

Investigation of phytochemical constituent of Plant *Sauromatum Venosum* Linn having anti-Acne activity

Shivani Lodhi¹, Deepak Kumar Basedia¹, Balkrishna Dubey¹,
Sunil Kumar shah², Sandra Goutam¹.

1. Research Scholar, Technocrats Institute of Technology – Pharmacy, Bhopal, MP.

2. Professor, TIT College of Pharmacy, Bhopal, MP.

Abstract: - Microbes are single-cell organisms. They are so tiny that, millions can fit into the eye of a needle and cannot be seen with the naked eyes. Microbes are the oldest form of life on earth. They are very small living things and are sometimes termed as micro-organisms. Microbes can only be seen using a microscope. Microbe fossils date back more than 3.5 billion years.

A few harmful microbes, for example less than 1% of bacteria, can invade our body (the host) and make us ill. Microbes cause infectious diseases such as flu and measles. There is also strong evidence that microbes may contribute to many non-infectious chronic diseases such as some forms of cancer and coronary heart disease. Different diseases are caused by different types of micro-organisms. First, the infectivity of an organism determines the number of individuals that will be infected compared to the number who are susceptible and exposed. Second, the pathogenicity is a measure of the potential for an infectious organism to cause disease. Acne vulgaris is a chronic inflammatory disease mainly affecting the teenagers. As it is chronic and affects face mainly, it always causes negative psychological impact on human life. In a study conducted in

2015 acne affected 650 million people globally making it the most common skin disease worldwide. In a study of “Global burden of disease” it is revealed that acne was the 8th common prevalent disease worldwide.

The aim of this study is to investigate the antimicrobial properties and in-vivo antiacne activity of the ethanolic extract derived from *Sauromatum venosum*. This botanical extract has shown promise in traditional medicine, and the primary objective is to scientifically evaluate its potential in combating microbial infections and alleviating acne symptoms.

In conclusion, this research establishes a compelling case for the therapeutic potential of *Sauromatum venosum* in the treatment of acne. The combined findings regarding the extraction yield, phytochemical profile, antimicrobial activity, and significant in vivo results collectively suggest that this plant may serve as a viable alternative or complementary treatment option for acne.

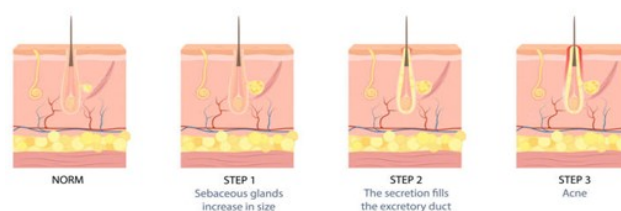
Keywords: Anti-Acne, Phytochemicals, ethenolic extract, *Sauromatum venosum*, Antimicrobial.

Introduction: Microbes are single-cell organisms. They are so tiny that, millions can

fit into the eye of a needle and cannot be seen with the naked eyes. Microbes are the oldest form of life on earth. They are very small living things and are sometimes termed as micro-organisms. Microbes can only be seen using a microscope. Microbe fossils date back more than 3.5 billion years. A few harmful microbes, for example less than 1% of bacteria, can invade our body (the host) and make us ill. Microbes cause infectious diseases such as flu and measles. There is also strong evidence that microbes may contribute to many non-infectious chronic diseases such as some forms of cancer and coronary heart disease. Different diseases are caused by different types of micro-organisms. Microbes that cause disease are called pathogen. Acne vulgaris is a chronic inflammatory disease mainly affecting the teenagers. As it is chronic and affects face mainly, it always causes negative psychological impact on human life. In a study conducted in 2015 acne affected 650 million people globally making it the most common skin disease worldwide. In a study of “Global burden of disease” it is revealed that acne was the 8th common prevalent disease worldwide.

Etiology of acne

Acne caused by clog up of follicles, hyper-keratinizations, keratin plug formation, enlargement of sebum gland, increase of sebum production. Enlargement of micro-comedo also responsible for open comedo (black heads) and closed comedo (white heads). Propionibacterium acne can be developed inflammation lesions.



Plant-derived chemicals

Plant-derived chemicals are a wide group of chemical compounds that have been found naturally in plants. The extensive existence of these compounds has demonstrated beneficial advantages in terms of antioxidant, antibacterial, and antifungal activities. They can restore the clinical application of older antibiotics by increasing their potency and as a consequent, avoid the development of resistance.

AIM AND OBJECTIVE

The aim of this study is to investigate the antimicrobial properties and in-vivo antiacne activity of the ethanolic extract derived from *Sauromatum venosum*. This botanical extract has shown promise in traditional medicine, and the primary objective is to scientifically evaluate its potential in combating microbial infections and alleviating acne symptoms.

PLANT PROFILE

Sauromatum venosum

Sauromatum venosum ‘Indian Giant’ better known as the ‘Giant Voodoo lily’ is a spectacular aroid from Himalayan India producing strong patterned leaf stems, petioles, emerging from the ground supporting large dissect palmate leaves up to 60cm or more across. Exotic enough you might think, but no its the outrageous flower

spike or caudix as they are correctly known that demands attention, deep purple and black up to 45m high they are otherworldly plus just in case you don't notice their arrival the 'perfume' will certainly grab you attention, not quite sure how to put this but boy it's an evil aroma if ever there was one. Designed to attract flies and carrion beetles it's one of nature's wonders able to generate heat too whilst trapping the hapless pollinators long enough to collect enough pollen ready to move on to the next flower farthing the reproductive cycle.



MATERIALS AND METHODS

1. **Collection of the plants sample**
2. **Preliminary phytochemical screening of extract**

Extraction of plant material using maceration process

3. **Qualitative chromatographic analysis**

Thin layer chromatography

4. ***In-vivo* anti-acne activity**

Measurement of ear thickness

Collection of plant material

Plants can be collected from either wild

woods or herbariums. However, there is a risk of erroneously recognized plants in the case of wild plants. They have the advantage of not containing any pesticides or herbicides. They are treated as quickly as possible after collection to avoid secondary metabolites from deteriorating.

Extraction of plant materials

Extraction from plant materials is an important step in phytochemical processing for discovering bioactive secondary metabolite. Maceration was a popular and inexpensive homemade technique for the preparation of tonic since a long time. Moreover, this technique was used for the extraction of essential oils and active compounds from plant materials. Generally, the maceration procedure consists of multiple steps in extraction.

Phytochemical Screening

Plants generate compounds known as phytochemicals. These are created by the primary and secondary metabolisms of the plant. These phytochemicals are necessary for plants to survive or to fend off other plants, animals, insects, microbial pests, and pathogens. They also protect plants from illness and damage induced by environmental threats such as pollution, UV, stress, and drought. They have been employed as traditional medicine and as poisons since ancient times phytochemical examinations were carried out for all the extract as per the standard methods

In vivo* antiacne activity of ethanolic extract of *Sauromatum venosum

Animals

Wistar rats (150-200g) were group-housed (n=6) under a standard 12 h light/dark cycle and controlled conditions of temperature and humidity (25±2°C, 55–65%). Rats received standard rodent chow and water *ad libitum*. Rats were acclimatized to laboratory conditions for 7 days before carrying out the experiments. All the experiments were carried in a noise-free room between 08.00 to 15.00 h. A separate group (n=6) of rats was used for each set of experiments. The animal studies were approved by the Institutional Animal Ethics Committee (IAEC), constituted for the purpose of control and supervision of experimental animals by Ministry of Environment and Forests, Government of India, New Delhi, India.

Experimental designs

Group –I: Control (acne induced)

Group -II: Ethanolic extract of *Sauromatum venosum* (100mg/kg, p.o.)

Group –III: Ethanolic extract of *Sauromatum venosum* (200mg/kg, p.o.)

Group –IV: Clindamycin (200mg/kg, p.o.)

The experimental model of acne-like inflammation was induced in rat ears through subcutaneous administration of 0.14 mg of heat-killed *Propionibacterium acnes*. The study comprised four experimental groups:

Group I served as the control with acne induction,

Group II received 100 mg/kg of ethanolic extract of *Sauromatum venosum* orally,

Group III received 200 mg/kg of the same extract orally, and

Group IV was administered Clindamycin at a dose of 200 mg/kg orally.

Statistical analysis

All statistical analysis is expressed as mean ± standard error of the mean (SEM). Data were analyzed by one way ANOVA, where applicable p<0.05 was considered statistically significant, compared with vehicle followed by Dunnett's test.

RESULTS AND DISCUSSION

Determination of percentage yield

S. No.	Extracts	% Yield (W/W)
1	Ethanol	5.70%
2	Aqueous	8.32%

In this study, the percentage yield of *Sauromatum venosum* extracts was determined for both ethanol and aqueous solvents, as presented in Table.

The results indicate that the aqueous extract produced a higher yield (8.32% W/W) compared to the ethanol extract, which yielded 5.70% W/W. This difference in extraction efficiency can be attributed to the polarity of the solvents.

Phytochemical screening of extract

S. No.	Constituents	Ethanolic
1.	Alkaloids	-ve
2.	Glycosides	+ve
3.	Flavonoids	+ve

4.	Phenol	-ve
5.	Amino acids	-ve
6.	Carbohydrates	+ve
7.	Tannins	-ve
8.	Proteins	+ve
9.	Saponins	+ve
10.	Diterpenes	+ve

The phytochemical screening of the ethanolic extract of *Sauromatum venosum*, as summarized in Table, revealed the presence of several important bioactive compounds. The extract tested positive for glycosides, flavonoids, carbohydrates, proteins, saponins, and diterpenes. These findings suggest that the ethanolic extract of *S. venosum* is rich in diverse phytochemicals, which could contribute to its pharmacological properties.

Results of thin layer chromatography of leaves extract of *Sauromatum venosum* the thin layer chromatography (TLC) analysis of the leaf extracts of *Sauromatum venosum* would typically provide insights into the diversity and separation of different phytochemicals present in the extract. TLC is a technique used to identify and isolate compounds based on their polarity and interaction with the stationary phase (silica) and mobile phase (solvent). From the Rf value it was confirmed the presence of Quercetin as Flavanoids compound in the extract.

TLC of leaves extract of *Sauromatum venosum*

S. No.	Extract	Rf Value
	Mobile phase (Toluene: Ethyl acetate: Formic acid; 5:4:1)	
1.	Quercetin	0.52
	Long UV=5 spot	0.16, 0.38,
	Short UV=5 spot	0.16, 0.38,
	Normal light =1 spot	0.52

Total flavonoid content estimation (TFC)

The total flavonoid content (TFC) was expressed as mg/100mg of quercetin equivalent of dry extract sample using the equation obtained from the calibration curve: $Y = 0.040X + 0.012$, $R^2 = 0.999$, where X is the quercetin equivalent (QE) and Y is the absorbance.

***In vitro* antimicrobial activity of *Sauromatum venosum* extract**

The lyophilized cultures of microbial strains upon culturing in nutrient broth for 24 hours at 37°C in an incubator resulted into turbid suspension of activated live microbial cell ready to be used for microbiological study. From the broth of respective revived cultures of bacteria loop full of inoculum is taken and streaked on to the nutrient agar medium and incubated again at same culture conditions and duration that yielded the pure culture colonies on to the surface of the agar culture that are successfully stored in refrigerated conditions at 4°C as stock culture to be used for further experimentation.

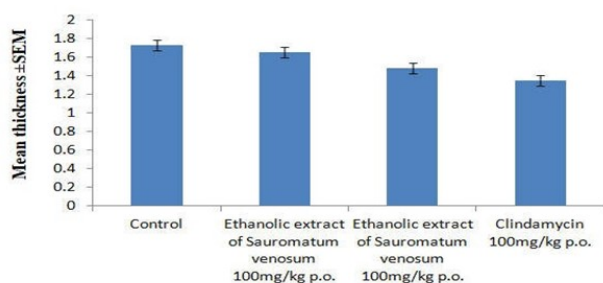
Results of *in vivo* antiacne activity of ethanolic extract of *Sauromatum venosum*

Based on the pilot screening, the following protocol was followed: In pilot screening, 6 rats were taken under study, which showed that the granulomatous inflammation remained constant from the 6th day to the 10th day.

Protocol study for *in-vivo* anti-acne activity on rats

Groups	Induction of Acne	Treatment
Control (acne induced)	Heat killed <i>Propionibacterium acnes</i>	Vehicle
Treated with ethanolic extract of <i>Sauromatum venosum</i>	Heat killed <i>Propionibacterium acnes</i>	100 mg/kg p.o.
Treated with ethanolic extract of <i>Sauromatum venosum</i>	Heat killed <i>Propionibacterium acnes</i>	200mg/kg p.o.
Treated with Clindamycin	Heat killed <i>Propionibacterium acnes</i>	200mg/kg p.o.

Effect of Clindamycin (Standard) and ethanolic extract of *Sauromatum venosum* induced acne by *Propionibacterium acnes* in rats



The significant reduction in acne lesion thickness with both the ethanolic extract and Clindamycin highlights their potential mechanisms in combating *Propionibacterium acnes*, possibly through antibacterial activity, anti-inflammatory effects, or a combination of both. The findings also warrant further investigation into the specific bioactive compounds present in *Sauromatum venosum*, as they may contribute to its therapeutic effects.

SUMMARY AND CONCLUSION

This study meticulously explores the efficacy of ethanolic extracts of *Sauromatum venosum* in mitigating acne induced by *Propionibacterium acnes*, particularly in comparison to the well-established antibiotic Clindamycin. The investigation encompasses a comprehensive analysis of the extraction yield, phytochemical constituents, antimicrobial properties, and the *in vivo* anti-acne effects of the plant extract. In the *in vivo* portion of the study, the effects of ethanolic extracts of *Sauromatum venosum* were assessed on acne-induced rats through the administration of heat-killed *Propionibacterium acnes*. The results demonstrated a significant reduction in the mean thickness of skin lesions over the treatment period. Specifically, the extract at a dosage of 200 mg/kg p.o. yielded a reduction in lesion thickness comparable to that of Clindamycin, which exhibited a marked decrease in mean lesion thickness over the same timeframe. On Day 10, the thickness in the control group was recorded at 1.86 mm,

while the group treated with 200 mg/kg of the extract exhibited a thickness of 0.59 mm, illustrating the extract's potent anti-acne properties.

In conclusion, this research establishes a compelling case for the therapeutic potential of *Sauromatum venosum* in the treatment of acne. The combined findings regarding the extraction yield, phytochemical profile, antimicrobial activity, and significant in vivo results collectively suggest that this plant may serve as a viable alternative or complementary treatment option for acne.

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