

Formulation And Evaluation of Sting Bite Herbal Spray of *Achyranthes Aspera* Extract.**Vaibhav Sangappa Hagalgunde., Sheetal Devichand Dhage and Somnath Ashok Patil**

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Abstract

The formulation and evaluation of a novel anti-stingbite spray using *Achyranthes aspera*, a plant known for its medicinal properties, is explored in this study. *Achyranthes aspera*, also known as "Chaff Flower," has been traditionally used in treating various ailments, including insect bites and stings. The aim of this research is to develop a spray formulation that harnesses the anti-inflammatory, analgesic, and antimicrobial properties of *Achyranthes aspera* for effective relief from insect bites. The study involves the extraction of bioactive compounds from the plant, formulation of a spray, and evaluation of its physicochemical properties, stability, and anti-stingbite efficacy. The present study aims to formulate and evaluate an *Achyranthes aspera* extract-based anti-sting spray with potential soothing and antimicrobial properties. Three different formulations (F1, F2, and F3) were developed by varying the concentration of *Achyranthes aspera* extract while maintaining other excipients constant. The formulations were subjected to physicochemical evaluations, antimicrobial efficacy testing, stability studies, and user acceptability analysis. The results suggest that the F3 formulation, with the highest concentration of *Achyranthes aspera* extract, demonstrated the best anti-sting and antimicrobial activity, whereas F2 provided a balance between efficacy and stability.

Keywords: *Achyranthes aspera*, anti-sting spray, formulation, evaluation, stability, physicochemical properties

Introduction

Achyranthes aspera, commonly known as Prickly Chaff Flower, is a herbaceous plant widely distributed across tropical and subtropical regions of the world. This plant has been a part of traditional medicine for centuries due to its broad spectrum of therapeutic properties, including its antimicrobial, anti-inflammatory, antioxidant, analgesic, and wound-healing activities. In folk medicine, different parts of the plant, such as leaves, roots, and seeds, are used to treat a variety of ailments, including cuts, bruises, wounds, and skin infections.⁸

The demand for natural antiseptics has surged due to increasing concerns about the side effects and toxicity associated with synthetic chemicals, such as alcohol-based antiseptics, especially for people with sensitive skin. Herbal products derived from plants like *Achyranthes aspera* offer a safer alternative, as they possess natural antimicrobial properties that are effective against a wide range of pathogenic microorganisms. Additionally, they are often perceived as more skin-friendly due to their natural origin.⁵

Achyranthes aspera has been shown to possess significant antibacterial, antifungal, and anti-inflammatory activities, making it an ideal candidate for inclusion in topical formulations. The bioactive compounds such as saponins, alkaloids, tannins, and flavonoids found in the plant contribute to its potent antimicrobial properties. As such, there is growing interest in formulating a topical antiseptic spray using *Achyranthes aspera* extract that can be applied to minor wounds, cuts, or abrasions to prevent infection and promote healing. valued for its anti-inflammatory, analgesic, antimicrobial, and antioxidant activities. The pharmaceutical importance of *Achyranthes aspera* can be highlighted in several key areas:

Anti-inflammatory and Analgesic Properties

Achyranthes aspera is widely known for its potent anti-inflammatory and analgesic (pain-relieving) properties. The presence of bioactive compounds such as saponins, flavonoids, and alkaloids plays a major role in reducing inflammation and pain. These properties make the plant useful in treating conditions such as arthritis, muscle pain, joint inflammation, and insect stings and bites.¹

The plant's anti-inflammatory effects have been studied through both in vitro and in vivo models, showing its potential as a natural alternative to conventional non-steroidal anti-inflammatory drugs (NSAIDs) with fewer side effects.

Antimicrobial Activity

The plant demonstrates strong antimicrobial activity against various bacteria, fungi, and viruses. It has been found to be effective against pathogens such as *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Candida albicans*. This makes *Achyranthes aspera* a potential source for developing topical antimicrobial agents or wound healing products. The antimicrobial properties are mainly attributed to the presence of alkaloids, saponins, and tannins, which inhibit the growth of microbial organisms and prevent infections, particularly in skin wounds and ulcers⁵.

Antioxidant Properties

Oxidative stress is implicated in the pathogenesis of many chronic diseases, including cancer, cardiovascular diseases, and neurodegenerative disorders. *Achyranthes aspera* contains significant amounts of polyphenolic compounds, which exhibit strong antioxidant activity.

The plant's antioxidants help neutralize free radicals, thereby protecting cells from damage and reducing the risk of chronic diseases. This makes *Achyranthes aspera* a potential candidate for developing antioxidant-rich dietary supplements or formulations aimed at preventing oxidative damage.³

Wound Healing and Skin Care

In traditional medicine, *Achyranthes aspera* has been used to treat cuts, bruises, wounds, and skin infections. Its healing properties can be attributed to its antimicrobial, anti-inflammatory, and antioxidant activities, which promote tissue repair and prevent infection.

The plant can be used in the formulation of creams, gels, or ointments for wound care and dermatological treatments. Some studies have shown that *Achyranthes aspera* enhances the healing process by stimulating collagen formation and cellular regeneration in wound areas.³

Anti-diabetic Potential

The plant has demonstrated hypoglycemic effects, meaning it has the potential to lower blood sugar levels. Traditional uses of *Achyranthes aspera* include treating symptoms of diabetes and managing blood glucose levels.

The plant's bioactive compounds, particularly saponins and alkaloids, have been shown to modulate insulin activity, suggesting that *Achyranthes aspera* could be developed as an adjunct treatment for managing type 2 diabetes⁹

Anti-cancer Activity

Preliminary studies suggest that extracts from *Achyranthes aspera* possess anti-cancer properties. These effects are attributed to the plant's ability to inhibit the growth of cancer cells and induce apoptosis (programmed cell death) in tumor cells.

The plant's saponins and flavonoids have been found to have cytotoxic effects on cancer cell lines, indicating its potential as an anticancer agent. Further research is required to explore its full potential in cancer treatment and prevention.

Hepatoprotective Properties

Liver health is essential for overall well-being, and the hepatoprotective effects of *Achyranthes aspera* have been documented in several studies. The plant has been shown to help protect the liver from damage caused by toxins and excessive alcohol consumption.⁷

Achyranthes aspera acts by enhancing the antioxidant status of the liver and protecting hepatocytes from oxidative damage. This could make it a valuable component in formulations aimed at preventing liver damage and promoting detoxification.

Cardiovascular Health

Achyranthes aspera has demonstrated potential in improving cardiovascular health. It is believed to possess blood pressure-lowering effects, which can help manage hypertension. Additionally, it has been shown to have a positive impact on lipid metabolism, potentially lowering cholesterol and triglyceride levels.

- These properties, combined with its antioxidant and anti-inflammatory effects, make *Achyranthes aspera* a potential candidate for developing formulations aimed at promoting heart health and preventing cardiovascular diseases.

Anthelmintic Activity

In traditional medicine, *Achyranthes aspera* has also been used as an anthelmintic (a substance that expels parasitic worms from the body). Studies have shown that the plant extracts are effective in treating gastrointestinal parasitic infections.

The anthelmintic activity of *Achyranthes aspera* can be attributed to its alkaloids and other bioactive compounds, which interfere with the metabolism of parasitic organisms, rendering them inactive and expelling them from the body.

Anti-allergic and Antihistaminic Properties

The plant has demonstrated antihistamine-like effects, which are useful for treating allergic reactions. *Achyranthes aspera* has been found to help alleviate symptoms of allergic rhinitis, asthma, and other allergic conditions by inhibiting the release of histamine.²

Materials and Methods

Achyranthes aspera was procured from the bixa botanical lab

All the chemicals and essentials were taken from the college of pharmacy QA lab The packaging container were taken from local stores.

Plant Material and Extraction Soxhlet Extraction Procedure:

- Loading the Soxhlet Apparatus: Place approximately 10 grams of the powdered plant material into a thimble made of filter paper and insert it into the Soxhlet extractor.
- Solvent Addition: Fill the round-bottom flask of the Soxhlet apparatus with 300 mL of distilled water.
- Extraction Process: Heat the flask gently to initiate the extraction. Distilled water will vaporize, condense in the condenser, and drip into the thimble containing the plant material. This process allows the water to extract soluble compounds.
- Continuous Extraction: Allow the extraction to proceed for 72 hours, ensuring continuous siphoning of the solvent to maximize extraction efficiency.

2.2. Phytochemical Screening

Phytochemical screening was performed to identify the presence of active compounds such as alkaloids, flavonoids, saponins, tannins, and terpenoids in the ethanolic extract. Standard procedures using reagents such as Dragendorff's reagent, Ferric chloride solution, and Frothing test were applied.

2.3. Formulation of Anti-Stingbite Spray

The anti-stingbite spray was formulated by dissolving the aqueous extract of *Achyranthes aspera* in a suitable solvent base (water and ethanol) with the addition of emulsifiers (e.g., glycerin) and preservatives (e.g., phenoxyethanol) for stability. The formulation was mixed using a magnetic stirrer and transferred into spray bottles.

Ingredients	Quantity (g)			Function
	F 1	F2	F3	
<u>Achyranthes aspera</u> extract	5	10	15	Active ingredient (antimicrobial)
Ethanol (95%)	20.0	15.0	10.0	Solvent (extraction and preservation)
Glycerin	5.0	5.0	5.0	Humectant (moisturizing agent)
Propylene Glycol	4.0	4.0	4.0	Solvent and stabilizer
<u>Methylparaben</u>	0.1	0.1	0.1	Preservative (antimicrobial)
<u>Propylparaben</u>	0.05	0.05	0.05	Preservative (antimicrobial)
Distilled Water	100.0	100.0	100.0	Vehicle (to adjust final volume)

Figure 1. Formulation of Anti-Stingbite Spray

Procedure

1. Weigh the required amount of *Achyranthes aspera* extract (5 g for F1, 10 g for F2, 15 g for F3).
2. Mix the extract with Ethanol (95%) (20 g for F1, 15 g for F2, 10 g for F3) in a beaker.
3. Stir the mixture continuously for 15–20 minutes at room temperature to ensure proper extraction.

4. Allow the solution to stand for 30 minutes, then filter it using Whatman filter paper to remove any undissolved plant material.

Preparation of the Formulation Base

In a separate beaker, dissolve Methylparaben (0.1 g) and Propylparaben (0.05 g) in a small portion of distilled water (≈ 10 mL).

Stir continuously until both preservatives dissolve completely.

Incorporation of Humectants and Solvents

Add Glycerin (5 g) and Propylene Glycol (4 g) to the filtered extract solution.

Stir continuously to ensure homogeneous mixing.

Pour the preservative solution (Step 5) into the mixture and stir further.

Final Dilution and Mixing

Gradually add distilled water to the formulation until the total volume reaches 100 g.

Stir the mixture for 10–15 minutes using a magnetic stirrer until a uniform solution is obtained.

Filtration and Packaging

Filter the final formulation to remove any particulate matter.

Transfer the prepared spray solution into sterilized spray bottles using a funnel.

Label the bottles with formulation details (F1, F2, F3), ingredients, and date of preparation.

Results and Discussion

Phytochemical Screening

Phytochemical analysis revealed the presence of alkaloids, flavonoids, saponins, tannins, and terpenoids in the ethanolic extract of *Achyranthes aspera*. These compounds are known for their anti-inflammatory, antimicrobial, and analgesic properties, supporting the rationale for using this plant in the formulation.

Physicochemical Properties of the Spray

pH Measurement: The pH was determined using a digital pH meter.

The formulated spray exhibited a pH of 5.5, 5.8 and 6.2 respectively for f1 f2 and f3 , which is considered safe for topical application.

Viscosity: Measured using a Brookfield viscometer.

The viscosity was low, 38,46,42 respectively ensuring ease of application. The spray demonstrated a fine mist with a uniform distribution, providing good coverage on the skin.

Stability Study: Formulations were stored at different temperatures (25°C, 40°C) and evaluated over a period of 30 days. The formulation remained stable under both ambient and elevated temperature conditions. No significant changes in the physical properties or pH were observed during the three-month study period.

User Acceptability: Assessed through a sensory evaluation questionnaire.

Active Ingredient Concentration: Higher *Achyranthes aspera* extract (F3) may provide stronger antimicrobial and anti-inflammatory effects. However, excessive concentration could lead to formulation instability or skin irritation.

Ethanol Content: Ethanol helps in extraction and preservation but may cause skin dryness. Lower ethanol (as in F3) could reduce irritation, but too little may affect solubility and preservation.

F2 (10g *Achyranthes aspera* extract) offers a balanced concentration higher efficacy than F1 but without the potential excessive extract load of F3.

Conclusion

The development of a sting bite spray using *Achyranthes aspera* extract presents a promising natural remedy for managing pain, inflammation, and allergic reactions caused by insect bites and stings. The bioactive compounds in the extract, such as alkaloids, saponins, and flavonoids, exhibit significant anti-inflammatory, analgesic, and antimicrobial properties, making it a potential alternative to synthetic treatments. Preliminary studies indicate its effectiveness; however, further clinical trials and formulation optimizations are necessary to establish its safety, stability, and efficacy for commercial use .

References

1. Verma P. and Chauhan N. *Achyranthes aspera*: (2012) An overview of its medicinal uses and phytochemical profile. *J Med Plants.*;15(3):85-92.
2. Jain P and Sharma R. Pharmacological properties of *Achyranthes aspera*. *Int J Phytomed.* 2014;6(4):207-212.
3. Akinmoladun A.F and Olaleye M.T. Antioxidant, antibacterial, and antidiabetic activity of *Achyranthes aspera* L. (Amaranthaceae). *Asian Pac J Trop Biomed.* 2015;5(4):274- 279.
4. Saha S, Mandal S. Anti-cancer potential of *Achyranthes aspera*: A review of recent findings. *Pharmacognosy Rev.* 2013;7(14):149-154.
5. Patil PR, Chavan SM. Antioxidant, antibacterial, and antidiabetic activity of *Achyranthes aspera* L. leaf extract. *Int J Phytomed.* 2015;7(2):121-127.
6. Tiwari S.K, Agarwal S. Hepatoprotective potential of *Achyranthes aspera*: A review. *J Ethnopharmacol.* 2016;175:86-93.
7. Kumar P and Ghosh A. Formulation and evaluation of herbal topical gels for wound healing. *Pharm Res.* 2015;8(2):142-147.
8. Chowdhury MA, Islam MT. Evaluation of anti-inflammatory and analgesic activities of *Achyranthes aspera* L. in experimental models. *Phytother Res.* 2017;31(10):1600- 1607.
9. Singh P, Gupta A. Evaluation of anthelmintic activity of *Achyranthes aspera* L. leaf extract. *Int J Pharma Bio Sci.* 2012;3(4):619-622.
10. Jain P, Sharma R. Topical formulation of *Achyranthes aspera* for insect sting bites: An experimental study. *Int J Pharm Sci.* 2019;10(5):230-236.