



Different Fractions of *Mimosa pudica* by Wound Healing Activity

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ABSTRACT

Tannins and Flavonoids Fractions of Mimosa pudica leaves were screened for phytochemical constituents. Phytochemical analysis of the extract revealed that the wound healing activity by wound excision method. The plant material is due to the presence of active constituents like tannins or flavonoids. Mimosa pudica is used in disease related to blood, bile, bilious fever, piles, jaundice, leprosy, ulcer and smallpox. In the present study Tannins and Flavonoids Fractions of Mimosa pudica leaves sample were obtained using Maceration (softening) Process. Phytochemical studies revealed that tannins and flavonoids are present in the sample.

Keywords: Tannins; Flavonoids

INTRODUCTION

Many plants have the medicinal values by the presence of active constituents and other substances. By the presence of this value they are named as medicinal plants. Since ancient times most of the products are obtained from the natural plants to cure the diseases. Due to the natural way of curing a disease, people look forward for a natural treatment. This phenomena has expanded the research on plants to cure different diseases. Now a days, most of the people are still unaware of this natural way of treating a chronic disease by natural plant products. Our aim is to spread the knowledge of different medicinal values of the natural plant products to the people to cure a chronic disease [1-5].

Mimosa pudica is commonly known as the sensitive plant, humble plant, touch-me-not plant it is a very popular plant around the world because it is enjoyed by many people due to its sensitivity nature. The leaves are of medicinal importance, the leaves are pulped into a paste which is then rubbed on to people as the treatment for them suffering pains (Figure 1).

Plant Profile

Botanical source: *Mimosa pudica*

Family: Fabaceae

Common name: touch-me-not plant

Parts used: leaves

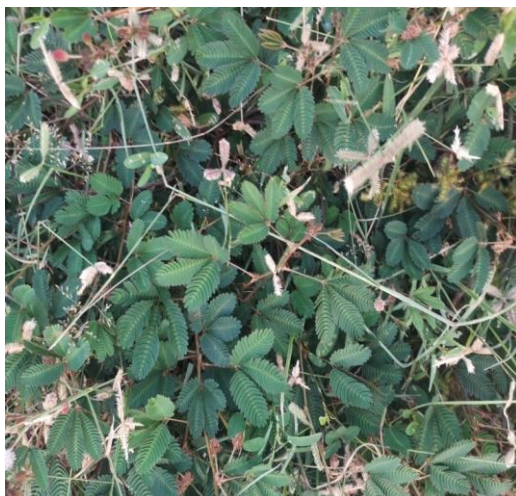


Figure 1. Whole plant

PHYTOCHEMICAL STUDIES**Collection and Authentication**

The plant specimen (Leaves) for the proposed study was collected during the month of July 2017 from the garden of Vaageswari College of pharmacy. It was identified and authenticated by L Rasingam, scientist incharge of Botanical Survey of India (BSI), Hyderabad. A voucher specimen no. BSI/DRC/2017-2018/Tech/470 has been deposited for further reference Animal Ethical Committee no: VCP/cology/006/11/2017 [6-9].

Extraction of Tannin and Flavonoid Leaf Fractions

The leaves of *Mimosa pudica* were shade dried and coarsely powdered. About 300 gm of powdered drug was extracted with Acetone + Water (7:3) by cold maceration method. After 72 hours of maceration it was filtered. To this filtrate, Petroleum Ether (it is generally used to remove waxy substances in leaf extract followed by acetone for removal of chlorophyll) is added in a separating funnel to remove the Chlorophyll. After removal of chlorophyll, petroleum ether layer was decanted. Again to this filtrate add a saturated solution of sodium chloride (salt water works to pull water from organic layer to water layer) and vitamin-C or Ascorbic Acid (methods used for bioactive compound extraction like flavonoids, tannins and phenolic acids), once again Filter the solution. To the filtered solution add Ethyl Acetate solvent (flavonoids separated in these solvent) in a separating funnel once again. After gradual shaking of both these solvents in a separating funnel, decanted ethyl acetate solvent gives the flavonoid. The separating funnel contains aqueous layer. After complete extraction, the extract was concentrated by distilling off the solvent and then evaporated to dryness under reduced pressure using vacuum flash evaporator which gives tannins. The results are shown in Table 1.

Table 1. Isolation of flavonoid and tannin leaf fraction

Extract/Fraction	Percentage yield (% w/w)	Color	Consistency
Flavonoid leaf fraction (FLF)	7.9	Light green	Greasy
Tannin leaf fraction (TLF)	8.9	Brownish	Hard

Phyto Chemical Screening

The preliminary phytochemical screening of crude extract have been done to detect the presence of active chemical constituents these phytochemicals are present in different parts of plants which have been utilized by both animals and humans.

The leaves of tannins and flavonoids fractions were subjected to qualitative phytochemical test for identification of constituents. The results are shown in Tables 2, 3 and Figures 2-5.

Table 2. Chemical tests for tannins

S. no	Test	Result
1	Ferric Chloride	+
2	Lead Acetate	+

Table 3. Chemical tests for flavonoids

S. No	Test	Result
1	Schinoda	+
2	Ferric Chloride	+

PHARMACOLOGICAL STUDIES

Wound Healing Activity

Many plants synthesize substances that are useful to the maintenance of health in humans and other animals. A number of traditions came to dominate the practice of herbal medicines for various effective human benefits at the end of the twentieth century. With a view to increasing the wide spectrum of medicinal usages, the present day requires a new biologically active fraction which exhibit wound healing activity as local applications. The availability of market products capable of stimulating the process of wound repair is still limited. Hence the present investigation was focused in the direction of establishment of tannin fraction in the form of ointment for wound healing activity. The results are shown in Table 4 and Figure 6 [10-15].

Table 4. Wound healing activity of *Mimosa pudica* and its leaf fractions of flavonoids and tannins

Control (length and width)							
4 th day		8 th day		12 th day		25 th day	
L	w	L	w	L	w	L	w
1 ± 0.43	1.46 ± 0.39	0.76 ± 0.42	1.41 ± 0.43	0.51 ± 0.31	0.7 ± 0.34	0.46 ± 0.19	0.53 ± 0.25
Standard (length and width)							
4th day		8th day		12th day		18th day	

L	W	L	W	L	W	L	W
1.15 ± 0.40	1.3 ± 0.34	0.86 ± 0.31	1.1 ± 0.34	0.58 ± 0.26	0.71 ± 0.31	0.45 ± 0.26	0.66 ± 0.26
Test drug-LFF (length and width)							
4th day		8th day		12th day		19th day	
L	W	L	W	L	W	L	W
1.1 ± 0.34	1.7 ± 0.34	0.6 ± 0.23	1.3 ± 0.34	0.53 ± 0.19	0.9 ± 0.34	0.41 ± 0.21	0.53 ± 0.25
Test drug-LTF (length and width)							
4th day		8th day		12th day		20th days	
L	W	L	W	L	W	L	W
1.66 ± 0.77	1.6 ± 0.34	1.05 ± 0.37	1.01 ± 0.31	0.8 ± 0.34	0.76 ± 0.42	0.35 ± 0.17	0.41 ± 0.24

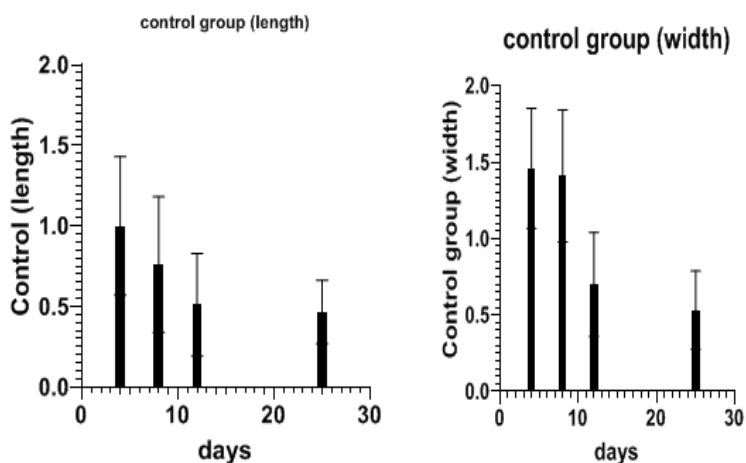


Figure 2. Wound healing activity of *Mimosa pudica* with control group (a) L-Length, (b) W-Width on 4th, 8th, 12th and 25th days

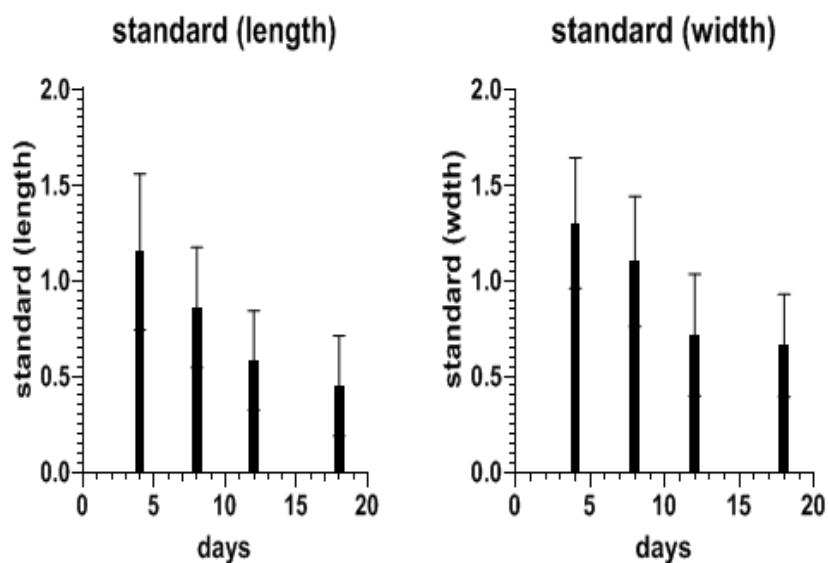


Figure 3. Wound healing activity of *Mimosa pudica* on standard (a) L-Length, (b) W-Width on 4th, 8th, 12th and 18th days

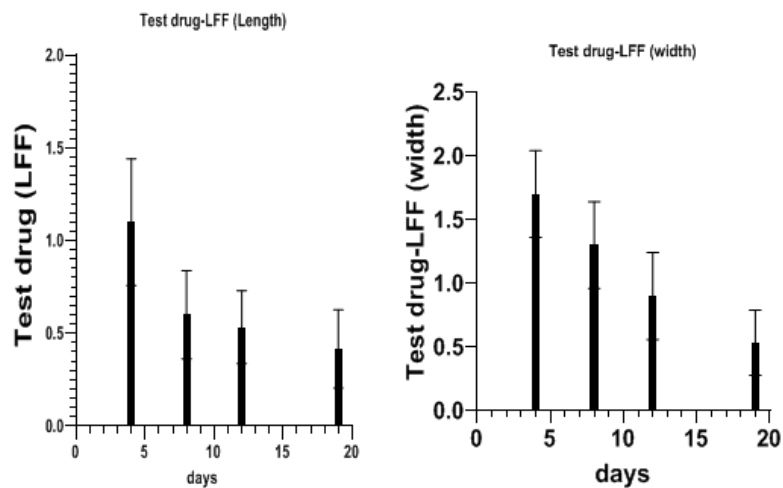


Figure 4. Wound healing activity of *Mimosa pudica* with test drug-LFF (a) L-Length, (b) W-Width on 4th, 8th, 12th and 19th days

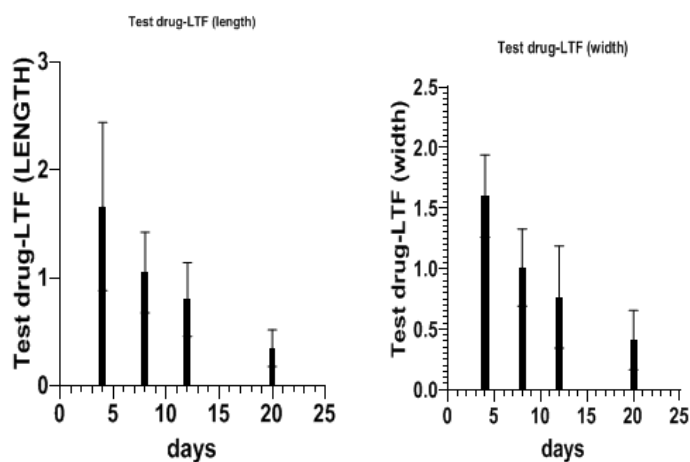


Figure 5. Wound healing activity of *Mimosa pudica* with test drug-LTF (a) L-Length, (b) W-Width on 4th, 8th, 12th and 20th days



Figure 6. Wound created rabbits

Formulation of Ointment

Ointment: Ointment is soft semisolid preparation meant for external application on to the skin or mucous membrane in which the drugs substances are emulsified and suspended [16-20].

Preparation of Ointment

Procedure: Cetostearyl alcohol, white soft paraffin and yellow soft paraffin were melted together. Wool fat is dissolved separately in purified water and warmed it to almost at temperature (about 60°C). Then add warmed 5% of tannin leaf fraction (TLF) and 5% flavonoid leaf fraction (FLF) separately on to the melted mixture and stir thoroughly, cool it and store it in a suitable container [21-25].

Excision Wound Model

On the shaved back of the rabbit light ether anesthesia is given. The skin of the impressed area was excised carefully. Animals are kept in separate cages. The day on which wound was made consider as day '0' (Zero). Animals divided into four groups of each with 4 animals. Group A considers as control and treated with simple ointment (eg. Bees wax, Cetostearyl alcohol etc.), group B consider as standard and treated with 5% w/w Povidone iodine ointment, group C and group D are *Mimosa pudica* treated group and applied ointment 5% respectively. The percentage of wound closure was recorded on day 4, 8, 12, 18, 19, 20 and 25. Wound area was traced and measured plan metrically with the help of sqmm graph paper [26-29].

RESULTS

The present work covers study on a wound healing activity of the leaves of *Mimosa pudica* Linn.

DISCUSSION

The ethno-botanical studies reviewed that the leaves of the plant *Mimosa pudica*, are used for wound healing activity. The young leaves are used as tonic in the diseases of the digestive function and is said to be remedy for toothache. It has a high content of tannin substances reviewed from literature. so the present work was focused to isolate tannin rich fraction and it was evaluated. The tannin rich fractions of the plant was formulated in the ointment form and studied for wound healing activity.

Phytochemical Study

Phytochemical screening was carried out to identify the phyto-constituents present in the leaf fraction which shows the presence of tannin. The total tannin content was estimated by using spectrophotometer method. The results indicate that the content of tannin was found to higher in the plant.

Wound Healing Activity

Wound healing, a complex sequence of events, is initiated by the stimulus of injury to the tissues. A positive result shows the release of some factors by wounding of tissues. The result of present study indicates that 70% hydro alcoholic leaf strengths (5% leaf) exhibited significant wound healing promoting activity. However, this effect was found to be concentration related fashion where 5% ointment promotes significant wound-healing activity by increasing cellular proliferation, formation of granulation tissue, synthesis of collagen and by increase in the rate of

wound contraction as compared to the control animals. This was evident by faster rate of wound closure and epithelization period in excision wound model. Further phytochemical studies are needed to isolate and characterize the individual tannin, and identify the compound which is responsible for wound healing activity.

CONCLUSION

From this study, it is concluded that *Mimosa pudica*. Tannin leaf fraction and flavonoid leaf fraction have significant wound healing models and the ointment form was formulated, screened for In vivo wound healing. It showed significant percentage wound protection at the tested concentration. The wound healing activity is probably due to the presence of tannins (gallic acid and tannic acid) and flavonoids (rutin and quercetin) Further studies need to be isolate individual tannin and flavonoid biological potency conducted by various preclinical and clinical trials of the isolated compounds.

In conclusion, the isolated plant leaf fractions contains high amount of flavonoids as the leaves are rich in flavonoid content than tannins.

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