



FORMULATION AND EVALUATION OF HERBAL SOAP USING EXTRACT OF *Suaeda fruticosa* (Forssk)

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ABSTRACT

Herbal soap is often created by hand using only organic ingredients that are suitable for the environment and the skin. *In this study S. fruticosa*, a fleshy eternal halophytic shrub of Amaranthaceae family were used the objectives of the study is To investigate phytochemical, physiochemical, antimicrobial and antioxidant activities of the herbal formulations methanolic extract of *Suaeda fruticosa* was used with other ingredients. The current study suggested that formulated herbal soap has good texture, has organic odour, pleasant scent, pH of 7.92, good thumb impression effect for bacterial growth, and it have good moisture content. The prepared soap and plant extract exhibit good results against *Bacillus cereus* at a concentration of 150µg/mL (30.97 ± 0.2 and 25.7 ± 0.7). At the maximum concentration of 150µg/mL, the plant extract and herbal soap showed growth inhibition of 34.95 ± 0.72mm and 28.6 ± 0.68mm against *A. niger*, respectively, and 36.9 ± 0.44mm and 23.65 ± 0.15mm against *A. flavus*. Soap shows good result against *A. flavus* while plant extract shows good result against *A. niger* at the concentration of 150µg/ml. As a negative control, DMSO was used. In test tubes used as positive controls (terbinafine), no fungal growth was seen. Plant extract and specially manufactured soap maximum scavenging capacities of 41% and 61% at 150 g/ml concentration. These results showed that the formulated herbal soap is suitable for skin and can be a therapeutic alternative to skin problems.

INTRODUCTION

Hands are the main method of transmission for bacteria and illnesses, maintaining good hand hygiene is the most important precaution to take in order to prevent the spread of harmful germs and diseases (Natarajan *et al.*, 2022). In general, the act of washing one's hands with water, soap, or another liquid is referred to as practicing good hand hygiene (Freitag *et al.*, 2001). Most often, soaps created by process known as saponification, which involves the alkaline hydrolysis of microbial, vegetable, and animal fats and oils. Chemicals in soap can cause a variety of skin conditions such as slowing the skin's natural regeneration process, they block skin pores, restrict cell respiration, and hasten skin aging (Fariha *et al.*, 2025). Herbal soap is often created by hand using only organic ingredients that are suitable for the environment and the skin (Patel *et al.*, 2023).

Because soap can disassemble complex compounds like lipids, colors, germs, and bacteria and prevent them from functioning, it is often used for sanitation and sterilization (Maidin *et al.*, 2020).

Herbal soap preparations are medicines or medications because they include antibacterial and antifungal chemicals that are primarily used to treat wounds, diseases, and promote good health using various parts of plants (Sindhu *et al.*, 2019). Fungi, *staphylococcus aureus*, and *streptococcus species* are the main causes of skin infections. In traditional medicine, juice and extract from plant leaves are applied topically as antibacterial and anti-inflammatory drugs to treat skin conditions such as eczemas (skin patches), ringworm, and pruritus (itchy skin) (Akuaden *et al.*, 2019).

Neem leaf and its extract have anti-inflammatory, antifungal, antibacterial, anti-ulcer, anti-malarial, immune modulatory and anti-carcinogenic properties (Sharma *et al.*, 2015). One of the most popular botanicals is *Aloe vera*. *Aloe vera* improves wound healing, boosts collagen production, and moisturizes the skin (Williams *et al.*, 2012). Furthermore, commercial antibacterial soap is a form of soap that commonly contains the chemicals triclosan, triclocarbon, and chloroxynol (Solanki *et al.*, 2020). According to research, these compounds are no more efficient in neutralizing viruses than any other type of soap or detergent (Sucharita *et al.*, 2022). It's interesting to note that the effectiveness of herbal-based soaps for treating skin conditions is driving up demand for them (Shakkarpude *et al.*, 2020). *Suaedafruticosa* (Forssk) a fleshy eternal halophytic shrub of Amaranthaceae family, on land and in coastal salt marshes are in great numbers (Ksouri *et al.*, 2012). Members of Suaeda widely recognized by usage in traditional medicinal practices (Towhidi *et al.*, 2011). The edible *S. fruticosa* in this situation produces hypoglycemic and hypolipidemic effects (Weber *et al.*, 2007), various plant components have anti-inflammatory, hypolipidemic, and hypoglycemic ability. *S. fruticosafarming* might aid for phytoremediation and recovery of hazardous metal-defiler soils (Bareen and Tahira, 2011) as well as saltiness (Khan *et al.*, 2009). The plant thrives in the 200–400 mMNaCl range and can withstand high salt concentrations (Khan *et al.*, 2000), through securing significant levels of cell's GB, which is distinctive property of the Amaranthaceae family and vacuolar sodium chloride (NaCl)

(Munns and Tester, 2008). Within ideal growing situations, it is reported that halophytes use enzymes and non-enzymatic antioxidants systems work together to keep the amounts of these damaging ROS in cells within a narrow, functionally meaningful range (Jithesh *et al.*, 2006).

METHODOLOGY

We put the required quantity of soap base in a beaker. Adjust and maintain the temperature when heating the soap base in a water bath. Heating causes the soap base to become liquid. Bring the mixture to a boil in a water bath, got the correct mixture at 70 °C using a magnetic stirrer. This mixture was poured into the soap mold. It was kept at room temperature to cool for a few hours. The soap was created.

Methanolic extract and formulated soap

Standard techniques were used in preliminary phytochemical screening to explain the existence of many bioactive components such as alkaloids, saponins, flavonoids, steroids, and terpenoids in methanolic extract (Joseph *et al.*, 2018).

Saponins identification

Frothing test

In 10 mL of dH₂O water, 10 mg of plant was heated for 5 minutes before being filtered. Once more, five ml of filtrate combined with five ml of dH₂O and then agitated till bubble formed. The froth was then supplied with 4 drops of olive oil, mixed vigorously, and allowed to form an emulsion.

Tannins determination

Ferric chloride test:

10 mL of dH₂O and 10 mg of plant crude extract were heated before filtered. Now, 0.1% ferric chloride was gradually added to the filtrate, and the color of the filtrate was checked.

Determination of flavonoids

By using the method described in (Sofowora, 1993), flavanoids were identified in plant extracts and herbal soap. 100 mL of dH₂O were combined with around 50 mg of plant extract before filtering to produce filtrate. A 5 mL diluted NH₃ aqueous solution was combined with around 10 mL of filtrate. Concentrated H₂SO₄ was now added to the mixture drop by drop while allowing for color change. The presence of flavonoids was shown by the yellow coloring.

Testing for alkaloids

To determine alkaloids according to the recommended method (Jeruto *et al.*, 2011). 8 mL of 1% HCl and 0.4 g of plant were combined. After being warmed, the mixture was filtered. Wagner's reagent (I₂+KI) was applied in small amount to 2 mL of filtrate to allow for coloring. Precipitate with a reddish-brown tint indicated the presence of alkaloids.

Coumarins identification

Alkali reagent test:

Coumarins were examined using (Kumah *et al.*, 2022) recommended protocol. A little amount of plant extract about 0.3 gm was collected, covered with filter paper, and then wet with 1 N NaOH. The solution was then warmed in a water bath following that for a short while. After boiling, the filter paper was taken out and examined with an ultra violet-2600 spectrophotometer. Yellow fluorescence's appearance indicated the presence of coumarins.

Test for steroids

100 mg of plant powder and 5 mL of CH₃OH were combined before being filtered. 2 mL of acetic anhydride were added to the filtrate. A few drops of concentrated H₂SO₄ were added to this mixture to allow for color shift. Green colour was used as a signal when steroids were present. And same

procedure was followed for steroids presence in herbal soap.

Test for anthraquinones

Hydrochloric acid test:

Conformation was checked for anthraquinones using the technique narrated in (Oludare *et al.*, 2015). Utilized water bath for boiling and filter a mixture containing around 20 mg of plant and 1 mL of 1% HCl. The filtrate was given a 5 mL C₆H₆ addition, and it was forcefully shaken. The top layer was removed, and 10% NH₄OH was then added. A hue that was pink, crimson, or violet indicated the presence of anthraquinone.

Physiochemical evaluation of herbal soap

Color, smell, and texture organoleptic qualities assessed manually or physically. The following criteria are used to evaluate herbal soap;

Determination of clarity, color, and odor

The determination of clarity, color, and odors is made. Clarity and color were inspected with the unaided eye on a white backdrop. In addition, an odor may be detected (Wongthongdee *et al.*, 2013).

pH

In 10ml of distilled water, 2g of the final soap was dissolved while being agitated until the sample was halved. Used pH meters or pH papers were used to determine the pH (Afsar and Khanam, 2016).

Calculating the Percent of Free Alkali

It was then transferred to a flask and 150 millilitre of filtered ddH₂O was put in. After that, the flask heated approx 30 minutes while maintaining reflux in a water bath (SISCO water bath). 250 ml were included in the beaker. Add 1 ml of the phenolphthalein indicator. As soon as the solution became colorless, titration was done using 0.1 M HCl (Haneefa *et al.*, 2019).

Retaining lather

A 100 ml graduated measuring cylinder was filled with 25 ml of the 1% soap solution. The cylinder was shaken ten times by hands over it. At 1-minute intervals for 4 minutes, the amount of foam was measured (Upadhyay *et al.*, 2021).

Moisture content

5g of soap were measured and either reported as wet weight or beginning weight. Used a hot air oven to dry a sample for one hour between 100 and 1150C. The sample was weighed after cooling. This measurement serves as the sample's dry weight. Using the formula below, obtained the moisture content (Haneefa *et al.*, 2019).

$$\% \text{ moisture content} = \frac{(\text{initial} - \text{Final weight})}{\text{initial}} \times 100$$

Cleaning efficiency by thumb impression test

Environment-exposed hands' thumbs were carefully put on a sterile nutritional agar medium plate while keeping the right spacing. The impressions of one thumb rinsed with the medicinal soap and the other thumb with the control soap are then carefully and separately placed on the same nutritional agar medium plate without overlapping. The way that microbial growth on the plates behaved after 24 hours at 37°C of incubation (Wijetunge *et al.*, 2015).

TFM (total fatty matter)

10 grams of herbal soap put in to 150 ml of dH₂O with 20 ml of 15% H₂SO₄. Bee's wax was used to solidify it, and after heating it again, it was allowed to form a cake. It was then taken out, allowed to dry, and weighed in order to calculate the total amount of fat using the formula below (Kumar *et al.*, 2022).

$$\text{Total Fatty Matter} = \frac{(a - b)}{c} \times 100$$

50 ml of warm ethanol and 5 g of the sample were added to a volumetric flask, and the mixture was shaken till the sample was fully dissolved. 20 ml of warm ethanol were

filtered through a tarred filter paper, which was then dried for one hour at 105 °C. The weight of dried paper was taken.

Skin Irritation Test

The skin of five peoples was thoroughly cleaned before applying a sample of soap, and then looked for any signs of itching, burning, or other symptoms.

Determination of Antibacterial activity of plant extract and formulated soap

The bacteria, *Klebsiella pneumonia* (gram negative), *Escherichia coli* (gram negative) and *Bacillus cereus* (gram positive) selected and these strains were cultured in nutrients broth incubated for 24hour at 37°C. An amount of 10g of nutrients agar was weighed, dissolved in 360ml of deionized water, autoclaved for 15 minutes at 121°C, and then allowed to cool to room temperature. 30ml of the media was added to each Petri plate, and it was left to solidify. A generous portion of each bacterial solution was extracted using a swab and evenly distributed onto Petri plates. A sterile cork borer (3–6 mm) was used to drill five wells per plate, and each well was appropriately labeled. To one of the wells, DMSO (20µl) was added as a negative control. To function as a positive control, the antibiotic ciprofloxacin (final concentration of 1 mg/ml in DMSO) was put to the center well. The remaining holes were filled with 20 µl of plant extract and formulated soap at varying concentrations (50, 100, and 150 µg/ml). After 24 hours, the graduated ruler was used to quantify a distinct zone of inhibition.

Determination of Antifungal activity of plant extract and formulated soap

The antifungal activity against *Aspergillus flavus* and *Aspergillus niger* was determined by applying the technique described in Ruparelia *et al.* (2008). Using a graduated cylinder, transfer about 7 ml of sterilized

SDA medium into each test tube. Next, add 67 µL of extract and prepared soap in each test tube, varying in concentrations (50, 100, and 150 µg/mL), and let to solidify. Terbinafine was utilized as the positive control and deionized water as the negative control. Every tube was positioned slantwise and kept at ambient temperature. Each test tube received an equal amount of the fungus before being sealed with cotton plugs. Following each procedure, the tubes were all put in incubators at 30°C. The graduated ruler was used to observe the inhibition after seven days.

Determination of Antioxidant activity of plant extract and formulated soap

Popularly, anti-oxidants being biomolecules that prevent other molecules from oxidizing. There are several antioxidants, including thiols and vitamin C (ascorbic acid).

DPPH assay

Ascorbic acid, prepared soap, and plant extract were combined in a 1:1 ratio with regard to the test. Stock solutions of plant extract and soap formulation was created in ddH₂O at a concentration of 1 mg/ml, and these solutions were subsequently diluted further to 50, 100, and 150 µg/ml. An ascorbic acid solution with a concentration of 1 mg/ml was used as a reference (control). In a separate test tube, same measurements were taken. 200 µl of each sample and control were mixed with 800 µl of DPPH solution (1.5 mg/50 ml methanol) from the various concentrations before being kept at room temperature for 30 min in the dark. Using a spectrophotometer, absorbance at 517 nm wavelength was recorded. The proportion of scavenging was calculated using the following equation:

(Absorbance of Control – Ab of test sample) ÷ Ab of control x 100.

RESULTS

Soap formation

In current study, soap is created by using extract of *Suaeda fruticosa*.

Phytochemical screening of *suaeda fruticosa* methanolic extract and formulated soap

The preliminary phytochemical analysis was carried out utilizing *Suaeda fruticosa* methanolic extract and herbal soap. The outcome demonstrates whether the chemical component is present or not.

Table 1- show presence (+) and absence (-) of phytochemical compounds in herbal soap and extract of *Suaeda fruticosa*.

Test	Presence/ Absence in Herbal Soap	Presence/ Absence in methanolic Extract of <i>Suaedafruticosa</i>
Saponins	+	+
Tannins	+	+
Flavonoids	+	+
Alkaloids	+	+
Coumarins	+	+
Steroids	+	+
Anthraquonions	+	-

Our findings indicate the existence of tannins, alkaloids, flavonoids, terpenoids, glycosides, and phenols. Alkaloids are present and are utilized to treat inflammation. Flavonoids are present and act as an antioxidant defense. Tannins have anti-inflammatory properties (kumar *et al.*, 2022).

Physiochemical evaluation of herbal soap

The herbal soap had a pH of 7.92. The formulated soap has 0.25 % of free alkali. The formulated soap's moisture content was 50%. For herbal soap, foam retention was 10 minutes. 40% of the total fatty matter was observed, which is acceptable amount (%). Skin-irritation testing is revealed no irritation.

Table 2- Physiochemical evaluation of herbal soap

Tests	Results
Ph	7.92
Colour	Dark brown
Odor	Organic odor of rose oil
Appearance	Dark brown
Moisture content	50%
Foam height	30 ml
Foam retention	10 min
% free alkali	0.25 %
TFM	40%
Skin irritation	No
Skin burning	No
Alcohol insoluble matter	20%

Determination of Antibacterial activity of plant extract and formulated soap

At the quantity of 50 µg/mL formulated soap and Plant extract pose zone of inhibition in mm of 10 ± 0.25 and 6.5 ± 0.34 against *K. pneumonia*, 12 ± 0.5 and 9.8 ± 0.5 against *E. coli*, 20.97 ± 0.2 and 16.7 ± 0.1 against *B.cereus* respectively. At the concentration of 100 µg/ml formulated soap and Plant extract exhibited zone of inhibition in mm of 15 ± 0.35 and 10 ± 0.4 against *K. pneumonia*, 20.4 ± 0.3 and 16.5 ± 0.65 against *E. coli*, 25.97 ± 0.2 and 20.7 ± 0.5 against *B. cereus*. At the quantity of 150µg/ml formulated soap and Plant extract exhibited zone of inhibition in mm of 20 ± 0.35 and 15 ± 0.54 , against *K. pneumonia*, 25.8 ± 0.3 and 21 ± 0.5 against *E. coli*, 30.97 ± 0.2 and 25.7 ± 0.7 against *B.cereus*, DMSO used as negative control and ciprofloxacin (1µg /mL) as a positive control which shows zone of inhibition in mm of 30 ± 0.32 against *K. pneumonia*, 34 ± 0.2 against *E. coli* and 39 ± 0.3 against *Bacillus cereus*. Both soap and plant extract shows good result against *Bacillus cereus* at the concentration of 150µg/ml.

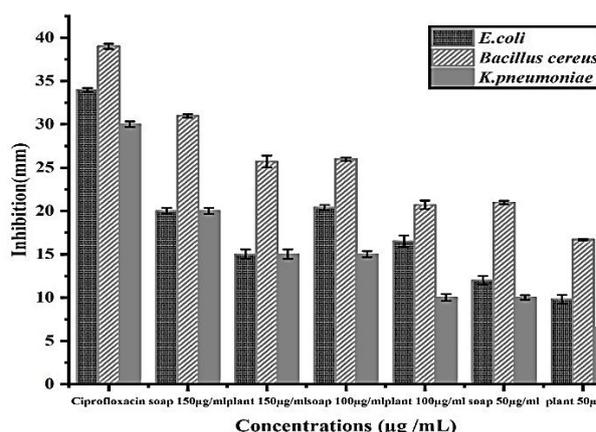


Fig. 1- Antibacterial activity of herbal soap and plant extract

Determination of Antifungal activity of plant extracts and formulated soap

The results of the current investigation showed that herbal soap greatly suppressed the development of tested fungal strains and proved to be a good, beneficial antifungal source. At concentration of 50µg/ml herbal soap and plant extract exhibited inhibition of 15.4±0.3mm and 10±0.22mm against *A.niger*, 17.3±0.3 and 13.4±0.25mm against *A. flavus*. At the quantity of 100µg/mL herbal soap and plant crude material showed growth inhibition of 22.4±0.33mm and 17.2±0.3mm against *A. niger*, 30±0.2mm and 25.5±0.3mm against *A. flavus*. The herbal soap and plant extract at the highest concentration of 150µg/ml exhibited growth inhibition of 34.95 ± 0.72mm and 28.6 ± 0.68mm against *A. niger*, while 36.9 ± 0.44mm and 23.65 ± 0.15mm inhibition against *A. flavus* respectively. DMSO served as a negative control. No fungal growth was observed in positive control test tubes. Soap shows good result against *A. flavus* while plant extract shows good result against *A. niger* at the concentration of 150µg/ml.

DPPH assay of herbal soap and plant extract

Concentrations of 50, 100, and 150 g/ml were used. Although plant extract's capacity

to scavenge DPPH increases dramatically as extract and soap concentrations rise to 150, 100, and 50 g/ml, respectively, so does their effectiveness. Ascorbic acid's maximal scavenging capability is 92.6%. Plant extract and formulated soap had scavenging capacities of 23% and 40%, 35%, and 51%, respectively, at 50 and 100 g/ml concentration, compared to ascorbic acid's capacities of 50% and 70%. Both plant and plant extract shows good result at 150 g/ml conc.

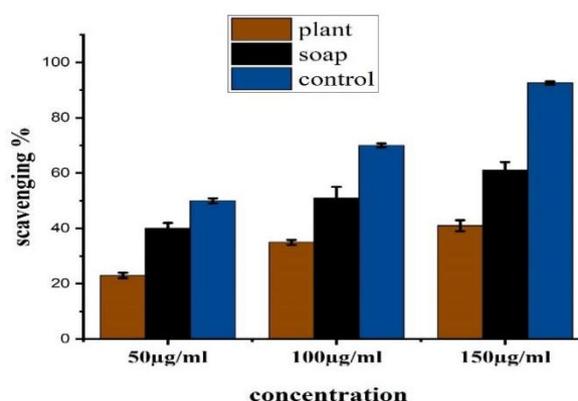


Fig. 2- DPPH scavenging activity of plant and formulated soap

DISCUSSION

In the current study, ingredients used in herbal soap includes *Suaeda fruticosa* methanolic extract as well as coconut oil, rose water, *Aloe vera* gel. The same procedure was also followed by studies of Rani *et al.*, (2023).

Flavonoids are present and act as an antioxidant defense (Agati *et al.*, 2007). Phenol acts as an anti-fungal (Simonetti *et al.*, 2020). Saponins have detergent properties and act as foaming agent (chen *et al.*, 2010). Tannins have anti-inflammatory properties (Ambreen *et al.*, 2020).

The pH of the herbal formulation was 7.92. The formulated soap has 40% TFM. TFM shows quantity of fatty material present in the soap. Dry skin needs high TFM in soap (Mensah *et al.*, 2011). The formulated soap had 50%moisture content (Tarasov *et al.*,

2021). Moisture content is greater in the formulated soap due to the addition of more *Aloe vera* (Sindhu *et al.*, 2019). Soap has a maximum foam retention time of 10 minutes. Several studies have suggested that longer foam retention provides a greater microbial reduction (Fischler *et al.*, 2007).

Klebsiella pneumoniae may lead to various infections, including skin and soft tissue infections (Paczosa and Mecsas 2016). Soap should be made from plant extracts that have antimicrobial properties in order to combat severe pathogenic diseases (Nadaroglu and Baran, 2020). In the current study at highest concentration of 150µg/ml exhibited zone of inhibition in mm of 20 ± 0.35 and 15 ± 0.54 , against *K. pneumoniae*, 25.8 ± 0.3 and 21 ± 0.5 against *E. coli*, 30.97 ± 0.2 and 25.7 ± 0.7 against *B. cereus* respectively.

Aspergillus niger and *Aspergillus flavus* are fungi that can cause various diseases in humans, primarily through respiratory infections (Samson *et al.*, 2014). *Aspergillus flavus* is a fungus known for producing mycotoxins, particularly aflatoxins, which can contaminate crops and pose health risks to humans and animals (Hedayati *et al.*, 2007).

These findings correlate with the study of Ejaz *et al.*, (2021) who studied the leaves extracts of *Suaeda vermiculata* contains compounds of fats that cause lysis of fungal cell wall which make them antifungal and medicinally important plants. In the current study the DPPH scavenging capacity of herbal soap and plant was identified and at maximum scavenging capacities of 41% and 61% at 150 g/ml conc. This observation was also supported by early study of (Febriani *et al.*, 2021) who also observed antioxidant capacity of herbal soap. Oueslati *et al.*, (2012) reported that *S. fruticosa* shoot extract contains valuable phenolic and abundant antioxidant capacity and *S.*

fruticosa is an interesting source of antioxidant and contains highest potency of phenols.

CONCLUSION

The plant *Suaeda fruticosa* methanolic extracts in this investigation show the potential for manufacturing soap. The herbal ingredients included in formulation, such as *Suaeda fruticosa* extract, *Aloe vera*, and coconut oil, have exhibited important actions including moisturizing, anti-aging, soothing, anti-acne, and anti-bacterial. The use of herbal medicine can treat a variety of skin conditions. Ayurvedic medicines are generally inexpensive compared to allopathic medications. Herbal remedies are a great source of active components that may cure skin infections like rashes and deadly skin, more safely and affordably.

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