



**FORMULATION AND EVALUATION OF HERBAL ANTIBACTERIAL
HANDWASH BY USING METHANOLIC EXTRACT OF ACACIA
AURICULIFORMIS FRUIT**

Avinash O. Maske, S.M.Sonwane, Miss. Priyanka K. Lanje, Dr. P. S. Raghu

Bajiraoji Karanjekar College of Pharmacy, Sakoli, Maharashtra.

* Corresponding Author. E-mail: avinashmaske59@gmail.com

ARTICLE INFO

Key words:

Herbal hand wash,
Earleaf acacia,
antimicrobial activity, agar
plate method

Access this article online

Website:

<https://www.jgtps.com/>

Quick Response Code:



ABSTRACT

A herbal handwash was prepared using extracts of the fruit of *Acaica auriculiformis*. *Acacia auriculiformis*, commonly known as auri, earleaf acacia, earpod wattle, northern black wattle, Papuan wattle, and tan wattle, akashmoni in Bengali, is a fast-growing, crooked, gnarly tree in the family Fabaceae. This study aimed to formulate effective herbal handwash using methanolic extract of *Acacia auriculiformis* fruits with emphasis on safety and efficacy and to avoid the risk posed by synthetic antiseptic. Evaluation of antimicrobial activity against microorganism was evaluated by agar plate method. The results showed the presence of various phytoconstituents and posses good antimicrobial activity against *S.aureus* & *E.coli*. So these work support the incorporation and utilization of herbs in the formulation for better effect & can be used by the healthacare professionals on commercial scale.

INTRODUCTION

Hand hygiene is a vital principle and exercise in the prevention, control, and reduction of healthcare acquired infections. Right hand washing and drying methods stop the chain of transmission of deadly pathogens (from the contaminated surface/site) form hands to other parts of the body. Hand sanitization is the preeminent aid in preventing nosocomial infections caused by different opportunistic microorganisms¹. Plants are the oldest resource of pharmacologically active compounds and have provided human kind with many medicinally valuable compounds from centuries². Plants are rich in a wide variety of secondary metabolites, such as phenolic compounds, tanins, terpenoids, alkaloids and flavonoids which

have been found to have antimicrobial properties. Hence there is an upserge of developing herbal disinfectants and evaluate its efficacy³. Literature survey of this plant shows that it is reported to posse's good antimicrobial properties. The chemical composition of *Acacia auriculiform* was reported to contain saponins, tannins, carbohydrates, anthocyanidins, flavonoids^{4,5}. Plants containing flavonoids and polypeptides used in traditional medicine have been found to be active against wide variety of microorganisms.⁶

The aim of present study was to formulate the herbal handwash by using extract of fruit of *acacia auriculiformis*. Furthermore to evaluate the formulation so

that it meets standard parameters & make easy to use commercially.

Materials & methods

Collection of plant material

The tree of *Acacia aruculiformis* widely found throughout the India. The fresh fruits of *Acacia auriculiformis* were collected from local area Sakoli district-Bhandara & dried and

its authenticity was confirmed by Dr.A.A.Jagia, Head of Department of Botany, Manoharbai Patel science college Sakoli Distt- Bhandara (M.S.)

Preparation of extract

The fruits of *Acacia auriculiformis* was collected, dried & washed with water. The extraction of fruit was carried out by simple maceration process. The fruit was then extracted by placing plant material into methanol for seven days. During this period shaking was done occasionally. After seven days, the liquid was strained and marc is pressed. The expressed liquid was mixed with strained liquid. It was then filtered to make clear liquid extract.

Formulation of herbal hand wash

The hand wash was prepared using 20 ml of methanolic extract filtrate. To this filtrate 8g of SLS, glycerin 40 ml, 0.5 g of methyl paraben, 5ml of rose oil is added and the volume is made up to 100ml with purified water.

Table No. 1 Formulation table of herbal handwash (Quantity)

Ingredients	Quantity taken
Methanolic extract of acacia auriculiformis fruit	20 ml
Sodium lauryl sulphate(SLS)	8 gm
Glycerine	40 ml
Methyl paraben	0.5 gm
Rose oil	5 ml
Purified water q.s.	100 ml

Preliminary Phytochemical Screening

The dried aqueous extract was analyzed for the various phytoconstituents like alkaloids, steroids, saponins, sugar, flavonoids, tannins, glycosides &

mucilage's.^{7,8,9} The concentration of the aqueous extract kept 1mg/ml for this study.

Test for sugar:

1 ml of test sample was taken in a test tube and mixed with 2 drops of Molisch's reagent. To this solution 1 ml of concentrated sulfuric acid was added. A red brown ring appears showing the presence of sugar.

Test for Saponins:

The extract was taken in a test tube and diluted with 2 ml of distilled water. It was shaken for 15 min. A foam layer was obtained at top layer that indicates the presence of saponins.

Test for Steroids:

The extract was dissolved with chloroform and an equal volume of concentrated sulfuric acid was added to the test tube by the sides. The upper layer in the test tube turns into red sulfuric acid layer showed yellow with green fluorescence which indicates the presence of steroids.

Test for Flavonoids:

One ml sample (1mg/1ml) was taken in a test tube and added a few drops of 10% NaOH solution. An intense yellow colour appears in the test tube. After addition of a few drops of dilute acid, it became colorless that indicates presence of flavonoids.

Test for tannins:

One ml of aqueous extract (1mg/1ml) was added to 1 ml of distilled water of a few drops of 5% ferric chloride (dissolved in 90% ethyl alcohol) solution was added. A dark green or blue green colour was formed, which showed presence of tannins.

Test for glycosides:

Crude plant extracts (1mg/ml) was taken in a test tube and added few drops of Molisch's reagent. Mixed it and add 2 ml of concentrated sulfuric acid carefully through the side of test tube. Formation of reddish violet ring indicates presence of glycoside.

Test for alkaloids :

1 ml of extract (1mg/1ml) was taken in a test tube and few drops of Mayer's reagent was added. Cream colour precipitates was formed, which indicates presence of alkaloid.

Evaluation of physicochemical parameters of hand wash

Determination of colour, odor

The color of formulation was determined by the naked eyes & odor was smelled.

Determination of viscosity:

The viscosity of formulation was determined by using digital Brookfield viscometer.

Determination of pH

The pH of prepared formulations was determined by using digital pH meter. The measurement of pH of formulations was carried out in previously calibrated pH meter.

Spreadability

Two glass slides were taken of known dimensions and an excess of gel (about 2g) under study was placed on the ground slide. The gel was then sandwiched between. This slide and another glass slide having the dimension of fixed ground slide. A 100g weighted was placed on the top of the two slides for 5 minutes to expel air and to provide a uniform film of the gel between the slides. Excess of the gel was scrapped off from the edges. The top slide was then separated with friction and the time (in seconds) required by the top slide to cover a distance of 7.5 cm be noted. A shorter interval indicates better spreadability. Spreadability was calculated using the following formula:

$S = ML/T$ Where, S = Spreadability, M = Weight in the pan (tied to the upper slide), L = Length moved by the glass slide T= Time (in sec.) Taken to separate the slide completely each other

Skin Irritation

The formulation was applied on skin and left for 30 min.

Evaluation of antimicrobial activity

The screening of anti-microbial efficacy of the formulated poly herbal hand wash was performed on various micro organisms by using agar plate method as per standard procedure. Four sterile petri plates were taken for testing the anti microbial activity against four different microorganisms, *Bacillus subtilis*, *Staphylococcus aureus*, *Psuedomonas aeruginosa* and *Escherchia coli*. The plates were filled with

nutrient agar solution and allowed for solidification. After solidification the microorganisms from the subculture were inoculated into the nutrient agar media and three cavities were made in it. The first cavity is filled with standard antibiotic amoxicillin, second one with formulated herbal hand wash. It was taken care that sample should be placed at the level of cavity. The plates are placed in incubator at 37°C to test the activity. After 24 hours the plates were observed for the formation of zone of inhibition. From the zone of inhibition the anti microbial activity of formulation is estimated^{10,11,12,13}.

RESULTS

Phytochemical screening of plant extracts

Phytochemical screening of plant extract of *Acacia auriculiformis* was carried out for the presence of various phytochemicals. Results were noted as shown in Table 2

Table No. 2 Phytochemical screening of *Acacia auriculiformis* Extract

Sr. No.	Phytochemical constituents	<i>Earleaf acacia extract</i>
1	Alkaloids	+
2	Tannins	+
3	Saponins	+
4	Steroids	+
5	Flavonoids	+
6	Glycosides	+
7	Carbohydrates	+

Preparation & Evaluation of physicochemical parameters of formulation

The physicochemical characterization was carried out. Parameters like color, odour, appearance, pH, viscosity were tested. The result of these parameters are shown in Table No. 3

Table No. 3 Physicochemical parameters

Formulation	Hand wash
Color	Green
Appearance	Good
Odour	Fragrant
pH	7.1-7.4
Viscosity	54±2
Skin irritation	No

Antimicrobial testing of the prepared formulations

Antimicrobial activity:

The Anti-microbial efficacy of the formulations of herbal hand wash was tested on *Staphylococcus aureus*, *Psuedomonas aeruginosa*, *Bacillus subtilis* and *Escherichia coli* by agar plate technique. From the result of zone of inhibition showed that the prepared hand wash from methanol extract have significant antimicrobial activity.

The data of zone of inhibition of formulations is shown in below table

Table 4: Antimicrobial activities of Polyherbal hand wash formulations

Organism	Zone of inhibition in cm	F
Bacillus subtilus	1.3	3.5
Staphylococcus aureus	2.9	4.5
<i>Psuedomonas aeruginosa</i>	2.1	3.8
<i>Escherichia coli</i>	1.9	4.1

CONCLUSION & DISCUSSION

From the above results it was concluded that methanolic extract of *Acacia auriculiformis* fruit extract shows the presence of various phytochemical constituents. Extract exhibiting good antimicrobial activity against *Bacillus subtilus*, *S. aureus*, *E. coli* & *Psuedomonas aeruginosa*. Furthermore the formulation showed good spreadability, good consistency & no evidence of phase separation. It is possible to develop antimicrobial herbal hand wash with *Acacia auriculiformis* fruit. Based on resultant data obtained from the different evaluation parameters it can be concluded that the prepared formulation was stable & safe to use.

REFERENCES

- Joshi MG, Kamat DV, Kamat SD. Evaluation of herbal handwash formulation. *Natural Products Radiance* 2008;7(5):413-415.
- Ravi, Pratibha, Kolhapure, Evaluation of the antimicrobial efficacy and safety of Pure Hands as a handsanitizer, *Indian Journal of Clinical Practice*, 15(10), 2005,19-27.
- Marjorie,. Plant products as antimicrobial agents, *Clinical Microbiology Reviews* 1999; 12(4):564-582.
- Mandal P, Sinhababu S P, Mandal N C. Antimicrobial activity of saponins from *Acacia auriculiformis*. *Fitoterapia*, 76:462-65,2005.
- Nidhi Sharma, Sumitra Singh, Shailendra K Singh. Review on Phytopharmacological Properties of *Acacia auriculiformis* A. Cunn. ex. Benth. *Inventi Rapid: Planta Activa*, 2016(1): 1-6, 2015.
- Abbiw DK. Useful plants of Ghana- West African use of wild and cultivated plants. Intermediate Technology Publications and the Royal Botanic Gardens; 1990.
- Depkes RI. *Materia Medika*. 6th ed. Jakarta: Ditjen POM; 1995. p. 297-307
- Farnsworth NR. Biological and phytochemical screening of plants. *J Pharm Sci* 1996;55:225-76.
- Harbone JB. *Metode Fitokimia: Penuntun Cara Modern Menganalisis Tumbuhan*. Vol. 2. Bandung: ITB; 1987. p. 6, 49
- Tamilarasi Tand Ananthi T. Phytochemical Analysis and Anti Microbial Activity of *Mimosa pudica* Linn. Res. *J. Chem. Sci.* 2(2); 2012: 72-74.
- Jyothi MJ, AV Praveen Kumar AV, Mohanalakshmi S, Prathyusha S. Formulation and evaluation of poly herbal hand wash. *Int J Pharm.* 2(2); 2012: 39-43.
- Ali H A, Shehab NG, Rasool BK. Formulation and evaluation of herbal handwash from *Matricaria chamomilla* flower extracts. *Int J Res Ayu and Pharm.* 2(6); 2011:1811-1813.
- Minakshi GJ, Kamat DV, Kamat SD. Evaluation of herbal handwash formulation. *Natural Product Radiance.* 7(5); 2008: 413-415.