

Formulation and Evaluation of Topical Gel Containing Nepalese Medicinal Plants

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ABSTRACT


Background: The formulation of herbal gel offers a great advantage in non-sticky products as compared to ointments and creams. Herbs are beneficial for human health containing several phytochemicals. Traditionally herbal preparations are used to enhance and facilitate the quality of the skin. Therefore, the aim of the present study was to prepare the herbal gel for the use of skin nourishing and help to cure skin related disease.

Methods: Different crude drugs like Aloe (*Aloe vera*), Neem (*Azadiracta indica*), Turmeric (*Curcuma longa*), Onion (*Allium cepa*), and Basil (*Ocimum tenuiflorum*) are used to formulate the gel. Selection of these herbs are based on literature review on their individual uses in skin related problems. Formulation was developed from polymer base by experimental design based on previous works. Different compositions of herbs were designed for the formulation study. The water extract of the herb was prepared individually and later on mixed with the gel base ingredients containing carbopol 934, methylparaben and propyl paraben. The gel is subjected to evaluation of organoleptic properties and consistency, pH, and spreadability. Disk diffusion method was used to evaluate the antimicrobial property of the extract. DPPH method was used to determine the antioxidant property of extracts.

Results: The average pH values of the formulations were 6.12 and 5.91, similarly spreadability of the two formulations F and S were 9.223 and 9.336 respectively. All the parameters were within the range. The antimicrobial study showed that both of the formulations have strong antimicrobial property to *E.coli*.

Conclusion: The study concludes that the developed formulation of different concentrations are safe and within the range of standard. Therefore, we can say that our formulation product is one of the good natural gels which can be considered for the antioxidant benefit.

Keywords: Herbal Gel, Formulation, Antimicrobial, Antioxidant, Skin Benefit

Access this article Online		Article Info.	
QR Code	How to cite this article in Vancouver Style?		
Scan Me	Journal of Karnali Academy of Health Sciences. 2021; 4(2)		
	Received: 2022	Accepted: 2022	Published Online: 30 December 2022
	Source of Support: No external source of support		Conflicts of Interest: None
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INTRODUCTION

Herbal gels are mostly utilized for improvement of skin and protection against numerous microorganisms. To achieve the desired effect, the polyherbal product

contains a variety of ingredients taken from plants. The combination of plant extract helps give multi advantages and solve a variety of skin problems via a formulation. Basically herbs having vitamin C, vitamin K, anti-

microbial properties, exfoliative properties, anti-inflammatory properties all can be incorporated into a single formulation to have a variety of properties and to solve a wide range of skin-related problems. Therapeutic benefits by addition of plant-based active ingredients such as alpha-hydroxy acid, retinoic acid, ferulic acid, ascorbic acid, and coenzyme Q10 are common. These properties from herbal extracts inhibit free radical formation and prevent skin ageing¹. The usage of the plant extracts included in the formula have benefit to skin biology, which slows skin aging by decreasing wrinkles, blocking UV rays, and preventing collagen breakdown. Medicinal cosmetics have long been sought after it for their effectiveness and lack of adverse effects that are common in synthetic cosmetics. Extracts are now widely used in manufacturing due to customer concerns about synthetic components and toxic compounds². Onion and aloe are well known for moisturizer, whereas neem, basil and turmeric are taken as antiseptic and anti-inflammatory natural products.

Herbal cosmetics are cosmetic formulations and products that use herbal elements to give defined cosmetic benefits. Natural herbs aid in the preservation and enhancement of human beauty. Many cosmetics are made up of herbal extract for anti-marks, fairness, cleansing and hair care which are very effective³. Hydrogels are water-immobilized gel systems made up of insoluble polymers. Hydrogels are made up of water and a hydrophilic but non-water soluble polymeric material. The dry polymer swells and absorbs liquid when exposed to water. Chemical or physical pressures are used to cross-link the polymer strands. Hydrogels can be classified based on the type of polymer used and/or the cross-linking mechanism⁴. Millions of natural product compounds show many useful medicinal and cosmeceutical properties. There is a wide scope of research on the properties of such plants to be used in cosmetics⁵. Formulation of herbal gel have many advantages including, non-sticky, easily washable, and beneficial for sensitive oil skin⁶.

There is no report on polyherbal gel formulation studies of these herbs particularly on locally available selected plants of Nepal. Therefore, the herbal gel was developed, and evaluated to collect the scientific evidences which can support such prototype in commercial product development.

MATERIALS AND METHODS

Plant materials

The plant parts, based on their traditional usage as folk medicine, were collected from the locality of

Kavrepalanchowk, Siraha, and Kathmandu district, Nepal during the month of January 2022 (Table 1). The herbarium of the plants were prepared and then plants were identified by lead author in his department of the University laboratory. The collected samples were dried in shade and stored at room temperature in polybags before starting the project.

Table 1: List of medicinal plants collected from different parts

Plants	Collection area
Neem (<i>Azadirachta indica</i>)	Siraha
Onion (<i>Allium cepa</i>)	Kavre
Aloe (<i>Aloe vera</i>)	Kathmandu
Basil (<i>Ocimum tenuiflorum</i>)	Kathmandu
Turmeric (<i>Curcuma longa</i>)	Kavre

Preparation of plant extract

The dried plant materials were powdered with the help of grinder and prepared into 80 mesh size. Total 50 gm of each powder is boiled in the 250 ml of water for 30 min. Extracts were filtered, and then concentrated under reduced pressure in rotary evaporator (Buchi R-215, Switzerland), 75-90 RPM, under 100 mbar pressure maintaining 40°C temperature of water bath and stored at 4 °C for further use.

Preparation of Gel

Methyl paraben and propyl paraben were carefully dissolved in distilled water by continuous heating and stirring until they dissolved as described previously with some modifications⁷. Carbopol was sieved and added to the above cooled solution and few drops of Triethanolamine were added to neutralize the solution. The solution was left whole night to soak and swell with carbopol. glycerin was added and volume was making up with distilled water. Firstly gel base was prepared with different concentration from which best one was selected (Table 2). Amount of glycerin used in the formulation was finalized after the physical inspection of sticky performance of the product. Herbal decoctions and essential oil were dispersed separately in PEG 400, added to gel base and mixed well. All the chemicals used in the experiments were lab grade.

Physical parameters

pH: The pH was measured by using the digital pH meter. Homogeneity: Visual observation technique was implemented to observe the homogeneity.

Spreadability: Slip and drag method was used to measure spreadability. During the test, a sample was placed in between glass plate and pressed with certain load. Time needed for the complete separation of two

plates were measured. In this case, it was determined by the formula;

Spreadability = $\frac{m \times l}{t}$ Where m = weight tied to the upper slide, l = length of the glass slide and t = time taken in seconds.

Antimicrobial Property

For the anti-microbial test *E.coli* was collected from Dhulikhel Hospital, Kathmandu University teaching hospital and was cultured. The bacteria were grown in nutrient broth at 37 °C and were maintained at 4 °C. Antimicrobial property of the extract, and formulated gel were evaluated after dissolving them in 1% (v/v) DMSO. The whatman filter paper with diameter of 6mm, that was made by using paper punching machine which were dipped in extracts. The petri plates with MacConkey agar and the subcultures, to which the filter disc with extract were placed and incubated for 24 hour at 37°C. The result was recorded by using Vernier Caliper for measuring Zone of Inhibition. DMSO at the concentration of 1% was considered as negative control. For comparison 30mcg ciprofloxacin disc was taken as a standard⁸. The assay was repeated trice.

Antioxidant Property

DPPH method was used to determine the antioxidant property of extract. Ascorbic acid was dissolved in methanol for the preparation of stock solution. A stock

solution of DPPH (0.04 mg/mL in methanol) was prepared. The concentration of herbal extract solution was 50 gm/250 mL. 1mL was taken in a volumetric flask from the stock solution and diluted by the same solvent up to 100 ml. This is stock solution was taken and diluted to 100 ml using the same solvent. Then 5, 10, 15, 20, and 25 mL of the stock solution was taken in different test tubes, whose concentration was then 10, 20, 30, 40, and 50 µg/mL, respectively. Freshly prepared DPPH solution 5 ml (1.3 mg/mL) was added in each of these volumetric flasks containing onion and basil extracts and kept in dark for 30 min, the absorbance was taken at 517 nm using a spectrophotometer. Different sample concentration was used in order to obtain antiradical curves for calculating IC₅₀. IC₅₀ was calculated from % inhibition. Ascorbic acid was used as a reference standard and dissolved in methanol to make the stock solution with the same concentration of onion and basil extracts. Control sample was prepared without any extracts, reference was ascorbic acid, and methanol was blank. Percentage of free radical scavenging was measured using the following equation^{9,10}.

$$\% \text{ inhibition} = \frac{(A_{\text{control}} - A_{\text{sample}})}{(A_{\text{control}})} \times 100$$

A control = absorbance of DPPH alone

A sample = absorbance of DPPH along with different concentrations of extracts.

Table 2: Poly herbal gel base formulation

Ingredients	F1(1%)	F2(1.5%)	F3(2%)	F4(1%)	F5(2%)	F6(3%)
Carbopol	1.00	1.00	1.00	1.00	1.00	1.00
Methyl paraben	0.20	0.20	0.20	0.20	0.20	0.20
Propyl paraben	0.02	0.02	0.02	0.02	0.02	0.02
Propylene glycol	1	1	1	1	1	1
PEG 400	1	1	1	1	1	1
Glycerin	10	10	10	10	10	10
Neem extract	1.0	1.5	2	-	-	-
Onion extract	1	1.5	2	-	-	-
Aloe extract	1.0	1.5	2	1.0	1.5	2
Basil extract	-	-	-	1.0	1.5	2
Turmeric extract	-	-	-	1.0	1.5	2
Mentha oil	-	q.s	-	-	-	-
Triethanolamine	q.s	q.s	q.s	q.s	q.s	q.s

RESULT

Physical characteristics

The gel was semisolid with a characteristic odour. After addition of herbal extract, it showed green color from white. pH-value was accordance to the skin (Table 3).

Antimicrobial Property

The antimicrobial results of the studies showed both formulations were with better zone of inhibition. The antimicrobial activity shown by 2% concentration of herbal extract was with great zone of inhibition i.e 12.75 ± 0.25. (Figure 1).

Antioxidant Property

This study concluded that these formulations show antimicrobial property and can be used safely on the human skin. The DPPH Assay was easy and rapid procedures to determine antioxidant activity of the

tested extract for this study which showed the basil has high antioxidant property which is beneficial for human use (Figure 2).

Table 3: Physical characteristics of the poly herbal gel with different formulation concentrations showing the appearance, color, odour, pH, and Spreadability

Formulations	Appearance	Color	Odour	pH	Spreadability
F1(1%)	Semisolid	Green	Characteristic	6.17	9.7402
F2(1.5%)	Semisolid	Green	Characteristic	5.99	9.168
F3(2%)	Semisolid	Green	Characteristic	6.2	8.761
F4(1%)	Semisolid	Green	Characteristic	5.79	9.469
F5 (1.5%)	Semisolid	Green	Characteristic	5.83	9.328
F6(2%)	Semisolid	Green	Characteristic	6.11	9.213

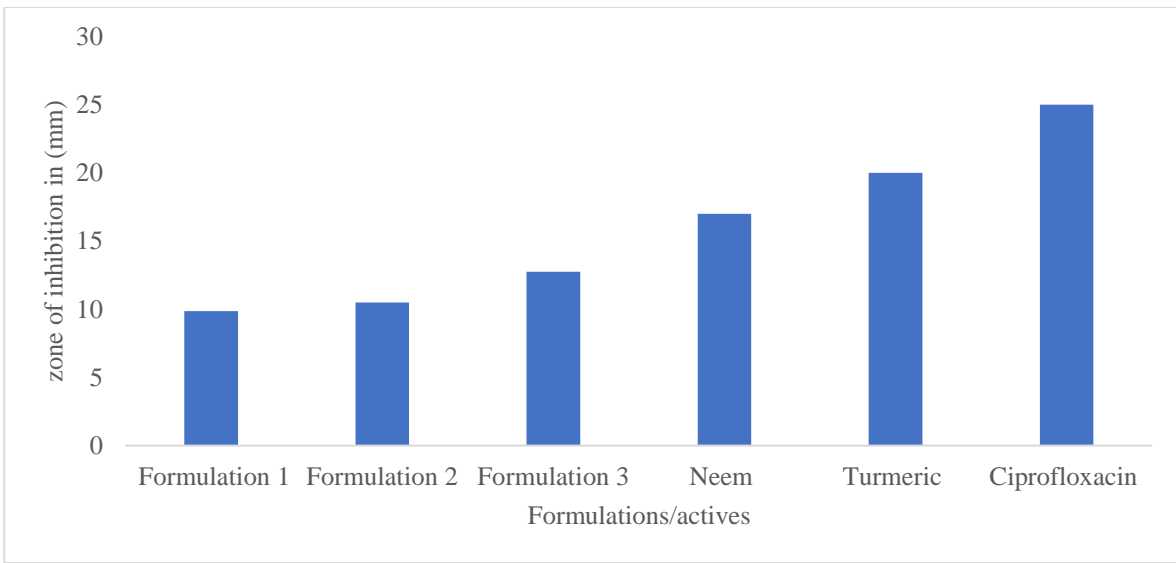


Figure 1: Study of antimicrobial property from zone of inhibition

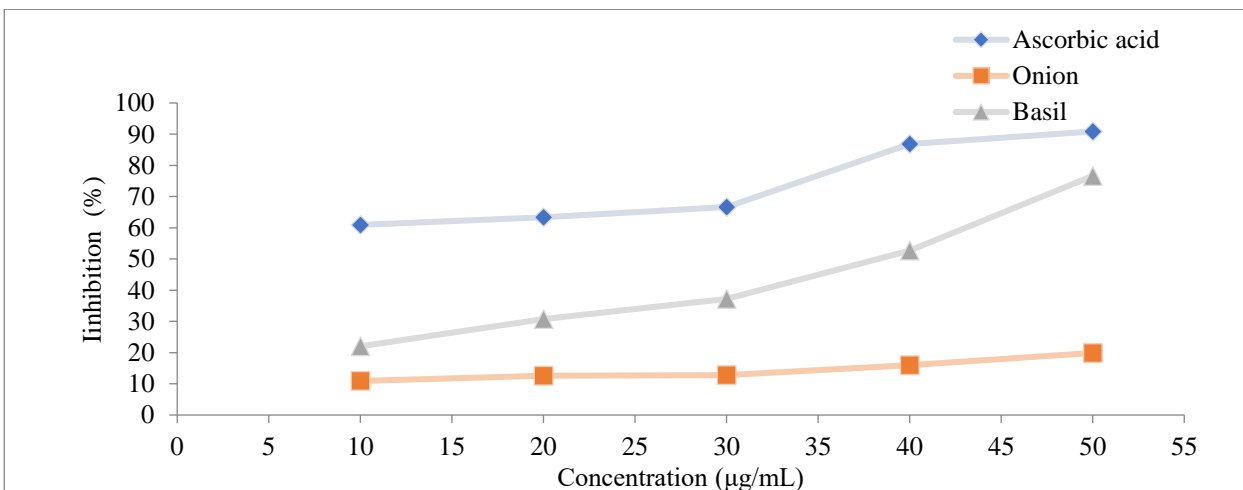


Figure 2: Antioxidant property of herbal extracts in comparison with Ascorbic acid

DISCUSSION

Alkaloids terpenoids, and tannins are useful for the antimicrobial property. Flavanoids which are considered

one of the best metabolite to prevent wide range of diseases are also used to protect the plant against microbial attacks and predation by animals^{11,12}. Basil extract shows the good antioxidant property as well as antimicrobial property. Basil essential oils and their principal constituents were found to exhibit antimicrobial activity against a wide range of Gram-negative and Gram-positive bacteria, yeast, and mold¹³. IC₅₀ of the extract was compared which showed 1.47 µg/mL, 34.60 µg/mL, 196.41 µg/mL of ascorbic acid, basil, and onion respectively which is quite similar with the previous findings¹⁴. Previous pharmacological data shows that several herbal gels are used in formulations and compatible with the skin. Though the excipients are same, the spreadability of the formulations were variable due to the presence of various class of plant secondary metabolites in different concentration. Similarly, due to the combination of various excipients with the herbal extracts, antagonistic effect may be exposed by medicinal plants. The topical formulation from are considered better if they have such strong scientific evidence. Gels are better options for their lower dermal irritations, less mechanical abrasion, and more accepted appearance for using compared to other topical dosage forms¹⁵. Skin beneficial constituents of turmeric a lipophilic polyphenol substance curcumin¹⁶, and phenolic and sulfur-containing compounds of

onion; cysteine sulfoxides, quercetin, and quercetin glucosides¹⁷, are the major bioactive constituents. Thus, herbal gel containing bioactive compounds and polymers can be used for the skin beneficial purpose. Previous studies revealed that the developed single herbal formulation was comparatively better while compared to various pharmacological properties¹⁸. Basil was detected as a one of the best antioxidant agent which have health promoting property.

CONCLUSION

The study concludes that the developed formulation of different concentration F1, F2, F3, F4, F5, and F6 are comparatively safe. According to efficacy of turmeric and basil, the F5 formulation product is one of the acceptable gel for further application.

Acknowledgment: We would like to express our deep gratitude to Department of Pharmacy, Kathmandu University for supporting this project. Thanks goes to the Kathmandu University Integrated Rural Development Project (KU-IRDP) funded by Korean International Cooperation Agency (KOICA) Nepal for the Research and Business Development (R&BD), herbal health program.

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