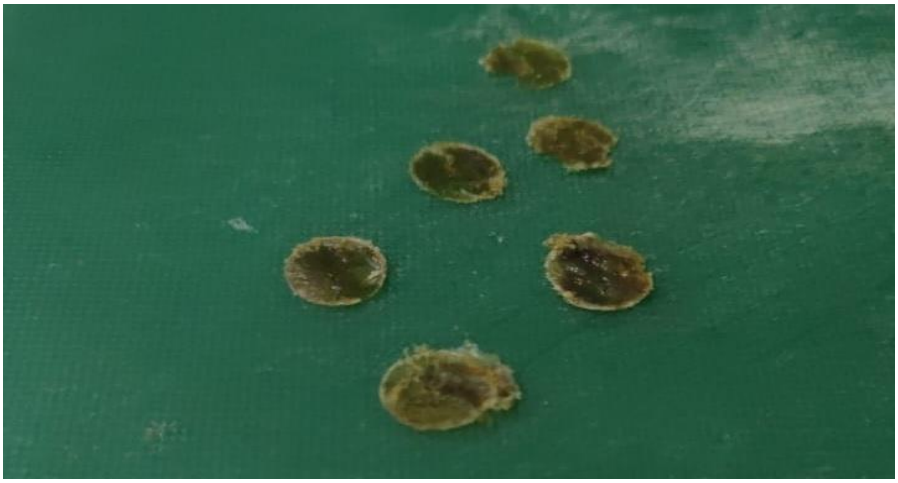


Figure 2: Hydrogel Disc Samples Prepared for Antibacterial Evaluation

Statistical analysis

The data obtained from the experiments were summarized and analyzed in MS-Excel. Physicochemical measurements were subjected to descriptive statistics to succinctly summarize the measurements and variation, and graphical presentations were prepared to compare the formulations. The physical, morphological, stability, and antibacterial results were analyzed and evaluated together to rank T1-T12 and determine the optimum hydrogel mask.

RESULTS AND DISCUSSION

Formulation Development and Optimization

The 12 formulations were clearly divided between films that resisted tearing and one that was a smooth peel-off system. The film produced by T1 was brittle with cracks, the film produced by T2 ruptured during handling, and the film produced by T3 shrank. The failures suggest that the early polymer networks were not cohesive enough to withstand the stresses imposed by the evaporation and removal of the solvent from the mould. This seems to be the interpretation of Luo et al, who attributed poor intermolecular association between composite hydrogels to poor network integrity[14]. When the solvent is lost, the polymer chains move closer together, and if not balanced with an appropriate amount of film former, elastic polymer, and retained water, then macroscopically it looks like cracking or dimensional instability. Increase in T4 to T11 demonstrates that formulation performance is not just a function of the total solids content of the product but also polymer balance. PVA enhanced the film-forming ability by forming a large amount of hydrogen bonds, while the gelatin component increased the film flexibility and decreased the brittleness. Hydrogen-bond-dominated interactions in PVA films were also related to improved mechanical stretchability, in a similar manner to what Fu et al. reported [15]. But if too much gelatin is used, it will cause softening of the matrix, resulting in poor dimensional control. The optimization then was a compromise between a highly rigid film, which would not deform and tear during peeling on one hand, and yet not be too soft to have shape retention on the other hand. The best compromise was T12, which exhibited clear formation, ease of handling, good flexibility, and uniformity. Selection was therefore not purely aesthetic but also based on mechanism.

Table 1: Observations of Hydrogel Formulations (T1–T12)

Formulation Code	Observation	Outcome
T1	Brittle film with cracks	Rejected
T2	Poor flexibility and tearing	Rejected
T3	Film shrinkage observed	Rejected
T4	Slightly improved flexibility	Modified
T5	Moderate film integrity	Modified
T6	Improved structure	Accepted for further optimization
T7	Uniform appearance	Accepted
T8	Good flexibility	Accepted
T9	Improved stability	Accepted
T10	Better film integrity	Accepted
T11	High flexibility and uniformity	Accepted
T12	Optimized formulation	Selected

The optimized formulation included: 4% PVA, 3% gelatin, 6% Aloe vera gel, and 2% neem extract. The function of the continuous film-forming phase was provided by PVA, and the skin compatibility and chain mobility were modified by the addition of gelatin, consistent with the function of the substances described by Zulkiflee and Fauzi (2021) [12]. The Aloe vera provided one hydrated polysaccharide-rich component, while the principal title-defined botanical antibacterial agent was provided by the neem. However, the formulations without neem or Aloe vera were not included in the study. Thus, it is impossible to determine the antibacterial effects of the other constituents of the optimized matrix later, as they reflect only the activity of the whole optimized matrix.

Visual and morphological characteristics

Systematic improvement was observed throughout T1-T12 through visual assessment in terms of colour, transparency, flexibility and homogeneity. Early formulations were pale yellow, poorly transparent, inflexible, and non-uniform compared to T9-T12, which were more transparent and uniform. T12 had a light-yellow color and was very transparent, very uniform, and very flexible. Analytically, it is important that the increase in transparency is related to the decrease in phase separation and to a more homogeneously dispersed composition of the polymeric and botanical components. It does not, however, establish molecular compatibility - evidence in the form of spectroscopy or thermal measurements would be required to make that claim.

Table 2: Visual Characteristics of Hydrogel Formulations

Formulation	Colour	Transparency	Flexibility	Homogeneity
T1	Pale yellow	Low	Poor	Non-uniform
T2	Pale yellow	Low	Poor	Non-uniform
T3	Yellow	Moderate	Low	Moderate
T4	Yellow	Moderate	Moderate	Moderate
T5	Yellow	Moderate	Moderate	Good
T6	Yellow	Good	Good	Good
T7	Light yellow	Good	Good	Good
T8	Light yellow	Good	Good	Good
T9	Light yellow	High	Good	Uniform
T10	Light yellow	High	Very good	Uniform

T11	Light yellow	High	Very good	Uniform
T12	Light yellow	High	Excellent	Highly uniform

The appearance of later formulations is smoother and more continuous, likely to suggest improved polymer mixing and solvent evaporation during their optimization. Asthana et al. (2021) found that optimization of the Aloe vera-PVA film system enhanced surface continuity, flexibility, and user acceptability [16]. In the present findings, that formulation logic was further extended to a neem-loaded system, but without an experimental isolation of the contribution of neem to morphology. The optimized-composition table was not reported for the presence of glycerol and pectin, although they were discussed during the development of the thesis. Not therefore be treated as proven factors at T12 until the actual levels used have been confirmed from laboratory records. This is important as it helps to keep the flexibility of the film to be reproducible even if the ingredients are not reported.
For peel-off products, this is of practical significance, since morphology is a key criterion for acceptability. Removal can induce crack formation, non-uniform areas can result in variable botanical loads, and too high and opacity can indicate aggregation. Because a smooth, flexible film enhances continuous contact with the application surface, it will be easier to obtain than a smooth yet inflexible film. Observations indicate suitability related to the physical qualities of the material, while microscopy was described in an informal manner. The results should not be considered as detailed microstructural characterization, but rather as preliminary morphological screening, if no pore size, roughness, or micrographs are provided with high-resolution.

Film thickness

The thickness of the film differed among the formulations, as is shown by the thesis graph. The direction of change agreed with a rise in polymer components and an increase in the network or matrix: the earlier polymer formulations were quite thin and mechanically weak, while the later formulations resulted in a thicker, more coherent film. This correlation is consistent with the findings by Asthana et al. (2021), which showed that the thickness and integrity of peel-off film are affected by the polymer concentrations.

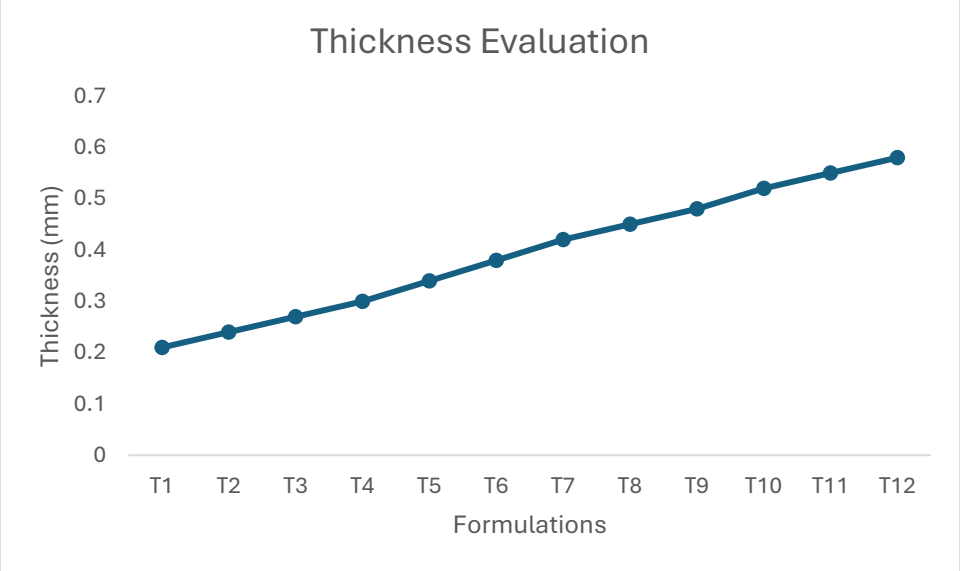


Figure 3: Comparison of Thickness

Thickness is not just a superficial kind of quality. A very thin film will dry quickly, but may rip when removed, while a very thick film will take longer to dry, not be comfortable to put on the body, and will hinder diffusion. The optimized value must therefore be a balance between mechanical stability, flexibility, and also drying behavior. T12 was found to be excellent in thickness and not brittle, as was noted in thinner early trials. The numerical values of the thickness and dispersion estimated in the thesis

text, however, are not shown in the graph. The graph data should therefore be published as the original graph data, preferably as mean ± standard deviation, n numbers of replicates. If these values are not available, differences may be discussed as trends but not as being statistically significant.

Dimensional uniformity

There were relatively small differences noted in the diameter measurements made in the formulations, indicating generally consistent casting. Later formulations (mostly T9 - T12) proved to be more stable dimensionally, and the low polymer formulations were more prone to shrinkage. The result is in line with the proposed mechanism by Boateng et al. (2008), which involved polymer composition and solvent evaporation affecting the film contraction and structural integrity [17].

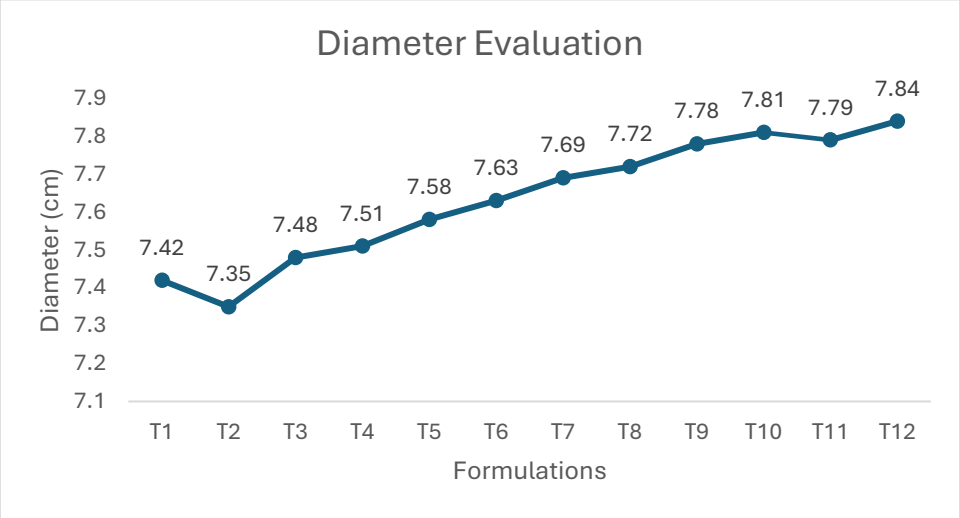


Figure 4: Comparison of diameter

The greatest markings in the overall rating that T12 collected were for the diameter, indicating that the polymer network was more resistant to contraction when drying. This may be important for dose and coverage consistency due to the effect of uncontrolled shrinkage on film area and local distribution of active material. However, bigger doesn't always mean better. It is desirable that the desired result is reproducibility based on the mould dimensions rather than the maximum size. The graph should then be reported as percentage shrinkage (when the original measurements are available) and interpreted against the casting diameter used. Absolute diameter alone would not necessarily represent good dimensional stability, whereas such normalization would offer more evidence.

pH and prospective skin compatibility

The pH values ranged from 5.21 to 5.79, with T12 reaching 5.79. This small difference suggests that polymer composition did not have a great influence on the acid-base stability. The pH values of all formulations were in the range considered to be of mild acidity that corresponds to the pH of facial cosmetics. Lambers et al. (2006) noted that the natural moisture barrier has, on average, a pH of less than 5 and that acidity is beneficial to resident flora and barrier function [18]. T12 in the present work was comparable to that average but was within the range of values generally admitted to being physiologically acceptable, namely 4.5-6.0, as reported in the thesis.

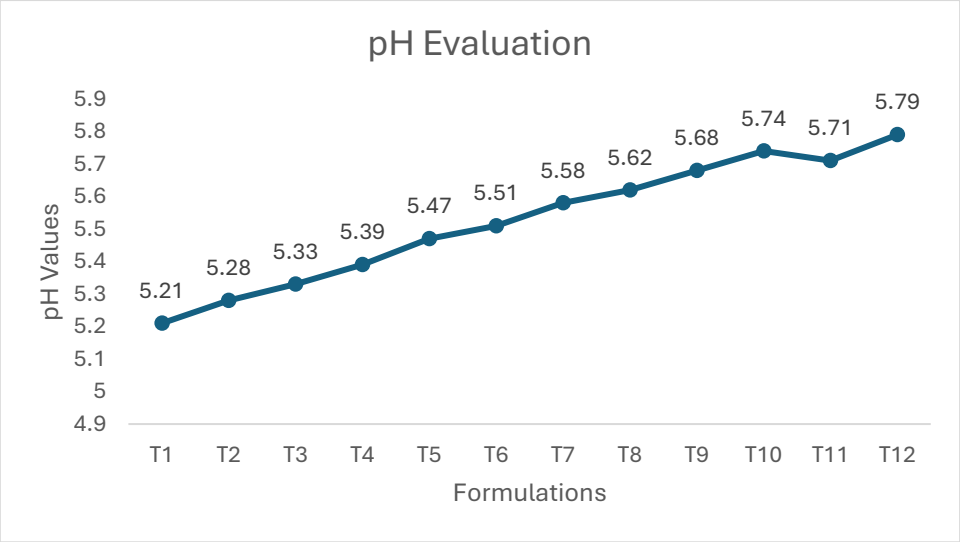


Figure 5: Comparison of pH

The result is favourable for compatibility, but it doesn't prove that there is no irritation. Tolerability is also pH-independent and may be affected by botanical constituents, residual extraction solvent, impurities, and/or some combined effects of these that may occur with prolonged occlusion. The finding should be reported as physicochemical compatibility, not clinical safety, accordingly. The pH trend in the formulations could be attributed to changes in the proportions and interactions of PVA, gelatin, aloe vera, and neem constituents with the formulations. The differences were fairly small, suggesting that the botanical extracts did not stray significantly from the range of relevance to skin.

Freeze–thaw stability

Weak formulations were differentiated from structurally resilient formulations using freeze–thaw testing. Slight shrinkage occurred in T1, slight cracking occurred in T2, and minor deformations occurred in T3. No significant changes were observed in T4-T8, and T9-T12 were unchanged, rated highly stable.

Table 3: Freeze Stability Results

Formulation	Before Test	After Test	Observation
T1	Uniform film	Slight shrinkage	Moderate stability
T2	Uniform film	Slight cracking	Moderate stability
T3	Uniform film	Minor deformation	Moderate stability
T4	Uniform film	No major change	Stable
T5	Uniform film	No major change	Stable
T6	Uniform film	No major change	Stable
T7	Uniform film	No major change	Stable
T8	Uniform film	No major change	Stable
T9	Uniform film	No major change	Highly stable
T10	Uniform film	No major change	Highly stable
T11	Uniform film	No major change	Highly stable
T12	Uniform film	No major change	Highly stable

The progressive resistance to temperature stress is in agreement with the suggested contribution of increased intermolecular forces and water retention in the optimized polymer network. Freeze–thaw cycling can lead to ice-crystal growth, water migration, local polymer concentration, and mechanical

disruption. A network architecture that determines the swelling and structural response of hydrogels and the sensitivity of PVA hydrogel properties to freeze–thaw conditions Waresindo et al., 2023 have been stressed. The lack of change in T12's appearance is then evidence of some physical stability at this early stage [19, 20].

This result cannot, however, be interpreted as confirmation of the shelf-life. There is no mention of cycle number, freezing and thawing temperatures, phase durations, or quantitative measurements after the cycle in the thesis. Additionally, stability in the visual sense does not preclude any chemical change or loss of antibiotic properties. Thus, it can be concluded that T12 had a better tolerance to the short-term temperature stress when applied than the previous formulations. Accelerated and real time stability studies are still required, along with pH, moisture, mechanical stability, phytochemical, and microbiological tests and measurements.

Antibacterial activity against Cutibacterium acnes

The antibacterial activity increased progressively with the formulations, with T1–T4 having relatively small inhibition zones and later formulations having larger inhibition zones. T12 exhibited the maximum zone of inhibition (17.2 mm); thus, it was ranked as the most effective in antibacterial activity.

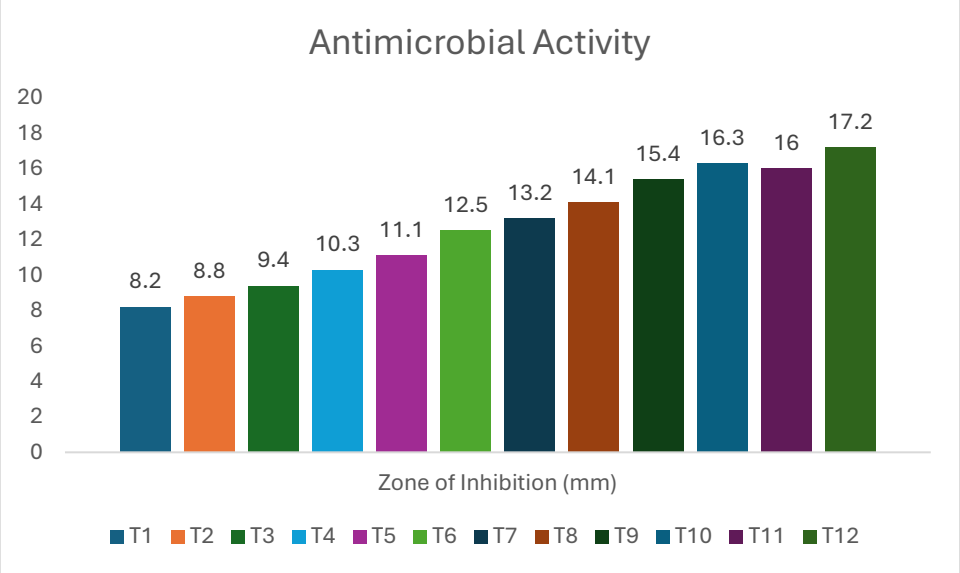


Figure 6 Antibacterial Activity

Optimized formulations show that the higher response could be due to better release and diffusion of botanical constituents in a polymer matrix to which water has been added. Neem has antimicrobial and anti-inflammatory properties due to the presence of limonoids, flavonoids, and tannins, and Aloe Vera has anthraquinones, saponins, flavonoids, and phenolics with reported biological properties. Hamman (2008) and Gupta et al. (2021) have reported bioactive components and antimicrobial activity of Aloe vera and based on those findings, explained the activity from the combined formulation. This observed inhibition is therefore biologically feasible [16, 21, 22].

However, the strength of the antimicrobial agent affects agar diffusion, as do solubility, the molecular size of the sample, its concentration, and diffusion through the agar. If the zone is larger, it does NOT necessarily mean that there will be more bactericidal activity. In addition, the thesis does not provide the results of measurements with a blank polymer film, control without neem, control without Aloe, control without the extraction solvent, and the marketed comparator. The omissions exclude separation of the effects of neem, Aloe vera, and the polymer matrix. The lack of minimum inhibitory concentration and viable-count assays also indicates that the data do not provide evidence for bactericidal activity but only growth inhibition.

The Zone of 17.2 mm is still within the range of the thesis reported by Gupta et al. (2021) for Aloe vera containing biomaterials (12- 18 mm). This comparison indicates the plausibility of the result and should

not be interpreted as proof of equivalence, as strains of microorganisms, density of inoculum on the plate, composition of the agar, loading of samples, and incubation conditions may vary. The most defensible conclusion from the study was that under the assay conditions used, T12 had some interesting in vitro anti-C. acnes activity.

Integrated selection of the optimized formulation

The ranking gradually increased from T1 to T12 within the framework of the integration. T12 was found to be the best performer on all the evaluated parameters, including high stability freeze–thaw behaviour, skin relevant pH, excellent appearance and thickness, and the greatest diameter uniformity; for these reasons, it ranked first. Indeed, T11 and T10 were second and third, respectively, and this was a result of optimization, demonstrating that the performance gradient was not an isolated favourable result.

Table 4 Overall Performance Ranking of Hydrogel Formulations

Formulation	Physical Appearance	Thickness	Diameter	pH	Stability	Antimicrobial Activity	Overall Rank
T1	Poor	Fair	Fair	Good	Moderate	Low	12
T2	Poor	Fair	Fair	Good	Moderate	Low	11
T3	Fair	Fair	Fair	Good	Moderate	Low	10
T4	Fair	Good	Good	Good	Stable	Moderate	9
T5	Good	Good	Good	Good	Stable	Moderate	8
T6	Good	Good	Good	Good	Stable	Moderate	7
T7	Good	Very Good	Good	Good	Stable	Good	6
T8	Very Good	Very Good	Good	Good	Stable	Good	5
T9	Very Good	Very Good	Very Good	Good	Highly Stable	Very Good	4
T10	Excellent	Very Good	Very Good	Very Good	Highly Stable	Excellent	3
T11	Excellent	Excellent	Very Good	Very Good	Highly Stable	Excellent	2
T12	Excellent	Excellent	Excellent	Very Good	Highly Stable	Excellent	1

Another restriction is associated with the ranking method. The categories “poor”, “good”, and “excellent” are a summary of performance, but there are no rules or thresholds reported in the thesis for weighting. Implicit in equal treatment of appearance, diameter, pH, stability, and antibacterial activity is that each of these properties is equally important to product quality. Future optimisation should involve a clear definition of acceptance limits prior to experimentation and a use of a desirability function (transparency) or a model for multiple criteria decision making. Replication level measurements and inferential testing would also be used to differentiate true formulation effects from experimental variation. These enhancements would not necessarily shift the T12’s position, as it showed similar results in all reported domains, but they would help to narrow the choice of formulation for selection, and when making a decision regarding scale up, less would rely on subjective assessment of the product.

This multi-parameter approach is suitable as a topical mask cannot be chosen by just one parameter: its antibacterial activity. Such a product as a highly active formulation that causes skin cracking, shrinking, or irritation will have little use in practice. However, a nice film that doesn't move would not have the desired anti-acne effect. T12 was the one that met both of these competing requirements most balanced.

CONCLUSION

The results of this study indicated the potential of preparing a neem-loaded PVA–gelatin biopolymer peel-off hydrogel mask that possesses acceptable physicochemical properties and in vitro antibacterial efficiency against *Cutibacterium acnes*. The film formation, flexibility, homogeneity, dimensional stability, and also the resistance to freeze–thaw stress was improved. T12 showed the most balanced results among the 12 formulations, delivering great appearance, uniform thickness/diameter, physical stability, and a skin-compatible pH of 5.79. It also generated the biggest zone of inhibition (17.2mm), which demonstrated high antibacterial activity under the experimental conditions applied. However, this activity can't be solely attributed to neem since aloe vera was also included in the formulation, and control tests for the components were not available. The results of the findings recognize T12 not as a clinically proven acne therapy, but rather as a potential therapy. Standardization of botanical extracts and evaluation of mechanical strength, long-term stability, release behaviour, skin permeation, irritation, minimum inhibitory concentration, and clinical effectiveness should be evaluated in future studies.

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