

Evaluation of phytochemical and antibacterial activity on *Tridax procumbens* extract for wound care applications

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Abstract

Background: The functional requirements of protective textiles have led to the innovative use of a variety of natural plants with wound healing properties for wound care applications. The *Tridax procumbens* is one of the incredibly important natural plants available in all areas with hygienic properties. **Materials and Methods:** The *T. procumbens* herb leaves were collected and extracted, and it was coated on bamboo fabric. **Results and Discussion:** The extract was tested under qualitative phytochemical screening and expressed the occurrence of phytochemicals. The antimicrobial property of *Tridax* ethanolic extract 8% and 16% concentrated treated samples was tested using standard AATCC 147 qualitative and AATCC 100 quantitative tests against both Gram-positive bacterial pathogens, namely *Staphylococcus aureus* and *Klebsiella pneumoniae*, and Gram-negative bacterial pathogens, namely *Pseudomonas aeruginosa* and *Escherichia coli*. **Conclusion:** The test result shows that the *Tridax* treated samples have superior antibacterial activity.

Key words: Agar well diffusion, antibacterial activity, ethanolic extract, microorganisms, phytochemical constituents, *Tridax procumbens*

INTRODUCTION

Wound healing is very important for survival mechanism, and it represents to maintain the normal structure and function of tissues. Wound healing is still a challenge for the pharmaceutical industry, despite being more complex nowadays. The 1–3% of drugs only used in western pharmacopeias for curing and healing of wounds. It is due to this challenge that medicinal plants possess enormous potential to come up with a widespread solution for the wound healing treatments.^[1]

Moreover, medicinal plants are now considered as a rich source for treatment of wounds and health-care systems and an impressive number of modern drugs have been isolated from natural resources.^[2] Natural plants have been used for centuries as a remedy for human diseases and propose a new source of biologically active chemical compounds as an antimicrobial agent.^[3] The wound care textile applications are directly related to the skin, and it should have the better hygienic effects. From the plant kingdom, one of the copious sources of the natural plant is *Tridax procumbens*. *Tridax* is known to be

a prosperous source of various secondary metabolites, and these higher plants are widely used for traditional medicine practices.^[4]

The plant *T. procumbens* are commonly used for wound healing purpose, and also it has numerous properties such as anticoagulant, antifungal, and insect repellent. The leaf extracted herbal solution is directly applied to wounds to get cure instantly and also used for various skin diseases such as boils and cut wounds. The plant kingdom contains many species of plants possessing substances of medicinal value that had to be discovered.^[5,6] Although large numbers of plants are constantly being analyzed for their antimicrobial effects, still there is a search for a natural antibiotic.^[7]

Plant-derived substances have recently become a variety of phytochemicals such as tannins, flavonoids, and phenols of

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Received: 07-08-2018

Revised: 17-09-2018

Accepted: 26-09-2018

great interest due to their versatile applications.^[8,9] Plants containing favorable phytochemicals may supplement the requirements of the human body by acting as natural antioxidants and the solvent ethanol which may have yielded a great number of active constituents responsible for the antimicrobial activity.^[10] Apart from wound applications, the plant is used to develop medicated products. The present research work aims at developing antimicrobial medicated fabrics for health-care applications.

MATERIALS AND METHODS

Materials Selection of Textile Fabric

The twill weave bamboo fabric with the count of 2×42 's was constructed and used for research work. Then, the developed fabric was forced to undergo some treatments such as desized, scoured, and bleached.^[11]

Sourcing of Natural Plant

The *T. procumbens* leaves were selected for the study on the basis of their wound healing and skin disease curing medicinal properties, and it was collected from Agriculture University.

Extraction of Herbal Extract

The *T. procumbens* leaves were dried in a dark condition for about 7 days and transformed into residue using automatic machines. 50 g of fine powder was mixed with 100 ml of ethanol for 7 h using Soxhlet apparatus by hot extraction method. After the extraction process, the solution was kept for solvent evaporation for about 8 hours. The obtained precipitate of the developed extract was stored in a refrigerator with the help of tight containers at 4°C. Based on the need of requirement, the extract was diluted and utilized for further end use.^[12,13]

Application of Coating on Fabric Using Extract

The bamboo fabric samples were finished with 8% and 16% concentrated herbal extract using the material liquor ratio of 1:10 with optimized conditions like 20-pascal pressure at 50°C temperature for about 1h time duration with pomegranate mordant as cross-linking agent. The extracts were applied on to the fabric by pad dry cure method using the padding mangle, and the fabric was dried and then cured at 140°C for 3 min.^[14]

Methods

The various methods are used for identifying the phytochemical constituents in the *T. procumbens* extracted

solution and antimicrobial activity on treated fabric samples

Preliminary Phytochemical Screening

The qualitative method of phytochemical analysis is used to identify the presence of active phytoconstituents in *Tridax* extract such as steroids, alkaloids, flavonoids, phenols, and tannins. The preliminary test was conducted as per standard test procedures.

Standard Test Method for Alkaloids - Meyer's Test

Few drops of Meyer's reagent were mixed with 1.0 ml of the extract. A yellow creamy precipitate indicates the presence of alkaloids.

Test for Flavonoids - Alkaline Reagent Test

A few drops of the extract were added with 4–5 drops of sodium hydroxide solution. The dark color of the yellow solution was obtained, and after some time that colored solution is modified in to colorless by adding dilute hydrochloric acid which indicates the presence of flavonoids.

Test for Phenols - Ferric Chloride Test

The 2.0 ml of distilled water was added with 1.0 ml of the extract followed by a few drops of 10% aqueous ferric chloride solution. Blue, green, or violet color has formed indicating the presence of phenols.

Test for Saponins - Foam Test

Drops of sodium bicarbonate solution were added with 1.0 ml of the extract, shaken vigorously and kept for 3 min. A honeycomb-like froth will be formed, and this indicates the presence of saponins.

Test for Tannins - Ferric Chloride Test

Few drops of aqueous 5% ferric chloride were mixed with 1.0 ml of the extract. A bluish-black color was formed which then gets disappeared in addition of few drops of dilute sulfuric acid and a yellowish brown precipitate was formed which indicates the presence of tannins.^[15,16]

Antibacterial Activity Assessment Method of *T. procumbens* Ethanolic Extract (AATCC - Test Method)

The antibacterial activity of the *T. procumbens* extract was analyzed using AATCC standard qualitative test method of agar well diffusion method.

Preparation of Bacterial Cultures

The different four bacterial cultures and two different fungal cultures were developed, namely Gram-positive bacterial pathogens such as *Staphylococcus aureus* and *Klebsiella pneumonia* and Gram-negative bacterial pathogens such as *Pseudomonas aeruginosa* and *Escherichia coli*. The developed bacterial cultures were maintained on nutrient agar slant and were also isolated, stored separately in a refrigerator at 4°C.

Antibacterial Activity Assessment by Agar Well Diffusion Method

The antibacterial activity of *T. procumbens* extract was evaluated using agar well diffusion method.^[17] 20 ml of nutrient agar was prepared and allowed for sterilization at 121°C for about 15 min. The Petri plates were autoclaved in a hot air oven at 121°C for 45 min. The leaf ethanolic extract has been converted into 100 µg/ml concentration. The nutrient agar of 20 ml was dropped into the Petri plates and was allowed to solidify and then plant extract was poured in the developed well, and it was incubated at 36°C for a time period of 24 h. After 24 h, the antibacterial activity was assessed against the test organisms *S. aureus*, *Klebsiella pneumoniae*, *P. Aeruginosa*, and *E. coli* by measuring the zone of inhibition.

RESULTS

The preliminary phytochemical screening and antimicrobial activity test results were discussed.

Preliminary Phytochemical Screening

The qualitative phytochemical screening of *T. procumbens* extract test results are shown in Table 1. The results revealed the presence of phytochemical constituents in the extract such as flavonoids, phenols, tannins, and alkaloids, whereas saponins were absent in the *T. procumbens*. The precipitate color represents the presence of these components induces either individually or in combination to possess' antimicrobial activity.

Antibacterial Activity Assessment by Agar Well Diffusion Method

The antibacterial activity of the *T. procumbens* leaf extract was assessed by agar diffusion method, and it has been shown in Table 2. The zone of inhibition test results of *Tridax* extract against Gram-positive bacterial pathogens, namely *K. pneumoniae* and *S. aureus* and Gram-negative bacterial pathogens, namely *P. aeruginosa* and *E. coli* by agar well diffusion method.

Table 1: Qualitative phytochemical analysis of the ethanolic extract of *Tridax procumbens*

Plant constituents	Ethanolic extract
Alkaloids	
Meyer's test	+
Flavonoids	
Alkaline reagent test	+
Phenol	
Ferric chloride test	+
Saponins	
Foam test	-
Tannins	
Ferric chloride test	+

Table 2: Antibacterial zone of inhibition in (mm) against Gram-positive and Gram-negative bacterial pathogens on ethanolic extract of *Tridax*

Test organisms	Zone of inhibition (in mm)	
	8% Conc. treated sample	16% Conc. treated sample
<i>Staphylococcus aureus</i>	17	21
<i>Klebsiella pneumonia</i>	15	16
<i>Pseudomonas aeruginosa</i>	14	15
<i>Escherichia coli</i>	12	14

DISCUSSION

The phytochemicals are the most important factor for depicting the antimicrobial activity.

Flavonoids are found to be active antimicrobial component against a wide range of microorganisms due to their ability to combine with extracellular, soluble proteins, and bacterial cell wall. The presence of tannins in the roots of *Tridax procumbens* implied that tannin may be the active compound which was responsible for antimicrobial activity in this study. The component tannin expresses better antibacterial activity.^[16-18]

The zone of inhibition test results of *Tridax* ethanolic extract showed good antibacterial activity against Gram-positive pathogens, namely *S. aureus* (20 mm) and *K. pneumoniae* (16 mm) than Gram-negative pathogens, namely *P. aeruginosa* (17 mm) and *E. coli* (16 mm). In 16% conc. extract treated fabric sample than 8% conc. extract treated fabric sample. The extract proved that it has better control over the positive pathogens than compared to the negative pathogens.

CONCLUSION

The study results proved the phytochemical constituents present in the extract includes flavonoids, phenols, alkaloids, and tannins. The presence of phytocomponents such as tannins and flavonoids in the extract provides better antimicrobial activity. The antibacterial test results also showed greater levels of antibacterial activity in 16% conc. extract treated fabric sample against Gram-positive bacterial pathogens, namely *S. aureus* and *K. pneumoniae* than Gram-negative bacterial pathogens, namely *P. auruginosa* and *E. coli*. Based on the qualitative phytochemical screening and antibacterial activity assessments, the extract treated fabric samples proved that it will be more suitable for the wound care applications. Hence, this research work will give an idea for developing sustainable antimicrobial coated wound dressing fabrics for wound care applications in the health-care field as well as a raw material is from 100% natural resources, it is renewable and environmental benefits.

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Source of Support: Nil. **Conflict of Interest:** None declared.