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FORMULATION, EVALUATION & COMPARATIVE ANTI-BACTERIAL STUDY OF POLYHERBAL SOAP WITH MARKETED SOAP

Sohil Bendre¹, Milind Buradkar², Twinkle Baisware³, Manjiri Bharasakare⁴, Ashwini Armarkar⁵

Shree Sainath College of Pharmacy, Dawalameti, Nagpur

Abstract: The present work involves the formulation, evaluation & comparative anti-bacterial study of polyherbal soap with marketed soap. The herbal soaps were formulated using *Azadirachta indica, Curcuma longa Linn.* And *Sapindus mukorossi* extract. They were extracted by the Soxhlet extraction method using ethanol as solvent. The phytochemical screening of all the extracts showed the presence of various phytochemical constituents like saponins, phenols, alkaloids, flavonoids, tannins and steroids. Six different formulations with varying concentrations of plant extracts and ingredients were formulated using factorial design. Physico-chemical evaluation parameters like pH, moisture content, % alcohol insoluble matter, foam height, foam retention and % free alkali gave satisfactory results for formulated soap with good appearance, uniform colour and odour. The polyherbal soap (F3) and marketed soap were evaluated for antibacterial study against E.coli & B.subtilis strain. The zone of inhibition for E.coli at 100mg/ml concentration were found to be 0.3 cm & 0.4 cm and that of B.subtilis were found to be 0.7 cm & 0.8 cm, respectively. Therefore, the formulated polyherbal soap (F3) showed pre-eminent antibacterial properties as compared to marketed soap and the phytochemicals present in the herbs strengthens, nourish and moisturize the skin.

Keywords: Antibacterial study, Factorial design, polyherbal soap, E. coli

Graphical abstract:



1. INTRODUCTION

The term "Herbal Cosmetics" refers to products that have been made with one or more herbal substances that are employed solely to deliver specified cosmetic benefits after a base of other legal cosmetic ingredients.

A drug or medications are contained in the herbal soap formulation. Antibacterial and antifungal medications are primarily created from plant materials including leaves, branches, roots, and fruits, and they're used to heal wounds, fend against illness, and enhance general health. This product is available in a range of dosage forms, like ointment, lotions and creams as well as gels, solvent extracts, and ointments, and has antibacterial qualities that are applied topically. Various skin conditions have been treated using the qualities of various creams and soaps. Fungi, staphylococcus aureus, and streptococcus species are the main causes of skin infections. In traditional medicine, juice and extract from plant leaves are administered topically as antibacterial and anti-inflammatory medicines to treat skin conditions such eczemas, ringworm, and itch.[2]

Soaps are one of the latest cosmetics for protecting and enhancing skin vibrancy. As a result, the Ayurvedic herbs listed under varnya are chosen for formulation. Galen, a physician, is credited with bringing up the idea of using soap for bathing and body preservation for the first time. However, the skin is commonly irritated and dried out by modern chemical soaps.[3]



Figure 1: Polyherbal soap

Herbal soaps are typically handmade and prepared with solely organic components, which are excellent for the skin and the environment. Some herbs are effective at organically colouring your items. Some plants are excellent for unwinding and relieving tension. Other herbal ingredients will assist the skin by decreasing acne or relaxing inflammation, for example. Additionally, certain plants contain a range of healthful minerals and vitamins that are highly advantageous. Organic natural ingredients used in herbal soaps help to smooth and rejuvenate the skin. Even the scent of herbal soap calms the mind without changing the surrounding environment. They won't include any synthetic flavour or colour. Chemical soaps also include animal fat and lack the essential oils that give them their natural and enticing aroma.[4]

2. REVIEW OF LITERATURE

- a. Francisco José González-Minero* et al., 2020- Rosemary is a plant native to the Mediterranean region that has been distributed throughout the world. It is a member of the Labiatae family, which includes many genera that are frequently used in cosmetics because of their high levels of antioxidant compounds, including Salvia, Lavandula, and Thymus. We assess the uses of rosemary in cosmetics while taking into consideration its preservation properties, the products it is employed in, its toxicological safety, as well as its current uses or potential future roles in topical preparations, in accordance with most recent and continuing studies.[5]
- b. Blessy Jacob* et al., 2019- The majority of industrial soaps and detergents contain chemicals that might harm skin. An excellent substitute is to use organic, herbal cleaning soap and detergents. They promote rest and stress relief. In this study, cleaning soap was made with herbal ingredients and tested for a number of physicochemical characteristics. Therefore, this strategy can be examined for treating contamination and nourishing skin due to its greatest influence on the skin and appropriate parameters.[4]
- c. Pandey Shivanand* et al., 2010-Herbal ingredients are all-natural and have no negative effects on the human body; instead, they bolster it with vitamins and other beneficial minerals. Natural substances such as ashwagandha, sandalwood (Chandan), saffron (kesar), and many more are used to herbal cosmetics together with nutritious sand and other essential components.[1]
- d. Ashlesha Ghanwat et al., 2020- *Azadirachta indica, Ocimum tenuiflorum, Sapindus mukorussi*, and powdered *Acacia concinna* leaf and bark extracts were utilised to create a herbal cleaning soap and hand sanitizer. Ayurvedic cosmetics are often known as natural cosmetics since the natural components of the herbs have no negative effects on the body.[2]

3. OBJECTIVES

Nowadays, various cosmeceuticals companies formulate soap preparations using lots of chemical preservatives and surfactant that cause skin irritation to the skin. Therefore, to minimize those side effects, we have formulated polyherbal soap of natural origin. The final aim of this research work was to develop and assess herbal soap employing methanolic extracts of three herbs of significant dermatological and ethnic value in Ayurveda, namely *Azadirachta indica, Curcuma longa Linn.*, and *Sapindus mukorossi* extract. The phytochemical screening of all the extracts was performed to determine the presence of active phytoconstituents that are present in the plant responsible for the therapeutic activity.

4. MATERIAL AND METHODS

4.1 Material

Powder of Neem, Reetha & Turmeric were purchased from MANAS AYURVEDA, located at PLOT NO. 09, "GOKUL" GHATATE CHOWK VIP ROAD, CIVIL LINES, NAGPUR. Sandalwood oil and rosemary liquid extract were purchased from GYATRI TRADERS, PLOT NO. 68, VAISHALI NAGAR, NAGPUR, 440017. They were made by BRM CHEMICAL, FLAT NO. 2 MCD BUILDING FLOOR, SHAKTI NAGAR, DELHI, 110007. Glycerine base was purchased from amazon.com.

4.2 Plant Profile

4.2.1 Neem

It consists of all arial part of known as *Azadirachta indica*, family maliaceae. It should not contain less than 1.0% w/w of Rutin. It's order Sapindales , genus azadirachta & kingdom plantae.

4.2.2 Turmeric

Turmeric consists of dried, as well as, fresh rhizomes of the plant known as *Curcuma longa Linn*. (*C. domestica*), belonging to family Zingiberaceae. It contains not less than 1.5 per cent of curcumin. It's order zingerberales , genus curcuma & kingdom plantae.

4.2.3 Sandalwood

Sandalwood describes a number of small tree species in the genus Santalum, which occur in south and Southeast Asia, Australia and the Pacific. The trees produce an oil, deposited in the heartwood, which when extracted by distillation is used in the international perfumery market. Its biological name is *Santalum album* belonging to family Santalaceae. Its order is Santalales, genus Santalum & kingdom plantea.

4.2.4 Rosemary

Rosemary is an evergreen plant (Fig. 14.39) about 2 meters in height. It bears blue-coloured racemes. The leaves of the plant are about 2.5cm. It is cultivated in Mediterranean basin and India. Its biological name is *Rosemary officinalis* belonging to family Labiatae. Its order is lamiales, genus salvia & kingdom plantae.

4.2.5 Reetha

Sapindus mukorossi Gaert, a member of the family Sapindaceae, is commonly known by several names such as soapnut, soapberry, washnut, reetha, aritha, dodan and doadni. It is a deciduous tree widely grown in upper reaches of Indo-Gangetic plains, Shivaliks and sub-Himalayan tracts at altitudes from 200 m to 1500 m. Its order Sapindales , genus sapindus (Soapberry) & kingdom plantae.

4.3 **Procedure For Extraction**

20g of coarsely powdered samples of neem, turmeric, and reetha were kept for maceration (7 days) on rotatory flask shaker using ethanol as a solvent of choice for extraction. The alcohol extract was then filtered using Whatman filter paper. The filtrate was distilled out by using Soxhlet apparatus to obtain concentrated extract.[10]

4.4 Formulation Procedure For Polyherbal Soap Using Factorial Design

The extracts of *Azadiracta indica, Curcuma longa, Santalum album, Sapindus mukorossi* and *Rosmarinus officinalis* were added to a basic glycerine soap that also consists of coconut oil, sunflower seed oil, olive oil, glycerine, honey, and water. 1.2g of stearic acid, 0.75g of soft paraffin, and 5ml of ethanol were weighed. After melting 45g of glycerine basic soap, 1.2g of stearic acid, 0.75g of soft paraffin, and 5ml of ethanol for a basic soap, 1.2g of stearic acid, 0.75g of soft paraffin, and 5ml of ethanol were mixed. After melting the solution, the extract were added and stirred continuously for 30 minutes to make the mixture uniform. The slurry was put into a mould and given time to solidify.[2]

]

Factorial	Neem	Turmeric	Sandalwood	Reetha	Rosemary
Design					
F1	1	2	1	3.2	2
F2	2	1	2	3.2	2
F3	2	2	2	3.2	2
F4	2	2	1	3.2	2
F5	1	1	2	3.2	2
F6	1	2	2	3.2	2

Table 1: Factorial design of six different formulations of polyherbal soap



Figure 2: Six different formulated polyherbal soap

5. ASSESSMENT OF PHYSIOCHEMICAL PROPERTIES OF POLYHERBAL SOAP

5.1 **Physical characteristics of formulated soap are examined:**

Table 2: Physical properties of formulated soap

Sr. No	Parameter	Test	J
1	Colour	By visually	
2	Odour	By smell	
3	Texture	By touch	
4	Clarity	By visually	[2

5.2 Determination of pH:

In 100 mL of water, 5 g of soap was dissolved. Using a digital pH metre (Systronics Digital pH metre MK VI), The pH level of the solution of soap was measured. pH for the commercial product was measured separately.[20]

5.3 Determination of percentage free alkali:

150 ml of filtered water was added after 10g of test soap had been measured with a digital scale (Essae Weighing balance, model DS-852G). Following that, the beaker was put in a water bath (a SISCO water bath) and boiled for 30 minutes while experiencing reflux. The volume was raised to 250 ml in a beaker. The indicator phenolphthalein, 1 ml, was added. As soon as possible, it was titrated with 0.1 M HCl till the solution became colourless. [20]

5.4 Determination of foam height:

25 ml of sterilised water were used to dissolve a 0.5g sample of soap. It was diluted with water to a volume of 50 ml before being put into a 100 ml measuring cylinder. It took 25 strokes. It was permitted to stand

until an aqueous volume of 50 ml has been measured. We measured the height of the foam above the aqueous volume.[20]

5.5 Determination of foam retention:

The 1% soap solution was made. In a graduated measuring cylinder with a volume of 100 ml, 25 ml of 1% soap solution was taken. Ten times the cylinder shaken with the cover off. The amount of time it took for the foam to vanish was noted. [20]

5.6 Determination of alcohol insoluble matter:

Under the test conditions, the majority of the alkaline salts, such as borates, carbonates, silicates, and phosphates, talc along with sulphates and starch, are insoluble in alcohol. Alcohol insoluble matter also includes substances that are insoluble in alcohol under other conditions. 50 ml of warm ethanol was added to a conical flask containing 5g of soap sample, and the flask was vigorously shaken until the sample was completely dissolved. The solution was passed through a tare filter paper along with 20 cc of warm ethanol and dried at 105°C for an hour. It was observed the dried paper's weight.

% Alcohol insoluble matter = Weight of residue/ Weight of sample \times 100 [20]

5.7 Determination of Sensitivity:

Through patch testing, it was tested. Apply the product to a 1-cm-long area of skin; if there are no rashes or swelling, it was regarded to be non-sensitive.[21]

5.8 Determination of Irritation:

Applying a product to the skin for 10 minutes is how it's done. It was regarded as a non-irritation product if there was no irritation.[21]

5.9 Moisture content:

The total amount of water in soap was determined using the moisture content. To calculate the amount of moisture in 5g of soap, the weight was noted as wet weight or initial weight. In a hot air oven set to 100 to 115°C during one hour, a sample was dried. Once the sample had cooled, it was weighed. The sample's dry weight is indicated by this measurement. The formula shown below was used to calculate the moisture content.

% Moisture content = Initial – Final weight/ Final weight \times 100[20]

5.10 Emolliency test:

The occlusiveness of soap compositions was assessed by an emolliency test. After each soap formulation's 2g portion was put on the surface of white sheets of paper across an area of around 5 cm² and let to stand on the laboratory shelf for 24 hours, the degree of translucency was evaluated into a three-level rating of mild, moderate, or strong translucency.[19]

6. RESULT AND DISCUSSION

6.1 Phytochemical Screening

Table 3: Phytochemi	cal screening of extracts
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Chemical constituents	Neem extract	Turmeric extract	Sandalwood extract	Reetha extract	Rosemary extract
Carbohydrate	+	+	+	+	+
Protein	+	+	+	+	+
Amino acid	+	+	+	-	+
Steroid	+	+	+	+	-
Glycoside	+	+	+	+	+
Flavonoid	+	+	+	+	+
Alkaloid	+	+	-	+	+
Tannin	+	+	+	+	+

6.2 TLC Profile

TLC Profile	Neem	Turmeric	Sandalwood	Reetha	Rosemary	
	extract	extract	extract	extract	extract	
Stationary	Silica Gel G	Silica Gel G	Silica Gel G	Silica Gel G	Silica Gel G	
Phase						
Mobile Phase	Isopropanol:	Chloroform:	Toluene: Ethyl	Chloroform:	Ethyl Acetate:	
	Hexane	Methanol	Acetate (93:7)	Glacial	Toulene (5:95)	
	((2:17.5)	(98:2)		Acetic Acid:		
				Methanol:		
				Water		
				(64:32:12:8)		
Spraying	Iodine	Visible Light	Vanillin Sulphuric	Vanillin	Valinin Reagent	
Agent	Chamber		Acid	Sulphuric	_	
				Acid		
RF Value	0.52	0.68	0.9	0.95	0.68	
Reference RF	055	0.75	0.88	0.76	0.63	
Value						

Table 4: TLC profile of extracts

6.3 Anti-bacterial Study

6.3.1 Comparative Anti-Bacterial Study Of Polyherbal Soap (F3) With Marketed Soap

Bact	erial	Antibiotic		Polyherbal	Soap	Marketed S	oap
Strai	in 🦳	(Azithromy	cin)	F3 Formula	tion		
		50mg/ml	100mg/ml	50mg/ml	100mg/ml	50mg/ml	100mg/ml
E.CO	OLI	0.3cm	0.5cm	0.2 cm	0.3 cm	0.3 cm	0.4 cm
B. SI	UBTILIS	0.7 cm	0.9 cm	0.6 cm	0.7 cm	0.8 cm	0.8 cm

Table 5: Zone of inhibition of certain strain of bacteria i.e., b. subtilis (gram +ve) and e. coli (gram -ve)

6.3.2 Anti-Bacterial Study Of Six Different Formulated Soaps

Table 6: Zone of inhibition for six different soap formulations at various concentrations

	SOAP CONCENTRATION				
DIFFERENT	C1	C2	C3	C4	
FORMULATED SOAP	10mg/ml	20mg/ml	30mg/ml	40mg/ml	
		ZONE OF INH	IBBITON		
SOAP F1	0.1cm	0.1cm	0.12cm	0.13cm	
SOAP F2	0.11cm	0.12cm	0.13cm	0.14cm	
SOAP F3	0.21cm	0.25cm	0.28cm	0.3cm	
SOAP F4	0.18cm	0.17cm	0.19cm	0.2cm	
SOAP F5	0.1cm	0.21cm	0.23cm	0.24cm	
SOAP F6	0.18cm	0.2cm	0.24cm	0.21cm	

6.4 Final Formulation

Table 7: Final formulation of polyherbal soap

Sr.	Ingredients	Quantity	Uses
No			
1	Neem Extract	2ml	Anti-Bacterial
2	Turmeric Extract	2ml	Antiseptic
3	Sandalwood Oil	2ml	Anti-Inflammatory & Fragrance
4	Reetha Extract	3.2ml	Cleanser & Surfactant
5	Rosemary Extract	2ml	Preservatives & Anti-Bacterial
			Activity
6	Stearic Acid	1.2g	Hardening Agent
7	Liquid Paraffin	0.75g	Hardening Agent
8	Glycerin Base	45g	Emollient

6.5 Evaluation Parameters

Table 8: Comparison of evaluation parameter between polyherbal soap and marketed soap

Sr. No.	Evaluation	Observation Results		
	parameters	Polyherbal Soap (F3 soap)	Marketed soap (Santoor)	
1.	Colour	Brownish red	Orange	
2.	Odour	Characteristics sandalwood like	Characteristics sandalwood like	
		odour	odour	
3.	Texture	Soft	Lightly hard	
4.	Clarity	Good	Good	
5.	рН	8.07	9.12	
6.	Percentage free alkali	5.07%	7.17%	
7.	Foam height	6cm	9cm	
8.	Foam retention	9min	14min	
9.	Alcohol insoluble	6%	7%	
	matter			
10.	Sensitivity	No inflammation & rashes	No inflammation & rashes	
11.	Irritation	No irritation	No irritation	
12.	Moisture content	3.12%	2.67%%	
13.	Emolliency test	High translucency	Mild or moderate translucency	

7. CONCLUSION

Polyherbal soap have less side effects as compared to marketed soap because the phytochemicals present in the plants strengthen, nourish, and moisturise the skin, making the herbal soaps significantly superior in antibacterial properties. Some contrast check of polyherbal soap offers optimal or equal outcomes than marketed soap. The quantity of rosemary extract we used in our formula which will act as a preservative and the quantity of reetha extract which produced vast amount of foam and cleansing properties.

But, F3 soap formulation had shown the satisfactory results for antibacterial activity against E. coli. It has optimum foaming property and least wide variety of impurities as per the popular of the Bureau of Indian Standard. The manageable use of the formulated soap in treating skin infections in addition can be explored. Naturally neem, turmeric. sandalwood, rosemary and reetha shows anti-bacterial properties. The other substances used have been tested to be dermatologically tested. Therefore, the formulated polyherbal soap (F3) showed pre-eminent antibacterial properties as compared to marketed soap and the phytochemicals present in the herbs strengthens, nourish and moisturize the skin.

8. CONFLICT OF INTEREST

The authors declare no conflict of interest.

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